Karp, O’Loughlin, Hanley, Tyndale and Paradis (2006) in their study on 1293 grade 7 students to study the incidence of conversion to tobacco dependence (TD) and the prevalence of TD state in relation to several potential determinants in adolescent smokers found out that 113 of 344 novice smokers converted to TD over 54 months follow up. Conversion to TD and TD status were associated with the intensity of recent cigarette consumption and subjects with slow nicotine metabolism and those with more depression symptoms are at increased risk of becoming tobacco dependent.

Kandel et al., (2006) in their study to examine the extent and sources of discrepancies between self-reported cigarette smoking and salivary cotinine concentration among adolescents found three patterns of inconsistencies. Self reported nonsmoking adolescents with cotinine concentration above 11.4ng/ml cut-point, low cotinine concentration among adolescents reporting having smoked within the last 3 days and high concentration among adolescents reporting not having smoked within the last three days were observed. The study observed an inverse relationship of cotinine concentration with depressive symptoms. Also suggested that depressed adolescent smokers might take in smaller dose of nicotine than non depressed smokers and alternatively depressed adolescents might metabolize nicotine more rapidly.

Baker et al., (2007) on their four placebo controlled smoking cessation trials to determine the relations between the Fagerström Test for Nicotine Dependence, Heaviness of Smoking Index, Nicotine Dependence Syndrome Scale and Wisconsin Inventory of Smoking Dependence Syndrome Scale with cessation success showed that the first item i.e. time to first cigarette in the morning had greater validity than any other measure. Among the treatment-seeking smokers, the majority of individuals (70%) indicated that they smoke within 30 minutes of awakening and the study concluded that time of first cigarette may be a good single item measure of nicotine dependence.

Ferketich, Wee, Shultz and Wewers (2007) in their study on evaluation of salivary cotinine concentration in smokeless tobacco users found out median cotinine concentration to be
The level of cotinine concentration was positively related to the years of smokeless tobacco use. Among the snuff only users, cotinine concentration was positively related to age, no quit attempts and tobacco dependence score and center for epidemiologic studies depression scale. Also the result concluded that the tobacco dependency scale (FTND) score was associated with cotinine, which indicated that these two measures of dependency are significantly related among the smokeless tobacco users.

Nuca, Amariei, Badea, Zaharia and Arendt (2011) on their cross sectional study of 286 participants to evaluate the salivary cotinine levels and self reported smoking status and nicotine dependence using Heaviness of Smoking Index (HIS) found out that the level of cotinine in saliva was higher in males than in females. Also there was a significant correlation between the cotinine levels and self reported smoking status (p<0.05) and also between cotinine levels and HIS. Nicotine dependence levels were low for 41 subjects, moderate for 48 subjects and high for 27 subjects.

Dhavan et al., (2011) in their study of validation of self reports of tobacco use by Indian youth living in low income neighborhoods using salivary cotinine on 1224 samples showed under reporting use of chewing and smoking tobacco. Self reports had a low sensitivity (36.3%) and a positive predictive value of 72.6%. The study concluded that biochemical validation of self reported tobacco use should be considered during prevention and cessation studies.

Manimunda et al., (2012) on their study to estimate the prevalence and determinants of tobacco use and nicotine dependency, using Fagerstrom Test for Nicotine Dependence (FTND) in 18,018 individuals of Andaman and Nicobar Islands, India found the prevalence of current tobacco use in any form was 48.9% and chewing alone was in 40.9 %. Around 9.7 % of the male population and 3% of the female population were found to be nicotine dependent.

Lim et al., (2012) on their study to evaluate the consistency between Heaviness of Smoking Index (HIS), number of cigarette smoked and FTND found out 20.1%, 13.2% and 20.5%, respectively of patients were high nicotine dependent smokers. The sensitivity and specificity of the heavy smoker category compared to find FTND was 67 % and 86.9 % respectively. The study
concluded that FTND and HIS can be used in screening for high nicotine dependence among daily smokers in large population based studies.

