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Nicotine in tobacco product aerosols: 'It's déjà vu all over again'

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ABSTRACT

Introduction The distribution of nicotine among its free-base (fb) and protonated forms in aerosolised nicotine affects inhalability. It has been manipulated in tobacco smoke and now in electronic cigarettes by the use of acids to de-freebase nicotine and form 'nicotine salts'.

Methods Measurements on electronic cigarette fluids (e-liquids) were carried out to determine (1) the fraction of nicotine in the free-base form (α_{fb}) and (2) the levels of organic acid(s) and nicotine. Samples included JUUL 'pods', 'look-a-like/knock-off' pods and some bottled 'nicotine salt' and 'non-salt' e-liquids.

Results $\alpha_{fb} = 0.12 \pm 0.01$ at 40°C ($\approx 37^\circ\text{C}$) for 10 JUUL products, which contain benzoic acid; nicotine protonation is extensive but incomplete.

Discussion First-generation e-liquids have $\alpha_{fb} \approx 1$. At cigarette-like total nicotine concentration (Nic_{tot}) values of ~ 60 mg/mL, e-liquid aerosol droplets with $\alpha_{fb} \approx 1$ are harsh upon inhalation. The design evolution for e-liquids has paralleled that for smoked tobacco, giving a 'déjà vu' trajectory for α_{fb} . For 17th-century 'air-cured' tobacco, α_{fb} in the smoke particles was likely ≥ 0.5 . The product $\alpha_{fb} \text{Nic}_{tot}$ in the smoke particles was high. 'Flue-curing' retains higher levels of leaf sugars, which are precursors for organic acids in tobacco smoke, resulting in $\alpha_{fb} \approx 0.02$ and lowered harshness. Some tobacco cigarette formulations/designs have been adjusted to restore some nicotine sensory 'kick/impact' with $\alpha_{fb} \approx 0.1$, as for Marlboro. Overall, for tobacco smoke, the de-freebasing trajectory was $\alpha_{fb} \geq 0.5 \rightarrow \sim 0 \rightarrow \sim 0.1$, as compared with $\alpha_{fb} = \sim 1 \rightarrow \sim 0.1$ for e-cigarettes. For JUUL, the result has been, perhaps, an optimised, flavoured nicotine delivery system. The design evolution for e-cigarettes has made them more effective as substitutes to get smokers off combustibles. However, this evolution has likely made e-cigarette products vastly more addictive for never-smokers.

INTRODUCTION

Nicotine can exist in a free-base (fb) form and in two protonated forms (figure 1). For electronic cigarette fluids (e-liquids) and the aerosolised droplets created therefrom, both the total nicotine concentration (Nic_{tot}) and the fraction of nicotine in the free-base form (α_{fb}) can vary.¹ Fb nicotine is volatile and gaseous fb nicotine is directly sensible. Protonated nicotine is not volatile and so has been referred to in the tobacco industry as 'bound nicotine'.² First-generation e-liquids were simply fb nicotine dissolved in a mix of propylene glycol (PG) and/or glycerol (GL), with $\alpha_{fb} = 1$, and Nic_{tot} in the range of 6–24 mg/mL. In comparison, in the droplets making up tobacco smoke particulate matter (PM), Nic_{tot} values are typically much higher (~ 60

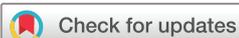
mg/mL).³ Electronic cigarette (e-cigarette) aerosols with high values of the product $\alpha_{fb} \text{Nic}_{tot}$ can be expected to be harsh upon inhalation, as with $\alpha_{fb} = 1$ and $\text{Nic}_{tot} = 60$ mg/mL.¹ Non-harsh cigarette-like nicotine levels in aerosolised e-liquids therefore require $\alpha_{fb} \ll 1$. This can be achieved by the addition of an acid to the PG/GL/nicotine mix, for example, benzoic acid, as in the JUUL product line.

Given the large market share quickly achieved by JUUL^{4 5} and its youth-oriented e-cigarette demographic,⁶ the goal of this work was to determine α_{fb} values and acid levels in the e-liquids from JUUL and look-a-like/knock-off product⁷ competitors, available as of October 2018, and thereby characterise the use of acid additives to moderate fb nicotine delivery, and thus harshness, while maintaining high total nicotine delivery. The measured α_{fb} values were compared with those for first-generation e-cigarette products. The first-generation e-cigarette \rightarrow JUUL trajectory is compared with that for the smoke aerosol from colonial-era air-cured tobacco \rightarrow flue-cured tobacco (1850s forward) \rightarrow the modern Marlboro cigarette. The measurements were carried out by application of ¹H NMR spectroscopy (hereafter, NMR).^{1 8 9} As outlined by Duell *et al*,¹ NMR is a method that allows the reliable determination of α_{fb} values in e-liquids without any alteration of the sample, for example, without water addition, which changes nicotine protonation chemistry. The e-liquid results are examined in the context of the acid+nicotine first protonation equilibrium constant.

Nicotine protonation and α_{fb}

Predicting the extent of nicotine protonation (including α_{fb} ¹⁰) in any solution requires knowledge of the governing acid/base concentrations and their medium-dependent equilibrium constants. Fully protonated nicotine carries two protons (figure 1) with acidity constants $K_a^{\text{NicH}_2^+} (=K_{a,1})$ and $K_a^{\text{NicH}^+} (=K_{a,2})$. Measurement of K_a values in tobacco smoke and e-liquids is very difficult but relatively easy in water. In water, reported values at 25°C are $\text{p}K_a^{\text{NicH}^+} = 8.01$ and $\text{p}K_a^{\text{NicH}_2^+} = 3.10$.¹¹ At 37°C, the values are 7.65 and 2.77, respectively.¹² In water, $\text{pH} \leq 4$ is required for significant ($\geq 10\%$) NicH_2^+ .

When conditions are such that there is not an equivalent excess of acid over nicotine (so that total molar-based concentration of monoprotic acid (C_{HA})/total molar-based concentration of nicotine (C_{Nic}) is ≤ 1), or the protonating acid is weak for the medium, NicH_2^+ can be neglected and the dominant protonation of fb nicotine (Nic) occurs according to



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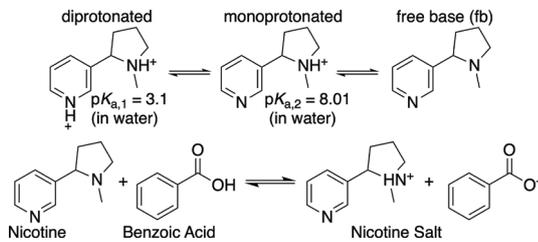


Figure 1 Top: the distribution of nicotine in vape and tobacco aerosols primarily involves two forms; centre: NicH^+ (monoprotonated), which is non-volatile; and right: free-base (fb) nicotine, which is volatile. The fraction of the fb (α_{fb}) depends on the acid/base conditions. In water at 25°C, $\text{p}K_{\text{a},2} = 8.01$. Bottom: so-called 'nicotine salts' in electronic cigarette liquids are formed by adding an organic acid (benzoic acid is depicted here) to the formulation, producing a lower α_{fb} that depends on the ratio of acid:nicotine, as well as temperature and solvent conditions.



