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Risk Factors of Nasopharyngeal Pneumococcus Carriage Post PCV-10 Era among Children Attending Gertrude's Children Hospital, Nairobi

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Abstract

Pneumococcal infections kill at least one million children under the age of five every year, > 70% of these deaths occurs in low and middle-income countries. Nasopharyngeal colonization begins very soon after birth. While nasopharyngeal colonization may occur naturally even in healthy children, certain risk factors associated with the host and the host's environment have been demonstrated to have a nexus with quick transmission of the bacteria in a population; a phenomena which is yet to be well profiled among children living in Kenya. We investigated risk factors associated with carriage of *Streptococcus pneumoniae* among children ≤5 years of age attending Gertrude's Childrens Hospital. Guardians to children diagnosed clinically with pneumococcal disease were approached and requested to fill a consent form. Those who consented to the study were subsequently requested to fill a questionnaire on socio-demographic factors of the study subjects and proposed risk factors associated with nasopharyngeal carriage of *Streptococcus pneumoniae*. Carriage was determined on the basis of laboratory recovery of *Streptococcus pneumoniae* in the collected nasopharyngeal swabs. Logistic regression and odds ratios (OR) were done to establish associations between nasopharyngeal pneumococcus carriage and the proposed risk factors. The risk of nasopharyngeal *Streptococcus pneumoniae* carriage decreased insignificantly when the subject was female (Odds ratio [OR]: 0.766, 95% CI: 0.388, 1.511, p-value=0.442). Children between the age of 25-36 months (OR: 1.147 (95% I: 483, 2.722) and 37-48 months (OR: 1, 95% CI: 0.286, 3.501) had an insignificant elevated risk of nasopharyngeal *Streptococcus pneumoniae* carriage. Children whose mothers were non-cigarrate smokers exhibited low odds of carriage (OR: 0.764 (95% CI: 0.077, 7.537; p=0.818). Use of a stove for cooking, consumption of antibiotics two weeks prior to collection of a sample, daycare attendance and house-hold overcrowding insginificantly increased odds of nasopharyngeal *Streptococcus pneumoniae* carriage at 95% CI. Breast feeding a child ≥4 times a day, reduced exposure to smoke and child's PCV-10 immunization status were associated with reduced risk of nasopharyngeal *Streptococcus pneumoniae* carriage at 95% CI. Mothers and caregivers should ensure that children are: adequately breastfed not exposed to both active and passive smoke and fully immunized with PCV-10.

Keywords: *Streptococcus pneumoniae*, Nasopharyngeal Carriage, PCV-10, Children, Age, Smoke, Breast-feeding

Background Information

Adhesion of the pneumococcus to the epithelium of the human nasopharyngeal surface is a major phase in the pathogenesis of pneumococcal disease and a pivotal basis of person to person transmission (Alonso *et al*., 1995; Gillespie & Balakrishnan, 2000). Interventions targeting reduction of nasopharyngeal carriage of the pneumococcus would therefore play a crucial role in extenuating the burden of pneumococcal disease in both children and adult cohorts (Obaro & Adegbola, 2002).

The World Health Organization (WHO) approximations show that Pneumococcus causes close to 5 million deceases among children ≤5 years of age every year; the greatest proportion occurring in low and middle income countries (LMICs) (Ramirez, 2014). The prevalence of the pneumococcus which spreads through aerosols is premier among children ≤2 years of age and adults ≥65 years of age (Bogaert *et al*.,, 2004). Most studies have found carriage of between 30 and 62% among children less than 2 years of age across the globe (Ercibengoa *et al.*, 2011). In Kenya, pneumococcal disease kills ≥21000 children below the age of two years annually; this makes it the principal cause of child morbidity and mortality ahead of the highly dreaded malaria and diarrhea (Sallam *et al.*, 2019).

While the pneumococcus is a very belligerent organism capable of thriving by use of its own virulence entitlements, it also to a very large extent relies on other factors to expedite its dissemination in the community (Assefa *et al.*, 2013). Studies have classified these factors as: host and environmental associated (Mehr & Wood, 2012; Jeffrey *et al.*, 2018).