Park, Lee, Song and Cho (2012) on their cross sectional study on age associated changes in nicotine dependence of 337,933 participants determined by FTND found out that the dependent variables FTND, partial FTND and Cigarettes per day increased with age until approximately 50 years of age, at which point they decreased. Both nicotine dependence and cigarettes per day displayed an inverse U shaped relationship with age, with a significant peak at 50 years of age and suggested that tobacco control policies should target issues related to nicotine dependence according to age group.

McNeill, Jarvis, West, Russell and Bryant (1987) in their study to measure the salivary cotinine and expired air carbon monoxide in 508 girls aged between 11-16 years found out that a saliva cotinine cut-point of 14.7ng/ml detected 99% of regular daily smokers and performed better than expired air CO in identifying smoking. The mean saliva cotinine among the regular daily smokers was 200.8ng/ml, and comparisons with adult smokers suggested that adolescents inhale similar dose of nicotine per cigarette. The study also found no evidence for an increase in smoke inhalation per cigarette with increasing age suggesting that inhalation develops early in smoking career.

Swan, Habina, Means, Jobe and Esposito (1993) conducted a study to examine the nature of the association between cotinine and reported number of cigarettes smoked in 116 smokers of mean age 37.4 years and smoked 20.1 cigarettes daily. The Pearson correlation between saliva cotinine and the logged number of cigarettes smoked in the previous 17 hours (the time window corresponding to the half-life of cotinine) accounted for significantly more of the variance in cotinine than did the average logged number of cigarettes smoked daily during 5 days. Age was also significantly associated with cotinine levels. The study suggested that adjustments for age and inclusion of a nonlinear component for cigarette consumption would result in more precise use of cotinine as a validation tool for existing differences in smoking levels.
van Poppel, Verhagen, van’t Veer, van Bladeren (1993) in their study on sister chromatid exchanges in peripheral lymphocytes and micronuclei in sputum cells for cytogenetic damage in smokers, which may reflect the increase in cancer risk; found out that in smokers sister chromatid exchange were weakly correlated with plasma cotinine \((r=0.186)\) but not with plasma antioxidants. Micronuclei in smokers were not correlated with either cotinine or antioxidants \((r<0.14)\).

Etter, Vu Duc and Perneger (2000) in their study to evaluate the relation between saliva cotinine concentration and self reported active and passive exposure to tobacco smoke, to identify factors associated with saliva cotinine levels in smokers and to determine an optimal cutoff for the cotinine concentration to distinguish smokers from non smokers found out that among the 207 cigarette only smokers smoked on an average of 10.7 cigarettes/day with a median concentration of cotinine of 113ng/ml and was moderately associated with the number of cigarettes smoked per day \((p<0.001)\). A cutoff of 7ng/ml of cotinine distinguished smokers from non smokers with a sensitivity of 92.3% and a specificity of 89.7% also it was found that each additional cigarette smoked per day was associated with an increase of 14ng/ml in saliva cotinine.

Jaakkola et al (2003) on their study to characterize the distribution of salivary cotinine levels in Chinese smokers and to investigate factors that influence salivary cotinine levels in smokers found out that among smokers consuming 20 per day the median cotinine concentration was higher among younger subjects smoking without filter and regular rather than light cigarettes and those inhaling frequently and deeply. The increase in cotinine per cigarette tended to be larger in those with lower median cotinine level. The study concluded that smoking behavior related factors modify the relation between number of cigarettes smoked and salivary cotinine concentration.

Maziak, Ward and Eissenberg (2006) in their study to assess exposure to environmental tobacco smoke (ETS) among non smokers in the community and examine the relation between various subjective and objective measures of exposure to ETS in a developing country on a sub sample
of 419 non smokers found out that 72.9% were women have detectable saliva cotinine (mean 1.5ng/ml ± 1.7). Correlation between self-reported exposure measures and saliva cotinine was moderate with the strongest observed for number of cigarette smokers in the house, average number of cigarettes smoked daily in the house, house policy regarding smoking, and total ETS score (r 0.3–0.4). These same variables were among the best predictors of saliva cotinine according to stepwise linear regression analysis, but their individual relevance differed between men and women reflecting underlying differences in gender-based behavior–mobility patterns. The study concluded that even in the environment of omnipresence of smoking, household restrictions seemed to offer protection against ETS exposure.

