Evaluation of Novel, Oral Tobacco-Derived Nicotine Products for HPHCs

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Abstract

VERVE® Discs and Chews are oral, non-dissolvable, tobacco-derived nicotine products. In May 2016, the U.S. Food and Drug Administration (FDA) issued a final rule to deem e-cigarettes, cigars and all other tobacco products to be subject to the Federal Food, Drug, and Cosmetic Act (the FD&C Act), as amended by the Family Smoking Prevention and Tobacco Control Act (Tobacco Control Act). Manufacturers of regulated tobacco products are required to report to FDA quantities of Harmful and Potentially Harmful Constituents (HPHCs) by November 8, 2019. FDA has not issued specific guidance for reporting HPHCs for novel tobacco products, such as VERVE®, as they have for certain other regulated tobacco products. Absent specific guidance from FDA, we measured HPHCs in VERVE® according to the requirements for smokeless tobacco, recognizing that these products do not meet the statutory definition of a smokeless tobacco product. The objective of this work was to measure HPHCs in VERVE® Discs and Chews products over time and compare those results to other commercially available oral tobacco products and an oral nicotine replacement therapy (NRT) product. Results show the absence of detectable levels or significant reductions in HPHCs compared to other oral tobacco products and comparable HPHC results to the NRT.

Background

- To date, FDA has not issued specific guidance regarding reporting of HPHC levels in novel products such as VERVE®. Even though these types of products do not meet the definition of a "smokeless tobacco product" (i.e., they do not consist of cut, ground, powdered, or leaf tobacco), we applied the abbreviated list of HPHCs for smokeless tobacco from the 2012 Draft Guidance," Reporting Harmful and Potentially Harmful Constituents in Tobacco Products and Tobacco Smoke Under Section 904(a)(3) of the Federal Food, Drug, and Cosmetic Act."
- This study establishes the level of HPHCs in VERVE® Discs and VERVE® Chews and also provides a comparison to other tobacco products as required under section 910(b)(1)(A) of the FD&C Act.

HPHC	FDA Classification
Acetaldehyde	Carcinogen, Respiratory Toxicant, Addictive
Arsenic	Carcinogen, Cardiovascular Toxicant, Reproductive or Developmental Toxicant
B[a]P	Carcinogen
Cadmium	Carcinogen, Respiratory Toxicant, Reproductive or Developmental Toxicant
Formaldehyde	Carcinogen, Respiratory Toxicant
Crotonaldehyde	Carcinogen
Nicotine (total and free)	Reproductive or Developmental Toxicant, Addictive
NNK	Carcinogen
NNN	Carcinogen

Methods

Nicotine (total and free)
- VERVE® Discs are milled to <1.25 mm using a Wiley Mill and an aliquot is extracted in a methanol solution containing internal standard. VERVE® Chews are extracted as-is, without any prior sample preparation, in a methanol and water solution containing internal standard. An aliquot of the extracts is analyzed by LC-MS.

pH
- VERVE® Discs are milled to <1.25 mm using a Wiley Mill and an aliquot of the sample is diluted with deionized water and stirred. The pH of the aqueous extract is then measured at 5 minutes.
- VERVE® Chews are flattened in a pasta roller and diluted with deionized water and stirred. The pH of the aqueous mixture is measured at 5 minutes.

Arsenic and Cadmium
- VERVE® Discs and Chews are digested in nitric acid and analyzed by ICP-MS.

Benzo[a]pyrene
- VERVE® Discs are milled to <1.25 mm using a Wiley Mill and an aliquot is extracted in acetonitrile and water. QuEChERS® salts are added and mixed thoroughly. A portion of the acetonitrile layer is extracted into hexanes and an aliquot is filtered and analyzed by GC-MS.
- VERVE® Chews are extracted in acetonitrile and water. QuEChERS® salts are added and mixed thoroughly. A portion of the acetonitrile layer is extracted into hexanes and an aliquot is filtered and analyzed by GC-MS.

