Comprehensive Evaluation of Aerosol Constituents from Juul Virginia Tobacco 5.0% Using Non-targeted Analysis: LC-HRMS Analysis of E-Vapor Aerosols, Data Acquisition and Processing

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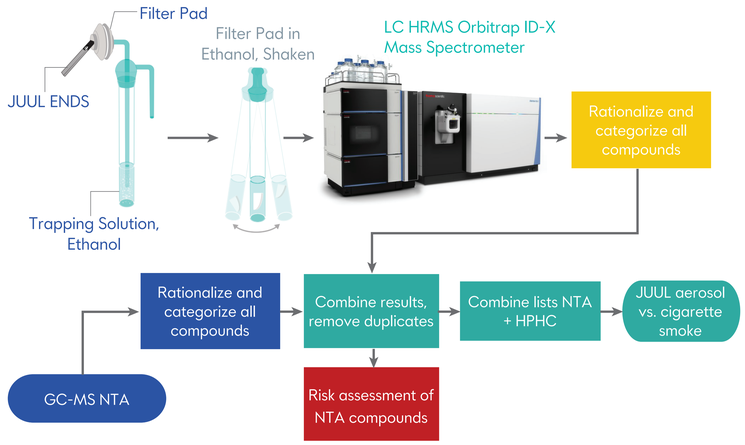
Introduction

Exposure to the smoke resulting from tobacco combustion is a primary risk factor for many diseases, including heart disease, chronic lung disease, and cancer[1]. Tobacco smoke from cigarettes causes the most premature and preventable death in the US and is an important public health consideration[2]. Chemical constituents resulting from tobacco combustion in conventional cigarettes have been well studied and characterized over the past few decades[3,4].

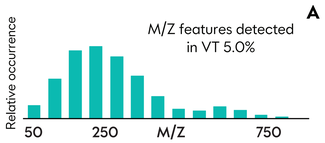
JUUL is an Electronic Nicotine Delivery Systems (ENDS) which aerosolizes a formulation of mostly propylene glycol and glycerol and delivers nicotine. ENDS were invented in recent years[5], therefore their aerosol composition has not been as extensively characterized as cigarette smoke. The objective of the work was to use a liquid chromatography-based non-targeted method, which is complimentary to a gas chromatography-based non-targeted method, to defensibly characterize as much of the chemical composition of the aerosol as possible.

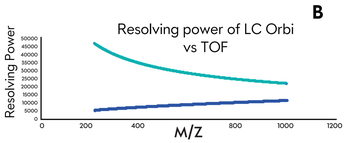
Overview of Methodology

In order to achieve our objective, JUULpods filled with Virginia Tobacco 5% nicotine (VT5) e-liquids were aerosolized, and the samples were analyzed high performance liquid chromatography and High Resolving power Mass Spectrometer (HRMS). The resulting data were processed in an automated fashion using Compound Discoverer software to provide descriptive statistics and allow for a defensible differential comparison of the aerosol from a JUUL versus an aerosol collection blank. Compounds which had a p-value of less than 0.05 and which were estimated at or above 0.5 ug/g were assigned identifications. Compound identifications were rationalized for a thorough understanding of the aerosol composition.

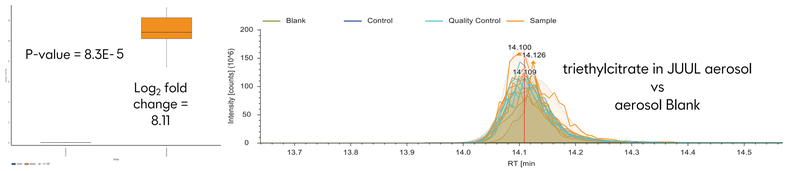


LC-HRMS Instrument Suitability



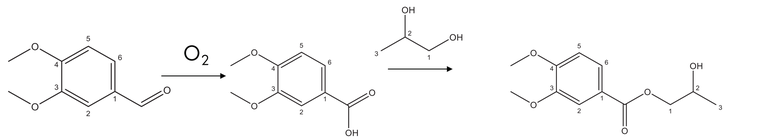


LC-HRMS Defensible Compound Detection



P-value is quantitative, accepted, defensible, objective, reproducible

Compound Rationalization VT5



Veratraldehyde (ingredient) oxidized to 3,4-dimethoxybenzoic acid which reacts with + PG to form 2-hydroxypropyl 3,4-dimethoxybenzoate

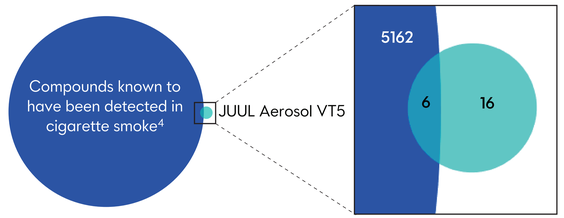
LC HRMS NTA Results for VT5

Category of Compounds Identified	Intense Number of Compounds	Non-Intense Number of Compounds	Average (ug/g) Intense	Average (ug/g) Non-Intense
Ingredients	3	3	780.0	733.3
Reaction Products	14	14	420.8	325.6
Not Rationalized	4	2	9.6	30.0
Total	21	19	1210.4	1088.9

Non-flavorant primary constituents quantitated in targeted analyses account for ~ 90% of aerosol collected mass (excluding water).

Unrationalized compounds make up approximately 0.00096% and 0.003% of aerosol mass for intense and non-intense aerosol collections, respectively.

LC HRMS NTA Results for VT5



Venn diagram showing that of the 22 total compounds detected by NTA using LC HRMS in JUUL aerosol, 6 have been detected in cigarette smoke[4]

Discussion

We presented the non-targeted LC-HRMS workflow, and approach to data processing, rationalization and LC HRMS- NTA results. JUUL nicotine-salt based e-liquids were vaped and aerosols were collected. Samples were analyzed using LC-HRMS and Compound Discoverer software was used to detect constituents, assign compound identification and perform a defensible differential analysis vs an aerosol blank. Approximately 5162 compounds were reportedly detected in cigarette smoke[4] and approximately 100 of these are known carcinogens, cocarcinogens, and/or mutagens[6]. Such chemical complexity has made it difficult to determine the active constituents responsible for the tobacco-related health risks of smoking and it is now being realized that the health effects of this complex mixture are likely to result from a combined effect of these chemicals through multiple mechanisms rather than as result of the effects of a single smoke constituent[6]. In contrast, for JUUL Virginia Tobacco with 5.0% nicotine, using LC HRMS, a total of 21 compounds were identified in the aerosol condensate using the intense puffing regime and 19 compounds in the aerosol condensate using the non-intense puffing regimen. Because 18 compounds were in common in intense and non-intense collections, there were a total of 22 unique compounds detected in both aerosols. Reaction products accounted for the greatest number of compounds, 66% and 73% in intense and non-intense, while ingredients, excluding nicotine, water, propylene glycol, benzoic acid and glycerol, were 14% and 16% of the total number of compounds, respectively. Unrationalized compounds make up approximately 0.00096% and 0.003% of aerosol mass for intense and non-intense aerosol collections.

References

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