Neonatal Tetanus in Rural Bangladesh: Risk Factors and Toxoid Efficacy

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A B S T R A C T

Objectives. Tetanus continues to be a leading cause of neonatal death in Bangladesh as in other developing countries, yet little is known about risk factors or the efficacy of tetanus toxoid in this setting.

Methods. In May 1990, mothers of 6148 infants born alive between March 15, 1989, and March 14, 1990, in 30 rural unions of Rajshahi Division in Bangladesh were interviewed. Three surviving controls for each neonatal tetanus death were matched for sex, residence, and date of birth.

Results. Of 330 neonatal deaths, 112 met the case definition for tetanus. Risk was increased with a history of neonatal tetanus in a previous child, application of coconut oil to the vagina, and use of multiple ties on the umbilical cord. Risk was reduced by the birth attendant washing hands and using a cleaned cord-cutting tool. Risk was not reduced by a maternal history of two doses of tetanus toxoid (TT2), although estimated efficacy of TT2 was 45% (95% confidence interval = 16% to 64%). Subsequent to the survey, a reference laboratory reported no potency in three consecutive lots of tetanus vaccine from the production laboratory in Bangladesh.

Conclusions. These findings identify high-risk mothers, stress the importance of washing hands and cleaning the cord-cutting tool, and demand improved quality control of tetanus vaccine production. (*Am J Public Health.* 1992;82:1365–1369)

Introduction

Neonatal tetanus continues to be a major cause of death in developing countries, accounting for more than 654 000 deaths in 1990,[1] yet few analytic studies have been done. The value of such studies was most recently demonstrated by a report from Pakistan, which suggested that the risk of neonatal tetanus could be significantly reduced by eliminating the application of ghee (clarified butter) to the umbilical stump during the first several days of life—a common practice in that country and a previously unidentified risk factor.[2] Neonatal tetanus can be totally prevented both by two injections of tetanus toxoid vaccine (TT2) to the mother, with the second dose at least 4 weeks after the first and at least 30 days prior to delivery, and by sterile cord care. But vaccination coverage of pregnant women in most developing countries remains low, and unsafe birth practices persist.[1]

Bangladesh has one of the highest rates of neonatal tetanus in the world.[1] The government has addressed this problem through a vigorous program of vaccination, training of traditional birth attendants, and promotion of safe-delivery kits. In addition, a national study was begun in February 1990 to identify risk factors for neonatal tetanus. This study, which was patterned after the study in Pakistan, was conducted by Global 2000, Inc, in collaboration with the Ministry of Health and Family Planning, and the Task Force for Child Survival and Development. We report here the results of the Rajshahi Division survey, with emphasis on the findings not only of potential risk factors but also of the low efficacy of tetanus toxoid, apparently due to the use of low-potency vaccine.

Methods

Study Population

Bangladesh is divided into four administrative divisions of approximately equal populations. The survey reported here was of Rajshahi Division, occupying the northwest quadrant of the country with a 1981 rural population of 20 009 351.[3] Cluster sampling was done according to World Health Organization (WHO) guidelines.[4] Thirty rural unions (average population 18 000) were systematically selected from the total of 1089 listed in the 1981 census by using a random start and a sampling interval equal to the cumulative population divided by 30, making the probability of selection proportionate to population size. Urban areas were excluded.

Each union is divided into three wards of approximately equal populations, and each ward is divided into a variable number of *mauzas* (average population 1400). The starting point for the survey in each selected union was the first

TABLE 1—Circumstances of Delivery for Neonatal Tetanus Cases (n = 112) and Matched Controls (n = 336)

