Am. J. Trop. Med. Hyg., 96(2), 2017, pp. 415–420
doi:10.4269/ajtmh.16-0452

Tetanus Immunity Gaps in Children 5–14 Years and Men ≥ 15 Years of Age Revealed by Integrated Disease Serosurveillance in Kenya, Tanzania, and Mozambique

Heather M. Scobie,[1]* Minal Patel,[1] Diana Martin,[2] Harran Mkocha,[3] Sammy M. Njenga,[4] Maurice R. Odiere,[5] Sonia Pelletreau,[2] Jeffrey W. Priest,[6] Ricardo Thompson,[7] Kimberly Y. Won,[2] and Patrick J. Lammie[2]

[1]*Global Immunization Division, Centers for Disease Control and Prevention, Atlanta, Georgia;* [2]*Division of Parasitic Diseases and Malaria, Centers for Disease Control and Prevention, Atlanta, Georgia;* [3]*Kongwa Trachoma Project, Kongwa, Tanzania;* [4]*Eastern and Southern Africa Centre of International of Parasite Control, Kenya Medical Research Institute, Nairobi, Kenya;* [5]*Centre for Global Health Research, Kenya Medical Research Institute, Kisumu, Kenya;* [6]*Division of Foodborne, Waterborne and Environmental Diseases, Centers for Disease Control and Prevention, Atlanta, Georgia;* [7]*Instituto Nacional de Saúde, Maputo, Mozambique*

Abstract. Recent tetanus cases associated with male circumcision in Eastern and Southern Africa (ESA) prompted an examination of tetanus immunity by age and sex using multiplex serologic data from community surveys in three ESA countries during 2012–2013. Tetanus seroprotection was lower among children 5–14 years versus 1–4 years of age in Kenya (66% versus 90%) and Tanzania (66% versus 89%), but not in Mozambique (91% versus 88%), where children receive two booster doses in school. Among males ≥ 15 years of age, tetanus seroprotection was lower than females in Kenya (45% versus 96%), Tanzania (28% versus 94%), and Mozambique (64% versus 90%). Tetanus immunity from infant vaccination doses wanes over time, and only women of reproductive age routinely receive booster doses. To prevent immunity gaps in older children, adolescents, and adult men, a life-course vaccination strategy is needed to provide the three recommended tetanus booster doses.

INTRODUCTION

Tetanus continues to be a global public health problem, with the highest disease burden remaining in Africa and southeast Asia.[1,2] Case fatality ratios for tetanus vary from 10% to 70%, depending on the treatment and age of the patient.[2,3] Accurate estimation of tetanus cases and deaths is challenged by underreporting of neonatal cases (occurring within the first 28 days of life) and lack of routine reporting of nonneonatal cases in many countries.[2]

Globally, tetanus vaccination started in 1974 as part of the Expanded Program on Immunization (EPI), which was implemented in all countries by the mid-1980s.[3] The programmatic goals of tetanus control are eliminating maternal and neonatal tetanus (MNT; < 1 case per 1,000 live births in every district) and preventing tetanus in all age groups.[3] In most countries, infants receive tetanus toxoid–containing vaccine (TTCV), for example, diphtheria–tetanus–pertussis (DTP) or DTP–hepatitis B–*Haemophilus influenzae* type b (pentavalent), through routine immunization (RI) services at 6, 10, and 14 weeks of age. Women of reproductive age (WRA), defined as 15–49 years of age, also receive up to five doses of TTCV, for example, tetanus toxoid (TT) or tetanus–diphtheria (Td), either during antenatal care visits, mass campaigns, or at school, depending on the country.[3] Since 2006, the World Health Organization (WHO) has recommended three booster doses for both sexes in childhood, adolescence, and early adulthood, but these doses are not provided in many countries, especially in sub-Saharan Africa.[3–5] After infancy, males might receive tetanus booster doses during treatment of injuries or military service.

