Effect of smokeless tobacco on oral cancer: A case-control study

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Abstract: Oral cancer is one of the most common life threatening diseases in Asian countries. Smoking, smokeless tobacco and alcohol are considered to be the most potent risk factor for oral cancer. This study was conducted to investigate the association of smokeless tobacco with oral cancer. A case-control study of 350 cases and 350 controls over a period of 19 months during February 2005 and September 2006 was carried out in Pune, India. The self-reported information about the consumption of smokeless tobacco, alcohol, dietary habits and demographic status was collected by structured questionnaires. Univariate and multivariate analysis was used to identify the risk of substances abuse. The frequency of smokeless tobacco in cases were significantly higher than controls ($P < 0.0001$). Among smokeless types, consumption of gutkha ($P < 0.0001$, OR = 12.8, 95% CI =7.0-23.7), chewing tobacco ($P < 0.0001$, OR = 8.3, 95% CI = 5.4-13.0), supari ($P < 0.0001$, OR = 6.6, 95% CI =3.0-14.8), mishiri ($P < 0.0001$, OR = 3.3, 95% CI =2.1-5.4), indicated strong association with oral cancer upon adjustment. Smokeless products were more frequent than smoking among individuals with oral cancer. Of smokeless type; gutkha, supar, i chewing tobacco and mishiri emerged significant in oral cancer development.
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Key words: Oral Cancer; Smokeless Tobacco; Case-Control Study

1. Introduction

Tobacco, particularly in association with alcohol, has been recognized as an important risk factor for oral cancer for almost half a century (Wynder and Bross, 1957; Alamdari et al., 2012; Thanaa et al., 2012). Whereas in most countries cigarettes are the main form of tobacco use, in India where oral cancer is a striking incidence, account for approximately 50% of all cancer cases (Khandekar et al., 2006), only less than one-fifth (19%) of tobacco consumed is in the form of cigarettes. On the other hand, about one-fourth of tobacco consumption is in smokeless form, such as chewing tobacco (tobacco flakes), and mishiri (tooth cleaner applied tobacco) (Panchamukhi, 2008).

Moreover, areca nut, seed of the fruit of the oriental palm *Areca catechu* is common to use in India and other South Asian countries. It is the basic ingredient of a variety of widely used chewed products. The basic forms of areca nut usage can be classified as the traditional form and the modern form. In the traditional form, naturally crud areca nut is used wrapped in leaves of piper betel, with lime, saffron and additives such as catechu, cinnamon, cloves. This preparation is referred as betel leaf or paan. When tobacco is added to this preparation it is referred to as betel leaf with tobacco. Since 1970's and 80's the areca nut industries, using traditional Indian technology, along with the tobacco industry has developed products similar to betel quid ready for immediate consumption, packed in small, beautiful and convenient sachets. This product without tobacco is known as 'panmasala' while the product with tobacco is known as 'gutkha'. People consume this abundantly, even by those who do not have the habit of smoking or any other form of tobacco addiction. The other forms of areca nut include indigenous preparations like supari (a naturally crud areca nut without adding other ingredients) (Rooban, 2005).

Despite this knowledge, there remain several outstanding issues to be clarified, including the determination of which type of tobacco and which kind of oral dips substances are exactly associated with oral cancer. Moreover, what is the estimation of the magnitude of the effect of tobacco and poly-ingredient oral dip products on oral cancer? A further issue is whether the effects seen elsewhere can be demonstrated in Pune, India, where smokeless tobacco products differ somewhat from those in other countries. Thus in this study we investigated how smokeless tobacco products consumption was associated with oral cancer.

