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GASTROINTESTINAL DISEASES

Use of tobacco products and gastrointestinal morbidity: an endoscopic population-based study (the Kalixanda study)

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Abstract The impact of snus (smokeless tobacco or snuff) on gastrointestinal symptoms and pathological findings is largely unknown. The authors aimed to investigate whether the exposure to different forms of tobacco influences upper gastrointestinal symptoms, histology and frequency of *Helicobacter pylori* infection. A random sample ($n = 2{,}860$) of the adult population of two northern Swedish municipalities Kalix and Haparanda ($n = 21{,}610$) was surveyed between December 1998 and June 2001 using a validated postal questionnaire assessing gastrointestinal symptoms (response rate 74.2%, $n = 2{,}122$) (The Kalixanda Study). A random sub-sample ($n = 1{,}001$) of the responders was invited to undergo an esophagogastroduodenoscopy (participation rate 73.3%) including biopsies, *Helicobacter pylori* culture and serology and symptom assessment and exploration of present and past use of tobacco products. No symptom groups were associated with snus use. Snus users had a significantly higher prevalence of macroscopic esophagitis univariately but snus use was not associated with esophagitis in multivariate analysis. Snus use was associated with basal cell hyperplasia (OR = 1.74, 95% CI: 1.02, 3.00) and with elongation of papillae (OR = 1.79, 95% CI: 1.05–3.05) of the squamous epithelium at the esophago-gastric junction. Current smoking cigarettes was associated with overall peptic ulcer disease (OR = 2.32, 95% CI: 1.04, 5.19) whereas snus use was not. There were no significant association between current *Helicobacter pylori* infection and different tobacco product user groups. Snus significantly alters the histology of the distal esophagus but does not impact on gastrointestinal symptoms or peptic ulcer disease.

Keywords Dyspepsia · Esophagitis · Gastroesophageal reflux symptom · Peptic ulcer disease · Smoking · Snus

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Abbreviations

ASQ	Abdominal symptom questionnaire
BMI	Body mass index
CI	Confidence interval
EGD	Esophagogastroduodenoscopy
GERD	Gastroesophageal reflux disease
GERS	Gastroesophageal reflux symptom
IBS	Irritable bowel syndrome
OR	Odds ratio
PUD	Peptic ulcer disease
SD	Standard deviation

Introduction

Smokeless tobacco or moist snuff, snus in this study, is widely used in Sweden (900,000 users, 21% of males and 4% of females, while 13% of males and 17% of females smoke cigarettes) [1] and also in other Nordic countries. There is a widespread use in India and the marketing is intense, while USA is the biggest market of snus in the world with approximately 12 million users [2, 3]. Results of a national US survey show that nearly 9% of male and 0.4% of the female college students used snus [4]. In Sweden marketing efforts have resulted in higher use among young males and females have tripled their use over 8 years [1].

Swedish snus products are predominantly finely ground moistened tobacco with an alkaline pH between 7.9 and 8.6 [5]. A pinch of approximately 1–2 g is placed under the upper lip, and some nicotine is absorbed through the buccal mucosa, while some is swallowed together with the saliva [2].

Epidemiological and experimental studies have shown that smoking leads to harmful effects on the gastric mucosa [6–9]. The mechanisms, however, are unclear and the effects might be mediated by inhaled nicotine, as well as by other chemical contents of tobacco. Many studies provide conflicting results but the overall evidence supports the hypothesis that nicotine per se is harmful to the gastric mucosa [7, 9].

There is little knowledge regarding the relationship between gastrointestinal symptoms and snus. The nicotine intake is usually higher than in smokers, and a great deal of tobacco juice contaminated saliva is swallowed during use. In a Swedish cross-sectional study of symptoms in 130,000 construction workers, smokers reported "ulcer-like" dyspepsia three times as often as the non tobacco users, while snus users reported significantly fewer symptoms than both smokers and non tobacco users [10].

It has been shown both in studies on patients and in population-based studies that smoking is a risk factor for peptic ulcer disease (PUD) [8, 11], but there are no data concerning snus on this issue. Data from the US show that cigarette smokers have a markedly increased risk for gastric cancer and that use of more than one tobacco product increases the risk in men [12]. Male users of smokeless or chewing tobacco have also been shown in US prospective studies to have higher death rates from all causes compared with non-users (CPS-I and II studies) and also higher death rates from cancer of the gastrointestinal tract overall (CPS-I study), but separate data on the upper gastrointestinal tract were not shown [13].

Although there is extensive exposure to swallowed tobacco juice contaminated saliva, and high serum levels of nicotine and nitrites, there have been no studies in terms of oesophageal or gastric histology or *Helicobacter pylori* infection in snus users.

The aim of the study was to investigate whether smoking cigarettes and snus use are associated with gastrointestinal symptomatology, macroscopic findings on endoscopy or histological signs of inflammation or cancer risk markers in the esophagus or stomach, including the prevalence of *Helicobacter pylori*.

