SYSTEMATIC REVIEW

Smokeless tobacco use and reproductive outcomes among women: a systematic review [version 1; peer review: 2 not approved]

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Abstract

Background: Both smoked and smokeless tobacco use have deleterious effects on most major organ systems including the reproductive system. We conducted a systematic review on smokeless tobacco (SLT) use and reproductive outcomes among women.

Methods: We searched Pubmed, ProQuest, Cochrane, Wiley and Emerald databases for studies involving smokeless tobacco use in women with any or a combination of three conditions:

infertility, menstrual disorders and pelvic inflammatory disease (PID). Eligibility criteria included

English language publications from 1st January 1990 - 31st October 2020. CADIMA software

used for filtering the studies and modified SIGN checklist for the quality assessment. The findings are reported as per the PRISMA guidelines. The AXIS and ROBIN E tool were used for assessment of risk of bias.

Results: In total, three studies addressed our research question. Two studies addressed infertility

(prospective cohort: n=501, cross sectional: n=192) of which, the cross-sectional study compared

the mean cotinine levels between those with infertility, menstrual disorders and PID. This study

also explored the association between SLT and PID. PID was the most common gynecological

complaint. Women with PID had significantly higher urinary cotinine levels = 24.95±12.259) ng/

ml (p=0.0144). Mean urinary cotinine in women with menstrual complaints was 19.32±10.29 ng/ml.

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Any reports and responses or comments on the article can be found at the end of the article.

The other study used population-based sampling of 501 couples who attempted pregnancy (enrolled in the LIFE Study).

Results showed that only 2% (n=28) of men and none of the women used smokeless tobacco.

Compared with never users of tobacco, smoking by females was individually associated with longer time-to-pregnancy; smoking among males remained significant when modeling partners together.

Conclusions: Available studies exploring associations between SLT and reproductive outcomes are inconclusive due to limitations in the study methodologies. More studies with robust study designs are required from low- and middle-income countries with high prevalence of SLT use.

Keywords

Smokeless Tobacco, infertility, menstruation, abnormal uterine bleeding, pelvic infections, reproductive health, women

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Introduction

Use of both smoked and smokeless forms of tobacco is a major cause of preventable morbidity and mortality. It kills half of all its lifetime users (WHO, 2011) and more than 8 million people each year, out of which, 1.2 million die due to second hand smoke (WHO, 2019). Toxic and carcinogenic chemicals in tobacco along with other ingredients that are added to them are known to be causally associated with several non-communicable diseases (NCDs) including cancer, especially oral cancer which is the leading cancer among men and the third most common cancer among women in India (Bhisey, 2012).

Smokeless tobacco (SLT) is “consumed without combustion at the time of use” (WHO, 2015). It is generally used orally (sucked, chewed, dipped or held in the mouth, used as dentifrice or toothpaste) or nasally resulting in nicotine absorption across the mucous membrane, along with other chemicals. Majority of SLT users, approximately 286 million people, live in low and middle-income countries in South-East Asia region. Three countries, namely India, Bangladesh, and Myanmar, host around 86% of the global users (NCI & CDC, 2014). As per Global Adult Tobacco survey 2016–17, women accounted for 2% among around 99.5 million adults current smokers. In contrast, 12.8% of women used SLT out of 199.4 million adults (GATS, 2017).

SLT use is addictive; it leads to oral health problems and plays a contributory role in the development of cardiovascular disorders, fatal ischemic heart disease, stroke, peripheral vascular diseases, peptic ulcers, type 2 diabetes, chronic rhinitis, foetal morbidity and mortality (WHO, 2015; Inamdar *et al.*, 2015; Suliankatchi and Sinha, 2016; Hossain *et al.*, 2017). The leading health consequences related to SLT use in Southeast Asia include cancers of numerous sites along with poor reproductive outcomes (World Health Organization. News release 11th Sept 2013).

Tobacco use in India is majorly considered a male-dominant behavior. However, over the past decade, the use of SLT products by Indian women is substantial and increasing, with adverse consequences for oral (Niaz *et al.*, 2017; Singh *et al.*, 2020) and perinatal health (Inamdar *et al.*, 2015; Suliankatchi and Sinha, 2016; Nair *et al.*, 2015). Women who use SLT are at risk of oral (Singh *et al.*, 2020) and pharyngeal cancers (Niaz *et al.*, 2017; Datta *et al.*, 2014; Sinha *et al.*, 2016; Awan and Patil, 2016), esophageal cancer (Niaz *et al.*, 2017; Datta *et al.*, 2014; Sinha *et al.*, 2016; Awan and Patil, 2016), cervical cancer, ischemic heart disease (IHD) (Sinha *et al.*, 2018) and osteoporosis (Ayo-Yusuf and Olutola, 2014). Compounds in SLT products such as nicotine act as neuro-teratogens as they can cross the placental barrier (Liao *et al.*, 2012) affecting the fetal development along with other pregnancy complications like pre-term delivery, low-birth weight (Inamdar *et al.*, 2015; Suliankatchi and Sinha, 2016) increased stillbirth risk (Hossain *et al.*, 2017) and risk of cancers in the developing fetus (Rogers *et al.*, 2009). However, there is little evidence that explores the association between SLT use and reproductive health of women. Therefore, we planned to systematically conduct a review on smokeless tobacco (SLT) use and reproductive health among women.