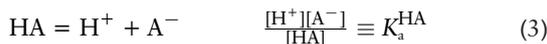
so that

$$\alpha_{\text{fb}} \equiv \frac{[\text{Nic}]}{[\text{Nic}] + [\text{NicH}^+] + [\text{NicH}_2^{2+}]} \approx \frac{[\text{Nic}]}{[\text{Nic}] + [\text{NicH}^+]} \quad (2)$$

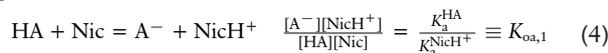
The diprotonated form may not be negligible for all e-liquids, including some non-JUUL high-acid brands examined experimentally here. Each bracketed term in equations (1) and (2) is a molar concentration (and not a chemical activity) so that $K_{\text{a}}^{\text{NicH}^+}$ and all the other K values herein are constant-medium-type equilibrium constants, analogous to K values as discussed by Pankow,¹³ and dependent on the nature of the particular solution medium.

Net protonation reaction

In a liquid medium (eg, the PG/GL matrix and water), the acid dissociation reaction of an acid, HA (eg, benzoic acid and acetic acid), is



The overall reaction for monoprotection of Nic by HA is given by equations (1) + (3), so that



$K_{\text{oa},1}$ is dimensionless because both the forward and backward reactions are bimolecular: any mol-proportional concentration scale can be used. For water, $K_{\text{oa},1}$ values for different acids can be calculated; $K_{\text{a}}^{\text{NicH}^+}$ values and K_{a}^{HA} values for many important acids are individually well known because pH is easily measured in water: at 37°C, for benzoic acid and vanillin (a common e-liquid flavour additive), $\text{p}K_{\text{a}}^{\text{HA}} = 4.20$ and 7.27, respectively.¹⁴ For these two acids with nicotine in water at 37°C, then $K_{\text{oa},1} = 10^{3.45}$ and $10^{0.38}$, respectively. In contrast, in PG and GL, either individually or as a mixture, $K_{\text{a}}^{\text{NicH}^+}$ and K_{a}^{HA} values for relevant acids are unknown. The species H^+ , however, does not appear in equation (4), and so $K_{\text{oa},1}$ values can be directly measured in PG and GL solutions/mixtures.

Let C_{HA} and C_{Nic} be the total molar-based concentrations of HA and nicotine as initially added to a PG/GL solution. (C_{Nic} and Nic_{tot} are proportional; Nic_{tot} has units of mg/mL.) Neglecting formation of the diprotonated species, establishment of a reaction equilibrium will lead to protonation such that $[\text{NicH}^+] = [\text{A}^-] = x$:

$$K_{\text{oa},1} = \frac{x^2}{(C_{\text{HA}} - x)(C_{\text{Nic}} - x)} \quad (5)$$

so that $\alpha_{\text{fb}} = (C_{\text{Nic}} - x) / C_{\text{Nic}}$. Because the reaction is bimolecular and $K_{\text{oa},1}$ is dimensionless, for any mass concentration of total nicotine, we can set $C_{\text{Nic}} = 1$ and $C_{\text{HA}} = C_{\text{HA}}/C_{\text{Nic}}$. Then $\alpha_{\text{fb}} = 1 - x$, and

$$K_{\text{oa},1} = \frac{(1 - \alpha_{\text{fb}})^2}{\left(\frac{C_{\text{HA}}}{C_{\text{Nic}}} - 1 + \alpha_{\text{fb}}\right) \alpha_{\text{fb}}} \quad (6)$$

When $K_{\text{oa},1}$ and $C_{\text{HA}}/C_{\text{Nic}}$ are known, then equation (6) can be solved for α_{fb} either numerically or by the quadratic equation. For the latter, $a = K_{\text{oa},1} - 1$, $b = (K_{\text{oa},1} C_{\text{HA}}/C_{\text{Nic}} - K_{\text{oa},1} + 2)$, and $c = -1$; the root $\alpha_{\text{fb}} = (-b + \sqrt{b^2 - 4ac}) / (2a)$ is chosen so that $\alpha_{\text{fb}} > 0$. Cases involving $K_{\text{oa},1} = 1$ are not second order ($a = 0$), and so reduce to $\alpha_{\text{fb}} = 1 / (1 + C_{\text{HA}}/C_{\text{Nic}})$. When protonation is favoured, the reliability of equations (5-6) will decrease for $C_{\text{HA}}/C_{\text{Nic}} > 1$ due to an increasing importance of NicH_2^{2+} . For the special case of $C_{\text{HA}}/C_{\text{Nic}} = 1$, then

$$K_{\text{oa},1} = \frac{(1 - \alpha_{\text{fb}})^2}{\alpha_{\text{fb}}^2} \quad (7)$$

$$\alpha_{\text{fb}} = \frac{1}{1 + \sqrt{K_{\text{oa},1}}} \quad (8)$$

LABORATORY METHODS

NMR determinations of α_{fb} , nicotine and acid concentrations

JUUL e-liquid 'pods' were purchased from JUUL. Other pod brands (ZOOR, SMPO, Myle, ZiiP and Eon Smoke) and bottles of e-liquids (Fuzion Vapor) were purchased from online suppliers. Bottles of 'nicotine salt' e-liquids (Salt Bae⁵⁰ and Pacha Mama Salts) were purchased from a vape shop in Portland, Oregon. Glacial acetic acid was obtained from Mallinckrodt Chemicals (Staines-upon-Thames, England). Tertbutylamine (98%) was obtained from Sigma-Aldrich (St. Louis, Missouri, USA). DMSO- d_6 , D 99.9%, was obtained from Cambridge Isotope Laboratories (Andover, Massachusetts, USA). Precision coaxial NMR inserts (WGS-5BL-SP and WGS-5BL) and precision NMR tubes (535-PP-7) were purchased from Wilmad (Vineland, New Jersey, USA).