Materials and methods

Setting

The Kalixanda study setting consisted of two neighbouring communities in Northern Sweden (Kalix and Haparanda) with 28,988 inhabitants (December 1998). The distribution of age and sex was similar to the national average in Sweden in both communities; although proportion of unemployment was slightly higher and income and the proportion with a higher education were slightly lower [14].

Participants

By using the computerized national population register, covering all citizens in the two communities by date of birth order, a representative stratified sample was generated. Every seventh adult ($n = 3{,}000$) from the target population (18–80 years of age, $n = 21{,}610$ in September 1998) was drawn. The sampled subjects were given an identification number (1–3,000) in a random order [14].

Study design and logistics

The original study population ($n = 3{,}000$) was invited by mail to take part. The invitation included information of the study design and of the aims of the study and a validated questionnaire, the Abdominal Symptom Questionnaire (ASQ) to be returned by mail [15]. Up to two remainders were sent when necessary; 140 subjects were unavailable at the time for invitation, thus 2,860 of the original study

population were eligible for inclusion [14]. The overall response rate was 74.2% ($n = 2,122$) of the eligible study population.

The original study population was divided into five groups according to their given identification number, 1–600, 601–1,200 and so forth, the first subset of study subjects was approached with the mailed ASQ in November 1998 and the study was completed in June 2001 [14]. In order to complete 1,001 esophagogastroduodenoscopies (EGDs), 1,563 responders to the ASQ had to be approached of whom 364 declined, 74 had moved or could not be reached and 124 were excluded according to the study protocol. Thus the overall response rate for those eligible for the EGD was 73.3% [14]. The exclusion criteria were the presence of serious physical or mental disorder, alcoholism, previous upper gastrointestinal surgery and pregnancy [14]. The biopsies for *Helicobacter pylori* culture and concomitant histology were available from 1,000 subjects. At the visit for the EGD, the participants filled in a more comprehensive ASQ, as described previously [14].

The study protocol was approved by Umeå University ethics committee and the study was conducted according to the declaration of Helsinki. Informed consent was obtained from all participants.

Assessments

Abdominal Symptom Questionnaire (ASQ) is a questionnaire assessing symptoms from the upper and lower part of the abdomen, and it has been found to be valid, reproducible and reliable [15]. All participants were asked if they had been troubled by abdominal pain or discomfort at any location or by any of the listed 33 other gastrointestinal symptoms [15]. The extended ASQ filled in at the EGD visit also included the grading of severity and the frequency of each symptom (daily, weekly or last 3 months). The participants' medication use during the previous 3 months was recorded.

Demographic data and history

Demographic data were collected at the clinic visit (sex, age, length and weight, use of different tobacco products, use of alcohol and use of medication). The subjects' level of education (low education = elementary, comprehensive or secondary school, high education = upper secondary school or university) was asked at the clinic visit.

Definition of body mass index

Body mass index (BMI) was calculated and categorized according to World Health Organization recommendations [16].

Definitions of symptom groups

Gastroesophageal reflux symptoms (GERS) were defined as the presence of any troublesome heartburn and/or acid regurgitation over the past 3 months [14, 17].

Dyspepsia was defined as epigastric troublesome pain or discomfort, and/or nausea, early satiety or uncomfortable feeling of fullness after meal. This is consistent with the Rome II definition (except for upper abdominal bloating which was not asked about in the ASQ) [18]. A simple definition of dyspepsia, labelled "epigastric pain or discomfort", based on the Rome I definition of dyspepsia, was also used [19].

Irritable bowel syndrome (IBS) was defined as troublesome abdominal pain or discomfort located at any site plus concomitant bowel habit disturbances (constipation, diarrhoea, or alternating constipation and diarrhoea). This definition has been used previously and shown to produce results reasonably concordant with the Rome criteria in Sweden [20].

Abdominal pain was defined as troublesome pain or discomfort indicated anywhere in the abdomen [14].

No or minor gastrointestinal symptoms were symptoms not fulfilling any of the above symptom classifications, or absence of symptoms in the ASQ.

Smoking cigarettes and snus use

A complete medical history was taken after the blinded upper endoscopy. The participants were asked about their present and past snus use and the current amounts/week and also about their smoking habits and the number of cigarettes per day in a standardized fashion.

Definitions of tobacco user categories

Current snus users were individuals using moist snuff or chewing tobacco ($n = 1$), without any present or former use of smoked tobacco.

Current smokers were individuals smoking cigarettes without any present or former snus use.

Current users of both were individuals currently both smoking cigarettes and using snus.

Former smokers were former cigarette smokers without present or former snus use.

Former snus users were former snus users without present or former smoking cigarettes.

Former users of both were former cigarette smokers and snus users without present smoking or snus use.

Non-users were individuals who had never used tobacco products.