Methods

The protocol was registered on PROSPERO on 2nd October 2020 (CRD42020207176). This paper is reported in line with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines (Radhika, 2021).

We did an extensive literature search that described the association between SLT use and selected reproductive outcomes in women. For this, we selected three common reproductive outcomes, namely infertility, menstrual disorders and pelvic inflammatory disease. The search terms revolved around P (population): women, E (exposure): exposure to SLT, C (control): women who were not exposed to SLT, O (outcome): reproductive outcomes in women that included infertility, menstrual disorders, and pelvic inflammatory disorder (PID). The reference period was from 01 January 1990 to 31st October 2020.

Eligibility criteria

Publications including reviews, original trials and conference proceedings documenting any form of SLT use along with the three selected reproductive outcomes in English language within the timeframe were considered for the inclusion.

Information sources

Studies were identified by searching electronic databases (Pubmed, Wiley, Cochrane Library, Emerald and ProQuest) in addition to scanning reference lists of articles using the chosen keywords for all three reproductive outcomes separately.

Search strategy

Search strings used for infertility among women and SLT were: (Smokeless OR Chew OR Chewing OR Dipping OR Snuff OR toothpaste OR dentifrice OR paan masala OR betel quid OR betelquid OR areca nut OR arecanut) AND (Tobacco OR Tobaccos OR Gutka OR Gutkas OR Snuff OR Mint) AND (Female OR Women) AND (Infertility OR Subfertility OR Sub Fertility OR sterility OR Infertile OR sterile).

Search strings used for Menstrual disorders and SLT were: (Smokeless OR Chew OR Chewing OR Dipping OR Snuff OR toothpaste OR dentifrice OR paan masala OR betel quid OR betelquid OR areca nut OR arecanut) AND (Tobacco OR Tobaccos OR Gutka OR Gutkas OR Snuff OR Mint) AND (Female OR Women) AND (menstrual OR menstruation OR bleeding OR menorrhagia OR Hypermenorrhea OR Hypermenorrhoea OR Hypomenorrhea OR Hypomenorrhoea OR dysmenorrhea OR dysmenorrhea OR Abnormal uterine bleeding OR amenorrhea OR menometrorrhagia OR metrorrhagia OR dysfunctional uterine bleeding).

For PID and SLT use, search strings used were (Smokeless OR Chew OR Chewing OR Dipping OR Snuff OR toothpaste OR dentifrice OR paan masala OR betel quid OR areca nut) AND (Tobacco OR Tobaccos OR Gutka OR Gutkas OR Snuff OR Mint) AND (Female OR Women) AND (infection OR inflammation OR pelvic inflammatory disease OR PID OR genital infections OR genital tuberculosis OR tubo-ovarian mass OR Salpingitis OR salpingo-oophoritis OR endometritis OR adnexitis OR parametritis).

Study selection

We included studies irrespective of sample size. After duplicate removal by CADIMA, every record was screened by two reviewers independently using the title. Those accepted by both were subjected to abstract review. In case of any disagreement, arbitration was done by a senior member from the team. For the selected abstracts, full articles were obtained, and quality check was performed by two reviewers independently using the modified SIGN checklist. Those selected at this stage were eligible for the review. The study selection is mentioned according to reproductive outcomes selected and reported according to PRISMA guidelines. The AXIS tool was used to assess risk of bias (Ma *et al.*, 2020) for cross sectional study and the ROBIN E tool was used for prospective studies.

Data items and data collection process

Full text appraisal for study selection was done by two authors independently. Title & abstract filtering was done with CADIMA. Information was extracted from all the eligible studies on a predesigned format (Tables 2–4) including a range of study variables relating to the design, objectives, and outcomes. For each of the reproductive outcomes, studies included were summarized separately.

Results

Search for infertility among women and SLT use resulted in 1093 results which were run through CADIMA for removal of the duplicate studies, which gave a final of 1062 results (9 from Pubmed; 44 from Wiley; 6 from Emerald; 11 from Cochrane and 992 from ProQuest). that for Menstrual disorders and SLT gave 1330 results. These results were run through CADIMA for removal of the duplicate studies, which gave a final of 1294 results (44 from Pubmed; 19 from Wiley; 25 from Cochrane and 1206 from ProQuest). For PID and SLT use, there were 3929 results. These results were run through CADIMA for removal of the duplicate studies, which gave a final of 3808 results (205 from pubmed; 27 from wiley; 46 from Cochrane and 3530 from proquest) (Table 1).

The search revealed a total of two studies (prospective cohort, n = 501 and cross sectional, n = 192) addressing our research question related to infertility (Figure 1, Table 2).