2. Material and Methods

Study subjects and protocol

The study was approved by local institutional ethical committee. Written informed consent forms were obtained from each participant. The study design was a hospital-based case-control where were conducted at Pune Cancer Institute within 19 months

from February, 2005 to September, 2006. A total of 700 age-gender grouping matched subjects including 350 cases and 350 controls were selected using simple random sampling. Cases were the newly diagnosed patients of oral cancer (tongue, gingival, buccal, retromolar trigone, lip, mouth floor and hard palate cancers) aged above 18 years; the patients first were screened by well-trained interviewers using 10-page structured questionnaire. To confirm the diagnosis, including and excluding criteria were implemented through physical and histopathological examination and classified by the standard International Classification of Diseases (ICD10) criterion. The participants were invited and then registered based on institute guidelines. In fact, we pulled the names of administered patients daily by chance. Written consent forms were obtained from each participant. These individuals were assigned as case, otherwise, the following one being pulled. Therefore, we carried out group matching to select controls randomly/by chance and the same as for cases.

The controls were selected from the relatives, friends and caretakers of cases, who were accompanied the patients at the hospital and were healthy and did not reportedly have cancer. Firstly, the patient's accompanied was invited and then registered as a control group. Then in case of their agreement a written consent forms were obtained from each control participant similar to cases. At the well-situated a trained interviewer interviewed the control participants. To reduce the recall bias interviewer asked them to first, think about the year before, then, to further rear years.

We used structured questionnaire to obtain complete information on demographical characteristics such as age, gender, education, income, place of residency, occupational history, religion, marital status, tobacco-related behavior, smokeless tobacoo, alcohol consumption and dietary habits. Medical record was available in patients' record files preserved in Morbai Naraindas Budhrani Cancer Institute.

Nevertheless, designing an instrument to collect the valid and precise data was very important. To build proper questionnaire, we took the following steps: Firstly, based on research literature and consulting expertise in the field on potential risk factors concerning oral cancer (Balaram, 2002), we then prepared a primary questionnaire with typically open-ended questions and were presented to 10 patients with oral cancer and 10 healthy subjects. Secondly, we prepared a structured questionnaire. It was then presented to 10 patients and 10 healthy subjects. Thirdly, we then reviewed the questionnaire with expertise and discussed all points that were in any way confusing and work together to solve the problems. To ensure content validity of the tool it was submitted to seven experts in field of oncology, public health, epidemiology and social medicine. We modified our questionnaire concerning to comments and suggestions of the experts. To control reliability of the instrument, we submitted the final questionnaire to 20 subjects (10 cases and 10 controls) of the Morbhai Naraindas Budhrani Cancer Institute, and were asked to answer the question with the help of a trained investigator and then we referred to them after approximately 10 days and were submitted to them to the same bank questionnaire and the process was repeated similarly. Responses to the questions in the first and tenth round were not notably different.

Statistical analysis

The data is presented as the numbers with percentage (prevalence). The significance of difference between the proportions of qualitative characteristics is tested using Chi-square test of independence of attributes. The multivariate associations with oral cancer were tested using multiple logistic regression analysis. The quantitative risk assessment was done by calculating the odds ratios (OR) with 95% confidence intervals. All the associations were adjusted for potential confounders like the use of other tobacco types, education, location and monthly household income as appropriate.

The entire data was analyzed using a statistical package for social sciences (SPSS) version 13.The statistical analysis was carried out by calculating the Odds Ratio (OR) for oral cancer with multiple logistic regression models, with a 95% confidence interval. In the first place, the variables were analyzed by univariate regression models, and then a global model was made with all the variables. Finally, a multiple logistic regression model was obtained with the variables having statistical significance. The regression models were made with the computer program.

3. Results

Cases and controls were similar with respect to age, gender and place of residence, but cases reported less education, lower monthly household income and difference in types of occupation. Data related to demographic characteristics of subjects are presented in Table 1. In general, overall smoking and smokeless forms as well as overall drinking alcohol, and non-vegetarian diet habit were significantly different between cases and controls ($p < 0.001$).

Smoking further categorized into 3 sub types viz. filtered cigarette, non-filtered cigarette and bidi; the prevalence of all types was significantly different for cases compare to controls ($P < 0.001$).