Esophagogastroduodenoscopy

The upper endoscopies were provided by three experienced endoscopists in Kalix and Haparanda which gave sole medical cover to the area. Internal validity was assessed by means of consensus sessions [14]. The endoscopists had been participating in regular quality assessment programs over several years. The endoscopists were unaware of the symptoms of the subjects before and during the endoscopy [14].

Definition of gastric and duodenal ulcer

Peptic ulcer was defined as a mucosal break at least 3 millimetres in diameter in either the stomach or duodenum [11].

Definition and classification of erosive esophagitis

The subjects with mucosal breaks in the esophagus were classified as having erosive esophagitis and graded according to the Los-Angeles classification [21].

Histology and *Helicobacter pylori*

Two experienced pathologists (M. V. and M. Stolte), who were unaware of the endoscopy findings, evaluated the biopsies and provided a common report. The biopsies were stained with haematoxylin and eosin. *Helicobacter pylori* was detected by Warthin-Starry silver staining [22].

Histological parameters of the gastric mucosa were assessed by using the updated Sydney System definitions [23].

Chemical-reactive gastritis proposed to be caused by aspirin, NSAIDs, alcohol or bile reflux was defined according to the updated Sydney System definitions [23, 24].

Samples from the antrum and corpus were cultured and analysed as described previously [22].

Current *Helicobacter pylori* infection was defined as a positive culture or histology. There was an overall agreement of 99.3 percent between the two methods [22].

Serology

Helicobacter pylori IgG antibodies were determined by EIA (Pyloriset EIA-G, Orion diagnostica, Espoo Finland) [25]. A positive test in the absence of *Helicobacter pylori* detection by culture or histology was considered indicative of a past infection.

Gastrin-17 (cut off $\geq$ 10 pmol/liter) and Pepsinogen-1 (cut off $<$ 25 µg/l for low and $>$ 100 µg/l for high levels) were analysed using specific EIA tests (Biohit Plc, Helsinki, Finland).

Statistical analysis

Pearson χ^2 test was used for testing comparisons in univariate analysis. The associations of tobacco user categories with GERS, dyspepsia, epigastric pain, abdominal pain, no or minor GI symptoms and IBS were analyzed applying logistic regression models including possible confounders; *Helicobacter pylori* infection, use of aspirin, use of non-steroidal anti-inflammatory drugs, use of acid reducing drugs, high alcohol consumption ($\geq$100 g/week), education level and categorized BMI and adjusting for age and sex and using non-users as the reference group (odds ratio (OR) = 1). The associations of tobacco user categories with esophagitis, gastric ulcer, duodenal ulcer, overall PUD, high Pepsinogen-1 level, high Gastrin-17 level and dichotomized histological variables from the esophagus, stomach and duodenum were analyzed applying multivariate logistic regression models including possible confounders; *Helicobacter pylori* infection, use of aspirin, use of non-steroidal anti-inflammatory drugs, high alcohol consumption, education level, categorized BMI, use of acid reducing drugs and GERS and adjusting for age and sex and using non-users as the reference group (OR = 1).

Associations with PUD were also analyzed using smokers as the reference group (OR = 1). The results were controlled for interactions. Age and gender in all analyses and use of aspirin/NSAIDs, high alcohol consumptin, gastroesophageal reflux and *Helicobacter pylori* infection were the variables tested as possible effect modifiers. Stepwise model improvement was applied in all analyses to determine the most suitable multivariate logistic regression model and all analyses were adjusted at least for categorized (15 year bands) age and sex. The results from crude logistic regression models are also presented. The results are presented as odds ratios (OR) with 95% confidence interval (95% CI). The goodness of fit of the models was judged from the Pearson χ^2 test (acceptable model when $P > 0.05$). A two sided P-value of < 0.05 was regarded as statistically significant. Fisher's exact test was used in appropriate analyses. The STATA 8 program (Stata Corporation. College Station, TX, USA) was used for the analyses [26].

Results

Of the 1,001 subjects endoscoped, 12 did not have data on the snus use or cigarette smoking, leaving 989 subjects for analysis. Of these, 96 (9.7%) were current snus users, 165 (16.7%) were current cigarette smokers, 22 (2.2%) were combined users, 209 (27.7%) were former smokers, 16 were former snus users and 49 former users of both. Overall 432 (43.7%) individuals had never smoked or used

snus. The snus users consumed on average 3.2 cans/week (1 can = 24–50 g), smokers consumed on average 11.5 cigarettes/day and combined snus users/smokers consumed 2.2 cans/week and 6.2 cigarettes/day.

The sex distribution, age groups in 15 year bands, mean BMI, use of aspirin, alcohol consumption and education level in different user and non-user groups are shown in Table 1.

Use of proton pump inhibitors, histamine-2 receptor antagonists and antacids during the last week or during the last 3 months before the EGD was not significantly associated with smoking cigarettes or snus use.