Blackford et al., (2006) in their study regarding salivary cotinine concentration in smokers from Brazil, China, Mexico and Poland and its relation with amount smoked and cigarette type using standard questionnaire, FTND and gas chromatography found out that cotinine concentration increased linearly with cigarettes smoked up to 20 per day (11.3ng/mL; 95% CI) and then stabilized as the number of cigarettes exceeded 20 (6.8ng/mL; 95% CI) for up to 40 cigarettes. The cotinine concentration per cigarette smoked did not differ between regular and light cigarette smokers. The study concluded that intake of nicotine per cigarette is comparable across countries and smokers of more than 20 cigarettes per tend to have lower nicotine intake per cigarette and the type of cigarette smoked has little effect on nicotine intake.

Huang, Lin and Yang (2007) on their study on evaluation of smoking characteristics and saliva cotinine levels between males and females found out that average saliva cotinine level to be significantly higher for males than females (p<0.0001). Family and friends smoking were risk factors of nicotine dependence among both men and women smokers. The study concluded that smoking characteristics and biochemical marker differ between males and females.

Fu et al., (2009) on their cross sectional study to describe and characterize the distribution of salivary cotinine concentration in a representative sample of adult daily smokers in Barcelona, Spain found out that salivary cotinine concentration was associated with the reported number of cigarettes smoked in the previous 24 hours ($R^2=0.339; p<0.05$). They concluded that that
salivary cotinine concentration was significantly associated with the number of cigarettes smoked and sex, with men having higher levels but not with other smoking related variables.

Ashley et al., (2010) on their study to analyze the effect of differing levels of tobacco specific nitrosamines in cigarette smoke on the levels of biomarkers in smokers found out that the relationship between mouth level exposure to nicotine and its salivary metabolite, cotinine was not statistically significant ($\beta=0.29, p=0.057$) likely due to the very limited range of differences in mouth level nicotine exposure in the study population. The study had created an impact that lowering the level of 4-(methylnitrosamino)-1-(-3pyridyl)-1- butane (NNK) in the mainstream smoke of cigarettes through the use of specific tobacco types and known curing practices can significantly affect the exposure of smokers to the known carcinogen.

Nesic, Rusted, Duka and Jackson (2011) in their study to investigate the effects of smoking on pre frontal cortical (PFC) mediated cognitive flexibility and subjective states in low dependent (LD) and high dependent (HD) smokers found out that compared to LD smokers, HD smokers had higher salivary cotinine and breath carbon monoxide levels, reported more craving and felt less vigorous, friendly and elated. The study demonstrated that PFC mediated cognitive effects of smoking as well as subjective reports vary accordingly to the degree of nicotine dependence.

Fu et al., (2012) in their cross sectional study to analyze the relationship between nicotine dependence measured by the Fagerstrom Test for Nicotine Dependence and salivary cotinine in smokers of Barcelona, Spain found out that around 17% of the subjects had high nicotine dependence. The findings suggested that salivary cotinine is associated with nicotine dependence. Salivary cotinine was associated related to daily tobacco consumption, time to first cigarette smoked after waking up, and smoking more in the first hours of the day ($R^2=0.414$).

Nuca et al., (2012) on their study on evaluation of passive smoking prevalence in non smoking adults from Romania, by Salivary NicAlert Strip test, found out that salivary cotinine level most frequently found was level 1. Based on the self-reported smoking status (questionnaire), 44.06% of the subjects (n=126) were classified as active smokers (constant and occasional) and
55.94% (n=160) as non-smokers. Using the salivary cotinine levels as standard, the real
distribution of the subjects according to their smoker status comprised 44.06% active smokers
(constant and occasional), 16.43% (n=47) non-smokers (non-exposed to tobacco smoke) and
39.50% (n=113) passive smokers.

Sarto et al., (1987) analyzed the aneugenic and clastogenic effects induced by tobacco on 25
individuals with such exposure (23 who smoked cigarettes and two who smoked cigars), and
compared them with the same number of non smokers. They observed a significantly higher
frequency of micronuclei consequent to chromosome breakage among the smokers.

