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## Streptococcal throat carriage in a population of nursery and primary school pupils in Benin City, Nigeria

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**Abstract** Background: Group A streptococcus (GAS) is a major cause of mortality in man. Regular disease surveillance can be achieved through evaluation of throat carriage.

Objectives: To evaluate GAS throat carriage amongst nursery and primary schools pupils in Benin City, Nigeria.

Method: This cross sectional study was carried out between September and November 2011. A multistage random sampling method was used to recruit the pupils. Throat swabs were obtained for microscopy and Lancefied grouping.

Results: 426 pupils were recruited of which 363(85.2) were in

primary and 63(14.8%) in nursery schools. The subjects were aged 2 to 15 years and 203(47.7%) were males. Of the 426 pupils, 123 (28.9%) had positive throat swabs for  $\beta$  haemolytic streptococcus (βHS). None of the isolate was GAS. The isolates were groups C (50.4%), D (38.2%) and G (11.4%).

Conclusion: Although BHS throat carriage rate in this study is high, there was no GAS isolate. This may suggest low prevalence of GAS related diseases in the community.

**Keywords:** group A Streptococcus; throat carriage; school children

### Introduction

Group A Streptococcus (GAS) is the ninth major infectious cause of deaths in man globally. 1 It causes several invasive illnesses in children including meningitis, septicaemia and toxic shock.<sup>2</sup> It is one of the three leading causes of bacteraemia in children aged less than 90 days in developing countries<sup>2</sup> and accounts for 15-30% of cases of pharyngitis in children .3,4 Its chronic sequelae are glomerulonephritis, acute rheumatic fever (ARF), rheumatic heart disease (RHD) and throat carriage. The majority of deaths from GAS are caused by RHD, accounting for 233,000 deaths per year globally.<sup>5</sup> Rheumatic heart disease is the greatest contributor to acquired heart disease in Nigeria. <sup>6,7</sup> On the other hand, post streptococcal acute glomerulonephritis (PSAGN) is the third most common cause of renal disorders in Nigerian children.8

The enormous disease burden arising from GAS emphasizes the need for active surveillance of the organism. Such surveillance may be achieved through evaluating the prevalence of GAS throat carriage in the community. 9,10 The reservoir of acute GAS disease is the carrier state11,12 and this emphasizes its public health significance.

Previous studies on GAS throat carriage rate have documented results as high as 19.5% amongst asymptomatic children aged 5 to 15 years. 13 It has also been found more recently that children less than 5 years of age may have high throat carriage rates in Southern Israel, 14 contrary to previous documentation that GAS pharyngitis and chronic sequelae are uncommon in children of this age group. Some authors have reported throat carriage rates as high as 8.5% and 17.8% amongst infants and toddlers respectively.14

In Nigeria, there is a dearth of data on GAS throat carriage. Different authors have reported different throat carriage rates in previous studies ranging from 0% to 32.7%, 15-17 The variations in these results, as well as the fact that none of these studies evaluated throat carriage in preschool aged pupils highlights the need for a new study which will include preschool aged children. This is particularly important as invasive GAS diseases in this age group may contribute to high underfive morbidity and mortality rates. In an earlier study in 1999 on streptococcal throat carriage amongst primary school pupils in mid-western Nigeria, although a prevalence of 9.87% of BHS was obtained, none of the isolates was GAS. The absence of GAS may reflect a low GAS

activity in the study locale in which the prevalence of RHD is expected to be high.

The present study set out to evaluate the streptococcal throat carriage rate amongst primary school pupils in Egor local government area of Edo State, in mid-Western Nigeria. It further aimed to evaluate the prevalence of BHS throat carriage in children less than five years which is as yet undocumented in the country to the best knowledge of the researcher.

#### Methods

This cross sectional study was carried out in Egor local government area (LGA) of Edo State of Nigeria. It is one of the three LGAs that comprise Benin City, the capital of Edo State. Benin City is a cosmopolitan city located in the tropical rain forest belt at 122 metres above sea level. The LGA has ten (10) political wards.

Ethical clearance was obtained from the Ethics and Research Committee of the University of Benin Teaching Hospital. Approval for the study was obtained from the Edo State Universal Basic Education Board. Written informed consent was obtained from each of the parents/ guardians of all the pupils that participated in the study.

