

Prevalence of Oral Potentially Pathogenic Microorganisms and Their Association with Oral Hygiene Practices and Socio-demographic Factors among Adults in Urban and Rural Communities: A Cross-sectional Study

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ABSTRACT

Background: The oral cavity harbours a diverse resident microbiota, and disruption of this balance permits overgrowth of potentially pathogenic microorganisms implicated in dental caries, periodontal disease, halitosis, and systemic complications. Oral hygiene practices and socio-demographic determinants substantially shape this microbial ecology, with urban–rural disparities in dental care access and health literacy further influencing the pattern of colonisation.

Objectives: To determine the prevalence of potentially pathogenic oral microorganisms among adults in urban and rural communities and to evaluate their association with oral hygiene practices and socio-demographic factors.

Materials and Methods: A community-based cross-sectional study was conducted among 100 adults (50 urban, 50 rural) selected by multistage stratified random sampling over a 12-month period. Oral swab specimens were cultured and organisms identified by standard microbiological techniques. Socio-demographic and oral hygiene data were collected using a structured, pretested questionnaire. Statistical analysis was performed using SPSS version 25, with chi-square testing and binary logistic regression.

Results: Potentially pathogenic microorganisms were isolated in 42.0% of participants, more frequently among rural (48.0%) than urban (36.0%) residents ($p = 0.04$). *Streptococcus mutans* (14.0%) was the most commonly isolated organism, followed by *Staphylococcus aureus* (10.0%) and *Candida albicans* (8.0%). Eighteen percent of participants were symptomatic, and 9.0% were laboratory-confirmed to harbour clinically significant colonisation. Irregular tooth-brushing (70.6% positive versus 26.3% among twice-daily brushers), tobacco use (61.5% versus 35.1%), and lower educational attainment (66.7% versus 33.3% among graduates) were significantly associated with pathogen positivity ($p < 0.05$ for each).

Conclusion: Potentially pathogenic oral microorganisms are prevalent among adults, particularly in rural communities with poorer oral hygiene practices. Strengthening oral health education and hygiene behaviour, especially among rural, tobacco-using, and less-educated populations, may substantially reduce oral pathogen burden.

Keywords: Oral microbiota; Potentially pathogenic microorganisms; Oral hygiene; Socio-demographic factors; Urban–rural; Cross-sectional study.

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INTRODUCTION

The oral cavity harbours one of the most diverse microbial ecosystems in the human body, comprising several hundred bacterial, fungal, and occasionally protozoal species that exist in dynamic equilibrium with the host. Under normal physiological conditions, this resident microbiota performs protective functions, including colonisation resistance against exogenous pathogens and modulation of local mucosal immunity. Disruption of this delicate balance—commonly termed oral dysbiosis—permits overgrowth of potentially pathogenic microorganisms capable of producing dental caries, periodontal disease, halitosis, mucosal infection, and, in susceptible individuals, systemic complications. [1–3]

Among the organisms most frequently implicated in oral disease are *Streptococcus mutans*, a principal aetiological agent of dental caries; *Staphylococcus aureus* and *Candida albicans*, opportunistic pathogens capable of colonising the oral mucosa and dorsal tongue; and enteric gram-negative organisms such as *Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Escherichia coli*, which are increasingly recovered from the oral cavity of individuals with poor oral hygiene, systemic illness, or hospitalisation. [3–6]

Dental caries and periodontal disease remain among the most prevalent non-communicable conditions worldwide, affecting an estimated 3.5 billion people according to the World Health Organization's Global Oral Health Status Report, with a disproportionate burden falling on populations in low- and middle-income countries where access to preventive dental care remains limited. [5–8]

Oral hygiene practices constitute the single most modifiable determinant of oral microbial ecology. Regular tooth-brushing, particularly with fluoride-containing toothpaste, mechanical plaque removal through flossing, and adjunctive use of antimicrobial mouth rinses have each been shown to reduce bacterial and fungal load and to lower the risk of colonisation by potentially pathogenic organisms. Conversely, irregular or inadequate oral hygiene, reliance on traditional non-fluoridated cleaning aids, and infrequent professional dental visits are consistently associated with higher rates of pathogen carriage. [7–10]

