



**International Journal of TROPICAL DISEASE  
& Health**

29(4): 1-6, 2018; Article no.IJTDH.39279  
ISSN: 2278-1005, NLM ID: 101632866

---

## **Assessment of Pneumococcal Conjugate Vaccination on Nasopharyngeal Pneumococcal Carriage: A Case Study of Vaccinated and Unvaccinated Children Under-Five Years Attending Mbale Municipality Health Centers, Eastern Uganda**

**Bournventure Onyango<sup>1,2</sup>, Kabaka Angurini<sup>1,3</sup>  
and Ivan Mugisha Taremwa<sup>1\*</sup>**

<sup>1</sup>Clarke International University, Kampala, Uganda.

<sup>2</sup>Cure International Hospital, Mbale, Uganda.

<sup>3</sup>Arua Regional Referral Hospital, Arua, Uganda.

### **Authors' contributions**

*This work was carried out in collaboration between all authors. Authors BO, KA and IMT designed the study, performed the statistical analysis, wrote the protocol and first draft of the manuscript. Author IMT managed the analyses of the study. Author BO, KA and IMT managed the literature searches. All authors read and approved the final manuscript.*

### **Article Information**

DOI: 10.9734/IJTDH/2018/39279

#### Editor(s):

(1) Giuseppe Murdaca, Clinical Immunology Unit, Department of Internal Medicine, University of Genoa, Italy.

#### Reviewers:

(1) Grienggrai Rajchakit, Maejo University, Thailand.

(2) Ketan Vagholkar, D.Y.Patil University School of Medicine, India.

(3) Dan Dano Ibrahim, Niger.

Complete Peer review History: <http://www.sciencedomain.org/review-history/23757>

**Original Research Article**

**Received 4<sup>th</sup> November 2017**

**Accepted 29<sup>th</sup> January 2018**

**Published 21<sup>st</sup> March 2018**

---

### **ABSTRACT**

**Aims:** To determine the nasopharyngeal carriage of *Streptococcus pneumoniae* among children under-5 years of age who have been vaccinated with Pneumococcal Conjugate Vaccine (PCV-10) and those unvaccinated in Mbale Municipality, Eastern Uganda.

**Study Design:** This was a cross sectional study.

**Place and Duration of Study:** The study was conducted in Mbale Municipality Health Centres, Eastern Uganda during the period of September 2015 and June, 2016.

---

\*Corresponding author: Email: [imugisha@ymail.com](mailto:imugisha@ymail.com);

**Methodology:** We enrolled 280 children (140 vaccinated with PCV 10 and 140 unvaccinated). Demographic data were obtained from the care givers, and vaccination status from the caregiver and the vaccination card. Nasopharyngeal swabs were tested for *Streptococcus pneumoniae* at least four weeks post immunization. Pneumococcal carriage was defined by a positive culture, with an identification of *Streptococcus pneumoniae*.

**Results:** The overall prevalence of *Streptococcus pneumoniae* carriage was 123 of 280 (43.93%; 95% Confidence interval: 39.88-47.69) with the rate among the immunized of 14.29% (95% CI: 11.33-16.73), compared to the 78.57% (95%CI: 81.34-88.61) among the unvaccinated children. The odds ratio of pneumococcal carriage was 7.4 among the unvaccinated children compared to the PCV immunized children.

**Conclusion:** Vaccination with PCV offers protection to the risk of pneumococcal carriage. This study emphasizes the value of monitoring for the impact of PCV vaccination program in order to reassure health care workers and families.

**Keywords:** Children under-five years; nasopharyngeal carriage; Pneumococcal Conjugate Vaccine; *Streptococcus pneumoniae*.

## ACRONYMS AND ABBREVIATIONS

PCV: Pneumococcal Conjugate Vaccine; PPSV: Pneumococcal Polysaccharide Vaccine; CDC: Centers for Disease Control and Prevention; WHO: World Health Organization; MOH: Ministry of Health; B.A: Blood agar; BHI: Brain Heart Infusion; CHO: Chocolate

## 1. INTRODUCTION

Worldwide, pneumonia accounts for 16% of the under-five mortalities [1]. Pneumonia is an independent predictor of childhood mortality and kills more children under-five years than human immune deficiency virus (HIV), malaria, and measles combined [1-3]. The major causative agents of pneumonia are *Streptococcus pneumoniae*, *Haemophilus influenzae type b* (Hib), and respiratory syncytial virus (RSV) [4-7].

