TOX/2019/11

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

Potential toxicological risks from electronic nicotine (and non-nicotine) delivery systems (E(N)NDS – e-cigarettes). Paper 9: Bystander exposure.

Background

1. The COT is reviewing the potential toxicity of electronic nicotine delivery systems (ENDS) and electronic non-nicotine delivery systems (ENNDS) (collectively abbreviated to E(N)NDS). As part of this review, a paper on characterisation of the aerosol particle fraction (TOX/2017/49) was discussed at the December 2017 COT meeting, and papers summarising evaluations of metals (TOX/2018/15) and other constituents (TOX/2018/16) in E(N)NDS products were presented at the March 2018 COT meeting. These 3 discussion papers focussed on constituents sampled in E(N)NDS liquids or in 'firsthand' E(N)NDS aerosols produced from the E(N)NDS product (without having been inhaled by a user), with relevance to evaluating potential levels of exposure to the E(N)NDS user. Further to this information, the present paper provides a review of the literature relating to exposures that non-E(N)NDS-users may experience as 'bystanders' in indoor spaces where E(N)NDS use is occurring or has taken place.

Introduction

2. E(N)NDS are battery-powered devices containing a liquid (E(N)NDS liquid or 'e-liquid'). The E(N)NDS liquid is heated on use to produce an aerosol that is inhaled by the user ('puffing', 'vaping'). E(N)NDS were first introduced commercially in China in 2004 and subsequently in the EU (2005) and USA (2007) as nicotine-delivery devices. The main constituent parts of an E(N)NDS device are a mouthpiece, cartridge (tank) containing E(N)NDS 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 (CC) 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 E(N)NDS 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). In addition, highly advanced 'fourth generation' E(N)NDS (innovative regulated mods) are now being described.

3. Constituents that have been identified in E(N)NDS liquids and/or aerosols include propylene glycol (PG), vegetable glycerine (VG, glycerin(e), glycerol), water, nicotine, carbonyls, volatile organic compound (VOC), tobacco-specific nitrosamines (TSNAs), polycyclic aromatic hydrocarbons (PAHs), metals, ethanol, ethylene glycol, di-ethylene glycol, flavouring compounds, flavour enhancers, sweeteners, and phenolics. Data on reported levels of some of these constituents in E(N)NDS liquids and aerosols were summarised in discussion papers, TOX/2018/15 and TOX/2018/16, presented at the February 2018 COT meeting.

4. Passive or 'bystander' exposure to CC smoke (secondhand smoke) is well established as a health hazard. However, unlike CC, the design of E(N)NDS products means that emissions from them will be different to those from the burning tip of CC. In terms of environmental exposure to bystanders, E(N)NDS should not produce sidestream emissions and thus the constituents released into ambient air upon E(N)NDS use should comprise only the residual product (particulates and/or vapour) exhaled by the user. Bystanders may be exposed to these components by inhalation of ambient air in the space where the E(N)NDS aerosol has been exhaled (secondhand exposure) or through inhalation of dust or via skin contact with contaminated surfaces where the exhaled E(N)NDS products settle and accumulate (thirdhand exposure). Some authors have noted that such deposited chemicals may persist for long periods and may undergo chemical reaction to form other species (*discussed in* Kuschner et al. (2011)).

Literature searches and scope of the review

5. Searches of the 'PubMed' and 'Scopus' databases were performed on 15/11/2018 to identify published literature relating to 'bystander' exposure to E(N)NDS products. A total of 302 citations were identified, of which 24 were selected for inclusion in this overview. An updated search of PubMed on 05/03/2019 identified 1 additional citation of relevance. Reports that discussed modelling of bystander exposure were not included. Details of the search strategy are provided at Annex A.

Studies of the constituents emitted from E(N)NDS use

6. The constituents emitted after 'real-life' E(N)NDS use (i.e. after inhalation and exhalation of aerosol by one or more users) have been measured in different settings, including in rooms or exposure chambers where E(N)NDS products are used under defined test conditions, or in households, cars, or public buildings where E(N)NDS are used. Measurements have, variously, been reported before, during, and/or after E(N)NDS use, and include sampling of constituents directly in exhaled breath, in ambient air, and deposited on surface materials. Studies have measured vapour and/or particulate phase constituents including PG, glycerine, nicotine, TSNAs, carbonyls, PAHs, carbon monoxide (CO), nitric oxide (NO), other VOCs, metals, fine particles (FP) and ultra-fine particles (UFP).

7. The following literature includes only those studies that have evaluated levels of constituents relating to E(N)NDS use (i.e. after exhalation by human users). Details of the studies of secondhand and thirdhand exposure to E(N)NDS reviewed in this paper are summarised in Table A, attached at Annex B.

Exhaled breath measurements

8. A small number of studies have investigated chemicals present in exhaled breath after E(N)NDS use.

9. A study reported by Long (2014)[1] evaluated levels of phenolics, carbonyls, water, glycerine, and nicotine in exhaled breath before (blank) and after E(N)NDS or CC use. Two disposable E(N)NDS products were used in the study, namely blu eCigs Classic Tobacco Disposable (blu CTD; 82% glycerine, 9% water, 2% nicotine, and 7% flavour), and blu eCigs Magnificant Menthol Disposable (blu MMD; 75% glycerine, 18% water, 2% nicotine, and 5% flavour). The CC brand used was Marlboro Gold King Box filtered cigarette. A total of 30 test subjects were recruited (10 per product type), all of whom were regular dual E(N)NDS and CC users. Subjects abstained from using E(N)NDS and CC for a minimum of 1 h prior to tests. Test sessions (maximum 2 h) were held in a 40 m^3 conference room, and consisted of an average of around 95-99 E(N)NDS puffs (3.7 V, 3.0 Ω), or smoking 3 CC (average around 30 puffs), and each subject underwent 9 test sessions. Exhaled breath constituents were captured on glass fibre filter pads before and after puffing. Room air background levels were determined for carbonyls. Average mass balance for nicotine, glyercine, and water in exhaled breath of E(N)NDS users was 104 ± 18% for blu CTD and 101 ± 7% for blu MMD. Average distribution of mass balances were reported as 73.3% water, 26.7% glycerine, 0.049% nicotine (blu CTD) or 75.7% water, 24.2% glycerine, and 0.057% nicotine (blu MMD), and the authors commented that a previous study had reported that machine-generated mainstream E(N)NDS aerosols contain approximately 86% glycerine and 8% water. The average mass balance for nicotine, glycerine, and water in exhaled breath after CC smoking was 83 ± 21%, with the rest attributed to particulates from combustion. The nicotine proportion in exhaled CC smoke was approximately 0.40%. Levels of individual carbonyls in exhaled breath after E(N)NDS use were described as being generally below the individual limit of detection (LOD) or limit of quantitation (LOQ) values[2] and summed levels of total carbonyls were not significantly different from those in exhaled breath blanks or in room air. Higher levels of total carbonyls, summed from individual measurements, were detected in exhaled breath after CC smoking (average total carbonyls, 242 µg/session, range 136–352 µg/session). The finding was similar for phenolics, with measurements described as generally below the individual LOD or LOQ values for E(N)NDS breath, but with quantifiable levels of

[1] The 'Conflicts of Interest' citation of this publication stated that: "The author is employed by Lorillard, a manufacturer of conventional cigarettes and the parent company of the manufacturer of the e-cigarette products used in this study."

[2] LOD and LOQ values, respectively, for 8 individual carbonyls in exhaled breath ranged from 0.21 and 1.80 µg/session (propionaldehyde) to 0.61 and 13.64 µg/session (acetone). Results of measured levels were only provided for acetadehyde.

individual phenolics present in exhaled CC breath[3] (summed average total phenols, 66 µg/session, range 36–117 µg/session). The authors summarised that: exhaled E(N)NDS aerosol comprised more than 99.9% water and glycerine, with a small amount (< 0.06%) of nicotine; that total levels of phenolics and total levels of carbonyls were not significantly different in exhaled breath before or after E(N)NDS use, but levels of these compounds were greater in exhaled breath after CC smoking. The authors concluded that exhaled E(N)NDS aerosol does not increase bystander exposure for phenolics and carbonyls above background levels in exhaled breath, in contrast to the quantifiable levels of these analytes in exhaled CC smoke.

10. Marco and Grimalt (2015) used a 'Bio-VOC' sampler to compare the presence of 156 VOCs in directly sampled room air, CC smoke, and E(N)NDS aerosol and corresponding exhaled breath samples. An American-brand, filter-containing CC with low nicotine content, low tar, and low CO was used, along with a disposable E(N)NDS product and a rechargeable E(N)NDS product. Exhaled breath samples were collected from volunteer users 30 min after product use. Further details of the products used, the usage protocols, or the numbers of volunteers/tests conducted were not given. Gas chromatography (GC) analysis showed the presence of many VOCs in CC smoke, with a similar spectrum in exhaled breath after CC use, but at generally lower levels. GC analysis of E(N)NDS aerosol showed peaks of PG and glycerine, nicotine and related compounds, and, for the rechargeable E(N)NDS product, vanillin and ethyl vanillin. In all cases (including exhaled air from control participants who had not smoked CC or used E(N)NDS), the main compounds in exhaled breath were acetone and isoprene, presumed to be from endogenous sources. The reported levels of nicotine measured were around 1300 µg/m^3 (CC) and 710-720 µg/m^3 (E(N)NDS) in directly sampled product and 7 µg/m^3 (CC), 1 µg/m^3 (E(N)NDS disposable), and 4 µg/m^3 (E(N)NDS rechargeable) in exhaled breath samples. Nicotine was not detected in room air or exhaled air from control participants. The authors concluded that the Bio-VOC system is suitable for the analysis of VOCs in exhaled breath of CC and E(N)NDS users.

Evaluation of ambient air and surfaces in exposure chambers or rooms after specified E(N)NDS use

11. Experimental studies that have evaluated levels of chemicals present in ambient air and/or on surfaces in rooms or exposure chambers in which human volunteers follow pre-specified protocols of E(N)NDS use are summarised in the following paragraphs.

12. A volunteer took six 3-s puffs of a tank-system E(N)NDS device at intervals of 60 s in an 8 m^3 test chamber. The air exchange rate in the chamber was 0.3/h. Three different E(N)NDS liquids were tested sequentially: Liquid 1 (apple flavour, 0 mg/mL nicotine); Liquid 2 (apple flavour, 18 mg/mL nicotine); Liquid 3 (tobacco flavour, 18 mg/mL nicotine). Following this, the volunteer smoked 1 CC (brand not

[3] LOD and LOQ values, respectively, for 6 individual phenolics in exhaled breath ranged from 0.09 and 0.32 µg/session (phenol) to 0.60 and 4.00 µg/session (*m,p*-Cresol). Results of measured levels were only provided for hydroquinone.

stated). Before and during the study, particle size distribution and levels of 20 VOCs in chamber air were measured. During E(N)NDS use, a bimodal particle size distribution was observed (maxima at 30 and 100 nm), considered by the authors to be due to changes that occurred in the lung of the user prior to exhalation. After subsequent smoking of a CC in the chamber, a single-mode particle size around 100 nm was measured. A set of 20 VOCs was analysed during the study. All 20 VOCs showed levels < 1 µg/m^3 in chamber air before test exposures except for isoprene (8 µg/m^3), and substantially higher levels at the end of the procedure (after CC smoking). During E(N)NDS use (i.e. prior to CC smoking), levels of 5 VOCs increased (2-butanone, 2 µg/m^3; acetic acid, 11-14 µg/m^3; acetone, 17-25 µg/m^3; formaldehyde, 8-16 µg/m^3; acetaldehyde, 2-3 µg/m^3), while values for the other 15 VOCs did not differ from pre-test baseline measurements (all < 1 µg/m^3, except for isoprene, 6–10 µg/m^3). The authors considered that emissions of some of these compounds associated with E(N)NDS use may have been due to endogenous production from the volunteer in the chamber. Levels of these compounds were substantially higher after the CC smoking section of the experiment (see Table A, attached at Annex B). To determine if compounds were emitted in exhaled breath that had not been detected in the large chamber study, a volunteer exhaled 1 puff from E(N)NDS Liquid 1 (apple flavour, 0 mg/mL nicotine) directly into a small (10 L) glass chamber (air exchange rate, 3/h) and VOC concentrations in chamber air were measured. In this case 1,2-propanediol (PG) (not detected in the large-chamber study) was measured at 'a high amount', with other main components being 1,2,3-propanetriol (glycerol), diacetin (flavour), and traces of apple oil and nicotine (Schripp et al. 2013).

13. Czogala et al. (2014)[4] compared levels of emissions of nicotine, particulate matter ($PM_{2.5}$), CO, and VOCs in a room before and after E(N)NDS or CC use. Five adult males who were regular, long-term dual users of E(N)NDS and CC underwent the following test procedure in a 39 m^3 exposure chamber with an air sampler (single user per test session). The air exchange rate in the chamber during this part of the study was not reported. Each participant used their own E(N)NDS product[5] (range 16–18 mg/mL nicotine) *ad libitum* for 2 x 5-min sessions with a 30-min interval, after which the room was decontaminated and ventilated for 5 min. Subsequently the user smoked 2 of their own CC (range 0.5–0.6 mg/cigarette nicotine), with a 30-min interval between each CC. Air concentration measurements were made for 3 x 1 h (at baseline, during E(N)NDS use, and during CC use; 1-h mean values reported). Air concentrations of chemicals measured varied substantially depending on the brand of E(N)NDS/CC used. Average nicotine levels emitted after CC use (31.60 ± 6.91 µg/m^3) were approximately 10-fold higher than those emitted after E(N)NDS use (3.32 ± 2.49 µg/m^3), while the baseline nicotine level was below the LOD of 0.22 µg/m^3. $PM_{2.5}$ levels emitted on E(N)NDS use (151.7 ± 86.8 µg/m^3) were also

[4] The 'Declaration of Interests' citation of this publication states that: "MLG received research funding from Pfizer, manufacturer of stop smoking medication, and was funded by the UK Centre for Tobacco Control Studies (UKCTCS) during the study. AS received research funds and travel expenses from Chic Group Ltd., manufacturer of electronic cigarettes in Poland. Other authors declare no conflicts of interest."
[5] Details of the E(N)NDS and CC products used by the participants are listed in Table A, attached at Annex B.

higher than baseline (32.4 ± 30.0 µg/m^3) but lower than those on CC smoking (819.3 ± 228.6 µg/m^3). CC smoking was associated with increased levels of CO and VOCs compared with baseline air, but E(N)NDS use was not associated with increased levels of these chemicals compared with those in baseline air. The authors commented that using E(N)NDS in an indoor environment may involuntarily expose nonusers to nicotine, and that secondhand exposure to nicotine from E(N)NDS is on average 10-fold less than from tobacco smoke.

14. Ruprecht et al. (2014) measured particulate matter emissions from E(N)NDS and CC into ambient air of a 50 m^3 room in an Italian research centre. The air exchange rate in the room was determined to be 0.80/h to 0.86/h. For this protocol, a volunteer used an E(N)NDS (Elips Serie C, Tank System) with 0 mg/mL or 16 mg/mL nicotine at a fixed rate of 1 puff/min during 7 min, then a 3-min interval, for 2–3 h. E(N)NDS tests were carried out on different days, after room ventilation, and the CC test was carried out afterwards (1 CC of unstated brand smoked in the room). Total suspended particulate (TSP) concentrations measured by Aerocet Model 531 optical particle counter were approximately 12 µg/m^3 after use of E(N)NDS without nicotine and 160 µg/m^3 after smoking 1 CC. TSP values measured after use of E(N)NDS containing nicotine were very low and/or negative (values were calculated by subtracting against outside ambient air), which the authors suggested may be due to different optical properties of the aerosol in the presence/absence of nicotine.

15. Saffari et al. (2014) found that particle-phase emissions were much lower from E(N)NDS use than CC smoking, PAHs were not detected from E(N)NDS use, and emission rates of organic compounds and inorganic elements were significantly lower from E(N)NDS use compared with CC smoking. This study compared indoor air emissions of particulate matter, several metals, organic species, and nicotine into a room in a cancer research centre in Italy during CC smoking or E(N)NDS use with levels in outdoor air on an adjacent terrace. The air exchange rate in the room during sampling was 1.1/h. Levels of TPM were approximately 50 µg/m^3 in outdoor air, 50 µg/m^3 indoors during E(N)NDS use, and 250 µg/m^3 indoors during CC smoking. Emission rates of metals into indoor air during E(N)NDS use were calculated for 10 of the 25 metals evaluated, as follows: boron, 963.8 ng/h; potassium, 7765 ng/h; titanium, 50.16 ng/h; chromium, 28.10 ng/h; nickel, 130.5 ng/h; zinc, 1142 ng/h; silver, 20.91 ng/h; cadmium, 0.480 ng/h; lanthanum, 3.210 ng/h; lead, 96.16 ng/h. The other 15 elements (magnesium, aluminium, sulphur, calcium, vanadium, manganese, iron, cobalt, copper, rubidium, strontium, molybdenum, tin, antimony, and tungsten) were not detected during E(N)NDS use. Of the 10 elements detected during E(N)NDS use, 4 (titanium, chromium, nickel, silver) were estimated to have higher indoor emission rates from E(N)NDS use compared with CC smoking[6] (during CC smoking titanium and chromium were not detected; emission rates were 36.39 ng/h for nickel and 14.65 ng/h for silver). Of the 25 metals evaluated, 6

[6] Emissions were detected for 11 elements during CC smoking, as follows: boron 23,680 ng/h; sulphur 34,540 ng/h; potassium 297,500 ng/h; nickel 36.39 ng/h; copper 1029 ng/h; zinc 8252 ng/h; rubidium 200.1 ng/h; silver 14.65 ng/h; cadmium 657.3 ng/h; lanthanum 1846 ng/h; lead 1012 ng/h.

showed higher levels indoors during E(N)NDS use than in outdoor air (ratio indoors during E(N)NDS use / outdoors: boron, 13.13; potassium, 1.53; nickel, 1.75; zinc, 1.26; silver, 3.39; lanthanum, 1.47). Analysis of the E(N)NDS liquid used indicated that this was not the source of the metal emissions during E(N)NDS use. PAHs were not detected during E(N)NDS use. Nicotine measurements in indoor air were 1524 ng/m^3 during CC smoking, 60.68 ng/m^3 during use of E(N)NDS containing 16 mg/mL nicotine, and 18.6 ng/m^3 during use of E(N)NDS containing no nicotine. The authors considered that, in the latter case, the observed (small) nicotine levels could be due to particle-bound nicotine resuspension in the room during sampling (i.e. 'background' or 'blank' emission level). A nicotine emission rate was calculated from E(N)NDS containing nicotine, of 4344 ng/h, and from this they estimated that 0.02% of total nicotine emission from this E(N)NDS product was released into the air as nicotine associated with particulate matter. They noted, however, that the study did not evaluate components in the vapour phase, and suggested that further investigation of gas-to-particulate partitioning of nicotine would be recommended. In conclusion, the authors commented that particle-phase emissions were much lower from E(N)NDS use than from CC smoking, PAHs were not detected from E(N)NDS use, and emission rates of organic compounds and inorganic elements were significantly reduced from E(N)NDS use compared with CC smoking. Some metals were noted to have higher emission rates from E(N)NDS than from CC smoking, with nickel and silver being of possible concern. Secondhand, particle-phase nicotine was estimated to be 0.02% of the total nicotine generation and emission from E(N)NDS use.

16. Schober et al. (2014) measured indoor air quality in a 45 m^3 ventilated room, before and during use of E(N)NDS (without and with 18 mg/mL nicotine). The air exchange rate in the room was 0.76/h for the control session and ranged from 0.37-0.74/h for the 6 different E(N)NDS sessions. For each set of test sessions, 3 volunteers concurrently used E(N)NDS in the room for a period of 2 h for 6 consecutive days. During the first session, E(N)NDS product without nicotine was used, and during the subsequent 5 sessions the same product with nicotine was used. In total, 3 E(N)NDS liquid types were tested (hence 9 volunteers in total). Control air samples were taken from the room the day before the first test session. Levels of particulate matter, CO and carbon dioxide (CO_2), PG, glycerine, nicotine, aldehydes, PAHs, and metals were measured. E(N)NDS use was associated with the emission of substantial amounts of PG, glycerine, and nicotine. Mean levels during E(N)NDS use were 199.2 ± 93.2 µg/m^3 (maximum, 395 µg/m^3) for PG; 72.7 ± 6.9 µg/m^3 (maximum, 81 µg/m^3) for glycerine; and 2.2 ± 1.7 µg/m^3 (maximum, 4.6 µg/m^3) for nicotine. Mean levels of all 3 chemicals in control air were < 0.04 µg/m^3. Increased levels of particulate matter were also observed during E(N)NDS use. Mean $PM_{2.5}$ concentrations were 197 µg/m^3 (maximum, 514 µg/m^3) during E(N)NDS use and 6 µg/m^3 in control air. There was a 2.4-fold increase in aluminium levels during E(N)NDS use (482.5 ± 158.6 ng/m^3) compared with control air (203.0 ng/m^3).

17. In a study reported by O'Connell et al. (2015)[7] levels of PG, glycerol, nicotine, VOCs, carbonyls, PAHs, TSNAs, and trace metals were measured in a 38.5 m^3 office meeting room, before, during, and after a 165-min session of E(N)NDS use. The average air exchange rate in the room was 0.8/h. In total, 5 participants were present in the room during the test session, of whom 3 used, *ad libitum*, 'Puritane 16 mg/g disposable original-flavoured, closed-system E(N)NDS' (67% PG, 30% glycerol, 1.6% nicotine, flavourings), while the other 2 participants did not use E(N)NDS. Measurements were made before participants entered the room ('background', for 60 min), after they entered the room ('occupancy', for 60 min), during E(N)NDS use ('during E(N)NDS', for 165 min), and after participants had left the room ('after', for 75 min). Levels of glycerol, nicotine, 16 PAHs, 31 trace elements, and 4 TSNAs were below the LODs at all time points, for the detection methods used. The authors considered that the lack of detection of nicotine (LOD, 7 µg/m^3) was likely due to high retention of nicotine in the body. Average levels of PG were below the LOD at background and in the occupied room before E(N)NDS use (< 0.5 µg/m^3), rose substantially during E(N)NDS use (203.6 µg/m^3), then fell in the empty room after E(N)NDS use (10 µg/m^3). The authors commented that levels were well below the UK workplace exposure limit (WEL) for PG of 474,000 µg/m^3. Glycerol was not detected, which may have been due to the relatively high LOD of the detection method (350 µg/m^3). Authors noted that the UK WEL for glycerol is 10,000 µg/m^3. Total volatile organic compounds (TVOC) rose from 65.0 µg/m^3 in background air to 237.0 µg/m^3 in occupied room, 379.0 µg/m^3 during E(N)NDS use, then fell to 129.0 µg/m^3 afterwards. The authors considered the increase in TVOC was likely to be related to occupation of the room (cosmetic and toiletry products worn by the participants) rather than to E(N)NDS use. Levels of formaldehyde (32.0 µg/m^3 background, 31.0 µg/m^3 occupancy, 37.6 µg/m^3 during E(N)NDS, 21.0 µg/m^3 after), acetaldehyde (9.0 µg/m^3 background, 6.5 µg/m^3 occupancy, 12.4 µg/m^3 during E(N)NDS, 6.0 µg/m^3 after), and acrolein (< LOD of 2.0 µg/m^3 at all times) did not rise substantially during E(N)NDS use. The authors noted these levels to be below WHO or EU indoor air quality guideline (AQG) levels. The authors also commented that exposure of bystanders to the chemicals in the exhaled E(N)NDS aerosol, at the levels measured in this study, would be below current regulatory standards that are used for workplaces or general indoor air quality (where these have been established), and they concluded that this finding supports the conclusion that there is no apparent risk to bystanders from exhaled E(N)NDS aerosols.