	Cases		Controls	
	No.	%	No.	%
Place of delivery				
In hospital or clinic	0	0	6	1.8
At home	112	100	330	98.2
Birth attendant				
Relative	63	56.3	182	54.2
Unrelated traditional birth attendant	23	20.5	84	25.0
Health care worker	0	0	8	2.4
Self (delivered without assistance)	26	23.2	62	18.4
Delivery practices[a]				
Attendant washed hands	45	40.2	182	54.2
Perineum washed	41	36.6	146	43.5
Mustard oil applied to vagina	22	19.6	69	20.5
Coconut oil applied to vagina	9	8.0	12	3.6
Safe-delivery kit used	2	1.8	7	2.1
Cord-cutting tool cleaned before use	55	49.1	231	68.8
Multiple cord ties used	41	36.6	89	26.5
Mother's vaccination status at delivery				
History of TT2[b]	33	29.5	122	36.3
No history of TT2	79	70.5	214	63.7

[a]Figures exceed total (100%) because categories are not exclusive.
[b]Two doses of tetanus toxoid at least 4 weeks apart, with the second dose at least 30 days before delivery.

household encountered in a *mauza* selected by use of a random number table. The survey then proceeded from house to house within the same ward until a target of 67 mothers of eligible infants were interviewed. An eligible infant was one who was born alive between March 15, 1989, and March 14, 1990. The survey continued then from the nearest house in the next nearest ward and so on until all three wards were surveyed, for a total target of 201 interviews in each selected union. Interviews completed in excess of the target (usually 3 or 4 per union) were also included in the analysis.

Interviews were conducted by 10 teams of six female interviewers using a standardized questionnaire. Interviewers received 6 days of instruction by Global 2000 staff and faculty of the National Institute of Preventive and Social Medicine, and 4 days of closely supervised field testing. The survey was completed between May 3 and May 28, 1990.

Case Definition

This was a case-control study. A case was an eligible infant who was normal at birth but who died at 3 to 30 days of age following illness, with generalized spasms and at least three of the following four signs: trouble opening mouth, cessation of sucking, clenched hands, and boardlike rigidity.

Infants who were seen at a health care facility and given a diagnosis other than tetanus (as reported by the mother) were excluded as cases.

Control Selection

Three living infants—matched for sex, residence (union), and date of birth to control for the effects of circumcision, regional variations in birth practices, and recall—were selected as controls for each case. When more than three infants were eligible as controls, priority for selection was given to those with birth dates closest to the case's and to those who resided in the same ward, in that order. Control selection differed from that used in the Pakistan study in that the mothers' vaccination status with tetanus toxoid was disregarded in selecting control infants.

Data Analysis

Data were double entered into an Epi Info, Version 5, data file and were analyzed using the same software package.[5] Univariate analysis of matched data used the same calculations as stratified Mantel-Haenszel analysis to calculate matched odds ratios, with each stratum consisting of a single matched case-control group. The 95% confidence intervals (CIs) for matched odds ratios were calculated using

standard methods,[6] and all probabilities were based on two-tailed tests.

Multivariate analysis was performed using EGRET software.[7] Data were defined by the DEF program for conditional logistic regression and analyzed by the PECAN program.[7] The log likelihood ratio statistics, degrees of freedom, and probability statistics for the model, and the 95% CIs and P values for each variable, were calculated by the program. Interaction was examined by evaluating the P value for main and product terms as well as significant differences in likelihood ratio statistics between noninteractive and interactive models. Multicolinearity among covariables was examined by modeling and correlation procedure.

Vaccine efficacy based on the entire study population was calculated using the following formula: $(Iu-Iv)/Iu$, where Iu is the incidence of neonatal tetanus in infants with no maternal history of tetanus toxoid vaccine prior to delivery and Iv is the incidence of neonatal tetanus in infants with a maternal history of TT2. This formula is equal to $1-(Iv/Iu)$, or 1-(risk ratio), and 95% CIs for the risk ratio were used to calculate confidence intervals for efficacy.[8] Because vaccine efficacy was not an anticipated issue, incidence rates could not be weighted to account for differences in sample union populations. However, the difference between weighted and unweighted rates using this sampling procedure (the EPI method) is considered small and acceptable.[9]

Results

Descriptive Statistics

A total of 43 791 households were surveyed, and the mothers of 6148 eligible infants were interviewed. The circumstances of delivery are summarized in Table 1. There were 330 neonatal deaths (occurring up to 30 days of age). One hundred twelve neonatal deaths met the case definition for tetanus, resulting in a neonatal tetanus death rate of 18.2 per 1000 live births in our study population. Three infants who otherwise met the case definition were excluded because they had been seen at a health care facility and the mothers reported a cause of death other than tetanus (two "evil spirits," one "milk disease"). Only 21 (18.8%) of the cases were seen at a health care facility.

