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COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT (COT)

Potential toxicological risks from e-cigarettes. Paper 1: Characterisation of the aerosol droplet particle fraction

Background

1. During a horizon scanning exercise at the COT meeting in February 2016, the Committee considered the subject of the possible human health effects of e-cigarettes (electronic nicotine delivery systems or electronic non-nicotine delivery systems; ENDS/ENNDS) as a potential item for review. Members considered that this was a topic that should be evaluated by the COT. It was decided that a full systematic review would not be an efficient way to proceed, and the Committee recommended a more focussed review of three key areas: additives, nitrosamines produced by ENDS/ENNDS, and secondary exposure to exhaled products.

2. A scoping document (TOX/2016/25, attached at Annex A) reviewing these three areas was discussed by the Committee in July 2016, with the aim to set priorities for more in-depth reviews. From these discussions, a number of areas were agreed for further consideration. These were:

- the composition of particles
- bystander exposure to key analytes
- effects of long term inhalation of the main constituents and emissions
- the situation regarding flavourings (exposure, thermal products, toxicity on inhalation)
- exposure to metals from the device components

3. The Committee agreed that further discussion papers should be prepared to address the above questions. This paper addresses the first of these topics and reports studies that investigated the particulate matter in the aerosol produced from e-cigarette use.

Introduction

4. E-cigarettes (EC) are battery-powered devices containing a liquid (‘e-liquid’). The e-liquid is heated on use to produce an aerosol that is inhaled by the user (‘puffing’, ‘vaping’). EC were first introduced commercially in China in 2004 and

subsequently in the EU (2005) and USA (2007) as nicotine-delivery devices (Bansal and Kim 2016). The main consituent parts of an EC device are a mouthpiece, cartridge (tank) containing e-liquid, a heating element/atomizer, a microprocessor, a battery, and sometimes an LED light. Commercially available devices are sometimes categorised as first, second, or third generation. First-generation devices look like conventional cigarettes and thus are termed 'cigalikes'. Initial models comprised three principal parts; a lithium-ion battery, a cartridge and an atomizer. However, more recent models mostly consist of a battery connected to a 'cartomizer' (cartridge/atomizer combined), which may be replaceable, but is not refillable. Second-generation EC are larger and have less resemblance to tobacco cigarettes. They often resemble pens or laser pointers (hence the name, 'vape pens'). They have a high-capacity rechargeable lithium-ion battery and a refillable atomizer (sometimes referred to as a 'clearomizer'). Third-generation models ('advanced personal vapers', 'mods') are also refillable, have very-high-capacity lithium-ion batteries and are highly customisable (different coil options, power settings, tank sizes) (see Annex B).

5. E-liquid normally comprises a base material of propylene glycol (PG), with or without glycerol (generally referred to as vegetable glycerine, VG), plus water and optional ingredients such as nicotine and flavourings. The majority of the mass (around 90-95%) is made up of PG/VG, for which the proportions can vary. Nicotine concentrations are generally in the range of up to around 20 mg/mL, although some reports have suggested that nicotine contents listed on commercially available ECs/e-liquids do not always correlate well with actual levels measured in the products (Bansal and Kim 2016). The Tobacco and Related Products Regulations 2016[1] states that "nicotine-containing liquid which is presented for retail sale in an electronic cigarette or refill container must not contain nicotine in excess of 20 milligrams per millilitre" (Part 6, section 36(4)). There is also contradictory information as to whether this nicotine fraction is translated to the aerosol phase.

6. Several detailed analyses of e-liquids have been reported, and this area has been reviewed in TOX/2016/25 (Annex A) and in several publications (Cheng (2014); Famele et al. (2015); Bansal and Kim (2016)). Some constituents that have been identified include PG, VG, water, nicotine, carbonyls, volatile organic compound (VOCs), tobacco-specific nitrosamines (TSNAs), polycyclic aromatic hydrocarbons (PAHs), metals, ethanol, ethylene glycol, di-ethylene glycol, flavouring compounds, and phenolics. In addition, studies have investigated the composition of the EC 'vape' product (aerosol) (see, for example, Goniewicz et al. (2014); Margham et al. (2016)). This aerosol in fact comprises two major parts – the gas phase (vapour) and a particulate phase of suspended liquid droplets. Several techniques have been used to sample and analyse these components and this is an area that is still in development (see the recent review by Bansal and Kim (2016)). Components of the

[1] http://www.legislation.gov.uk/uksi/2016/507/contents/made

vapour phase include VOCs and carbonyls. The particulate phase comprises droplets that are formed when components within the e-liquid are heated and vapourise, then condense back into liquid aerosol as the gas cools. In addition, metal particles derived from the EC device or e-liquid may also be present (Hess et al. (2017); Williams et al. (2017)).

7. The following narrative focuses on investigations of the particulate droplet phase of EC mainstream aerosol. Studies of metal particle content and of second-hand EC aerosols (i.e. after exhalation) are not included as these aspects will be reviewed in future papers.

Evaluation of the particulate fraction of mainstream aerosol

8. Studies of EC aerosol particulate matter have focussed on evaluation of the physical characteristics (size distribution) of the particles. Size distribution of EC aerosol droplets is an important determinant of the amount and location of EC aerosol deposition within the respiratory tract, and an understanding of droplet size distribution is necessary to be able to model distribution and deposition in airway. This area is still relatively early in development for EC. A number of studies have been performed in recent years, using a range of different:

- methodologies and instruments (aerosol production, collection, and analysis);
- test materials (homemade or commercial EC devices and e-liquids, with/without nicotine, flavourings, varying PG/VG carrier proportions); and
- test parameters (aerosol dilution, temperature and relative humidity, puff parameters, real-time or delayed analysis).

9. Because of the current lack of standardisation in protocols and testing devices, the observations that have been reported are often inconsistent. For this reason, the studies summarised below are listed chronologically to follow the development of the field, followed by a short commentary of the main findings to date. Study details are also summarised in Table 1 (Annex C).

10. Ingebrethsen, Cole and Alderman (2012) demonstrated that the particle size measured in EC aerosol is highly dependent on the analytical method used, noting in particular the impact of dilution of the aerosol. In a comparison of two methods, namely spectral transmission (used to measure the undiluted sample) and ‘fast’ electrical mobility analysis (used to measure particle size distribution following high dilution of the aerosols; measurement range 5–1000 nm), the average mass particle diameters for EC aerosol (‘cartomizer’, e-liquid not described) were approximately ten-fold lower when measured in the diluted sample (approximately 20–50 nm at around 1:5000 dilution) compared with the undiluted sample (approximately 250–450 nm). Particle number concentration (PNC) calculated for mainstream aerosol was in

the $10^9/cm^3$ range in all cases. Smoke from conventional cigarettes (CC) showed a particle diameter range of approximately 200–400 nm. The smaller particle diameters reported in the diluted samples was attributed by the authors to an almost complete evaporation of aerosol particles at high dilution and under the conditions of the electrical mobility analysis. This was supported by gravimetric analysis, which indicated total particulate mass values similar to those obtained by spectral transmission. Authors thus suggested that data obtained via spectral transmission are the more useful starting basis for modelling of respiratory deposition of EC aerosols.