18. Maloney et al. (2016)[8] used active integrated air sampling, near-real-time and direct-measurement techniques to evaluate emissions of nicotine, menthol, PG, glycerol, formaldehyde, and total particulates associated with the use of E(N)NDS ('MarkTen' prototype, menthol and non-menthol flavours containing 1.5–2.5%

[7] The 'Conflicts of Interest' listing of this publication states that "All authors are employees of Imperial Tobacco Group. The work in this manuscript was supported by Imperial Tobacco Group. Imperial Tobacco Group is the parent company of Fontem Ventures B.V., the manufacturer of the e-cigarette products used in this study."
[8] The 'Funding' acknowledgement section of this publication notes that "All authors of this study are current or retired employees of Altria Client Services which is a subsidiary of Altria Group. NuMark, a subsidiary of Altria Group, is a manufacturer of electronic cigarettes. Funding for this project was provided by Altria Client Services."

nicotine) in a 137 m^3 room. Data were collected over 2 separate study periods. The air exchange rate in the room was 1.47/h during Study 1 and 1.56/h during Study 2. Test sessions comprised the use of E(N)NDS product by 2–12 users for 6 periods of 1 h (total, 36–216 puffs per 1-h session), interspersed with 1-h non-use periods. Levels of particulate matter were noted to spike during test sessions, returning to background after approximately 1 h of non-use, which the authors took as a 'good indication of product usage'. For the other measurements taken, except for formaldehyde, all were below the LOQ for the test methods used (ranges of LOQ values over the 2 separate study periods were 180–210 µg/m^3 for menthol, 10–15 µg/m^3 for nicotine, 45–65 µg/m^3 for PG, 53–77 µg/m^3 for glycerol, 39–153 µg/m^3 for TSP, and 1 µg/m^3 for formaldehyde (Study 2 only)). Formaldehyde levels during E(N)NDS use were similar to background (around 5–8 µg/m^3), suggesting that E(N)NDS use was not the source of formaldehyde in the room air. The authors concluded that indoor use of MarkTen prototype E(N)NDS does not produce chemical constituents at quantifiable levels or background levels using standard industrial hygiene collection techniques and analytical methods.

19. Liu et al. (2017)[9] measured levels of nicotine, PG, glycerol, 15 carbonyls, 12 VOCs, and 4 trace elements (chromium, nickel, cadmium, arsenic) under controlled and *ad libitum* use conditions of 3 different E(N)NDS products or CC. The studies were carried out in a 114 m^3 exposure chamber, with groups of 8–10 participants using MarkTen (2.5% nicotine) or a prototype, cartridge-based E(N)NDS (2.4% nicotine) under pre-specified conditions (total 80 puffs per user) or *ad libitum* conditions (1224 and 747 puffs total for MarkTen and cartridge, respectively), or a tank-system (approximately 6–24 mg/mL nicotine) (1649 puffs total) or CC under *ad libitum* conditions (45 CC total), for 4-h periods. Fresh air was supplied to the room at a rate of 7.5 L/s. Prior to product use, measurements were taken in the exposure chamber with participants absent (control) and then present (baseline). Measurements during product use were reported as mean change compared with the baseline. Apart from nicotine, PG, and glycerol, the majority of the other measured chemicals were below the LOQ during E(N)NDS use. For the remaining chemicals that showed measurable changes during E(N)NDS use, both positive and negative changes compared with baseline were observed. In general, the changes were small and considered by the authors not to be of concern. Levels of PG and glycerol, although significantly increased during E(N)NDS use, were nevertheless noted by the authors to be well below WELs. The ranges of mean increase over baseline associated with use of the different E(N)NDS products, over the different usage conditions, were as follows: nicotine, 0.38–2.83 µg/m^3 (LOQ, 0.25 µg/m^3); PG, 33.06–317.06 µg/m^3 (LOQ, 3.63 µg/m^3); glycerol, 67.89–242.00 µg/m^3 (LOQ, 4.11 µg/m^3). For comparison, during *ad libitum* CC smoking, the mean increases over baseline were 40.65 µg/m^3 (nicotine) and 56.21 µg/m^3 (PG), while glycerol was not

[9] The 'Conflicts of Interest' section of the publication states that "The study was funded by Altria Client Services LLC. The authors, Mohamadi Sarkar, Jianmin Liu, Qiwei Liang, Michael J. Oldham, Ali A. Rostami and Karl A. Wagner are employees of ALCS. I. Gene Gillman, Piyush Patel and Rebecca Savioz are paid contractors. The study was conducted on behalf of NuMark LLC., (Richmond, VA, USA) a subsidiary of Altria Group, that produces and markets e-vapor products."

detected. Full results are given in Annex B, Table A. The authors concluded that under the conditions of this study, E(N)NDS products did not generate chemicals at levels that could likely pose health concerns for nonusers.

20. Melstrom et al. (2017) reported that 2-h *ad libitum* E(N)NDS use (disposable and tank-style test products[10] containing ≥ 18 mg/mL nicotine), by 3 experienced users in a 52.6 m^3 room, produced elevated air levels of $PM_{2.5}$, UFP, and nicotine, and accumulation of nicotine on surfaces and clothing compared with baseline measurements. Ambient air levels of particulates were measured at table-level in the exposure room, while air nicotine in the gas phase was measured using personal air samplers at mouth-level of nonusers present in the room. Measurements were made before, during, and after the 2-h E(N)NDS use sessions. The mean air exchange rate in the room was approximately 5/h. Values recorded are shown in Table 1, below. $PM_{2.5}$, UFP, and nicotine levels were all significantly increased during E(N)NDS product use, as compared with before or after use. $PM_{2.5}$ levels were significantly higher when using the tank-style product compared with the disposable product, while the converse was true for levels of UFP. Median accumulation rates of nicotine using disposable and tank-style E(N)NDS were calculated as 2.1 and 4.0 ng/100 cm^2/h on surfaces, and 44.4 and 69.6 ng/cm^2/h on cloth samples, respectively. The authors commented that short-term E(N)NDS use can produce elevated $PM_{2.5}$, UFP, and nicotine in air, and accumulation of nicotine on surfaces and clothing. This could lead to dermal absorption of nicotine and to secondhand inhalation of particles and associated chemicals by bystanders.

Table 1. Ambient air concentrations of $PM_{2.5}$, UFP, and nicotine before, during, and after 2-h *ad libitum* use of disposable and tank-style E(N)NDS containing ≥ 18 mg/mL nicotine by 3 experienced users in a 52.6 m^3 room (6 non-users were also present in the room) (from the study of Melstrom et al. 2017).

		Before		During		After	
		Disposable	Tank	Disposable	Tank	Disposable	Tank
$PM_{2.5}$ (mg/m^3)	Mean (SD)	0.131 (0.019)	0.018 (0.015)	0.788 (2.147)	1.454 (2.683)	0.008 (0.008)	0.017 (0.011)
	Median	0.010	0.015	0.035	0.515	0.006	0.014
	Range	0.003–0.761	0.006–0.827	0.002–19.961	0.007–19.972	0.001–0.228	0.005–0.390
UFP (particles/cm^3)	Mean	4092	3323	38,695	31,227	2829	3216
	Range	0–10,700	2190–4680	1800–239,000	3360–225,000	1360–4730	2130–4100
Nicotine* (µg/m^3)	Mean	0.004	0.011	0.717	1.680	0.114	0.145
	Range	0.003–0.005	0.009–0.014	0.445–0.989	1.158–2.047	0.100–0.124	0.129–0.168

*LOQ, 0.2 ng/sampling air tube

21. A study to evaluate the release of submicronic particles (SMPs) into ambient air in a 52.7 m^3 room during use of combustion (CC; hand-rolled CC; cigar; pipe with tobacco) or non-combustion (IQOS heated tobacco product; Smooke E-SMART (L) E(N)NDS with Smooke Light e-liquid containing 9 mg/mL nicotine) tobacco products was reported by Protano et al. (2017). Test sessions comprised use of the product

[10] Further product details were not given.

by volunteers for 4 min, except for cigar (30 min) and pipe (45 min), at 3 x 1-h intervals. Aerosol particle number size distributions were measured by Fast Mobility Particle Sizer (FMPC) in the range 5.6–560 nm. The air exchange rate in the room was 0.67/h. Results, as described narratively in the publication, noted that peak concentrations were in the range of 1.2 x 10^5 to 2.9 x 10^5 particles/cm^3 with combustion products, and were < 4.7 x 10^4 particles/cm^3 for IQOS and E(N)NDS. The 1-h time interval was sufficient for the particle number to fall to background for E(N)NDS, but not the other products. The authors commented that as both IQOS and E(N)NDS emitted SMPs, a ban on their use indoors should be supported.

22. Protano et al. (2018) published a further evaluation of levels of particulate matter released into air on E(N)NDS use. This study evaluated 4 generations of E(N)NDS products (both with and without nicotine), using a similar test setting as that reported by Protano et al. (2017) (52.7 m^3 room), described in paragraph 21, above. PM_1 levels were sampled before and during E(N)NDS use by portable, laser-operated aerosol mass analyser (0.1–10 µm) placed at approximately 1.5 m from a volunteer who took 12 puffs from an E(N)NDS product over a period of 5.5 min. The air exchange rate in the room was not reported. PM_1 levels were significantly higher during E(N)NDS use compared with before use for all test sessions. PM_1 emissions during a range of 8 different test conditions using 1st, 2nd, or 3rd generation E(N)NDS products were in the following ranges. *Before E(N)NDS use:*18–44 µg/m^3 (arithmetic mean), 18–38 µg/m^3 (median). *During E(N)NDS use:* 54–3429 µg/m^3 (arithmetic mean),17–648 µg/m^3 (median). In tests with 4th generation products, over a total of 12 test conditions, emission levels varied widely, with ranges as follows: *Before E(N)NDS use:* 21–45 µg/m^3 (arithmetic mean), 20–43 µg/m^3 (median). *During E(N)NDS use:* 75–14,887 µg/m^3 (arithmetic mean), 39–3475 µg/m^3 (median). The highest levels were measured during use of a 4th generation product operated at 0.4 Ω/80 W, an approach known as sub-ohming, using a nicotine-containing e-liquid. The authors commented that particulate matter emissions occur during use of all generations of E(N)NDS products and that the emissions of nanoparticles should be investigated further.

Evaluation of ambient air and surfaces in real-life settings where E(N)NDS use takes place

23. In addition to experimental studies of ambient-air levels of chemicals in association with E(N)NDS use, other studies have been carried out that have measured levels of chemicals in ‘real-life’ environments where E(N)NDS use occurs, including private households, vehicles, shops where E(N)NDS products are sold, and convention centres holding ‘vaping conventions’.

Household

24. Ballbe et al. (2014) reported that nicotine levels were significantly different in ambient air of the main family room in households where CC smoking, E(N)NDS use, or neither (control) took place. The air exchange rates in the rooms were not reported. Geometric mean (GM) nicotine concentrations were 0.74 µg/m^3 in CC-

smoking households (n = 25 sampled), 0.13 µg/m^3 in E(N)NDS-use households (n = 5 sampled), and 0.02 µg/m^3 in control households (n = 24 sampled). The reported LOQ for nicotine was 5 ng/filter (suspended from the ceiling for 1 week), equivalent to 0.02 µg/m^3 air concentration over 1 week. Further findings from this study on biomarkers of nicotine exposure are summarised in paragraph 35.

25. Surface nicotine levels in 8 homes where regular use of E(N)NDS products containing 10–15 mg/mL nicotine (50–500 puffs/day) occurred were similar to those taken from 8 control homes with no E(N)NDS use and no CC smoking (7.7 ± 17.2 and 7.2 ± 13.8 µg/m^2, respectively). Conversely, levels in 6 homes where regular CC smoking took place were > 200-fold higher (1303 ± 2676 µg/m^2). Details of room ventilation were not reported. The authors concluded that thirdhand nicotine exposure from E(N)NDS use is low (Bush and Goniewicz 2015).

26. The median $PM_{2.5}$ level in a household during active E(N)NDS use (*ad libitum*, 42 puffs during 1 h) was not significantly different to those measured in air of 2 households of non-CC/non-E(N)NDS users (in all cases, levels were around 9–10 µg/m^3). The median $PM_{2.5}$ level in air in a household where CC smoking was occurring (3 CC during 1 h) was 572 µg/m^3. Measurements were made 2 m from the subject actively using E(N)NDS or smoking CC. Details of room ventilation were not reported (Fernandez et al. 2015).

Vehicles

27. Schober et al. (2019) measured levels of PM and VOCs emitted into indoor air in cars during CC smoking or use of a heated tobacco or E(N)NDS product. Measurements were made in a total of 7 cars, with interior volume classed as large (5–7 m^3; n=2), medium (3–4 m^3; n=3), or small (2–3 m^3; n=2). Tests were performed with front passenger windows open 2 cm or 5 cm, all other windows closed, and air recirculation set to 'off'. The ventilation speed in the vehicles was described as '0–50 km/h'. Two subjects were present in the car during each test; 1 driver and 1 front-seat passenger who used the test product. Seven tests were performed per vehicle, each while driving around the same 8.5-km circuit in Munich, Germany, as follows. 'IQOS': Passenger used IQOS heated tobacco product with 2 Bronze Label Heet sticks (0.5 mg/Heet stick) (2 test circuits: window open 2 cm, window open 5 cm). 'E(N)NDS': Passenger continually used E(N)NDS (SubTwin Neo tank model with tobacco-flavoured e-liquid containing 18 mg/mL nicotine) (2 test circuits: window open 2 cm, window open 5 cm). 'CC': Passenger smoked 2 CC (Marlboro Red, 0.8 mg nicotine/CC) (2 test circuits: window open 2 cm, window open 5 cm). 'Control': No product use, window open 5 cm. The vehicle was fully ventilated for 10 min in-between each test. Measurements were made in the rear passenger seat area, for particulate matter (PNC and $PM_{2.5}$), VOCs (including PG and nicotine), and carbonyls. Results were presented as individual measurements for each test circuit performed. LOD values were not reported except for carbonyls (1.5 or 2.0 µg/m^3 per individual chemical). Neither the car size nor the extent to which the windows were open were significantly correlated with levels of chemicals measured in the air inside

during tests. Particulate matter levels increased over background during use of all test products, with the highest levels measured during CC smoking. $PM_{2.5}$ ranges were: 4–11 µg/m^3 (control), 4–34 µg/m^3 (IQOS), 8–490 µg/m^3 (E(N)NDS), and 64–1988 µg/m^3 (CC). PG was not detected during control or IQOS tests. During E(N)NDS use, PG was not detected in the 2 small cars, while levels were in the range of 50–762 µg/m^3 in the medium and large vehicles. During CC smoking, PG was mostly not detected, although a few (4/14) positive readings were noted, in the range of 36–94 µg/m^3. Nicotine was < LOD in all of the control tests. During use of IQOS, nicotine levels were < LOD in 4 vehicles, while some positive readings were noted in the other 3 cars (4/14 positive readings, in the range of 4–12 µg/m^3). During E(N)NDS use, the nicotine concentration was < LOD in 3 vehicles, with some positive readings noted for the other 4 vehicles (6/14 readings positive in total, in the range of 4–10 µg/m^3). During CC smoking, nicotine was detected in 6 of 7 vehicles (12/14 positive readings), in the range of 8–140 µg/m^3. Levels of aldehydes and ketones increased significantly during CC smoking but did not increase significantly over background levels during IQOS or E(N)NDS use. Authors concluded that, overall, CC, E(N)NDS, and IQOS are avoidable sources of indoor air pollutants, in particular $PM_{2.5}$ and nicotine, and to protect the health of occupants, in particular children and pregnant women, these products should not be used in cars.

Public buildings and vaping conventions

28. Soule et al. (2017) measured $PM_{2.5}$ levels in a 4023 m^3 event room in a hotel during a 2-day E(N)NDS event. Details of room ventilation were not reported. At 6 different time-points during the event, between 59–86 individuals were noted to be actively using E(N)NDS in the room. The overall median $PM_{2.5}$ concentration during the event over 6 measurement sessions was 595.31 µg/m^3 (mean, 607.12 µg/m^3). The range of median levels measured the day before the event was 1.92–3.20 µg/m^3 and the day after the event, 12.80–15.52 µg/m^3. The authors commented that indoor E(N)NDS use exposes nonusers to secondhand E(N)NDS aerosol, and that regulatory bodies should consider establishing policies that prohibit E(N)NDS use in places where CC smoking is prohibited.

29. Chen et al. (2017) measured PM_{10}, VOC, CO_2, NO_2, and nicotine concentrations at a vaping convention held in a 13,475m^3 venue in Maryland, USA during April 2016. PM_{10}, total VOCs, CO_2, and nitrogen dioxide (NO_2) were measured in real time outdoors, indoors, and during plume competitions (2 trick 'artistic plume' competitions, 1 'big plume size' competition). Nicotine and PM_{10} were measured by time-integrated monitoring (overall measurement from 400 minutes of monitoring, of which 58 minutes outdoors and 343 min indoors). Results of real-time and time-integrated sampling are shown in Tables 2 and 3, below. Median PM_{10}, TVOC, and CO_2 concentrations were higher indoors than outdoors and increased further during plume events. CO_2 and TVOC levels were significantly correlated with PM_{10} levels, which the authors considered to support the conclusion that exhaling was the major source of PM_{10} and TVOCs. The estimated 24-h time-weighted average (TWA) for PM_{10} (including all indoor and outdoor measurements) was

1800 µg/m^3, which was noted by the authors to be 12-fold higher than the EPA 24-h limit of 150 µg/m^3. Mean PM_{10} levels were in the range 8500–9000 µg/m^3 by both real-time and integrated sampling. The time-integrated nicotine concentrations were 109 µg/m^3 and 140 µg/m^3 for each of 2 monitors carried around the venue during the sampling period (mean, 124.7 µg/m^3). Although actual values for the air exchange rate in the venue during the vaping convention were not reported, the authors commented that the venue was probably not adequately ventilated; despite the fact that CO_2 monitoring indicated adequate ventilation, the authors considered that the high presence of other aerosol components in the air was likely to have interfered with these CO_2 measurements. The authors described how, for example, ventilation was deliberately reduced during competitions (organisers requesting closing of doors and windows) to increase the visibility of the generated plumes. Overall, the authors concluded that E(N)NDS aerosol in a vaping convention that congregates many E(N)NDS users is a major source of PM_{10}, air nicotine, and VOCs, impairing indoor air quality, and that the findings also raise occupational concerns for E(N)NDS vendors and other venue staff workers.

Table 2. Real-time sampling results reported by Chen et al. (2017).

	Median (Inter-quartile range (IQR))			
	Outdoor	Indoor	Trick competition	Plume size competition
PM_{10} (µg/m^3)	228 (14–8,468)	11,327 (9050–13,182)	7,987 (9,425–11,727)	11,928 (11,245–14,298)
TVOCs (ppm)	0.06 (0.05–0.1)	0.13 (0.11–0.19)	0.10 (0.10–0.10)	0.20 (0.19–0.25)
NO_2 (ppm)	0.25 (0.1–0.32)	0.09 (0.06–0.11)	0.11 (0.05–0.13)	0.06 (0.05–0.09)
CO_2 (ppm)	448 (346–715)	870 (740–1,035)	737 (668–766)	1,051 (1,003–1,150)

Table 3. Time-integrated samples (58 min outdoor + 342 min indoor), results reported by Chen et al. (2017).

	Measurement		
Sampling technique* (sample no.)	Active (1)	Active (2)	Passive (1,2)
PM_{10} (µg/m^3)	8,850	8,429	Not reported
Nicotine (µg/m^3)	109.2	140.2	< LOD^

*Active sampling (sampler flow rate, 3 L/min); passive sampling (sampler flow rate, 25 mL/min); ^ LOD value not reported

30. Johnson et al. (2018) reported a NIOSH-funded study to measure levels of PG, nicotine, formaldehyde, acetaldehyde, and acrolein at 4 indoor vaping events[11] in the USA. Ventilation rates in the venues were not reported. However, the authors noted that venues for Events 1 and 3 were modern buildings with high ceilings typical of a large convention centre (approximately 30–45 feet [9–14 m]) and a noticeable air draft that indicated a highly functioning heating, ventilation, and air

[11] Event 1: Daytona Beach, Florida, April 2016: 42,146 sq.ft convention centre, ceiling height 45 ft, ventilation and air conditioning; Event 2: Athens, Georgia, August 2016: 5100 sq.ft concert hall, 35 ft ceiling height; Event 3: Chattanooga, Tennessee, October 2016: 36,000 sq.ft convention centre, 30 ft ceiling height, ventilation and air conditioning; Event 4: Atlanta, Georgia, March 2017: 205,000 sq.ft exhibition/tradeshow, 13 ft ceiling height.

conditioning system. Event 4 venue was a tradeshow with lower ceilings (approximately 13 feet [4 m]). Event 2 venue was a small concert hall with 2 levels of balconies and a high ceiling (approximately 35 feet [11 m]). All venues had doors open during the events. Levels were measured by collection in sorbent tubes carried in portable backpacks. Reporting limits were described as 20 µg/sample (PG), 0.1 µg/sample (nicotine), 0.1 µg/sample (formaldehyde), 0.5 µg/sample (acetaldehyde), and 2.0 µg/sample (acrolein). PG was detected at high levels (median, 305 µg/m^3; range, < LOD–490 µg/m^3; 25% and 75%, 230 and 410 µg/m^3) at E(N)NDS conventions, but not when venues were empty or at crowded non-E(N)NDS events. The median level of 305 µg/m^3 during E(N)NDS events was noted by the authors to be much higher than the ATSDR intermediate minimum risk level (MRL) for inhalation of PG of 28.01 µg/m^3, and they considered that in light of the growing use of E(N)NDS, guideline values for inhalation of PG may need to be re-assessed. Nicotine was detected at low levels (median, 1.1 µg/m^3; range, < 0.36–2.2 µg/m^3) during E(N)NDS events, but not when venues were empty or crowded at non-E(N)NDS events. The authors considered this finding to be in keeping with other reports in the literature for secondhand exposure to nicotine in settings where E(N)NDS use occurs. Levels of formaldehyde (median, 12.0 µg/m^3 at E(N)NDS event, 10.5 µg/m^3 at crowded non-E(N)NDS event, and 12.5 µg/m^3 at empty venue), and acetaldehyde (median, 9.7 µg/m^3 at E(N)NDS event, 15.5 µg/m^3 at crowded non-E(N)NDS event, and 3.5 µg/m^3 at empty venue) were generally similar at crowded events with or without E(N)NDS use. Acrolein was not detected in any of the samples. The authors noted that the study used sorbent tubes, which are only designed to capture chemicals in the gas phase, thus chemicals present in the particle phase may not have been measured.

31. Khachatoorian et al. (2018) reported that residues of nicotine, other alkaloids, and TSNAs were identified from sample materials (paper towels, terrycloth towels, test filters) taken from an actively trading shop in a shopping centre that was close to a vaping shop in which active vaping occurred, and with active air recirculation from the vaping shop to the actively trading shop. Air flow through the site was reported as approximately 39.67–48.39 m^3/min, and there was an air filter in the return vent from the vaping shop to the actively trading shop where measurements were performed. Levels of the compounds measured generally increased with the time that the sample materials were left in place. The authors concluded that in a multi-tenant retail building, chemicals in E(N)NDS aerosol travelled from a vape shop into an adjacent business where they deposited forming E(N)NDS exhaled aerosol residue. However, measurements from the vaping shop or other sites were not provided for comparison.

Biomarkers of bystander exposure to E(N)NDS aerosols

32. A small number of studies have investigated alterations in biomarkers in subjects with possible exposure to E(N)NDS products as bystanders.

33. Flouris et al. (2012) reported that exposure to CC smoke, both through active smoking by CC smokers and also from passive exposure of never-smokers to secondhand smoke, was associated with changes that the authors report are significant in white blood cell (WBC), lymphocyte and granulocyte counts after exposure, as compared with measurements at baseline (before exposure). Conversely, no significant changes from baseline values were observed after either active (CC smokers) or passive (never-smokers) exposure to E(N)NDS aerosol containing 11 mg/mL nicotine. However, from the figures in which the results are presented it is difficult to judge how statistical significance could be determined.