Monoprotonated and fb nicotine standards, which were used to calculate the fb nicotine fraction in each sample, were prepared by adding acetic acid or tertbutylamine to the e-liquids until the limiting NMR chemical shifts were achieved. In the present study, standards were prepared using the following commercial e-liquids: 'Mango'-flavoured JUUL, 'Apple'-flavoured ZOOR, 'Cake'-flavoured ZOOR and 'Blue Raspberry Lemonade'-flavoured Salt Bae⁵⁰. In our previous work, standards were prepared from nicotine-containing PG/GL samples rather than actual commercial e-liquids, resulting in small differences in the α_{fb} values reported here. Various commercial e-liquid standards were prepared because dissimilarities in the e-liquid compositions (such as the presence of benzoic acid or levulinic acid) can result in slightly different limiting chemical shifts for the mono-protonated and fb nicotine reference samples. Appropriate reference samples were matched to the tested commercial e-liquids by using the most similar compositions as determined by analysis of ^1H NMR spectra. In particular, this was executed by matching samples and reference standards containing the same primary acid(s) (if present), that is, benzoic acid or levulinic acid. Details for the references used for each sample can be found in online supplementary table S-1. α_{fb} was calculated using the difference between the chemical shifts of two aromatic nicotine protons and the nicotine methyl resonance, respectively. The average was

Table 1 Listed versus measured nicotine contents, molar acid/nicotine ratios and free-base fraction (α_{fb}) for a selection of JUUL pod liquids, 'look-a-like/knock-off' pod liquids, and bottled e-liquids (additional details can be found in online supplementary table S-1).

Brand 'flavour'	Nicotine wt%* (listed/measured)	Nic _{tot} (mg/mL) (measured)	Acid†	Molar acid‡/nicotine ratio (C _{HA} /C _{Nic})	α_{fb} ‡
JUUL 'pods' (5% nicotine by wt.)					
JUUL 'Cool Mint'/JUUL(8) group	5.0/5.1	60	BA	0.97	0.13
JUUL 'Classic Menthol'/JUUL(8) group	5.0/4.9	58	BA	0.98	0.13
JUUL 'Crème Brûlée'/JUUL(8) group	5.0/5.1	60	BA	0.97	0.12
JUUL 'Fruit Medley'/JUUL(8) group	5.0/5.0	59	BA	0.99	0.12
JUUL 'Cool Cucumber'/JUUL(8) group	5.0/5.0	59	BA	1.00	0.11
JUUL 'Classic Tobacco'/JUUL(8) group	5.0/5.0	59	BA	1.00	0.11
JUUL 'Virginia Tobacco'/JUUL(8) group	5.0/5.1	60	BA	1.00	0.11
JUUL 'Mango'/JUUL(8) group	5.0/5.2	62	BA	0.99	0.09
JUUL 'pods' (3% nicotine by wight)					
JUUL 'Virginia Tobacco'/JUUL(2) group	3.0/3.0	35	BA	0.94	0.14
JUUL 'Mint'/JUUL(2) group	3.0/3.0	35	BA	1.04	0.11
				For 10 JUUL liquids: ave.±1 SD	0.12±0.01
Other 'nicotine salt' formulation 'pods'					
EM	6.0/4.0	47	BA	3.43	0.00§
SS	5.0/4.3	51	BA	1.02	0.09
ZiC	5.0/3.3	38	BA	4.03	0.01§
ZiM	5.0/3.5	41	BA	3.71	0.00§
FF	5.0/2.3	27	BA	0.76	0.15
ZA	5.0/4.5	53	LA	0.22¶	0.19§
ZMI	5.0/4.4	52	LA	0.29¶	0.17
ZC	5.0/4.8	57	LA	0.25¶	0.14
Nicotine salt bottled e-liquids					
Fuji-50	4.2/4.2	50	U	U	0.08
BRL-50	4.2/4.1	49	U	U	0.01
Fuji-25	2.1/2.0	24	U	U	0.08
BRL-25	2.1/2.1	25	U	U	0.02
Non-'salt' bottled e-liquids					
Ec-24	2.0/1.0	12	–	U	0.98§
RwC-24	2.0/2.8	33	U	U	0.70
UB-24	2.0/0.9	11	U	U	0.84
Ec-6	0.5/0.5	5	–	U	0.96
UBP-6	0.5/0.4	5	U	U	0.53
UB-6	0.5/0.4	5	U	U	0.43
RwC-6	0.5/0.5	6	U	U	0.08

* Calculated by integrating ^1H NMR resonances for nicotine relative to propylene glycol and glycerol resonances in each e-liquid and obtaining the mole per cent values, which were then converted into wt% values. These values do not reflect the variable presence of water, accounting for water affecting nicotine mg/mL by less than ~10%.

† Values by liquid chromatography for JUUL products, by NMR for all others. Ratio computed based on the main acid contributor for each liquid.

‡ Different chemical shift references were used based on composition. The value presented is that for the average at 40°C. For details, see the Methods section.

§ Only one α_{fb} value was obtained by ^1H NMR due to either resonance overlap or peak broadening.

¶ Incomplete characterisation of the acid content.

ave., average; BA, benzoic acid; BRL-25, Salt Bae⁵⁰ 'Blue Raspberry Lemonade'; BRL-50, Salt Bae⁵⁰ 'Blue Raspberry Lemonade'; C_{HA}, total molar-based concentrations of HA; C_{Nic}, total molar-based concentrations of nicotine; Ec-6, Fuzion Vapor 'Ectoplasm'; Ec-24, Fuzion Vapor 'Ectoplasm'; EM, Eon Smoke 'Mango'; α_{fb} , fraction of nicotine in the free-base form; FF, SMPO 'Full Fruit'; Fuji-25, Pacha Mama Salts 'Fuji'; Fuji-50, Pacha Mama Salts 'Fuji'; LA, levulinic acid; Nic_{tot}, total nicotine concentration; NMR, ^1H NMR spectroscopy; RwC-6, Fuzion Vapor 'Roundhouse with Cream'; RwC-24, Fuzion Vapor 'Roundhouse with Cream'; SS, Myle 'Summer Strawberry'; U, unknown/undetected; UB-6, Fuzion Vapor 'Unicorn Blood'; UB-24, Fuzion Vapor 'Unicorn Blood'; UB-6, Fuzion Vapor 'Unicorn Blood Prime'; wt%, weight per cent; ZA, ZOOR 'Apple'; ZC, ZOOR 'Cake'; ZiC, ZiP 'Cappuccino'; ZiM, ZiP 'Mango'; ZMI, ZOOR 'Mint Ice'.

then calculated (\pm the difference between the two values divided by 2).¹

Concentric tube samples containing each e-liquid were prepared for α_{fb} analyses per previous methods,¹ and samples containing a single drop of each e-liquid in 500 μL of DMSO- d_6 were used for composition analysis, owing to the better shim that can be achieved with a lower sample concentration. A 600 MHz NMR spectrometer was used to execute zg30 ^1H experiments using parameters reported previously and heteronuclear single quantum coherence spectroscopy (HSQC) experiments, as needed.¹ Thus, each e-liquid sample was placed in a precision coaxial NMR insert and the lock solvent, DMSO- d_6 , was placed in the outer 5 mm NMR tube. ^1H NMR experiments were conducted using a TXI ("Triple Resonance") probe and at 40°C in order to increase the molecular tumbling rate, improving the shim. Sixteen scans were collected using the zg30 pulse sequence;

a relaxation delay (D1) of 3 s between each scan was used; the size of the real spectrum (TD) was 65 536 data points; and the spectral width (SW) was 15 ppm, with the transmitter frequency offset (O1P) set to 6 ppm, giving a total experiment time of 2 min per sample.