Symptoms

Symptom prevalences in different tobacco user categories are shown in Table 2. Associations with GERS, IBS, dyspepsia, epigastric pain, overall abdominal pain and no or minor symptoms are shown in Table 3. No symptom groups were associated with snus use.

Endoscopy and histology

The prevalences of esophagitis, gastric ulcer, duodenal ulcer and overall PUD, split by tobacco use category, are presented in Table 4 and the associations of different tobacco user categories with esophagitis, gastric ulcer, duodenal ulcer and overall PUD are presented in Table 5.

Esophagus

Snus users had a significantly higher prevalence of macroscopic esophagitis in univariate analyze compared with non-users ($P = 0.04$, Table 4). However, snus was not associated with esophagitis in multivariate analyze adjusting for age and sex (Table 5).

Former snus use was associated with hyperplasia of the basal cell layer 2 cm above the esophago-gastric junction (crude logistic regression model, OR = 4.43, 95% CI: 1.54, 12.74) and logistic regression model ajdusting for GERS, categorized age (15 year bands) and sex (OR = 3.79, 95% CI: 1.28, 11.16). Also former cigarette smoking shows an association with hyperplasia of the basal cell layer in crude model (OR = 1.50, 95% CI: 1.03, 2.17) and in logistic regression model adjusting for GERS, *Helicobacter pylori* infection, categorized age and sex the result is nearly significant (OR = 1.41, 95% CI: 0.97, 2.06). Male sex was also associated with hyperplasia of basal cell layer at this location (OR = 1.39, 95% CI: 1.02, 1.89) as was the oldest age group (≥65 years old

Table 1 Demographic data of tobacco user/non-user groups

Demographic variable	Non-user ($n = 432$)	Current snus user ($n = 96$)	Current smoker ($n = 165$)	Using both ($n = 22$)	Former snus user ($n = 16$)	Former smoker ($n = 209$)	Former user of both ($n = 49$)
Proportion of men	38.2	84.4[a]	34.5	54.5	100.0	50.2	89.8
95% CI	33.6, 42,8	77.1, 91.7	27.2, 41.8	33.7, 75.3		43.4, 57.0	81.3, 98.3
Age 20–34 years	8.3	18.8	11.5	22.7	12.5	4.3	10.2
	5.7, 10.9	11.0, 26.6	6.6, 16.4	5.2, 40.2	0.0, 28.7	1.5, 7.1	1.7, 18.7
Age 35–49 years	25.9	28.1	33.9	22.7	37.5	23.0	24.5
	21.8, 30.0	19.1, 37.1	26.7, 41.1	5.2, 40.2	13.8, 61.2	17.3, 28.7	12.5, 36.5
Age 50–64 years	34.0	42.7	41.8	45.5	25.0	40.7	38.8
	29.5, 38.5	32.8, 52.6	34.3, 49.3	24.7, 66.3	3.8, 46.2	34.0, 47.4	25.2, 52.4
Age ≥65	31.7	10.4	12.7	9.1	25.0	32.1	26.5
	27.3, 36.1	4.3, 16.5	7.6, 17.8	0.0, 21.1	3.8, 46.2	25.8, 38.4	14.1, 38.9
High alcohol consumption (>100 g/week)	7.4	25.0	13.3	27.3	6.3	14.4	20.4
	4.9, 9.9	16.3, 33.7	8.1, 18.5	8.7, 45.9	0.0, 18.2	9.6, 19.2	9.1, 31.7
Use of aspirin	10.7	8.3	9.1	13.6	6.3	12.9	10.2
	7.8, 13.6	2.8, 13.8	4.7, 13.5	0.0, 27.9	0.0, 18.2	8.4, 17.4	1.7, 18.7
Use of PPI	5.1	1.0	3.6	4.6	0.0	6.7	8.2
	3.0, 7.2	0.0, 3,0	0.8, 6.4	0.0, 13.4	0.0, 0.0	3.3, 10.1	0.5, 15.9
Mean BMI (SD)	26.6 (3.8)	26.2 (3.5)	25.8[a] (4.5)	26.5 (3.9)	26.3 (3.2)	26.7 (4.2)	27.5 (4.2)
Proportion of low education 95% CI	57.1	54.7	58.0	56.8	56.3	63.8	59.2
	52.4, 61.8	44.7, 64.7	50.5, 65.5	36.1, 77.5	32.0, 80.6	57.3, 70.3	45.4, 73.0

Randomised, population-based, endoscopic study, Kalix and Haparanda, Sweden (the Kalixanda study), December 1998 to June 2001

95% CI 95 percent confidence interval, *SD* standard deviation, *BMI* body mass index

[a] Significant difference compared with non-users ($P < 0.05$)

Table 2 Three months period symptom group prevalences split by tobacco user category