Male cases tended to be seen more often than female cases at a health care facility (23% vs 15%, $P = .3$), but the proportion of males among cases (50.9%) did not differ from that among all live births (51.1%).

None of the male infants was circumcised before 30 days of age.

The dates of birth for cases and for all live births are shown in Figure 1. The cyclic pattern of live births is identical with that observed prospectively in Matlab, Bangladesh.[10] The July through November peak in neonatal tetanus deaths has also been previously observed in India.[11-13] The mean age at death for cases was 10.7 days (SD = 6.3); the median age was 8 days.

Application of substances to the vagina and to the cord stump at the time of delivery were common practices and usually involved the use of mustard oil, which was heated before use. Mustard oil was also commonly applied several times to the cord stump during the first few days of life. Ghee was never applied to either the vagina or the cord stump.[2] Cow dung was applied to the cord stumps of only three infants on the day of delivery, one of whom died of tetanus. Cow dung was also applied to the cord stumps of eight infants only on days subsequent to birth, and none of those infants died of tetanus.

Risk Factors

The results of a multivariate analysis of risk factors for neonatal tetanus are shown in Table 2. The striking finding was that a maternal history of TT2 failed to be protective. However, hand washing and the use of a cleaned tool to cut the umbilical cord, regardless of what tool was used, were both associated with decreased risk. The risk increased when coconut oil was applied to the vagina and when the umbilical cord was tied in more than one place.

A history of neonatal tetanus in a previous child was a significant risk factor; its odds ratio, obtained by conditional logistic regression as a single variable in the model, was 3.63 (95% CI = 1.30, 10.14; P = .01), but it was excluded from the final model because of colinearity with hand washing. This statistical association was also confirmed by stratified Mantel-Haenszel analysis. Such a history was present in 27.7% (31/112) of cases and in only 7.7% (26/336) of controls. Of previous children born to case mothers, 14.3% (39/273) were reported to have died of neonatal tetanus, compared with 4.5% (33/740) of previous children born to control mothers (x^2 = 29.14, P < 10^{-6}). The mean total number of live births was 3.4 (SD = 2.6) for mothers of cases and 3.2 (SD = 2.2) for mothers of controls. Similarly, mean maternal age was 26.1 years (SD = 6.3) for cases and 25.5 years (SD = 5.7) for controls.
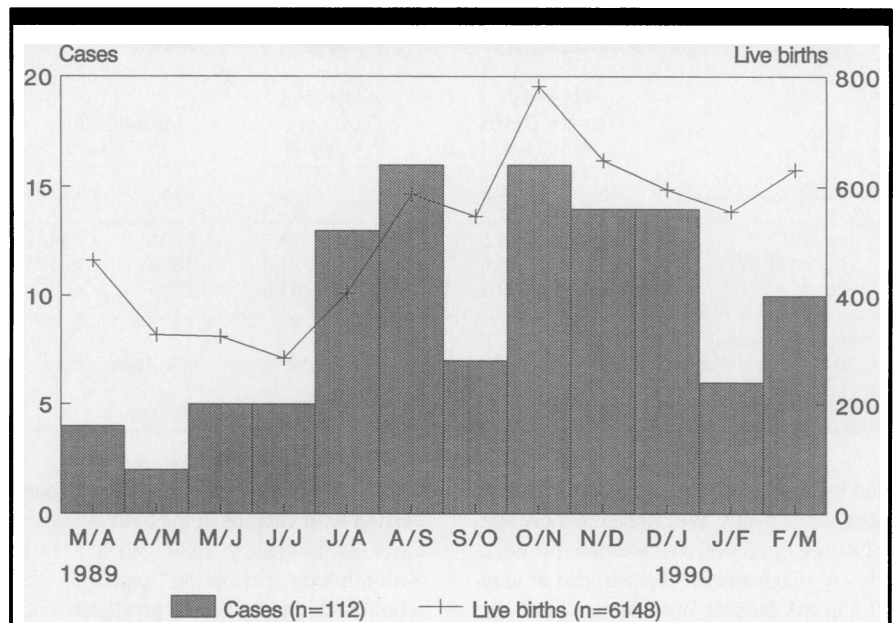