Spectra for composition determinations were assessed using integration analysis. After phasing and baseline correction, the chemical components (eg, PG, GL, nicotine, and benzoate or levulinate) were analysed using the resonance(s) with the least overlap. The resulting integrations were used to calculate the mole percent of each component, which was then used to calculate the weight percents (wt%). Other details about the calculation of α_{fb} have been reported previously,¹ except with a modification to the fb and monoprotonated nicotine standards used as described above.

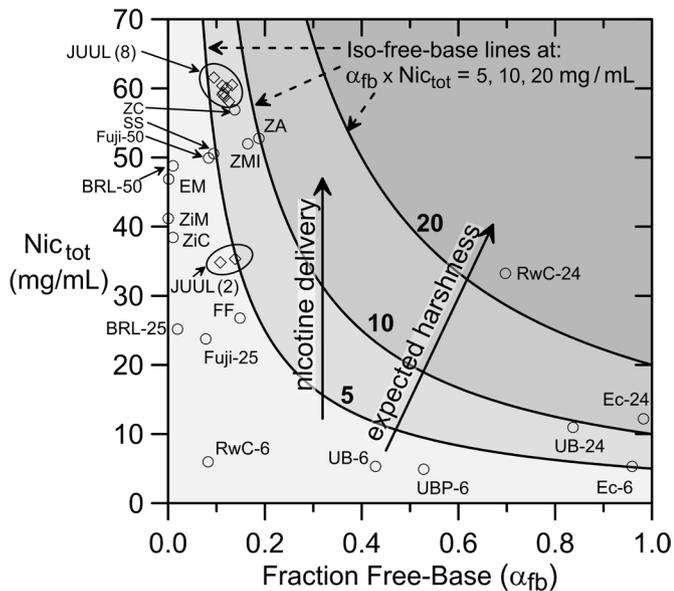


Figure 2 The expected harshness of a nicotine-containing product is influenced by both the free-base fraction (α_{fb}) and the total nicotine concentration (Nic_{tot}). BRL-25, Salt Bae⁵⁰ 'Blue Raspberry Lemonade'; BRL-50, Salt Bae⁵⁰ 'Blue Raspberry Lemonade'; Ec-6, Fuzion Vapor 'Ectoplasm'; Ec-24, Fuzion Vapor 'Ectoplasm'; EM, Eon Smoke 'Mango'; FF, SMPO 'Full Fruit'; Fuji-25, Pacha Mama Salts 'Fuji'; Fuji-50, Pacha Mama Salts 'Fuji'; Nic_{tot} total nicotine concentration; RwC-6, Fuzion Vapor 'Roundhouse with Cream'; RwC-24, Fuzion Vapor 'Roundhouse with Cream'; SS, Myle 'Summer Strawberry'; UB-6, Fuzion Vapor 'Unicorn Blood'; UB-24, Fuzion Vapor 'Unicorn Blood'; UB-6, Fuzion Vapor 'Unicorn Blood Prime'; ZA, ZOOR 'Apple'; ZC, ZOOR 'Cake'; ZIC, ZiP 'Cappuccino'; ZiM, ZiP 'Mango'; ZMI, ZOOR 'Mint Ice'.

$K_{oa,1}$ determinations

Based on equation (6), values of $K_{oa,1}$ were determined for benzoic acid at 40°C in 43/57 PG/GL by weight (48/52 by mol). The mixture was amended with benzoic acid and nicotine to give $C_{HA} = 3.31 \times 10^{-4}$ mol/mL and $C_{Nic} = 3.28 \times 10^{-4}$ mol/mL ($C_{HA}/C_{Nic} = 1.01$, nicotine at 4.6 wt%). A second mixture was prepared with a PG/GL ratio of 32/68 by weight (36/64 by mol) (similar to that currently represented by JUUL) and amended with benzoic acid to give $C_{HA} = 3.38 \times 10^{-4}$ mol/mL and nicotine at $C_{Nic} = 3.30 \times 10^{-4}$ mol/mL ($C_{HA}/C_{Nic} = 1.03$, nicotine 4.6 wt%). To investigate the effects of water on nicotine protonation, an aliquot of the second mixture was amended with water at 5% (by volume). Values of $K_{oa,1}$ were also calculated for benzoic acid at 40°C based on the data for the JUUL products in table 1, with $C_{HA}/C_{Nic} \approx 1$, as verified here by a liquid chromatography (LC) method discussed elsewhere.¹⁵ ¹H NMR results gave slightly different $C_{HA}:C_{Nic}$ ratios (online supplementary table S-1); because NMR spectra can be subject to resonance overlap in these cases, due to the presence of flavourants, the LC-determined $C_{HA}:C_{Nic}$ ratios were used for the calculations herein.

$K_{oa,1}$ values were also determined for vanillin at 40°C in 45/55 PG/GL by weight (49/51 by mol). The mixture was amended with nicotine and three levels of vanillin. The three solutions were characterised by (1) $C_{HA} = 1.80 \times 10^{-4}$ and $C_{Nic} = 3.61 \times 10^{-4}$ mol/mL ($C_{HA}/C_{Nic} = 0.50$) (nicotine at 5.1 wt%), (2) $C_{HA} = 3.67 \times 10^{-4}$ and $C_{Nic} = 3.59 \times 10^{-4}$ mol/mL ($C_{HA}/C_{Nic} = 1.02$) (nicotine at 5.1 wt%) and (3) $C_{HA} = 5.15 \times 10^{-4}$ and $C_{Nic} = 3.41 \times 10^{-4}$ mol/mL ($C_{HA}/C_{Nic} = 1.51$) (nicotine at 4.9 wt%).