The sample size was determined using the formula<sup>18</sup>

The sample size was determined using the for 
$$n = Z^{2}_{(1-\alpha)/2} \frac{P(1-P)}{d^{2}}$$
 where n = minimum

sample size,  $Z^2(1-\alpha)/2$ = 1.96, P = 9.78% which was the prevalence of streptococcal throat carriage in the Benin study.<sup>17</sup> d = 5% or 0.05. A minimum sample size of 136 was obtained. For better representation of the population, the minimum sample size was multiplied by a factor of 3, thus a final sample size of 410 was used. The study was carried out between September and November 2011, when schools were in session.

The study subjects were nursery and primary school pupils in Egor LGA of Edo State. The pupils who used mouthwash on the day of examination and pupils who had antibiotics during the preceding two weeks of the study were excluded.

### Sampling method

A multistage sampling technique was used to select the schools in the Egor LGA. In the first stage, one nursery and one primary school were selected by simple random sampling technique from each of the ten political wards in Egor LGA. Twenty schools were thus selected comprising of 8 private nursery and 2 public nursery schools. This was because there were more private than public nursery schools. The selected primary schools comprised of four public and six private schools. The number of pupils to be selected from nursery and primary schools was the second stage. It was determined

by a ratio of the total number of pupils in the ten selected nursery (1,238) and primary (7,879) schools which was 1:6.36. Thus, 410 was distributed into 56 nursery and 354 primary school pupils. The number of pupils to be selected from each of the twenty schools was determined by proportion. The school sample size determined using formula the

$$N2 = \frac{a \times N1}{n}$$

Where:  $N_2$  = number of pupils recruited from a given nursery or primary school, a = population of index school, n = total number of pupils in all the selected nursery or primary schools (i.e. 1,238 for the nursery schools and 7,879 for the primary schools). = sample proportion calculated above (i.e.57 for the nursery schools and 363 for the primary schools). In each school, one arm of classes was selected randomly from which pupils were selected. The number of pupils obtained from the chosen arm in a given school was calculated using the list of pupils in each arm comprising of all the classes in the arm e.g nursery 2 had two classes. The calculated sample size of each school (N<sub>2</sub>) was distributed among the classes by proportion

$$N3 = \frac{a \times N2}{b}$$

using this formula: where  $N_3$  = Class sample size, a =Class population (all arms), b = Population of all classes in the school (either nursery or primary),  $N_2$  = School sample size (as above).

The first pupil was randomly selected from the list of pupils in that class by balloting. All subsequent pupils that were recruited were selected using a sampling interval. The list of pupils in the class arranged alphabetically was used as the sampling frame. The selected pupils were then given a consent form to take to their parents/guardians.

For each selected pupil, with the aid of a proforma, information on biodata and household number was obtained. The socio-economic class (SEC) was determined by Olusanya et al<sup>19</sup> method. The pupil's weight and height were measured using a Seca digital electronic and Seca portable stadiometer, using standard methods. The pupils had a complete physical examination in the school sick bay. Throat swab was obtained after depressing the tongue to expose the oropharynx with a cotton swab stick rubbed gently on the posterior pharyngeal wall. Thereafter, the swab stick was returned into its sterile container and transported in that manner to avoid contamination. The swabs were obtained between the hours of 11am and 1 pm. The younger children were sampled when the parents/caregivers were available at the close of school to enlist their cooperation.

### Processing of throat specimens

Within two hours of swabbing the throats, the swab sticks were taken to the laboratory for gram staining and culture.20 Growth with clear zones of haemolysis indicated beta haemolytic Streptococcus.

The Lancefield group was determined using PROLEX<sup>TM</sup> STREPTOCOCCAL GROUPING LATEX KIT, China, which identifies Groups A, B, C, D, F and G streptococci, according to the manufacturer's recommendations The cultures and Lancefield grouping were done in University of Benin Teaching Hospital, Benin City, Nigeria by a senior laboratory scientist.

### Follow-up of subjects

Pupils with positive throat swabs were followed up on a twice-weekly basis for six weeks, looking out for features of pharyngitis, PSAGN or ARF.