The prospective cohort study used population-based sampling with 501 couples who attempted pregnancy in Michigan and Texas, 2005–2009 (enrolled in the LIFE Study). Results showed that only 2% (n = 28) of men were SLT users and no women used smokeless tobacco. Smokers showed higher cadmium levels than SLT, adjusted for cadmium attenuated the cigarette–time-to-pregnancy (TTP) association, especially among women. Shorter TTP was observed among SLT users in comparison to smokers (FOR [fecundability odds ratio]: 2.86, 95% confidence interval [CI]: 1.47, 5.57). Compared

Table 1. Number of search results from each database and dates of search performed.

Reproductive outcomes among women		Pubmed	Wiley	Emrald	Cochrane	Proquest	Total
Infertility	No. of results	9	47	6	11	1020	1093
	Date	10-11-20	11-11-20	11-11-20	12-11-20	15-11-20	
Menstruation	No. of results	46	26	0	25	1233	1330
	Date	11-11-20	12-11-20	12-11-20	13-11-20	13-11-20	
PID	No. of results	211	37	0	46	3635	3929
	Date	11-11-20	12-11-20	12-11-20	13-11-20	25-11-20

Figure 1. Study selection for smokeless tobacco (SLT) use and infertility among women. IVF = in vitro fertilization.

Table 2. Data extraction sheet for smokeless tobacco (SLT) use and infertility. TTP = time-to-pregnancy; FOR = fecundability odds ratio; OR = odds ratio; CI = confidence interval.

S. no.	4I	6I
Title	Time-to-Pregnancy Associated with Couples' Use of Tobacco Products	Assessment of urinary cotinine levels in women with gynecological complaints at a tertiary care hospital: A pilot study
Authors	Katherine J. Sapra, Dana B. Barr, José M. Maisog, Rajeshwari Sundaram, Germaine M. Buck Louis	A. G. Radhika, Sruthi Bhaskaran, Jagdish Kaur, Anshuja Singla, Tusha Sharma, B. D. Banerjee
Year of Publication	2016	2017
Country	USA	India
Study objective	<ol style="list-style-type: none"> 1. To assess the relationship between couples' preconception tobacco use (smokeless and combustible) and prospectively observed TTP. 2. To evaluate blood heavy metal and serum cotinine concentrations among various tobacco product users and nonusers to determine if specific chemicals may contribute to changes in TTP 	To evaluate the urinary cotinine levels in women of reproductive age with common gynecological conditions (PID, infertility, and AUB).
Study design	Prospective cohort	Cross-sectional pilot

Table 2. *Continued*

S. no.	4I	6I
Study setting (hospital/ community)	Community	Hospital
Study population	Population-based sampling, 501 couples who discontinued contraception for becoming pregnant in 16 counties in Michigan and Texas, 2005–2009, enrolled in the LIFE Study. Inclusion criteria - Both partners spoke English or Spanish, were discontinuing contraception to attempt pregnancy or were off contraception not more than 2 months, males aged 18 years and above, females aged 18–40 years, had menstrual cycle length of 21–42 days, and had not received injectable contraception in the past year.	Women presenting at gynecological outpatient clinic of GTB hospital among non-pregnant women of reproductive age having any of the complaints namely menstrual disturbance, infertility or pelvic inflammatory disease. Sample size 192.
Primary exposure	lifetime and current use of tobacco in any form - cigarette, cigar, and chew/snuff (smokeless) use	tobacco use or exposure (any form) in self or family members
Method of ascertainment	Questionnaire administered by strained study personnel, fertility monitor, Regular use of highly sensitive home pregnancy tests and blood samples for quantification of heavy metals and cotinine	Interview/questionnaire, urinary cotinine (spot urine test)
Secondary exposure(s)	None	Secondhand Exposure to smoked tobacco
Method of ascertainment	NA	Interview/questionnaire, urinary cotinine (spot urine test)
Intervention (if applicable)	None	None
Control (if applicable)	never users (participants who never used any tobacco product)	None
Primary outcome	11% of females and 15% of males smoked cigarettes. Among men, 14% smoked cigars, 9% used snuff, and 2% used chew. Cadmium levels were higher in smokers than smokeless tobacco and never users; adjusting for cadmium attenuated the cigarette–TTP association, particularly among women. TTP was shorter among smokeless tobacco users relative to smokers (FOR: 2.86, 95% CI: 1.47, 5.57).	Assessment of urinary cotinine: The mean urinary cotinine level in women exposed to SHS was 23.82 ± 12.67 ng/ml. PID was the most common gynecological complaint. Women with PID had significantly higher urinary cotinine levels compared to those with Mean urinary cotinine levels in infertile women were 22.42 ± 12.72 ng/ml
Secondary outcome(s)	Compared with never tobacco users, male (FOR: 0.41, 95% CI: 0.24, 0.68) and female (FOR: 0.53, 95% CI: 0.33, 0.85) smoking were individually associated with longer TTP; males' smoking remained significant (FOR: 0.46, 95% CI: 0.27, 0.79) when modeling partners together.	NA
Sample size (group1)	293 couples	192
Sample size (group2)	208 couples	NA
Sampling strategy	Participants were selected from marketing databases (Michigan) or state wildlife and fish registry (Texas)	Urinary cotinine levels in three common categories of gynecological complaints, among non-pregnant women of reproductive age residing in East Delhi who sought treatment from gynecological outpatient clinic of Guru Tegh Bahadur Hospital. The hospital caters to patients living in urban slums in NandNagri, Seema Puri, and Seelampur. The population density in these areas is very high, with congested living conditions. Majority belong to low-income group.