NNN and NNK
- VERVE® Discs are milled to <1.25 mm using a Wiley Mill and an aliquot is extracted in acetonitrile/water containing internal standards followed by solid phase extraction (SPE) and analysis by LC-MS-MS.
- VERVE® Chews are extracted in acetonitrile/water containing internal standards followed by solid phase extraction (SPE) and analysis by LC-MS-MS

Acetaldehyde, Crotonaldehyde and Formaldehyde
- VERVE® Discs are milled to <1.25 mm using a Wiley Mill and an aliquot is extracted in acetonitrile then derivatized with 2,4-dinitrophenylhydrazine (DNPH). The derivatized extract is analyzed for the respective hydrazones using ultra-high performance liquid chromatography (UPLC) with detection by mass spectrometry (MS).
- VERVE® Chews are extracted in acetonitrile then derivatized with 2,4-dinitrophenylhydrazine (DNPH). The derivatized extract is analyzed for the respective hydrazones using ultra-high performance liquid chromatography (UPLC) with detection by mass spectrometry (MS).

Study Summary

Studies were conducted to assess the HPHCs of VERVE® products over a 12 month period. Samples in commercial packaging were stored at room temperature upon receipt until they were analyzed for the first study time point at approximately 1 month post manufacturing. Samples were placed into environmental chambers at the time of analytical testing for the first study time point. The environmental chamber conditions were set to 25 °C ± 2 °C / 60% relative humidity ± 5%. These storage conditions represent ICH Q1A long-term conditions for product intended to be stored at ambient temperatures. Samples were again tested for HPHCs at 6 and 12 months post manufacturing. We also measured HPHCs in Nicorette® Fresh Mint™ Gum (2 mg) for comparison to VERVE®. Data for HPHCs in other tobacco products were taken from published literature and compared to VERVE®.

Results

HPHC (unit/portion)	Time (months)	Chews		Analytical Limits		Discs		Analytical Limits	
		Blue Mint	Green Mint	LOQ	LOD	Blue Mint	Green Mint	LOQ	LOD
Acetaldehyde (µg) CAS# 75-07-7	1	BLOQ	BLOQ	0.100	0.056	BLOQ	BLOQ	0.025	0.014
	6	BLOQ	BLOQ			BLOQ	BLOQ		
	12	0.488 (0.034)	0.472 (0.071)			BLOQ	BLOQ		
Arsenic (ng) CAS# 7440-38-2	1	20.72 (1.84)	BLOQ	20.32	6.70	ND	ND	5.08	1.68
	6	BLOQ	20.64 (1.77)			ND	ND		
	12	20.45 (1.27)	21.53 (1.54)			ND	ND		
B[a]P (ng) CAS# 50-32-8	1	ND	ND	5.0	2.0	ND	ND	5.0	2.0
	6	ND	ND			ND	ND		
	12	ND	ND			ND	ND		
Cadmium (ng) CAS# 7440-43-9	1	ND	ND	14.12	4.66	7.26 (1.93)	7.56 (1.03)	3.53	1.17
	6	ND	ND			4.55 (1.47)	4.15 (1.01)		
	12	ND	ND			4.66 (3.30)	BLOQ		
Crotonaldehyde(mg) CAS# 4170-30-3	1	BLOQ	BLOQ	0.100	0.040	ND	ND	0.025	0.010
	6	ND	ND			ND	ND		
	12	ND (NA)	ND			ND	ND		
Formaldehyde (mg) CAS# 50-00-0	1	0.393 (0.028)	0.602 (0.054)	0.100	0.064	0.859 (0.211)	0.861 (0.155)	0.025	0.016
	6	BLOQ	0.424 (0.050)			0.561 (0.036)	0.541 (0.034)		
	12	BLOQ	BLOQ			BLOQ	BLOQ		
Nicotine total (mg) CAS# 54-11-5	1	1.35 (0.04)	1.37 (0.04)	0.48	0.12	1.37 (0.02)	1.42 (0.03)	0.50	0.12
	6	1.47 (0.03)	1.54 (0.03)			1.38 (0.04)	1.45 (0.02)		
	12	1.33 (0.03)	1.36 (0.03)			1.33 (0.01)	1.33 (0.01)		
Nicotine free (mg) CAS# 54-11-5	1	0.06 (0.01)	0.08 (0.01)	NA	NA	0.55 (0.04)	0.58 (0.02)	NA	NA
	6	0.03 (0.003)	0.04 (0.003)			0.38 (0.01)	0.44 (0.01)		
	12	0.01 (0.001)	0.02 (0.001)			0.31 (0.01)	0.33 (0.01)		
NNK (ng) CAS# 64091-91-4	1	ND	BLOQ	1.0	0.2	0.27 (0.04)	BLOQ	0.25	0.05
	6	BLOQ	BLOQ			0.38 (0.02)	0.34 (0.04)		
	12	ND	ND			0.38 (0.04)	0.32 (0.04)		
NNN (ng) CAS# 16543-55-8	1	ND	ND	2.0	0.6	0.65 (0.04)	1.27 (0.27)	0.50	0.20
	6	ND	ND			0.95 (0.07)	1.48 (0.23)		
	12	ND	ND			0.74 (0.00)	1.32 (0.22)		