*Streptococcus pneumoniae* accounts for a third of all childhood pneumonia-associated deaths [8]. The microbe can be found in the child's nose or throat; from where it is then aspirated into the respiratory secretions to colonize the nasopharynx and ultimately cause blood-borne disease [6,9].

Vaccination against *S. pneumoniae* has been shown to be very effective in decreasing hospital cases of pneumonia in high income countries as well as low income countries [10-13]. Given this impact, Uganda added the 10-valent Pneumococcal Conjugate Vaccine (PCV-10) administered at 4 weeks, 10 weeks and 14 weeks into the National Immunization Program in 2014 [2]. However, the major drop in local hospitalization for pneumonia has not yet been

seen and the burden of pneumonia among children under-five remains high. Research conducted in Eastern Uganda revealed an increase of documented pneumococcal infections from 56% in 2011 to 70% of hospitalized children under five years of age in 2016 [14,15]. As the roll out of PCV-10 intensifies, there is a need to demonstrate locally that it is having an impact. Studies in other settings have shown that pneumococcal vaccine can decrease nasopharyngeal carriage [10-13, 16]. Therefore, we examined pneumococcal carriage burden among children vaccinated with PCV-10 and those unvaccinated in Mbale Municipality Health Centres, Eastern Uganda.

## 2. MATERIALS AND METHODS

### 2.1 Study Design, Site and Population

This was a cross sectional laboratory based study, involving bacteriologic examination of nasopharyngeal swab samples among children under 60 months of age attending Namakwekwe, Namatala, Maluku, Busamaga and Wanale health centers in 2016-2017. The study enrolled children who presented to the health facility with signs and symptoms of pneumonia such as fever, wheezing, fast breath and negative malaria rapid diagnostic test (RDT). We excluded children who had malaria, cough or runny nose without fever. Our population comprised children under-five years, whose care givers consented to the study. As vaccination with PCV had already been incorporated into the Uganda National Expanded Program on Immunization (UNEPI), we considered children to be in the vaccinated group if at least 4 weeks or more has passed since the third PCV10 dose.

## 2.2 Sample Size Estimation and Sampling

The sample size was determined based on the formula for the prevalence surveys, with finite population correction,  $n = Z\alpha^2 (p) (1-p)/e^2$  [17]. Using  $Z\alpha^2$  as the normal standard distribution at 95% confidence interval,  $p$  the estimated prevalence of pneumococci among children under five years of age reported at 66% [14], and  $e$  as the margin of error estimated at 10% for the prevalence surveys; a sample size of 140 was considered for the unvaccinated group and unequal number for the vaccinated. Children were purposively enrolled into the study, through the week days of Monday to Friday.

## 2.3 Sample

The swabs were taken by trained technicians using a pre-packed sterile calcium alginate flexible swab (*Copan, Brescia, Italy*). Swabbing was done at the back of the nasopharynx as guided by World Health Organization (2013). Following collection, swabs were placed in a millilitre of skim-milk tryptone glucose glycerol (STGG) transport media, and transported to the Microbiology laboratory at Mbale Regional Referral Hospital at  $-20^{\circ}\text{C}$ . All

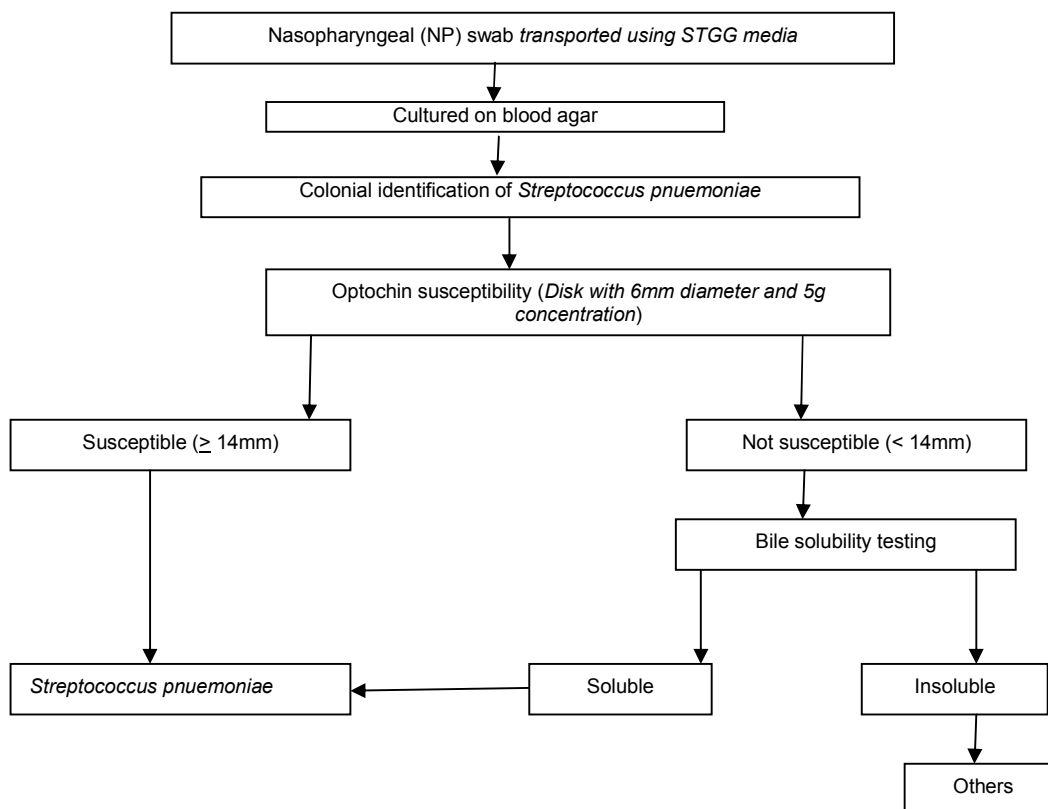
swabs were processed within 8 hours of collection for primary culture, isolation and identification.