### Data analysis

Data were analysed using Statistical Package for Social Sciences (SPSS) 16.0 (Chicago IL). Student t test was used to compare the means of continuous variables. More than two means were compared using one way ANOVA. Fisher's exact test and Pearson's Chi-square statistical test of significance were used to determine the associations between throat carriage and the sociodemographic variables. The level of significance was set at p < 0.05.

# **Results**Characteristics of the study population

Four hundred and seventy eight pupils were recruited for the study, of which 52 were excluded because of antibiotics use within the preceding two weeks of the study. Hence, 426 pupils' data were analysed. There were 203 (47.7%) males and 223 (52.3%) females. The mean age of the subject was  $8.0\pm2.8$  (range; 2-15) years. The mean age of the female pupils  $8.1\pm2.8$  years was not significantly higher than that of the males  $8.0\pm2.7$  years, p=0.72. The mean age of the nursery pupils was  $3.4\pm0.7$  (range; 2-5) years while that of the primary school pupils was  $8.8\pm2.1$  (range; 5-15) years.

There were 363 pupils from the primary schools and 63 from the nursery schools. Of all the pupils, 216 (50.7) were in public schools and 147(34.5%) in the private schools. The SEC distribution of the subjects showed that 133 (33.7%), 158 (40.0%) and 104 (26.3%) pupils respectively were from high, middle and low SECs. The socio-demographic distribution by gender of the study population is shown in table 1. A total of 396 pupils provided data on their household sizes. The average (range) number of persons per household was 7 (range; 2-20). 220 (57.0%) of the pupils were from households with less than 7 persons while 146 (37.8%) and 20 (5.2%) were from households with 7-11 persons and greater than 11 persons respectively.

### Streptococcal throat carriage

Of the 426 pupils, 123 (28.9%) had positive throat swabs for  $\beta$ HS. Of the 123 pupils with positive isolates,

74 (33.2%) were female carriers while 49 (24.1%) were the male carriers, p=0.04. Table 2 illustrates the distribution of streptococcal throat carriage amongst the pupils in the study. Most of the carriers 61 (49.6%) were in the 9-12 years age group while the least 4 (3.3%) were between 13-15 years of age. Table 2

| <b>Table 1:</b> Socio-demographic characteristics of the study population |           |              |           |  |  |  |  |
|---|-----------|--------------|-----------|--|--|--|--|
| Characteristic  | Male      | Female Total |           |  |  |  |  |
|   | n(%)      | n(%)         | N(%)      |  |  |  |  |
| Age group (years)   |           |              |           |  |  |  |  |
| 2 - 4   | 28(6.6)   | 34(8.0)      | 62(14.6)  |  |  |  |  |
| 5 - 8   | 60(14.1)  | 67(15.7)     | 127(29.8) |  |  |  |  |
| 9 - 12  | 113(26.5) | 118(27.7)    | 231(54.2) |  |  |  |  |
| 13 - 15   | 2(0.5)    | 4(0.9)       | 6(1.4)    |  |  |  |  |
| Nursery $(n = 63)$  |           |              |           |  |  |  |  |
| Public  | 10(2.3)   | 11(2.6)      | 21(4.9)   |  |  |  |  |
| Private   | 19(4.5)   | 23(5.4)      | 42(9.9)   |  |  |  |  |
| Primary (n = 363)   |           |              |           |  |  |  |  |
| Public  | 105(24.6) | 111(26.1)    | 216(50.7) |  |  |  |  |
| Private   | 69(16.2)  | 78(18.3)     | 147(34.5) |  |  |  |  |
| Socio-economic status   |           |              |           |  |  |  |  |
| High  | 64(34.0)  | 69(33.3)     | 133(33.7) |  |  |  |  |
| Medium  | 79(42.0)  | 79(38.2)     | 158(40.0) |  |  |  |  |
| Low   | 45(23.9)  | 59(28.5)     | 104(26.3) |  |  |  |  |