Table 2. *Continued*

S. no.	4I	6I
Co variates/ confounders	Age, race/ethnicity, educational attainment, income, and alcohol and caffeine use. Height, weight, Metals with significantly different blood level concentrations across tobacco types.	Secondhand smoking
Biases	Sampling bias (only 28 men smokeless tobacco users, no women used smokeless tobacco)	Sample not enough and other confounding variables not considered; None of the participants admitted to smoking or use of SLT.
Measure of association (OR/RR)	OR (adjusted Fecund ability Odds Ratio); FOR: 2.86, 95%	Mean±SD
Lower bound	1.47	Infertility – 9.70 Menstrual disorders – 9.03 PID – 12.96
Upper bound	5.57	Infertility – 35.14 Menstrual disorders – 29.61 PID – 37.21
Sub group analysis	None	None
Results of sub group analysis	NA	NA

with never users of tobacco, smoking by females (FOR: 0.53, 95% CI: 0.33, 0.85) was individually associated with longer TTP; for males, smoking remained significant (FOR: 0.46, 95% CI: 0.27, 0.79) when modeling partners together (Sapra *et al.*, 2016).

A cross sectional study carried out in India to evaluate the urinary cotinine levels in three common categories of gynecological complaints, among 192 women of reproductive age, who were not pregnant, and sought treatment from a Government Medical college. Results showed that mean urinary cotinine level in women exposed to secondhand smoking (SHS) was 23.82 ± 12.67 ng/ml. PID was the most common gynecological complaint. Mean urinary cotinine levels in infertile women were 22.42 ± 12.72 ng/ml. The limitations of this study were that the sample size was not enough, other confounding variables were not considered and none of the participants admitted to smoking or use of SLT (Radhika *et al.*, 2017).

The same study found that mean urinary cotinine levels in women with menstrual complaints was 19.32 ± 10.29 ng/ml. Out of 1330 articles obtained on initial search, this was the only study selected in our review to study the association between SLT and menstrual problems (Figure 2, Table 3). For the question related to PID, another study (Simen-Kapeu *et al.*, 2009) in addition to this study was selected (Figure 3, Table 4). Women with PID had significantly higher urinary cotinine levels = $24.95 (\pm 12.259)$ ng/ml ($p = 0.0144$) (Radhika *et al.*, 2017).

Another study (Simen-Kapeu *et al.*, 2009) compared the association between tobacco use (smoking and chewing) and the risk of multiple human papillomavirus (HPV) infections and cervical squamous intraepithelial lesions (SILs) in two populations with different exposure. For this, baseline data from 2144 women from Cote d'Ivoire, West Africa and 415 women from Finland, Northern Europe regarding cervical screening, HPV positivity and tobacco use (smoking and chewing habits) was re-analyzed to determine the association between tobacco chewing in Cote d'Ivoire and tobacco smoking in Finland and the age stratified risk of multiple HPV infections and cervical SIL. Results show that in Côte d'Ivoire tobacco chewing (2.6%) was more common than tobacco smoking (1.4%). In 236 cases (eligible women with SIL), mean age of the women was 28.4 ± 6.6 years with low-grade SIL in 165 and high-grade SIL in 71. Tobacco users (smokers and chewers) showed an increased risk of LSIL. Tobacco chewers were at 5 times higher risk for HSIL in both younger age group (<30 years) with OR: 5.5, 95% CI: 1.2-26 and older age group (≥ 30 years of age) with OR: 5.5, 95% CI: 2.1-14) in comparison to non-chewers. Age-adjusted OR of cervical HSIL was significantly higher among tobacco chewers. Increased risk of LSIL and HSIL (not significant) was found in HPV positive women ≥ 30 years of age who were actively exposed to tobacco through smoking or chewing was seen.

Having multiple HPV infections was common in HPV16 and/or HPV18 infected women (60.4% in Finland and 47.2% in Côte d'Ivoire). There was no increased risk of multiple HPV infections among tobacco consumers. It was found that

Figure 2. Study selection for smokeless tobacco (SLT) use and menstrual disorders

Table 3. Data extraction matrix for smokeless tobacco (SLT) use and menstrual problems. PID = pelvic inflammatory disease. SD = standard deviation.