11. Schripp et al. (2013) reported that both heating temperature and 'aging[2]' time affected the size distributions of EC (tank-system/e-liquid without nicotine) aerosol particles collected in a 10 L emission chamber after a 3 s puff, measured by Fast Mobility Particle Sizer (FMPS) (measurement range, 5.6-560 nm). EC aerosol transferred directly to the collection chamber was aged for periods of 1–10 min before analysis. Two sets of conditions were investigated. Firstly, aging time-period was set at 5 min and the temperature in the collection chamber was maintained at either 23, 37 or 50 °C. A bimodal size distribution (maxima at around 60 nm and 110 nm) was observed at a chamber temperature of 23 °C, but the distribution was unimodal at higher temperatures (maximum around 60 nm at 37 °C; 40 nm at 50 °C). Secondly, the collection-chamber temperature was maintained at a constant 37 °C, whilst aging occurred for different time periods. A bimodal size distribution was observed after 1 min aging (maxima at around 50 nm and 110 nm) but the distribution was unimodal (around 60 nm) after aging for longer time periods (3, 5, 7, 10 min). Similarly to Ingebrethsen et al. (2012), Schripp and colleagues suggested that this 'particle shrinking' could be attributed to evaporation under the experimental conditions.

12. A study by Zhang, Sumner and Chen (2013) indicated that particles produced by a single puff from an EC cartomizer (with 16 mg/mL nicotine) containing either PG or VG e-liquids had peaks at diameters of 117 nm and 180 nm, respectively, measured by Scanning Mobility Particle Sizer (SMPS) (measurement range, 10–1000 nm, dilution not reported). Similar results were obtained from a CC. Aging for 10 s or 40 s was associated with slightly increased measured particle sizes for PG (121 nm and 131 nm, respectively) but not VG. Bimodal particle size distributions with peak diameters at that of the single puff and also at around 400 nm were observed for both PG and VG in steady-state vapour (continuous puffing into the 50 mL test chamber). The authors estimated that deposition of EC aerosol by volume in the human lung (20-27%) would be slightly less than that of tobacco cigarette smoke (25-35%).

[2] Time between aerosol production and measurements.

13. Alderman et al. (2014) used cascade impaction to measure particle size distribution of undiluted aerosol from three EC, two of which had been evaluated in a previous study by the same group (Ingebrethsen et al. 2012). Ninety-five percent of the particle mass was observed in the size range 280–1420 nm, and count median diameter (CMD[3]) values correlated relatively well with those reported previously: e.g. 262 nm (Alderman) vs. 339 nm (Ingebrethsen); and 261 nm (Alderman) vs. 265 nm (Ingebrethsen), for the two ECs evaluated in both studies. Analysis of EC aerosol components (glycerine, PG, nicotine, water) captured by a Cambridge filter pad (used for gravimetric analysis) indicated that the majority of the mass of glycerine, PG, and nicotine was in the condensed particulate phase, although results for water were inconclusive.

14. Fuoco et al. (2014) reported that PNC increased with EC nicotine content and with puff duration (2, 3, 4 s). Size-distribution measurements of diluted (1:4400[4]) aerosol were made in real time by FMPS (range 5.6–560 nm), and PNC values were measured by Condensation Particle Counter (CPC). Average 2-s-puff peak PNCs calculated for undiluted aerosol were around 4.4 x 10^9 particles/cm^3 (averaged across several EC/e-liquids) and 3.1 x 10^9 particles/cm^3 (for CC). Particle size distribution modes were observed in the range 120-165 nm for EC and at 165 nm for CC. A second mode at around 10 nm for EC was rejected by the authors as a measurement artefact. Particle size distribution was not affected significantly by EC type (tank system; atomizer phantom; cartomizer), nicotine content, or e-liquid (four flavours), but PNC was higher with 12–18 mg nicotine/mL compared with zero nicotine content.

15. Blair et al. (2015) reported similar PNCs measured in the mainstream product of an EC (4.0 x 10^9 particles/cm^3) and two reference CCs (4.8 x 10^9 and 5.7 x 10^9 particles/cm^3). A small peak particle diameter of 30 nm reported for the EC was attributed by the authors to the approximately 1:1000 dilution of the sample necessary for the real-time SMPS measurements. Particle diameter increased with increasing number of puffs to approximately double the single-puff value at steady state. MOUDI cascade impactor measurements at different dilution ratios showed that increased dilution (from 13:1 to 190:1) resulted in a reduction in mass peak diameter from ~350 nm to ~150 nm, again attributed to greater evaporation from particles at higher dilutions.

16. Lerner et al. (2015) reported a particle mass median aerodynamic diameter (MMAD[5]) of 1.03 µm (range 0.45-2.02 µm; geometric standard deviation (GSD[6])

[3] Calculated diameter in a population of particles for which 50% of particles have larger diameter and 50% have smaller diameter

[4] Not stated by Fuoco, but reported in a table in Baassari et al. (2017)

[5] The diameter at which 50% of the particles by mass have a larger diameter and 50% smaller

[6] Standard deviation of the logarithms, which describes the spread of particle diameters in a distribution; in general values ≤ 1.25 indicate a monodisperse population and ≥ 1.25 polydisperse

1.71) in aerosols produced from a ‘Blu’ EC, calculated from particle deposition weight measurements made by a cascade particle impactor. The lower measurement detection limit of the technique used in this study was 0.45 µm.

17. Ji et al. (2016) measured particles in the range 7–289 nm by SMPS in aerosols from menthol- or tobacco-flavoured e-liquids with or without nicotine (24 mg/mL). Peak particle diameters were in the range 25-35 nm. The authors reported that menthol flavour produced overall fewer nanoparticles than tobacco flavour, and the co-presence of nicotine appeared to affect aerosol PNC differently for the menthol- and tobacco-flavoured products. The aerosol dilution level was not stated, however puffs were measured in a large (320 L) chamber, and a paper cited by the same authors with apparently the same setup also cited in this report suggests high dilution ratios (around 10,000:1 to 4000:1).