FIGURE 1—Month (English equivalent of Bengali month) of birth for neonatal tetanus cases and all live births.

TABLE 2—Multivariate Analysis[a] of Risk Factors for Neonatal Tetanus

	Coefficient	Standard Error	Odds Ratio	95% CI	P
History of TT2[b]	−0.2712	.267	0.76	0.45, 1.29	.3
Hand washing	−0.7150	.257	0.49	0.30, 0.81	.005
Cleaned cutting tool	−0.9234	.258	0.40	0.24, 0.66	<.001
Vaginal coconut oil	1.150	.526	3.16	1.13, 8.86	.03
Multiple cord ties	0.6919	.276	2.00	1.16, 3.43	.01

Note. Likelihood ratio statistic on 5 df = 34.711, P < .001.
[a]Conditional logistic regression by PECAN procedure.[7]
[b]Two doses of tetanus toxoid at least 4 weeks apart, with the second dose at least 30 days before delivery.

Other factors of interest, including use of a safe-delivery kit, washing of the perineum, application of substances to the umbilical cord, and the presence of a trained birth attendant (8 cases, 35 controls), were not associated with risk and thus were not included in the final regression model.

Efficacy of TT2

The maternal vaccination status at the time of delivery for neonatal tetanus cases and all other live births in our study population is shown in Table 3. The efficacy of a maternal history of TT2, based on these figures, was 45% (95% CI = 16%, 64%). The efficacy of a maternal history of TT2, based on the odds ratio obtained by multivariate analysis of the case-control data, was 24% (95% CI = −29%, 55%). Although less precise, the latter efficacy estimate may be more valid as it is adjusted for the effects of other risk factors. However, both estimates

are far below the level expected for a potent vaccine. Verification of vaccination by medical records was difficult to achieve because the medical records were kept with mothers, and only 48.3% of all mothers with a history of TT2 (1209/2505) possessed a medical record at the time of interview.

Discussion

The accuracy of our case ascertainment is supported by the fact that the rate of neonatal tetanus death observed in our study population is similar to that reported by others.[14-16] Had we used a less restrictive case definition of only a history of generalized spasms preceding death during the first 30 days of life, as has been used elsewhere,[17] the number of neonatal tetanus deaths in our sample would have increased to 259 (42.1 per 1000 live births). A similar increase would have resulted

TABLE 3—Maternal Vaccination Status at Time of Delivery						
	Neonatal Tetanus Cases (n = 112)		No Neonatal Tetanus (n = 6036)		All live births (n = 6148)	
Vaccination Status	No.	%	No.	%	No.	%
None[a]	56	50.0	2278	37.7	2334	38.0
History of TT1[b]	23	20.5	1286	21.3	1309	21.3
History of TT2[c]	33	29.5	2472	41.0	2505	40.7

[a]Never vaccinated.
[b]Received one dose of tetanus toxoid at least 30 days before delivery. (May have received a second dose less than 30 days before delivery.)
[c]Received two doses of tetanus toxoid, with second dose at least 30 days before delivery.

had we relied only on the mothers' report of cause of death. We chose the more specific case definition to maximize the accuracy of diagnosis for the purpose of identifying risk factors, but this may have also resulted in an underestimate of the incidence of neonatal tetanus death.