34. In a companion report describing a similar set of studies, Flouris et al. (2013) noted that serum cotinine levels were similar after active exposure of smokers to either CC smoke (60.6 ng/mL) or E(N)NDS aerosol containing 11 mg/mL nicotine (61.3 ng/mL). Serum cotinine levels were also similar after passive exposure of nonsmokers to either CC smoke (2.4 ng/mL) or E(N)NDS aerosol (2.6 ng/mL), although these were lower than with active exposures. Statistical analysis indicated that cotinine levels were higher immediately after, as well as 1 h after passive exposure of nonsmokers to either CC smoke or E(N)NDS aerosol, as compared with serum levels in controls (nonsmokers exposed to room air). Neither the active nor the passive E(N)NDS exposures were associated with significant changes in any of the other parameters measured in this study, which included lung function tests and exhaled CO levels. Conversely, lung function (FEV_1/FVC) was significantly reduced after the active CC exposure, and exhaled CO was significantly increased after both the active and the passive CC exposures.

35. A study reported by Ballbe et al. (2014) that evaluated nicotine levels in households in which E(N)NDS use (n = 5), CC smoking (n = 25), or neither (n = 24) took place is described in paragraph 24, above. This study also evaluated biomarkers of nicotine exposure in subjects living in these households who did not smoke CC or use E(N)NDS ('nonusers'). Details of ventilation in the homes were not reported. Salivary and urinary cotinine levels were significantly higher in nonusers living in homes where E(N)NDS use took place, compared with levels in nonusers living in control homes (no E(N)NDS use or CC smoking). Similarly, salivary and urinary cotinine levels were significantly higher in nonusers living in homes where CC smoking occurred compared with levels in nonusers living in control homes. Conversely, salivary and urinary cotinine levels were not significantly different between nonusers in homes where E(N)NDS took place compared with homes where CC smoking occurred, suggesting passive absorption of nicotine in both settings. Measured GM (or median) cotinine levels in nonusers living in control, E(N)NDS use, and CC smoking homes, respectively, were 0.07 (0.05), 0.19 (0.24), and 0.38 (0.32) ng/mL in saliva, and 0.70 (0.72), 1.75 (2.64), and 2.46 (2.58) ng/mL in urine. Air nicotine, nonuser salivary cotinine, and nonuser urinary cotinine levels were highly correlated ($p < 0.001$). The authors concluded that nonsmokers passively exposed to E(N)NDS absorb nicotine.

36. Melstrom et al. (2018) measured serum, saliva, and urinary cotinine levels in 6 non-E(N)NDS-users who were present during 2-h test sessions where 3 volunteers used E(N)NDS (2 separate sessions with either disposable or tank-style products with e-liquid nicotine concentrations ranging from 12–20.5 mg/mL). Tests were conducted in a 52.6 m^3 room, with a mean air exchange rate of approximately 5/h. Baseline cotinine levels, measured in nonusers before secondhand exposure, were variable and showed an overall higher level than that reported for the general population (based on data from the 2007–2008 US National Health and Nutrition Examination Survey (NHANES) (CDC 2010, *cited in* Melstrom et al. 2018)[12]. In subjects with low baseline cotinine, secondhand exposure was associated with subsequently increased values, but similar increases were not noted in subjects with higher baseline values. Median cotinine levels were 0.007 ng/mL in serum, 0.033 ng/mL in saliva, and 0.316 ng/mg creatinine in urine (disposable E(N)NDS session) and 0.041 ng/mL in serum, 0.060 ng/mL in saliva, and 0.948 ng/mg creatinine in urine (tank-style E(N)NDS session). The authors concluded that nonusers can systemically absorb nicotine following acute exposure to secondhand E(N)NDS aerosol.

37. Martínez-Sánchez et al. (2015) measured urinary levels of the nicotine-derived nitrosamine ketone (NNK) metabolite, 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol (NNAL) in pairs of subjects comprising 1 E(N)NDS user and 1 nonuser (no tobacco product use) in 6 different households. Details of ventilation in the households were not reported. The E(N)NDS products used contained nicotine in the range of 6–24 mg/mL. NNAL levels were also measured in urine samples from nonusers (no tobacco products) living in households where CC smoking took place (n = 25) and in control homes where tobacco products were not used (n = 24). Overall, 29% (control households), 67% (E(N)NDS-use households), and 76% (CC-use households) of samples from non-users in each type of household had quantifiable levels of NNAL. Median NNAL levels in nonusers (adjusted for urinary creatinine) were 0.33 pg/mL (control households), 0.55 pg/mL (E(N)NDS-use households), and 0.46 pg/mL (CC-use households). From an evaluation of NNAL concentrations in urine of individual E(N)NDS users (range 0.37–9.4 pg/mL) and nonusers (range, < 0.44–3.0 pg/mL) in each of the 6 E(N)NDS-use households, the authors calculated a Spearman's correlation of 0.943 (p = 0.005) for levels in users with nonusers.

[12] In NHANES (2007-2008), 63.3% of the nonsmoking US population ≥20 years old had serum cotinine values below 0.05 ng/mL, while in the study of Melstrom et al. (2018), 33% of the nonusers (two/six) had levels below 0.05 ng/mL in each exposure. A median serum cotinine of 0.034 ng/ml (95% confidence interval, 0.024–0.038) in nonsmokers ≥20 years of age was calculated using 2001–2002 NHANES data (Pirkle et al. 2006, *cited in* Melstrom et al. 2018). Baseline median serum cotinine levels for the study of Melstrom et al. (2018) were 0.089 ng/mL for session one and 0.052 ng/mL for session two. One nonuser's serum cotinine level in the second session was 2.94 ng/mL, similar to a smoker baseline level of > 3 ng/ml, and was omitted from analyses. Authors noted that it is likely that some nonusers had some exposure to secondhand sources of nicotine, despite their self-reported negative histories, and that this highlights a challenge for similar future studies.

Summary of data by type of chemical or species analysed

38. A total of 25 publications (to 05/03/2019) were reviewed that evaluated potential bystander exposure to chemicals and/or particulates as a result of use of E(N)NDS products by human subjects. These included analyses of constituents in exhaled breath, in a chamber or a room under defined test conditions of E(N)NDS use, and in real-life situations where E(N)NDS use took place. An overview of the study settings and evaluations is given in Table 4, below.

Table 4. Studies that have reported evaluation of potential for bystander exposure associated with use of E(N)NDS products. (Studies in italics reported a conflict of interest in terms of funding of the study)

	Exhaled breath of user	**Ambient air**		**Biomarkers of exposure in bystanders**
		Test chamber or room	Household, car, or public building	
Particulate matter		Schripp et al. (2013) Czogala et al. (2014)* Ruprecht et al. (2014) Saffari et al. (2014) Schober et al. (2014) *Maloney et al. (2016)* Melstrom et al. (2017) Protano et al. (2017) Protano et al. (2018)	Fernandez et al. (2015) Chen et al. (2017) Soule et al. (2017) Schober et al. (2019)	
PG and/or glycerine	*Long (2014)* Marco and Grimalt (2015) Oldham et al. (2017)	Schober et al. (2014) *Maloney et al. (2016)* *O'Connell et al. (2015)* *Liu et al. (2017)*	Johnson et al. (2018) Schober et al. (2019)	
Nicotine and nicotine-related compounds	*Long (2014)* Marco and Grimalt (2015) Oldham et al. (2017)	Czogala (2014)* Saffari et al. (2014) Schober et al. (2014) *O'Connell et al. (2015)* *Maloney et al. (2016)* *Liu et al. (2017)* Melstrom et al. (2017)	Ballbe et al. (2014) Bush and Goniewicz (2015) Chen et al. (2017) Johnson et al. (2018) Khachatooria et al. (2018) Schober et al. (2019)	Flouris et al. (2013) Ballbe et al. (2014) Martínez-Sánchez et al. (2015) Melstrom et al. (2017)
Carbonyls	*Long (2014)* Oldham et al. (2017)	Schober et al. (2014) *O'Connell et al. (2015)* *Maloney et al. (2016)* *Liu et al. (2017)*	Johnson et al. (2018)	
PAHs		Schober et al. (2014) *O'Connell (2015)*		
Metals		Saffari (2014) Schober (2014) *O'Connell (2015)* *Liu ((2017)*		
Others	*Long (2014)*	Schripp et al. (2013) Czogala et al. (2014)* Saffari et al. (2014) Schober et al. (2014) *O'Connell et al. (2015)* *Liu et al. (2017)*	Chen et al. (2017)	Flouris et al. (2012) Flouris et al. (2013)

* Authors reported: "MLG received research funding from Pfizer, manufacturer of stop smoking medication, and was funded by the UK Centre for Tobacco Control Studies (UKCTCS) during the study. AS received research funds and travel expenses from Chic Group Ltd., manufacturer of electronic cigarettes in Poland. Other authors declare no conflicts of interest". This study was funded by the Ministry of Science and Higher Education of Poland.

39. Study data are difficult to compare due to the lack of standardisation and the wide range of test conditions and methodologies used. These include variations in the products tested (types of E(N)NDS devices and E(N)NDS liquids), usage parameters (type of user, puffing parameters, numbers of puffs taken during the test period), test environment (location, size, ventilation, background/pre-test sampling), number of users present, sampling methodologies (equipment, numbers of samples taken/duration of sampling, 'washout' between tests, LOD/LOQs of the methods used, controls), constituents sampled (particulate and/or gas phase, specific chemicals/species or groups of chemicals/species), and statistical analyses and reporting of data.

40. Taking into account the limitations of the available literature, as far as is possible from the data set identified, levels of chemicals and/or particulate matter that have been reported in ambient air following use (inhalation/exhalation) of E(N)NDS products are summarised below[13].

Particulate matter

PM_1

41. Ruprecht et al. (2014) reported that PM_1 levels in a 50 m^3 office (air exchange rate, 0.80–0.85/h) where 1 person used E(N)NDS at a rate of 1 puff/min for 7 min, then a 3-min break, for 3 h, were not substantially different from those measured in outdoor air. Mean increases in PM_1 concentration over outside air were 3.5 µg/m^3 during use of E(N)NDS without nicotine and 0.0 µg/m^3 during use of E(N)NDS containing 16 mg/mL nicotine.

42. Protano et al. (2018) reported levels of PM_1 emissions during use of 1st, 2nd, and 3rd generation E(N)NDS products (1 user; 12 x 30-s puffs during 5.5 min in a 52.7 m^3 room; air exchange rate not reported), with or without nicotine, and under various different operating conditions (a total of 8 test conditions, 2x 1st generation, 2x 2nd generation, 4x 3rd generation). The following ranges were reported for PM_1 levels. *Before E(N)NDS use:*18–44 µg/m^3 (arithmetic mean), 18–38 µg/m^3 (median). *During E(N)NDS use:* 54–3429 µg/m^3 (arithmetic mean),17–648 µg/m^3 (median). The highest levels were measured during use of a 2nd generation product containing nicotine.

43. In the same study as described in paragraph 42 above, Protano et al. (2018) reported PM_1 emissions during use of 4th generation E(N)NDS products (1 user; 12 x 30-s puffs during 5.5 min in a 52.7 m^3 room; air exchange rate not reported), with or without nicotine, and under various different operating conditions (a total of 12 test conditions, including devices used on low resistance/high power settings, a practice known as 'sub-ohming'). Emission levels varied widely, with PM_1 ranges as follows: *Before E(N)NDS use:* 21–45 µg/m^3 (arithmetic mean), 20–43 µg/m^3 (median). *During E(N)NDS use:* 75–14,887 µg/m^3 (arithmetic mean), 39–3475 µg/m^3 (median). The

[13] Study data underlying these summaries can be found in the earlier narrative of this discussion paper and/or in Table A attached at Annex B.

highest levels were noted during use of a product with liquid containing 9 mg/mL nicotine under 'sub-ohming' conditions (0.4 Ω/80 W). Pre-test PM_1 concentrations in the room during this latter test-set were 41.66 µg/m^3 (mean) and 39.00 µg/m^3 (median).

Highest reported average PM_1 concentration:

1st, 2nd, 3rd generation E(N)NDS products. **3420 µg/m^3** (arithmetic mean); **630 µg/m^3** (median) during use of a 2nd generation E(N)NDS product containing nicotine for 12 x 30-s puffs over 5.5 min in a 52.7 m^3 room (air exchange rate not reported) (Protano et al. 2018).

4th generation E(N)NDS product. **14,845 µg/m^3** (arithmetic mean); **3436 µg/m^3** (median) during use of 4th generation E(N)NDS device containing nicotine, at 0.4 Ω/80 W ('sub-ohming'), by 1 user, for 12 x 30-s puffs over 5.5 min in a 52.7 m^3 room (air exchange rate not reported) (Protano et al. 2018).

$PM_{2.5}$

44. Ruprecht et al. (2014) reported that $PM_{2.5}$ levels in a 50 m^3 office (air exchange rate, 0.80–0.85/h) where 1 person used E(N)NDS at 1 puff/min for 7 mins, then a 3-min break, for 3 h, were not substantially different to those in outdoor air. Mean increases in $PM_{2.5}$ concentration over outside air were 7.2 µg/m^3 during use of E(N)NDS without nicotine and 0.5 µg/m^3 during use of E(N)NDS containing 16 mg/mL nicotine.

45. Fernandez et al. (2015) reported a median $PM_{2.5}$ level of 9.88 µg/m^3 in a household during active *ad libitum* use of E(N)NDS containing 18 mg/mL nicotine (42 puffs in 1 h) by 1 user. This measurement was similar to levels measured in 2 non-E(N)NDS-user households (9.36 and 9.53 µg/m^3). Details of room ventilation were not reported.

46. Czogala et al. (2014)[14] reported a mean $PM_{2.5}$ concentration of 151.7 µg/m^3 (range, 63.3–272 µg/m^3) measured during 2 x 5-min *ad libitum* use sessions over 1 h of E(N)NDS containing 16–18 mg/mL nicotine (1 user per session) in a 39 m^3 exposure chamber. The air exchange rate in the chamber during this part of the study was not reported. The mean $PM_{2.5}$ concentration in air at baseline was 32.40 µg/m^3 (range 8.0–80 µg/m^3).

47. Schober et al. (2014) reported a mean $PM_{2.5}$ concentration of 197 µg/m^3 (maximum, 514 µg/m^3) in a 45 m^3 room (range of air exchange rate 0.37-0.74/h) while 3 people used E(N)NDS, with or without 18 mg/mL nicotine, *ad libitum* for 2 h.

[14] Authors reported: "MLG received research funding from Pfizer, manufacturer of stop smoking medication, and was funded by the UK Centre for Tobacco Control Studies (UKCTCS) during the study. AS received research funds and travel expenses from Chic Group Ltd., manufacturer of electronic cigarettes in Poland. Other authors declare no conflicts of interest". The study was funded by the Ministry of Science and Higher Education of Poland.

The $PM_{2.5}$ concentration in the room prior to E(N)NDS use was 6 µg/m^3 (air exchange rate 0.76/h). $PM_{2.5}$ levels were highest during use of E(N)NDS without nicotine.

48. Schober et al. (2019) reported ranges of mean $PM_{2.5}$ concentrations of 4–11 µg/m^3 (control), and 8–490 µg/m^3 (tank-style E(N)NDS with 18 mg/mL nicotine) during use of products in car interiors during an 8.5-km journey (1 user and 1 driver in the front of the car; measurement in the back of the car; front windows open 2 cm or 5 cm; total of 14 tests for E(N)NDS product and 7 for control).

49. Soule et al. (2017) measured $PM_{2.5}$ levels in a 4023 m^3 hotel event-room where a 2-day E(N)NDS event was held. Details of room ventilation were not reported. Median $PM_{2.5}$ concentrations were 1.92–3.20 µg/m^3 the day before the event and 595.31 µg/m^3 during the event.

50. Melstrom et al. (2017) measured $PM_{2.5}$ levels in air prior to and during E(N)NDS use in a 52.6 m^3 room where E(N)NDS containing ≥ 18 mg/mL nicotine were used *ad libitum* by 3 users during 2 h. Mean air exchange rate in the room was approximately 5/h. Mean (SD) levels were 788 (2147) µg/m^3 (disposable E(N)NDS product) and 1454 (2683) µg/m^3 (tank-style product) during E(N)NDS use, and 131 (19) µg/m^3 (disposable) and 18 (15) µg/m^3 (tank-style) before use. Median (range) values were 35 (2–19,961) µg/m^3 (disposable E(N)NDS product) and 515 (7–19,972) µg/m^3 (tank-style product) during E(N)NDS use, and 10 (3–761) µg/m^3 (disposable) and 15 (6–827) µg/m^3 (tank-style) before use.

Highest reported average $PM_{2.5}$ concentration:

1436 µg/m^3 (mean); **500 µg/m^3** (median) during *ad libitum* use of tank-style E(N)NDS product containing ≥ 18 mg/mL nicotine by 3 users for 2 h in a 52.6 m^3 room (air exchange rate, approximately 5/h) (Melstrom et al. 2017).

PM_7 and PM_{10}

51. Ruprecht et al. (2014) reported that PM_7 and PM_{10} levels in a 50 m^3 office (air exchange rate, 0.80–0.85/h) where 1 person used E(N)NDS, with or without 16 mg/mL nicotine, at a rate of 1 puff/min for 7 min, then a 3-min break, for 3 h, were not substantially different to those in outdoor air. Mean increases in levels over outdoor air were 8.7 µg/m^3 (PM_7) and 9.9 µg/m^3 (PM_{10}) for E(N)NDS without nicotine, and -0.3 µg/m^3 (PM_7) and -0.6 µg/m^3 (PM_{10}) µg/m^3 for E(N)NDS containing nicotine.

52. Chen et al. (2017) reported mean PM_{10} concentrations in the range of 8429 µg/m^3 and 8850 µg/m^3 (for each of 2 sampling monitors) measured by time-integrated monitoring (342 min indoor plus 58 min outdoor) of ambient air during a vaping event at a 13,475 m^3 indoor venue. Air exchange rates were not reported, but the authors were of the opinion that the venue was not well ventilated. From their measurements, the authors estimated a 24-h TWA for PM_{10} of 1800 µg/m^3.

Highest reported average PM_{10} concentrations:

Office room. **9.9 µg/m³** higher than level in outdoor air in a 50 m^3 office room (air exchange rate, 0.80–0.85/h) where 1 person used E(N)NDS without nicotine at a rate of 1 puff/min for 7 min, then a 3-min break, for 3 h (Ruprecht et al. 2014).

Vaping event. **8640 µg/m³** (mean) in a 13,475 m^3 venue during a vaping event (Chen et al. 2017).

Propylene glycol and glycerine

53. During use of E(N)NDS containing 1.5–2.5% nicotine by 2–12 users for 6 periods of 1 h (total, 36–216 puffs per 1-h session) in a 137 m^3 room (air exchange rate 1.47–1.56/h), Maloney et al. (2016)[15] noted that levels of PG and glycerol were below the measurable LOQ of the techniques used (45–65 µg/m^3 for PG, 53–77 µg/m^3 for glycerol).

54. Schober et al. (2014) reported mean PG and glycerine concentrations of 199.2 µg/m^3 (maximum, 395 µg/m^3) and 72.7 µg/m^3 (maximum, 81 µg/m^3), respectively in a 45 m^3 room (range of air exchange rate 0.37-0.74/h) while 3 people used E(N)NDS with or without 18 mg/mL nicotine, *ad libitum*, for 2 h. The pre-test levels of PG and glycerine in the room air were below the LOD for detection of individual VOCs in the study (< 0.04 µg/m^3).

55. An increase in levels of PG, but not glycerol, was detected in a 38.5 m^3 office meeting room (average air exchange rate, 0.8/h) during a 165-min session of *ad libitum* use by 3 people of E(N)NDS containing 16 mg/g nicotine. Average levels of PG were < 0.5 µg/m^3 at background and in the occupied room before E(N)NDS use, 203.6 µg/m^3 during E(N)NDS use, and 10 µg/m^3 in the empty room after E(N)NDS use. Glycerol was not detected, but the LOD for the detection method was high (150–350 µg/m^3). The LOD for PG was not stated (O'Connell et al. 2015)[16].

56. The median measured air PG concentration across 4 vaping events (indoors) in the USA during 2016–2017 was 305 µg/m^3 (range, < LOD–490 µg/m^3), while PG was not detected during non-E(N)NDS events or when the venues were empty). Air exchange rates were not reported, but the authors considered that the venues were all well ventilated, with doors open during events. The LOD for PG was not specified in the report, but the 'reporting limit' was noted as 20 µg/sample (Johnson et al. 2018).

[15] The 'Funding' acknowledgement section of this publication notes that "All authors of this study are current or retired employees of Altria Client Services which is a subsidiary of Altria Group. NuMark, a subsidiary of Altria Group, is a manufacturer of electronic cigarettes. Funding for this project was provided by Altria Client Services."
[16] The 'Conflicts of Interest' listing of this publication states that "All authors are employees of Imperial Tobacco Group. The work in this manuscript was supported by Imperial Tobacco Group. Imperial Tobacco Group is the parent company of Fontem Ventures B.V., the manufacturer of the e-cigarette products used in this study."

57. Liu et al. (2017)[17] carried out tests under controlled and *ad libitum* use conditions of 3 different E(N)NDS products, all containing nicotine, in a 114 m^3 exposure chamber (fresh air was supplied to the room at a rate of 7.5 L/s). The highest reported mean increase in air PG and glycerol concentrations over baseline were 317 µg/m^3 and 242 µg/m^3, respectively, when using a tank-style E(N)NDS product[18] *ad libitum* (a group of 9 users present in the chamber at the same time took a combined total of 1649 puffs over a 4-h period). LOQs were 3.63 µg/m^3 (PG) and 4.11 µg/m^3 (glycerol).

58. Schober et al. (2019) measured a range of mean PG concentrations from < LOD (value not specified) to 762 µg/m^3 during continuous use of an E(N)NDS product containing nicotine in car interiors during an 8.5-km journey (1 user and 1 driver in the front of the car; measurement in the back of the car; front windows open 2 cm or 5 cm; total of 14 tests for E(N)NDS product and 7 for control). PG levels were < LOD during control tests.

Highest reported average concentrations:

Room/exposure chamber setting:

PG. Mean increase in air PG concentration over baseline of **317 µg/m^3** during *ad libitum* use of a tank-style E(N)NDS product by a group of 9 users present in a 114 m^3 exposure chamber (air supplied at 7.5 L/s) (combined total of 1649 puffs over a 4-h period) (Liu et al. 2017).

Glycerine. Mean increase in air glycerol concentration over baseline of **242 µg/m^3** during *ad libitum* use of a tank-style E(N)NDS product by a group of 9 users present in a 114 m^3 exposure chamber (air supplied at 7.5 L/s) (combined total of 1649 puffs over a 4-h period) (Liu et al. 2017).

Other setting:

PG. Maximum mean concentration of **762 µg/m^3** (with control < LOD) measured during continuous use of an E(N)NDS product containing nicotine in car interiors during an 8.5-km journey (1 user and 1 driver in the front of the car; measurement in the back of the car; front windows open 2 cm or 5 cm; total of 14 tests carried out during use of E(N)NDS product and 7 tests under control condition (no product use).

Nicotine

59. Nicotine was not detected (< LOD of 7 µg/m^3) in a 38.5 m^3 office meeting room (average air exchange rate, 0.8/h), before, during, and after a 165-min session

[17] The 'Conflicts of Interest' section of the publication states that "The study was funded by Altria Client Services LLC. The authors, Mohamadi Sarkar, Jianmin Liu, Qiwei Liang, Michael J. Oldham, Ali A. Rostami and Karl A. Wagner are employees of ALCS. I. Gene Gillman, Piyush Patel and Rebecca Savioz are paid contractors. The study was conducted on behalf of NuMark LLC., (Richmond, VA, USA) a subsidiary of Altria Group, that produces and markets e-vapor products."