JUUL aerosol PM determinations

A fully charged JUUL device was equipped with a JUUL 'Classic Menthol' 5% nicotine pod and vaped using the CORESTA puff method (55 mL puff volume, 3 s long) and employed vaping methods described previously.^{16,17} The JUUL device (+e-liquid pod) was weighed before and after the generation of five puffs to obtain the mass of aerosol produced over the five puffs.

RESULTS

Protonation in e-liquids

Table 1 lists the measured (by ¹H NMR) versus manufacturer-listed nicotine concentrations and the measured α_{fb} values (online supplementary figure S-3 visually depicts the data in a bar chart). The e-liquids tested included those for JUUL pods, other look-a-like/knock-off pods, bottled nicotine salt e-liquids and early-generation (ie, non-salt) bottled e-liquids. The agreement between the listed and actual nicotine contents varied among brands; in this work, the measured values were used; online supplementary figure S-1 illustrates the differences among the e-liquids. Table 1 also gives C_{HA}/C_{Nic} ; the acids were fully identifiable by NMR for the first 14 e-liquids, and the presence of at least one acid was identified for the first 18 e-liquids. C_{HA}/C_{Nic} values varied widely among the brands (see also online supplementary figure S-2). Online supplementary figure S-5 is a comparison of the ¹H NMR spectra for two e-liquids with differing ratios of benzoic acid relative to nicotine; for one, $C_{HA}/C_{Nic} \approx 1$, and for the other, $C_{HA}/C_{Nic} \approx 4$.

Figure 2 is a plot of measured Nic_{tot} versus α_{fb} . Lines of constant fb concentration as given by the product $\alpha_{fb} Nic_{tot}$ plot as hyperbolas (see also the issue cover graphic for Duell *et al*).¹ All the e-liquids with $C_{HA}/C_{Nic} \approx 1$ with benzoic acid were found to be characterised by similar α_{fb} values (0.09–0.14). As noted earlier, the inhalation harshness of a nicotine aerosol is related to the fb concentration in the aerosol liquid, as given by $\alpha_{fb} Nic_{tot}$. Values for $\alpha_{fb} Nic_{tot}$ can be computed from the data in table 1 (see also online supplementary figure S-4). Bookending these values, e-liquids with $C_{HA}/C_{Nic} > 1$ gave $\alpha_{fb} \approx 0$, and some e-liquids that were not marketed as nicotine salts gave α_{fb} values as high as 0.98.

Besides carboxylic acids (eg, benzoic acid and levulinic acid) as protonating agents, the prevalent flavour phenols vanillin and ethyl vanillin can contribute to protonation of nicotine; these two weak acids can be found at high concentrations in some e-liquids.¹⁸ Such an effect on α_{fb} may be indicated in the α_{fb} values for the 'Roundhouse with Cream' flavour formulations for two different Nic_{tot} values, 33 and 6 mg/mL, with $\alpha_{fb} = 0.70$ and 0.08, respectively. Assuming a constant phenol flavourant level, the lower α_{fb} for the lower nicotine-level may have been caused in part by a higher total acids:nicotine ratio.

$K_{oa,1}$ determinations

The $K_{oa,1}$ values (40°C \approx 37°C) obtained here are provided in table 2. For benzoic acid, values were determined in JUUL liquids and in two laboratory-prepared mixtures (with added $\sim 1:1$, by mol, benzoic acid:nicotine): 43/57 PG/GL and 32/68 PG/GL (similar to JUUL) by weight. The average $K_{oa,1}$ value for the JUUL e-liquids tested was 67, which is within a factor of 3 of $K_{oa,1}$ for 43/57 PG/GL by weight, where $K_{oa,1} = 26$ and with $K_{oa,1}$ for 32/68 PG/GL by weight, where $K_{oa,1} = 31$. When 5 vol% water was added to the 32/68 PG/GL (by weight) mixture, $K_{oa,1} = 51$; this sample may be the most comparable to the JUUL liquids, which contain some water. For vanillin in $\sim 45/55$ PG/GL by weight, $K_{oa,1}$ averaged 0.0089, about 6000 times smaller than that for benzoic acid. (At constant C_{Nic} , the $K_{oa,1}$ values

Table 2 Values of $K_{oa,1}$ (40°C) for benzoic acid and vanillin in e-liquid formulations.

	C_{HA}/C_{Nic}	α_{fb}	$K_{oa,1}$	Log $K_{oa,1}$
Benzoic acid				
JUUL 'Cool Mint' (5% nicotine)	0.97*	0.13	58	1.77
JUUL 'Classic Menthol' (5% nicotine)	0.98*	0.13	53	1.72
JUUL 'Crème Brûlée' (5% nicotine)	0.97*	0.12	72	1.86
JUUL 'Fruit Medley' (5% nicotine)	0.99*	0.12	59	1.77
JUUL 'Cool Cucumber' (5% nicotine)	1.00*	0.11	65	1.82
JUUL 'Classic Tobacco' (5% nicotine)	1.00*	0.11	65	1.82
JUUL 'Virginia Tobacco' (5% nicotine)	1.00*	0.11	65	1.82
JUUL "Mango" (5% nicotine)	0.99*	0.09	115	2.06
JUUL "Virginia Tobacco" (3% nicotine)	0.94*	0.14	66	1.82
JUUL 'Mint' (3% nicotine)	1.04*	0.11	48	1.68
Averages for JUUL	0.99±0.03 SD	0.12±0.01	67±18	1.81±0.10
43/57 PG/GL (by wt.)+nicotine+benzoic acid (final nicotine level=4.6 wt%)	1.01	0.16	26	1.41
32/68 PG/GL (by wt.)+nicotine+benzoic acid (final nicotine level=4.6 wt%)	1.03	0.14	31	1.49
32/68 PG/GL (by wt.)+nicotine+benzoic acid+5% (by vol.) water (final nicotine level=4.5 wt%)	1.03	0.11	51	1.71
Vanillin				
45/55 PG/GL (by wt.)+nicotine+vanillin (final nicotine level=5.1%)	0.50	0.95	0.0058	-2.23
45/55 PG/GL (by wt.)+nicotine+vanillin (final nicotine level=5.1%)	1.02	0.91	0.0089	-2.05
45/55 PG/GL (by wt.)+nicotine+vanillin (final nicotine level=4.9%)	1.51	0.88	0.0120	-1.92
Averages for vanillin		Ave.±1 SD	0.0089±0.0025	-2.07±0.13

*By liquid chromatography for both nicotine and benzoic acid, using a method discussed elsewhere.¹⁵

ave, average; C_{HA} , total molar-based concentrations of HA; C_{Nic} , total molar-based concentrations of nicotine; α_{fb} , fraction of nicotine in the free-base form; GL, glycerol; PG, propylene glycol; vol., volume; wt., weight.

for vanillin may indicate some tendency to increase with an increasing $C_{HA}:C_{Nic}$ ratio; an increasingly ionic medium would be expected to favour the $HA+Nic=A^{-}+NicH^{+}$ reaction, due to Debye-Hückel effects.)