18. Mikheev et al. (2016) commented that the dilution methods employed in previous studies would be likely to lead to particle transformation during sampling and analysis, leading to altered aerosol size parameters. These authors emphasised the requirement for low dilution, quick sample delivery, and real-time measurements to analyse EC aerosol size distribution characteristics, due to the highly dynamic nature of the mixture which can result in rapid particle growth or evaporation, depending on the conditions. They used an electrical mobility particle sizer (Differential Mobility Spectrometer DMS500 and Smoke Cycle Simulator, Cambustion) to evaluate aerosol particles generated from 5 s puffs of several ECs (NJOY King, V2, and blu disposable ‘cigalikes’, and the tank-style ‘Joyetech’ EC; various flavours; with or without nicotine) at low (1:30) and high (1:3000) dilutions. Bimodal aerosol size distributions were observed, with a high concentration of nanoparticles (11-25 nm CMD, GSD 1.9) as well as submicron particles (96-175 nm CMD, GSD 1.76), with PNCs of 10^7-10^8 particles/cm^3 for each type. This bimodal size distribution was observed at both high (1:3000) and low (1:30) aerosol dilutions. The highest concentration of nanoparticles occurred at the beginning of the puff, and the authors hypothesised that they may be derived from the metal wire, the compounds that collect around the wire (so called ‘coil gunk’), and/or primary, less-volatile compounds in the e-liquid (glycerine, nicotine, possibly flavourings). Calculations based on total particulate mass measurements indicated that metals made up an estimated 10% of the nanoparticle mass. Tests of ‘dry puffing’ (puffing from a tank containing no e-liquid) produced only nanoparticles.

19. Pratte, Cosandey and Goujon-Ginglinger (2016) used laser aerosol spectrometry (measurement range 0.09–7.5 µm) for the optical measurement of EC aerosol particles. MMAD values ranged from 225–293 nm, with CMDs in the range 158–191 nm (GSDs, 1.41–1.46) for four commercially available EC containing various proportions of PG, VG, water, nicotine, and flavour, and an aerosol residence time of 3.4 s. The ranges were slightly lower (17-33% for MMAD) using an alternative aerosol capture system with lower residence time. The authors pointed

out that these study results could not be compared directly with previously published study findings due to the use of different measurement techniques (e.g. aerodynamic, mobility, optical), dilutions, and sampling times.

20. Sosnowski and Kramek-Romanowska (2016) analysed undiluted aerosol samples generated from a PG/VG e-liquid (with 18 mg/mL nicotine and up to 15% flavours) using eGO clearomizer EC devices. Measurement by laser diffraction spectrometry (measurement range, 0.1-100 µm) indicated aerosol particle size distributions were in the ranges of 180-220 nm CMD and 405-420 nm volumetric mean diameter (VMD[7]) with a GSD of 1.5-1.7. The authors noted that these findings were similar to those of Fuoco et al. (2014), and in a similar range to tobacco smoke. Deposition in the respiratory system was modelled (see section 3).

21. Wright et al. (2016) performed studies to measure the variation in growth and evaporation kinetics and hygroscopic growth of glycerol or PG aerosol with temperature, relative humidity, and residence time. In this model, peak particle numbers for glycerol were observed at around 180 nm and 300 nm. Modelled time required to evaporate a 350 nm glycerol or PG particle to half its mass was much longer at a high water saturation ratio than in dry conditions, which was longer for glycerol than for PG, and shorter at higher temperature (37 °C vs 'room temperature'). The authors commented on the importance of understanding glycerol and/or PG and H_2O vapour fields at high spatial and temporal resolution to model plume dynamics and compositional changes of EC aerosol from the device condenser through the bronchial tract and to exhaled plume.

22. Zhao et al. (2016) conducted studies to assess how the characteristics of aerosol produced from EC may vary with the temperature of the heating coil and with puff topography. The temperatures of the coils from four different brands of EC, measured repeatedly using different cartridges, for a 2-s puff, were in the range of 140–230 °C. The authors commented that standard coil temperatures are often reported as being lower than this (e.g. 60–70 °C): this may be due to the fact that the temperature falls sharply with distance from the coil (i.e. 60–70 °C measured at 1 mm from the coil in these experiments), presumably due to a cooling effect of the aerosol droplets produced. PNC values varied within and between brands, in the range of around 0.6–1.6 x 10^9 particles/cm^3, and CMD values were in the range of 18–29 nm (measured in samples at high dilution). Increasing puff length (2, 3, 4, 5 s) was correlated with increases in coil temperature, PNC and CMD. Conversely, increasing the flow-rate led to decrease in coil temperature and CMD, although it was positively correlated with PNC.

23. Baassiri et al. (2017) reported that the e-liquid VG/PG ratio has a substantial effect on aerosol total particulate mass, particle size distribution and nicotine content.

[7] The diameter of a particle of mean volume of the sample

A second-generation tank-machine system was used with a digital vaping machine. Particle mass distribution was measured with a six-stage cascade impactor, and number size distribution by FMPS at high (1450:1) dilution. Lower VG proportions produced lower levels of total particulate matter and lower nicotine yield. Mass median diameter measured from the cascade impactor ranged from 2.28–3.57 µm. Analysis of FMPS particle size distribution showed bimodal particle number distributions with modes around 10, 30, and/or 160 nm depending on the VG/PG ratio used. CMDs were in the range 44–97 nm, with PG associated with smaller diameters. The authors concluded that the e-liquid composition (VG/PG) has a large impact on nicotine and total particulate matter emissions and on aerosol particle size.

24. Belka et al. (2017) reported a unimodal distribution of fine/ultrafine particles, which appears to be roughly in the range 50–600 nm, emitted from a Czech Joyetech EC (0 or 16 mg/mL nicotine). The CMD, measured by SMPS at approximately 1:4000 dilution and with a 90 s delay, was approximately 150 nm (a CMD of 200 nm was measured for CC mainstream smoke). The authors noted that they did not observe the bimodal distribution reported in some other studies, however a slight deviation around 70 nm was noted for the zero-nicotine-content EC. PNC values for mainstream aerosol, measured by CPC, were in the range of 5×10^9 particles/cm^3, which was approximately double the concentration observed in mainstream CC smoke.

25. Kim et al. (2017) developed a reference e-liquid for analytical studies. Characterisation of the aerosol, produced using a custom-built testing device, revealed a mean PNC of 1903 ± 492 particles/cm^3 and mean particle diameter of 1.40 ± 0.06 µm, of which the authors state 38% had a diameter less than 0.5 µm. However, this study used an Aerodynamic Particle Sizer (APS), an instrument which is not specifically designed to investigate particles below 0.5 µm and hence the number of particles reported in this size fraction is subject to significant uncertainty. The particle concentrations here are thus not directly comparable with other studies using aerosol mobility-based techniques.