Table 1. Demographic characteristic of the participants

Characteristics	Cases n (%)	Controls n (%)	*P* value
	n= 350	n= 350	
Gender			
Male	251 (71.7)	254 (72.6)	0.8
Female	99 (28.3)	96 (26.4)	
Age			
< 40	78 (22.3)	75 (21.4)	0.78
41-50	85 (24.3)	76 (21.7)	
51-60	94 (26.9)	104 (29.7)	
61 +	93 (26.6)	95 (27.1)	
Location			
Rural	94 (26.9)	87 (24.9)	0.4
Urban & semi urban	256 (73.1)	263 (75.1)	
Education			
Literate	269 (76.9)	323 (92.3)	0.001
Illiterate	81 (23.1)	27 (7.7)	
Primary school	97 (27.7)	30 (8.6)	
Middle school	59 (16.9)	58 (16.6)	
High school	59 (16.9)	134 (38.3)	
Undergraduate	33 (9.4)	72 (20.6)	
Postgraduate	21 (6.0)	29 (8.3)	
Occupation			
[1]Agriculture	89 (25.4)	42 (12.0)	0.001
[2]Blue collar	82 (23.4)	63 (18.0)	
[3]White collar	32 (9.1)	54 (15.4)	
[4]Self-employed	49 (14)	74 (21.1)	
Professional	21 (6.0)	29 (8.3)	
Unemployed	77 (22)	88 (25.3)	
[5]**Monthly Income**			
Rs < 5000	183 (75.6)	136 (51.7)	0.001
> 5000	59 (24.4)	127 (48.3)	

Values are n (%), p-by Chi-square test, P>0.05 = Not Significant. 1. Farm's worker; 2. Manual/industrial laborers, skilled/unskilled, building/construction, and mechanical worker. 3. Non-manual labor working in office 4. Businesspersons / contractors, property owners 5. Monthly household income in Indian currency

Similarly, smokeless products further categorized in to sub-types viz, chewing tobacco, mishiri, pan parag, gutkha, supari, and betel leaf (paan); all except betel leaf, (*P=0.112)*, and pan parag (*p=0.621*), shown significantly different by cases compare to controls (Table 2). Uni-variate analysis revealed that oral dipping was significant risk compare to smoking in overall (OR= 7.2; 95% CI= 4.4-11.8 v/s OR= 2.6; 95% CI=1.8-3.7). In terms of smokeless forms, the risk was 7.3 (OR= 7.2; 95% CI, 4.4-11.8) for consumption of gutkha, OR= 5.3 (95% CI, 3.7-7.6) for consumption of chewing tobacco and OR= 4 (95% CI, 2.1-7.8) for consumption of supari (pure areca nut) and the lower risk was found for mishiri (OR= 2.2; 95% CI, 1.5-3.1). In the case of smoking, bidi smoking was significant risk only (OR= 4.1; 95% CI, 2.4-6.9). However, we found no significant risk of filtered cigarette. Multivariate analysis model approved the risk of studied smokeless tobacco products after adjusting for possible confounders like, alcohol, non-vegetarian habit, education, monthly household income, occupation, and other tobacco types (adjusted OR, Table 3).

Table 2. Distribution of subjects by selected habits

Characteristics	Cases n (%)	Controls n (%)	*P* value
	n= 350	n= 350	
Smoking (overall)	125 (35.7)	61 (17.4)	0.001
Filtered cigarette	44 (12.6)	33 (9.4)	0.149
Non-filtered cigarette	15 (4.3)	6 (1.7)	0.046
Bidi	70 (20.0)	20 (5.7)	0.001
Smokeless (overall)[1]	514[1]	202[1]	0.001
Chewing tobacco	175 (50.0)	55 (15.7)	0.001
Betel leaf (Paan)	44 (12.6)	31 (8.9)	0.112
Pan Parag	21 (6.0)	18 (5.1)	0.621
Gutkha	112 (32.0)	21 (6.0)	0.001
Supari	44 (9.1)	12 (3.4)	0.002
Mishri	118 (33.7)	65 (18.6)	0.001
Alcohol (overall)	106 (30.3)	48 (13.7)	0.001
Diet			
Vegetarian	52 (26.4)	145 (73.6)	0.001
Non-Vegetarian	298 (59.2)	205 (40.8)	

[1]There is some overlap in the use of various oral dip products. Values are n (%), p-by Chi-square test.

