

HHS Public Access

Author manuscript *Tob Control.* Author manuscript; available in PMC 2016 December 01.

Published in final edited form as:

Tob Control. 2015 December ; 24(E4): e271-e274. doi:10.1136/tobaccocontrol-2014-051744.

High levels of tobacco-specific *N*-nitrosamines and nicotine in Chaini Khaini, a product marketed as snus

Irina Stepanov, PhD,

Masonic Cancer Center and Division of Environmental Health Sciences, University of Minnesota, Minneapolis, MN 55455, USA

Prakash C. Gupta, PhD,

Healis - Sekhsaria Institute for Public Health, Belapur Navi Mumbai 400 614, India

Gauri Dhumal, MSc,

Healis - Sekhsaria Institute for Public Health, Belapur Navi Mumbai 400 614, India

Katrina Yershova, BS,

Masonic Cancer Center, University of Minnesota, Minneapolis, Minnesota, 55455, USA

William Toscano, PhD,

Division of Environmental Health Sciences, University of Minnesota, Minneapolis, Minnesota, 55455, USA

Dorothy Hatsukami, PhD, and

Masonic Cancer Center, University of Minnesota, Minneapolis, Minnesota 55455, USA

Mark Parascandola, PhD

Tobacco Control Research Branch, National Cancer Institute, Bethesda, MD 20892

Abstract

Introduction—Recently, a tobacco product, Chaini Khaini, identified as *snus* appeared in India. The product marketing emphasizes its discreet nature and explicitly claims safety by referring to the existing evidence on Swedish snus. We here analyzed tobacco-specific nitrosamines and nicotine in 12 samples of Chaini Khaini purchased in 2013 at open markets in India.

Methods—Samples were purchased twice: in March 2013 from Mumbai and in November 2013 from Mumbai and Ahmedabad. Chemical constituents were measured by our routine validated methods.

Corresponding author: Irina Stepanov, Ph.D. Masonic Cancer Center, University of Minnesota, Cancer and Cardiovascular Research Building, 2231 6th Street SE - Room 2-140, Minneapolis, MN 55455, USA, phone: 1-612-624-4998 ; fax: 1-612-624-3869; stepa011@umn.edu.

Contributions. IS participated in development of study concept and design, analyzed data, drafted and revised the manuscript, and approved the final version for publication. PCG contributed to study concept and design, data collection, and manuscript preparation. GD contributed to data collection and manuscript preparation. KY performed laboratory analyses and contributed to data interpretation and manuscript preparation. WT contributed to data collection and manuscript revision. DH developed study concept and design, revised the manuscript, and approved the final version for publication. MP contributed to study concept and design, interpretation of data, and preparation of the manuscript.

Competing interests. There are no competing interests associated with this study.

Results—Levels of carcinogenic nitrosamines NNN, NNK, and NNAL averaged 22.9 (\pm 4.9) μ g/g, 2.6 (\pm 1.0) μ g/g, and 3.1 (\pm 1.5) μ g/g tobacco (wet weight), respectively. The levels of NAB, which is normally present in trace levels in tobacco products, ranged from 3.9 to 12.9 μ g/g tobacco. Total nicotine levels in all samples averaged 10.0 mg/g tobacco and unprotonated nicotine accounted for an average 95.4% of the total nicotine content.

Conclusions—Chaini Khaini, which is labeled as *snus* and is marketed as a safe alternative to other tobacco products contains very high levels of carcinogenic nitrosamines and biologically available nicotine. Interventions are urgently needed to educate current and potential consumers of this product.

Keywords

Smokeless tobacco; snus; tobacco-specific nitrosamines; nicotine; toxicology

INTRODUCTION

The use of smokeless tobacco has been evaluated as a causal factor for oral, pancreatic, and esophageal cancer.¹ However, existing epidemiologic studies indicate that exclusive use of Swedish moist snuff, which is called snus, is associated with the relatively low overall cancer risks.² An increased risk of pancreatic cancer has been reported in snus users when compared to never-users of any tobacco; however, the evidence suggests that it is not associated with significant risk of oral cancer.