26. Lee et al. (2017), in a chamber study, reported that diluted aerosol produced from two different flavours of a ‘cigalike’, cartomizer-type EC aerosol contained approximately double the particle number and mass concentrations of fine (< 2.5 µm) and nano (< 100 nm) particles in the tobacco flavour compared with the menthol flavour (at 1 puff/min, PNC was approx. 8000 and 16 000 particles/cm^3; nanoparticles approx. 5500 and 12 000 particles/cm^3, for menthol and tobacco flavours, respectively).

27. Prévôt et al. (2017) reported similar MMAD values to Lerner et al. (2015), i.e. 0.75–0.8 µm for aerosol particles produced from a third-generation tank-system

atomiser EC with homemade e-liquid (VG/PG mix + 18 mg/mL nicotine[8]), measured by cascade impaction using a high flow rate and low dilution (1:5). Values did not vary with VG/PG ratio (20/80 or 80/20). Nicotine concentration measured in the aerosol was around 20–40% less than in the e-liquid, but was distributed equally over the aerosol particle size distribution (12 fractions).

28. Sundahl, Berg and Svensson (2017) used a particle impactor (with filter capture) designed for the evaluation of inhaled pharmaceutical products (Next Generation Impactor) to investigate particle size distributions of a range of 13 commercially available EC. All tests were carried out at relative humidity of 100%, to represent the human respiratory system, although the experiments were mostly carried out at low temperature (4–8 °C). MMADs were in the range of approx. 0.5–0.9 µm. Nicotine was measured in droplets and gas phase, and was found to be distributed mostly to the droplet phase when experiments were run at 4-8 °C, but with a shift towards the vapour phase at a higher temperature (20–25 °C). Theoretical modelling of lung deposition based on these data using the multipath particle dosimetry (MPPD) model (see below, 'Modelling aerosol dynamics in the airways' section) suggested that 10–25 % of droplets would be deposited in the respiratory system and 75–90 % would be exhaled. Pronounced deposition was predicted in the alveolar region (generations 16 and below), with little mouth deposition. However, the authors noted that this model may overestimate the exhaled nicotine fraction as nicotine is semi-volatile and dilution during inhalation would drive the equilibrium towards gaseous nicotine. In addition, the authors suggested that the large surface area of the lower respiratory system might capture substantial levels of gaseous nicotine, particularly in individuals who practise 'breath-holding' to maximise nicotine uptake. The exhaled fraction could theoretically also be reduced by droplet hygroscopic growth during transport through the airway, although this was not considered in the model.

29. Pankow (2017) discussed the importance of considering gas/particle phase partitioning of components of EC aerosols, which would affect their deposition in the respiratory tract. Theoretical gas/particle partitioning coefficients in 1:1 PG/glycerol were calculated for several compounds, including formaldehyde and derivatives (the hydrate, methandiol; the hemiacetal with PG), acetaldehyde, acrolein, benzene, diacetyl, limonene, benzaldehyde, and nicotine. Formaldehyde would be expected to exist almost exclusively in the gas phase, even at high levels of total particulate matter, while its two adducts formed by 'vaping' would be mostly in aerosol droplets, even at low total particulate matter levels.

[8] The publication by Prévôt et al. (2017) reported a value of 18 mg/L nicotine, but this is assumed to be a typographical error, as nicotine concentrations used are generally in the range of 18 mg/mL

Commentary

30. The nature of the aerosol emitted in regular EC use, i.e. the high and variable particle number concentration and volatility of the particulate matter, makes it difficult to perform standardised, comparable studies. In particular, a number of investigators have noted that measurement procedures requiring dilution of the aerosol are likely to result in substantial evaporation, and thus alter the particle size distribution compared with the particulate matter to which an EC user would actually be exposed. Comparison of results within groups of studies performed with similar instrumentation and methodologies may be possible if sufficient information is provided as to the specific operational parameters employed. However, caution is advised in comparing results, especially particle size data, obtained from different classes of instruments, including (low-flow) SMPS, (high-flow) 'fast' mobility particle sizers (FMPS, DMS), impaction and direct sampling techniques. Information about the nature of the particles produced by EC, e.g. composition, volatility, may be inferred by examining the differences in particle size, number and other properties from different types of study, where sufficient detail is provided on the experimental method.

31. Impaction or filtration with subsequent gravimetric mass measurement may suffer to some extent from evaporation of volatile material during or after sampling, depending on the pressure at collection (some instruments, e.g. MOUDI, ELPI, operate at low pressure which encourages evaporation of material, especially at smaller particle size) and/or treatment, although assessment of collection efficiencies within studies can clarify the extent of, or rule out, this problem for the specific methodology applied.

32. SMPS measurements require a relatively long sample collection time, often more than one minute, which is not ideal in the measurement of transient EC emissions without either 'smoothing' of concentrations by using a holding chamber, which itself often introduces dilution and potentially also particle size change, or at the least, concurrent measurement of particle concentrations via another method. In cases where there is reason to suspect changing particle concentrations during the SMPS scan period, the operational conditions should be reported. If not, interpretation of the true size distribution may be very uncertain. In addition, in aging studies conducted with SMPS, e.g. Zhang, Sumner and Chen (2013) the true aging time in reality is spread over a range beginning and ending with the start and end of the relevant SMPS scan(s) and may be different to the times reported directly in studies if they report only the measurement start time.

33. Studies have measured either undiluted, low-dilution, or high-dilution aerosol. Particle number concentrations in the range of 10^9 particles/cm^3 have been reported from several studies, although results vary and it is not always clear whether the measurements described relate to diluted sample or the undiluted mainstream aerosol. Measurements, including those that do not explicitly report dilution, may

introduce some dilution of the EC aerosol by virtue of the flow rates necessary to operate the experimental instrumentation used, or by introduction into a chamber for subsequent measurements. Care must be taken in interpreting experimental results, especially where dilution ratios and/or flow rates through both EC and instrumentation are not provided.

34. A unimodal particle size distribution in the submicron range, similar to that observed for CC smoke, has been reported. However, a number of studies have revealed a bimodal size range distribution, with the presence of both submicron and nano particles. Interpretation of the results in relation to the 'real' situation in the mainstream EC aerosol that is inhaled by an EC user is an area that is still uncertain.

35. In addition to possible effects of sample dilution, other factors such as higher temperature, lower relative humidity, and sample aging have been associated with a shift to smaller particle size, suggesting an effect of evaporation. The major constituents of e-liquid, PG and VG, are described to partition mostly to the particulate phase of the aerosol, and findings suggest that nicotine also partitions mostly to the droplet phase. One study estimated theoretical gas/particle phase partitioning coefficients for components of EC aerosols under various conditions, including potential adducts formed during vaping.

36. The effects of variation in e-liquid constituents on the particulate phase, for example PG:VG ratio, nicotine and/or flavouring content, have been investigated in some studies, with variable findings reported.