Product Comparisons

HPHC	Units/ Portion	VERVE® Maximum Levels[1]	Smokeless Tobacco (General® Snus)[2,3]	Cigarettes[4]	Nicorette® Fresh Mint™ Gum
Acetaldehyde	µg	0.488	20.7	1690	BLOQ
Arsenic	ng	21.53	74.63	11.3	47.3
B(a)P	ng	ND	ND	20.5	ND
Cadmium	ng	7.561	179.22	114	50.7
Crotonaldehyde	µg	<0.1	0.685	59.2	ND
Formaldehyde	µg	0.861	5.55	91.2	BLOQ
Nicotine (Total)	mg	1.54	10.9	2.59	2.02
Nicotine (Free)	mg	0.58	5.03	NR	2.00
NNK	ng	0.38	304	145	ND
NNN	ng	1.48	1080	284	ND

ND=not detected; NR=not reported; BLOQ=below limit of quantitation.
[1] Data represent the maximum values (mean (95% CI)) for different products analyzed during 12 months storage time. B(a)P levels were below the detection limit of 2.00 ng/portion
[2] Data obtained from Stepanov et al., (2008) (Average levels), based on average levels reported for General® snus. Note that the observations were converted from µg/portion to ng/portion.
[3] Data for HPHCs not reported by Stepanov et al. (2008), i.e., arsenic and cadmium, therefore we include values reported for General® snus Original Portion from Borgerding et al. (2012). Note that the observations were converted from ng/g of tobacco to ng/portion (since General® snus contains 1 g of tobacco) and from dry weight basis to as-is basis using the moisture content reported (50.9%).
[4] Data represent the mean (S.D.) for Marlboro 100's Box cigarettes smoked under Health Canada smoking conditions ((Oldham et al., 2014)(Appendix Table 2)).

Observations & Conclusions

- In the absence of an established list for novel, oral tobacco-derived nicotine products, we tested the products for constituents described in the abbreviated list of HPHCs in smokeless tobacco products from the 2012 Draft Guidance, Reporting Harmful and Potentially Harmful Constituents in Tobacco Products and Tobacco Smoke Under Section 904(a)(3) of the Federal Food, Drug, and Cosmetic Act.
- Chemical analyses of VERVE® products demonstrated low levels of HPHCs. Differences in HPHC levels (e.g., arsenic or cadmium) between product format (disc or chew) are possibly related to differences in ingredients (source unknown). Tobacco specific nitrosamine (TSNA) levels, NNN and NNK, were below levels of analytical quantitation in the chew product format and only slightly above levels of analytical quantitation in the discs product format.
- We demonstrate that the HPHCs listed in the 2012 FDA Draft Guidance for Smokeless Tobacco products are at levels substantially lower in VERVE® products than levels found in cigarette smoke with the exception of arsenic. Additionally, the levels of HPHCs in the products are lower than those reported for the recently FDA authorized General® snus products.
- The VERVE® products contain nicotine (~1.5 mg) at levels similar to nicotine polacrilex (NRT) gum (2-4 mg). While Nicorette® Gum is not a tobacco product and VERVE® is not intended for cessation indications, a comparison of HPHCs is relevant since both contain similar levels of USP grade tobacco-derived nicotine and similar oral routes of administration. As noted above, we observe higher levels of arsenic in VERVE® Chews than cigarette smoke, however, arsenic levels were lower than those measured in Nicorette® Gum.
- Since the products are noncombustible products, we expect that combustion-related HPHCs (e.g., carbon monoxide) would be absent.
- HPHC measurements over 1-year showed minimal variations in HPHCs over time. We conclude from these observations that the products do not present any additional risks contributed by HPHCs during the shelf life of the products.
- A toxicological risk assessment was conducted for all quantifiable HPHCs (except nicotine), with an estimated daily exposure assumption of 100% bioavailability of the VERVE® maximum level from 16 portions/day. When compared to an established regulatory value (i.e., California OEHHA No Significant Risk Levels or ICH Q3D Permissible Daily Exposures for elemental impurities), estimated daily exposure to each of the HPHCs was below the established lifetime regulatory limit for each analyte which suggests negligible toxicological concern for lifetime exposure to VERVE® products.

References

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