Swedish snus is a moist finely ground product made with pasteurized air- or sun-cured tobacco. It is available in either loose form or pre-portioned in small teabag-like sachets and is typically used by placing it between the upper lip and gum for approximately 30 minutes. The relatively low risk of cancer associated with the use of Swedish snus is attributed, at least in part, to the fact that it contains low levels of tobacco-specific *N*-nitrosamines (TSNA) – a major group of potent carcinogens in smokeless tobacco." Indeed, in laboratory animal studies, the two carcinogenic TSNA, *N'*-nitrosonornicotine (NNN) and 4- (methylnitrosamino)-1-(3-pyridyl)-1-butanone (NNK), cause cancers that are most strongly associated with smokeless tobacco use,' and there is strong and consistent evidence of an increased risk of oral cancer with the use of smokeless tobacco that is highly contaminated with TSNA.⁻⁻ The specifics of Swedish snus manufacturing prevent the formation of high levels of TSNA in this product.' Thus, based on the "Swedish experience", encouraging smokers to switch to the Swedish-type low-nitrosamine snus is seen by some as a potential harm reduction strategy.'

While Sweden is the home of snus, products with this name started appearing in other parts of the world. For instance, several new tobacco products called snus entered the U.S. market in 2006. More recently, a tobacco product Chaini Khaini that is also marketed as snus appeared in India. The product package and website seem to employ marketing features similar to those used for the U.S. snus: the statements emphasize its discreet nature, and a 3-step instruction on how to use this product is provided. In addition, Chaini Khaini is explicitly equated with Swedish snus and is claimed to offer "safety from smoking and chewing tobacco" (http://chainikhaini.com/index.htm, Figure 1). However, traditional Khaini

Stepanov et al.

- the product that is the most popular in India – was reported to contain high levels of TSNA.[•] On the other hand, a recent publication described Chaini Khaini "snus" as not being the traditional khaini, and it was suggested that cultural attributes are being exploited in the promotion of this product.

To explore whether Chaini Khaini is similar to Swedish snus and is simply called "khaini" to appeal to Indian consumers, or it is actually more similar to khaini and is marketed as snus, we analyzed TSNA, nicotine and unprotonated nicotine in samples of this product purchased in 2013 in India.

METHODS

Tobacco samples

Sample collection was carried out twice: in March 2013 in Mumbai and in November 2013 in Mumbai and Ahmedabad. At all locations, Chaini Khaini was exposed to high ambient temperatures and humidity, similar to the routine handling of other smokeless tobacco products. We sought to obtain representative averages for constituent levels by purchasing samples from three different markets in each location. After the purchase, samples were labeled and handled according to our standardized sampling and labeling procedures. In the laboratory, samples were sealed in plastic sleeves and stored at 4 °C until analysis.

Tobacco analysis

Samples were prepared according to our routine validated methods. Analysis of five TSNA – NNN, NNK, 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol (NNAL), *N*'-nitrosoanatabine (NAT), and *N*'-nitrosoanabasine (NAB) – were performed by liquid chromatography-tandem mass-spectrometry as described. Nicotine was analyzed by gas chromatography-mass spectrometry-selected ion monitoring. The amount of unprotonated nicotine was calculated using the Henderson-Hasselbalch equation. Moisture content and pH were measured as previously described.

RESULTS

A total of 12 samples, all produced by the same manufacturer, have been purchased and analyzed. The first set of 6 samples was purchased in March 2013 in three markets in Mumbai, two samples per vendor. To confirm the results of chemical analyses obtained for this first set, additional samples were obtained in November 2013 from three markets in Mumbai and three markets in Ahmedabad, one sample per location.

Constituent levels in all samples are summarized in Table 1. The results are expressed per gram tobacco (wet weight). Nicotine levels in all samples ranged from 7.9 to 13.4 mg/g tobacco, and pH ranged from 9.01 to 9.92. On average, unprotonated nicotine accounted for 95% of total nicotine content. The sum of all five measured TSNA ranged from 23.1 to 61.2 μ g/g tobacco. Levels of carcinogenic NNN, NNK, and NNAL averaged 22.9 (±4.9) μ g/g, 2.6 (±1.0) μ g/g, and 3.1 (±1.5) μ g/g tobacco, respectively. The levels of NAB, which is normally present in trace levels in tobacco products, ranged from 3.9 to 12.9 μ g/g tobacco.