37. Post puffing, the EC aerosol would be expected to be dynamic, but measurements have mostly not addressed the subsequent dynamics of the input aerosol during puffing and inhalation. However, based on the particle size distribution data that have been obtained, some investigators have modelled the distribution of EC particulates to the human airways.

Modelling aerosol dynamics in the airways

38. Manigrasso and colleagues performed modelling studies of EC aerosol deposition in the human respiratory tract, based on aerosol particle size distribution parameters reported by the same group (Fuoco et al. (2014), described in the 'Evaluation of the particulate fraction of mainstream aerosol' section, above. Using an MPPD model, which calculates deposition and clearance of mono- and polydisperse aerosols, they calculated that 6.25×10^{10} particles would be deposited in the respiratory tree from one 50 cm^3, 2 s puff, with maximum deposition in the alveolar region. Models suggested maximum deposition at a slightly higher particle diameter with nicotine (124 nm) than without (93 nm) (Manigrasso et al. 2015).

39. Sosnowski and Kramek-Romanowska (2016) measured EC aerosol particle size distribution parameters from refillable clearomizer ECs containing a

PG/VG/nicotine/flavour e-liquid (described in the 'Evaluation of the particulate fraction of mainstream aerosol' section, above) and used these measurements, along with measurements of intrinsic aerodynamic resistance of the EC devices, as the basis of modelling studies to predict deposition of EC aerosol in the human airway. Two computational methods were used: MPPD and Finlay-Martin correlations. Based on an MMAD of 410 nm, predicted total deposition of mainstream aerosol was relatively low, generally in the range 10–30%, and varied depending on the breathing scheme tested. This would imply that a substantial proportion of the product is exhaled. Aerodynamic resistance of the EC devices was found to be high compared with both CC and medical dry powder inhalers, implying that substantial effort and the inhalation of large amounts of EC product are required to achieve an effective dose of nicotine to the lungs from EC. The authors pointed out that their applied models did not take into account aerosol particle-specific factors such as hygroscopic growth and coagulation.

40. Pichelstorfer et al. (2016) concluded that modelling without consideration of EC aerosol particle dynamics would be expected to lead to an underestimation of droplet deposition in the lung, and a vast underestimation of nicotine deposition, due to failure to take into account the vapour phase. The authors used their Aerosol Dynamics in Contaminants model, which takes into account aerosol particle dynamics (coagulation, condensation of water vapour, evaporation of semi-volatile components, chemical reactions, and deposition), to estimate particulate and vapour-phase deposition of CC and EC products in the human lung. EC aerosol median particle diameter (163 nm) and usage parameters (puffing, mouth-hold, inhalation, expiration) were taken as equivalent to those for CC. These studies indicated a shift towards larger particle size during the breathing cycle, mostly due to particle coagulation in the mouth. Lung deposition of particles was predicted to be higher for EC than for CC due to the higher hygroscopic growth rates of EC particles. Around 99% of nicotine from EC would be deposited in the vapour phase, while for glycerol, approximately 40% would be deposited in the condensed phase and 2% in the vapour form. Maximum particle deposition was estimated to occur at airway generation 11 (EC and CC), with vapour-phase maxima at generation 12 for CC and 15 for EC.

41. Feng and colleagues reported detailed theoretical modelling studies of EC aerosol droplet distribution for e-liquids with various theoretical PG/glycerol/nicotine concentrations in human respiratory segment models. EC droplets were found to exhibit greater hygroscopic growth than CC smoke particles, and droplets would undergo size changes due to numerous factors (Feng, Kleinstreuer and Rostami 2015). These studies were extended with detailed modelling in a subsequent report (Feng et al. 2016).

42. A report by Nordlund et al. (2017) described experimental modelling of EC aerosol in a segmented human respiratory tract cast model. GC-MS was used to

quantify regional deposition in a 32-segment model of components of aerosol particles from two liquids: 1) monodisperse glycerol (CMD 2.1 µm, MMAD 2.3 µm) (1.5–2 h exposure, flow rate 1.5 L/min), 2) a nebulised multicomponent liquid (52.6% PG, 28.1% glycerine, 1.9% nicotine, 17.4% water; CMD 0.307 µm, MMAD 0.497 µm) (1 h exposure, 15 L/min). Deposition patterns and fractions were similar for all components of the multicomponent aerosol as for the glycerol aerosol, indicating that the compounds are likely to be deposited as part of the same droplets. The highest deposition fraction was seen in the upper airways.

Summary and conclusions

43. Studies of EC aerosol particulate matter suggest that it comprises submicronic particles with a similar size distribution to CC, and also nanoparticles. Particle number concentrations in undiluted mainstream aerosol are generally reported in the range of 10^9 particles/cm^3. The relative proportions of submicron and nanoparticles are difficult to estimate due to experimental limitations, including substantial evaporation of larger particles at high dilution ratios, and limited capability of spectral transmission methods to detect nanoparticles. Solid particles (e.g. metal nanoparticles) may also be present, and this area will be reviewed in a future paper.

44. Studies are difficult to compare, due to their variability in test conditions (including aerosol dilution, temperature, puff parameters, real-time or delayed measurements, instrumentation and measurement ranges), numbers, and types of ECs and e-liquids (brand, flavour, presence or absence of nicotine) tested. Standardised and validated testing devices and protocols are needed.

Questions for the Committee

45. Members are asked to consider the paper and in particular:

i. Whether there are any particular aspects which should be captured when a COT statement on e-cigarettes is prepared?

NCET at WRc/IEH-C under contract supporting the PHE COT Secretariat
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Abbreviations and Glossary

CC:	Conventional cigarette
CMD:	Count median diameter (calculated diameter in a population of particles for which 50% of particles have larger diameter and 50% have smaller diameter)
CPC:	Condensation Particle Counter
EC:	Electronic cigarette; e-cigarette
FMPS:	Fast Mobility Particle Sizer
GSD:	Geometric standard deviation (standard deviation of the logarithms, which describes the spread of particle diameters in a distribution; in general values ≤ 1.25 indicate a monodisperse population and ≥ 1.25 polydisperse)
MMAD:	Mass median aerodynamic diameter (the diameter at which 50% of the particles by mass have a larger diameter and 50% smaller)
MPPD:	Multipath particle dosimetry
PAH:	Polycyclic aromatic hydrocarbon
PG:	Propylene glycol
PNC:	Particle number concentration
SMPS:	Scanning Mobility Particle Sizer
TSNA:	Tobacco-specific nitrosamine
VG:	Vegetable glycerine (glycerol)
VMD:	Volume mean diameter (the diameter of a particle of mean volume of the sample)
VOC:	Volatile organic compound

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COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT (COT)

Potential toxicological risks from e-cigarettes. Paper 1: Characterisation of the aerosol droplet particle fraction

COT discussion paper TOX/2016/25: Scoping paper on the potential risks from electronic nicotine (or non-nicotine) device systems in users and non-users (bystanders): a focused overview.