The prevalence of carriage of the *Streptococcus pneumoniae* has been demonstrated to vary largely between 19-86% on the basis of a range of factors like: the subjects' age, immune status and ecological location (Weiser *et al.,* 2018). According to Regev *et al.* (2004) and Simell *et al.* (2012), the pneumococcus can colonize the nasopharyngeal epithelium for a couple of weeks or even months before becoming invasive; the immunological status, recent viral infections, acute respiratory tract infection and age of the host having been described to play a fundamental role in the development of disease from carriage to invasive (Adegbola *et al.,* 2014). Further, Berkley *et al.* (2005) and Abdullahi *et al.* (2008), both reported that children of African origin have a slightly elevated risk of suffering from the various forms of pneumococcal disease following nasopharyngeal colonization as compared to their American counterparts; they are also likely to contract the disease at a young age and mostly carry serotypes 1 and 5 in their NP reservoir.

While a range of risk factors have been extensively and precisely associated with the blossoming of pneumococcal infections in other communities especially in developed economies, the phenomena is yet to be well profiled in Kenya. The pneumococcal-conjugate vaccine was introduced in Kenya in 2011 and it is administered across the country. The vaccine is given to under 5 children at 6, 10 and 14 weeks of age along with other childhood vaccines (Hammitt *et al.*, 2014). Considering that prevention and control can only be effective after understanding the dissemination faculties of any infectious agents; the current paucity of data regarding the precise role of certain risk factors in the spread of the pneumococcus impedes the containment efforts in entirety.

We investigated the role of age, gender, daycare attendance, smoke, household size, recent use and/or use of antimicrobial agents, breastfeeding frequency and waste disposal methods in the spread of pneumococcal infections among children below 5 years of age and attending Gertrude's Childrens Hospital.

Methods

Study site

This study was conducted at the Gertrude's Children's Hospital, Muthaiga (GCH) from May 2017 to February 2018. GCH is the largest Pediatric Hospital in East and Central Africa. The hospital is based in Muthaiga, Nairobi with various outpatient clinics. The hospital has a bed capacity of 105. It currently offers both primary and tertiary care services with its clientele largely comprising of individuals residing in Nairobi and its environs and within the Eastern Africa region. The hospital is accredited by the Joint Commission International (JCI).

Study Design and Population

This was a descriptive cross sectional study (Aagaard & Hauer, 2003). The study population included both PCV-10 vaccinated and unvaccinated children between 6 months and 5 years of age attending GCH. Consent was sought from biological mothers or guardians to target study subjects before data collection. Children with known immunosuppressive or terminal conditions were excluded from the study.

Sampling Technique

A purposive convenient sampling technique was utilised to recruit subjects to the study. Children to be included were assessed against a range of factors; they include: age of between 6 months to 5 years; clinical diagnosis of pneumococcal disease by the resident physician; informed assent from a biological parent or legal guardian and; lack of history of any immunosuppressive condition like cancer and HIV/AIDS. On qualifying these requirements, children were recruited to the study on first come, first recruited basis.

Sample Size Determination

To determine the minimum sample size, the formula of Fisher (1998) was used with a prevalence rate of 16% (WHO & UNICEF, 2017).

$n = \frac{z^2 \hat{p}(1-\hat{p})}{m^2}$. Where:

N= Desired minimal sample size.
Z= Standard normal deviation = 1.96 (from the tailed normal table).
P= Prevalence rate
M= the desired degree of accuracy @ 95% confidence level= 0.05
$N=1.96^2$ x 0.16 (1-0.16) / 0.05^2=206
Sample size (*n*) = **206**

Specimen collection and isolation

A standardised questionnaire containing questions on a range of selected risk factors was used to collect data. Information collected was on the following parameters: mothers age, mother's level of education, whether mother smokes, house size, waste disposal method, type of fuel used, mother's occupation, gender of the child, breast-feeding frequency, recent use/consumption of antibiotics, child attendance at day-care center, house hold sharing.

Documented vaccination history from the immunisation cards of children, was used to confirm birth weight and as prove of immunization with PCV-10.

Identification of *S. pneumoniae*

A nasopharyngeal sample was collected from each child using Copan flocked swabs and temporarily suspended in Amies medium for transportation to the main laboratory at GCH, within 3 hours of collection. Each swab was inoculated onto a selective gentamicin with 5% sheep blood agar (BA) plate. All swabs were plated within 24 hours of collection. The plates were incubated at 37°C in a 5% CO_2 atmosphere and examined at 16–24 hours and then again at 40–48 hours for growth of *S. pneumoniae*. Isolates were identified as *S. pneumoniae* by colony morphology (Mucoid, draughtsman appearance, α-haemolysis) and susceptibility to optochin (positive, ≥14 mm zone of inhibition; negative, <14 mm zone of inhibition). Plates with colonies akin to *S. pneumoniae* morphological features but with optochin clearance zones below 14 mm were further subjected to solubility in bile salts (positive, bile soluble; negative, bile insoluble).