DISCUSSION

As manufactured in Sweden, snus contains relatively low levels of TSNA and is associated with relatively low risk of cancer compared to other forms of smokeless tobacco. Thus, there is an opinion shared by many public health researchers that Swedish-type snus has a potential to reduce tobacco-induced morbidity and mortality in people who switch to this product instead of using other, more harmful, forms of tobacco.¹ We here demonstrate that Chaini Khaini, a product marketed as snus in India with explicit reference to "Swedish experience", contains remarkably high levels of TSNA and biologically available nicotine. This is an example of how the notion of snus "harm reduction" can be misused in the marketing of a highly carcinogenic and addictive product.

The reported levels of TSNA and nicotine in traditional Khaini and in Swedish snus are listed in Table 1 for comparison with the data obtained in the current study. We found that the levels of TSNA in Chaini Khaini are similar to those found in traditional Khaini.[,] These levels are among the highest reported to be present in tobacco products, second only to those found in Sudanese toombak in the recently analyzed sample of products from the global market. It is also notable that the levels of NNAL, which is a metabolite of NNK and demonstrates carcinogenicity that is comparable to NNK in laboratory animals, is unusually high in this product; the levels are similar to those of NNK (Table 1). Typically, NNAL levels in tobacco comprise about 10% of the NNK content. However, relative amounts of NNK and NNAL found in Chaini Khaini in this study are also similar to those previously reported for traditional khaini. In addition, the levels of NAB, a weak esophageal carcinogen which is normally present in trace levels in tobacco products, averaged 8.4 μ g/g tobacco. Whereas, given its weak carcinogenicity and typically low levels NAB is usually not referred to as an essential carcinogenic TSNA, such high levels of NAB in Chaini Khaini may contribute to the carcinogenicity of this product. Similarly high levels of NAB are also found in traditional khaini.[,]

Similar to traditional Kahini, pH of Chaini Khaini is highly alkaline, leading to more than 90% of total nicotine being present in the biologically available unprotonated form (Table 1). Furthermore, total nicotine levels in Chaini Khaini are higher than those reported for traditional khaini. Thus, the levels of unprotonated nicotine, which defines the addictive potential of smokeless tobacco, are 2–3 times higher in Chaini Khaini than in traditional khaini, and are among the highest reported for smokeless tobacco products.

Together, our analyses demonstrate that, in its carcinogenic and addictive potential, Chaini Khaini is very close to traditional khaini. Thus, labeling this product as *snus* and claiming its safety as compared to other forms of tobacco use is unsubstantiated. Such labeling represents an example of dangerous misuse of the available evidence on Swedish snus in the marketing of a highly carcinogenic and addictive tobacco product. The misleading marketing of Chainin Khaini is aggravated by the fact that, according to Global Adult Tobacco Survey, khaini is the most popular smokeless tobacco product in India. It is the most commonly used smokeless product by both exclusive smokeless users and dual users (with bidi smoking).¹ There is no published data available on the prevalence of use or the consumer perception of Chaini Khaini. However, marketing of this product seems to be a

"perfect storm" that combines an appeal of a popular tobacco product type, emphasis on the use of native tobacco and indigenous flavors, trendiness of packaging, discreet nature of the pouched form, and a promise of safety. Given that the tobacco use-associated oral cancer rates in India are among the highest in the world, this issue requires immediate attention.

In summary, we here report that the product that is identified by its manufacturer as snus and is marketed as a safe alternative to other tobacco products contains very high levels of carcinogenic nitrosamines and biologically available nicotine. Interventions are urgently needed to educate current and potential consumers of this product. Furthermore, regulatory measures are warranted to prevent mislabeling of similarly harmful products as snus and claims, implied or explicit, of risk reduction.

Acknowledgments

The authors thank Bob Carlson for editorial assistance.

Funding. This work was supported by the National Cancer Institute contract HHSN261201200392P and by startup funds to IS from the Masonic Cancer Center, University of Minnesota.