## 2.4 Sample Analysis

Nasopharyngeal swabs were processed as described by Rutebemberwa et al. [15]. Pneumococci were identified using a zone of inhibition around the optochin disk, and bacteriologic characteristics which included: alpha hemolysis, flat and irregularly shaped colonies. In instances where two or more morphologically different colonies were identified, these were separately isolated.

## 2.5 Identification of *S. pneumoniae*

*S. pneumoniae* was identified as Gram-positive cocci, alpha hemolytic colonies on the plates. Further identification was done as described by Rutebemberwa et al [15]. The overall flow chart explaining identification algorithm for *S. pneumoniae* isolates was adopted from the standard operating procedure of Mbale Regional Referral Hospital as given below;



## 2.6 Data Analysis

Data was analyzed using SPSS version 18.0, and presented as a bar graph and a table. Pearson's square and p-value was used to interpret the statistical association. A p-value less than 0.05 was considered statistically significant.

## 2.7 Quality Control Measures

Known strain of *Streptococcus pneumoniae* (ATCC 49619) was used. In addition, quality control of the culture media was ensured by performing sterility and fertility testing. Sample analysis was conducted in strict adherence to the Standard operating procedures (SOPs).

## 3. RESULTS

### 3.1 Characteristics of Study Participants

Of 364 approached, 280 caregivers consented to have their child enrolled. The main reason for declining participation was concern about the invasiveness of the nasopharyngeal swabbing procedure. These were equal numbers (n=140) of PCV vaccinated and unvaccinated children. The mean age was 23 months (range: 4-59 months). By vaccination status, the mean age of the vaccinated group was 14.1 months, while the unvaccinated was 39.7 months. There were 151 males and 129 females. Additionally, 81 (53.64%) of the male participants were vaccinated, while 59 (45.74%) were unvaccinated. There were 59 (45.74%) vaccinated female participants and 70 (54.26%) were unvaccinated as indicated in Fig. 1.

### 3.2 Isolation of *Streptococcus pneumoniae* among Study Participants

The overall prevalence of *Streptococcus pneumoniae* was 123 of the 280, 43.93% (95% Confidence interval: 39.88-47.69). *Streptococcus pneumoniae* was isolated in only 13 (9.29%) of the vaccinated participants, while it was isolated among 110(78.57%) children who had not been vaccinated with PCV as indicated in Table 1.

## 4. DISCUSSION

This study revealed a high nasopharyngeal carriage rate (78.57%) of *Streptococcus pneumoniae* in Mbale Municipality, Eastern Uganda amongst unvaccinated children under 60 months of age. In Contrast the rate amongst the immunized was less than 10%. These findings are in agreement with a systematic review of pneumococcal carriage in sub-Saharan Africa that showed carriage rates of 21-94% [16]. The overall prevalence in the unimmunized is similar to that noted among the Gambian communities (72%) [16]. The unimmunized children were on average older than the immunized, mostly likely reflecting when the PCV 10 program had been inaugurated.