Study ID	13M
Name of the study	Assessment of urinary cotinine levels in women with gynecological complaints at a tertiary care hospital: A pilot study
Authors	A. G. Radhika, Sruthi Bhaskaran, Jagdish Kaur, Anshuja Singla, Tusha Sharma, B. D. Banerjee
Year of Publication	2017
Country	India
Study objective	To evaluate the urinary cotinine levels in women of reproductive age with common gynecological conditions (PID, infertility, and AUB).
Study design	Cross-sectional pilot
Study setting (hospital/ community)	Hospital
Study population	Women presenting at gynecological outpatient clinic of GTB hospital among non-pregnant women of reproductive age having any of the complaints namely menstrual disturbance, infertility or pelvic inflammatory disease. Sample size 192.
Primary exposure	Tobacco use or exposure (any form) in self or family members
Method of ascertainment	Interview/questionnaire, urinary cotinine (spot urine test)
Secondary exposure(s)	Secondhand exposure to smoked tobacco
Method of ascertainment	Interview/questionnaire, urinary cotinine (spot urine test)
Intervention (if applicable)	None

Table 3. *Continued*

Study ID	13M
Control (if applicable)	None
Primary outcome	Assessment of urinary cotinine: The mean urinary cotinine level of women exposed to SHS was 23.82 ± 12.67 ng/ml. Women with menstrual complaints 19.32 ± 10.29 ng/ml, $P = 0.029$
Secondary outcome(s)	NA
Sample size (group1)	192
Sample size (group2)	NA
Sampling strategy	Urinary cotinine levels in three common categories of gynecological complaints, among non-pregnant women of reproductive age residing in East Delhi who sought treatment from gynecological outpatient clinic of Guru Tegh Bahadur Hospital. The hospital caters to patients living in urban slums in NandNagri, Seema Puri, and Seelampur. The population density in these areas is very high, with congested living conditions. Majority belongs to low-income group. Consecutive, spot urine sample.
Co variates/confounders	Second-hand smoking
Biases	Patients presenting to the hospital with specific gynecological complaints. Sample not enough and other confounding variables not considered
Measure of association (OR/RR)	Mean $\pm$ SD
Lower bound	Infertility – 9.70 Menstrual disorders – 9.03 PID – 12.96
Upper bound	Infertility – 35.14 Menstrual disorders – 29.61 PID – 37.21
Sub group analysis	None
Results of sub group analysis	NA

Figure 3. Study selection for smokeless tobacco (SLT) use and pelvic inflammatory disease (PID) among women.

Table 4. Data extraction matrix for smokeless tobacco (SLT) use and pelvic inflammatory disorder. HPV = human papillomavirus; OR = odds ratio; CI = confidence interval; LSIL = low-grade squamous intraepithelial lesion; HSIL = high-grade squamous intraepithelial lesion.

Study ID	426P	3697P	
Name of the study	Assessment of urinary cotinine levels in women with gynecological complaints at a tertiary care hospital: A pilot study	Tobacco smoking and chewing as risk factors for multiple human papillomavirus infections and cervical squamous intraepithelial lesions in two countries (Côte d'Ivoire and Finland) with different tobacco exposure.	
Authors	A. G. Radhika, Sruthi Bhaskaran, Jagdish Kaur, Anshuja Singla, Tusha Sharma, B. D. Banerjee	Aline Simen- Kapeu, Guy La Ruche, VesaKataja, MerjaYliskoski, Christine Bergeron, Apollinaire Horo, Kari Syrjanen, Seppo Saarikoski, Matti Lehtinen, Francois Dabis, Annie J. Sasco	
Year of Publication	2017	2008	
Country	India	Cote d'Ivoire	Finland
Study objective	To evaluate the urinary cotinine levels in women of reproductive age with common gynecological conditions (PID, infertility, and AUB).	To compare the association between tobacco smoking and chewing and the risk of multiple human papillomavirus infections and cervical squamous intraepithelial lesions (SILs) in two populations with different exposure.	
Study design	cross-sectional pilot	Comparative	
		(Prospective follow up study)	(long term prospective study)
Study setting (hospital/ community)	Hospital	Hospital	
Study population	Women presenting at gynecological outpatient clinic of GTB hospital among non-pregnant women of reproductive age having any of the complaints namely menstrual disturbance, infertility or pelvic inflammatory disease. Sample size 192.	2144 women attending 3 outpatient gynecology clinics of Abidjan, age - 20 to 50 years, no h/o lower genital tract neoplasias and absence of active pregnancy.	415 Finnish women - Kuopio University Hospital between 1981 and 1998; 15-69 years of age attending municipal health centers in Kuopio province.
Primary exposure	tobacco use or exposure (any form) in self or family members	Actively exposed to tobacco (any form - smoking and/or chewing)	Actively exposed to tobacco (any form - smoking and/or chewing)
Method of ascertainment	Interview/questionnaire, urinary cotinine (spot urine test)	Interview and Cytological diagnosis - cervical specimen for HPV detection - collected with a plastic brush (Viba-Brush, CML, Nemours, France - using PCR for HPV positive	Interview and gynecological examination - colposcopy, repeat PAP smear and directed punch biopsy; formalin fixed, paraffin embedded biopsies analyzed using DNA in situ hybridization (ISH) technique.
Secondary exposure(s)	Secondhand Exposure to smoked tobacco	None	None
Method of ascertainment	Interview/questionnaire, urinary cotinine (spot urine test)	NA	NA
Intervention (if applicable)	None	NA	NA
Control (if applicable)	None	women with absence of cervical SIL	Normal cervical smears at the first study visit