Among the 20 countries in the WHO subregion of Eastern and Southern Africa (ESA), all provide the primary series of TTCV to infants (6-, 10-, and 14-week schedule; except for Botswana with a 2-, 3-, and 4-month schedule; and Seychelles with a 3-, 4-, and 5-month schedule) and TTCV booster doses to WRA.[4] MNT elimination (MNTE) has been achieved by all ESA countries, except Kenya, South Sudan, and the Somali region of Ethiopia. Tetanus booster doses for both sexes in childhood and adolescence are included in the immunization schedules of nine ESA countries, with four countries providing the three TTCV booster doses needed for lifelong immunity when vaccination begins in infancy (Table 1).[3–5] Voluntary medical male circumcision (VMMC) is performed for adolescent and adult males in 14 ESA countries as part of human immunodeficiency virus prevention programs (Table 1).[10] During 2012–2016, 12 tetanus cases were determined to be causally associated with VMMC among clients 11–47 years of age in five ESA countries.[2,10,11] Tetanus immunity gaps likely exist due to waning immunity from infant doses, but recent serologic evidence from the region is too limited to inform immunization strategies among VMMC clients.[11]

In the three ESA countries of Tanzania, Mozambique, and Kenya, EPI began in 1975, 1979, and 1980, and WHO–United Nations Children's Fund (UNICEF) estimates of national coverage with the third dose of DTP (DTP3) vaccine among children < 12 months of age in 2013 were 91%, 78%, and 76%, respectively.[12] Of the three countries, only Mozambique provides two childhood TTCV booster doses to both sexes in the first and second grades of school (Table 1).[4,5] To evaluate immunity gaps in Kenya, Tanzania, and Mozambique, we assessed tetanus seroprotection by age group and sex. Serologic data were collected in the context of community surveys conducted in rural areas to examine the broad health impact of treatment programs for neglected tropical diseases (NTDs), for example, schistosomiasis, trachoma, soil-transmitted helminths (STH), and lymphatic filariasis (LF). The evaluation included integrated disease serosurveillance with a multiplex bead assay (MBA) for ≥ 30 bacterial, viral, and parasitic antigens, including tetanus.

MATERIALS AND METHODS

Survey methods. In Kenya during May–July 2012, we enrolled and collected venous blood from a convenience

*Address correspondence to Heather M. Scobie, Global Immunization Division, Centers for Disease Control and Prevention, 1600 Clifton Road, MS-E98, Atlanta, GA 30333. E-mail: hscobie@cdc.gov

TABLE 1
TTCV booster doses provided to both sexes in 20 Eastern and Southern African countries

Country	No. of TTCV booster doses	TTCV booster schedule
Botswana*	3	DTP at 18 months; DT at 7 years (grade 1); TT at 13 years (grade 7)
Comoros	–	–
Eritrea	–	–
Ethiopia*	–	–
Kenya*	–	–
Lesotho*	1	DT at 18 months
Madagascar	–	–
Malawi*	–	–
Mauritius	3	DT at 18 months and 5 years (grade 1); TT at 10–11 years (grade 6)
Mozambique*	2	TT at grade 1 and grade 2†
Namibia*	2	DT at 5 years (grade 1) and 10 years (grade 5)
Rwanda*	–	(historically provided DTP at 15 months that was discontinued)[6]
Seychelles	3	DTP at 18 months; DT at 6 years (grade 1); TT at 15 (secondary grade 3) and 25 + years
South Africa*	3	DTP-Hib-IPV/DTP-Hib-HepB-IPV at 18 months; Td at 6 and 12 years (in school)
South Sudan	–	–
Swaziland*	1	(historically provided DTP at 18 months that was discontinued)[7]; DT at 5 years
Tanzania*	–	–
Uganda*	–	–
Zambia*	–	(historically provided DTP at 12 months that was discontinued)[6]
Zimbabwe*	1	DTP at 18 months; (DT at 5 years discontinued 2008)[8]

DT = diphtheria–tetanus; DTP = diphtheria–tetanus–pertussis; HepB = hepatitis B; Hib = *Haemophilus influenzae* type b; IPV = inactivated poliovirus vaccine; Td = tetanus–diphtheria (with reduced content of diphtheria antigen); TT = tetanus toxoid; TTCV = tetanus toxoid–containing vaccine. Sources: WHO immunization schedule data, WHO school-based immunization data, UNICEF immunization summaries.[4,5,9]
*Country performing voluntary medical male circumcision.
†Not in official national immunization schedule, but provided through school.

