RELATIONSHIP BETWEEN GUTKHA CHEWING PEOPLE AND THEIR SALIVA COMPOSITION INCLUDING BACTERIAL MICROFLORA

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ABSTRACT

Background: Chewing tobacco has been a tradition in India for centuries. But oral diseases are still a neglected epidemic. Of the total amount of tobacco produced in the country, around 48% is in the form of chewing tobacco, 38% as bidis, and 14% as cigarettes. Chewing mixes without tobacco are termed as ‘Pan Masala’ and with tobacco as ‘Gutkha’. In the present study an attempt was made to find a relationship between gutkha chewing people and their saliva composition including bacterial microflora.

Materials and Methods: 50 subjects were chosen randomly in a college which also included gutkha chewing people. Saliva was collected, inoculated on media and observed.

Results: Decreased salivation and mucous formation was observed in 20 gutkha chewing subjects. Hence reduced number of normal symbiotic salivary microflora. Many bacterial species were commonly present both in the healthy and gutkha chewing subjects.

Conclusion: We expect the exploration of composition and microbial diversity in the saliva will direct the development of efficient and noble prevention methods of oral diseases due to eating gutkha.

KEYWORDS Gutkha, Oral Cavity, Microflora

INTRODUCTION - Tobacco- a plant native to America, introduced by Portuguese in India in 1605, was used initially for ceremonial and medical purposes. It is now used in various forms all over the world. Chewing tobacco has been a tradition in India for centuries. It is the third largest tobacco-producing country (13%) after China (40%) and USA (15%). The Indian National Sample Survey revealed that 35% men and 12% women use tobacco in one form or another. Several types such as smoking practices (e.g. cigarette, bidi, hukka and pipe), tobacco chewing (e.g. tobacco-beetle i.e. Pan, gutkha and khaini), rubbing tobacco powder/paste on gums and teeth, and snuffing tobacco are prevalent with varying frequency in different communities. Thus, bidis, sniff and chewing tobacco (such as gutka, khaini and zarda) form the bulk (86%) of India’s total tobacco production. Chewing mixes without tobacco are termed as ‘Pan Masala’ and with tobacco as ‘Gutkha’. Oral diseases are still a neglected epidemic. About 8 lakhs deaths occur every year in India directly as a result of cardiovascular diseases, chronic obstructive lung diseases and tobacco related cancers. So tobacco causes almost one-third of all deaths. During the recent years, in India, both in urban and rural, consumption of tobacco in the form of direct chewing of gutkha is alarmingly increasing especially in the young adults as which is major reason for subsistence of oral cancer. Saliva is a complex biological fluid composed of a wide variety of organic and inorganic constituents. The major organic constituents of saliva are proteins and glycoproteins while main inorganic constituents are phosphate and calcium ions. Interest in saliva as a diagnostic fluid has grown exponentially in recent years. The oral cavity provides two types of surfaces for microorganisms’ colonization – soft tissue and hard tooth enamel/exposed root surfaces. It comprises of different environments such as the tongue, cheek, palate, tooth surfaces and gingival crevice, which are colonized by different microfloras. These microfloras are influenced by host factors such as tooth eruption.
MATERIALS AND METHODS

The study was performed at PL Mishra Public Inter College, Gooja, Kanpur. Subjects were chosen randomly which included students, lecturers and other office staff. The control subjects were selected from healthy subjects. Data was collected by distributing questionnaire to all the subjects. Dental check up was conducted. Saliva was collected from a group of 50 subjects by mouth rinse with 10 mL sterile saline for 30 sec. The saline solution had been UV-irradiated to avoid DNA contamination. Samples collected in sterilized eppendorf tubes were placed on ice immediately. They were plated as soon as possible otherwise stored at -80°C until further use. Salivation and mucous formation was observed under microscope. Ten fold serial dilutions of samples were prepared in saline solution. Microorganisms were isolated by spreading diluted samples (100μl) in small petri dishes containing Nutrient Agar (NA). All plates were incubated at 37°C for 3-5 days. Following incubation, microbial colonies were purified by sub-culturing on the same media used for isolation. In total, 70 bacterial microflora strains were isolated.

