Quantification of Nicotine Related Impurities in Novel, Oral Tobacco-Derived Nicotine Products

Timothy L. Danielson, Christopher B. McFarlane, Michael R. Noe, Yezdi B. Pithawalla, Willie J. McKinney Jr.

Altria Client Services LLC, 601 East Jackson Street, Richmond, VA 23219 USA

72nd Tobacco Science Research Conference, 16-19 September 2018 - Memphis, Tennessee USA

Altria
Altria Client Services

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, such as VERVE®, 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). As a result, these deemed tobacco products will ultimately require a market authorization from FDA through the premarket tobacco application (PMTA) pathway to either remain on the market or to enter interstate commerce. Section 910(b)(1)(B) of the FD&C Act requires "that your PMTA include a full statement of the properties of the new tobacco product. The 'full statement of the properties' of the new tobacco product should include a full narrative description of the tobacco product, including: Established shelf life of the product to include data establishing the stability of the product through the stated shelf life." The US and European Pharmacopeias recommend purity specifications for nicotine intended for pharmaceutical products; however, there are no official purity specifications recommended for the nicotine used in tobacco products. We developed a sensitive, selective, and robust analytical method (LC-MS) to quantify nicotine-related impurities listed in Pharmacopeia guidelines in VERVE® Discs and Chews and monitored these impurities over time. This selective and sensitive method provides data suitable for quantitative risk assessments and for stability studies.

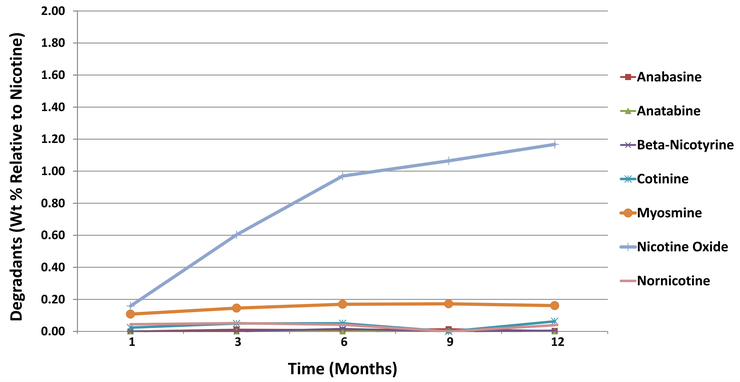
Methodological Relevance

- FDA has not issued guidance to inform the evaluation of product/nicotine impurities or stability for oral tobacco products containing tobacco-derived nicotine.
- We measured 8 nicotine and nicotine-related impurities based on specifications in the European Pharmacopeia Council of Europe.[1]
- The U.S. Pharmacopeia (USP) recommends the measurement of nicotine degradants and impurities in nicotine intended for pharmaceuticals.[2]
- USP-grade nicotine has no single impurities greater than 0.3% (3 mg/g) and total impurities less than 0.8% (8 mg/g) on a weight percent basis relative to nicotine at the time of manufacture.
- An acceptance limit approach consistent with the International Conference on Harmonization (ICH) Guidance Q3B(R2) – Impurities in New Drug Products, July 2006 Revision 2, provides a reasonable and appropriate approach for evaluating nicotine stability.[3]
- Both the Center for Drug Evaluation and Research (CDER) and the Center for Biologics Evaluation and Research (CBER) also adopted this guidance.
- Per ICH, "For a given degradation product, its acceptance criterion should be established by taking into account its acceptance criterion in the drug substance (if applicable), its qualified level, its increase during stability studies..." Furthermore, "each acceptance criterion should be set no higher than the qualified level of the given degradation product."
- Considering that nicotine-N´-oxide is a metabolite of nicotine and is also less acutely toxic than nicotine, we determine a higher threshold level of 3.0% for nicotine-N´-oxide as acceptable because it would not result in additional toxicological concern.
- 4% to 7% of metabolized nicotine is excreted as nicotine-N´-oxide[4,5] and nicotine-N´-oxide is a less acutely toxic metabolite compared to nicotine.[6,7]