sample of 6,238 individuals living in 30 villages in Mbita District, Homa Bay County, as part of an evaluation of schistosomiasis and STH control programs. We used the MBA to test a random selection of 397 serum samples from individuals 1–85 years of age living in 12 of the 30 villages. During October–December 2012 in Tanzania, we collected capillary blood as dried blood spots (DBS) from a convenience sample of 2,308 individuals 1–90 years of age and living in eight villages of Kongwa District, Dodoma Region, as part of an evaluation of trachoma and LF control programs; only 1,585 (69%) had enough serum for MBA testing. In Mozambique, we collected DBS from a convenience sample of 1,423 individuals ≥ 12 months of age in two villages in January and August 2013, respectively, as part of an evaluation of LF and schistosomiasis control programs in Morrupula District, Nampula Province; only 1,231 (87%) had enough serum for MBA testing. Evaluation sites were chosen based on the presence of endemic NTD burden and/or initiation of treatment programs.

Laboratory. For DBS, blood was collected onto a filter paper with six extensions, each calibrated to absorb 10 µL volume (TropBio Pty Ltd, Queensland, Australia), and then allowed to air-dry ≥ 4 hours. Depending on the country, serum was prepared from whole blood by centrifugation, or serum was eluted from DBS as previously described.[13,14] TT (Massachusetts Biologic Laboratories, Boston, MA) was coupled to SeroMap microspheres (Luminex Corp., Austin, TX) at the Centers for Disease Control and Prevention (CDC) with 12.5 µg of toxoid per 12.5×10^6 beads in buffer containing 50 mM 2-(N-morpholino) ethanesulfonic acid and 0.85% NaCl at pH 5.0, as previously described.[13,15] Serum samples were diluted 1:400 and assayed in duplicate for IgG (along with control sera) against multiple bacterial, viral, and parasitic disease antigens at CDC in Atlanta, GA, or the Kenya Medical Research Institute (KEMRI) in Nairobi, Kenya, using the conditions previously described.[13] Beads suspended in 100 µL of phosphate-buffered saline in microtiter plates were read in a Bio-Plex 200 instrument equipped with Bio-Plex Manager 6.1 software (Bio-Rad, Hercules, CA), and the average of the median fluorescent intensity values of duplicate wells minus the background fluorescence (MFI-bg) was recorded. Laboratory testing was standardized by running a dilution series of the TE-3 WHO reference serum (National Institute for Biological Standards and Control, United Kingdom). The tetanus MBA was previously validated against the gold-standard double antigen enzyme-linked immunosorbent assay with 99% sensitivity and 92% specificity.[13]

Ethical considerations. For all sites, adults ≥ 18 years of age provided written consent, children 6–17 years of age provided verbal assent, and adults provided written consent for children < 18 years of age. This evaluation was approved by the Scientific Steering and Ethics Review Committees of KEMRI in Kenya, the National Health Research Ethics Review Committee of the National Medical Research Institute in Tanzania, and the National Bioethics for Health Committee and Instituto Nacional de Saúde in Mozambique. The Human Research Protection Office of the U.S. CDC approved the evaluations in Kenya and Tanzania through reliance on the respective institutional review boards and determined the evaluation in Mozambique to be a nonresearch, public health program evaluation activity according to its human subjects' procedures.

Statistical analysis. MBA results in MFI-bg values were transformed to international units per ml (IU/mL) using least squares regression of a log–log plot of a standard curve generated with the TE-3 reference serum. We calculated the percentage of survey participants seroprotected (≥ 0.01 IU/mL) and by antibody-level category (< 0.01, 0.01–0.099, ≥ 0.1–0.9 ≥ 1.0 IU/mL).[13] Absolute differences in seroprotection were calculated by sex and across age groups with 95% Wald asymptotic confidence limits; median antibody concentrations were also calculated for subpopulations.

RESULTS

Tetanus seroprotection among children 1–4 years, 5–14 years, and persons ≥ 15 years of age was 90%, 66%, and 91% in Mbita District, Kenya, respectively; 89%, 66%, and 73% in Kongwa District, Tanzania; and 88%, 91%, and 79% in Morrupula District, Mozambique. Seroprotection was significantly lower among children 5–14 years versus 1–4 years of age by 24% (95% confidence interval [CI]: 13–35%) in Kenya, and 23% (95% CI: 16–29%) in Tanzania; there was no significant difference (95% CI: –8 to 3%) in Mozambique (Table 2).