Ethical Considerations

The study was approved by the Kenyatta University Ethics Review Committee (KU/ERC/APPROVAL/VOL 1(12) and permission obtained from the National Commission of Science Technology and Innovation (Ref No. Ref No. P/17/65428/15801). Gertrude's Childrens Hospital research committee issued permission for the study to be conducted at their clinics (GCH/ERB/VOLMMXVII/121). Informed & written parental or guardian ansent was sought before subjects were recruited to the study. Data from subjects was handled with high level confidence and only the study lead had access to the actual data.

Data Analysis

Bivariate and multi-variate logistic regression analyses were carried out to identify potential risk factors of pneumococcal nasopharyngeal carriage. Adjusted odds ratio with the corresponding 95% confidence intervals (CI) was used to measure the association between potential risk factors and nasopharyngeal carriage. *P-values* less than 0.05 were considered statistically significant.

Results

A total of 206 children were enrolled in the study. Of the 206, fifty one percent (51%, *n*=106) had received a full dose of PCV-10 while 49% (*n*=100) had not. *S. pneumoniae* was isolated from 11% (*n*=22) and 10% (n=20) of the PCV-10 vaccinated and unvaccinated cohorts respectively (Table: 1).

Socio-Demographic Factors of Study Subjects

Fourty seven percent (*n*=97) of the study children were males and 52.9% (*n*=109) were of female gender. 33% (*n*=68) of the children were within the age bracket of 6-12 months, 22.8% (*n*=47) were between the ages of 13-24 months, 22.3% (*n*=46) were between the ages of 25-36 months, 8.3% (*n*=17) were between the ages of 37-48 months and 13.6% (*n*=28) were between the ages of 49-60 months. Twenty three percent (*n*=47) of the biological mothers to the study children were between the ages of 18-24 years, 33.5% (n=69) were between 25 to 29 years, 26.2% (*n*=54) were between 30 to 34 years, 10.7% (*n*=22) were between 35 to 39 years and 6.8% (*n*=14) were between the age of 40 to 45 years. Most of the children sampled were between the age of 6-12 months (n=68, 33%).

One hundred and fifteen (*n*=115) families where study children came from had a monthly income of between 70-140USD, 28.6% (n=59) earned between 150-250USD, 7.3% (*n*=15) earned between 260-350USD, 2.9% (*n*=6) earned between 360-450USD and 4.9% (*n*=10) earned >450USD.

Further, about 43% (*n*=88) of the subjects' mothers & guardians had obtained secondary school education, 30% (*n*=61) had primary school education while 19% (*n*=38) and 9% (*n*=19) had obtained tertiary and university education. About 98% (*n*=202) of the subjects mothers/guardians were non-smokers as compared to 2% (*n*=4) who were smokers. About 96% (*n*=197) of the subject's households had no member who consumed alcohol while 4% (*n*=9) had. Fifty five percent (*n*=113) of the subjects lived in single rooms, 20% (*n*=42) lived in one bedroom houses, 22% (n=46) lived in two bedroom houses while 2% (*n*=5) lived three or more bed roomed houses. About 46% (*n*=94), 30% (*n*=62), 18% (*n*=38), 4% (*n*=9) and .5% (*n*=1) of the subjects cooked using gas, stove, charcoal, firewood and electricity respectively.

Seventy five percent (*n*=154) of the subjects households disposed their waste in public sewerage system, 23% (*n*=47) disposed their waste in private septic tanks and 2% (*n*=5) disposed waste using other undisclosed ways. About 90% of the subjects lived in households that used electricity as their source of light, 5% *(n*=11) used traditional lamps as their source of light, 2% (*n*=5) used candles for lighting, 1% (*n*=3) used solar while 4% (*n*=1) used lanterns. Table 4 reveals that 54% (*n*=112) of the subjects had consumed antibiotics within two weeks preceding visit to the clinic while 46% (*n*=94) had not.