Our observation that tetanus toxoid failed to provide the expected high level of protection was not the first to indicate that there may be a problem with the Bangladesh vaccine. Anecdotal reports from physicians had already described apparent vaccine failures; and a 1989 WHO survey of more than 4000 live births, equally divided between areas with high vaccine coverage and areas with low vaccine coverage, found almost twice the expected rate of neonatal tetanus death in the high coverage area.[18]

Subsequent to our findings, a WHO reference laboratory reported no potency in samples from three consecutive lots of tetanus toxoid produced in Bangladesh in July 1990 (J. Milstien, WHO, personal communication, October 17, 1990), and a serosurvey of 100 women vaccinated in September and October 1990 with two doses of tetanus toxoid vaccine found that only 31% had developed protective levels of antibody (N. Abiprojo, UNICEF, personal communication, July 29, 1991).

Since 1983, routine potency testing of tetanus toxoid in Bangladesh has been performed in the same facility by the same staff that produced all the tetanus toxoid vaccine for the national immunization program. There is no national control authority in Bangladesh to certify vaccine safety or potency. Currently, tetanus toxoid production in Bangladesh has been suspended, and the immunization program has continued with the use of imported vaccine.

It has not yet been determined when the problem with vaccine quality control began. Documented vaccination histories

from Rajshahi suggest that the problem existed with vaccine in the field at least as early as February 1989. With a 1- to 3-month-long distribution "pipeline," this would include all vaccine produced since January 1989—approximately two thirds of all tetanus vaccine ever produced in Bangladesh—with more than 40 000 000 doses administered. It is possible that the problem was intermittent, but this cannot be determined from available information as no records are kept of the distribution of vaccine by lot number.

Nevertheless, a potent preparation of tetanus toxoid is one of the safest, most effective vaccines available and is the primary element of any neonatal tetanus control program.[1,14] Bangladesh has achieved a remarkably high level of vaccine distribution. If immunization activities are sustained, corresponding decreases in tetanus mortality can be expected with the introduction of a potent vaccine. The very high risk associated with a maternal history of previous neonatal tetanus death, which was initially observed elsewhere[2] and was confirmed in this study, should make this group of women a particularly high priority for immunization.

Failure of the tetanus vaccine in Bangladesh, however, must also serve as a reminder that such high-technology interventions are not always effectively implemented and should not be relied upon solely, especially where simple measures such as hand washing and cleaning of the cord-cutting tool have proven effective and reliable.

The use of a trained traditional birth attendant was not observed to be protective, possibly because of inaccuracies in both the mothers' ascertainment of attendant training and the low number of trained attendants involved. For these reasons and because certain behaviors of the birth attendants were significantly related to decreased risk, our results should

not discourage the training of traditional birth attendants as a control measure for neonatal tetanus. Similarly, the low number of safe-delivery kits used did not allow a meaningful evaluation of their effect.

The increased risk associated with the use of multiple cord ties has not been observed elsewhere and awaits further clarification. However, the increased risk associated with the vaginal use of coconut oil is an important finding and is in line with the risk associated with the vaginal use of ghee in Pakistan (H. Traverso, personal communication, December 18, 1990). The mechanism by which the vaginal use of coconut oil increases the risk of neonatal tetanus is unknown, but the increased risk could be due to contamination of the oil by the uncleaned hands of the birth attendant.

Unfortunately, control of neonatal tetanus in Bangladesh will be delayed due to the failure to maintain vaccine potency. Given that fewer than one in five neonatal tetanus cases had contact with the health care system and that the problem with the tetanus vaccine was not brought to light earlier, the need for improved surveillance to evaluate program effectiveness and to monitor vaccine quality and distribution in the field is indicated. Such improvement should include the creation of a separate quality control laboratory and a national control authority. More work is needed to determine how surveillance might best be structured and what thresholds for action are appropriate. □

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