[18] 5.92–23.9 mg/mL nicotine, 362–688 mg/mL PG, 212–700 mg/mL glycerol

of *ad libitum* use by 3 people of E(N)NDS containing 16 mg/g nicotine (O'Connell et al. 2015)[19].

60. During use of E(N)NDS containing 1.5–2.5% nicotine by 2–12 users for 6 periods of 1 h (total, 36–216 puffs per 1-h session) in a 137 m^3 room (air exchange rate, 1.47–1.56/h), Maloney et al. (2016)[20] noted that nicotine levels were below the measurable LOQ of the techniques used (10–15 $\mu g/m^3$).

61. Saffari et al. (2014) reported average nicotine levels of 0.061 $\mu g/m^3$ (E(N)NDS without nicotine) and 0.123 $\mu g/m^3$ (E(N)NDS with nicotine) during a time when 1 volunteer used E(N)NDS in a 48 m^3 room (air exchange rate, 1.1/h) at rate of 1 puff/min for periods of 7 min, with 3-min intervals. The authors considered that for the product without nicotine the observed (small) nicotine levels could be due to particle-bound nicotine resuspension in the room during sampling (i.e. 'background' or 'blank' emission level).

62. Ballbe et al. (2014) reported an average nicotine level of 0.13 $\mu g/m^3$ (GM) or 0.11 $\mu g/m^3$ (median) in ambient air of the main family room in 5 households where E(N)NDS use took place. Average levels in 24 control households with no E(N)NDS use were 0.02 $\mu g/m^3$ (GM) and 0.01 $\mu g/m^3$ (median). Details of ventilation in the households were not reported. The reported LOQ for nicotine was 5 ng/filter (suspended from the ceiling for 1 week), equivalent to 0.02 $\mu g/m^3$ air concentration over 1 week.

63. Melstrom et al. (2017) measured mean (range) nicotine levels of 0.717 (0.445–0.989) $\mu g/m^3$ (disposable product) and 1.680 (1.158–2.047) $\mu g/m^3$ (tank-style product) in a 52.6 m^3 room (air exchange rate, approximately 5/h) where E(N)NDS containing nicotine was used *ad libitum* by 3 users during 2 h. Background mean nicotine level in the room air prior to E(N)NDS use were 0.004 (0.003–0.005) $\mu g/m^3$ (disposable) and 0.011 (0.009–0.014) $\mu g/m^3$ (tank). The LOQ was reported as 0.2 ng/air tube.

64. The median air nicotine concentration across 4 vaping events (indoors) in the USA during 2016–2017 was 1.1 $\mu g/m^3$ (range, 0.36–2.2 $\mu g/m^3$), while nicotine was not detected during non-E(N)NDS events or when the venues were empty). Air exchange rates were not reported, but the authors considered that the venues were all well ventilated, with doors open during events. The 'reporting limit' for nicotine was 0.1 µg/sample (Johnson et al. 2018).

65. Schober et al. (2014) reported a mean nicotine concentration of 2.2 $\mu g/m^3$ (maximum, 4.6 $\mu g/m^3$) in a 45 m^3 room (range of air exchange rate 0.37-0.74/h) while 3 people used E(N)NDS with or without 18 mg/mL nicotine, *ad libitum*, for 2 h.

[19] The 'Conflicts of Interest' listing of this publication states that "All authors are employees of Imperial Tobacco Group. The work in this manuscript was supported by Imperial Tobacco Group. Imperial Tobacco Group is the parent company of Fontem Ventures B.V., the manufacturer of the e-cigarette products used in this study."
[20] The 'Funding' acknowledgement section of this publication notes that "All authors of this study are current or retired employees of Altria Client Services which is a subsidiary of Altria Group. NuMark, a subsidiary of Altria Group, is a manufacturer of electronic cigarettes. Funding for this project was provided by Altria Client Services."

The pre-test level of nicotine in the room air was below the LOD for detection of individual VOCs in the study (< 0.04 µg/m^3).

66. Liu et al. (2017)[21] carried out tests under controlled and *ad libitum* use conditions of 3 different E(N)NDS products, all containing nicotine, in a 114 m^3 exposure chamber (fresh air was supplied to the chamber at a rate of 7.5 L/s). The highest reported mean increase in air nicotine concentration over baseline was 2.83 µg/m^3, when using a cartridge-based E(N)NDS product containing 2.4% nicotine, under a pre-specified protocol (8 users present in the chamber at the same time each took 80 puffs over a 4-h period). The LOQ for nicotine was 0.25 µg/m^3.

67. In the study of Czogala et al. (2014)[22], during 2 x 5-min *ad libitum* use, with 30-min interval, of E(N)NDS containing 16–18 mg/mL nicotine by 1 user in a 39 m^3 exposure chamber (air exchange rate not reported in this part of the study), the 1-h mean nicotine level was 3.32 ± 2.49 µg/m^3 (average of 5 tests, each with a different user; range of 1-h means over the 5 users, 0.65–6.23 µg/m^3), with baseline level below the LOD (0.22 µg/m^3).

68. Schober et al. (2019) reported mean nicotine concentrations in the range of 4–10 µg/m^3 for readings taken during 6 of a total of 14 test sessions in cars during use of E(N)NDS containing 18 mg/mL nicotine (1 user and 1 driver in the front of the car; measurement in the back of the car; front windows open 2 cm or 5 cm; total of 14 tests for E(N)NDS product and 7 for control). Nicotine levels during the other 8 test sessions, and during control sessions where no product was used, were all < LOD (value not specified).

69. Chen et al. (2017) measured nicotine concentrations at a vaping convention held in a 13,475 m^3 indoor venue in the USA in 2016. Air exchange rates were not reported, but the authors were of the opinion that the venue was not well ventilated. The time-integrated nicotine concentrations (overall measurement from 400 minutes of monitoring, of which 58 minutes outside and 343 min inside the venue) were 109 µg/m^3 and 140 µg/m^3 for each of 2 monitors carried around the venue during the sampling period.

Highest reported average concentration:

Room/exposure-chamber setting. **3.32 µg/m^3** during 2 x 5-min *ad libitum* use of E(N)NDS containing 16–18 mg/mL nicotine by 1 user in a 39 m^3 chamber (air exchange rate not reported in this part of the study) (Czogala et al. 2014).

[21] The 'Conflicts of Interest' section of the publication states that "The study was funded by Altria Client Services LLC. The authors, Mohamadi Sarkar, Jianmin Liu, Qiwei Liang, Michael J. Oldham, Ali A. Rostami and Karl A. Wagner are employees of ALCS. I. Gene Gillman, Piyush Patel and Rebecca Savioz are paid contractors. The study was conducted on behalf of NuMark LLC., (Richmond, VA, USA) a subsidiary of Altria Group, that produces and markets e-vapor products."

[22] Authors reported: "MLG received research funding from Pfizer, manufacturer of stop smoking medication, and was funded by the UK Centre for Tobacco Control Studies (UKCTCS) during the study. AS received research funds and travel expenses from Chic Group Ltd., manufacturer of electronic cigarettes in Poland. Other authors declare no conflicts of interest". The study was funded by the Ministry of Science and Higher Education of Poland.

Car interior. **10 µg/m^3** during continuous use by 1 user of E(N)NDS containing 18 mg/mL nicotine, in a moving car with front windows slightly open (Schober et al. 2019).

Vaping convention. **124.7 µg/m^3** (average of 2 time-integrated measurements) measured during a vaping convention (Chen et al. 2017).

Carbonyls

70. During use of E(N)NDS containing 1.5–2.5% nicotine by 2–12 users for 6 periods of 1 h (total, 36–216 puffs per 1-h session) in a 137 m^3 room (air exchange rate, 1.47–1.56/h), Maloney et al. (2016)[23] noted that levels of formaldehyde did not change substantially from background (5–8 µg/m^3). The LOQ for formaldehyde was 1 µg/m^3.

71. Johnson et al. (2018) reported that formaldehyde and acetaldehyde concentrations during E(N)NDS events were comparable to concentrations present when the venue was empty and during non-E(N)NDS events, while acrolein was not detected. Reported median levels for the E(N)NDS event, crowded non-E(N)NDS event, and empty venue, respectively, were 12.0, 10.5, and 12.5 µg/m^3 for formaldehyde, and 9.7, 15.5, and 3.5 µg/m^3 for acetaldehyde. Reporting limits were 0.1, 0.5, and 2.0 µg/sample for formaldehyde, acetaldehyde, and acrolein, respectively. Air exchange rates were not reported, but the authors considered that the venues were all well ventilated, with doors open during events.

72. Levels of formaldehyde, acetaldehyde, and acrolein in a 38.5 m^3 office meeting room (average air exchange rate 0.8/h) were similar before, during, and after a 165-min session of *ad libitum* use by 3 people of E(N)NDS containing 16 mg/g nicotine. Measured levels before E(N)NDS use in unoccupied room, before E(N)NDS use in occupied room, during E(N)NDS use, and afterwards, respectively, were 32.0, 31.0, 37.6, and 21.0 µg/m^3 for formaldehyde, 9.0, 6.5, 12.4, and 6.0 µg/m^3 for acetaldehyde, and < LOD (2.0 µg/m^3) at all times for acrolein (O'Connell et al. 2015)[24].

73. Schober et al. (2014) reported levels of aldehydes measured in a 45 m^3 room (range of air exchange rate 0.37-0.74/h) while 3 people used E(N)NDS, with or without 18 mg/mL nicotine, *ad libitum*, for 2 h (6 test sessions each of nicotine-free and nicotine-containing products). Levels were not significantly higher during E(N)NDS use than in control air sampled the day before test sessions began. There was an exception, during one session using an E(N)NDS liquid containing nicotine,

[23] The 'Funding' acknowledgement section of this publication notes that "All authors of this study are current or retired employees of Altria Client Services which is a subsidiary of Altria Group. NuMark, a subsidiary of Altria Group, is a manufacturer of electronic cigarettes. Funding for this project was provided by Altria Client Services."

[24] The 'Conflicts of Interest' listing of this publication states that "All authors are employees of Imperial Tobacco Group. The work in this manuscript was supported by Imperial Tobacco Group. Imperial Tobacco Group is the parent company of Fontem Ventures B.V., the manufacturer of the e-cigarette products used in this study."

the level of formaldehyde was 55 µg/m^3 (pre-test background level, 25 µg/m^3). However, the LOD was reported as 30 µg/m^3 aldehyde per air sample.

74. Liu et al. (2017)[25] carried out tests of 3 different E(N)NDS products under controlled (MarkTen product; cartridge-style product) and *ad libitum* (MarkTen; cartridge-style product; tank-style product) use conditions, all containing nicotine, in a 114 m^3 exposure chamber (fresh air was supplied to the room at a rate of 7.5 L/s). Of the 15 measured carbonyls over the 5 different test scenarios, only 2 measurements were significantly higher than baseline; hexaldehyde during *ad libitum* use of 'MarkTen' (mean increase of 2.07 µg/m^3 over baseline), and acetaldehyde during *ad libitum* use of the tank-style product (mean increase of 1.10 µg/m^3 over baseline). In other cases, levels of carbonyls were either not significantly different, or were significantly lower, than baseline.

75. In the study reported by Schripp et al. (2013), a volunteer in an 8 m^3 test chamber (air exchange rate, 0.3/h) took six 3-s puffs of E(N)NDS containing 0 or 18 mg/mL nicotine, at intervals of 60 s. Air sampled in the chamber for 15 minutes (starting at puff 4) had higher levels of formaldehyde (8-16 µg/m^3) and acetaldehyde (2-3 µg/m^3) than air in the chamber prior to E(N)NDS use (< 1 µg/m^3).

Highest reported average concentrations

Studies mostly indicated no significant increases in levels of carbonyls in ambient air in association with use of E(N)NDS products, compared with control samples. One study reported levels of formaldehyde (**8–16 µg/m^3**) and acetaldehyde (**2–3 µg/m^3**) that were increased over background during/immediately after 6 x 3-s puffs by 1 user in an 8 m^3 exposure chamber (air exchange rate, 0.3/h) (Schripp et al. 2013), but these levels were within the range of ambient air background levels reported in other studies.

Metals

76. All metals (US EPA 'Method 29' metals plus aluminium) were below the LOD (1–2 µg/m^3) in a 38.5 m^3 office meeting room (average air exchange rate, 0.8/h), before, during, and after a 165-min session of *ad libitum* use of E(N)NDS containing 16 mg/g nicotine by 3 participants (O'Connell et al. 2015)[26].

77. Liu et al. (2017)[27] carried out tests under controlled and *ad libitum* use conditions of 3 different E(N)NDS products, all containing nicotine, for 4 h in a

[25] The 'Conflicts of Interest' section of the publication states that "The study was funded by Altria Client Services LLC. The authors, Mohamadi Sarkar, Jianmin Liu, Qiwei Liang, Michael J. Oldham, Ali A. Rostami and Karl A. Wagner are employees of ALCS. I. Gene Gillman, Piyush Patel and Rebecca Savioz are paid contractors. The study was conducted on behalf of NuMark LLC., (Richmond, VA, USA) a subsidiary of Altria Group, that produces and markets e-vapor products."

[26] The 'Conflicts of Interest' listing of this publication states that "All authors are employees of Imperial Tobacco Group. The work in this manuscript was supported by Imperial Tobacco Group. Imperial Tobacco Group is the parent company of Fontem Ventures B.V., the manufacturer of the e-cigarette products used in this study."

[27] The 'Conflicts of Interest' section of the publication states that "The study was funded by Altria Client Services LLC. The authors, Mohamadi Sarkar, Jianmin Liu, Qiwei Liang, Michael J. Oldham, Ali A. Rostami and Karl A. Wagner are employees of ALCS. I. Gene Gillman, Piyush Patel and Rebecca Savioz are paid contractors. The

114 m^3 exposure chamber (fresh air supplied to the room at a rate of 7.5 L/s). Of the 4 metals analysed, cadmium and arsenic were below the LOQ (0.12 µg/m^3 for each metal), while no significant increases over baseline were observed for chromium or nickel.

78. Saffari et al. (2014) reported indoor to outdoor air concentration ratios for 25 metals during a period when 1 volunteer used E(N)NDS (with and without nicotine) in a 48 m^3 room (air exchange rate, 1.1/h) at rate of 1 puff/min for periods of 7 min, with 3-min intervals. Ratios were > 1 for boron (13.13), potassium (1.53), nickel (1.75), zinc (1.26), silver (3.39), and lanthanum (1.47), and < 1 for all other metals. However, actual concentrations of metals measured in air were not reported.

79. Schober et al. (2014) reported that there was a wide variation between measurements for levels of individual metals during different test sessions in a 45 m^3 room (range of air exchange rate 0.37-0.74/h) while 3 people used E(N)NDS, with or without 18 mg/mL nicotine, *ad libitum* for 2 h, with values both higher and lower than control air noted.

Highest reported average concentrations

Although some increases in levels of metals were reported, in general the limited available data set did not indicate significant increases in levels of metals in ambient air in association with use of E(N)NDS products by users compared with controls.

Assessment of potential bystander exposure to $PM_{2.5}$, propylene glycol, glycerine, and nicotine associated with use of E(N)NDS products indoors

80. Overall, the chemicals/species which often showed increased concentrations in ambient indoor air in association with E(N)NDS use were particulate matter, PG, glycerol, and nicotine. Data on ranges of levels reported are summarised below, along with a summary of data on regulations and guideline values, where available.

$PM_{2.5}$

Bystander exposure

81. In studies where $PM_{2.5}$ levels were measured both at baseline and during E(N)NDS use, under pre-specified conditions in a room or exposure chamber, mean levels of increase over baseline ranged from:

- 151.7 µg/m^3 (1 user, 2 x 5-min periods of *ad libitum* use over 1 h, 39 m^3 chamber, air exchange rate not reported in this part of the study) (Czogala et al. 2014), to

study was conducted on behalf of NuMark LLC., (Richmond, VA, USA) a subsidiary of Altria Group, that produces and markets e-vapor products."

- **1436 µg/m^3** (3 users, *ad libitum* use during 2 h, 52.6 m^3 room, air exchange rate approximately 5/h) (Melstrom et al. 2017).

Regulations and guideline values

82. The WHO AQG levels for $PM_{2.5}$ are 25 µg/m^3 (24-h mean) and 10 µg/m^3 (annual mean) (WHO 2010). The basis of the AQG is 'the lowest levels at which total, cardiopulmonary and lung cancer mortality have been shown to increase with more than 95% confidence in response to long-term exposure to $PM_{2.5}$.

Propylene glycol

Bystander exposure

83. In studies where PG levels were measured at baseline and during E(N)NDS use, under pre-specified conditions in a room or exposure chamber, mean levels of increase over baseline ranged from:

- 0.199 mg/m^3 (3 users, *ad libitum* use for 2 h, 45 m^3 room, range of air exchange rate 0.37-0.74/h) (Schober et al. 2014), to
- **0.317 mg/m^3** (9 users (total 1649 puffs) over 4 h,114 m^3 exposure chamber, fresh air supplied at 7.5 L/s) (Liu et al. 2017).

Regulations and guideline values

84. From discussions at the July 2018 COT meeting (TOX/2018/23), the COT established a health-based guidance value (HBGV) for continuous exposure to PG of 2.9 mg/m^3. This was based on a lowest observed adverse effect level (LOAEL) of 160 mg/m^3 for nasal haemorrhaging from the study of Suber et al. (1989), with adjustment for continuous exposure (x5.6) and using an uncertainty factor (UF) of 10 for inter-individual variation.

85. Other agencies have established HBGVs for PG based on the study of Suber et al. (1989) (see TOX/2018/23 for more details), as summarised in the following paragraphs.

86. The Dutch Expert Committee on Occupational Standards established an 8-h TWA (vapour + aerosol) of 50 mg/m^3 based on a no observed adverse effect level (NOAEL) of 160 mg/m^3 for increased numbers of goblet cells. The Committee also recommended that health-based occupational exposure limits for inhalable and respirable dust should be applied to aerosols of PG (HCN 2007).

87. The German Committee on Indoor Guide Values recommended a health precaution guide value (RW I, guideline value I[28]) of 0.06 mg/m^3 for PG, based on a

[28] RW I represents the concentration of a substance in indoor air for which, when considered individually, there is no evidence that life-long exposure would have an adverse health impact. RW II represents the concentration of a substance that, if reached or exceeded, requires immediate action as this concentration could pose a health hazard. It may be defined as a short-term value (RW II K) or a long-term value (RW II L). For more information,

health hazard guide value (RW II, guideline value II) of 0.6 mg/m^3, derived using a LOAEL of 160 mg/m^3 for nasal haemorrhage (Umweltbundesamtes 2017).

88. The US Agency for Toxic Substances and Disease Registry established an intermediate-duration MRL for PG of 0.009 ppm [0.028 mg/m^3], based on a LOAEL of 51 ppm [160 mg/m^3] for nasal haemorrhaging (ATSDR 1997).

Glycerol

Bystander exposure

89. In studies where glycerol levels were measured at baseline and during E(N)NDS use under pre-specified conditions in a room or exposure chamber, mean levels of increase over baseline ranged from:

- 0.073 mg/m^3 (3 users, *ad libitum* use for 2 h, 45 m^3 room, range of air exchange rate 0.37-0.74/h) (Schober et al. 2014), to
- **0.242 mg/m^3** (9 users (total 1649 puffs) over 4 h,114 m^3 exposure chamber, fresh air supplied at 7.5 L/s) (Liu et al. 2017).

Regulations and guideline values

90. From discussions at the July 2018 COT meeting (TOX/2018/23), the COT established an HBGV for continuous exposure to glycerol of 11.8 mg/m^3. This was based on a PoD of 662 mg/m^3 (NOAEL) from the rat inhalation study of Renne et al. (1992), with an adjustment for continuous exposure (x5.6) and using an UF of 10 for inter-individual variation.

91. One other agency has established an HBGV for PG based on the study of Renne et al. (1992) (see TOX/2018/23 for more details). DFG in Germany set a maximum workplace concentration (MAK value) of 200 mg/m^3, based on a NOAEL of 662 mg/m^3 (Hartwig A 2017).

Nicotine

Bystander exposure

92. In a study where nicotine levels were measured during 4 vaping events, which took place in convention centres that were reported to be well ventilated, the median nicotine level was 1.1 µg/m^3 (range < 0.36–2.2 µg/m^3) (Johnson et al. 2018). In another study conducted during an indoor vaping event that the authors considered to be poorly ventilated, ambient air nicotine levels of 124.7 µg/m^3 were reported (Chen et al. 2017).

---

see: https://www.umweltbundesamt.de/en/topics/health/commissions-working-groups/german-committee-on-indoor-guide-values#textpart-3 (accessed 06/04/18).

93. One study measured nicotine in ambient air of the main family room of 5 households where E(N)NDS were used. The GM concentration was 0.13 µg/m^3 (Ballbe et al. 2014).

94. One study measured nicotine levels in cars (moving, with front windows slightly open) during continuous use by 1 passenger of an E(N)NDS product containing 18 mg/mL nicotine. For a total of 14 test sessions, nicotine levels were < LOD (value not specified) during 8 tests, and in the range of 4–10 µg/m^3 during the other 6 tests (Schober et al. 2019).

95. A total of 7 studies measured nicotine levels in ambient air in association with E(N)NDS use under pre-specified conditions, in rooms or exposure chambers. In 2 of these studies nicotine was not detected, but these studies used detection methods with relatively high LOD/LOQ values (O'Connell et al. 2015, nicotine < LOD of 7 µg/m^3; Maloney et al., nicotine < LOQ of 10–15 µg/m^3). The remaining 5 studies reported mean ambient air nicotine levels as follows:

- 0.123 ± 34.5 µg/m^3 (1 user, 1 puff/7min, 48 m^3 room, air exchange rate 1.1/h (Saffari et al. 2014)
- 0.717 ± 0.195 µg/m^3 (range, 0.445–0.989 µg/m^3) (3 users, *ad libitum* use for 2 h, 52.6 m^3 room, air exchange rate 5/h) (Melstrom et al. 2017)
- 2.2 ±1.7 µg/m^3 (maximum, 4.6 µg/m^3) (3 users, *ad libitum* use for 2 h, 45 m^3 room, range of air exchange rate 0.37-0.74/h) (Schober et al. 2014)
- 2.83 ± 0.44 µg/m^3 (8 users, 80 puffs each over 4 h, 114 m^3 room, air supplied at 7.5 L/s) (Liu et al. 2017)
- **3.32 ± 2.49 µg/m^3** (range, 0.65–6.23 µg/m^3) (1 user, 2 x 5-min periods of *ad libitum* use over 1 h, 39 m^3 chamber, air exchange rate not reported in this part of the study) (Czogala et al. 2014).

96. Taking the mean value of 3.32 µg/m^3 (0.00332 mg/m^3) from the study of Czogala et al. (2014), assuming that a 70 kg individual inhales 20 m^3 air during 24 h, this would lead to a nicotine intake of 66.4 µg/day = **0.00095 mg/kg bw/day**. Assuming a 13.3 kg, 1–6 year old child inhales 8.8 m^3 air during 24 h, this would lead to a nicotine intake of **0.0022 mg/kg bw/day**.