Socio-demographic characteristics exert a profound influence on oral hygiene behaviour and, consequently, on the composition of the oral microbiota. Age, gender, educational attainment, occupation, and household income have each been shown to shape oral health literacy, frequency of dental attendance, and adoption of preventive practices. Lower educational status and occupational groups with limited health awareness have repeatedly been identified as risk factors for

poor oral hygiene and higher pathogen positivity in community-based surveys. [9–12]

Place of residence—urban versus rural—adds a further important dimension to this relationship. Urban populations generally enjoy greater access to dental infrastructure, oral health education, and fluoridated dental products, whereas rural communities often contend with limited availability of dental services, lower health literacy, and continued reliance on traditional oral hygiene methods. These disparities are compounded by differences in tobacco and alcohol consumption patterns, dietary habits, and health-seeking behaviour between the two settings, all of which independently influence oral microbial colonisation. [10–13]

Beyond their strictly local effects, potentially pathogenic oral microorganisms have been increasingly implicated in systemic disease. Aspiration of oral pathogens has been linked to community-acquired and nosocomial pneumonia, while chronic periodontal infection has been associated with adverse cardiovascular outcomes, poor glycaemic control in diabetes mellitus, and adverse pregnancy outcomes. These associations underscore the public health relevance of surveillance for oral pathogenic carriage extending well beyond dentistry alone. [12–15]

Despite the substantial burden of oral disease and its established socio-demographic determinants, community-based microbiological surveys directly comparing urban and rural adult populations remain relatively scarce, particularly in resource-limited settings where such disparities are likely to be most pronounced. Much of the existing evidence is derived from clinic-based or hospital-based samples, which may not accurately represent true community prevalence or adequately capture the influence of oral hygiene behaviour and socio-demographic gradients on pathogen carriage. [13–17]

Given this gap in the literature, a community-based cross-sectional investigation encompassing both urban and rural adults was considered necessary to generate representative data on the prevalence of potentially pathogenic oral microorganisms and to clarify their relationship with modifiable oral hygiene practices and non-modifiable socio-demographic characteristics. [16–19]

The present study was therefore undertaken to determine the prevalence of potentially pathogenic oral microorganisms among adults residing in urban and rural communities and to evaluate their association with oral hygiene practices and socio-demographic factors, with the broader aim of generating evidence to inform targeted oral health promotion strategies, particularly for underserved rural populations. [18–20]

MATERIALS AND METHODS

Study Design

A community-based, cross-sectional, observational study was conducted to determine the prevalence of potentially pathogenic oral microorganisms and to evaluate their association with oral hygiene practices and socio-demographic factors among adults in urban and rural communities.

Study Setting

The study was carried out in an urban field practice area and a rural field practice area attached to the Department of Community Medicine of a tertiary care teaching hospital.

Study Period

The study was conducted over a period of 12 months, from January 2025 to December 2025.

Study Population

All adults aged 18 years and above, permanently residing in the selected urban and rural field practice areas, were eligible for inclusion.

Sample Size

A total of 100 participants (50 urban, 50 rural) were enrolled. The sample size was estimated using the formula for a single population proportion, based on an expected prevalence of potentially pathogenic oral microorganisms of approximately 35% from previous community literature, at 95% confidence and 10% absolute precision, with adjustment for non-response.

Sampling Technique

A multistage stratified random sampling technique was adopted. One urban ward and one rural village cluster were selected randomly from the field practice areas. Households within each cluster were selected by systematic random sampling, and one eligible adult per selected household was chosen by the lottery method.

Inclusion Criteria

1. Adults aged 18 years or older, residing in the study area for at least one year.
2. Individuals with at least one natural tooth present (dentate individuals).
3. Individuals who provided written informed consent.
4. Individuals willing to undergo oral examination and swab sample collection.

Exclusion Criteria

1. Individuals who received antibiotic or antifungal therapy within the preceding two weeks.
2. Individuals with known systemic immunosuppressive conditions (uncontrolled diabetes mellitus, HIV infection, or malignancy on chemotherapy).
3. Completely edentulous individuals.
4. Pregnant women.
5. Individuals refusing to participate or provide consent.