Eighty one percent (*n*=167) of the subjects did not attend day-care centers as compared to 19% (*n*=39) who attended. About 30% (*n*=61) of the subjects' shared their households with 3 people other than parents/guardians, 25% (*n*=52) shared with two other people, 18% (*n*=37) shared with one other person, 16% (*n*=33) shared with four other people, 6% (*n*=12) shared with five other people and 5% (*n*=10) shared with >5 other people. Sixty five percent (n=133) of the subjects were exclusively breastfed, 35% (*n*=72) were moderately breastfed while 4% (n=1) had never been breastfed. About 51% (*n*=106) of the subjects had received the GAVI recommended full dose of PCV-10 while 49% (*n*=100) of the subjects had not (Table 1a,b & c).

Logistic Regression Analysis of the Risk Factors against Nasopharyngeal Carriage of *S. pneumoniae*

Table 2 below shows the relationship between occurrences of nasopharyngeal carriage of *Streptococcus pneumoniae* (dependent variable) with various selected risk factors (independent variables). The ORs demonstrate the nature of the relationship (OR=1: exposure does not affect odds of outcome, OR>1: exposure associated with higher odds of outcome, OR<1: exposure associated with lower rates of outcome) while P-Values demonstrate the degree of significance of the relationships at 95% CI. The odds of occurrence of nasopharyngeal carriage of *Streptococcus pneumoniae* when the subject was female decreased (OR: 0.766 (95% CI: 0.388, 1.511) although the decrease was insignificant (P-Value at 95% CI: 0.442). The odds of occurence of nasopharyngeal carriage of *Streptococcus pneumoniae* when the subject was between 25-36 Months and 37-48 Months increased insignificantly (OR: 1.147 (95% I: 483, 2.722) and (OR: 1 (95% CI: 0.286, 3.501) respectively.

The odds decreased when the subject was between 13-24 Months and 49-60 Months (OR: 0.667 (95% CI: 0.259, 1.715) and (OR: 0.413 (95% CI: 0.104, 1.634) respectively. However the

degrees of increase and decrease of odds of occurence of nasopharyngeal carriage of *Streptococcus pneumoniae* under the various predictors were insignificant (95% CI: 0.756, 1, 0.4 and 0.078) respectively. Although insignificant (p-Value at 95% CI: 0.818), the odds of occurence of nasopharyngeal carriage of *Streptococcus pneumoniae* when the mother to the subject was a non-smoker decreased (OR: 0.764 (95% CI: 0.077, 7.537) as compared to when she was a smoker.

Table 3 below shows that the odds of occurence of nasopharyngeal carriage of *Streptococcus pneumoniae* when a stove was used for cooking at the subject's home increased (OR: 1.079 (95% CI: 0.498, 2.339) but the odds decreased when the fuel type was charcoal (OR: 0.835 (95% CI: 0.321, 2.176). P-Values in both instances were insignificant (95% CI, p-value: 0.847 and 0.713). The odds of occurence of nasopharyngeal carriage of *Streptococcus pneumoniae* at 95% CI insignificantly (P-Value: 0.686) decreased (OR: 0.868 (95% CI: 0.438, 1.721)) when the subject had consumed antibiotics two weeks prior to visiting the health facility.

Odds of occuerence of nasopharyngeal carriage of *Streptococcus pneumoniae* when the subjects' attendance of daycare center was the the predictor variable insignificantly (95% CI, P-Value: 0.182) decreased (OR: 0.58 (0.261, 1.290) when the subject had not been attending. The odds insignificantly (95% CI, P-Value: 0.559) reduced (OR: 0.81 (0.40, 1.64) when the subject had been exclusively breastfed. The odds of occurence of nasopharyngeal carriage of *Streptococcus pneumoniae* insignificantly (95% CI, P-Value: 0.893) reduced (OR: 0.955 (0.484, 1.881) when the subject had not received 3 doses of PCV-10.

Overcrowding index (household members / household rooms) insignificantly (95% CI, P-Value: 0.352) reduced (OR: 0.70 (0.326, 1.491) odds of nasopharyngeal carriage of *Streptococcus pneumoniae* occurence when ≥3 people shared the same room as the subject (Table: 4)

Discussion

Nasopharyngeal carriage of *Streptococcus pneumoniae* is thought to be caused by a combination of exposure to risk factors related to the host, the environment and infections. We report that lack of PCV10 vaccination is not a significant risk factor for pneumococcal carriage.