Among persons ≥ 15 years of age, seroprotection among females and males was 96% and 45% in Kenya, 94% and 28% in Tanzania, and 90% and 64% in Mozambique. Seroprotection was lower among males ≥ 15 years of age than females by 51% (95% CI: 29–73%) in Kenya, 67% (95% CI: 59–75%) in Tanzania, and 26% (95% CI: 20–33%) in Mozambique. In all three countries, there was no significant difference in seroprotection by sex for children 5–14 years and 1–4 years of age (Table 2).

Median antibody levels among children 1–4 years and 5–14 years of age were 0.139 IU/mL and 0.028 IU/mL in Kenya, 0.113 IU/mL and 0.019 IU/mL in Tanzania, and 0.244 IU/mL and 0.793 IU/mL in Mozambique. Among persons ≥ 15 years of age, median antibody levels for females and males were 0.950 IU/mL and 0.007 IU/mL in Kenya, 0.966 IU/mL and 0.002 IU/mL in Tanzania, and 0.844 IU/mL and 0.047 IU/mL in Mozambique (Table 2). Antibody levels by age group, 1–4 years, 5–14 years, and ≥ 15 years (males and females), are shown in Table 3.

DISCUSSION

In summary, we observed evidence of tetanus immunity gaps among men ≥ 15 years of age in community surveys in Kenya, Tanzania, and Mozambique, and among children 5–14 years of age in Kenya and Tanzania. Among these groups, immunity gaps were characterized by low tetanus seroprotection (< 70%) and low antibody concentrations (median ≤ 0.05 IU/mL), which generally correlate with the robustness and duration of protection.[3] Immunity differences in men and women ≥ 15 years of age exist because WRA receive routine TTCV booster doses and men do not, and immunity gaps in older children occur from waning immunity from infant doses.[3] We observed no immunity gap among children 5–14 years of age in Mozambique, who receive two TTCV booster doses through school in first and second grades.[5]

Observed sex differences in tetanus seroprotection among persons ≥ 15 years of age in all three ESA countries (range: 26–67%) were similar to the results of historic serosurveys. Adult males had lower tetanus seroprotection than females in a 1990–1991 serosurvey in rural western Kenya (19% versus 69%).[16] In Dar-es-Salaam, Tanzania, 64% seroprotection was observed in a 1999 serosurvey of adult males, whereas 99% seroprotection was observed in a 1997 serosurvey of adult females.[17,18] Reviews of hospital records from tetanus cases among adolescents and adults support serologic results. The male-to-female ratio of tetanus patients was 3:1 in Maputo, Mozambique, during 1987–1992; 12:1 in Bugando, Tanzania, during 2001–2010; and 10:1 in Dar-es-Salaam, Tanzania, during 2004.[19–21] In our survey, the smaller difference in tetanus seroprotection (26%) between females and males ≥ 15 years of age in Mozambique is likely explained by provision of two childhood TTCV booster doses. Providing the third WHO-recommended booster in adolescence may be needed to eliminate the sex difference in tetanus immunity among persons ≥ 15 years of age in Mozambique.[3]

Differences observed in tetanus seroprotection among children 5–14 years of age compared with 1–4 years of age in Kenya (66% versus 90%) and Tanzania (66% versus 89%) were similar to results from a 1997 serosurvey in Tanzania (54% versus 96%).[22] The low seroprotection we observed among Kenyan children aged 5–14 years of age (66%) was similar to results of 1998 serosurvey in eastern Kenya (63% for 5–14 years of age) and a 1990–1991 serosurvey in western Kenya (58% for 8–11 years of age).[16,23] These results indicate chronic tetanus immunity gaps in older children from stagnating DTP3 coverage and waning immunity of infant doses.[3,12]

Seroprotection of children 1–4 years of age was similar across the three countries (range: 88–90%). National DTP3 coverage estimated by WHO–UNICEF for annual birth cohorts corresponding to this age group were slightly lower

TABLE 2
Tetanus serologic protection by age and sex among participants in community surveys: Kenya, Tanzania, and Mozambique