Symptom	Non-user ($n = 432$)	Current snus user ($n = 96$)	Current smoker ($n = 165$)	Using both ($n = 22$)	Former smoker ($n = 209$)	Former snus user ($n = 16$)	Former user of both ($n = 49$)
GERS%[b]	39.1	38.5	37.6	50.0	42.6	50.0	34.7
95% CI	34.5, 43.7	28.8, 48.2	30.2, 45.0	29.1, 70.9	35.9, 49.3	25.5, 74.5	21.4, 48.0
Dyspepsia %[b]	34.5	32.3	42.3	59.1	37.8	37.5	26.5
95% CI	30.0, 39.0	22.9, 41.7	34.8, 49.8	38.6, 79.9	31.2, 44.4	13.8, 61.2	14.1, 38.9
IBS%[b]	27.6	20.8	30.3	54.6[a]	35.9	43.8	16.3
95% CI	23.4, 31.8	12.7, 28.9	23.3, 37.3	33.8, 75.4	29.4, 42.4	19.5, 68.1	6.0, 26.6
Epigastric pain %[b]	18.5	16.7	26.7	50.0[a]	20.1	25.0	16.3
95% CI	14.8, 22.2	9.2, 24.2	19.9, 33.5	29.1, 70.9	14.7, 25.5	3.8, 46.2	6.0, 26.6
Abdominal pain %[b]	49.5	42.7	52.1	68.2	56.5	62.5	42.9
95% CI	44.8, 54.2	32.8, 52.6	44.5, 59.7	48.7, 87.7	49.8, 63.2	38.8, 86.2	29.0, 56.8
No GI symptoms %[b]	38.2	39.6	37.0	27.3	30.1	18.8	38.8
95% CI	36.6, 42.8	29.8, 49.4	29.6, 44.4	8.7, 45.9	23.9, 36.3	0.00, 37.9	25.2, 52.4

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95% CI 95 percent confidence interval, *GERS* gastroesophageal reflux symptoms, *IBS* irritable bowel syndrome, *GI* gastrointestinal

[a] Significant difference compared with non-users ($P < 0.05$)

[b] Prevalence

Table 3 Tobacco use and association with gastrointestinal symptoms (OR = 1 for non-users, $n = 432$)

Symptom	Current snus user		Current smoker		Using both		Former snus user		Former smoker		Former user of both	
	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI
GERS	1.22	0.72, 2.06	0.9	0.60, 1.39	2.40	0.89, 6.56	1.89	0.65, 5.51	1.12	0.77, 1.65	0.83	0.40, 1.74
Dyspepsia	1.29	0.76, 2.19	**1.61**[a]	**1.09, 2.19**	**2.78**[a]	**1.06, 7.28**	1.93	0.64, 5.86	1.18	0.81, 1.73	0.84	0.40, 1.77
IBS	0.87	0.49, 1.55	1.12	0.74, 1.70	**3.25**[a]	**1.28, 8.22**	2.60	0.91, 7.41	1.62	1.12, 2.35	0.59	0.26, 1.35
Epigastric pain	1.48	0.77, 2.85	1.49	0.94, 2.35	**5.66**[a]	**2.18, 14.69**	3.15	0.91, 10.96	1.17	0.75, 1.83	1.28	0.52, 3.11
Abdominal pain	1.05	0.64, 1.73	1.06	0.72, 1.57	2.08	0.77, 5.66	2.54	0.87, 7.47	**1.48**[a]	**1.03, 2.12**	0.96	0.49, 1.87
No or minor GI symptoms	0.81	0.49, 1.35	0.97	0.65, 1.46	0.63	0.22, 1.80	**0.27**[b]	**0.07, 0.99**	**0.63**[b]	**0.43, 0.93**	0.79	0.40, 1.57

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OR odds ratio, *95% CI* 95 percent confidence interval, *GERS* gastroesophageal reflux symptoms, *IBS* irritable bowel syndrome, *GI* gastrointestinal

[a] Significant association

[b] Significant negative association

individuals). *Helicobacter pylori* infection did not have any impact on histological changes 2 cm above the esophago-gastric junction.

Snus use was associated with hyperplasia of the basal cell layer (crude logistic regression model, OR = 1.98, 95% CI: 1.19, 3.31 and logistic regression model adjusting for GERS, *Helicobacter pylori* infection, categorized age and sex, OR = 1.74, 95% CI: 1.02, 3.00) and with elongation of papillae (crude model. OR = 2.02, 95% CI: 1.23, 3.35 and logistic regression model adjusting for GERS, *Helicobacter pylori* infection, categorized age and sex, OR = 1.79, 95% CI: 1.05–3.05) of the squamous epithelium at the esophago-gastric junction; both are histological markers of cell turnover due to chronic chemical irritation such as occurs in GERD. GERS was also a associated both with hyperplasia of basal cell layer and elongation of papillae at this location (OR = 1.42, 95% CI: 1.07, 1.88 and OR = 1.66, 95% CI: 1.26, 2.21, respectively). *Helicobacter pylori* infection was negatively associated with both these histological changes (OR = 0.71, 95% CI: 0.53, 0.95 and OR = 0.69, 95% CI: 0.51, 0.92). These results were not confounded by sex or age.