Table 4. Continued

Study ID	426P	3697P	
Primary outcome	PID (n=139) most common gynecological complaint. Assessment of urinary cotinine Mean urinary cotinine levels in women exposed to environmental tobacco smoke (SHS) was 23.82 ± 12.67 ng/ml. Women with PID had significantly higher urinary cotinine levels = $24.9548 (\pm 12.259)$ ng/ml ($p=0.0144$) unpaired student t test	236 cases; mean age 28.4 ± 6.6 ; LSIL (n=165), HSIL (n=71). Tobacco users (smokers and chewers) - increased risk of LSIL; Tobacco chewers, 5 times higher risk in both younger (OR: 5.5, 95% CI: 1.2-26) and older (OR: 5.5, 95% CI: 2.1-14) in comparison to non-chewers. Age-adjusted OR of cervical HSIL significantly high among tobacco chewers.	88 cases; mean age 27.9 ± 7.5 ; LSIL (n=60), HSIL (n=28). Tobacco users (smokers and chewers)- increased risk of LSIL; Tobacco smokers; LSIL - younger (OR:1.2, 95% CI: 0.6-2.5) and older (OR:2.5, 95% CI: 0.9-6.8); HSIL - younger (OR:1.1, 95% CI: 0.4-3.1) and older (OR:2.2, 95% CI: 0.5-8.7) in comparison to non-chewers.
Secondary outcome(s)	NA	chlamydia trachomatis detection	chlamydia trachomatis detection, serum cotinine detection
Sample size (group1)	192	236	88
Sample size (group2)	NA	1908	327
Sampling strategy	Urinary cotinine levels in three common categories of gynecological complaints, among non-pregnant women of reproductive age residing in East Delhi who sought treatment from gynecological outpatient clinic of Guru Tegh Bahadur Hospital. The hospital caters to patients living in urban slums in Nand Nagri, Seema Puri, and Seelampur. The population density in these areas is very high, with congested living conditions. Majority belongs to low-income group. Consecutive, spot urine sample.	2144 out of 2198 women attending 3 outpatient gynecology clinics of Abidjan in the time frame April 1995 to February 1996, age - 20 to 50 years, no h/o lower genital tract neoplasias and absence of active pregnancy	415 Finnish women - Kuopio University Hospital between 1981 and 1998; 15-69 years of age attending municipal health centers in Kuopio province (cervical swab taken as part of their contraception counselling)
Co variates/ confounders	Secondhand smoking	None	None
Biases	Patients presenting to the hospital with specific gynecological complaints. Sample not enough and other confounding variables not considered	Sampling bias: very few Ivorian women reported smoking evaluation, for this habit alone, in the regression analysis was restricted to Finnish women.	Sampling bias: very few Ivorian women reported smoking evaluation, for this habit alone, in the regression analysis was restricted to Finnish women.
Measure of association (OR/RR)	Mean $\pm$ SD	Tobacco chewers, 5 times higher risk in both younger (OR: 5.5, 95% CI: 1.2-26) and older (OR: 5.5, 95% CI: 2.1-14) in comparison to non-chewers.	OR Tobacco smokers; LSIL - younger (OR:1.2, 95% CI: 0.6-2.5) and older (OR:2.5, 95% CI: 0.9-6.8); HSIL - younger (OR:1.1, 95% CI: 0.4-3.1) and older (OR:2.2, 95% CI: 0.5-8.7) in comparison to non-chewers.
Lower bound	Infertility - 9.70 Menstrual disorders - 9.03 PID - 12.96	Younger - 1.2 Older - 2.1	LSIL younger - 0.6 older - 0.9 HSIL younger - 0.4 older - 0.5
Upper bound	Infertility - 35.14 Menstrual disorders - 29.61 PID - 37.21	Younger - 26 Older - 14	LSIL younger - 2.5 older - 6.8 HSIL younger - 3.1 older - 8.7

Table 4. *Continued*

Study ID	426P	3697P	
Sub group analysis	None	Analysis LSIL and HSIL in HPV positive women was done - age and tobacco exposure being the variables	Analysis LSIL and HSIL in HPV positive women was done - age and tobacco exposure being the variables
Results of sub group analysis	NA	Increased risk of LSIL and HSIL (not significant) in HPV positive women ≥ 30 years of age and actively exposed to tobacco through smoking or chewing)	Increased risk of LSIL and HSIL (not significant) in HPV positive women ≥ 30 years of age and actively exposed to tobacco through smoking or chewing)

women ≥ 30 years of age exposed to tobacco through smoking in Finland (OR: 2.2, 95% CI: 0.5-8.7) and chewing in Côte d'Ivoire (OR: 5.5, 95% CI: 2.1-14) had a moderately or highly increased risk of high-grade SIL, respectively. In the latter, the risk was statistically significant. Sampling bias was seen in the study as very few Ivorian women reported smoking evaluation, for this habit alone, and the regression analysis was restricted to Finnish women (Simen-Kapeu *et al.*, 2009).