This paper is available here: https://cot.food.gov.uk/sites/default/files/tox2016-25.pdf

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COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT (COT)

Potential toxicological risks from e-cigarettes. Paper 1: Characterisation of the aerosol droplet particle fraction

Illustrations of examples of first, second and third generation e-cigarette devices

Images are attached, they are not being made publicly available for copyright reasons

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COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT (COT)

Potential toxicological risks from e-cigarettes. Paper 1: Characterisation of the aerosol droplet particle fraction

Table 1 - Summary of studies of the physical properties of EC mainstream aerosol particulate fraction

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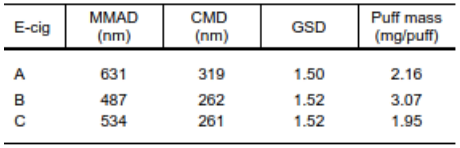
Table 1. Summary of studies of the physical properties of EC mainstream aerosol particulate fraction

Reference	Investigation	Test materials	Experimental methods	Results / aerosol particle characteristics
Ingebrethsen, Cole and Alderman (2012)	Compared two different real-time methods to measure EC aerosol particle size distribution: spectral transmission (undiluted aerosol); electrical mobility analysis (two-step dilution)	2 x cartomizer EC (1 rechargeable, 1 disposable, brands not stated) 3R4F reference CC	2, 3, 4 s puffs, 30 s interval, 55 cm^3 total volume (EC) 2 s puff, 30 (55cm^3) or 60 s (35 cm^3) interval (CC) Real-time measurements Spectral transmission measured with linear diode array spectrometer (Ocean Optics) at approx. 550, 650, 750, and 850 nm (no dilution) Electrical mobility measured with Cambustion DMS500 differential mobility spectrometer coupled to Cambustion Smoking Cycle Simulator (range 5-1000 nm) (two-step dilution, 1:3400-1:5500) Gravimetric measurements using Cambridge filter pad	<u>Average PNC</u> In the range approx. 10^9 particles/cm^3 for EC and CC, by both measurement methods. <u>Particle diameter of average mass</u> EC: in the range of around 250–450 nm by spectral transmission; in the range of around 20–50 nm by electrical mobility CC: in the range of around 200–400 nm by both methods (lower by a factor of ≤ 2 by electrical mobility) Similar results obtained for the two different cartomizers <u>TPM (per 2 s puff)</u> EC: in the range approx. 0.95–2.4 mg by spectral transmission in the range approx. 0.002–0.01 mg by electrical mobility in the range approx. 1.4–2.5 mg by gravimetric analysis
Schripp et al. (2013)	Measured EC aerosol size	3 EC delivery systems (2 tank, 1 cotton), apple-flavoured e-liquid without nicotine	Particle size distribution measurements by fast FMPS spectrometry (TSI) (detection range 5.6-560 nm) (flow rate 8 L/min) 3 s puff into a 10 L glass chamber; Near-real-time measurements (1, 3, 5, 7, 10 min at 23,37, or 50 °C);	Bimodal size distribution (maxima at 60 nm and 100 nm) at 23 °C after 5 min shifted to unimodal distribution (maximum at 45 nm) at 50 °C after 5 min Bimodal size distribution (maxima at 60 nm and 100 nm) at 37 °C after 1 min shifted to unimodal distribution (maximum at 45 nm) at 37 °C after 3 min

Reference	Investigation	Test materials	Experimental methods	Results / aerosol particle characteristics
Zhang, Sumner and Chen (2013)	Measured EC aerosol size	Bloog MaxX Fusion EC with cartridges filled with 16 mg/mL nicotine in PG or VG Kentucky reference CC	SMPS 3936 (TSI) (measurement range, 10–1000 nm, scan time not reported) Single puff (30 s, cycle, actual puff duration not stated), 50 mL chamber, aging for 0, 10, or 40 s, 22 °C (no dilution reported); steady-state measurements	Peak particle diameter (nm) at 0 s, (10 s, 40 s) (single puff) PG: 117 (121, 131) VG: 180 (181, 181) CC: 215 Values increased with aging time Steady-state aerosol, bimodal particle distribution with peak diameters compared with a single puff and at around 400 nm
Alderman et al. (2014)	Measurement of EC aerosol particle size distribution (to verify the results of Ingebrethsen et al., 2012)	Three EC (two rechargeable cartomizer-type (A,B); one disposable (C))	Cascade impactor (MiniMOULDI MSP), measurement range 0.056-10 µm, flow rate, 2 L/min, no dilution 3 s puff, 50 mL puff volume	
Fuoco et al. (2014)	Measured EC mainstream aerosol particle number and size distribution	Tank system (rechargeable); Atomiser phantom (rechargeable); Cartom (disposable) CC (Marlboro; 0.8 mg/cigarette nicotine) 4 x e-liquid flavours +/- nicotine (zero 0 mg/mL; medium 8-9 mg/mL; high 12-18 mg/mL)	PNC (≥ 4 nm diameter) measured with CPC 3775, TSI Inc) (flow rate 1.5 L/min) Particle size distribution (range 5.6-560 nm diameter) measured with FMPS 3091 (TSI Inc) (flow rate 10 L/min); and TSI 3080 classifier + 3775 CPC, range 5.83-583 nm diameter, 14 channels, dilution 16:1) Volatility analysis (FMPS 3091) (37, 100, 150 °C) 2 s puff (for other variables tested); real-time measurement; two-step dilution 1:4400 (37 °C) Puff length (2, 3, 4 s)	PNC (x 10^9 particles/cm^3) EC (average): 4.39 ± 0.42 CC: 3.14 ± 0.61 EC (zero nicotine): 3.26-4.09 EC (high nicotine): 5.08-5.29 Increased with increased puff length (3 s, 4 s), not affected by EC type or e-liquid flavour Particle size distribution Unimodal, in the range of 120-165 nm, similar to those from CC No effect of nicotine concentration Volatility analysis No effect of temperature on amount of evaporation over size distribution.