Stomach

Current smoking cigarettes was associated with overall PUD (crude logistic regression model, OR = 2.38, 95% CI: 1.11, 5.11 and logistic regression model adjusting for categorized (15 year bands) age, *Helicobacter pylori* infection, use of aspirin, obesity and sex, OR = 2.32, 95%

Table 4 Prevalences of gastrointestinal findings split by tobacco user category

Endoscopic finding	Non-user ($n = 432$)		Current snus user ($n = 96$)		Current smoker ($n = 165$)		Using both ($n = 22$)		Former snus user ($n = 16$)		Former smoker ($n = 209$)		Former user of both ($n = 49$)	
	%[c]	95% CI	%[c]	95% CI	%[c]	95% CI	%[c]	95% CI	%[c]	95% CI	%[c]	95% CI	%[c]	95% CI
Esophagitis	13.7	10.5, 16.9	21.9[a]	13.6, 30.2	12.7	7.6, 17.8	18.2	2.1, 34.3	25.0	3.8, 46.2	16.3	11.3, 21.3	18.4	7.6, 29.2
Gastric ulcer	1.6	0.4, 2.8	1.0[b]	0.0, 1.8	3.6	0.8, 6.4	4.6	0.0, 13.2	0.0	–	2.4	0.3, 4.5	0.0	–
Duodenal ulcer	1.9	0.6, 3.2	0.0	–	4.2	1.1, 7.3	4.6	0.0, 13.2	0.0	–	1.4	0.0, 3.0	2.0	0.0, 5.9
Overall PUD	3.5	1.8, 5.2	1.0[b]	0.0, 1.8	7.9	3.8, 12.0	9.1	0.0, 21.1	0.0	–	3.8	1.2, 6.4	2.0	0.0, 5.9

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PUD peptic ulcer disease, *95% CI* 95 percent confidence interval

[a] Significant difference $P < 0.05$

[b] A user of chewing tobacco and using only <5 g/week

[c] Prevalence

Table 5 Tobacco use and associations with gastrointestinal findings (OR[#] = 1 for non-users, $n = 432$)

Endoscopic finding	Current snus user ($n = 96$)		Current smoker ($n = 165$)		Using both ($n = 22$)		Former smoker ($n = 209$)		Former snus user ($n = 16$)		Former user of both ($n = 49$)	
	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI	OR	95% CI
Esophagitis	1.13	0.62, 2.08	1.07	0.61, 1.88	1.08	0.33, 3.60	1.09	0.66, 1.78	1.33	0.40, 4.43	0.74	0.32, 1.69
Gastric ulcer	0.93	0.11, 8.08[b]	2.6	0.84, 8.08	2.88	0.32, 26.23	1.49	0.46, 4.87	–	–	–	–
Duodenal ulcer	–	–	2.20	0.77, 6.30	2.12	0.23, 19.46	0.64	0.17, 2.51	–	–	0.93	0.11, 8.11
Overall PUD	0.34	0.04, 2.69	**2.32**[a]	**1.04, 5.19**	2.57	0.49, 13.55	1.00	0.41, 2.44	–	–	0.64	0.08, 5.23

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PUD peptic ulcer disease, *95% CI* 95 percent confidence interval, *OR* odds ratio

[a] Significant association

[b] A user of chewing tobacco and using only <5 g/week

CI: 1.04, 5.19) whereas snus use was not. Snus users had lower odds of PUD than smokers (OR = 0.12, 95% CI: 0.02, 0.99 in logistic regression model adjusting for age and sex and OR = 0.13, 95% CI: 0.02, 1.09 analyzed with all possible confounders), but the odds of snus users was not significantly lower than non-users had. Current smoking and former smoking cigarettes were associated with chemical reactive gastritis in the antrum (crude model OR = 1.34, 95% CI: 0.89, 2.00 and OR = 1.36, 95% CI: 0.94, 1.98, respectivley, and logistic regression model adjusting for *H. pylori* infection, high alcohol consumption, use of aspirin, categorized age and sex, OR = 1.62, 95% CI: 1.02, 2.60 and OR = 1.56, 95% CI: 1.02, 2.55, respectively). Sex was not associated with chemical reactive gastritis in the antrum but age groups older than 34 years were associated (35–49 years OR = 1.78, 95% CI: 1.00, 3.16, 50–64 years OR = 2.10, 95% CI 1.19, 3.68 and ≥65 years OR = 2.52, 95% CI 1.35, 4.69). Use of any tobacco product was not associated with high granulocyte or lymphocyte counts in antrum or corpus and neither with atrophy or intestinal metaplasia at these locations.