| <b>Table 2:</b> Distribution of socio-demographic characteristics of carriers and non carriers |                   |                  |         |  |  |  |
|--|-------------------|------------------|---------|--|--|--|
| Characteristics  | carriers<br>Nn(%) | non-carrier n(%) | p value |  |  |  |
| Males  | 49(24.1)          | 154(75.9)        | 0.04    |  |  |  |
| Females  | 74(33.2)          | 149(66.8)        |         |  |  |  |
| Age group (years)  | )                 |                  |         |  |  |  |
| 2 - 4  | 18(14.6)          | 44(14.5)         |         |  |  |  |
| 5 - 8  | 40(32.5)          | 87(28.7)         |         |  |  |  |
| 9 - 12   | 61(49.6)          | 170(56.1)        | 0.15    |  |  |  |
| 13 - 15  | 4(3.3)            | 2(0.7)           |         |  |  |  |
| High SEC   | 36(31.6)          | 97(34.5)         |         |  |  |  |
| Middle SEC   | 46(40.4)          | 112(39.9)        | 0.82    |  |  |  |
| Low SEC  | 32(28.1)          | 72(25.6)         |         |  |  |  |
| Household size   |                   |                  |         |  |  |  |
| <7 persons   | 58(53.7)          | 162(58.3)        |         |  |  |  |
| 7 - 11   | 43(39.8)          | 103(37.1)        | 0.63    |  |  |  |
| >11  | 7(6.5)            | 13(4.6)          |         |  |  |  |

Of the 123 carriers, 46 (40.4%) were of the middle SEC while 32 (28.1%) and 36 (31.6) were of the low and high SEC respectively, p=0.82. The prevalence of throat carriage among the nursery pupils 19 (30.2%) was higher than in primary school pupils 104 (28.7%). The difference was not statistically significant, p=0.80. The carriage rate was highest 7 (35.0%) amongst pupils from households with greater than 11 persons and was least 58 (26.4%) amongst those from households with less than 7 persons, p=0.63. Seventy two (30.4%) of the 237 public school pupils and 51 (27%) of the 189 private school pupils were carriers, p=0.357.

### Lancefield group of the isolates

Of the 123  $\beta$  haemolytic Streptococci isolated in the present study, 62 (50.4%) were of group C, 47 (38.2%)

were of group D and 14 (11.4%) were of group G. There were no group A, group B nor group F isolates. The mean ages of children with Lancefield groups C, D and G were  $8.5 \pm 2.7 \text{years}$ ,  $7.6 \pm 2.9$  years and  $7.5 \pm 3.5$  years respectively and their mean ages did not differ significantly, p = 0.21.

Table 3: Gender and age group distribution of LancefieldgroupsLancefield groupC(%)D(%)G(%)p valueGenderMale28(58.3)18(37.5)2(4.2)0.19

|           | C(%)     | D(%)     | G(%)     | p value |
|-----------|----------|----------|----------|---------|
| Gender    |          |          |          |         |
| Male      | 28(58.3) | 18(37.5) | 2(4.2)   | 0.19    |
| Female    | 34(45.3) | 29(38.7) | 12(16.0) |         |
| Age group |          |          |          |         |
| 2 - 4     | 5(27.8)  | 9(50.0)  | 4(22.2)  |         |
| 5 - 8     | 20(50.0) | 16(40.0) | 4(10.0)  | 0.78    |
| 9 - 12    | 35(57.4) | 21(34.4) | 5(8.2)   |         |
| 13 – 15   | 29(93.6) | 1(3.2)   | 1(3.2)   |         |

Follow-up

None of the pupils in the study with streptococcal isolate developed symptoms of pharyngitis, ARF or PSGN during follow-up.

### Discussion

The present study noted a  $\beta$ HS carriage rate of 28.9%. A similarly high value of 32.7% was reported by Odigwe et al<sup>15</sup> in Calabar. The latter study evaluated only GAS strains implying that the overall  $\beta$ HS carriage rate would have been higher. High  $\beta$ HS carriage rates infer that the burden of disease such as skin infections and invasive illnesses attributable to  $\beta$ HS in the community under study may also be high.<sup>9,10</sup>

Some other Nigerian studies however reported lower  $\beta HS$  throat carriage rates. In 1999, Sadoh and co worker<sup>17</sup> reported a  $\beta HS$  carriage rate of 9.8% in Edo State while Ogunbi et al<sup>16</sup> reported a carriage rate of 13.3% in 1978 in Lagos State. Studies done in other countries reveal a wide range of  $\beta HS$  carriage rates including 8.3% in Croatia, <sup>21</sup> 19.5% in South India <sup>13</sup> and 28.5% in Lucknow. <sup>22</sup> These rates may reflect the disparity in disease burden in the different communities during the period of the research.