E-cig	MMAD (nm)	CMD (nm)	GSD	Puff mass (mg/puff)
A	631	319	1.50	2.16
B	487	262	1.52	3.07
C	534	261	1.52	1.95

Reference	Investigation	Test materials	Experimental methods	Results / aerosol particle characteristics
Blair et al. (2015)	Compared standard technology with a 'fast-flow tube system' to study VOCs and particulates	EC (brand not specified, 18 mg nicotine/cartridge in PG, 3.6 V) Reference CCs (IR5F, 3R4F)	CPC SMPS Real-time analysis, 2 s puff, high dilution (1000:1) Cascade impactor (MOUDI, dilution ratio 13:1 or 190:1)	PNC (particles/cm^3) EC: 4.0 x 10^9 IR5F: 4.8 x 10^9 3R4F: 5.7 x 10^9 Peak number particle diameter EC: 30 nm (SMPS) Peak mass particle diameter EC (dilution 13:1): 350 nm EC (dilution 190:1): 150 nm
Lerner et al. (2015)	Studied oxidants in EC (measured aerosol size distribution and levels of copper)	Blu (Lorillard Technologies, Inc.)	Cascade particle impactor (range, 0.450–2.02 µm), flow rate 5 L/min 4 s puff	MMAD: 1.03 µm, GSD 1.71
Ji et al. (2016)	Characterised EC aerosol	Menthol or tobacco flavour EC 0 or 24 mg/mL nicotine	CPC 3785 (TSI) to measure PNC SMPS 3080 (TSI) to measure size distribution (measurement range, 7–289 nm, flow rate 0.6 L/min) 2 to 5 s puff, 320 L chamber, 24 °C, relative humidity 30%, dilution not reported	Peak particle diameters measured in the range 25–35 nm

Reference	Investigation	Test materials	Experimental methods	Results / aerosol particle characteristics
Mikheev et al. (2016)	Measured: aerosol particle size distribution (from e-liquid or 'dry puffing'[9]) by real-time, low-dilution methodology	3 brands of fixed-power, non-refillable 'cigalikes' (NJOY King, V2, blu) 1 adjustable-power, refillable 'tank-style' (Joyetech) Reference CC (3R4F)	Machine vaping (Smoke Cycle Simulator, Cambustion Ltd) Particle size distribution measured with Differential Mobility Spectrometer, DMS500, (Cambustion Ltd) <u>e-liquid</u> 5 s puffs at 1 min intervals, 3 flow rates (15, 20, 25 mL/s) High (1/3000) and/or low (1/30) sample dilution <u>Dry puff (tank-style 'Joyetech', empty tank)</u> 5 s puffs at 1 min intervals, flow rate 15 mL/s Real-time measurements (temperature not stated)	<u>e-liquid</u> Puffing e-liquid at low dilution produced aerosol with a bimodal particle distribution (10^7-10^8 particles/cm^3 each type) - nanoparticles (11-25 nm CMD, geometric SD within 1.9) - submicron particles (96-175 nm CMD, geometric SD within 1.76) Higher dilution led to a higher proportion of nanoparticles. CC produced mainly submicron particles <u>Dry puff</u> Dry puff produced only nanoparticles (10^7 particles/cm^3; 7.5 nm CMD).
Pratte, Cosandey and Goujon-Ginglinger (2016)	Evaluated Laser Aerosol Spectrometer technology for determination of EC aerosol droplet size distribution and diameter range	e-cigarette A (75% glycerine, 25% water) e-cigarette B (1.14% nicotine, 50.96% glycerine, 27.86% PG, 15.91% water, 4.13% others) e-cigarette C (1.18%nicotine, 2.29% menthol, 50.96% glycerine, 26.36% PG, 15.94% water, 3.27% others) e-cigarette D (20.20% glycerine, 74.13% PG, 5.96% water)	Laser aerosol spectrometer 3340 (TSI) (measurement range, 0.09–7.5 µm) Health Canada puffing regime (55 mL puff volume, 2 s puff), dilution 1:10,000, 3.4 s sampling delay	MMADs in the range, 225–293 nm CMDs in the range 158–191 nm (GSDs, 1.41–1.46)

[9] No e-liquid

Reference	Investigation	Test materials	Experimental methods	Results / aerosol particle characteristics
Sosnowski and Kramek-Romanowska (2016)	Measured aerosol particle size distribution and modelled lung deposition	2 x clearomizer-type refillable devices (eGO-CE5, eGO-W) e-liquid (50–55% PG, 30–35% VG, 18 mg/mL nicotine, up to 15% flavours)	Aerosol particle size distribution measured by laser diffraction spectrometer (Spraytec, Malvern Instruments). Measurements performed on-line on undiluted aerosol sample (range of analysis, 0.1– 100 µm), flow rate 5 L/min, 500 ms sampling with 1 s interval	<u>Particle size distribution</u> CMD, 180-220 nm, Volumetric median diameter (VMD) 405-420 nm, GSD, 1.5-1.7
Wright et al. (2016)	Measured the growth and evaporation kinetics and hygroscopic growth of glycerol aerosol	Glycerol PG	TDMA CPC 3773 (TSI) (measurement range > 10 nm) Aerosol electrometer 3036 (TSI) Variation in temperature, relative humidity, and residence time	Peak particle numbers at approximately 180 nm and 300 nm. Modelled time required to evaporate a 350 nm glycerol particle to half its mass at high H_2O saturation was much higher than in dry conditions (approximately 200 s vs 3 s at room temperature[10]), and decreased with temperature (33 s and 0.5 s at 37 °C). Modelled values for a 350 nm PG particle were 800 s and approx. 1.5 s (room temp.); 45 s and 0.5 ms (37 °C)
Zhao et al. (2016)	Assessed how EC aerosol characteristics vary with heating coil temperature and puff topography	4 rechargeable EC brands (not stated), tobacco flavour, no nicotine; several cartridges tested from each brand	Combinations of 2, 3, 4, 5 s puff, 0.5-1, 1.5, 2 L/min flow rate; 30 s puff interval Heating coil temperature measured by thermocouple thermometer Homemade puffing machine; Aerosol diluted in 320 L chamber, approx. 30% relative humidity and 24 °C; PNC and particle size distribution measured by SMPS 3080 (TSI)	<u>At 3 s puff, 1 L/min</u>, Peak heating coil temperatures measured using different brands/cartridges ranged from around 140–230 °C; PNC ranged from 0.58–1.64 x 10^9 particles/cm^3, high variation between different cartridges within the same brand; CMD varied from 18 nm to 29 nm <u>Effects of puff topography</u> Increasing puff duration led to: increased coil temperature; higher PNC; higher CMD Increasing flow rate led to: decreased coil temperature; increased PNC; decreased CMD

[10] value not stated

<table>
<tr><th>Reference</th><th>Investigation</th><th>Test materials</th><th>Experimental methods</th><th>Results / aerosol particle characteristics</th></tr>
<tr><td>Baassiri et al. (2017)</td><td>Investigated the effect of PG/VG ratio on aerosol total particulate matter, particle size distribution and nicotine content</td><td>Vapor-Fi second-generation tank system

Liquid batches of analytical-grade PG/VG at ratios from 0/100 to 100/0 + 18 mg/mL nicotine</td><td>Particle mass measurements by cascade impactor (CI) (dilution 1.75:1)