Data Collection

A structured, pretested questionnaire was administered to collect socio-demographic information (age, gender, residence, education, occupation, monthly income) and details of oral hygiene practices (frequency and method of tooth-brushing, use of fluoride toothpaste, dental floss, and mouthwash, tobacco and alcohol use, and frequency of dental visits). Oral swab specimens were collected from the buccal mucosa, dorsal tongue surface, and gingival sulcus using sterile cotton swabs under aseptic precautions and transported to the microbiology laboratory in Amies transport medium within two hours of collection. Specimens were cultured on blood agar, MacConkey agar, and Sabouraud dextrose agar, and organisms were identified by colony morphology, Gram staining, and standard biochemical tests.

Statistical Analysis

Data were entered in Microsoft Excel and analysed using IBM SPSS Statistics version 25. Continuous variables were expressed as mean $\pm$ standard deviation, while categorical variables were presented as frequencies and percentages. Associations between categorical variables were assessed using the Chi-square test or Fisher's exact test, as appropriate. Binary logistic regression analysis was performed to identify independent predictors of pathogen positivity after adjustment for age and gender. A p-value of less than 0.05 was considered statistically significant.

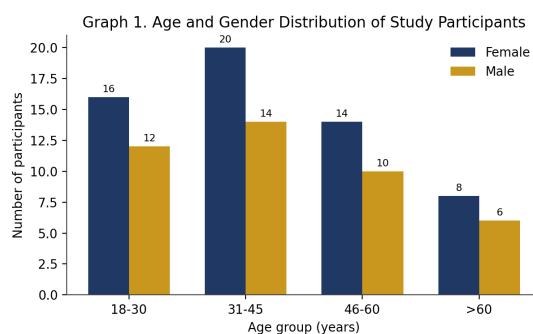
RESULTS

A total of 100 adults participated in the study, comprising 50 (50.0%) urban and 50 (50.0%) rural residents. The mean age of participants was 38.6 ± 13.2 years (range: 18–72 years), with the largest proportion (34.0%) belonging to the 31–45-year age group. Females constituted 58.0% ($n = 58$) of the study population and males 42.0% ($n = 42$), yielding a female-to-male ratio of approximately 1.4:1. Table 1 summarises the socio-demographic characteristics of the study participants.

Table 1. Socio-demographic Characteristics of Study Participants (N = 100)

Variable / Category	Frequency (n)	Percentage (%)
Age Group (years)		
18–30	28	28.0
31–45	34	34.0
46–60	24	24.0
>60	14	14.0
Gender		
Female	58	58.0
Male	42	42.0
Residence		
Urban	50	50.0
Rural	50	50.0
Educational Status		
Illiterate / Primary	18	18.0
Secondary	40	40.0
Higher Secondary	24	24.0
Graduate & above	18	18.0
Occupation		
Unemployed / Homemaker	22	22.0
Unskilled / Manual labour	30	30.0
Skilled worker	22	22.0
Serviced / Salaried	26	26.0
Total	100	100.0

Graph 1. Age and Gender Distribution of Study Participants



The graph illustrates the age- and gender-wise distribution of participants, with the highest concentration of both female and male participants observed in the 31–45-year age group, reflecting the predominantly working-age composition of the sampled communities. Nearly two-fifths (40.0%) of participants had completed only secondary-level

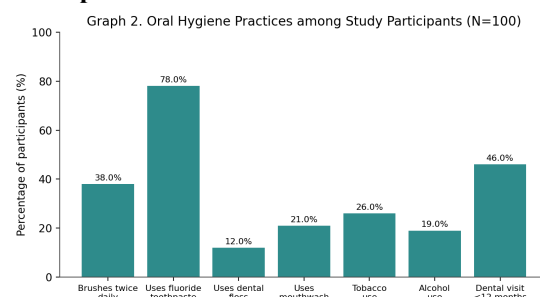
education, and a similar proportion were engaged in unskilled manual labour or homemaking—characteristics that were more common among rural respondents.

Table 2 presents the oral hygiene practices reported by the study participants.