RESULTS

A total of 50 questionnaires were administered. 25 were students, 15 lecturers and 10 office staff. Total of 27 males and 23 females, out of which 20 were gutkha chewing subjects. 70 bacterial strains were isolated. Many bacterial species were commonly present both in the healthy and gutkha chewing subjects. Such as Peptococcus niger, Fusobacterium, Streptococcus, Campylobacter, Pseudomonas, Veillonella and Prevotella species. Three genera-Prevotella, Streptococcus and Veillonella constituted about 50% of the salivary microflora. Streptococcus species were more abundant in gutkha chewing subjects. Prevotella and Veillonella species were abundant in non gutkha chewing subjects. It was also found that gutkha chewing subjects had decreased salivation and mucous formation. So overall there was reduced normal symbiotic microflora in gutkha chewing subjects (Figure 1)

DISCUSSION

During the recent years, in India, both in urban and rural, consumption of tobacco in the form of direct chewing of gutkha is alarmingly increasing especially in the young adults as which is major reason for subsistence of oral cancer. Sangita Sushree Nayak found that more males use gutkha than females.\textsuperscript{10} We too found the same during our study. Out of 20 subjects, 16 were males and 4 females. Our main focus, however, was to find relationship between gutkha chewing people and their saliva and bacteria microflora and how it is different from that of non-gutkha chewing people. It was found that even teenagers too had gutkha as time pass. Avasn et al. conducted this sort of study for gutkha chewing including smoking and oral micro flora in some slum dwellers of Visakhapatnam. The results revealed that decrease in salivation and mucous formation in gutkha chewers, which further resulted in reduction in number of oral micro flora.\textsuperscript{1} Our findings were the same. Genotoxicity of Pan masala and gutkha among chewers was examined by Smita Jyoti et al.\textsuperscript{11} In the young, changes in the oral microflora that have been related directly to age are reasonably well established and are usually associated with major perturbations to the habitat (e.g., tooth eruption, hormonal changes, etc.) while in adults, the resident microflora is believed to remain relatively stable unless the environment is disturbed, as occurs during disease was examined by Percival et al.\textsuperscript{5} We used nutrient agar media to isolate bacteria from saliva samples. A total of 70 bacterial strains were isolated. Three genera-Prevotella, Streptococcus and Veillonella constituted about 50% of the salivary microflora. Similar findings were by Jung-Gyu Kang et al.\textsuperscript{5} Streptococcus species were more abundant in gutkha chewing subjects. Prevotella and Veillonella species were abundant in non gutkha chewing subjects. It was also found that gutkha chewing subjects had decreased salivation and mucous formation. It was also observed that gutkha chewing subjects had one or the other dental issues. Therefore, a saliva-based test could prove very useful for early detecting and monitoring of health effects of these habits since saliva is a readily available specimen\textsuperscript{7} so we can say that an understanding of the composition of the oral
microbial ecosystem in relation to oral health is essential for the prevention and treatment of oral diseases.

CONCLUSION

Drug addiction in general and gutkha use in particular represent a complex phenomenon, which has various social, cultural, biological, geographical, historical and economic aspects. The use of gutkha is growing day by day and teenagers are the main consumers. The main users are males and people belonging to nuclear families. Gutkha chewing may lead to an increase in the oral pathogens by reducing the normal symbiotic microflora. Our results provide a framework for further studies of saliva microflora and for investigations of various factors that influence salivary microbiome. These in turn will direct the development of efficient and noble prevention methods of oral diseases due to eating gutkha. The alarming scenario demands that federal regulatory and health agencies and NGOs should launch awareness programmes to inform and educate the public regarding the adverse health consequences and possible cancer risk associated with gutkha.

![Figure 1: Mean CFUs of various microflora in gutkha and non-gutkha chewing subjects](image)

REFERENCES


5) Jung-Gyu Kang, Seong Hwan Kim, and Tae-Young Ahn. Bacterial Diversity in the Human Saliva