Ambient air levels of nicotine associated with secondhand CC smoke

97. In a 2006 review 'The Health Consequences of Involuntary Exposure to Tobacco Smoke' published by the US Surgeon General, Chapter 4 reviewed 'Prevalence of Exposure to Secondhand Smoke', with a focus on measured concentrations of airborne nicotine (CDC 2006). This publication summarised data from numerous studies that had measured air nicotine levels in different settings

where CC smoking was permitted, restricted, or banned, including homes, restaurants and bars, offices and other workplaces. Detailed information can be found in the report, at https://www.ncbi.nlm.nih.gov/books/NBK44325/ (accessed 04/03/2019). As a brief summary, in homes where CC smoking occurred, average nicotine levels were often in the range of 1–3 µg/m^3, with higher ranges measured during active smoking (e.g. 5–15 µg/m^3). Workplace studies showed a wide range of nicotine concentrations, with mean levels often in the range of 1–10 µg/m^3 but ranging up to around 50 µg/m^3 where smoking was allowed, and levels generally less < 1 µg/m^3 where smoking was banned. In public places such as restaurants, bars, lounges, and other venues, nicotine levels ranged from less than detectable up to around 70 µg/m^3. A study of waiters exposed to secondhand smoke showed average nicotine levels of 5.8 µg/m^3, with an upper range of 68 µg/m^3, while a study in a cafeteria showed nicotine concentrations of 25-40 µg/m^3 in a smoking section, 2–5 µg/m^3 in a proximal non-smoking section, and < 0.5 µg/m^3 in a more-distant non-smoking section. Nicotine levels in bars and lounges were generally > 10 µg/m^3 and often > 50 µg/m^3. A study conducted in prisons indicated average nicotine levels of 3–10 µg/m^3 in living and sleeping areas. A figure summarising the data presented in Chapter 4 of the report is reproduced in Figure 1.

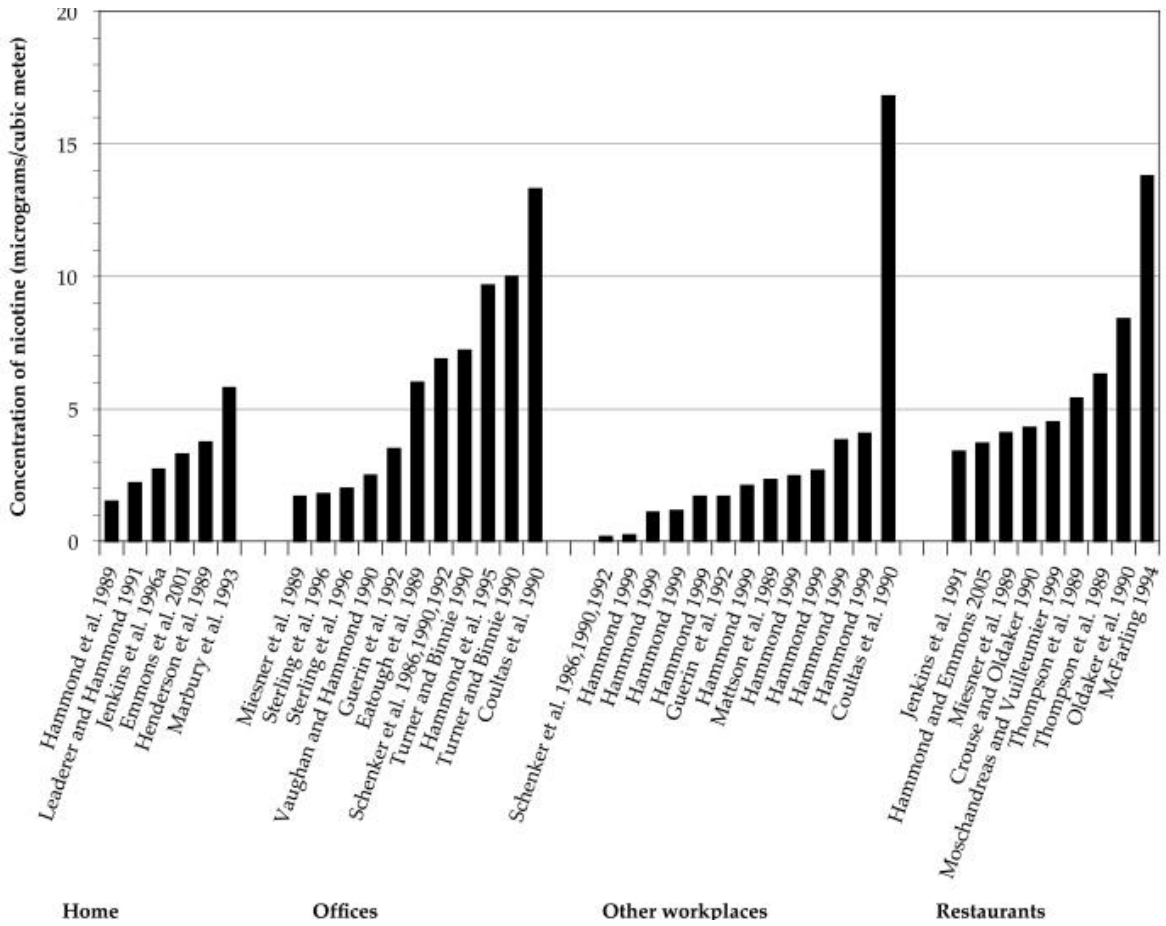

Figure 1. Average concentrations of nicotine in homes, offices, other workplaces, and restaurants where smoking is permitted. (Reproduced from: The Health Consequences of Involuntary Exposure to Tobacco Smoke: A Report of the Surgeon General. Office on Smoking and Health (US). Atlanta (GA): Centers for Disease Control and Prevention (US), 2006. Chapter 4. Prevalence of Exposure to Secondhand Smoke, Figure 4.6).

Regulations and guideline values

98. The UK WEL for nicotine is 0.5 mg/m^3 8-h TWA, with a 15-min STEL of 1.5 mg/m^3 (HSE 2018). Workplace exposure limits in many other EU countries are also 0.5 mg/m^3 8-h TWA, except for Sweden (0.1 mg/m^3 8-h TWA)[29]. The National Institute for Occupational Safety and Health (NIOSH) recommended exposure level (REL) and Occupational Safety and Health Administration (OSHA) recommended permissible exposure limit (PEL) values for nicotine are 0.5 mg/m^3 TWA [skin][30]. The NIOSH IDLH (immediately dangerous to life or health) is 5 mg/m^3, based on a fatal human oral dose estimated as 50 to 60 mg[31].

99. The United States Environmental Protection Agency (US EPA) evaluated occupational risk of short- and intermediate-term use of nicotine as a pesticide (in the format of smoke-generating canisters), by certified applicators, in greenhouses (only) for a reregistration application eligibility decision. A NOAEL of 1.25 mg/kg bw/day was identified for hepatotoxicity in a 10-day rat drinking-water study (Yuen et al. 1995) (EPA 2008). The report by EPA (2008) can be found at https://archive.epa.gov/pesticides/reregistration/web/pdf/nicotine_red.pdf (accessed 01/03/2019) and is reproduced at Annex C. The Agency described using a margin of exposure (MOE) approach. The Agency determined that an MOE of 1000 would be considered to be protective of human health (10x for inter-species extrapolation, 10x for intra-species variability, 10x for database uncertainty). The major potential source of risk for exposure was considered to be by inhalation, with relative smaller exposure dermally.

100. The European Food Safety Authority (EFSA) set an oral acute reference dose ARfD of 0.0008 mg/kg/bw/day for nicotine (EFSA 2009). This value was based on the study of Lindgren et al. (1999) (*cited in* EFSA 2009), in which a dose-response relationship for electroencephalographic parameters and heart-rate frequency over a range of nicotine doses (i.v. infusion) were evaluated in 14 regular CC smokers. From these data, EFSA determined a LOAEL of 0.0035 mg/kg bw/day for pharmacological effects (slight, transient and rapidly reversible increase of the heart rate in humans), using an overall UF of x10 and a correction factor of x0.44 for oral bioavailability of nicotine (extrapolation from i.v to oral route[32]). Given that nicotine has a short biological half-life and does not accumulate in the body, and that the most sensitive effect was considered to be the pharmacological effect on the cardiovascular system, EFSA considered that the value set for the ARfD would be suitable to protect from chronic effects and could also be applied as the acceptable

[29] See http://limitvalue.ifa.dguv.de/, accessed 28/02/2019.
[30] The " [skin] " designation indicates the potential for dermal absorption; skin exposure should be prevented as necessary through the use of good work practices, gloves, coveralls, goggles, and other appropriate equipment.
[31] See https://www.cdc.gov/niosh/idlh/54115.html, accessed 28/02/2019.
[32] For comparison, in a report of 'Metabolism and Disposition Kinetics of Nicotine', Hukkanen et al. (2005) report % bioavailability for nicotine administered as single doses by various routes as follows: Smoking 1 CC (80-90%); i.v. approx 5.1 mg (100%); Nasal spray 1 mg (60-80%); Gum 2-4 mg (55-78%; Inhaler 4 mg (51-56%); Lozenge 2-4 mg (50-79%); Transdermal patch 14-21 mg/24 h (68-100%); s.c. injection 2.4 mg (100%); Oral capsule 3-4 mg (44%); Oral solution approx. 3 mg (20%); Enema approx. 3.5 mg (15-25%). See https://pdfs.semanticscholar.org/9043/b736c593390f4389409f8051c95b75e1de97.pdf (accessed 04/03/2019).

daily intake (ADI). The EFSA (2009) report can be found at https://www.efsa.europa.eu/en/efsajournal/pub/rn-286 (accessed 01/03/2019) and is reproduced at Annex D.

101. The report of EFSA (2009) noted that in 2009 the German Federal Institute for Risk Assessment (BfR) also established an ARfD for nicotine of 0.0008 mg/kg bw/day, based on the study of Lindgren et al. (1999). The PoD was a LOAEL of 0.0035 mg/kg bw/day for increased heart rate, with a safety factor of x10 for intra-species variability, and a correction of x0.44 for oral bioavailability (*data cited in* EFSA 2009).

102. EFSA (2009) also noted that in an assessment of nicotine under the EU peer review process for pesticides, in 2007, a UK Rapporteur proposed an ARfD and ADI of 0.0001 mg/kg bw/day for nicotine. This was based on data from a report by Woolf et al. (1997) (*cited in* EFSA 2009), a post-marketing surveillance study of data collected at US poison centres, including 36 children aged 0–15 y exposed to transdermal nicotine patches (by either dermal or oral route). Clinical signs of toxicity were reported at approximately 0.03–0.8 mg/kg/bw day. The lowest estimated systemic exposures of nicotine associated with adverse effects were reported to be < 0.01 mg/kg bw/day. The value of 0.01 mg/kg bw/day was taken as a lowest observed effect level (LOEL), with application of a UF of 10 for intra-species variability and 10 for use of a limited data set (UK DAR 2007, *data cited in* EFSA 2009).

103. Finally, the report of EFSA (2009) also noted that the Agence Française de Sécurité Sanitaire (AFSSA) had prepared a report providing scientific and technical advice concerning mushroom contamination by nicotine. AFSSA endorsed the ADI and ARfD of 0.0001 mg/kg bw/day proposed by the UK in 2007 (*information cited in* EFSA 2009).

Data on developmental toxicity of nicotine

104. At the December 2018 COT meeting, the Committee considered the discussion paper, TOX/2018/45, which reviewed literature on developmental toxicity associated with nicotine exposure. The report noted that animal studies have demonstrated developmental toxicity following prenatal and/or early postnatal nicotine exposure on several organ systems, notably the neurological and respiratory systems. Studies had used exposure regimes including continuous subcutaneous infusion, drinking water, or bolus injection, but not inhalation. Exposure levels had been evaluated to model systemic nicotine exposures that would be achieved via direct CC smoking or from secondhand smoke exposure. Recent studies had indicated effects in offspring on brain cholinergic and serotonergic signalling systems, and on cognitive and behavioural function, associated with a maternal dose of 0.2 mg/kg bw/day nicotine (by continuous subcutaneous infusion), which was considered to represent a nicotine exposure level equivalent to that from secondhand CC smoke (Slotkin et al. 2015a, Hall et al. 2016, *cited in* TOX/2018/45).

Summary

105. This report has reviewed the literature relating to exposures that individuals may experience as 'bystanders' in indoor spaces where E(N)NDS use is occurring or has taken place.

106. As noted in previous COT discussion papers on E(N)NDS, study data may be difficult to compare due to the lack of standardisation and the wide range of test conditions and methodologies used. These include variations in the parameters and microenvironment of the locations where sampling took place (including room size, ventilation, location of windows/doors, height of ceiling), E(N)NDS products (devices, liquids), usage parameters (including number of users present in the room, rate of product use per user), sampling and detection methodologies (including whether gas phase or particulate matter is sampled), LOD/LOQs of the methods used, controls, and statistical analysis and reporting of data.

107. Data on concentrations of chemicals/species present in ambient air (secondhand) or on surfaces (thirdhand) in association with E(N)NDS use were obtained from the study reports. Where possible, highest reported ambient air levels were noted for 'standard' (e.g. moderate levels of E(N)NDS use in locations such as office rooms or exposure chambers) and 'non-standard' (e.g. within convention centres during dedicated vaping events) situations.

108. The principal chemicals/species that were often reported to show increased levels in ambient air during E(N)NDS use were particulate matter, the major E(N)NDS liquid constituents, PG and glycerol, and nicotine.

Questions for the Committee

109. Members are invited to comment on the information provided in this paper and to consider the following questions:

i. Do Members consider that the data presented indicate any specific chemicals that may be of particular concern in relation to bystander exposure?

ii. Based on the information that has been provided, is there an appropriate value for nicotine to which exposures by inhalation can be compared?

iii. Is the Committee able to draw any conclusions from the data presented on potential health risks associated with exposure of bystanders to ambient air in situations where E(N)NDS use takes place?

iv. Are there any particular aspects of this paper that should be captured when a COT statement on E(N)NDS is prepared?

NCET at WRc/IEH-C under contract supporting the PHE COT Secretariat
March 2019

Abbreviations

ADI	Acceptable daily intake
AFSSA	Agence Française de Sécurité Sanitaire
AQG	Air quality guideline
ARfD	Acute reference dose
CC	Conventional cigarette
CO	Carbon monoxide
CO2	Carbon dioxide
CTD	Classic Tobacco Disposable
EBS	Exhaled breath system
EFSA	European Food Safety Authority
E(N)NDS	Electronic nicotine (or non-nicotine) delivery system
ENDS	Electronic nicotine delivery system
ENNDS	Electronic non-nicotine delivery system
FMPC	Fast mobility particle sizer
FP	Fine particles (< 2.5 µm in diameter)
GC	Gas chromatography
GM	Geometric mean
GSD	Geometric standard deviation
HBGV	Health-based guidance value
IDLH	Immediately dangerous to life or health
IQR	Inter-quartile range
LOAEL	Lowest observed adverse effect level
LOEL	Lowest observed effect level
LOD	Limit of detection
LOQ	Limit of quantitation
MAK	Maximum workplace concentration
MOE	Margin of exposure
MMD	Magnificant Menthol Disposable
MRL	Minimum risk level
NIOSH	National Institute for Occupational Safety and Health
NHANES	US National Health and Nutrition Examination Survey
NOAEL	No observed adverse effect level
NNAL	4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol
NNK	Nicotine-derived nitrosamine ketone
NO	Nitric oxide
NO2	Nitrogen dioxide
OSHA	Occupational Safety and Health Administration
PAH	Polycyclic aromatic hydrocarbon
PEL	Permissible exposure limit
PG	Propylene glycol
PM1	Particulate matter 1 µm or less in diameter
PM2.5	Particulate matter 2.5 µm or less in diameter
PM7	Particulate matter 7 µm or less in diameter
PM10	Particulate matter 10 µm or less in diameter
REL	Recommended exposure level

SMP	Submicronic particle
TSNA	Tobacco-specific nitrosamine
TSP	Total suspended particulates
TVOCs	Total volatile organic compounds
TWA	Time-weighted average
UF	Uncertainty factor
UFP	Ultra fine particles (< 100 nm in diameter)
VG	Vegetable glycerine (glycerol)
VOC	Volatile organic compound
WBC	White blood cell
WEL	Workplace exposure limit

References

ATSDR. 1997. Toxicological Profile for Propylene Glycol. Atlanta, GA.: d. U. S. D. o. H. a. H. S. Agency for Toxic Substances and Disease Registry, Public Health Service.

Ballbe, M., J. M. Martinez-Sanchez, X. Sureda, M. Fu, R. Perez-Ortuno, J. A. Pascual, E. Salto & E. Fernandez (2014) Cigarettes vs. e-cigarettes: Passive exposure at home measured by means of airborne marker and biomarkers. *Environ Res,* 135**,** 76-80.

Bush, D. & M. L. Goniewicz (2015) A pilot study on nicotine residues in houses of electronic cigarette users, tobacco smokers, and non-users of nicotine-containing products. *Int J Drug Policy,* 26**,** 609-11.

CDC (2006) Office on Smoking and Health (US). The Health Consequences of Involuntary Exposure to Tobacco Smoke: A Report of the Surgeon General. Atlanta (GA): Centers for Disease Control and Prevention (US).

Chen, R., A. Aherrera, C. Isichei, P. Olmedo, S. Jarmul, J. E. Cohen, A. Navas-Acien & A. M. Rule (2017) Assessment of indoor air quality at an electronic cigarette (Vaping) convention. *J Expo Sci Environ Epidemiol*.

Czogala, J., M. L. Goniewicz, B. Fidelus, W. Zielinska-Danch, M. J. Travers & A. Sobczak (2014) Secondhand exposure to vapors from electronic cigarettes. *Nicotine Tob Res,* 16**,** 655-62.

EFSA (2009) EFSA Statement. Potential risks for public health due to the presence of nicotine in wild mushrooms. *The EFSA Journal,* RN-286**,** 1-47.

EPA, U. (2008) Reregistration Eligibility Decision for Nicotine. List B. Case No. 2460. March 2008.

Fernandez, E., M. Ballbe, X. Sureda, M. Fu, E. Salto & J. M. Martinez-Sanchez (2015) Particulate Matter from Electronic Cigarettes and Conventional Cigarettes: a Systematic Review and Observational Study. *Curr Environ Health Rep,* 2**,** 423-9.

Flouris, A. D., M. S. Chorti, K. P. Poulianiti, A. Z. Jamurtas, K. Kostikas, M. N. Tzatzarakis, A. Wallace Hayes, A. M. Tsatsakis & Y. Koutedakis (2013) Acute impact of active and passive electronic cigarette smoking on serum cotinine and lung function. *Inhal Toxicol,* 25**,** 91-101.

Flouris, A. D., K. P. Poulianiti, M. S. Chorti, A. Z. Jamurtas, D. Kouretas, E. O. Owolabi, M. N. Tzatzarakis, A. M. Tsatsakis & Y. Koutedakis (2012) Acute effects of electronic and tobacco cigarette smoking on complete blood count. *Food and Chemical Toxicology,* 50**,** 3600-3603.

Hartwig A, M. C. (2017) Glycerin / Propane-1,2,3-triol MAK Value Documentation. In The MAK Collection for Occupational Health and Safety 2017, Vol 2, No 2.

HCN (2007) Propylene glycol (1,2-Propanediol); Health-based recommended occupational exposure limit. The Hague:: Health Council of the Netherlands.

HSE (2018) Health and Safety Executive. Workplace exposure limits. *http://www.hse.gov.uk/coshh/basics/exposurelimits.htm*.

Johnson, J. M., L. P. Naeher, X. Yu, S. L. Rathbun, J. L. Muilenburg & J. S. Wang (2018) Air monitoring at large public electronic cigarette events. *Int J Hyg Environ Health,* 221**,** 541-547.

Khachatoorian, C., P. Jacob Iii, N. L. Benowitz & P. Talbot (2018) Electronic cigarette chemicals transfer from a vape shop to a nearby business in a multiple-tenant retail building. *Tob Control*.

Kuschner, W. G., S. Reddy, N. Mehrotra & H. S. Paintal (2011) Electronic cigarettes and thirdhand tobacco smoke: two emerging health care challenges for the primary care provider. *Int J Gen Med,* 4**,** 115-20.

Lindgren, M., L. Molander, C. Verbaan, E. Lunell & I. Rosen (1999) Electroencephalographic effects of intravenous nicotine--a dose-response study. *Psychopharmacology (Berl),* 145**,** 342-50.

Liu, J., Q. Liang, M. J. Oldham, A. A. Rostami, K. A. Wagner, I. G. Gillman, P. Patel, R. Savioz & M. Sarkar (2017) Determination of Selected Chemical Levels in Room Air and on Surfaces after the Use of Cartridge- and Tank-Based E-Vapor Products or Conventional Cigarettes. *Int J Environ Res Public Health,* 14.

Long, G. A. (2014) Comparison of select analytes in exhaled aerosol from e-cigarettes with exhaled smoke from a conventional cigarette and exhaled breaths. *International Journal of Environmental Research and Public Health,* 11**,** 11177-11191.

Maloney, J. C., M. K. Thompson, M. J. Oldham, C. L. Stiff, P. D. Lilly, G. J. Patskan, K. H. Shafer & M. A. Sarkar (2016) Insights from two industrial hygiene pilot e-cigarette passive vaping studies. *J Occup Environ Hyg,* 13**,** 275-83.

Marco, E. & J. O. Grimalt (2015) A rapid method for the chromatographic analysis of volatile organic compounds in exhaled breath of tobacco cigarette and electronic cigarette smokers. *J Chromatogr A,* 1410**,** 51-9.

Martínez-Sánchez, J. M., M. Fu, J. C. Martín-Sánchez, M. Ballbè, E. Saltó & E. Fernández (2015) Perception of electronic cigarettes in the general population: does their usefulness outweigh their risks? *BMJ open,* 5**,** e009218.

Melstrom, P., B. Koszowski, M. H. Thanner, E. Hoh, B. King, R. Bunnell & T. McAfee (2017) Measuring PM2.5, Ultrafine Particles, Nicotine Air and Wipe Samples Following the Use of Electronic Cigarettes. *Nicotine & Tobacco Research,* 19**,** 1055-1061.

Melstrom, P., C. Sosnoff, B. Koszowski, B. A. King, R. Bunnell, G. Le, L. Wang, M. H. Thanner, B. Kenemer, S. Cox, B. R. DeCastro & T. McAfee (2018) Systemic absorption of nicotine following acute secondhand exposure to electronic cigarette aerosol in a realistic social setting. *Int J Hyg Environ Health,* 221**,** 816-822.

O'connell, G., S. Colard, X. Cahours & J. D. Pritchard (2015) An assessment of indoor air quality before, during and after unrestricted use of E-cigarettes in a small room. *International Journal of Environmental Research and Public Health,* 12**,** 4889-4907.

Protano, C., P. Avino, M. Manigrasso, V. Vivaldi, F. Perna, F. Valeriani & M. Vitali (2018) Environmental electronic vape exposure from four different generations of

electronic cigarettes: Airborne particulate matter levels. *International Journal of Environmental Research and Public Health,* 15.

Protano, C., M. Manigrasso, P. Avino & M. Vitali (2017) Second-hand smoke generated by combustion and electronic smoking devices used in real scenarios: Ultrafine particle pollution and age-related dose assessment. *Environ Int,* 107**,** 190-195.

Renne, R. A., A. P. Wehner, B. J. Greenspan, H. S. Deford, H. A. Ragan, R. B. Westerberg, R. L. Buschbom, G. T. Burger, A. W. Hayes, R. L. Suber & A. T. Mosberg (1992) 2-Week and 13-Week Inhalation Studies of Aerosolized Glycerol in Rats. *Inhalation Toxicology,* 4**,** 95-111.

Ruprecht, A. A., C. De Marco, P. Pozzi, E. Munarini, R. Mazza, G. Angellotti, F. Turla & R. Boffi (2014) Comparison between particulate matter and ultrafine particle emission by electronic and normal cigarettes in real-life conditions. *Tumori,* 100**,** e24-7.

Saffari, A., N. Daher, A. Ruprecht, C. De Marco, P. Pozzi, R. Boffi, S. H. Hamad, M. M. Shafer, J. J. Schauer, D. Westerdahl & C. Sioutas (2014) Particulate metals and organic compounds from electronic and tobacco-containing cigarettes: comparison of emission rates and secondhand exposure. *Environ Sci Process Impacts,* 16**,** 2259-67.

Schober, W., L. Fembacher, A. Frenzen & H. Fromme (2019) Passive exposure to pollutants from conventional cigarettes and new electronic smoking devices (IQOS, e-cigarette) in passenger cars. *Int J Hyg Environ Health*.