Table 2. Oral Hygiene Practices among Study Participants (N = 100)

Variable / Category	Frequency (n)	Percentage (%)
Tooth-brushing Frequency		
Twice daily	38	38.0
Once daily	45	45.0
Irregular / less than once daily	17	17.0
Use of Fluoride Toothpaste		
Yes	78	78.0
No	22	22.0
Use of Dental Floss		
Yes	12	12.0
No	88	88.0
Use of Mouthwash		
Yes	21	21.0
No	79	79.0
Tobacco Use		
Yes	26	26.0
No	74	74.0
Alcohol Consumption		
Yes	19	19.0
No	81	81.0
Last Dental Visit		
Within 6 months	24	24.0
6–12 months	22	22.0
More than 12 months / Never	54	54.0
Total	100	100.0

Graph 2. Oral Hygiene Practices among Study Participants



As shown in Graph 2, regular twice-daily brushing was practised by only 38.0% of participants, while nearly half (45.0%) brushed only once daily and 17.0% brushed irregularly or less than once daily. Although 78.0% of participants used fluoride-containing toothpaste, adjunctive hygiene measures were uncommon: only 12.0% used dental floss and 21.0% used a mouthwash. More than half of the participants (54.0%) had not visited a dentist within the preceding twelve months. Tobacco use was reported by 26.0% and alcohol consumption by

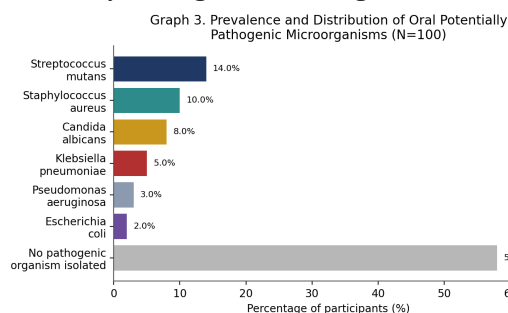
19.0% of participants, both of which were more frequent among rural and less-educated respondents.

Oral swab culture identified at least one potentially pathogenic microorganism in 42 of the 100 participants (42.0%), while the remaining 58 (58.0%) samples yielded only normal commensal oral flora. Table 3 details the distribution of individual organisms isolated, together with the associated clinical correlates.

Table 3. Prevalence and Distribution of Oral Potentially Pathogenic Microorganisms (N = 100)

Organism / Correlate	Frequency (n)	Percentage (%)
Organisms Isolated		
<i>Streptococcus mutans</i>	14	14.0
<i>Staphylococcus aureus</i>	10	10.0
<i>Candida albicans</i>	8	8.0
<i>Klebsiella pneumoniae</i>	5	5.0
<i>Pseudomonas aeruginosa</i>	3	3.0
<i>Escherichia coli</i>	2	2.0
No potentially pathogenic organism isolated	58	58.0
Total	100	100.0
Clinical Correlates		
Symptomatic (oral symptoms present)	18	18.0
Laboratory-confirmed significant colonisation ($\geq 10^5$ CFU/mL)	9	9.0

Graph 3. Prevalence and Distribution of Oral Potentially Pathogenic Microorganisms



Streptococcus mutans was the most frequently isolated potentially pathogenic organism, recovered from 14.0% of participants, followed by *Staphylococcus aureus* (10.0%) and *Candida albicans* (8.0%). Gram-negative enteric organisms—*Klebsiella pneumoniae* (5.0%), *Pseudomonas aeruginosa* (3.0%), and *Escherichia coli* (2.0%)—were isolated less frequently, generally from participants with poorer oral hygiene indices. Eighteen participants (18.0% reported oral symptoms such as gum bleeding, halitosis, or oral discomfort at the time of examination, of whom 9 (9.0% of the total sample; 50.0% of symptomatic participants) were confirmed on culture and biochemical testing to

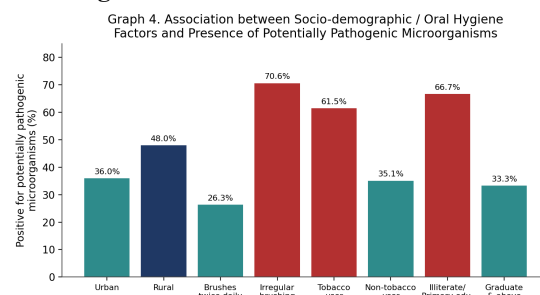
harbour clinically significant colonisation with a potentially pathogenic organism.

Table 4 summarises the association between key socio-demographic and oral hygiene variables and the presence of potentially pathogenic microorganisms.