JUUL aerosol PM determinations

The average mass lost per puff, for five puffs, was 4.4 mg, which when divided by the puff volume (55 mL) results in an average aerosol PM of ~80 mg/L, or $80 \times 10^6 \mu\text{g}/\text{m}^3$. This is only slightly greater than the high end of the range for tobacco cigarettes, from 13 to $63 \times 10^6 \mu\text{g}/\text{m}^3$.¹⁹

DISCUSSION

Past was prologue: Vu – tobacco smoke

The chemistry changes during the rapid evolution of e-cigarettes closely parallel the events that occurred during the centuries-long development of smoked tobacco. The tobacco that the English colony of Jamestown in Virginia exported to England beginning in 1619 was dark, 'air-cured' tobacco. Air curing occurs by slow drying (6–8 weeks) in ventilated barns. Air-cured (aka 'dark', 'brun', 'black')^{20 21} tobacco generally produces tobacco smoke that is much more basic than other tobacco types.^{20 22} Leaf sugars, which are precursors of tobacco-smoke organic acids, are generally lost during slow air curing; it is this loss that accounts for the relatively high proportions of fb nicotine in the smoke aerosol droplets from air-cured tobacco²³ (figure 3). Regardless of smoke basicity/acidity, most tobacco smoke nicotine is in the smoke PM, distributed among the fb and protonated nicotine forms.¹⁰

Nicotine-related harshness of tobacco smoke has long been viewed as being correlated with smoke basicity, with basicity favouring PM nicotine being in the volatilisable and therefore sensible (harsh) fb form. Consider:

'...The presence of unprotonated nicotine in the smoke of French cigarettes and the observation that French smokers of black tobacco inhale less frequently than smokers in England

and the USA ... support our hypothesis that the pH is a determining factor in the "inhalability" of tobacco smoke'.²⁰

'...increasing the pH ... introduces a smoke with high physiological impact and a harsh bite, which would seem to offset the advantages gained from increased nicotine'.²⁴

'Flue-cured' (aka 'bright') tobacco was developed in the 1850s after the accidental discovery that rapid drying with heat yields a bright yellow leaf that produces a noticeably milder smoke.^{25–27} Indeed, flue-cured tobacco remains high in leaf sugars so that the resulting smoke contains numerous organic acids.^{22 27} While historical measurements of 'smoke pH' both inside and outside the industry were indisputably flawed in absolute terms, within

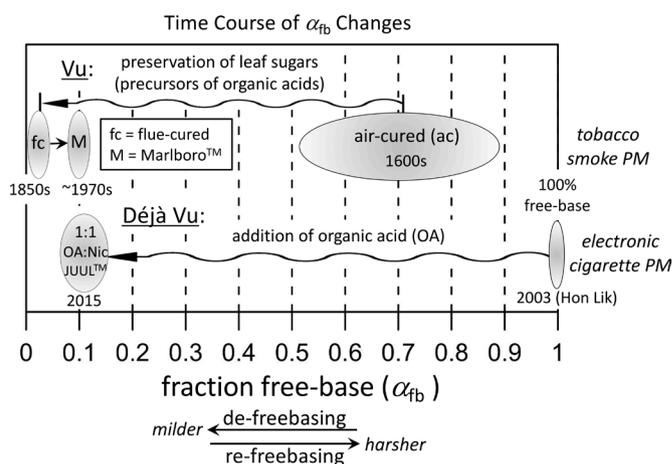


Figure 3 A visual representation of the historical changes in α_{fb} in tobacco smoke PM (top) in comparison to how electronic cigarette fluids and their associated aerosols have been changed (bottom). fc, flue-cured; α_{fb} , fraction of nicotine in the free-base form; M, Marlboro; Nic, nicotine; OA, organic acid; PM, particulate matter.

a given protocol (eg, the ‘pH electrode’ method), relative comparisons have likely been meaningful, so it is relevant that ‘smoke pH’ was found by the industry to be strongly negatively correlated with both leaf sugar levels and leaf sugar/leaf nicotine ratios.²⁸ In 1970, Armitage and Turner²⁹ wrote:

‘It is usually believed that the majority of cigarette smokers inhale to varying degrees the smoke which they take into their mouths, whereas the majority of cigar smokers do not.... One of the most striking differences between cigarette and cigar smoke is the pH of the smoke. The pH of T 29 cigarettes by the method of Grob... was 5.35, whereas the pH of the C 1 cigars was 8.5’.²⁹

Overall, as compared with tobacco smoke from air-cured tobacco, for flue-cured tobacco, the fraction of the PM nicotine in the fb form is much lower. The role of acids in converting nicotine to a protonated, ‘salt’ form in tobacco smoke has long been understood. In 1909, Garner²³ wrote:

‘Apparently the only possible explanation of this pronounced effect on the sharpness of the smoke is that in the presence of the citric acid the nicotine enters the smoke in the form of a salt rather than in the free state, and thereby loses its pungency while still exerting the usual physiological effect’.²³

Modern measurement of α_{fb} values in cigarette smoke PM began ~15 years ago.^{3 30} In ‘American blend’ cigarettes, flue-cured tobacco dominates. Thus, in measurements with tobacco smoke PM from nine commercial brands of cigarettes sold in the USA, Pankow *et al*³ reported relatively low α_{fb} values, ranging between ~0.01 (GPC) and ~0.10 (Marlboro). Two other, atypical commercial brands gave higher α_{fb} values: Gauloises Brunes (relatively high in air-cured tobacco) at $\alpha_{fb} = 0.25$ and American Spirit/Maroon at $\alpha_{fb} = 0.36$.³ Overall, together with historical evidence, it can be concluded that air-cured tobacco was characterised by very high α_{fb} values (≥ 0.4 and perhaps ≥ 0.5).