This is the first study to report the risk factors associated with pneumococcal carriage/colonization in Nairobi, Kenya. Although other studies have reported that PCV10 vaccine induced immunological protection at a level sufficient to prevent invasive disease and reduced transmission of vaccine-serotype pneumococci within the population Regev *et al.* (2004), the herd immunity proposed may need further evaluation. While the various Pneumococcal Diseases are caused by invasion of the body with *S. pneumoniae*, RSV and *H. influenzae*, its fast spread across any given population has been reported to be dependent on several risk factors (Cardozo *et al.*, 2008). The risk factors include: Immunization status, age, gender, breastfeeding type, attendance at day-care center, recent consumption of antibiotics (two weeks prior to study), type of cooking fuel, whether mother smokes cigarettes or not and overcrowding index (number of household occupants against number of rooms in that household) (Kuo *et al.*, 2011).

The role of gender as a risk factor for pneumococcal carriage is yet to be understood, and no accord has been reached in literature. In this study, females (0R: 0.766, p=0.44) were less vulnerable to developing lower respiratory tract infections compared to males.

This difference could be explained by the fact that females have enhanced Th1 immune responses (Marks *et al.*, 2012). The influence of the female gender to nasopharyngeal

pneumococcal carriage was however not significant. It has also been shown that children who are between the ages of 25-36 months had increased but insignificant chances of pneumococcal nasopharyngeal carriage (OR: 1.147; p= 0.756). This could be because maternal antibodies have started waning from the system and the subjects are yet to gain sufficient self mucosal-immunity towards the pathogen (Koliou *et al.,* 2018).

These results are coherent with those of Coles *et al*. (2009), which reported that age, PCV10 vaccination, attendance at day care or school, previous respiratory infection and non-susceptibility to penicillin were associated with nasopharyngeal pneumococcal carriage due to certain serotypes. Exposure to cigarette smoke as a risk factor has also been demonstrated to augment the spread of the pneumococcus. There were reduced odds for pneumococcus carriage when the mother was a nonsmoker. However the reduced risk was not significant (OR: O.764, p=0.818). This result is in tandem with that of (Schaumburg *et al.,* 2013). The relevance of this association has been emphasized (Cheung *et al.,* 2009). Passive cigarette smoke exposure is an established risk factor for lower respiratory tract infections among children (Millar *et al.,* 2009).

Indoor air pollution has been described as an important risk factor for respiratory diseases including pneumococcus in children (Abdullahi *et al.,* 2008). In the current study, exposure to air pollution via use of different cooking fuels was associated with an increased risk for pneumococcus carriage.

The carriage of pneumococcus when a stove was used for cooking insignificantly increased (OR: 1.079, p=0.847). However, the odds decreased when the fuel type was charcoal (OR: 0.835; p=0.713). This could be attributed to the fact that a cooking stove emits more smoke containing carbon impurities that interfere with the capacity of mucosal immunity thereby predisposing it to pneumococcal infection (Assefa *et al.,* 2013). The odds of occurence of nasopharyngeal carriage

of *Streptococcus pneumoniae* at 95% CI insignificantly (p-value: 0.686) decreased (OR: 0.868 (95% CI: 0.438, 1.721)) when the subject had consumed antibiotics two weeks prior to visiting the health facility. This result is in tandem with that of Assefa *et al.* (2013) whose study found that receiving antibiotics reduced the risks of getting infected with *S. pneumoniae*.

Results demonstrate that attendance at day-care centre affect occurrence of Pneumococcus carriage. Those who did not attend day-care centres had decreased odds for pneumococcal infection, however, this result did not intimate the existence of a significant relationship (OR: 0.58, p=0.182). While studies done elsewhere have reported direct relationships between attendances at day-care centers to have direct relationships with occurrence of nasopharyngeal carriage of *Streptococcus pneumoniae*, the current study only reports a very minimal relationship.

This study reports that the odds for nasopharyngeal carriage of *Streptococcus pneumoniae* insignificantly (95% CI, p-value: 0.559) reduced (OR: 0.81) when the subject had been exclusively breastfed. This is in agreement with other studies in developing countries that have evaluated the consequence of breast feeding on predisposition to acute lower respiratory infections, pneumonia in particular. These studies show protective effect breast milk has on pneumonia, even though causality has not yet been shown (Weinberger *et al.*, 2010). This result could be correlated to the protective nature of maternal antibodies on infants.