Table 3. Crude odds ratios (OR) of tobacco and oral dip products

Factors	Cases n (%)	Control n (%)	OR (95% CI)*	*P* value
Bidi	70	20	4.1 (2.4-6.9)	0.0001
Chewing tobacco	175	55	5.3 (3.7-7.6)	0.0001
Gutkha	112	21	7.3(4.5-12.1)	0.0001
Supari	44	12	4.0 (2.1-7.8)	0.002
Mishiri	118	65	2.2 (1.5-3.1)	0.0001

*Un-adjusted OR, P-by Chi-Square test, 95% CI of OR< 1 = Not significant

Table 4. Adjusted odds ratio for tobacco and oral dip products

Characteristics	Cases n (%)	Controls n (%)	*P* value
Bidi	4.1	2.4-6.9	0.0001
Chewing Tobacco	8.3	5.4-13.0	0.0001
Gutkha	12.8	7.0-23.7	0.0001
Supari	6.6	3.0-14.8	0.002
Mishiri	3.3	2.1-5.4	0.0001

[1]Adjusted OR for other tobacco and oral dip products, alcohol, non-vegetarian habit, education, occupation, age and gender. P-by Chi-Square test, 95% CI of OR < 1; not significant

4. Discussion

The aim of this study was to determine the risk of tobacco and smokeless tobacco products associated to oral cancer. Similar to prior studies, we demonstrated that use of tobacco, in overall, was prevalent in cases compare to controls. Comparing

smoking with smokeless, our results showed that smokeless tobacco users, in overall, were more at risk to develop oral cancer. Our finding confirmed the previous result from India (Znaor et al., 2003; Sankaranarayanan et al., 1989a,b; Sankaranarayanan et al., 1990; Nandakumar et al., 1990; and Bundgaard, 1995) as well as in some case-control studies in other countries (Stefani et al., 1998; Macfarlane et al., 1995; Weinberg and Stefan, 1996).

Smokeless tobacco may have a stronger effect than a smoking because of the direct contact of the ingredient carcinogens with the oral epithelium However, the etiologic role of these factors is not well understood, and further methods for modifying them need to be developed (Babu et al., 1996). It is very interesting to know that 112 out of 350 (32%) of cases used gutkha while only 21 out of 350 (6%) of controls used this substance with a maximum association of 7.3 time in development oral cancer. Our results confirm prior findings from India that have shown gutkha as the strong risk for oral submucous fibrosis (OSMF) (Shah and Sharma, 1998) and produce OSMF earlier as compared to supari (crude areca nut) and other products (Gupta and Ray, 2003; Van Wyck et al., 1993). It was found that average betel leaf (areca nut, tobacco, catechu and lime wrapped in betel leaf) approximately weighing 3.5-4 grams has 70 per cent moisture and dry weight of areca nut and tobacco is only 1.14 grams whereas the gutkha sachet weighing 3.5 gram has only 7 per cent moisture and dry weight 3.26 grams (Shah and Sharma, 1998). Since habitual chewers tend to consume more dry weight of areca nut and tobacco when they use gutkha so they probably develop more fibrosis of the oral mucosa, particularly the disorder afflicting quite earlier as well.

We have also found a great risk of oral cancer in chewing tobacco consumers, which is used with or without lime and kept in the mouth for different duration of time depending on the personal habits. Our study confirmed the previous findings that showed chewing of tobacco as the strong risk factor for oral cancer (Znaor et al., 2003; Sankaranarayanan et al., 1989a,b, and 1990; Nandakumar et al., 1990; and Bundgaard, 1995).