Schober, W., K. Szendrei, W. Matzen, H. Osiander-Fuchs, D. Heitmann, T. Schettgen, R. A. Jorres & H. Fromme (2014) Use of electronic cigarettes (e-cigarettes) impairs indoor air quality and increases FeNO levels of e-cigarette consumers. *Int J Hyg Environ Health,* 217**,** 628-37.

Schripp, T., D. Markewitz, E. Uhde & T. Salthammer (2013) Does e-cigarette consumption cause passive vaping? *Indoor Air,* 23**,** 25-31.

Soule, E. K., S. F. Maloney, T. R. Spindle, A. K. Rudy, M. M. Hiler & C. O. Cobb (2017) Electronic cigarette use and indoor air quality in a natural setting. *Tob Control,* 26**,** 109-112.

Suber, R. L., R. Deskin, I. Nikiforov, X. Fouillet & C. R. Coggins (1989) Subchronic nose-only inhalation study of propylene glycol in Sprague-Dawley rats. *Food Chem Toxicol,* 27**,** 573-83.

Umweltbundesamtes, B. d. (2017) Indoor air guide value for propan-1,2-diol (propylene glycol) : Communication from the Committee on Indoor Guide Values. *Bundesgesundheitsblatt, Gesundheitsforschung, Gesundheitsschutz,* 60**,** 1298-1304.

WHO. 2010. WHO Guidelines Approved by the Guidelines Review Committee. In *WHO Guidelines for Indoor Air Quality: Selected Pollutants*. Geneva: World Health Organization

Yuen, S. T., A. R. Gogo, Jr., I. S. Luk, C. H. Cho, J. C. Ho & T. T. Loh (1995) The effect of nicotine and its interaction with carbon tetrachloride in the rat liver. *Pharmacol Toxicol,* 77**,** 225-30.

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

Potential toxicological risks from electronic nicotine (and non-nicotine) delivery systems (E(N)NDS – e-cigarettes). Paper 9: Bystander exposure.

Details of Literature search carried out by NCET at WRc/IEH-C

Searches were carried out on 14/11/2018 to identify published literature relating to 'bystander' exposure to E(N)NDS products, as follows.

PubMed

(((("e-cig*" [Title/Abstract] OR "electronic cigarette*" [Title/Abstract] OR "electronic nicotine delivery system*"[Title/Abstract] OR vape[Title/Abstract] OR vaping[Title/Abstract])) AND (bystander[Title/Abstract] OR passive[Title/Abstract] OR secondhand[Title/Abstract] OR thirdhand[Title/Abstract] OR "second hand"[Title/Abstract] OR "second-hand"[Title/Abstract] OR "third hand"[Title/Abstract] OR "third-hand"[Title/Abstract])) AND english[Language]: 83 citations.

Scopus

(TITLE-ABS-KEY ("e-cig*" OR "electronic cigarette*" OR "electronic nicotine delivery system*" OR vape OR vaping) AND TITLE-ABS-KEY (bystander OR passive OR secondhand OR thirdhand OR "second-hand" OR "third-hand" OR "second hand" OR "third hand")) AND (LIMIT-TO (LANGUAGE , "English")) AND (EXCLUDE (SUBJAREA , "SOCI") OR EXCLUDE (SUBJAREA , "PSYC") OR EXCLUDE (SUBJAREA , "NURS") OR EXCLUDE (SUBJAREA , "AGRI") OR EXCLUDE (SUBJAREA , "ENGI") OR EXCLUDE (SUBJAREA , "CHEM") OR EXCLUDE (SUBJAREA , "HEAL") OR EXCLUDE (SUBJAREA , "MATE") OR EXCLUDE (SUBJAREA , "ARTS") OR EXCLUDE (SUBJAREA , "COMP") OR EXCLUDE (SUBJAREA , "EART") OR EXCLUDE (SUBJAREA , "CENG")) AND (EXCLUDE (LANGUAGE , "French") OR EXCLUDE (LANGUAGE , "German") OR EXCLUDE (LANGUAGE , "Polish") OR EXCLUDE (LANGUAGE , "Spanish")): 270 citations.

The text and reference lists of selected publications were also inspected for any further literature of relevance.

An updated search of PubMed carried out on 05/03/2019 identified 1 additional citation of relevance.

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

Potential toxicological risks from electronic nicotine (and non-nicotine) delivery systems (E(N)NDS – e-cigarettes). Paper 9: Bystander exposure.

Table A. Summary of studies of potential bystander exposure to E(N)NDS emissions.

Table A. Summary of studies of potential bystander exposure to E(N)NDS emissions.

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
Exhaled breath measurements				
Long (2014) Comparison of levels of chemicals in exhaled breath before and after use of E(N)NDS or CC [author employed by Lorillard Tobacco]	Exhaled breath: 6 phenolics 8 carbonyls Water Glycerine Nicotine Ambient air: carbonyls	blu CTD blu eCigs Classic Tobacco Disposable (82% glycerine, 9% water, 2% nicotine, and 7% flavour) blue MMD blu eCigs Magnificant Menthol Disposable (75% glycerine, 18% water, 2% nicotine, and 5% flavour) CC Marlboro Gold King Box filtered cigarette Conducted at Eastcoast Research facility, NC, USA	Three groups of 10 regular dual CC smokers / E(N)NDS users (abstinent for 1 h prior to testing) underwent 9 sessions of 1 of the following test protocols: E(N)NDS use (average around 95–99 puffs/session) Smoked 3 x CC (average around 30 puffs/session) Exhaled breath was collected on filter pads and analysed for levels of phenolics, carbonyls, water, glycerine, and nicotine. Room air was also evaluated for levels of carbonyls (before product use).	*Exhaled breath measurements:* Water, glycerine, and nicotine Average distribution of mass balances: blu CTD: 73.3% water, 26.7% glycerine, 0.049% nicotine blu MMD: 75.7% water, 24.2% glycerine, and 0.057% nicotine CC: 83 ± 21% nicotine/glycerine/water (0.40% nicotine) Total phenolics Blanks (prior to product use) and E(N)NDS (both brands): generally < LOQ CC: 66 µg/session (range 36–117 µg/session) Total carbonyls Blanks (before product use) and E(N)NDS (both brands): generally < LOQ CC: average 242 µg/session (range 136–352 µg/session) *In room air blanks, total carbonyls < LOQ* Authors concluded that exhaled E(N)NDS aerosol does not increase bystander exposure for phenolics and carbonyls above the levels observed in exhaled breaths of air.
Marco and Grimalt (2015) Identification and evaluation of volatile compounds in directly sampled and exhaled-breath CC smoke and E(N)NDS vapour	VOCS (presence or absence of 156 compounds)	Disposable E(N)NDS Rechargeable E(N)NDS Blend-type American cigarettes with filter and low nicotine content, low tar, and low CO Barcelona, Spain	Exhaled breath was sampled 30 min after smoking CC or using E(N)NDS (total 5 breaths collected per test). The authors stated that "To avoid metabolic differences all volunteers were asked to smoke with the tobacco cigarettes and Type 1 and 2 e-cigarettes considered in this study." However, the number of volunteers/tests performed was not stated.	Exhaled air (no CC or E(N)NDS use) Acetone and isoprene were the main compounds in exhaled breath Direct sampling of smoke/aerosol *CC*: The chromatogram indicated a large number of compounds in directly sampled CC smoke, including VOCs, nicotine and related compounds, aromatic compounds (e.g. benzene, toluene), polar compounds (e.g. ethanol, aldehydes and ketones), and unsaturated compounds (e.g. buta-1,3-diene, isoprene) *E(N)NDS*: Analysis revealed mainly PG and glycerine, and also nicotine and related compounds. Product from the rechargeable

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td></td><td></td><td>CC smoke, E(N)NDS aerosol, and room air were also sampled directly (without inhalation by user)

VOCs: collection with Bio-VOC, absorption in Tenax cartridges and analysis by TD-GC-MS</td><td>E(N)NDS also contained vanillin and ethyl vanillin.

<u>Exhaled breath sampling</u>
CC: Chromatogram indicated similar profile to that of directly sampled CC smoke, except that the results suggested that most of the original smoke components were retained in the lungs. Nicotine, nicotyrine and 2,5-dimethylfuran were noted to be present in exhaled breath. Isoprene (endogenous compound) was the most abundant exhaled breath peak.

E(N)NDS: For exhaled E(N)NDS product, the chromatographic peaks of PG and glycerine were absent. Peaks for nicotine and related compounds were also substantially decreased compared with direct sampled aerosol. The main peaks were acetone and isoprene (endogenous).

<u>Nicotine concentrations are shown in the table below µg/m^3)</u>

<table><tr><td></td><td colspan="2">Nicotine concentration (µg/m^3)</td></tr><tr><td></td><td>Direct sample</td><td>Exhaled breath</td></tr><tr><td>Room air</td><td>not detected</td><td>not detected</td></tr><tr><td>CC</td><td>1300</td><td>7</td></tr><tr><td>Disposable E(N)NDS</td><td>720</td><td>4</td></tr><tr><td>Rechargeable E(N)NDS</td><td>710</td><td>1</td></tr></table>
Authors concluded that the Bio-VOC system is suitable for the analysis of VOCs in exhaled breath of CC and E(N)NDS users, and that the comparisons made indicated a higher burden of VOC exposure to tobacco smokers than to E(N)NDS users.</td></tr>
<tr><td colspan="5">Evaluation of ambient air and surfaces in exposure chambers or rooms after specified E(N)NDS use</td></tr>
<tr><td>Schripp et al. (2013)

Evaluation of particulate matter and VOCs in air in an exposure chamber during E(N)NDS or CC use.</td><td>VOCs

FP/ UFP</td><td>Tank system with 3 E(N)NDS liquids:

Liquid 1: apple-flavoured (main flavour compound 2-methylbutyl-3-methylbutanoate), 0 mg/mL nicotine</td><td><u>Large chamber study</u>
Volunteer in an 8 m^3 stainless steel emission test chamber (23 ^{o}C, 44.5% relative humidity, air exchange rate 0.3/h) took 6 deep-lung, 3-s puffs at intervals of 60 s. Air sampling started at puff 4 and continued for 15 min. Particle size distribution</td><td><u>Large chamber study</u>
A bimodal particle size distribution (maxima at 30 and 100 nm) was noted.

<u>Pre-test chamber air levels</u>
All 20 VOCs were < 1 µg/m^3, except isoprene, 8 µg/m^3

<u>E(N)NDS</u>
Of the 20 VOCs analysed, 5 (listed below) were detected above</td></tr>
</table>

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
Measurement of VOC concentrations in exhaled E(N)NDS aerosol		Liquid 2: apple-flavoured (main flavour compound 2-methylbutyl-3-methylbutanoate), 18 mg/mL nicotine Liquid 3: tobacco-flavoured (main flavour compound, ethyl maltol), 18 mg/mL nicotine and CC (brand not stated), 0.8 mg/cigarette nicotine	measurements were made by fast FMPS spectrometry (TSI) (detection range 5.6-560 nm). Tests were conducted in the order, Liquid 1, Liquid 2, Liquid 3, CC. Air levels of 20 VOCs were measured by GC/MS after sampling on Tenax tubes or by HPLC after collection using silica gel cartridges <u>Small chamber study</u> Volunteer exhaled 1 E(N)NDS puff (Liquid 1) into a 10-L glass chamber (37 °C, 27.2% relative humidity, air exchange rate 3/h). VOC concentrations in chamber air were measured by GC/MS after sampling on Tenax TA tubes (6L, 150 mL/min).	pre-test chamber air levels during E(N)NDS use, while the other 15 did not increase. 1,2-propanediol (PG) was not detected. 2-butanone: 2 µg/m^3 acetic acid: 11-14 µg/m^3 acetone: 17-25 µg/m^3 formaldehyde: 8-16 µg/m^3 acetaldehyde: 2-3 µg/m^3 <u>CC</u> Levels of emissions after CC use were higher than pre-test for all 20 VOCs, including: 1,2-propanediol: 112 µg/m^3 2-butanone: 19 µg/m^3 acetic acid: 68 µg/m^3 acetone: 64 µg/m^3 formaldehyde: 86 µg/m^3 acetaldehyde: 119 µg/m^3 <u>Small chamber study</u> 1,2-propanediol (PG): 53,000–175,000 µg/m^3 1,2,3-propanetriol (glycerol): 161–326 µg/m^3 3-methylbutyl-3-methylbutanoate (apple oil): 3–35 µg/m^3 diacetin (flavour): 1–2 µg/m^3 nicotine: 4–7 µg/m^3 Authors commented that inhaled aerosol undergoes changes in the human lung that is assumed to be attributed to deposition and evaporation. From the small-chamber exhaled breath analysis, they concluded that some ‘passive vaping’ must be expected from E(N)NDS use.
Czogala et al. (2014) Evaluation of nicotine, particles, CO, and VOCs in air in an exposure	Ambient air concentrations of: Nicotine $PM_{2.5}$	Users own brands: <u>E(N)NDS</u> M201 pen-style (18 mg/mL nicotine; Mild brand) (2 users)	5 adult male experienced dual users of CC and E(N)NDS followed (singly) the following protocol: E(N)NDS used *ad libitum* for 5 min, twice, with a 30-min interval, after which the room was decontaminated and	The LOD for nicotine was 0.22 µg/m^3. Substantial variation in emission levels between brands. Mean (range) concentrations over 5 experiments (users) were: <u>Nicotine</u>: (µg/m^3) Baseline: < 0.22 (< LOD)

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
chamber during E(N)NDS or CC use. [One of the authors declared receipt of funding for travel from Chic Group Ltd., an E(N)NDS manufacturer]	CO VOCs	eGo (16 mg/mL nicotine; Janty brand) (2 users) M401 (18 mg/mL nicotine; Nicore brand, Atina Poland) CC L&M Blue Label brand (ISO yield/cigarette: 0.6 mg nicotine, 8 mg tar, 9 mg CO) (3 users) Marlboro Gold (ISO yield/cigarette: 0.5 mg nicotine, 7 mg tar, 7 mg CO) (2 users)	ventilated for 5 min. Then 2 x CC smoked, with a 30-min interval between each CC. Tests were conducted in a 39 m^3 exposure chamber equipped with exhaust, ventilation system, fans for mixing indoor air, and an air sampling station. The air exchange rate in the chamber during this part of the study was not reported 1-h mean concentrations of nicotine, aerosol particles ($PM_{2.5}$), CO, VOCs were determined at baseline, during E(N)NDS use, and during CC use (3 h total). p values were calculated for air concentrations, E(N)NDS vs. baseline and E(N)NDS vs. CC. Nicotine was measured using GC with nitrogen phosphorus detector following active sampling on XAD-4 sorption tubes, according to NIOSH method 2551. PM2.5 were measured continuously with SidePak AM510 Personal air monitor. CO was measured with Q-Trak Indoor Air Quality 8550 monitor. VOCs were analysed with GC-MS following active sampling on CSC sorption tubes according to OSHA reference method.	E(N)NDS: 3.32 ± 2.49 (0.65–6.23) CC: 31.60 ± 6.91 (25.6–41.8) $p < 0.05$ for E(N)NDS vs. baseline and for E(N)NDS vs. CC $PM_{2.5}$ (µg/m^3) Baseline: 32.4 ± 30.0 (8.0–80.0) E(N)NDS: 151.7 ± 86.8 (63.3–272.7) CC: 819.3 ± 228.6 (661.3–1217.3) $p < 0.05$ for E(N)NDS vs. baseline and for E(N)NDS vs. CC CO (ppm) Baseline: 1.40 ± 0.55 (1–2) E(N)NDS: 1.40 ± 0.55 (1–2) CC: 3.80 ± 0.84 (3–5) $p < 0.05$ for E(N)NDS vs. CC VOCs (µg/m^3) *Toluene* Baseline: 4.09 ± 2.12 E(N)NDS: 3.79 ± 2.16 CC: 14.75 ± 6.02 $p < 0.05$ for E(N)NDS vs. CC *Ethylbenzene, m,p-xylene, and o-xylene*: Baseline: not identified (LOD not stated) E(N)NDS: not identified CC: 1.17 ± 1.44, 1.94 ± 1.14, and 0.48 ± 0.95, respectively $p < 0.05$ for E(N)NDS vs. CC Authors concluded that E(N)NDS emit significant amounts of nicotine but do not emit significant amounts of CO and VOCs, and that emissions of nicotine from E(N)NDS are significantly lower than those from CC. Authors also commented that limitations of the study included: chamber input air was not filtered; chamber ventilation rate was higher than that of a standard residential room; levels of other potential emissions were not measured.

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr>
<td>Ruprecht et al. (2014)

Measured particulate matter emissions into ambient air from E(N)NDS and CC use</td>
<td><u>Ambient air</u>:

Particulate matter</td>
<td>Elips Serie C tank system (Ovale Europe) (0 or 16 mg nicotine)

CC (brand not stated)

Conduction at ‘Fondazione IRCCS Istituto Nazionale dei Tumori’ (a cancer research centre), Milan, Italy</td>
<td>3 volunteers (1 per test type) used E(N)NDS at 1 puff/min for 7 min, 3 min interval, for 2–3 h, or smoked 1 CC

Particles in ambient air of a 50 m^3 room (air exchange rate 0.80/h to 0.86/h) were evaluated using:

Aerocet Model 531 optical particle counter (Met One Instruments) to measure PM and total suspended particles (TSP)

Condensation Particle Counter (3007, TSI) to measure UFP</td>
<td><u>Particulate matter</u> (µg/m^3 ± SD) (subtracted against outside ambient air)

<table>
<tr><th></th><th>E(N)NDS (- nicotine)</th><th>E(N)NDS (+ nicotine)</th><th>CC</th></tr>
<tr><td>PM_1</td><td>3.5±7.3</td><td>0.0±0.3</td><td>76±18</td></tr>
<tr><td>$PM_{2.5}$</td><td>7.2±9.6</td><td>0.5±1.1</td><td>139±32</td></tr>
<tr><td>PM_7</td><td>8.7±9.9</td><td>-0.3±3.1</td><td>155±36</td></tr>
<tr><td>PM_{10}</td><td>9.9±10.3</td><td>-0.6±4.4</td><td>158±37</td></tr>
<tr><td>TSP</td><td>11.6±15.5</td><td>1.2±10.1</td><td>160±37</td></tr>
</table>

<u>UFP</u> approximate fold increase over background (outside ambient air), particles/m^3

CC: 1.5 x 10^5
EC (- nicotine): 650
EC (+ nicotine): 550

Authors concluded that E(N)NDS generate less indoor air pollution (particulate matter) than CC.</td>
</tr>
<tr>
<td>Saffari et al. (2014)

Comparison of indoor air during E(N)NDS or CC use with outdoor air in the vicinity of the same building</td>
<td><u>Ambient air</u>:

Total particulate matter (TPM)

PAHs

N-alkanes and organic acids

Metals

Nicotine</td>
<td>Elips Serie C, tank system, Ovale Europe

E(N)NDS liquid (PG, glycerol, aroma, water, 0 or 16 mg/mL nicotine)

CC, brand not stated

Conduction at ‘Fondazione IRCCS Istituto Nazionale dei Tumori’ (a cancer research centre), Milan, Italy</td>
<td>A volunteer used E(N)NDS (1 puff/min for periods of 7 min, with 3 min intervals) or smoked CC ad libitum (average 7 min/CC, with a 3 min pause between each CC) in a 48 m^3 room. The air exchange rate in the room during sampling was 1.1/h.

Approximately 1.3 mL E(N)NDS liquid consumed per hour
TSPs were collected on quartz filters using a high-volume PM sampler (flow-rate, 240 L/min). Black carbon concentration measured using an Aethalometer, CO measured using an indoor air analyser; inorganic elements collected on quartz filters at flow-rate of 240 L/min and analysed by ICP-MS. Organic species measured by</td>
<td>Results were presented relative to measurements in outdoor air on a terrace adjacent to the building where the indoor experiments were conducted.

<u>TPM</u>
Average levels reported as a figure in the paper were approximately 50 µg/m^3 indoors during E(N)NDS use and in outdoor air, and 250 µg/m^3 indoors during CC smoking.

<u>Organic species</u>
PAHs were detected indoors during CC smoking, but not during E(N)NDS use.
35 out of 36 N-alkanes and organic acids for which measurement ratios were reported were detected at higher concentrations indoors during E(N)NDS use than in outdoor air (all except levoglucosan). However, the narrative notes that in most cases indoor air levels during E(N)NDS use were < 100 ng/m^3, that these compounds were not detected in E(N)NDS liquids, and thus the compounds were likely to derive from a source unrelated to E(N)NDS use.</td>
</tr>
</table>

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
			GC-MS.	Metals Indoor (during E(N)NDS use) /outdoor ratios were calculated for 25 metals. Indoor/outdoor ratios were > 1 for boron (13.13), potassium (1.53), nickel (1.75), zinc (1.26), silver (3.39), and lanthanum (1.47). For all other metals, indoor/outdoor ratios were < 1. For the 6 metals that showed higher levels indoors during E(N)NDS use than outdoors, boron, potassium, zinc, and lanthanum levels were lower indoors during E(N)NDS use than during CC smoking, while nickel and silver were of the same (low) order of magnitude during E(N)NDS use and CC smoking. Modelling suggested that the amount of E(N)NDS liquid vaporised during the experiments was not sufficient to be the unique source of these metals measured in the room air. Using the data obtained from indoor and outdoor air analyses, the authors estimated average emission rates (ng/h) for 25 metals, and 32 alkanes and organic acids during CC smoking or E(N)NDS use. Detectable emission rates were reported for 10 metals (boron, 963.8 ng/h; potassium, 7765 ng/h; titanium, 50.16 ng/h; chromium, 28.10 ng/h; nickel, 130.5 ng/h; zinc, 1142 ng/h; silver, 20.91 ng/h; cadmium, 0.480 ng/h; lanthanum, 3.210 ng/h; lead, 96.16 ng/h). Of these, 4 (titanium, chromium, nickel, silver) were estimated to have indoor emission rates from E(N)NDS use that were higher than from CC smoking. Emission rates of alkanes and organic acids calculated for E(N)NDS use were in all cases much lower than from CC smoking. It was noted, however, that components in the vapour phase were not evaluated in this study. Nicotine Particle-phase nicotine levels, mass ratios, and emission rates were reported as follows (outdoor concentrations were not reported):

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td></td><td></td><td></td><td>
<table>
<tr><td>Nicotine (average ± SE)</td><td>CC smoking</td><td>E(N)NDS use (no nicotine)</td><td>E(N)NDS use (with nicotine)</td></tr>
<tr><td>Indoor concentration (ng/m^3)</td><td>1524 ± 80.4</td><td>60.68 ± 20.91</td><td>123.0 ± 34.5</td></tr>
<tr><td>Indoor/outdoor mass ratio</td><td>254.3 ± 13.4</td><td>6.7 ± 3.5</td><td>18.6 ± 7</td></tr>
<tr><td>Emission rate (ng/h)</td><td>91,161 ± 2170</td><td>2759 ± 93</td><td>7103 ± 172</td></tr>
</table>
Detection of nicotine in particulate matter during use of E(N)NDS containing no nicotine was suggested to be due to incorporation onto particles of residual nicotine on surfaces in the room. Subtraction of this background (thirdhand) nicotine emission from that measured during use of E(N)NDS containing nicotine gave an emission rate of 4344 ng/h, which the authors estimated would account for a very small amount (0.02%) of total nicotine emission from E(N)NDS being released into particulate emissions. However, this would not take into account nicotine in the vapour phase, and further investigation of gas-to-particulate partitioning of nicotine was recommended by the authors.
Authors commented that particle phase emissions were much lower from E(N)NDS use than CC smoking, PAHs were not detected from E(N)NDS use, and emission rates of organic compounds and inorganic elements were significantly reduced from E(N)NDS use compared with CC smoking. Some metals were noted to have higher emission rates from E(N)NDS than from CC smoking, with nickel and silver being of possible concern. Secondhand particle-phase nicotine was estimated to be 0.02% of the total nicotine generation and emission from E(N)NDS use.
The authors concluded that E(N)NDS represent an improvement on CC from a public health perspective, but that manufacturing processes for E(N)NDS should be improved and standardised, with implementation of quality control regulations to avoid the presence of non-desirable materials.
</td></tr>
<tr><td>Schober et al. (2014)</td><td>PM