Casartelli et al., (2000) on their study had observed MN frequencies in exfoliated buccal cells in
normal mucosa, precancerous lesions and squamous cell carcinoma. They concluded that the
gradual increase in MN counts from normal mucosal to precancerous lesions to carcinoma
suggested a link of this biomarker with neoplastic progression.

Bloching, Hofmann, Lautenschläger, Berghaus, & Grummt (2000) in their study carried out in
55 patients with oral squamous cell carcinoma, 16 patients with leukoplakia and 99 as healthy
individuals to predict the relative risk of cancer in the upper aerodigestive tract using the MN
assay found out that there was a direct correlation between tobacco abuse and increasing MN
count as a sign of cytogenetic damage of buccal mucosal cells but found out that alcohol did
not influence the formation of MN. A significant correlation between increasing pack years of
cigarette and a higher MN rate (p<0.0001; r>0.5) was observed. A significant difference in the
mean MN value of healthy subjects and patients (p<0.05) was observed.

Etter and Perneger (2001) conducted a study to develop accurate measures of self reported
active exposure to cigarette smoke by self reported questions intended to assess active
exposure to cigarette smoke and compared with saliva cotinine and Fagerström test for
nicotine dependence and self rated dependence in population sample from Geneva. Measures
associated with salivary cotinine were number of cigarette smoked per day (r²=0.36), smoking
intensity (r²=0.40), type of cigarette smoked (r²=0.04), smoking when ill (r²=0.15) and a single
item rating of the total quantity of smoke inhaled ($r^2=0.27$) and concluded other measures of exposure to smoke that reflect saliva cotinine better than number of cigarettes per day.

Wu et al., (2004) on their study to investigate the clastogenic effects of areca quid chewing and cigarette smoking using micronuclei from exfoliated buccal mucosal cells found out that heavy smoking was positively associated with MN frequency. A significant positive trend was demonstrated for the relationship between MN frequency and either daily cigarette consumption or cumulative smoking pack years. But MN were not associated with areca quid. They suggested that the carcinogenesis of oral cancer induced by areca nut chewing in Taiwan may be though a pathway other than genotoxicity.

Bohrer, Filho, Oaiva, da Silva and Rados (2005) in their study to assess the presence of micronuclei in exfoliated oral mucosal cells collected from 3 anatomic sites (lower lip, tongue border and floor of the mouth) in patients exposed to tobacco and alcohol found out that in comparison with the three different sites the mean number of cell undergoing karyorrhexis was higher on the lower lip than on the tongue border or floor of mouth. The three groups were similar in terms of the mean number of micronucleated cells. But a significant higher number of broken eggs were observed in the control when compared to the tobacco and tobacco/alcohol groups suggesting it’s association with DNA repair or healthy mucosa. They concluded that MN quantification is useful to detect and monitor genetic damage in populations exposed to genotoxic agents as tobacco and alcohol.

Jadhav, Gupta and Ahmed (2011) on their study correlated the frequency of micronuclei with different grades of histopathologically diagnosed cases of oral squamous cell carcinoma. A significant increase in the frequency of micronuclei was observed in oral squamous cell carcinoma patients as compared to healthy controls. They concluded that there was a correlation of frequency of micronuclei and histopathological grading in oral squamous cell carcinoma suggesting that the micronuclear count can be used for grading of oral squamous cell carcinoma.
Saeed and Younis (2012) examined the micronuclear expression among heavy smokers, light smokers and non smokers and correlated it with oral health variables of these groups. The oral health status was evaluated by using the plaque, gingival, calculus indices in addition to the amalgam and composite restorations. The observed a statistically significant increase in the micronuclear expression from the non smoker group to the light smokers and the heavy smokers group. They also observed a strong correlation between the oral health status variables and the micronuclei expression in the non-smokers' group. They concluded that the micronuclear assay could be used as a useful biomarker to detect the people at high risk of oral mutations due to the harmful effect of the smoking.

Bansal, Sandhu, Bhandari and Sharma (2012) on their study to detect micronuclei in exfoliated buccal mucosal cells using Papanicolaou stain in 75 healthy individuals equally divided within individuals with habit of tobacco smoking, chewing and non tobacco users found out that mean value of MN was highest in smokeless tobacco (24.13 ±10.68) users followed by smokers (11.96±4.23) and non tobacco users (4.17±2.99). It was significantly higher in smokeless tobacco users than in smokers and controls. The study concluded that micronucleus assay could be used as a biomarker of genotoxicity.