Chewing of supari; pure areca nut, has also been implicated as one of the ingredients that can cause oral cancer. Van Wyck et al. (1993) have shown evidence on the carcinogenicity of areca nut, where 68% of South African Indians with cheek cancer are areca nut chewers who do not smoke or drink. Similarly, there is a strong cause and effect relationship between areca nut chewing and oral submucous fibrosis in the South African and Indian studies (Gupta et al., 1998; Stich et al., 1986). Conversely, Stich reported that in Guam where areca nut is chewed alone or with leaf only, there is apparently no increase in oral cancer (Balaram et al., 2002). It may be due to the procedure of preparing the product (El-Domyati et al., 2012).

Surprisingly, despite to previous studies in this study we found no significant results of betel leaf in associated to oral cancer. It may be explained by the protective effects of betel leaf to the oral mucosa against the harmful alkaloids present in the areca nut in betel leaf chewers, because betel leaf (paan) is known to be rich in beta-carotene, which have the capacity to quench free radicals that are toxic. Our data suggests that it is not only smoking tobacco, but also dipping smokeless tobacco and areca nut leads to the development of oral cancer. The hierarchy of importance of effects was related to gutkha, chewing tobacco, supari and mishiri respectively. Our findings give emphasis to public health initiatives targeted to prevent and/or reduce smokeless tobacco products particularly, gutkha, chewing tobacco, supari and mishiri.

Limitations of the study

We might likely have faced overmatching because of selecting controls of relatives, friends and caretakers instead of population source. A combination of population-based and other control source like relatives, friends and caretakers might be a more appropriate approach to reduce the weakness of case-control study. Moreover, same as to the other case-control studies, main limitation of this study was recall bias. To minimize the recall bias we prepared a well-situation for the participants and a trained interviewer asked them to first, think about the year before, then, to go back to further rear years.

5. Conclusion

The statistical data on the relationship between smoking and smokeless tobacco and oral cancer provide strong evidence that smokeless products could be in a straight line responsible for developing oral cancer. Gutkha, supari -areca nut-, chewing tobacco and tobacco powder which applied as a tooth and gum cleaner (mishiri) are independent risk for oral cancer. The public should be aware of the high risk of oral cancer attributed to use above mentioned substances. Further studies are required in other parts of India to demonstrate the similar effects of these consumption also to find out the actual prevalence of oral cancer to get an idea of the burden of the underlying health problem. There is a need for appropriate prevention and planning strategies for gutkha, supari and chewing tobacco consumption.