	Total			Females			Males			Difference		
Country	Total	Seroprotected*		Total	Seroprotected*		Total	Seroprotected*		(Females – males)		
Age (years)	No.	No.	%	No.	No.	%	No.	No.	%	%	LCL	UCL
Kenya												
1–4	110	99	90	47	44	94	63	55	87	6	–4	17
5–14	95	63	66	40	28	70	55	35	64	6	–13	25
≥ 15	192	174	91	172	165	96	20	9	45	51	29	73
Tanzania												
1–4	115	102	89	56	49	88	59	53	90	–2	–14	9
5–14	1,018	671	66	540	358	66	478	313	65	1	–5	7
≥ 15	452	330	73	307	290	94	145	40	28	67	59	75
Mozambique												
1–4	178	157	88	75	65	87	103	92	89	–3	–12	7
5–14	427	388	91	232	207	89	195	181	93	–4	–9	2
≥ 15	626	493	79	350	316	90	276	177	64	26	20	33

IU/mL = international units per milliliter; LCL = lower confidence limit; UCL = upper confidence limit.
*Seroprotection was defined as ≥ 0.01 IU/mL tetanus toxoid IgG.

TABLE 3
Tetanus antibody concentration by age and sex among participants in community surveys: Kenya, Tanzania, and Mozambique

Country		Percentage of participants by tetanus toxoid IgG category (IU/mL)			
Age (years) and sex	Median tetanus toxoid IgG concentration (IU/mL)	< 0.01	0.01–0.09	0.1–0.99	≥ 1.0
Kenya					
1–4	0.139	10	33	51	6
5–14	0.028	34	35	26	5
≥ 15 males	0.007	55	5	10	30
≥ 15 females	0.950	4	18	38	40
Tanzania					
1–4	0.113	11	35	46	8
5–14	0.019	34	53	11	2
≥ 15 males	0.002	72	16	9	3
≥ 15 females	0.966	6	14	32	48
Mozambique					
1–4	0.244	12	25	39	24
5–14	0.793	9	23	24	44
≥ 15 males	0.047	36	21	26	17
≥ 15 females	0.844	10	11	40	39

IgG = immunoglobulin G; IU/mL = international units per milliliter.

in Mozambique (range: 74–76%) than in Kenya (range: 85–88%) and Tanzania (range: 86–91%).[12] Variation in vaccination coverage at the district level might explain the relatively high seroprotection we observed among children 1–4 years of age in Mozambique despite lower national coverage.

These serologic data were collected as part of integrated disease serosurveillance and were useful in documenting tetanus immunity gaps in children 5–14 years and men ≥ 15 years of age in ESA countries during the VMMC-associated tetanus outbreak. Our results were considered during the WHO Technical Consultation on Tetanus and VMMC when formulating recommendations for tetanus risk mitigation.[11] Tetanus and other NTD control programs have the potential to benefit from multiplex testing in integrated serologic surveillance because of limited resources, attention, and disease-specific data.[13,24,25] Including tetanus IgG testing by MBA in national serosurveys can help with monitoring progress towards and maintenance of MNTE.[13]

WHO recommends integrating vaccinations given during health service contacts where possible, and opportunities to provide the WHO-recommended TTCV booster doses increasingly exist as countries introduce a second dose of measles-containing vaccine (MCV2) during the second year of life (2YL) and work to develop robust school-based programs for new vaccines that target preadolescents, such as human papillomavirus virus vaccine.[3,26,27] Kenya, Tanzania, and Mozambique have established national 2YL delivery platforms with the introduction of MCV2 within the past 3 years. MCV2 coverage by the end of the 2YL in Tanzania improved from 29% in 2014 to 57% in 2015, whereas MCV2 coverage in Kenya was 28% in 2015; data were unavailable for Mozambique where MCV2 was introduced in 2015.[4,12] All three countries have high net primary school enrollment (> 80%) and gender parity of enrollment (> 90%), making school-based platforms attractive for delivering vaccines at school entry.[27,28] Existing school-based immunization programs in Mozambique and other ESA countries demonstrate the general feasibility of this approach, but country-specific factors enabling success can vary.[27,29] Country programs may benefit from technical guidance on how best to introduce TTCV booster doses into the national immunization schedule. Though TTCV is relatively inexpensive (~$0.10 per dose for TT/Td, ~$0.20 for DTP, and ~$1.00 per dose for pentavalent), funding support for operational costs related to the introduction of new vaccination opportunities beyond the first year of life may be helpful in some lower income countries (e.g., grants through GAVI, the Vaccine Alliance).[30,31]