References

- IARC Monographs on the Evaluation of Carcinogenic Risks to Humans. Vol. 89. Lyon, FR: IARC; 2007. International Agency for Research on Cancer Smokeless tobacco and tobacco-specific nitrosamines.
- Zhou J, Michaud DS, Langevin SM, et al. Smokeless tobacco and risk of head and neck cancer: Evidence from a case-control study in New England. Int J Cancer. 2013; 132:1911–1917. [PubMed: 22987222]
- 3. Foulds J, Ramstrom L, Burke M, et al. Effect of smokeless tobacco (snus) on smoking and public health in Sweden. Tob Control. 2003; 12:349–359. [PubMed: 14660766]
- 4. Luo J, Ye W, Zandehdel K, et al. Oral use of Swedish moist snuff (snus) and risk for cancer of the mouth, lung, and pancreas in male construction workers: a retrospective cohort study. The Lancet. 2007; 369:2015–2020.
- Rutqvist LE, Curvall M, Hassler T, et al. Swedish snus and the GothiaTek standard. Harm Reduct J. 2011; 8:11. [PubMed: 21575206]
- Hecht SS. Biochemistry, biology, and carcinogenicity of tobacco-specific N-nitrosamines. Chem Res Toxicol. 1998; 11:559–603. [PubMed: 9625726]
- 7. Gupta PC, Murti PR, Bhonsle RB. Epidemiology of cancer by tobacco products and the significance of TSNA. Crit Rev Toxicol. 1996; 26:183–198. [PubMed: 8688160]
- 8. Stepanov I, Hecht SS, Ramakrishnan S, et al. Tobacco-specific nitrosamines in smokeless tobacco products marketed in India. Intl J Cancer. 2005; 116:16–19.
- Winn DM, Blot WJ, Shy CM, et al. Snuff dipping and oral cancer among women in the Southern United States. N Engl J Med. 1981; 304:745–749. [PubMed: 7193288]
- Hoffmann D, Adams JD. Carcinogenic tobacco-specific N-nitrosamines in snuff and saliva of snuff-dippers. Cancer Res. 1981; 41:4305–4308. [PubMed: 7198005]
- Osterdahl BG, Jansson C, Paccou A. Decreased levels of tobacco-specific N-nitrosamines in moist snuff on the Swedish market. J Agric Food Chem. 2004; 52:5085–5088. [PubMed: 15291479]
- Levy DT, Mumford EA, Cummings KM, et al. The relative risks of a low-nitrosamine smokeless tobacco product compared with smoking cigarettes: estimates of a panel of experts. Cancer Epidemiol Biomarkers & Prev. 2004; 13:2035–2042.
- 13. Savitz DA, Meyer RE, Tanzer JM, et al. Public health implications of smokeless tobacco use as a harm reduction strategy. Am J Public Health. 2006; 96:1934–1939. [PubMed: 17018821]

Stepanov et al.

- Hatsukami DK, Feuer RM, Ebbert JO, et al. Changing smokeless tobacco products: new tobacco delivery systems. Am J Prev Med. 2007; 33:S368–S378. [PubMed: 18021912]
- 15. Mukherjea A. Tobacco industry co-optation of culture? Converging culturally specific and mainstream tobacco products in India. Tob Control. 2012; 21:63–64. [PubMed: 21724744]
- Bhawna G. Burden of smoked and smokeless tobacco consumption in India results from the Global Adult Tobacco Survey India (GATS-India) - 2009–2011. Asian Pac J Cancer Prev. 2013; 14:3323–3329. [PubMed: 23803124]
- Stanfill SB, Connoly GN, Zhang L, et al. Global surveillance of oral tobacco products: total nicotine, unionized nicotine and tobacco-specific *N*-nitrosamines. Tob Control. 2011; 20:e2.doi: 10.1136/tc.2010.037465 [PubMed: 21109685]
- Stepanov I, Biener L, Knezevich A, et al. Monitoring tobacco-specific *N*-nitrosamines and nicotine in novel Marlboro and Camel smokeless tobacco products: Findings from Round I of the New Product Watch. Nicotine Tob Res. 2012; 14:274–281. [PubMed: 22039075]
- Lawler TS, Stanfill SB, Zhang L, et al. Chemical characterization of domestic oral tobacco products: total nicotine, pH, unprotonated nicotine and tobacco-specific *N*-nitrosamines. Food Chem Toxicol. 2013; 57:380–386. [PubMed: 23517910]
- Richter P, Hodge K, Stanfill S, et al. Surveillance of moist snuff: total nicotine, moisture, pH, unionized nicotine, and tobacco-specific nitrosamines. Nicotine Tob Res. 2008; 10:1645–1652. [PubMed: 18988077]
- 21. Tomar SL, Henningfield JE. Review of the evidence that pH is a determinant of nicotine dosage from oral use of smokeless tobacco. Tob Control. 1997; 6:219–225. [PubMed: 9396107]
- 22. Gupta PC, Ray CS, Narake SS, et al. Profile of dual tobacco users in India: an analysis from Global Adult Tobacco Survey, 2009–10. Indian J Cancer. 2012; 49:393–400. [PubMed: 23442404]
- 23. Moore SR, Johnson NW, Pierce AM, et al. The epidemiology of mouth cancer: a review of global incidence. Oral Dis. 2000; 6:65–74. [PubMed: 10702782]