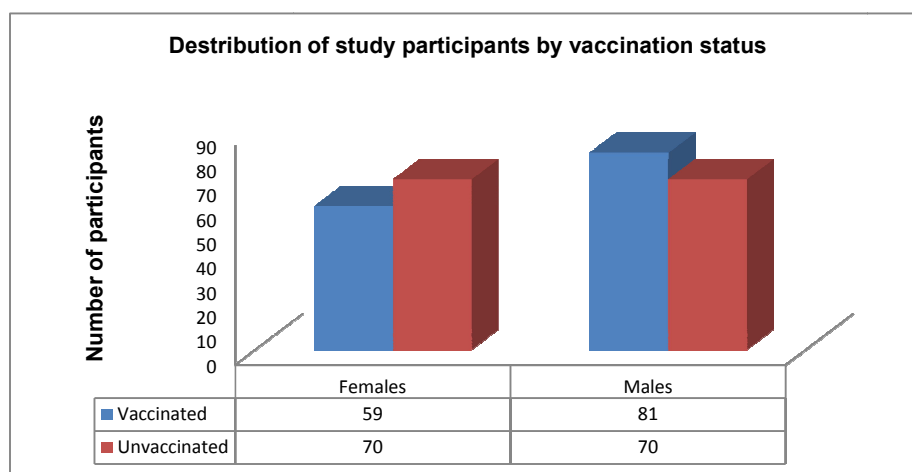
*Streptococcus pneumoniae* was isolated in less than 10% of the PCV vaccinated participants. This observation affirms the protective effect to the *Streptococcus pneumoniae* infection post immunization as has been shown in other studies [11,13,15,18,19]. Since the *Streptococcus pneumoniae* microbes were not serotyped we do not know if this represented carriage of organisms in PCV 10 or other serotypes. These findings provide local evidence of impact of PCV10 immunization which is reassuring for the immunization program and for families.

These data also emphasize the high burden of pneumococcal carriage is Mbale Municipality, and therefore risks more children under-five to pneumococcal infections. This observation is similar to what has been reported elsewhere in Uganda [8,15,19]. Until immunization rates become high, many children under five remain vulnerable. Management of pneumonia must be done with care keeping the possibility of pneumococcal infection in mind.

Based on our findings, we report that pneumococcal carriage remains a threat to children under-five in our community despite the introduction of universal access to PCV vaccination. Given the ease with which unimmunized children were recruited, it is clear

**Table 1. Showing *Streptococcus pneumoniae* isolated**

Immunization status	<i>Streptococcus pneumoniae</i>			
	No	Yes	Total	Odds ratio (P-value)
Immunized	127 (90.71)	13 (9.29)	140 (100)	7.4259 (0.000)
Not immunized	30 (21.43)	110 (78.57)	140 (100)	
Total	157 (56.07)	123 (43.93)	280 (100)	



**Fig. 1. Showing the vaccination status of study participants**

that there are still problems with access and/or caregiver acceptance of this vaccine and older children under 60 months are less likely to be immunized.

There are limitations to this study. We do not know if there is a seasonal variation in carriage. We do not know the overall coverage rate of PCV10 in this population. We also do not know the serotypes for either the immunized or the unimmunized children. Hence we cannot judge the full impact of PCV10 on carriage.

## 5. CONCLUSION

Based on our study findings, we report a high prevalence of pneumococcal carriage among unimmunized children under-five years in Mbale Municipality. PCV 10 immunization was associated with significantly less colonization. Not surprisingly, given when the PCV10 program commenced, the mean age of the immunized children was lower than for the unimmunized. PCV10 is having a demonstrable effect in this community even if the rate of pneumococcal hospitalization amongst young children has not yet dropped. This failure to see a decline is most likely due to inadequate population coverage levels given the ease with which unimmunized children were recruited. This lower rate of colonization among children who had been immunized with PCV10 should be reassuring for health care workers and families. Concerted efforts are needed to ensure wide availability and access to PCV10 for young children in this region in order to lessen childhood mortality risk due to pneumococcal infections.

## ETHICAL APPROVAL AND CONSENT

Ethical approval was obtained from research and ethics committee of Clarke International University. We ensured strict confidentiality and high ethical standards. We obtained written informed consent from each caregiver. All authors hereby declare that all experiments have been examined and approved by the appropriate ethics committee and have therefore been performed in accordance with the ethical standards laid down in the 1964 Declaration of Helsinki.

## ACKNOWLEDGEMENT

The authors wish to thank the children and caregivers who participated in the study. We are grateful to Micro Research ([www.Microresearch.ca](http://www.Microresearch.ca)) particularly Noni E MacDonald for the help with drafting and final editing of the manuscript.

## COMPETING INTERESTS

Authors have declared that no competing interests exist.