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References

1. Alamdari Sh, Hossein panah F, Amiri P, Alamdari A, Azizi F. Plan to design Policies For Science, Technology and innovation in the field of obesity Prevention and control measures in 20 years national vision. *Life Sci J* 2012;9(4):5884-5889.
2. Babu S, Bhat RV, Kumar PU, Sesikaran B, Rao KV, Aruna P, et al: A comparative clinico-pathological study of oral submucous fibrosis in habitual chewers of panmasala and betel quid. *Clin Toxicol* 1996; 34: 317-22.
3. Balaram P, Sridhar H, Rajkumar T, Vaccarella S, Herrero R, Nandakumar A. et al: Oral cancer in southern India: the influence of smoking, drinking, paan-chewing and oral hygiene. *Int J Cancer* 2002; 98 (3): 440–445.
4. Bundgaard T, Wildt J,. Frydenberg M, Elbrønd O and Nielsen J.E: Case-control study of squamous cell cancer of the oral cavity in Denmark. *Cancer Causes and Control* 1995; 6: 57–67.
5. El-Domyati F.M., Ramadan A.M., Gadalla N.O., Edris S., Shokry A.M., et al., Identification of molecular markers for flower characteristics in *Catharanthus roseus* producing anticancer compounds. *Life Sci J* 2012; 9(4):5949-5960
6. Gupta P.C, Sinor P.N, Bhonsle R.B, Pawar V.S and Mehta H.C: Oral submucous fibrosis in India: a new epidemic. *Nat Med J India*.1998; 11:113–116.
7. Gupta PC, Ray CS: Smokeless tobacco and health in Indian and South Asia. Respirology 2003; 8:419-31.
8. Khandekar S.P., Bagdey P.S., Tiwari R.R: Oral cancer and some epidemiological factors: A hospital based study. *Indian J Community Med* 2006; 31(3):154-159.
9. Macfarlane G.J, Zheng T, Marshall J.R, Boffetta P, Niu S, Brasure J, Meretti F and Boyle P: Alcohol, tobacco, diet and the risk of oral cancer: a pooled analysis of three case-control studies. *Eur J Cancer* 1995; 31 B (3): 181–187.
10. Nandakumar A., Thimmasetty K.T, Sreeramareddy N.M, Venugopal T.C, Rajanna M.A and Vinutha A.T. et al: A population-based case-control investigation on cancers of the oral cavity in Bangalore, *India. Br J Cancer* 1990; 62 (5): 847–851.
11. Panchamukhi, P. R., Woolery, T. and Nayantara, S. N: Economics of bidis in India. In *Bidi Smoking and Public Health* (eds Gupta, P. C. and Asma, S.), Ministry of Health and Family Welfare, Government of India, 2008; 167–195.
12. Rooban T, Elizabeth J, Anusa R, Girish K. G: Health hazards of chewing arecanut and products containing arecanut. *Calicut Med J* 2005; 3 (2): e 3.
13. Sankaranarayanan R, Duffy SW, Padmakumary G, Day N.E and, Padmanabhan T.K: Tobacco chewing, alcohol and nasal snuff in cancer of the gingiva in Kerala, India. *Br J Cancer* 1989a; 60 (4): 638–643.
14. Sankaranarayanan R, Duffy SW, Padmakumary G, Day N.E and Nair M.K: A case-control investigation of cancer of the oral tongue and the floor of the mouth in southern India. *Int J Cancer* 1989b; 44 (4): 617–621.
15. Sankaranarayanan R, Duffy SW, Padmakumary G, Day N.E and Krishan N.M: Risk factors for cancer of the buccal and labial mucosa in Kerala, southern India. *J Epidemiol Commun Health* 1990; 44 (4): 286–292.
16. Stefani E. De, Boffetta P, Oreggia F, Fierro L and Mendilaharsu M: Hard liquor drinking is associated with higher risk of cancer of the oral cavity and pharynx than wine drinking. A case control study in Uruguay. *Oral Oncology* 1998; 34: 99–104.
17. Stich HF, Rosin MP, Brunnermann KD: Oral lesions, genotoxicity and nitrosamines in betel quid chewers with no obvious increase in oral cancer risk. Cancer Lett. 1986; 31:15-25.
18. Shah N, Sharma PP: Role of chewing and smoking habits in the etiology of oral submucous fibrosis: A case control study. *J Oral Pathol Med* 1998; 27: 475-9.
19. Thanaa. A. El-kholy; Hatim Ali Al Abbadi; A.K.Alghamidi; Hesah Al- Qahtani; Morooj Al- Abya and Noha Mujalli. The Nutritional Status in Patients with Colorectal Cancer Pre and Post Different Modulates Of Treatment. *Life Sci J* 2012;9(4):2219-2230
20. Van Wyck C.W, Stander I, Padayachee A and Grobler-Rabie A.F: The areca nut chewing habit and oral squamous cell carcinoma in South African Indians. *SAfr Med J* 1993; 83:425–429.
21. Weinberg MA, Stefan DJ: Assessing oral malignancies. American physician family 1996; (7): 65-84.
22. Wynder E.L and Bross I.J: Aetiological factors in mouth cancer; an approach to its prevention. *Br Med J* 1957; 18: 1137–1143.
23. Znaor A, Brennan P, Gajalakshmi V, Mathew A, Shanta V, Varghese C et al: Independent and combined effects of tobacco smoking, chewing and alcohol drinking on the risk of oral, pharyngeal and esophageal cancers in Indian men. *Int J Cancer* 2003; 105 (5):681–686.

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