To prevent pneumococcus, the WHO recommends immunization using pneumococcal conjugate vaccines (Satzke *et al.*, 2013). It is expected that subjects who had not received three doses of PCV-10 vaccination would be more vulnerable to occurrence of nasopharyngeal carriage of *Streptococcus pneumoniae*. However, this is not the case according to results from the current study as even subjects who had received the full dose were not protected. This could be

attributed to serotype replacement whereby vaccine serotypes are no longer in circulation while those in circulation are not included in the conjugate vaccine.

According to Lipsitch (2001), nasopharyngeal carriage of *Streptococcus pneumoniae* due to serotypes included in the vaccine can be replaced by nasopharyngeal carriage of *Streptococcus pneumoniae* due to serotypes not included in the vaccine under a phenomenon called serotype replacement. This happens mostly when conjugate vaccines are used (Simell *et al.,* 2012). Household crowding in the current study is defined as two or more persons sleeping in the same room as the child. This was what closely reflected the circumstances informed by the guardians or parents of the children comprised in the study. In the current study, it is concluded that living standards in homes with few rooms was likely to facilitate the transmission of this pathogen. However, this reduction in the transmission of *S. pneumoniae* was insignificant (p=0.352).

Conclusion

While associations between the assessed risk factors and nasopharyngeal carriage of the pneumococcus have been noted, the strength of that association needs to be well studied and profiled owing to the limited sample size in our study. We therefore recommend further studies to better understand the avidity between age, attendance at day-care, and smoke on nasopharyngeal carriage of the pneumococci.

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Competing interests

The authors declare no competing interests.

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TABLES

Table 1a: Socio-Economic Demographic Data of PCV-10 Vaccinated and Unvaccinated Children Attending GCH

Factor	Description	*n*=206	Valid Percent (%)
Child's age (months)	6-12	68	33.0
	13-24	47	22.8
	25-36	46	22.3
	37-48	17	8.3
	49-60	28	13.6
Child's gender	Male	97	47.1
	Female	109	52.9
Age of mother (years)	18-24	47	22.8
	25-29	69	33.5
	30-34	54	26.2
	35-39	22	10.7
	40-45	14	6.8
Family's income per month (USD)	70-140	115	56.1
	150-250	59	28.8
	260-350	15	7.3
	360-450	6	2.9
	>460	10	4.9
	Total	205	100.0
Missing	System	1	

n: Total number of subjects per category
%: Percentage of subjects per category
USD: United States Dollars

Table 1b: Socio-Economic Demographic Data of PCV-10 Vaccinated and Unvaccinated Children Attending GCH

Characteristic	Description	*n*=206	Percent (%)
Education level mother	Primary School	61	29.61
	Secondary School	88	42.72
	Tertiary College	38	18.45
	University	19	9.22
Mother's smoking status	Smoker	4	1.94
	Non smoker	202	98.06
Alcoholic in the house	Yes	9	4.37
	No	197	95.63
Size of the house of residence	Single room	113	54.85
	One bedroom	42	20.39
	Two bedroom	46	22.33
	Three bedrooms	5	2.43
Cooking fuel	Firewood	9	4.37
	Stove	62	30.1
	Charcoal	38	18.45
	Gas	94	45.63
	Electricity	3	1.46
Waste disposal method	Public sewerage system	154	74.76
	Private septic tank	47	22.82
	Other	5	2.43
Source of light	Candle	5	2.43
	Traditional lamp	11	5.34
	Lantern	1	0.49
	Solar	3	1.46
	Electricity	185	89.81
	Missing	1	0.49

n: Total number of subjects per category
%: Percentage of subjects per category

Table 1c: Socio-Economic Demographic Data of PCV-10 Vaccinated and Unvaccinated Children Attending GCH

Factor	Description	*n*=206	Percent (%)
Recent consumption of antibiotics (two weeks prior to study)	Yes	112	54.37
	No	94	45.63
Attendance at day care center	Yes	39	18.93
	No	167	81.07
No of household occupants	One	37	17.96
	Two	52	25.24
	Three	61	29.61
	Four	33	16.02
	Five	12	5.83
	≥five	10	4.85
Breast feeding type	none	1	0.49
	Moderate	72	34.95
	Exclusive	133	64.56
Child immunization	Yes	106	51.46
	No	100	48.54

n: Total number of subjects per category
%: Percentage of subjects per category

Table 2: Overall *Streptococcus pneumoniae* NP Carriage among PCV-10 Vaccinated and Unvaccinated Children Attending GCH