There are limitations of this study. First, the surveys represent convenience samples taken from one district in each country, and are not nationally representative. Second, the limited volume eluted from some DBS collected in Tanzania and Mozambique prevented MBA testing among all participants, especially children 1–4 years of age. Third, other data regarding tetanus immunity from the ESA subregion were limited; there were no nationally representative tetanus serosurveys, and only a small number of surveys with convenience samples were available for comparison. Historic administrative DTP3 coverage from the districts included in our surveys were not available, and we did not collect vaccination data from survey participants, preventing correlation. General limitations of using serology to monitor vaccination programs include failure to seroconvert after vaccination and waning immunity after seroconversion.

In conclusion, tetanus immunity gaps among children 5–14 years and men ≥ 15 years of age have occurred where the WHO-recommended TTCV booster doses are not provided for both sexes in childhood and adolescence.[3] Although our survey results are not generalizable to entire countries or the ESA region, similar immunity gaps may exist in any country without TTCV booster doses. Recent serosurvey results from Bamako, Mali, demonstrated waning tetanus immunity in older children and a sex disparity in adult immunity before the mass campaign with a serogroup A meningococcal TT-conjugate vaccine (MenAfriVac), and serosurvey results after the campaign highlighted the potential to increase tetanus population immunity through use of TT-conjugate vaccines.[32] Moving toward a life course of vaccination approach for tetanus would help countries in sustaining MNTE initially achieved through intensive TT campaigns targeting WRA. Inclusion of TTCV booster doses in national RI schedules, for example, during the 2YL and as part of school-based immunization programs where appropriate, would also help reduce tetanus disease burden in older children and adult males.

Received June 7, 2016. Accepted for publication October 21, 2016.

Published online December 5, 2016.

Acknowledgments: We dedicate this publication to the memory of Martha H Roper, who provided editorial input, and more importantly, devoted her public health career to preventing illness and deaths from tetanus. We thank families and communities for their participation in the surveys, field teams for collecting the samples, Delynn Moss for preparing MBA beads, and Amanda Barry, Brook Goodhew, Henry Kanyi, and Faith Mwende for running the MBA samples. We also thank Kathleen Wannemuehler and Howard Gary for input on presentation of results. This article is published with the permission of the Director of KEMRI.

Financial support: This work was supported by a grant from the Bill & Melinda Gates Foundation (OPP 1022543).

Disclaimer: Use of trade names is for identification only and does not imply endorsement by the Public Health Service or by the U.S. Department of Health and Human Services. The findings and conclusions in this report are those of the authors and do not necessarily represent the official position of the U.S. Centers for Disease Control and Prevention.

Authors' addresses: Heather M. Scobie and Minal Patel, Global Immunization Division, Centers for Disease Control and Prevention, Atlanta, GA, E-mails: vih8@cdc.gov and hgo9@cdc.gov. Diana Martin, Jeffrey W. Priest, Kimberly Y. Won, and Patrick J. Lammie, Division of Parasitic Diseases and Malaria, Centers for Disease Control and Prevention, Atlanta, GA, E-mails: hzx3@cdc.gov, jip8@cdc.gov, kfw7@cdc.gov, and pjl1@cdc.gov. Harran Mkocha, Kongwa Trachoma Project, Kongwa, Tanzania, E-mail: hmkocha@yahoo.com. Sammy M. Njenga, Eastern and Southern Africa Centre of International of Parasite Control, Kenya Medical Research Institute, Nairobi, Kenya, E-mail: snjenga@kemri.org. Maurice R. Odiere, Neglected Tropical Diseases Branch, Centre for Global Health Research, Kenya Medical Research Institute, Kisumu, Kenya, E-mail: modiere@kemricdc.org. Sonia Pelletreau, Children's Investment Fund Foundation, London, United Kingdom, E-mail: spelletreau@ciff.org. Ricardo Thompson, Instituto Nacional de Saúde, Maputo, MZ, E-mail: rthompsonmz@gmail.com.