Helicobacter pylori and serology

Helicobacter pylori infection prevalence and seropositivity are presented in Table 6. There were no significant associations between current *Helicobacter pylori* infection or seropositivity and different tobacco product user groups. The proportion of cag-A positive *Helicobacter pylori* genotypes did not differ significantly between non-users and different tobacco product user groups.

Gastrin-17 and Pepsinogen-1

Snus use or smoking cigarettes were not associated with abnormal gastrin-17 level but both were associated with high Pepsinogen-1 level (crude logistic regression model for snus OR = 2.13, 95% CI: 1.35, 3.36 and logistic regression model adjusting for GERS, age and sex, OR = 2.01, 95% CI: 1.23, 3.28 and crude model for smoking cigarettes, OR = 2.60, 95% CI: 1.79, 3.77 adjusted logistic regression model OR = 3.01, 95% CI:

Table 6 *Helicobacter pylori* prevalences in tobacco user/non-user groups (culture/histology and serology)

Hp infection and serology in age groups	Non-user (*n*) Prevalence (95% CI)	Current snus user (*n*) Prevalence (95% CI)	Current smoker (*n*) Prevalence (95% CI)	Using both (*n*) Prevalence (95% CI)	Former smoker (*n*) Prevalence (95% CI)	Former user of snus (*n*) Prevalence (95% CI)	Former user of both (*n*) Prevalence (95% CI)
Age 20–49	$n = 148$	$n = 45$	$n = 75$	$n = 10$	$n = 57$	$n = 8$	$n = 17$
Current *Hp* infection	16.2 (10.3, 22.1)	17.8 (6.6, 29.0)	21.6 (12.3, 30.9)	10.0 (0.0, 28.6)	29.8 (17.9, 41.7)	25.0 (0.00, 55.0)	35.3 (12.6, 58.0)
Positive *Hp serology*	21.6 (15.0, 28.2)	26.7 (13.8, 39.6)	25.3 (15.5, 35.1)	20.0 (0.0, 44.8)	36.8 (24.3, 49.3)	25.0 (0.00, 55.0)	35.3 (12.6, 58.0)
Age 50+	$n = 284$	$n = 51$	$n = 90$	$n = 12$	$n = 152$	$n = 8$	$n = 32$
Current *Hp* infection	44.0 (38.2, 49.8)	33.3 (20.4, 46.2)	47.8 (37.5, 58.19)	33.3 (6.6, 60.0)	41.5 (33.7, 49.3)	37.5 (4.0, 71.0)	21.9 (7.6, 36.2)
Positive *Hp serology*	53.2 (47.4, 59.0)	47.1 (33.4, 60.8)	60.0 (49.9, 70.1)	41.7 (13.8, 69.6)	55.3 (47.4, 63.2)	37.5 (4.0, 71.0)	31.3 (15.2, 47.4)
All ages	$n = 432$	$n = 96$	$n = 165$	$n = 22$	$n = 209$	$n = 16$	$n = 49$
Current *Hp* infection	34.5 (30.0, 39.0)	26.0 (17.2, 34.8)	36.0 (28.7, 43.3)	22.7 (5.2, 40.2)	38.3 (31.7, 44.9)	31.3 (8.6, 54.0)	32.7 (19.6, 45.8)
Positive *Hp serology*	42.4 (37.7, 47.1)	37.5 (27.8, 47.2)	44.2 (36.6, 51.8)	31.8 (12.3, 51.3)	50.2 (43.4, 57.0)	31.3 (8.6, 54.0)	32.7 (19.6, 45.8)

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Hp Helicobacter pylori, 95% CI 95 percent confidence interval

2.04, 4.44, respectively) without changing the Pepsinogen-1/Pepsinogen-2 ratio significantly.

When all *Helicobacter pylori* infected, all with histological mucosal atrophy in the stomach and all proton pump inhibitor users were excluded, both snus use and smoking cigarettes were associated with high Pepsinogen-1 (crude logistic regression model for snus OR = 2.71, 95% CI: 1.47, 5.02 and logistic regression model adjusting for GERS, age and sex OR = 2.47, 95% CI: 1.27, 4.79 and (crude logistic regression model for smoking cigarettes, OR = 3.88, 95% CI: 2.29, 6.56 and adjusted logistic regression model, OR = 5.03, 95% CI: 2.89, 8.80, respectively) but not with abnormal Gastrin-17 levels. Male sex was a confounder (OR = 1.9, 95% CI: 1.26, 2.96) as was age groups older than 34 years (35–49 years OR = 2.25, 95% CI: 1.03, 4.88, 50–64 years OR = 2.90, 95% CI 1.35, 6.25 and ≥65 years OR = 4.67, 95% CI 2.00, 10.90). The mean value of Pepsinogen-1 for snus users was 91.8 μg/l (SD 36.4) and for cigarette smokers 102.8 μg/l (SD 37.9). The difference is not significant.