	All children		**PCV-10 Vaccinated children**		**PCV-10 Unvaccinated children**	
	n	**%**	***n***	**%**	***n***	**%**
Overall *Streptococcus pneumoniae* carriage	42	20.3 9	22	10.68	20	9.71
Proportion of *Streptococcus pneumoniae* Serotypes	*n*	%	*n*	%	*n*	%
PCV-10	0	0.00	0	0.00	0	0.00
Non PCV-10 serotypes	41	19.9 0	41	19.90	41	19.90
Non typeable	1	0.49	1	0.49	1	0.49

PCV-10: 10-valent pneumococcal conjugate vaccine
n: Total number of Pneumococci isolated
%: Percentage of Pneumococci isolates recovered

Table 3a: Logistic Regression Analysis of the Risk Factors Associated with Nasopharyngeal Carriage of *Streptococcus pneumoniae* among PCV-10 Vaccinated and Unvaccinated Children Attending GCH

Risk Factors	*Streptococcus pneumoniae* Colonization		Univariate Analysis	
	No	Yes		
	n (%)	*n* (%)	OR (95% CI)	P-Value
Gender				
Male	75 (77.32)	22 (22.68)	1	
Female	89 (81.65)	20 (18.35)	0.766(0.388, 1.511)	0.442
Age (months)				
6-12	52 (76.47)	16 (23.53)	1	
13-24	39 (82.98)	8 (17.02)	0.667(0.259, 1.715)	0.4
25-36	34 (73.91)	12 (26.09)	1.147(0.483, 2.722)	0.756
37-48	13 (76.47)	4 (23.53)	1(0.286, 3.501)	1
49-60	26 (92.86)	2 (7.14)	0.25(0.053, 1.170)	0.078
Mother's smoking status				
Smoker	3 (75)	1 (25)	1	
Non smoker	161 (79.7)	41 (20.3)	0.764(0.077, 7.537)	0.818

This was done at 95% CI. OR=1: Exposure does not affect odds of outcome. OR>1: Exposure associated with higher odds of outcome, OR<1: Exposure associated with lower rates of outcome

Table 3b: Logistic Regression Analysis of the Risk Factors Associated with Nasopharyngeal Carriage of *Streptococcus pneumoniae* among PCV-10 Vaccinated and Unvaccinated Children Attending GCH

Risk Factors	*Streptococcus pneumoniae* Colonization		Univariate Analysis	
	No	Yes		
	n (%)	*n* (%)	OR (95% CI)	p-value
Cooking method				
Gas	74 (78.72)	20 (47.62)	1	
Charcoal	31 (81.58)	7(16.67))	0.835(0.321, 2.176)	0.713
Stove	48 (77.42)	14 (22.58)	1.079(0.498, 2.339)	0.847
Electricity	3(7.14)	0(.00)		0.675
Firewood	8(19.05)	1(2.38)		0.546
Recent antibiotics use (two weeks)				
Yes	88 (78.57)	24 (21.43)	1	
No	76 (80.85)	18 (19.15)	0.868(0.438, 1.721)	0.686
Day care attendance				
Yes	28 (71.79)	11 (28.21)	1	
No	136 (81.44)	31 (18.56)	0.58(0.261, 1.290)	0.182
Breast feeding type				
Moderate	56 (77.78)	17 (22.22)	1	
Exclusive	108 (81.20)	25 (18.80)	0.81 (0.40, 1.64)	0.559
Child immunization				
Yes	84 (79.25)	22 (20.75)	1	
No	80 (80)	20 (20)	0.955(0.484, 1.881)	0.893
Overcrowding index				
2 or less	68 (77.27)	20 (22.73)	1	
3+	55 (83.33)	11 (16.67)	0.70(0.326, 1.491)	0.352

At 95% CI. OR=1: Exposure does not affect odds of outcome, OR>1: Exposure associated with higher odds of outcome, OR<1: Exposure associated with lower rates of outcome. Overcrowding index=household members / household rooms