REFERENCES

1. Roper MH, Vandelaer JH, Gasse FL, 2007. Maternal and neonatal tetanus. *Lancet 370:* 1947–1959.
2. Dalal S, Samuelson J, Reed J, Yakubu A, Ncube B, Baggaley R, 2016. Tetanus disease and deaths in men reveal need for vaccination. *Bull World Health Organ 94:* 613–621.
3. World Health Organization (WHO), 2006. Tetanus vaccine: WHO position paper. *Wkly Epidemiol Rec 20:* 198–208.
4. World Health Organization Department on Immunization Vaccines and Biologicals, Data, Statistics and Graphics, 2016. *Reported Immunization Schedules by Vaccine*. Available at: http://www.who.int/entity/immunization/monitoring_surveillance/data/schedule_data.xls?ua=1. Accessed August 31, 2016.
5. World Health Organization Department on Immunization Vaccines and Biologicals, Data, statistics and graphics, 2016. *Reported Data of Immunization Provided at School*. Available at: http://www.who.int/immunization/monitoring_surveillance/data/Immunization_at_school.xls?ua=1. Accessed August 31, 2016.
6. World Health Organization, 1996. Expanded programme on immunization (EPI). Immunization schedules in the WHO African region, 1995. *Wkly Epidemiol Rec 71:* 90–94.
7. Daly AD, Nxumalo MP, Biellik RJ, 2003. Missed opportunities for vaccination in health facilities in Swaziland. *S Afr Med J 93:* 606–610.
8. Zimbabas, Zimbabwe Expanded Programme on Immunisations, 2012. *New Vaccination Schedule Starting 20th June 2012*. Available at: http://www.zimbabas.co.zw/new-vaccination-schedule-starting-20th-june-2012/. Accessed September 16, 2016.
9. United Children's Fund (UNICEF), 2013. *Immunization Summary*. Available at: http://www.unicef.org/immunization/files/EN-ImmSumm-2013.pdf. Accessed September 16, 2016.
10. Grund JM, Toledo C, Davis SM, Ridzon R, Moturi E, Scobie H, Naouri B, Reed JB, Njeuhmeli E, Thomas AG, Benson FN, Sirengo MW, Muyenzi LN, Lija GJ, Rogers JH, Mwanasalli S, Odoyo-June E, Wamai N, Kabuye G, Zulu JE, Aceng JR, Bock N, 2016. Notes from the field: tetanus cases after voluntary medical male circumcision for HIV prevention—Eastern and Southern Africa, 2012–2015. *Morb Mortal Wkly Rep 65:* 36–37.
11. World Health Organization (WHO), 2015. *Meeting Report: WHO Informal Consultation on Tetanus and Voluntary Medical Male Circumcision, 9–10 March 2015*. Geneva, Switzerland. Available at: http://apps.who.int/iris/bitstream/10665/181812/1/9789241509237_eng.pdf?ua=1. Accessed August 20, 2015.
12. World Health Organization, 2015. *WHO Vaccine-Preventable Diseases Monitoring System (2015 Global Summary): WHO/UNICEF Estimates of National Immunization Coverage*. Available at: http://www.who.int/entity/immunization/monitoring_surveillance/data/coverage_estimates_series.xls?ua=1. Accessed August 7, 2015.
13. Scobie HM, Mao B, Buth S, Wannemuehler K, Sorenson C, Kannarath C, Jenks MH, Moss DM, Priest JW, Soeung SC, Quick L, Deming MS, Lammie PJ, Gregory CJ, 2016. Tetanus immunity among women aged 15 to 39 years in Cambodia: a national population-based survey, 2012. *Clin Vaccine Immunol 23:* 546–554.
14. Goodhew EB, Priest JW, Moss DM, Zhong G, Munoz B, Mkocha H, Martin DL, West SK, Gaydos C, Lammie PJ, 2012. CT694 and pgp3 as serological tools for monitoring trachoma programs. *PLoS Negl Trop Dis 6:* e1873.
15. Moss DM, Priest JW, Boyd A, Weinkopff T, Kucerova Z, Beach MJ, Lammie PJ, 2011. Multiplex bead assay for serum samples from children in Haiti enrolled in a drug study for the treatment of lymphatic filariasis. *Am J Trop Med Hyg 85:* 229–237.
16. Kurtzhals JA, Kjeldsen K, Hey AS, Okong'o-Odera EA, Heron I, 1997. Immunity to tetanus and diphtheria in rural Africa. *Am J Trop Med Hyg 56:* 576–579.
17. Aboud S, Lyamuya EF, Kristoffersen EK, Matre R, 2002. Immunity to tetanus in male adults in Dar es Salaam, Tanzania. *East Afr Med J 79:* 73–76.
18. Aboud S, Matre R, Lyamuya EF, Kristoffersen EK, 2001. Antibodies to tetanus toxoid in women of childbearing age in Dar es Salaam and Bagamoyo, Tanzania. *Trop Med Int Health 6:* 119–125.
19. Chalya PL, Mabula JB, Dass RM, Mbelenge N, Mshana SE, Gilyoma JM, 2011. Ten-year experiences with tetanus at a tertiary hospital in northwestern Tanzania: a retrospective review of 102 cases. *World J Emerg Surg 6:* 20.
20. Mchembe MD, Mwafongo V, 2005. Tetanus and its treatment outcome in Dar es Salaam: need for male vaccination. *East Afr J Public Health 2:* 22–23.
21. Vaz RG, Neves O, 1993. Tétano no Departamento de Medicina do Hospital Central de Maputo: revisao de 119 casos. *Rev Med Moamb 4:* 23–26.
22. Aboud S, Matre R, Lyamuya EF, Kristoffersen EK, 2000. Levels and avidity of antibodies to tetanus toxoid in children aged 1–15 years in Dar es Salaam and Bagamoyo, Tanzania. *Ann Trop Paediatr 20:* 313–322.
23. Siekmann JH, Allen LH, Watnik MR, Nestel P, Neumann CG, Shoenfeld Y, Peter JB, Patnik M, Ansari AA, Coppel RL, Gershwin ME, 2003. Titers of antibody to common pathogens: relation to food-based interventions in rural Kenyan schoolchildren. *Am J Clin Nutr 77:* 242–249.
24. Lammie PJ, Moss DM, Brook Goodhew E, Hamlin K, Krolewiecki A, West SK, Priest JW, 2012. Development of a new platform for neglected tropical disease surveillance. *Int J Parasitol 42:* 797–800.
25. Priest JW, Jenks MH, Moss DM, Mao B, Buth S, Wannemuehler K, Soeung SC, Lucchi NW, Udhayakumar V, Gregory CJ, Huy R, Muth S, Lammie PJ, 2016. Integration of multiplex bead assays for parasitic diseases into a national, population-based serosurvey of women 15–39 years of age in Cambodia. *PLoS Negl Trop Dis 10:* e0004699.
26. Subaiya S, Dumolard L, Lydon P, Gacic-Dobo M, Eggers R, Conklin L, 2015. Global routine vaccination coverage, 2014. *Morb Mortal Wkly Rep 64:* 1252–1255.