## REFERENCES

1. Wardlaw TM, Johansson EW, Hodge M. World Health Organization; UNICEF. Pneumonia: the forgotten killer of children. 2006;40. ISBN: 9789280640489/ 9280640488
2. The Global Burden of Disease. Mortality and Causes of Death Collaborators. Global, regional, and national life

- expectancy, all-cause mortality, and cause-specific mortality for 249 causes of death, 1980-2015: A systematic analysis for the Global Burden of Disease Study. *Lancet*. 2015;388:1459-1544.
3. Fischer CL, Rudan I, Liu L, Nair H, Theodoratou E, Bhutta ZA, et al. Global burden of childhood pneumonia and diarrhoea. *Lancet*. 2013;381:1405–16.
  4. Rudan I, O'Brien KL, Nair H, Liu L, Theodoratou E, et al. Epidemiology and etiology of childhood pneumonia in 2010: Estimates of incidence, severe morbidity, mortality, underlying risk factors and causative pathogens for 192 countries. *Journal of Global Health*. 2013;3.
  5. Kinney K, Johnson C, Papasian RT, Hall CG, Kurth MA, Jackson. Early onset Haemophilus influenzae sepsis in the newborn infant *Pediatr Infect Dis J*. 1993; 12(9):739-743.
  6. Madhi SA, Klugman KP Vaccine Trialist Group. A role for *Streptococcus pneumoniae* in virus-associated pneumonia. *Nat Med*. 2004;10:811–813.
  7. Ampofo K, Bender J, Sheng X, Korgenski K, Daly J, Pavia AT, Byington CL. Seasonal invasive pneumococcal disease in children: Role of preceding respiratory viral infection. *Pediatrics*. 2008;122:229–237.
  8. O'Brien KL, Wolfson LJ, Watt JP, Henkle E, Deloria-Knoll M, et al. Burden of disease caused by *Streptococcus pneumoniae* in children younger than 5 years: Global estimates. *The Lancet*. 2009;374:893–902.
  9. Stensballe LG, Hjuler T, Andersen A, Kaltoft M, Ravn H, Aaby P, Simoes EA. Hospitalization for respiratory syncytial virus infection and invasive pneumococcal disease in Danish children aged <2 years: A population-based cohort study. *Clin Infect Dis*. 2008;46:1165–1171.
  10. Lee LA, et al. The estimated mortality impact of vaccinations forecast to be administered during 2011-2020 in 73 countries supported by the GAVI alliance. *Vaccine*. 2013;18(31):035.
  11. Vucina VV, et al. Cost-effectiveness of pneumococcal conjugate vaccination in Croatia. *Vaccine*. 2015;7(33):043.
  12. Warren JL, et al. Impact of pneumococcal conjugate vaccines on pneumonia hospitalizations in high- and low-income subpopulations in Brazil. *Clinical Infectious Diseases*. 2017;65(11):1813-1818.
  13. Zar HJ, et al. Pneumonia in low and middle income countries: Progress and challenges. *Thorax*. 2013;68(11):1052-1056.
  14. Adegbola RA, DeAntonio R, Hill PC, Roca A, Usuf E, et al. Carriage of *Streptococcus pneumoniae* and Other Respiratory Bacterial Pathogens in Low and Lower-Middle Income Countries: A Systematic Review and Meta-Analysis. *PLoS ONE*. 2014;9.
  15. Rutebemberwa E, Mpeka B, Pariyo G, Peterson S, Mworozi E, et al. High prevalence of antibiotic resistance in nasopharyngeal bacterial isolates from healthy children in rural Uganda: A cross-sectional study. *Ups J Med Sci*. 2015;120: 249–256.
  16. Usuf E, Bottomley C, Adegbola RA, Hall A. Pneumococcal Carriage in Sub-Saharan Africa—A Systematic Review. *PLoS ONE*. 2014;9.
  17. Lwanga SK LS, WHO Geneva. Sample size determination in health studies; 1991.
  18. Simell B, Auranen K, Kayhty H, Goldblatt D, Dagan R, O'Brien KL. The fundamental link between pneumococcal carriage and disease. *Expert Rev Vaccines*. 2012; 11:841–55.
  19. Lindstrand A, Kalyango J, Alfvén T, Darenberg J, Kadobera D, Bwanga F, et al. Pneumococcal Carriage in Children under Five Years in Uganda-Will Present Pneumococcal Conjugate Vaccines Be Appropriate? *PLoS ONE*; 2016.

© 2018 Onyango et al.; This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<http://creativecommons.org/licenses/by/4.0>), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history:  
 The peer review history for this paper can be accessed here:  
<http://www.sciencedomain.org/review-history/23757>