The risk of bias assessed using appraisal tool for cross sectional studies (AXIS) for one and ROBIN E for observational studies. Risk of bias for cross sectional study was high due to the small sample size but the those for the prospective studies was low.

Discussion

This systematically conducted rapid review to study association between SLT use and reproductive health of women yielded a total of three studies with findings from four different countries, namely India, USA, Finland and Cote d'Ivoire and Finland. The differences in the study methodologies precluded us from combining the study findings.

Infertility was measured in terms of TTP and Urinary cotinine levels and both were seen to be higher in SLT users. Women with PID had highest mean urinary cotinine levels among the three, followed by infertility and menstrual complaints respectively (Radhika *et al.*, 2017). Another study showed longer TTP for cotinine levels more than 10ng/ml for SLT users (Sapra *et al.*, 2016). Another study comparing association between tobacco smoking and chewing with risk of PID in two populations with different exposures showed that SLT users were at a five times higher risk of SIL in comparison to non-chewers irrespective of age (Simen-Kapeu *et al.*, 2009). Results were however inconclusive regarding strong associations between SLT use and reproductive health in women.

Biologic fertility can be assessed using TTP. A study based in a community setting showed a remarkably comprehensible association between female smoking and sub-fecundity during the most recent waiting TTP (Kassa and Kebede, 2018).

Cigarette smoking was also seen to be associated with an adverse effect on ovarian function and hence on fertility among women. Evidence suggests that there is an association between cigarette smoking and reduced fertility (risk of delayed conception), even at low doses (Hatch *et al.*, 2012; Gormack *et al.*, 2015). Since the active metabolite is similar, it is natural to expect some effects on the reproductive outcomes i.e. menstrual function, infertility, PID and pregnancy outcomes with SLT use. However, there is a dearth of good quality studies to ascertain such an association.

Nicotine levels are found to be highest in bidis, followed by chewed tobacco and cigarettes (Amith *et al.*, 2018). Nevertheless, SLT products are more often abused than smoked tobacco products (Sharma *et al.*, 2015) and serum nicotine levels remain in significant amounts for a longer time period (Mala *et al.*, 2016). Like smoked form, SLT use causes alteration of the immune response to infections and has a damaging effect on majority of organ systems in the body including the reproductive system (Willis *et al.*, 2012). This justifies the biological plausibility between SLT and reproductive outcomes.

To conclude, the number of studies about an association between SLT use and reproductive health of women were very few despite the high prevalence of use in the South East Asia region. Impact of SLT on reproductive health of women requires more research. In this rapid review, we followed the principles of systematic review that offers strength to the study. We have considered the major reproductive outcomes comprehensively. However, we restricted our search to English papers that poses a limitation. In addition, our reference period was from 1990, though it possibly would not have made a difference even if we went beyond that timeline. Though our results show that there might be an association

between the SLT and poor reproductive outcomes, we recommend more studies on this topic with robust study designs for conclusive results.

Data availability

Underlying data

All data underlying the results are available as part of the article and no additional source data are required.

Reporting guidelines

Figshare: PRISMA checklist for ‘Smokeless tobacco use and reproductive outcomes among women: a systematic review’. <https://doi.org/10.6084/m9.figshare.16819102> (Radhika, 2021).

Data are available under the terms of the [Creative Commons Zero “No rights reserved” data waiver](#) (CC0 1.0 Public domain dedication).

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Sonali Jhanjee

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The authors use the term reproductive outcomes in the title and then use the terms “reproductive outcomes” and “reproductive health” interchangeably throughout the review. It may be beneficial to use standardized definitions in the review. Rationale also needs to be provided why these 3 particular outcomes namely infertility, menstrual disorders, and pelvic inflammatory disease have been the focus of the review rather than the more commonly reported reproductive outcomes low birth weight, stillbirth, spontaneous abortion, preterm birth and so on in studies related to smokeless tobacco.

In two of the studies, included in the review, the women participants were not consuming smokeless tobacco which is the focus of the review. So the authors need to clarify whether these studies fulfilled their inclusion and exclusion criteria.

All the above points have a bearing on the conclusions drawn from the review and hence need to be addressed.

The discussion also focuses more on smoking rather than smokeless tobacco.

Are the rationale for, and objectives of, the Systematic Review clearly stated?

Partly

Are sufficient details of the methods and analysis provided to allow replication by others?

Partly

Is the statistical analysis and its interpretation appropriate?

Partly

Are the conclusions drawn adequately supported by the results presented in the review?

Partly

Competing Interests: No competing interests were disclosed.