Particle number</td><td>E(N)NDS device comprising a rechargeable lithium-</td><td>45 m^3 room with air exchange rate of 0.76/h during the control session and ranging from 0.37 to</td><td>PM levels were generally higher during E(N)NDS use than in air tested on the control day, with highest levels mostly associated with use of E(N)NDS without nicotine.</td></tr>
</table>

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
Evaluation of emissions into indoor air associated with use of E(N)NDS with and without nicotine	concentration (PNC) PG, glycerine Nicotine Carbonyls PAHs Metals	ion battery, electronic circuit, vaporiser, mouthpiece and refillable tank) 3x Red Kiwi (Seevetal, Germany) tobacco-flavour E(N)NDS liquids with or without 18 mg/mL nicotine (PG/glycerine ratios were approximately 50:50) Study conducted in an office building of the Bavarian Health and Food Safety Authority, Munich, Germany	0.74/h for the E(N)NDS test sessions. For each tested E(N)NDS product, 3 subjects simultaneously used the product during a 2-h test session each day for 6 days. In each case, the nicotine-free product was tested on the first day, followed by 5 sessions using the nicotine-containing product. Air was sampled the day before the first E(N)NDS use session and then during each daily session, at a distance of approximately 1 m from the users, and 1 m height from the floor. Particle mass (PM_{10}, $PM_{2.5}$ and $PM_{1.0}$) was measured with optical laser aerosol spectrometer (LAS) (size range, 0.300–20 µm). PNC was measured with a wide range aerosol spectrometer (WRAS) (range 0.005–>20 µm). VOCs were collected at 0.2 L/min flow rate with Tenax GR adsorbant and analysed with a thermodesorption unit coupled to GC/MS (LOD 0.04 µg/m^3). Aldehydes and ketones were measured by method according to NIOSH Method 2018 (LOD, 30 µg/m^3). 16 PAHs were collected by quartz fibre filter and polyurethane foam, extracted with toluene, silica-	Mean $PM_{2.5}$ concentration was 197 µg/m^3 (maximum, 514 µg/m^3) during E(N)NDS use and 6 µg/m^3 in control air. Median PNC was in the range 48,620 – 88,386 particles/cm^3, with peak values at diameter 24–36 nm. There was no increase in levels of CO or carbon dioxide (CO_2). For PG, glycerine, and nicotine: Mean levels in control air were all < 0.04 µg/m^3 Mean ± SD levels during E(N)NDS use were 199.2 ± 93.2 (maximum, 395) µg/m^3 (PG), 72.7 ± 6.9 (maximum, 81) µg/m^3 (glycerine), and 2.2 ± 1.7 (maximum, 4.6) µg/m^3 (nicotine). Formaldehyde, benzene, acrolein and acetone were not higher during E(N)NDS use than control air, except during one session, where formaldehyde increased to 55 µg/m^3 (baseline level reported as 25 µg/m^3). Levels of 16 PAHs were 30–90% higher during E(N)NDS use than in control air, while levels of a subset of 7 PAHs that are classified as carcinogenic by IARC were raised by around 20% compared with control air. There was a wide variation between measurements reported for levels of individual metals during different test sessions, with values both higher and lower than control air noted. The authors commented that there was a 2.4-fold increase in aluminium levels during E(N)NDS use (482.5 ± 158.6 ng/m^3) compared with control air (203.0 ng/m^3). The authors noted that analysis of indoor air quality during E(N)NDS use showed that E(N)NDS products are not emission-free. Substantial amounts of PG, glycerine, and nicotine were emitted in the gas phase, as well as high concentrations of fine and ultrafine particles, and a 2.4-fold increase in aluminium.

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
			purified and analysed by GC/MS (LOD, 0.1 ng/m^3). Metals (aluminium, antimony, arsenic, bismuth, cadmium, calcium, cerium, chromium, cobalt, copper, iron, lanthanum, lead, magnesium, manganese, molybdenum, nickel, potassium, sodium, thallium, tin, titanium, vanadium, zinc) were collected on quartz filter pads and measured by ICP-MS.	
O'Connell et al. (2015) Evaluation of indoor air before, during, and after E(N)NDS use [Study carried out by Imperial Tobacco Group, the parent company of Fontem Ventures B.V., the manufacturer of the E(N)NDS products tested in the study]	PG, glycerol Nicotine VOCs Carbonyls (formaldehyde, acetaldehyde, acrolein) 16 PAHs (US EPA 'priority list') TSNAs (NNN, NNK, NAT, NAB) Metals/elements (US EPA 'Method 29' metals: antimony, arsenic, barium, beryllium, calcium, chromium, cobalt, copper, lead, manganese, mercury, nickel, selenium, silver, thallium, zinc; plus	Puritane 16 mg/g disposable original-flavoured, closed-system E(N)NDS with e-liquid containing 67% PG, 30% glycerol, 1.6% nicotine, flavourings) UK	38.5 m^3 office room, average air exchange rate of 0.8/h. Analysis of ambient air before (1 h empty room, then 1 h room + participants), during a 165 min session of *ad libitum* E(N)NDS use, and for 75 min immediately after the participants stopped using E(N)NDS and left the room In total, 5 participants were present in the room; 3 used E(N)NDS at an average puff rate of 3.2 puffs//min across the 3 users; 2 participants were present but did not use E(N)NDS Nicotine collected at 1 L/min into XAD2 sorbent tubes, and analysed by GC-MS (LOD 7.0 µg/m^3). VOCs sampled at 0.15 L/min and analysed according to ISO 16000-6 international standard	Average levels of chemicals determined in ambient air before, during and after E(N)NDS use (µg/m^3) *PG*: Average levels were < LOD (< 0.5 µg/m^3) at background and occupied room before E(N)NDS use; 203.6 µg/m^3 during E(N)NDS use; 10 µg/m^3 in the empty room after E(N)NDS use. Authors noted that the UK WEL for PG is 474,000 µg/m^3. *Glycerol*: Glycerol was not detected, which may have been due to the relatively high LOD of the detection method (350 µg/m^3). Authors noted that the UK WEL for glycerol is 10,000 µg/m^3. *Nicotine*: Levels were below the LOD of 7.0 µg/m^3 at all times. Authors noted that the UK WEL for nicotine is 500 µg/m^3. Authors considered that the lack of nicotine detected in the room air during E(N)NDS use may be attributable to high retention rate of nicotine in the body. Total VOCs (TVOC) rose from 65.0 µg/m^3 in background air to 237.0 µg/m^3 in occupied room, 379.0 µg/m^3 during E(N)NDS use, then fell to 129.0 µg/m^3 afterwards. Authors considered that increases were likely related to cosmetic and toiletry products used by the volunteers. *Carbonyls*: Levels of formaldehyde (32.0 µg/m^3 before/unoccupied, 31.0 µg/m^3 before/occupied, 37.6 µg/m^3

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
	aluminium and phosphorus)		(LODs for individual VOCs, 0.5-1.0 µg/m^3). Glycerol collected at 1 L/min into XAD7 sorbent tubes and analysed by thermodesorption coupled to solvent extraction and GC-MS (LOD 150-350 µg/m^3). Carbonyls sampled at 1.5 L/min and analysed according to ISO 16000-3 international standard (LOD 2.0 µg/m^3). PAHs sampled at 2 L/min, collected into XAD2 sorbent tubes, and analysed by solvent extraction and high-resolution GC-MS (LOD for each PAH, 1.25 µg/m^3). Trace elements sampled at 1.5 L/min, collected on mixed cellulose ester (MCE) filters and analysed by inductively coupled plasma optical emission spectroscopy (ICP-OES) (LODs in the range 1–2 µg/m^3 except 10 µg/m^3 for phosphorus).	during E(N)NDS, 21.0 µg/m^3 after), acetaldehyde (9.0 µg/m^3 before/unoccupied, 6.5 µg/m^3 before/occupied, 12.4 µg/m^3 during E(N)NDS, 6.0 µg/m^3 after), and acrolein ((< LOD at all times), did not rise substantially during E(N)NDS use, and were all noted by authors to be below WHO or EU indoor AQG levels. *PAHs*: Levels of all 16 PAHs were < 1.25 µg/m^3 at all times. *Trace metals*: All measurements were below the LODs. Authors noted that levels were all below UK workplace WELs where these values had been established. *TSNAs*: All measurements were below the LOD for the TSNAs evaluated. Authors commented that exposure of bystanders to the chemicals in the exhaled E(N)NDS aerosol, at the levels measured in this study, would be below current regulatory standards that are used for workplaces or general indoor air quality. They concluded that this finding supports the conclusion that there is no apparent risk to bystanders from exhaled E(N)NDS aerosols.
Maloney et al. (2016) Measurement of indoor air concentrations of nicotine, menthol, PG, glycerol, and total particulates during E(N)NDS use	Nicotine Menthol, PG Glycerol TSP	MarkTen brand E(N)NDS containing 1.5–2.5% nicotine, PG, glycerine, and proprietary flavourings 2 products tested: menthol MarkTen and non-menthol	Room of 137 m^3; Test sessions comprised the use of E(N)NDS product by 2–12 users for 6 periods of 1 h (total, 36–216 puffs per 1-h session), interspersed with 1-h non-use periods, for 4 days (2 days per product type). Background sampling the day before and the day after each 4-day study.	*Active (integrated) sampling* <u>LOQ</u> Study 1: 180 µg/m^3 (menthol), 15 µg/m^3 (nicotine), 65 µg/m^3 (PG), 77 µg/m^3 (glycerol), 153 µg/m^3 (TSP) Study 2: 210 µg/m^3 (menthol), 10 µg/m^3 (nicotine), 45 µg/m^3 (PG), 53 µg/m^3 (glycerol), 39 µg/m^3 (TSP), 1 µg/m^3 (formaldehyde) <u>Measurements</u> All measurements before, during, and after E(N)NDS use were

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
[Study funded by Altria Client Services, a subsidiary of Altria Group. NuMark, a subsidiary of Altria Group, is a manufacturer of E(N)NDS products]		MarkTen Study 1 (early 2014) Study 2 (mid 2014) Richmond, Virginia, USA	The air exchange rate in the room was 1.47/h during Study 1 and 1.56/h during Study 2. Active integrated air sampling during the 12-h session: Nicotine, menthol, and formaldehyde (Study 2 only for the latter) were collected using 'Pocket Pumps' (XAD4 for nicotine, Anasorb coconut charcoal for menthol, XAD-2 for formaldehyde) at a flow rate of 150 cm^3/min. PG and glycerol were collected on XAD-7 using AirCheck sampling pumps at a flow rate of 1.0 L/min. Particulates were collected on polyvinyl chloride. Near-real-time sampling and direct-reading measurements: For nicotine and menthol using MINICAMS portable gas chromatograph (GC), flow rate 200 cm^3/min for 3 min, flame ionization detection analysis. Particulates counted using a P-Trak 8525 particle counter (0.02–1 µm). Formaldehyde 30-min average concentrations (Study 2 only) using near-real-time monitor (30-min average). Active sampling on walls approximately 20 inches above E(N)NDS users. Sampling was carried out according to NIOSH methods.	below the LOQ values except for formaldehyde (Study 2), for which values were quantifiable but did not change substantially from background (5–8 µg/m^3). *Near-real-time and direct-reading measurements* From these readings, TWAs were calculated as: Menthol, range 59–90 µg/m^3 Nicotine, range < 1.2–6.2 µg/m^3 Formaldehyde, all results below LOD Particle counts, ranged from around 2000–220,000 particles per cm^3 on E(N)NDS use test days, not measured on the day prior to tests, and approximately 10,000–15,000 particles per cm^3 on the day after E(N)NDS use tests. Authors commented that, except for background levels of formaldehyde, measurements for nicotine, menthol, PG, and glycerine were all below the LOQ for the MarkTen prototype E(N)NDS products used in these studies. They noted that levels of formaldehyde were in the same range during active E(N)NDS use as background measurements the day before E(N)NDS use. Particle counts were taken to be a good indicator of E(N)NDS use, with levels spiking during active use sessions and returning to background levels approximately 1 h after the end of the use session.

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr>
<td>Liu et al. (2017)

Evaluation of ambient air and surface samples in an exposure chamber during use of E(N)NDS products or CC.

[Study funded by Altria Client Services, and conducted on behalf of the subsidiary of Altria Group: NuMark LLC, which produces and markets e-vapour products]</td>
<td>34 chemicals:

PG

Glycerol

nicotine

15 carbonyls

12 VOCs

4 trace elements (chromium, nickel, cadmium, arsenic)</td>
<td><u>Group I</u>
MarkTen (2.5% nicotine, 306 mg/mL PG, 600 mg/mL glycerol, proprietary flavours)

<u>Group II</u>
Prototype cartridge-based E(N)NDS (2.4% nicotine, 465 mg/mL PG, 480 mg/mL glycerol, proprietary flavours) (Group II)

<u>Group III</u>
Compact Ego RBC Tank device (900 mAh, 3.7 V) with participant’s own E(N)NDS liquid (5.92–23.9 mg/mL nicotine, 362–688 mg/mL PG, 212–700 mg/mL glycerol)

<u>Group IV</u>
CC (participant’s own brand)

December 2014, High Point, NC, USA</td>
<td>114 m^3 exposure chamber. Fresh air was supplied to the room at a rate of 7.5 L/s.

Participants were experienced users of the product of their test group (n=8 for group I, n=9 for groups II and III; n=10 for group IV; all participants for each group were present in the test chamber at the same time)

Pre-specified product use (groups I and II only; total 80 puffs per each user over 4 h)

Ad libitum product use (total: group I, 1224 puffs; group II, 747 puffs; group III, 1649 puffs; group IV, 45 CC)

Control = room air before product use, firstly without (background) and then with (baseline) participants present in the room

Air sampling: nicotine, PG, glycerol collected on XAD-7 adsorbent tubes at 1000 mL/min and analysed by GC-mass selection; carbonyls collected on DNPH-coated silica gel adsorbant tubes at 200 mL/min and analysed by HPLC-UV; VOCs collected on SVI thermal desorption tubes at 60 mL/min and analysed by GC-flame ionization; trace elements collected on quartz filters at</td>
<td>LOQ: 0.25 µg/m^3 (nicotine); 3.63 µg/m^3 (PG), 4.11 µg/m^3 (glycerol); 0.12 µg/m^3 (individual metals); 0.62–1.45 µg/m^3 (different individual carbonyls); 0.18–6.33 µg/m^3 (various other individual VOCs)

<u>Primary outcome – mean change in air level of measured constituents over baseline (34 analytes)</u>

Results for nicotine, PG, and glycerol are shown in the table below.
<table>
<tr><td></td><td colspan="6">Mean change over baseline under product use conditions (µg/m^3)</td></tr>
<tr><td></td><td colspan="5">E(N)NDS</td><td>CC</td></tr>
<tr><td></td><td colspan="2">Group I (MarkTen)</td><td colspan="2">Group II (cartridge)</td><td>Group III (tank)</td><td>Group IV</td></tr>
<tr><td>Product-use regime</td><td>PS</td><td>AL</td><td>PS</td><td>AL</td><td>AL</td><td>AL</td></tr>
<tr><td>Nicotine</td><td>0.48 ± 0.16</td><td>0.38 ± 0.07</td><td>2.83 ± 0.44</td><td>0.96 ± 0.22</td><td>1.47 ± 0.32</td><td>40.65 ± 6.40</td></tr>
<tr><td>PG</td><td>44.86 ± 3.84</td><td>33.06 ± 1.97</td><td>211.51 ± 14.23</td><td>68.51 ± 4.58</td><td>317.06 ± 12.45</td><td>56.21 ± 4.22</td></tr>
<tr><td>Glycerol</td><td>67.89 ± 16.81</td><td>98.90 ± 28.14</td><td>126.75 ± 12.71</td><td>78.65 ± 8.75</td><td>242.00 ± 7.62</td><td>< LOQ</td></tr>
</table>
PS, pre-specified; AL, ad libitum

Significant increases were noted for all 3 compounds under all tests, except glycerol in group IV (CC).

For the other chemicals tested, although some significant differences were noted between baseline and test conditions, both positive and negative changes were noted during E(N)NDS use, with no major overall consistent pattern across the different products. In addition, many of the chemicals were around or below the LOQ value during E(N)NDS use.

<u>Secondary outcome – mean change in surface levels of nicotine, PG, and glycerol</u>

Results are shown in the table below:</td>
</tr>
</table>

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td></td><td></td><td>1700 mL/min and analysed by ICP-MS

Surface sampling: nicotine, PG, glycerol collected on 15 cm petri dishes</td><td>
<table>
<tr><td></td><td colspan="5">Mean change from baseline to product use conditions (µg/cm²)</td></tr>
<tr><td></td><td colspan="4">E(N)NDS</td><td>CC</td></tr>
<tr><td></td><td colspan="2">Group I (MarkTen)</td><td colspan="2">Group II (cartridge)</td><td>Group III (tank)</td><td>Group IV</td></tr>
<tr><td>Product-use regime</td><td>PS</td><td>AL</td><td>PS</td><td>AL</td><td>AL</td><td>AL</td></tr>
<tr><td>Nicotine</td><td>0.001 ± 0.002</td><td>< LOQ (0.001)</td><td>< LOQ</td><td>< LOQ</td><td>< LOQ</td><td>0.001 ± 0.002</td></tr>
<tr><td>PG</td><td>0.01</td><td>< LOQ (0.019)</td><td>0.05</td><td>0.01</td><td>< LOQ</td><td>< LOQ</td></tr>
<tr><td>Glycerol</td><td>< LOQ</td><td>0.02 ± 0.01</td><td>0.07 ± 0.02</td><td>< LOQ</td><td>0.35 ± 0.12</td><td>0.00 ± 0.00</td></tr>
</table>
PS, pre-specified; AL, ad libitum

The change was statistically significant ($p < 0.05$) for glycerol/group III

Authors noted that although PG and glycerol were detected in air upon E(N)NDS use, levels were several-fold below workplace exposure limits.

Authors concluded that under the conditions of this study, E(N)NDS products did not generate chemicals at levels that could likely pose health concerns for non-users.</td></tr>
<tr><td>Melstrom et al. (2017)

Measurement of second-hand and third-hand PM and nicotine levels associated with short-term E(N)NDS use in a room.</td><td>PM2.5

UFP (< 100 nm)

Nicotine

(second-hand and third-hand exposure)</td><td>Disposable E(N)NDS with ≥ 18 mg/mL nicotine

Tank-style E(N)NDS with ≥ 18 mg/mL nicotine

(brands not stated)</td><td>52.6 m³ room. Mean ventilation rate, approximately 5/h.

2 x 2-h E(N)NDS use test sessions: 3 experienced E(N)NDS users used the test product ad libitum; 6 non-users were also present in the room. Measurements were made before, during, and after the 2-h sessions.

Ambient air measurements: $PM_{2.5}$ (SidePak) and UFP, < 100 nm (P-Trak) by aerosol monitors on the table in the exposure room; air nicotine collected by</td><td><u>$PM_{2.5}$, mean (SD; range), mg/m³</u>
Disposable, before E(N)NDS use: 0.131 (0.019; 0.003–0.761)
Tank, before E(N)NDS use: 0.018 (0.015; 0.006–0.827)
Disposable, during E(N)NDS use: 0.788 (2.147; 0.002–19.961)
Tank, during E(N)NDS use: 1.454 (2.683; 0.007–19.972)
Disposable, after E(N)NDS use: 0.008 (0.008; 0.001–0.228)
Tank, after E(N)NDS use: 0.017 (0.011; 0.005–0.390)
$p < 0.0001$ for during vs. before/after E(N)NDS use and
$p < 0.0001$ for during tank vs. during disposable E(N)NDS use

<u>UFP mean (SD; range), particles/cm³</u>
Disposable, before E(N)NDS use: 4092 (1048; 0–10,700)
Tank, before E(N)NDS use: 3323 (507; 2190–4680)
Disposable, during E(N)NDS use: 38,695 (31,437; 1800–239,000)
Tank, during E(N)NDS use: 31,227 (24,060; 3360–225,000)
Disposable, after E(N)NDS use: 2829 (1000; 1360–4730)</td></tr>
</table>

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
			personal air samplers (SKC XAD-4 canisters) at mouth level of non-users in the room (LOQ, 0.2 ng/air tube) (sampling flow rates were not reported), extracted, and analysed by LC/MS/MS with a HILIC silica column. Surface measurements: 100 cm^2 wipe samples taken before and after test sessions, and wipe cloths worn during E(N)NDS use	Tank, after E(N)NDS use: 3216 (289; 2130–4100) $p < 0.0001$ for during vs. before/after E(N)NDS use and $p < 0.0001$ for during tank vs. during disposable E(N)NDS use. <u>Air nicotine concentration sampled at mouth level of non-user, median, mean (SD; range), µg/m^3</u> Disposable, before E(N)NDS use: 0.004, 0.004 (0.001; 0.003–0.005) Tank, before E(N)NDS use: 0.010, 0.011 (0.002; 0.009–0.014) Disposable, during E(N)NDS use: 0.697, 0.717 (0.195; 0.445–0.989) Tank, during E(N)NDS use: 1.833,1.680 (0.379; 1.158–2.047) Disposable, after E(N)NDS use: 0.115, 0.114 (0.009; 0.100–0.124) Tank, after E(N)NDS use: 0.147, 0.145 (0.014; 0.129–0.168) $p < 0.05$ for during vs. before/after E(N)NDS use for both disposable and tank E(N)NDS <u>Nicotine, median accumulation rate (range), ng/100 cm^2/h</u> Surface: 2.1 (0.313–59.7) (disposable), 4.0 (0.562–35.8) (tank); LOQ, 0.3 ng Cloth: 44.4 (disposable), 69.9 (tank); LOQ, 0.5 ng $p < 0.01$ for disposable vs. tank Authors concluded that short-term E(N)NDS use can produce elevated air levels of PM2.5, UFP and nicotine, and accumulation of nicotine on surfaces and clothing. They commented that this could lead to dermal absorption of nicotine and to second-hand inhalation of particles and associated chemicals.
Protano et al. (2017) Evaluation of particulate matter in ambient air in room during use of	SMPs in the range of 5–560 nm	Smooke E-SMART (L) E(N)NDS with Smooke Light e-liquid (9 mg/mL nicotine)	In a 52.7 m^3 test room, 3 x test use sessions at 1-h intervals, during which either 12 puffs E(N)NDS were taken or 1 IQOS was used, or 1 CC or 1 hand-roll CC or 1 cigar or 1 pipe	<u>Peak particle number concentration (PNC)</u> Results were described in the narrative as follows: Peak concentrations ranging from 1.2 x 10^5 to 2.9 x 10^5 particles/cm^3 were reached when combustion occurred. Temporal trends indicated that 1 h was not sufficient time for particle number

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
E(N)NDS, IQOS or CC		IQOS heat-not-burn device used with a Marlboro Balance stick CC (Pall Mall) Hand-rolled CC with Golden Virginia tobacco Italian Toscanello cigar Pipe with tobacco Rome, Italy	was smoked. Smoking sessions were approximately 4 min for all test materials except cigar (30 min) and pipe (45 min). The air exchange rate in the room was 0.67/h. Aerosol measurements made from 5 min before first use session for 200 min Aerosol number size distribution measured by FMPS in the range 5.6–560 nm, 1 s resolution, flow rate 10 L/min	to fall to background level after use of CC/hand-rolled CC. Peak concentrations for IQOS and E(N)NDS devices were below 4.7 x 10^4 particles/cm^3. Overall particle emissions were lower from E(N)NDS than from IQOS, although the transient peak from E(N)NDS was higher. A 1-h time interval was sufficient for particle number to fall to background for E(N)NDS, but not for IQOS. Modelling was performed to estimate doses to passively exposed subjects of different ages. In their summary, with regard non-combustion products, the authors concluded that both non-combustion devices (IQOS and E(N)NDS) emitted SMPs, supporting a ban on their use indoors.
Protano et al. (2018) Evolution of PM1 fraction in emissions from use of different generations of E(N)NDS products, with and without nicotine	PM_1	<u>E(N)NDS products tested</u> 1st generation with 0 or 24 mg/mL nicotine 2nd generation with 0 or 18 mg/mL nicotine 3rd generation with 0 or 9 mg/mL nicotine, various resistance/ voltage settings 4th generation with 0 or 9 mg/mL nicotine, various resistance/ voltage settings Further details of devices, products, e-liquids, and test	In a 52.7 m^3 room, test sessions comprised 12 x 30-s E(N)NDS puffs taken over 5.5 min, by a volunteer user. Aerosol measurements were made from 5 min before until 1 h after the E(N)NDS use session. The air exchange rate in the room was not reported. Aerosol concentrations for size fractions (PM_1, $PM_{2.5}$, PM_4, PM_{10}) were measured by portable, laser-operated aerosol mass analyser (0.1–10 µm) placed at approximately 1.5 m height and 1.5 m from the user.	Results were only reported for PM_1 fraction. The authors noted that a significant increase ($p < 0.001$) in PM_1 levels was detected during compared with before E(N)NDS use for all products and test conditions. In most cases, nicotine-containing e-liquids produced statistically significantly higher PM1 levels than nicotine-free e-liquids. PM_1 emissions were generally highest from 4th generation products and with increasing operating power. <u>Ranges of arithmetic mean and median PM_1 levels measured over all products/ use conditions were</u>: *Before E(N)NDS use* Arithmetic mean:18.33–44.67 µg/m^3; Median: 18.00–43.00 µg/m^3 *During E(N)NDS use* Arithmetic mean: 54.39 µg/m^3 (3rd generation product + nicotine, operated at 1.6 Ω/3.4 V) to 14,887 µg/m^3 (4th generation product + nicotine, operated at 0.4 Ω/80 W ('sub-ohming'); Median: 17 µg/m^3 (3rd generation product + nicotine, operated at

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td></td><td>conditions can be found in Table 1 of the publication by Protano et al. (2018)</td><td></td><td>1.6 Ω/3.4 V) to 3475 µg/m^3 (4th generation product + nicotine, operated at 0.4 Ω/80 W ('sub-ohming')

Authors concluded that PM exposure occurs from the use of all E(N)NDS models, and that the emission of nanoparticles should be investigated further.</td></tr>
<tr><td colspan="5">Evaluation of ambient air and surfaces in real-life settings where E(N)NDS use takes place</td></tr>
<tr><td>Household</td><td></td><td></td><td></td><td></td></tr>
<tr><td>Ballbe et al. (2014)

Evaluation of passive exposure to nicotine in the home from CC smoking or E(N)NDS use</td><td>Nicotine in ambient air

Biomarkers of (passive) nicotine exposure: salivary and urinary cotinine</td><td>Nonsmokers/non-E(N)NDS users living in a home environment where CC smoking (n=25), or E(N)NDS use (n=5), or neither (n=24) took place (non-users exposed ≥ 2 h per day)

All E(N)NDS were tank systems with PG-based liquids

Barcelona, Spain (2011-2012)</td><td>Nicotine was sampled for a period of 1 week in the main family room using a filter hanging from the ceiling (flow rate, 24 x 10^{-6} m^3/min); analysis by GC-MS. The air exchange rate in the room was not reported.