Naderi, Darhadi and Sarshar (2012) on their study to evaluate the micronucleus assay of buccal mucosa cells in smokers who smoked less or more than 10 years found out that there was a significant difference (p<0.002) in the mean number of micronucleus of buccal mucosa cells between non smoker, smokers for less than and more than 10 years (0.94±0.94, 1.89±0.62 and 2.01±0.93 respectively). The study concluded that increasing the smoking duration could increase the frequency of micronucleus though not significant.

Chandirasekar et al., (2012) in their study to evaluate the genotoxic effects of tobacco use by analyzing the cytogenetic end points such as chromosome aberrations in peripheral blood and micronucleus in peripheral blood and buccal cells revealed that tobacco users displayed varied levels of elevated chromosomal damage and micronucleated cells than non tobacco users which was dependent on the duration of the tobacco use.
Caplash, Chaudhary and Kaur (2013) in their study to evaluate micronuclei in exfoliated buccal mucosal cells of individuals with tobacco chewing and/or smoking found that mean frequency of MN was significantly higher (p<0.05) in tobacco exposed individuals (0.76±0.74) compared to control group (0.03±0.06). They found that two confounding factors; age and duration of exposure to tobacco showed significant association with MN frequency.

Mohanta, Mohanty and Parida (2013) on their hospital based case-control study evaluated the genotoxicity of tobacco and alcohol on oral mucosal cells. They observed a significantly higher percentage of micronucleated cells in the chewer-smoker-alcoholics group as compared to only smokers group. They also observed a high percentage of micronuclei in the cancerous group than the single addicted groups suggesting the association with genetic susceptibility and modern lifestyle factors.

Kamath, Anigol and Setlur (2014) conducted a study to assess the micronuclear assay as a prognostic indicator in cytological smears. The study was conducted between two groups of smokers and non-smokers. They observed a significant increase in the number of micronuclei in the smokers group as compared to non smokers and suggested a positive correlation of occurrence of micronuclei with age; frequency and duration of smoking.

Khanna et al., (2014) evaluated and compared the frequency of micronuclei in smears of oral exfoliated cells among healthy controls, leukoplakia, and squamous cell carcinoma groups. They concluded that the micronuclei count was highest in the squamous cell carcinoma group followed by the leukoplakia group and lowest in the healthy controls group suggesting that the micronuclear assay can be used as a biomarker for cytogenetic damage in oral leukoplakia and oral squamous cell carcinoma.

Motgi et al., (2014) in their study to assess the cytogenic damage in form of micronuclei in oral epithelial cells in patients using smokeless and smoked form of tobacco and non tobacco users and its relevance to oral cancer included 100 patients each and buccal smears stained with Papanicolaou stain found no significant differences between the smokeless and smoked
tobacco groups, though higher in smokeless tobacco users. The micronuclei count was significantly lower in non tobacco users compared to tobacco users.

MR et al., (2014) on their study to compare the genotoxicity of different tobacco related habits evaluated micronuclei in exfoliated buccal epithelial cells. They found that the mean micronuclei count in individual with smoking habit were 3.11 which was 2.6 times more compared to normal controls. The study concluded that tobacco in any form is genotoxic especially the tobacco smokers and suggested the use of micronucleus assay as a reliable marker for genotoxic evaluation.

Chandirasekar et al., (2014) in their study to test whether smoking and smokeless tobacco affect the frequency of Comet assay and Micronuclei in peripheral blood lymphocytes and of MN in buccal epithelial cells and also to analyze specific genetic polymorphisms in smokers and smokeless tobacco users found out that MN frequency was higher (p<0.05) in all exposed subjects compared to controls. The individuals with both smoking and smokeless tobacco use showed higher numbers of micronucleated cells in their buccal mucosa and blood cells. They also stated that confounding factors such as age, exposure duration, and number of cigarettes might influence comet assay and micronuclei frequencies.