Discussion

This is the first population-based, endoscopic study on the effects of snus use and smoking cigarettes in the upper gastrointestinal tract. The snus use was associated with histological markers of chronic chemical irritation alike in gastroesophageal reflux disease (GERD) at the esophagogastric junction. Smoking cigarettes, but not use of snus, was associated with overall PUD.

In this study, 19% of men and 5% of women were current snus users, which are consistent with the Swedish average, of 21% of men and 4% of women. In addition the amounts consumed are also comparable to use in Sweden (3 cans/week) [2]. The study population has been shown to be representative of the Swedish general population [14]. We believe that the results are likely to be generalizable to most Western populations including those were snus is used.

Smoking was a significant risk for PUD while snus users had less PUD than expected although the latter observation did not reach statistical significance. The reasons for this difference are uncertain. Snus contains high amounts of nitrate [27], which is associated with an increased nitrite formation in the oral cavity and further to formation of nitric oxide in the stomach [28–30]. In contrast, cigarette smoking seems to be related to reduced levels of nitrite in saliva [31]. Salivary nitrite has marked gastro-protective effects through nitric oxide formation [32]. These effects include elevated gastric mucosal blood flow and increased mucus thickness. Acidified nitrite has bactericidal effects [33], possibly including *Helicobacter pylori*. Snus did not

impact on *Helicobacter pylori* status significantly in this population even though there was a non-significant trend towards less *Helicobacter pylori* infection along with rising age in snus users. We observed that both snus use and cigarette smoking caused significantly higher levels of Pepsinogen-1 without affecting Gastrin-17 level. Thus, the difference in PUD prevalences seems not to be due to the Gastrin-17-acid axis.

Intravenously infused nicotine has been shown to decrease pancreatic bicarbonate secretion in animals [34]. Similarly, reduced bicarbonate has been causally related to a higher risk of PUD in cigarette smokers [6]. There are no studies examining this effect in snus users, whose serum nicotine levels are similar to or higher than those of smokers. The role of swallowed alkaline tobacco-contaminated saliva against the development of PUD is unclear in snus users.

Snus use was associated with histological findings in the esophagus consistent with GERD. In a human study the frequency of transient lower esophageal sphincter relaxation was increased by the nitric oxide generating solution. The nitric oxide generating solution also increased esophageal acid exposure [35]. Whether swallowed alkaline tobacco-contaminated saliva, comprising tobacco specific nitrosamine compounds [27], has any role in the development of esophagitis is unclear.

Juice from snus contains high amounts of nitrate which can be reduced to nitrite in the oral cavity [28–30]. When saliva, including dietary nitrate, converted to nitrite, meets acidic gastric juice, the nitrite is converted to nitrous acid, nitrosative species and nitric oxide [29, 30, 36]. In healthy volunteers this potentially mutagenic chemistry appears to be focused at the gastric cardia [36]. A study from the UK showed that both acid and nitric oxide alone can induce double-strand DNA breaks in non-dysplastic Barrett's esophagus and thus may contribute to the genetic rearrangements in the progression from Barrett's esophagus to esophageal adenocarcinoma [37].

A recent Swedish study could observe an increased risk for esophageal squamous cell cancer and a slightly elevated risk for non-cardiac stomach cancer among snus users who had never smoked [38]. We could, however, not verify any premalignant mucosal changes in the stomach. A retrospective cohort study showed that snus use was associated with increased risk of pancreatic cancer as was also cigarette smoking [39]. A Swedish review on the possible harmful effects of snus use found that the carcinogenic effect is probably due to the content of tobacco specific nitrosamines [40].

The strength of our study is the population-based study design. The response rate to all parts of the study was high, suggesting that the results are likely to be reliable and representative. The ASQ-questionnaire is valid, reliable and reproducible [15]. Each participant was specifically asked about use of tobacco products in a face to face interview, and therefore underreporting use is less likely. The weakness is that we cannot provide any physiological data, aside from Gastrin-17 and Pepsinogen-1 levels, and due to the cross-sectional study design it is not possible to draw definite conclusions about any causal connections between exposure and different gastrointestinal disorders. The small number of individuals in some sub-groups is a limitation. Some of the analyses attaining levels of statistical significance ($P < 0.05$) could be chance given the number of tests.

The possibility of higher odds of histological changes, as shown by us, and the possible risk of cancer shown in earlier studies [38, 39] must be taken in account when advocating for using snus in smoking quitting programs [41]. There is no reason to believe, with Sweden as a vivid example, that addictive snus on an open market would be used only by ex-smokers. It is also important to note that concomitant smoking cigarettes and snus use seems to be more harmful than snus use only [12].

In conclusion the snus use was associated with histological markers of increased proliferation of the squamous epithelium consistent with GERD at the esophago-gastric junction but snus does not increase the risk for self-reported upper gastrointestinal symptom groups or risk for PUD.

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