27. Vandelaer J, Olaniran M, 2015. Using a school-based approach to deliver immunization: global update. *Vaccine 33:* 719–725.
28. UNESCO Institute for Statistics, Data centre, 2016. *Education: Gross Enrolment Ratio by Level of Education*. Available at: http://data.uis.unesco.org/index.aspx?queryid=142. Accessed August 31, 2016.
29. World Health Organization (WHO), 2015. *School-Based Immunization*. Available at: http://www.who.int/immunization/programmes_systems/policies_strategies/school_based_immunization/en/. Accessed September 16, 2016.
30. United Nations Children's Fund (UNICEF), 2016. *Supplies and Logistics: Vaccine Price Data, Tetanus Toxoid (TT)*. Available at: http://www.unicef.org/supply/files/TT.pdf. Accessed September 16, 2016.
31. Gavi, the Vaccine Alliance, 2016. *Vaccine Introduction Grants and Operational Support for Campaigns Policy*. Available at: http://www.gavi.org/about/governance/programme-policies/vaccine-introduction-grants-and-operational-support-for-campaigns/. Accessed October 4, 2016.
32. Basta NE, Borrow R, Berthe A, Onwuchekwa U, Dembele AT, Almond R, Frankland S, Patel S, Wood D, Nascimento M, Manigart O, Trotter CL, Greenwood B, Sow SO, 2015. Higher tetanus toxoid immunity 2 years after PsA-TT introduction in Mali. *Clin Infect Dis 61 (Suppl 5):* S578–S585.