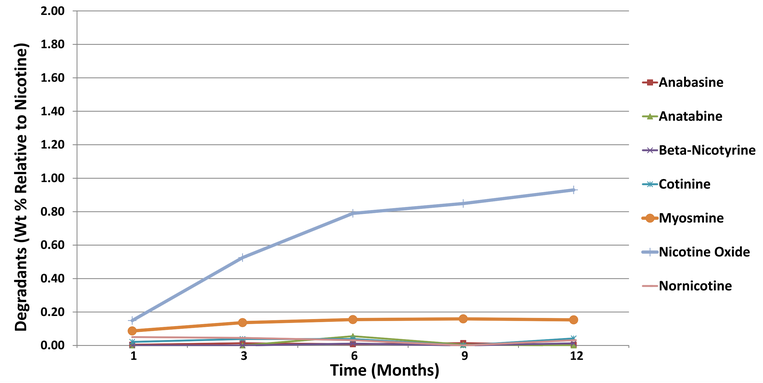
Impurity	Acceptance Limit (expressed as % of total nicotine)
Anabasine	0.5%
Anatabine	0.5%
Nornicotine	0.5%
β-Nicotyrine	0.5%
Cotinine	0.5%
Myosmine	0.5%
Nicotine-N'Oxide	0.5% at manufacture and 3.0% at end of shelf-life

Results

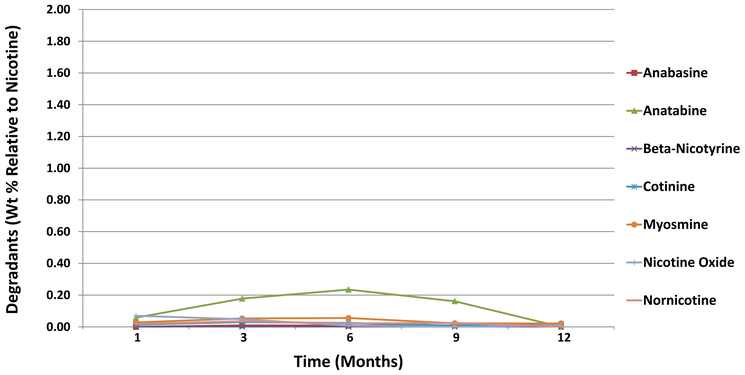
VERVE® Blue Mint Discs



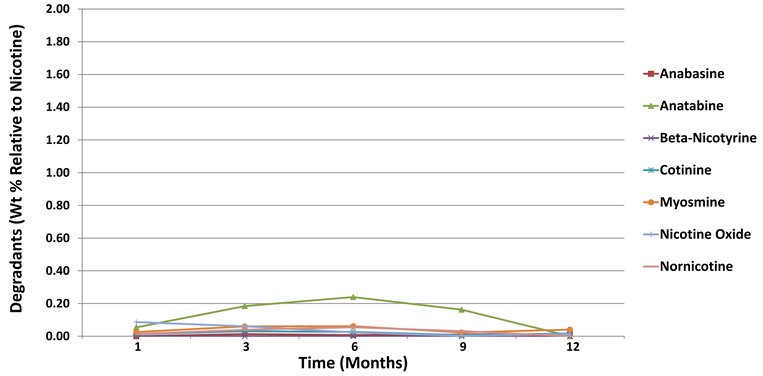
VERVE® Green Mint Discs



VERVE® Blue Mint Chews



VERVE® Green Mint Chews



Data shown are the mean value of 3 production lots. Product was stored in commercial packaging at 25 °C ± 2 °C / 60% RH ± 5% RH environmental conditions for 12 months duration. Each lot of product was tested at N=3 or 7.