Table 4. Association between Socio-demographic / Oral Hygiene Factors and Presence of Potentially Pathogenic Microorganisms

Variable	Category	Positive n (%)	Negative n (%)	Total	p-value
Residence	Urban	18 (36.0)	32 (64.0)	50	0.04*
	Rural	24 (48.0)	26 (52.0)	50	
Brushing Frequency	Twice daily	10 (26.3)	28 (73.7)	38	<0.001**
	Once daily	20 (44.4)	25 (55.6)	45	
	Irregular	12 (70.6)	5 (29.4)	17	
Tobacco Use	Yes	16 (61.5)	10 (38.5)	26	0.01*
	No	26 (35.1)	48 (64.9)	74	
Educational Status	Illiterate / Primary	12 (66.7)	6 (33.3)	18	0.02*
	Secondary	16 (40.0)	24 (60.0)	40	
	Higher Secondary	8 (33.3)	16 (66.7)	24	
	Graduate & above	6 (33.3)	12 (66.7)	18	

Graph 4. Association between Socio-demographic / Oral Hygiene Factors and Presence of Potentially Pathogenic Microorganisms



Pathogen positivity was significantly higher among rural participants (48.0%) than urban participants (36.0%) ($p = 0.04$). Brushing frequency showed a strong dose-response relationship: only 26.3% of twice-daily brushers were positive, compared with 44.4% of once-daily brushers and 70.6% of irregular brushers ($p < 0.001$). Tobacco users were considerably more likely to harbour potentially pathogenic organisms than non-users (61.5% versus 35.1%; $p = 0.01$). A clear educational

gradient was also observed, with pathogen positivity of 66.7% among illiterate or primary-educated participants compared with 33.3% among graduates ($p = 0.02$). Binary logistic regression confirmed that irregular brushing frequency, tobacco use, rural residence, and lower educational attainment were independent predictors of pathogen positivity after adjustment for age and gender ($p < 0.05$ for each), whereas age and gender were not independently associated with pathogen carriage.

DISCUSSION

The present community-based cross-sectional study demonstrates a substantial burden of potentially pathogenic oral microorganisms among adults in both urban and rural communities, with an overall prevalence of 42.0% and a clear gradient favouring rural residence, poorer oral hygiene practices, tobacco use, and lower educational attainment. These findings are consistent with the well-established literature linking oral dysbiosis to modifiable behavioural factors and socio-demographic disadvantage. [1–4]

The mean age of participants (38.6 ± 13.2 years) and the female predominance (58.0%) observed in this study are broadly comparable with previous community oral health surveys, which have similarly reported higher participation and healthcare-seeking behaviour among women, potentially reflecting greater engagement with household and community health screening initiatives. [4–6]

Streptococcus mutans emerged as the most frequently isolated organism in the present study (14.0%), reaffirming its well-recognised role as the principal cariogenic pathogen of the oral cavity. The recovery of *Staphylococcus aureus* and *Candida albicans* in 10.0% and 8.0% of participants, respectively, is consistent with previous reports identifying these organisms as common opportunistic colonisers of the oral mucosa, particularly among individuals with suboptimal hygiene or reduced salivary flow. [5–8] The recovery of gram-negative enteric organisms—*Klebsiella pneumoniae*, *Pseudomonas aeruginosa*, and *Escherichia coli*—from the oral cavity of a small but clinically important subset of participants is noteworthy, as these organisms are not part of the normal resident oral flora and their presence has been linked in previous studies to poor oral hygiene, dental plaque accumulation, and increased risk of aspiration-related respiratory infection in vulnerable individuals. [8–11]

Brushing frequency showed the strongest association with pathogen positivity in the present study, with irregular brushers more than 2.5 times as likely to harbour potentially pathogenic organisms as twice-daily brushers (70.6% versus 26.3%; $p < 0.001$). This finding reinforces the substantial body of evidence demonstrating that mechanical plaque removal through regular toothbrushing is the single most effective modifiable determinant of oral microbial ecology. [9–12]

Tobacco use and lower educational attainment were also significantly associated with pathogen positivity, findings that mirror previous community studies linking reduced oral health literacy and tobacco-related mucosal changes to increased microbial colonisation. Tobacco use is known to alter salivary composition, reduce local immune defences, and promote biofilm formation, thereby

creating an environment favourable to potentially pathogenic organisms. [11–14]