Particle number size distribution measurements by mobility particle sizer (TSI EEPS 3090), measurement range 5.6–560 nm (dilution 1450:1)

Nicotine measurements by GC-MS

4 s puff, flow-rate 29.2 mL/s</td><td>
<table>
<tr><th>PG/VG ratio</th><th>100/0</th><th>70/30</th><th>0/100</th></tr>
<tr><td>Mass concentration (µg/cm^3)</td><td>73.3±5</td><td>52.1±4.2</td><td>38.6±1.4</td></tr>
<tr><td>Mass median diameter (nm)</td><td>2279±92</td><td>3105±480</td><td>3573±380</td></tr>
<tr><td>PNC (part/cm^3, x10^9)</td><td>7.80±1.04</td><td>1.41±0.13</td><td>1.50±0.24</td></tr>
<tr><td>CMD (nm)</td><td>44±2</td><td>81±4</td><td>97±10</td></tr>
</table>
</td></tr>
<tr><td>(Belka et al.) 2017</td><td>Measured EC aerosol particle size distribution</td><td>Joyetech refillable, variable wattage (up to 9.6 W) EC; e-liquid containing 0 or 16 mg/mL nicotine

Marlboro light CC (0.6/cig nicotine)</td><td>SMPS 3936 (TSI) (flow rate 0.3 L/min)

4 s puff, total volume 60 cm^3, 90 s delay, 2-step dilution (1:4000)</td><td>
<table>
<tr><th></th><th>Averaged total number concentration (particles/cm^3, x10^9)</th><th>Averaged CMD (nm)</th></tr>
<tr><td>EC, 0 mg/mL nicotine</td><td>4.81±0.1</td><td>147.1±4.5</td></tr>
<tr><td>EC, 16 mg/mL nicotine</td><td>4.63±0.05</td><td>157.6±3.8</td></tr>
<tr><td>CC</td><td>2.41±0.15</td><td>201.6±0.3</td></tr>
</table>
</td></tr>
<tr><td>Kim et al. (2017)</td><td>Developed and analysed a reference e-liquid for analytical studies; characterisation of the aerosol for physical properties</td><td>Custom-built testing device

e-liquid containing PG/glycerine (1:1, 8:2, 2:8, v/v); 10 mg/mL nicotine</td><td>Physical analysis by Aerodynamic Particle Sizer (APS) 3321 (TSI) (size range 0.5-2.0 µm, 4 L chamber, flow rate 1.0 L/min, dilution not reported)

4 s puff, 50 ml puff volume, 21 °C</td><td>Mean particle size (diameter): 1.40 ± 0.06 µm

Mean particle concentration: 1903 ± 492 particles/cm^3</td></tr>
</table>

Reference	Investigation	Test materials	Experimental methods	Results / aerosol particle characteristics
Lee et al. (2017)	Analysed EC aerosol content: effects of flavour and puffing time	EC V2 'cigalike' cartomizer devices (VMR Products): tobacco flavour, menthol flavour; 1.8% nicotine	Automated smoking machine (Modified TE-2 system) Dilution 1:172, approx. 34 °C and 19% humidity 1, 2 puffs/min $PM_{2.5}$ (< 2.5 µm diameter) mass concentration measured with SidePak light-scattering integrating nephelometer AM510 (TSI) and Personal Exposure Monitor PNC (0.02–1 µm diameter) measured with P-Trak Ultrafine Particle Counter 8582 (TSI) (flow rate 0.1 L/min) Nanoparticle (< 100 nm diameter) mass and number concentrations measured with SMPS 3936 (TSI) (flow rate 0.3 L/min, measurement range, 10–1000 nm).	PNC (approximate mean values; particles/cm^3, 1-puff test) Tobacco flavour: 16,000 Menthol flavour: 8,000 Nanoparticles - number (approximate mean values; particles/cm^3, 1-puff test) Tobacco flavour: 12,000 Menthol flavour: 5,500 Nanoparticles- mass (approximate mean values, ng/m^3) Tobacco flavour: 1,500 Menthol flavour: 500
Pankow (2017)	Modelled studies of gas/particle phase partitioning in PG/VG droplets of other components of EC aerosols.	Theoretical modelling studies based on 1:1 PG/glycerol	-	Formaldehyde would be expected to partition almost exclusively to the vapour phase, even at high total particulate mass (TPM), while its potential adducts formed by vaping (methanediol, the hemiacetal with PG) would exist mostly in the particle phase, even at low TPM. Gas/particle partition coefficients reported for other components including acetaldehyde, acrolein, benzene, diacetyl, limonene, benzaldehyde, and nicotine (see Table 1 in Pankow et al., 2017).

Reference	Investigation	Test materials	Experimental methods	Results / aerosol particle characteristics
Prévôt et al. (2017)	Assessed nicotine delivery from EC	3rd generation iStick TC40W battery with GS Tank atomizer (Eleaf) Homemade e-liquid: 18 mg/L nicotine in VG/PG at ratio 20/80 or 80/20, no flavouring	Measurement of MMAD by cascade impaction (DLPI) and size fractionation (12 size fractions from 30 nm to 10 µm) Nicotine in fractions measured by LC-MS Low dilution (1:5), 4 s puff, flow-rate 500 mL/s	MMAD 20%VG/80%PG: 0.79 ± 0.01 µm (GSD, 1.43 ± 0.04) 80%VG/20%PG: 0.76 ± 0.03 µm (GSD, 1.49 ± 0.03) Nicotine distributed equally in the different-sized aerosol fractions; approximately 20-40% nicotine loss compared with levels in e-liquid
Sundahl, Berg and Svensson (2017)	Evaluated EC aerosol particle size distribution	13 different commercially available EC (blu Premium, Blu, SKYCIG, GAMUCCI Micro original, Liberro Realis Lite, Liberro Classic regular Black, Liberro Realis Regular High, Zebra, Supersmoker, Intellcig, Blood, Vapestick, E-lites) e-liquids comprising water, VG and/or PG, nicotine, and flavours	Particle impactor for pharmaceutical inhalers (NGI) (flow rate 15, 30, 60 mL/min) Residence time approx. 1 s	MMADs in the range 0.5–0.9 µm Theoretical modelling predicted nicotine partitioning mostly to the droplet rather than gas phase

Abbreviations: CC, conventional cigarette; EC, electronic cigarette; PNC, particle number concentration; FMPS, fast mobility particle sizer; SMPS, scanning mobility particle sizer; MMAD, mass median aerodynamic diameter; CMD, count median diameter; GSD, geometric standard deviation; CPC, condensation particle counter; VMD, volumetric mean diameter; PG, propylene glycol; VG, vegetable glycerine; TPM, total particulate mass; TDMA, tandem differential mobility analysis;