Constituent	CAS #	Units	Discs LOQ	Discs LOD	Chews LOQ	Chews LOD
Myosmine	532-12-7	% (Wt/Wt)	0.175	0.070	0.175	0.070
Nornicotine	5746-86-1	% (Wt/Wt)	0.175	0.070	0.175	0.070
Anatabine	2743-90-0	% (Wt/Wt)	0.175	0.070	0.175	0.070
Anabasine	13078-04-1	% (Wt/Wt)	0.175	0.070	0.175	0.070
β-Nicotyrine	487-19-4	% (Wt/Wt)	0.175	0.070	0.175	0.070
Cotinine	486-56-6	% (Wt/Wt)	0.175	0.070	0.175	0.070
Nicotine-oxide	51095-86-4	% (Wt/Wt)	0.175	0.070	0.175	0.070

Methods

- Determination of nicotine impurities and degradants in VERVE® Products was conducted by LC-MS. Prior to analysis, VERVE® Discs are milled to <1.25 mm using a Wiley Mill. VERVE® Chews are extracted as-is, without any sample preparation. An aliquot of the sample is extracted with a methanol and water solution (Chews) or methanol solution (Discs) containing internal standard. Samples are mixed thoroughly and an aliquot of the extract is analyzed by LC-MS. Nicotine degradants are reported in units of weight percent relative to the nicotine target value.

- Methods are validated according to the ALCS Analytical Sciences Method Validation Guidelines, which follow the ICH Harmonised Tripartite Guideline, Validation of Analytical Procedures: Methodology Q2B, November 1996, in addition to the U.S. Department of Health and Human Services Food and Drug Administration Guidance for Industry, Bioanalytical Method Validation, Draft Guidance, Revision 1, September 2013. All ALCS analytical methods used for stability testing are accredited to the ISO 17025:2005 standard by the American Association for Laboratory Accreditation (A2LA, accreditation No. 0660.01).

Observations and Conclusions

- 1-year shelf-life studies showed minimal variations in nicotine related degradants and impurities over time. At the end of one year, all nicotine impurities measured were <0.5% (expressed as % nicotine) with the exception of nicotine N-oxide in VERVE® Discs which was <3.0% (expressed as % nicotine).
- Within the targeted shelf life of one year, the nicotine-N'-oxide levels in the Discs products did not exceed the ALCS acceptance threshold of 3.0% wt/wt of USP grade tobacco-derived nicotine added to the product.
- With the exception of anatabine in Chews and nicotine N'-oxide in Discs, the majority of nicotine degradants and impurities measured were either not detected (below the limit of detection, BLOD) or below the limit of quantitation (BLOQ).
- In the absence of Guidance from FDA regarding acceptable levels of nicotine degradants over the shelf-life of tobacco products containing USP grade tobacco-derived nicotine, we have developed an approach adapted from ICH Guidelines Q3B(R2) and based on sound scientific principles.

References

1. Council of Europe. (2012) European Pharmacopoeia 7.0 European Directorate for the Quality of Medicines and Healthcare. Strasbourg, France.
2. United States Pharmacopeia and the National Formulary. (2015) USP 38-NF 33. Rockville, Maryland: The United States Pharmacopeia Convention Inc.
3. ICH. (2006) International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use Guidelines Q3B(R2): Impurities in New Drug Products.
4. Benowitz N, et al. (2009) Nicotine chemistry, metabolism, kinetics and biomarkers. Handb Exp Pharmacol 192, 29-60.
5. Hukkanen J, et al. (2005) Metabolism and disposition kinetics of nicotine. Pharmacol Rev 57, 79-115.
6. Doolittle DJ, et al. (1995) The genotoxic potential of nicotine and its major metabolites. Mutat Res, 344, 95-102.
7. Uwai K. (2003) Synthesis and reduction of (S)-(-)-nicotine-N'-oxide and N,N'-dioxides by rat liver S-9 fraction. ARKIVOC online Journal of Organic Chemistry, 2003, 211.

Acknowledgments

The authors would like to thank **Susan Plunkett** and **Lara Baker** for their significant contributions to this work.

This poster may be accessed at www.altria.com/ALCS-Science