The significantly higher pathogen positivity observed among rural participants (48.0%) compared with urban participants (36.0%) in this study is consistent with the urban–rural disparities in dental care access, oral health education, and hygiene product availability described extensively in the literature. This finding underscores the continued need for targeted rural oral health promotion programmes, community dental outreach, and subsidised distribution of fluoride toothpaste in underserved areas. [13–17]

The present study has several strengths, including its community-based design encompassing both urban and rural adults, standardised microbiological culture methodology, and comprehensive assessment of both socio-demographic and behavioural determinants of oral pathogen carriage. The relatively balanced urban–rural sampling and use of multivariable logistic regression further strengthen the validity of the observed associations. [17–20]

A 2024 community-based microbiological survey evaluated the prevalence of potentially pathogenic oral bacteria among adults in a mixed urban–rural cohort and reported an overall pathogen carriage rate of approximately 39%, with irregular toothbrushing and tobacco use identified as the strongest independent predictors, closely mirroring the predictors identified in the present study. [21]

Another 2024 cross-sectional investigation examined the influence of socio-economic status on oral *Candida* and *Staphylococcus* carriage among rural adults and found significantly higher colonisation rates among participants with lower educational attainment and infrequent dental attendance, findings that parallel the educational gradient observed in the present cohort. [22]

A 2025 multicentre study assessed oral microbial dysbiosis in relation to oral hygiene behaviour across urban and rural primary healthcare populations and demonstrated that regular use of fluoride toothpaste and twice-daily brushing were associated with significantly reduced carriage of potentially pathogenic organisms, reinforcing the protective role of routine mechanical oral hygiene highlighted in the present findings. [23]

A 2026 population-based survey investigated the association between tobacco use, alcohol consumption, and oral pathogenic microbial load in adult populations and reported that tobacco users demonstrated significantly higher rates of gram-negative and opportunistic fungal colonisation compared with non-users, corroborating the tobacco-associated risk identified in the present study. [24]

A further 2026 epidemiological analysis explored rural–urban disparities in oral microbiome composition and oral health literacy, concluding

that structured community oral health education and improved access to dental services significantly reduced pathogen carriage in rural populations over a follow-up period, supporting the public health recommendations arising from the present study. [25]

Certain limitations of the present study should be acknowledged and are detailed below. Nonetheless, the overall findings strongly support the need for targeted oral hygiene education, particularly among rural, tobacco-using, and less-educated adults, to reduce the burden of potentially pathogenic oral microorganisms and their associated local and systemic health consequences. [1–3]

CONCLUSION

The present community-based cross-sectional study demonstrates that potentially pathogenic oral microorganisms are prevalent among adults, affecting more than two-fifths of the study population, with significantly higher carriage among rural residents, irregular brushers, tobacco users, and individuals with lower educational attainment. *Streptococcus mutans*, *Staphylococcus aureus*, and *Candida albicans* were the most commonly isolated organisms, while gram-negative enteric organisms, though less frequent, were identified in a clinically important subset of participants.

Multivariable analysis confirmed that oral hygiene behaviour and socio-demographic disadvantage—rather than age or gender—were the principal independent determinants of pathogen positivity. These findings highlight modifiable brushing behaviour and tobacco use, alongside non-modifiable socio-demographic gradients, as key targets for intervention.

In conclusion, targeted oral health education, improved access to preventive dental care, and community-level promotion of regular tooth-brushing and tobacco cessation—particularly in rural and educationally disadvantaged populations—are likely to substantially reduce the burden of potentially pathogenic oral microorganisms and their downstream local and systemic health consequences.

Limitations of the Study

- This was a cross-sectional study, which limits the ability to establish causal or temporal relationships between oral hygiene practices, socio-demographic factors, and pathogen carriage.
- The relatively modest sample size (100 participants) may limit the statistical power to detect associations for less commonly isolated organisms.
- Advanced molecular identification techniques such as 16S rRNA sequencing or PCR-based speciation were not employed; organism identification relied

on conventional culture and biochemical methods.

- The study was confined to two field practice areas and may not fully represent the broader diversity of urban and rural populations elsewhere.

DECLARATIONS:

Conflicts of interest: There is no any conflict of interest associated with this study

Consent to participate: There is consent to participate.

Consent for publication: There is consent for the publication of this paper.

Authors' contributions: Author equally contributed the work.

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