Figure 3 summarises the main tobacco product development stages: (1) Aerosol PM produced from smoked tobacco products in the early 1600s contained high levels of fb nicotine and so was harsh on inhalation; the α_{fb} in the PM was likely greater than 0.5. (2) Flue-curing allowed retention of plant acids in the leaf during the curing process, bringing α_{fb} values in smoke PM to ~0.01 (very mild). (Note here that Proctor has aptly commented that manufacturers of cigars giving high fb smoke might similarly make their products more inhalable by adding acids, a process that he has termed ‘de-freebasing’.²⁷) (3) For Marlboro, by using additives and/or blend manipulation^{31 32} to accomplish a Goldilocks principle solution (ie, not too harsh, not too mild), α_{fb} was brought to ~0.1 for a tolerable/desired level of impact/harshness. Consider, by analogy, human affinity for the sensory ‘bite’ of carbonated beverages.³³ Much has been written on the technical efforts of Philip Morris and its competitors to understand and provide some nicotine ‘impact’.^{31 32} Overall, the tobacco smoke trajectory was $\alpha_{fb} \geq 0.5 \rightarrow \sim 0 \rightarrow \sim 0.1$.

Present: ‘Déjà Vu’ – e-cigarette aerosols

Stepanov and Fujioka³⁴ were the first to consider the acid/base chemistry of nicotine in e-liquids. Most early versions of e-cigarettes used PG/GL-based fluids with total nicotine levels of 6–24 mg/mL and $\alpha_{fb} \approx 1$ (nicotine+PG/GL is characterised by $\alpha_{fb} \approx 1$).¹⁶ It has been verified that such e-liquids correspondingly generate e-cigarette aerosol PM with $\alpha_{fb} \approx 1$.¹⁶ When e-liquids including some acid and their resulting aerosol PM are compared, total nicotine levels have been found to be similar,^{1 35} as have the α_{fb} values.¹ Following our prior work,¹ the product $\alpha_{fb} \text{Nic}_{\text{tot}}$ can be used to compare e-liquid fb delivery values, with

JUUL products having been found to be de-freebasing to $\alpha_{fb} \approx 0.1$.

Cigarette smoke PM generally contains nicotine levels that are much higher than those in early e-liquids. Assuming unit density for cigarette smoke PM, values of ~54 mg/mL for the GPC brand and 72 mg/mL for Marlboro (‘red’) have been reported.³ If e-cigarettes were to attempt cigarette-like nicotine levels along with $\alpha_{fb} \approx 1$, then with $\alpha_{fb} \text{Nic}_{\text{tot}} \approx 50\text{--}70$ mg/mL, the aerosol would be expected to be exceedingly harsh on inhalation. Enter JUUL, which was launched in 2015, offering its nicotine+benzoic acid pods (5% (w/w) nicotine, ~59 mg/mL); table 1 (and the results of Pankow *et al*¹⁵) indicate a $\approx 1:1$ molar ratio of benzoic acid to nicotine. As indicated earlier for $K_{\text{oa},1} = 38.5$ (table 2), equation (8) then gives $\alpha_{fb} = 0.14$ (see therefore figure 3), so that $\alpha_{fb} \text{Nic}_{\text{tot}} \approx 8.3$ mg/mL. This is very similar to what has been found for Marlboro cigarettes ($\alpha_{fb} \text{Nic}_{\text{tot}} \approx 0.10 \times 72$ mg/mL = 7.2 mg/mL).³ The trajectory for e-cigarettes has then been a partial de-freebasing according to $\alpha_{fb} = \sim 1 \rightarrow \sim 0.1$ (as compared with $\alpha_{fb} \geq 0.5 \rightarrow \sim 0 \rightarrow \sim 0.1$ for most smoked tobacco). Thus, taken with the PM results discussed earlier, the JUUL design characteristics provide effective cigarette-like delivery of nicotine, including (1) high total nicotine concentration in the liquid (Nic_{tot} , mg/mL); (2) low but not zero fb fraction (α_{fb}); (3) cigarette-like concentrations of fb nicotine in the

What this paper adds

- ▶ The chemistry of nicotine in aerosols from smoked tobacco and electronic cigarette (e-cigarette) products underlie their parallel product developments and popularities, and therefore their abuse liabilities.
- ▶ The development over more than four centuries of smoked tobacco products (de-freebasing then partial re-freebasing) is compared with the development of e-cigarette products during the last 16 years (extensive but incomplete de-freebasing). An explanation is provided of what has been perceived by some as inconsistent that (1) tobacco companies during the mid-20th century were interested in increasing the value of the free-base nicotine fraction (by the partial re-freebasing step) in the products’ smoke aerosol particulate matter, denoted α_{fb} , while (2) some e-cigarette manufacturers have moved to decrease it (by the extensive but incomplete de-freebasing).
- ▶ Values of α_{fb} are measured by ¹H nuclear magnetic resonance spectroscopy for a total of 29 products, including JUUL, JUUL look-a-like/knock-off products, as well as bottles of ‘nicotine salt’ and ‘non-salt’ e-liquids.
- ▶ The overall trajectory of smoked tobacco development is discussed as having been $\alpha_{fb} \geq 0.5 \rightarrow \sim 0.02 \rightarrow \sim 0.1$. A ‘Déjà Vu’ trajectory of $\alpha_{fb} \approx 1 \rightarrow \sim 0.1$ has been followed in the design of the nicotine-containing liquids used in e-cigarettes, as supported by the measurements of α_{fb} .
- ▶ A mathematical framework and equilibrium chemistry model are developed for understanding nicotine protonation chemistry in e-cigarette fluids in terms of $K_{\text{oa},1}$, the first overall nicotine protonation constant.
- ▶ De-freebasing has undoubtedly made e-cigarettes more effective as substitutes to get smokers off combustibles. However, as with smoked tobacco, it is likely that e-cigarettes have also been made vastly more addictive for never-smokers. The full public health implications of widely prevalent e-cigarette use will only become fully apparent perhaps a decade hence.

aerosol droplets ($\alpha_{\text{nb}}\text{Nic}_{\text{tot}}$, mg/mL); and (4) relatively low, cigarette-like PM; along with (5) optional flavours and no tobacco-smoke odour: a flavoured (at present) e-cigarette analogue of Marlboro.

The trajectory in figure 3 for smoked tobacco allowed cigarettes to become much more addictive, abused, and deadly than would have been the case if smoked tobacco remained of an air-cured type. The evolution of e-cigarettes has followed a similar overall trajectory. It is undoubtedly true that this evolution has made e-cigarettes more effective as substitutes to get smokers off combustibles. However, exactly as occurred with smoked tobacco, this evolution has made e-cigarette products vastly more addictive for never-smokers. The full public health implications of widely prevalent e-cigarette use will only become fully apparent perhaps a decade hence.