I confirm that I have read this submission and believe that I have an appropriate level of expertise to state that I do not consider it to be of an acceptable scientific standard, for reasons outlined above.

Reviewer Report 05 January 2022

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Ruchika Gupta

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The objective of the systematic review, as stated by the authors, is to review the association of SLT use and reproductive health among women. The methodology, search strings, eligibility criteria are all suitable to the objective. However, I have major concerns about the results.

1. What were the exclusion criteria? If the E in PECO is an exposure to SLT, then studies reporting on smoking or secondhand smoke should be excluded from the review.
2. Of the three studies that the authors claim to have 'addressed' their research question, the study from India addresses secondhand smoke. Neither of the participants admitted to smoking or SLT use. Hence, this study does not fulfill the eligibility criteria. In the report from Michigan and Texas, none of the female participants used SLT while only a small fraction of males consumed SLT in the form of snuff or chewing products. A close review of their results shows that impact of SLT on infertility is compared to smokers but not to never-users (could have been insignificant due to the low numbers of SLT users). Hence, this study also, in my opinion is not worth being included in the review.

That leaves only one study from Cote d'Ivoire, West Africa and Finland, Northern Europe that evaluated the risk of HPV and SIL in tobacco users, including SLT users.

3. The discussion section also pertains to smoking rather than SLT which is the actual topic of the review.
4. When talking of reproductive outcomes, why did the authors not include pregnancy outcomes like prematurity, stillbirth, low birth weight etc.?

Are the rationale for, and objectives of, the Systematic Review clearly stated?

Yes

Are sufficient details of the methods and analysis provided to allow replication by others?

Yes

Is the statistical analysis and its interpretation appropriate?

No

Are the conclusions drawn adequately supported by the results presented in the review?

No

Competing Interests: No competing interests were disclosed.

Reviewer Expertise: Smokeless tobacco and health impact

I confirm that I have read this submission and believe that I have an appropriate level of expertise to state that I do not consider it to be of an acceptable scientific standard, for reasons outlined above.

Comments on this article

Version 1

Author Response 18 Apr 2022

A.G. Radhika, University College of Medical Sciences & Guru Teg Bahadur Hospital, Delhi, India

Response to reviewers comments

Reviewer 1

1. What were the exclusion criteria? If the E in PECO is an exposure to SLT, then studies reporting on smoking or secondhand smoke should be excluded from the review.

Response: The query is well placed. Exposure to SLT is only qualified for inclusion.

2. Of the three studies that the authors claim to have 'addressed' their research question, the study from India addresses secondhand smoke. Neither of the participants admitted to smoking or SLT use. Hence, this study does not fulfill the eligibility criteria. In the report from Michigan and Texas, none of the female participants used SLT while only a small fraction of males consumed SLT in the form of snuff or chewing products. A close review of their results shows that impact of SLT on infertility is compared to smokers but not to never-users (could have been insignificant due to the low numbers of SLT users). Hence, this study also, in my opinion is not worth being included in the review.

Response: Thanks for this important observation.

1. The study from India found that even though none of the patients consented to use SLT, the urinary cotinine levels were quite high ("Urinary cotinine levels in women not exposed to SHS (162/192) ranged widely from 8.09 to 63.44 ng/ml with a mean of 23.60 * 12.00 ng/ml"). Following secondhand exposure to tobacco smoke, cotinine levels rarely exceed 10 ng/ml. In addition, patients' histories of addictions can be very inconsistent and unreliable. Therefore, the study was included in the review.
2. There are few studies on SLT and infertility, and their sample sizes are small. Despite its small numbers, the result was included for review due to its importance

That leaves only one study from Cote d'Ivoire, West Africa and Finland, Northern Europe that evaluated the risk of HPV and SIL in tobacco users, including SLT users.

3. The discussion section also pertains to smoking rather than SLT which is the actual topic of the review.

Response: Thanks for the suggestion, it has now been revised.

4. When talking of reproductive outcomes, why did the authors not include pregnancy outcomes like prematurity, stillbirth, low birth weight etc.?

Response: Yes, this aspect was considered when planning the study. We planned that gynecological aspects can be emphasized in the present review. Pregnancy outcomes will be examined in another study.

Are the rationale for, and objectives of, the Systematic Review clearly stated?

Yes

Are sufficient details of the methods and analysis provided to allow replication by others?

Yes

Is the statistical analysis and its interpretation appropriate?

No

Response: We request to have better clarity please specially the specific point of concern.

Are the conclusions drawn adequately supported by the results presented in the review?

No

Response: It has been revised.

Reviewer 2

Are the rationale for, and objectives of, the Systematic Review clearly stated?

Partly

Are sufficient details of the methods and analysis provided to allow replication by others?

Partly

Is the statistical analysis and its interpretation appropriate?

Partly

Are the conclusions drawn adequately supported by the results presented in the review?

Partly

Competing Interests

No competing interests were disclosed.

Responses to Reviewer 1 apply to the observations noted by Reviewer 2

Competing Interests: None

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