It is of note that the  $\beta$ HS carriage rate in the present study is higher than that of Sadoh and Omokhodion<sup>17</sup> carried out in the same LGA twelve years earlier. A plausible explanation is that seasonal factors may have influenced the results as neither study was carried out all through the year to accommodate seasonal variations in carriage rate and disease burden. It may also reflect a change in the epidemiology of  $\beta$ HS in the LGA over the years. The high  $\beta$ HS throat carriage rate of 29.0% amongst the nursery pupils in this study is considerable. Other workers have earlier reported high  $\beta$ HS throat carriage rates in preschool aged children. In 2002, Odigwe et al<sup>15</sup> reported a carriage rate of 45% in chil-

dren <5 years while Yagupsky et al<sup>14</sup> reported a carriage rate of 17.8% amongst toddlers in Southern Israel. In both of these studies, evaluation was only for GAS implying that the overall  $\beta$ HS would likely have been higher. The high streptococcal throat carriage rate in the preschool aged children in the present study may be due to their early exposure to  $\beta$ HS as a result of early patronage of day care facilities and nursery schools by the increasing number of working mothers.<sup>23</sup>

There was no GAS isolate in this study. This is similar to the earlier report by Sadoh and Omokhodion in 1999<sup>17</sup> where an absence of GAS was similarly noted in the same Egor L.G.A. This is in contrast to all previous studies which isolated GAS strains in India and Fiji Island. 13,24,25 The absence of GAS in the present study may stem from the preferential use of penicillin antibiotics for the management of acute pharyngitis in children in Benin City,<sup>26</sup> which is known to reduce the prevalence of GAS throat carriage in susceptible individuals.<sup>27</sup> The widespread antibiotic use may also account for the high group D found in this study compared to previous studies. 13,17,21,24,28 resulting in change in the microbiota of the throat, favouring the preferential growth of group D streptococcus over GAS. This is possible because group D streptococcus is mostly resistant to penicillin antibiotics.<sup>29</sup>

A previous study has demonstrated that low carriage rate in a community is associated with low prevalence of rheumatic fever and rheumatic heart disease in the same community. Another study done during an outbreak of invasive GAS disease in a community showed that the clone responsible for the outbreak was prevalent amongst throat carriers in the same community. The throat carriage rate of the GAS clone was significantly more amongst school children within the outbreak area than amongst those from other localities. These findings support the usefulness of GAS throat carriage as an index of GAS disease burden (both acute diseases and chronic sequelae), thus suggesting that the burden of GAS may be low in the community where the present study was carried out since throat carriage was absent.

A predominance of Lancefield group C and G *streptococci* as compared to GAS is consistent with previous studies in the tropics. <sup>16,17,22,24</sup> Groups C and G streptococci have been implicated in pharyngitis and skin infections and may cause invasive illnesses in man. <sup>28</sup>

### Conclusion

The prevalence of streptococcal throat carriage amongst nursery and primary school pupils in Egor LGA of Edo State was 28.9%. This high prevalence rate suggests that the burden of  $\beta$ HS diseases in Egor LGA may be high. However GAS was not isolated, which implies a likely low burden of group A Streptococcal diseases and sequelae in the study community. This is more so since a similar study conducted in the same locale over a decade

ago yielded no GAS isolate. The high streptococcal throat carriage rate amongst pupils less than 5 years of age suggests similarly high burden  $\beta$ HS activity in the age group perhaps because of early exposure.

Considering the enormous disease burden of invasive and RHD thought to be associated with the country where the study was done, this may mean a reduction in prevalence of GAS related diseases. This possible change in epidemiology needs further evaluation by conducting similar studies in other parts of the country to further describe the current epidemiology of GAS activity.

### **Authors' contribution**

ACO, WES,OO: Data collection, analysis, interpretation, Conceptualisation and design of the study ACO, WES: Writing of the initial draft ACO, WES, OO: Contribution of intellectual input to the draft, Approval of the manuscript

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