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REFERENCES

- Duell AK, Pankow JF, Peyton DH. Free-Base Nicotine Determination in Electronic Cigarette Liquids by ^1H NMR Spectroscopy. *Chem Res Toxicol* 2018;31:431–4.
- Morie GP. Fraction of protonated and unprotonated nicotine in tobacco smoke at various pH values. *Tobacco Sci* 1972;16.
- Pankow JF, Tavakoli AD, Luo W, et al. Percent free base nicotine in the tobacco smoke particulate matter of selected commercial and reference cigarettes. *Chem Res Toxicol* 2003;16:1014–8.
- Krishnan-Sarin S, Jackson A, Morean M, et al. E-cigarette devices used by high-school youth. *Drug Alcohol Depend* 2019;194:395–400.
- Ramamurthi D, Chau C, Jackler RK. JUUL and other stealth vaporisers: hiding the habit from parents and teachers. *Tob Control* 2019;28:610–6.
- Gentzke AS, Creamer M, Cullen KA, et al. *Vital signs: tobacco product use among Middle and high school students - United States, 2011–2018*. *MMWR Morb Mortal Wkly Rep* 2019;68:157–64.
- La Vito A. Juul asks regulators to stop 18 companies from selling lookalike e-cigarettes, 2018. Available: <https://www.cnn.com/2018/10/03/juul-asks-regulatorsto-stop-companies-selling-lookalike-e-cigarettes.html> [Accessed 23 Sep 2019].
- Barsanti KC, Luo W, Isabelle LM, et al. Tobacco smoke particulate matter chemistry by NMR. *Magn Reson Chem* 2007;45:167–70.
- Pankow JF, Barsanti KC, Peyton DH. Fraction of free-base nicotine in fresh smoke particulate matter from the Eclipse "cigarette" by ^1H NMR spectroscopy. *Chem Res Toxicol* 2003;16:23–7.
- Pankow JF. A consideration of the role of gas/particle partitioning in the deposition of nicotine and other tobacco smoke compounds in the respiratory tract. *Chem Res Toxicol* 2001;14:1465–81.
- Barlow RB, Hamilton JT. Effects of pH on the activity of nicotine and nicotine monomethiodide on the rat diaphragm preparation. *Br J Pharmacol Chemother* 1962;18:543–9.
- Clayton PM, Vas CA, Bui TTT, et al. Spectroscopic studies on nicotine and nornicotine in the UV region. *Chirality* 2013;25:288–93.
- Pankow JF. *Aquatic chemistry concepts*. 2nd edn. Boca Raton, FL: Taylor and Francis, 2019.
- Christensen JJ, Hansen LD. *Handbook of proton ionization heats and related thermodynamic quantities*. New York: J. Wiley and Sons, 1976.
- Pankow JF, Kim K, McWhirter KJ, et al. Benzene formation in electronic cigarettes. *PLoS One* 2017;12:e0173055.
- Duell AK, McWhirter KJ, Korzun T, et al. Sucralose-enhanced degradation of electronic cigarette liquids during Vaping. *Chem Res Toxicol* 2019;32:1241–9.
- Duell AK, Pankow JF, Gillette SM, et al. Boiling points of the propylene glycol + glycerol system at 1 atmosphere pressure: 188.6–292 °C without and with added water or nicotine. *Chem Eng Commun* 2018;205:1691–700.
- Omaie EE, McWhirter KJ, Luo W, et al. High concentrations of flavor chemicals are present in electronic cigarette refill fluids. *Sci Rep* 2019;9:2468.
- Pankow JF. Calculating compound dependent gas-droplet distributions in aerosols of propylene glycol and glycerol from electronic cigarettes. *J Aerosol Sci* 2017;107:9–13.
- Brunnemann KD, Hoffmann D. The pH of tobacco smoke. *Food Cosmet Toxicol* 1974;12:115–24.
- De Stefani E, Fierro L, Barrios E. Black (air-cured) and blond (flue-cured) tobacco and cancer risk III: oesophageal cancer. *Eur J Cancer* 1993;29:763–6.
- Elson LA, Betts TE, Passey RD. The sugar content and the pH of the smoke of cigarette, cigar and pipe tobaccos in relation to lung cancer. *Int J Cancer* 1972;9:666–75.
- Garner WW. The Relation of Nicotine to the Burning Quality of Tobacco, Bureau of Plant Industry. In: *Bureau of plant industry – Bulletin no 141, miscellaneous papers*, 1909: 5–16.
- Chen L. pH of smoke: a review, report number N-170, internal document of Lorillard tobacco company, 18 PP, Bates NOS. 00118164/8181. Available: <https://www.industrydocumentslibrary.ucsf.edu/tobacco/docs/#id=ykhn01011976> [Accessed 1 May 2019].
- Caswell County Genealogy. Slade, Abisha b.15 SEP 1799, Caswell County genealogy. Available: <https://caswellcountync.org/genealogy/getperson.php?personID=I2238&tree=tree1> [Accessed 25 Jun 2019].
- Caswell County Hist Association, from the Caswell County Historical Association. Bright leaf tobacco process, 2010. Available: <http://ncchca.blogspot.com/2010/01/bright-leaf-tobacco-process.html> [Accessed 14 May 2019].
- Proctor RN. *Golden holocaust: origins of the cigarette catastrophe and the case for abolition*. Berkeley and Los Angeles, California: University of California Press, 2012.
- Correlation of maximum smoke pH of Flue-cured tobacco with leaf and smoke constituents, Bates number 504428668-504428671. Available: <https://www.industrydocuments.ucsf.edu/docs/#id=yqbg00081977> [Accessed 1 May 2019].
- Armitage AK, Turner DM. Absorption of nicotine in cigarette and cigar smoke through the oral mucosa. *Nature* 1970;226:1231–2.
- Watson CH, Trommel JS, Ashley DL. Solid-Phase microextraction-based approach to determine free-base nicotine in trapped mainstream cigarette smoke total particulate matter. *J Agric Food Chem* 2004;52:7240–5.
- Kessler D. The control and manipulation of nicotine and cigarettes (21 June 1994 testimony before the Subcommittee on health and the environment, Committee on energy and Commerce, us house of representatives). *Tob Control* 1994;3:362–9.
- Stevenson T, Proctor RN. The secret and soul of Marlboro: Phillip Morris and the origins, spread, and denial of nicotine freebasing. *Am J Public Health* 2008;98:1184–94.
- Wise PM, Wolf M, Thom SR, et al. The influence of bubbles on the perception carbonation bite. *PLoS One* 2013;8:e71488.
- Stepanov I, Fujioka N. Bringing attention to e-cigarette pH as an important element for research and regulation: Table 1. *Tob Control* 2015;24:413–4.
- El-Hellani A, Salman R, El-Hage R, et al. Nicotine and carbonyl emissions from popular electronic cigarette products: correlation to liquid composition and design characteristics. *Nicotine Tob Res* 2018;20:215–23.