What this paper adds

- Epidemiologic studies indicate that exclusive use of Swedish snus, which contains low levels of carcinogenic tobacco-specific nitrosamines (TSNA), is associated with relatively low risk of cancer compared to other forms of smokeless tobacco. Encouraging smokers to switch to the Swedish-type snus is seen by some as a potential harm reduction strategy.
- This study shows that a product marketed in India as snus and explicitly claimed to be a safe alternative to other tobacco products contains remarkably high levels of TSNA and biologically available nicotine.
- Interventions are urgently needed to educate current and potential consumers of this product, and to prevent mislabeling of similarly harmful products as snus with claims of risk reduction.



Figure 1.

Image of the website that claims safety of Chaini Khaini use and implies similarity of this product with Swedish snus (retrieved from—http://chainikhaini.com/index.htm—on 7 August 2014).

Table 1

Summary of chemical analyses performed on Chaini Khaini and comparison with published data on traditional Khaini and Swedish snus.

Stepanov et al.

		/0 · · · · · · · · · · · · · · · · · · ·			du D	Unprotonated nicotine			ug/g	ug/g wet weight	ht	
vendor	Sample #	Moisture content, %	ЪН	lotal nicotine, mg/g wet weight	%	mg/g wet weight	NNN	NNK	NNAL	NAT	NAB	Total TSNA
				Analyzed in this study	s study							
Mumbai, Ind	Mumbai, India (March 2013)											
M1	1	26.1	9.01	8.2	90.6	7.5	19.5	2.3	2.3	5.7	7.5	37.2
	2	26.1	9.20	7.9	93.7	7.4	17.4	2.4	2.3	5.3	7.3	34.8
M2	3	25.0	9.60	13.4	97.4	13.1	29.4	2.0	4.9	11.6	12.9	60.8
	4	21.5	9.39	12.9	95.9	12.4	27.4	5.0	5.7	10.2	12.9	61.2
M3	S	22.9	9.50	10.0	96.8	9.7	26.7	3.5	5.5	6.1	11.0	52.9
	9	25.1	9.92	9.1	98.7	9.0	25.1	1.9	4.9	6.3	11.0	49.3
Mumbai, Ind	Mumbai, India (November 2013)	013)										
M4	7	25.6	9.41	11.0	96.1	10.6	26.2	3.2	2.4	8.5	7.9	48.2
M5	8	26.9	9.35	9.6	95.5	9.2	24.5	2.1	1.9	6.4	7.0	41.9
M6	6	27.9	9.64	12.0	97.7	11.7	24.9	3.3	2.0	9.1	7.3	46.5
Ahmedabad,	Ahmedabad, India (November 2013)	ər 2013)										
A1	10	25.2	9.24	8.3	94.3	7.8	16.0	1.2	1.7	3.1	4.2	26.3
A2	11	26.1	9.13	8.7	92.8	8.1	13.2	1.5	1.8	2.8	3.9	23.1
A3	12	26.4	9.36	8.3	95.6	7.9	25.0	2.7	2.3	6.1	7.8	43.9
Average for (Average for Chaini Khaini	25.4	9.4	10.0	95.4	9.5	22.9	2.6	3.1	6.8	8.4	37.6
SD		1.7	0.2	1.8	2.2	1.9	4.9	1.0	1.5	2.5	2.9	18.7
				Previously published data	hed dat	a						
Traditional Khaini	<i>Thaini</i>											
Super Raja Khaini	haini	n/r *	9.65	4.79	7.76	4.7	16.8	0.502	1.44	2.22	2.58	23.5
Spitt Raja Chap Khaini	ap Khaini	n/r	9.79	2.53	98.3	2.5	17.5	0.288	1.35	0.303	2.19	21.6
Swedish snus	(*											
General Original Snus	inal Snus	n/r	7.01	8.34	8.98	0.75	0.345	0.096	0.013	0.248	0.021	0.723
General Whit	General White Portion Snus	n/r	6.86	8.09	6.48	0.52	0.296	0.097	0.013	0.225	0.018	0.648