At the end of the 1-week period, saliva and urine samples were collected from nonsmoker/non-E(N)NDS-user volunteers and analysed for cotinine levels; a secondhand smoke questionnaire was administered</td><td>The LOQ for nicotine was 5 ng/filter (equivalent to 0.02 µg/m^3 air concentration over 1 week)

Measured nicotine levels are shown in the table below:
<table>
<tr><th></th><th></th><th>Air nicotine (µg/m^3)</th><th>Non-user salivary cotinine (ng/mL)</th><th>Non-user urinary cotinine (ng/mL)</th></tr>
<tr><td rowspan="2">Control home (n=24)</td><td>Median (IQR)</td><td>0.01 [0.01; 0.05]</td><td>0.05 [0.05; 0.13]</td><td>0.72 [0.55; 1.09]</td></tr>
<tr><td>GM (GSD)</td><td>0.02 (3.51)</td><td>0.07 (1.79)</td><td>0.70 (1.76)</td></tr>
<tr><td rowspan="2">E(N)NDS-use home (n=5)</td><td>Median (IQR)</td><td>0.11 [0.06; 0.32]</td><td>0.24 [0.15; 0.31]</td><td>2.64 [0.70; 4.04]</td></tr>
<tr><td>GM (GSD)</td><td>0.13 (2.4)</td><td>0.19 (2.17)</td><td>1.75 (2.67)</td></tr>
<tr><td rowspan="2">CC-smoker home (n=25)</td><td>Median (IQR)</td><td>1.03 [0.21; 1.99]</td><td>0.32 [0.23; 0.63]</td><td>2.58 [1.13; 4.85]</td></tr>
<tr><td>GM (GSD)</td><td>0.74 (4.05)</td><td>0.38 (2.34)</td><td>2.46 (2.67)</td></tr>
</table>
GSD, geometric standard deviation

Air nicotine, salivary cotinine, and urinary cotinine levels were highly correlated (p < 0.001).
There was a statistically significant difference of all 3 measurements according to type of home (ANOVA < 0.001).
There was a statistically significant difference in measured values for:
Control home vs. E(N)NDS home (for nicotine, salivary cotinine, and urinary cotinine);
Control home vs. CC home (for nicotine, salivary cotinine, and urinary cotinine);</td></tr>
</table>

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
				E(N)NDS home vs. CC home (for nicotine only) Authors concluded that nonsmokers passively exposed to E(N)NDS emissions absorb nicotine.
Bush and Goniewicz (2015) Evaluation of nicotine levels on surfaces (thirdhand exposure) in households with E(N)NDS or CC use, or neither	Nicotine (thirdhand deposit)	Households with regular E(N)NDS use (E(N)NDS, n=8) or CC smoking (CC, n=6) or neither (nonusers, n=8) E(N)NDS users used liquids containing 10–15 mg/mL nicotine, with a range of 50–500 puffs/day indoors. CC smokers smoked 5–40 CC/day indoors. Buffalo City, New York	Surfaces wipe samples taken using KimWipe from floor, wall, window of room where CC smoking /E(N)NDS use occurred or an equivalent room in control home. Details of room ventilation rates were not reported. Nicotine extracted and analysed by GC with a nitrogen phosphorus detector (NPD) (calibration in the range 0.05–100 µg).	Average nicotine concentration (LOQ, 5 µg/m^2): Non-users: 7.2 ± 13.8 µg/m^2 E(N)NDS: 7.7 ± 17.2 µg/m^2 CC: 1303 ± 2676 µg/m^2 Non-users vs. E(N)NDS, $p > 0.05$ Non-users vs. CC, $p < 0.05$ E(N)NDS vs. CC, $p > 0.05$ Authors concluded that nicotine is a common contaminant on indoor surfaces, but that thirdhand exposure to nicotine from E(N)NDS use is low compared to that from CC smoking
Fernandez et al. (2015) Measurement of household air $PM_{2.5}$ concentrations associated with active E(N)NDS use or CC smoking.	$PM_{2.5}$	Households of: Nonsmoker/non-E(N)NDS user (n=2) CC smoker (n=1). E(N)NDS user (Tornado device with Totally Wicked liquid, 18 mg/mL nicotine) (n=1), Barcelona, Spain	Air samples were collected during 1 h within the household at a distance of 2 m while subject actively used E(N)NDS (42 puffs, *ad libitum* use), smoked 3 CC, or neither (nonuser controls). Details of room ventilation were not reported. $PM_{2.5}$ levels were analysed using a TSI SidePak Personal Air Monitor model AM510 (light scattering) and 60-s average concentrations over the 1-h period were plotted. Median and IQR concentrations were reported	Household $PM_{2.5}$ concentration: median [IQR] Nonuser-1: 9.53 [8.32–10.50] µg/m^3 Nonuser-2: 9.36 [8.84–10.40] µg/m^3 E(N)NDS: 9.88 [8.84–11.96] µg/m^3 CC: 572.52 [431.09–747.24] µg/m^3 A plot of 60-s average $PM_{2.5}$ concentrations over the 1-h period showed peak concentration spikes correlated with time points of CC smoking and E(N)NDS puffing (numerical data are not reported, but from the graph, peak $PM_{2.5}$ levels associated with E(N)NDS puffing appear to be in the range of 10–20 µg/m^3, and peak levels associated with smoking a CC reaching 1000 µg/m^3 or more. Authors noted that median levels of $PM_{2.5}$ in the nonusers and

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td></td><td></td><td>for each household.</td><td>E(N)NDS household were below the WHO AQG for long-term exposure of 10 µg/m^3, while the concentration in the CC smoker household was 58-fold that in the E(N)NDS user household.</td></tr>
<tr><td>Car interior</td><td></td><td></td><td></td><td></td></tr>
<tr><td>Schober et al. (2019)

Measurement of levels of PM, VOCs, and carbonyls in vehicles durings use of E(N)NDS, heated tobacco product, or CC.</td><td>$PM_{2.5}$

VOCs (including PG and nicotine)

Carbonyls</td><td>E(N)NDS: SubTwin Neo tank model; tobacco-flavoured e-liquid containing 18 mg/mL nicotine

Heated tobacco product: IQOS with Bronze Label Heet, 0.5 mg/Heet

CC: Marlboro Red, 0.8 mg nicotine/CC

7 vehicles: large (5–7 m^3 interior volume; n=2), medium (3–4 m^3; n=3), small (2–3 m^3; n=2).

2 occupants per vehicle (driver; front passenger who used the test product)

Munich, Germany</td><td>7 tests were conducted per vehicle as follows:
Drive 1: No smoking, passenger window (PW): 5 cm open; Drive 2: Passenger used 2 heated tobacco products, PW: 5 cm open; Drive 3: Passenger used 2 heated tobacco products, PW: 2 cm open; Drive 4: Passenger continually used, PW: 5 cm open; Drive 5: Passenger continually used E(N)NDS, PW: 2 cm open; Drive 6: Passenger smoked 2 CC, PW: 5 cm open; Drive 7: Passenger smoked 2 CC, PW: 2 cm open

Sampling was performed in the rear passenger seat.

$PM_{2.5}$ were measured continually with an optical laser aerosol spectrometer (15 particle size fractions from 300 nm to > 20 µm) and nanoparticle counter (25–300 nm) (flow rate, 1.2 L/min).

VOCs were sampled via thermal desorption tubes (1 L or 2 L samples) with Tenax TA sorbent (flow rate, 55 or 110 mL/min), and analysed by GC/MS.

Aldehydes and ketones were collected via DNPH cartridge</td><td>LODs were 2 µg/m^3 for formaldehyde and acetaldehyde; 1.5 µg/m^3 for butyraldehyde, acetone, acrolein, 2-butanone.
LODs for other measurements were not reported.

Findings were not significantly correlated with either the car size or the extent to which the windows were open.

Ranges of mean values measured over all tests for the 7 vehicles:
<table>
<tr><th></th><th>Control</th><th>IQOS</th><th>E(N)NDS</th><th>CC</th></tr>
<tr><td>PNC (25–300 nm) (N/cm^3)</td><td>8434 – 73,791</td><td>16,726–123,655</td><td>10,248–73,954</td><td>24,319–236,167</td></tr>
<tr><td>PNC (300 nm – > 2 µm) (N/cm^3)</td><td>14–41</td><td>10–144</td><td>28–2145</td><td>288–9048</td></tr>
<tr><td>$PM_{2.5}$ (µg/m^3)</td><td>4–11</td><td>4–34</td><td>8–490</td><td>64–1988</td></tr>
<tr><td>PG (µg/m^3)</td><td>< LOD</td><td>< LOD</td><td>< LOD–762</td><td>< LOD–94</td></tr>
<tr><td>Nicotine (µg/m^3)</td><td>< LOD</td><td>< LOD–12</td><td>< LOD–10</td><td>< LOD–140</td></tr>
</table>
Levels of aldehydes and ketones increased substantially over background during CC smoking, but not during use of E(N)NDS or IQOS.

Authors concluded that, overall, CC, E(N)NDS, and IQOS are avoidable sources of indoor air pollutants, in particular $PM_{2.5}$ and nicotine, and to protect the health of occupants, in particular children and pregnant women, these products should not be used in cars.</td></tr>
</table>

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td></td><td></td><td>(flow rate, 1.115 L/min; 20 L air collected), stored in a refrigerator, then eluted and detected by HPLS/UV analysis</td><td></td></tr>
<tr><td colspan="2">Public buildings and vaping conventions</td><td></td><td></td><td></td></tr>
<tr><td>Soule et al. (2017)

Measurement of $PM_{2.5}$ levels associated with an indoor E(N)NDS event</td><td>$PM_{2.5}$</td><td>2-day E(N)NDS event in a large room in a hotel.

Summer 2015, Minnesota, USA.</td><td>$PM_{2.5}$ levels were measured 1] in the event room (4023 m^3), 2] in an indoor restaurant within the event hotel (596 m^3), and 3] in an indoor restaurant in a different hotel adjacent to the venue (942 m^3). CC smoking was prohibited in all 3 rooms. Details of room ventilation were not reported.

1-min average measurements were made for sessions of 30 min using 2 TSI Sidepak AM510 Personal Aerosol monitors (devices 1 and 2) (particle size ≥ 100 nm).

Measurements were made the day before the E(N)NDS event (1 session), during day 1 of the event (6 sessions over approximately 4 h), and 17 h after the event (1 session).</td><td>Active E(N)NDS use was observed to be performed by:
59–86 users at the 6 monitored time-points in the event room during the event,
0 users in the event room before or after the event,
2 users in the event-hotel restaurant,
0 users in the non-event-hotel restaurant.
Measurements were similar between devices 1 and 2.

Measured $PM_{2.5}$ concentrations:
<table>
<tr><td></td><td colspan="3">Range of median $PM_{2.5}$ measurements over time points and devices) (µg/m^3)</td></tr>
<tr><td></td><td>Before event</td><td>During event</td><td>After event</td></tr>
<tr><td>Event room</td><td>1.92–3.20</td><td>311.68–818.88</td><td>12.80–15.52</td></tr>
<tr><td>Event-hotel restaurant</td><td>1.60–2.56</td><td>4.80</td><td>NR</td></tr>
<tr><td>Non-event-hotel restaurant</td><td>NR</td><td>5.76</td><td>NR</td></tr>
</table>
NR, not reported

The average $PM_{2.5}$ concentration during the event over the 6 measurement sessions was 595.31 µg/m^3 (median), 607.12 µg/m^3 (mean).

Authors commented that E(N)NDS use can generate fine PM in high concentrations during ‘natural use conditions’ in indoor environments. Levels were noted to be 60-fold those measured in the home of an E(N)NDS user, and were also higher than levels that have been reported for hookah cafés (369–384 µg/m^3) and bars that allow indoor smoking (119 µg/m^3).

Authors concluded that indoor E(N)NDS use exposes non-users to second-hand E(N)NDS aerosol, and that regulatory bodies should consider establishing policies that prohibit E(N)NDS use in places where CC smoking is prohibited.</td></tr>
</table>

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr>
<td>Chen et al. (2017)

Measurement of indoor air concentrations of PM, VOCs, and nicotine at a vaping convention</td>
<td>PM_{10}

VOCs

NO_2

Nicotine</td>
<td>Indoor vaping convention

Venue size, 55×49×5m^2 (13,475m^3)

Number of attendees estimated to be in the range of 75–600 at any single observation time

During the convention, specific events included: 2 trick, ‘artistic plume’ competitions; 1 big plume size competition

April 2016, Maryland, USA</td>
<td>Real-time and integrated sampling via 3 teams of 2 people carrying backpacks around the convention during 7 h; outdoor air was sampled 33 min before entering the venue and 2 more times during the sampling period. Total sampling time was 58 min (outdoor) and 342 min (indoor). Time-integrated samples reflect a combination of indoor and outdoor sampling (total 400 min).

PM_{10} levels were measured with personal aerosol particle monitors (SidePak AM510, at a flow rate of 1.7 L/min); integrated sampling of PM_{10} was conducted with personal environmental monitors (PEM) with PVC filters, at a flow rate of 4 L/min;
Nicotine was collected with 37 mm glass fibre filters impregnated with sodium bisulphate and assembled in modified polystyrene sampling cassettes, covered with a porous diffusion membrane. Two samplers were used in passive mode (flow rate, 25 mL/min) and 2 were connected to pumps for active mode (flow rate, 3 L/min). Nicotine was extracted in heptane with 1% triethylamine and analysed by GC with flame thermoionic detector. Airborne nicotine concentrations were calculated by dividing ng nicotine on filter by volume of air sampled (m^3);</td>
<td>Mean temperature and relative humidity: 77 ºF, 38% (indoor), 74 ºF, 39% (outdoor)

At peak occupancy, visibility was very low and the smell was strong.

Measured concentrations from real-time sampling:
<table>
<tr><th></th><th colspan="4">Median (IQR)</th></tr>
<tr><th></th><th>Outdoor</th><th>Indoor</th><th>Trick competition</th><th>Plume size competition</th></tr>
<tr><td>PM_{10} (µg/m^3)</td><td>228 (14–8,468)</td><td>11,327 (9050–13,182)</td><td>7,987 (9,425–11,727)</td><td>11,928 (11,245–14,298)</td></tr>
<tr><td>VOCs (ppm)</td><td>0.06 (0.05–0.1)</td><td>0.13 (0.11–0.19)</td><td>0.10 (0.10–0.10)</td><td>0.20 (0.19–0.25)</td></tr>
<tr><td>NO_2 (ppm)</td><td>0.25 (0.1–0.32)</td><td>0.09 (0.06–0.11)</td><td>0.11 (0.05–0.13)</td><td>0.06 (0.05–0.09)</td></tr>
<tr><td>CO_2 (ppm)</td><td>448 (346–715)</td><td>870 (740–1,035)</td><td>737 (668–766)</td><td>1,051 (1,003–1,150)</td></tr>
</table>
Measured concentrations of time-integrated samples (58 min outdoor + 342 min indoor):
<table>
<tr><th></th><th colspan="3">Measurement</th></tr>
<tr><td>Sampling technique (sample)</td><td>Active (1)</td><td>Active (2)</td><td>Passive (1,2)</td></tr>
<tr><td>PM_{10} (µg/m^3)</td><td>8,850</td><td>8,429</td><td>Not reported</td></tr>
<tr><td>Nicotine (µg/m^3)</td><td>109.2</td><td>140.2</td><td>< LOD*</td></tr>
</table>
*The LOD value for nicotine was not stated in the report

PM_{10} and VOC concentrations were correlated with CO_2 concentrations during all sampling periods (r = 0.76, p < 0.0001 for PM_{10}; r = 0.81, p < 0.0001 for VOCs).
NO_2 concentrations were negatively correlated with other measurements (higher outdoor than indoor).

Authors noted that the mean measured nicotine concentration (124.7 µg/m^3) was of a similar magnitude to that reported historically for clubs and nightclubs when smoking CCs was permitted (94.5 µg/m^3), and 88-fold higher than an average (1.42 µg/m^3) that was reported for waterpipe cafes in Baltimore. They also commented that the venue was probably not ventilated adequately. For example, ventilation was deliberately reduced during competitions (organisers requesting closing of doors and windows) to increase the visibility of the generated plumes. They</td>
</tr>
</table>

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td></td><td></td><td>Total VOCs, CO_2, NO_2, temperature and RH were measured using a multi-gas monitor

Information on the estimated number of attendees, visibility, and smell was recorded every 15 min</td><td>considered that the correlation between PM_{10}, TVOC, and CO_2 concentrations supports the conclusion that exhaling was the major source of PM_{10} and TVOCs.

Authors concluded that E(N)NDS aerosol in a vaping convention that congregates many E(N)NDS users is a major source of PM_{10}, air nicotine, and VOCs, impairing indoor air quality, and that the findings also raise occupational concerns for E(N)NDS vendors and other venue staff workers.</td></tr>
<tr><td>Johnson et al. (2018)

Measurement of PG, nicotine, formaldehyde, acetaldehyde, and acrolein levels in air at vaping conventions</td><td>PG

Nicotine

Formaldehyde

Acetaldehyde</td><td>4 indoor vaping events in the USA:

<u>Event 1</u>
Daytona Beach, Florida, April 2016: 42,146 sq.ft convention centre, ceiling height 45 ft, ventilation and air conditioning

<u>Event 2</u>
Athens, Georgia, August 2016: 5100 sq.ft concert hall, 35 ft ceiling height

<u>Event 3</u>
Chattanooga, Tennessee, October 2016: 36,000 sq.ft convention centre, 30 ft ceiling height, ventilation and air conditioning

<u>Event 4</u>
Atlanta, Georgia, March 2017:</td><td>Air was sampled for an average of 5.3 h at each site: 1] in the empty venue; 2] during a crowded, non-vaping convention; 3] during a crowded vaping convention. Ventilation rates in the venues were not reported. However, the authors noted that venues for Events 1 and 3 were modern buildings with high ceilings typical of a large convention center (approximately 30–45 feet [9–14 m]) and a noticeable air draft that indicated a highly functioning heating, ventilation, and air conditioning system. Event 4 venue was a tradeshow with lower ceilings (approximately 13 feet [4 m]). Event 2 venue was a small concert hall with 2 levels of balconies and a high ceiling (approximately 35 feet [11 m]). All venues had doors open during the events.

Air sampling was conducted using active air sampling pumps carried in backpacks (n=21). Three types of sampling pumps</td><td>Reporting limits: PG, 20 µg/sample; nicotine, 0.1 µg/sample; formaldehyde, 0.1 µg/sample; acetaldehyde, 0.5 µg/sample; acrolein, 2.0 µg/sample

Low concentrations of nicotine and high concentrations of PG were present only during E(N)NDS events.
Formaldehyde and acetaldehyde concentrations during E(N)NDS events were comparable to concentrations present when the venue was empty and during non-E(N)NDS events.
Acrolein was not detected.

Detected levels of PG, nicotine, formaldehyde, and acetaldehyde:
<table>
<tr><th></th><th></th><th>Empty venue</th><th>Crowded venue</th><th>Crowded venue E(N)NDS convention</th></tr>
<tr><td></td><td></td><td colspan="3">(µg/m^3)</td></tr>
<tr><td>PG</td><td>Median</td><td>ND</td><td>ND</td><td>305</td></tr>
<tr><td></td><td>25th, 75th</td><td>ND</td><td>ND</td><td>230.0, 410.0</td></tr>
<tr><td></td><td>Range</td><td>ND</td><td>ND</td><td>< LOD*–490</td></tr>
<tr><td>Nicotine</td><td>Median</td><td>ND</td><td>ND</td><td>1.1</td></tr>
<tr><td></td><td>25th, 75th</td><td>ND</td><td>ND</td><td>< 0.37, 1.8</td></tr>
<tr><td></td><td>Range</td><td>ND</td><td>ND</td><td>< 0.36–2.2</td></tr>
<tr><td>Formaldehyde</td><td>Median</td><td>12.5</td><td>10.5</td><td>12.0</td></tr>
<tr><td></td><td>25th, 75th</td><td>11.0, 16.5</td><td>9.6, 17.0</td><td>10.0, 29.0</td></tr>
<tr><td></td><td>Range</td><td>9.9–20</td><td>9.0–45</td><td>6.5–59</td></tr>
<tr><td>Acetaldehyde</td><td>Median</td><td>3.5</td><td>15.5</td><td>9.7</td></tr>
<tr><td></td><td>25th, 75th</td><td>2.1, 4.2</td><td>8.0, 24.0</td><td>7.0, 14.0</td></tr>
<tr><td></td><td>Range</td><td>< 2.7–4.2</td><td>4.3–29</td><td>5.3–18</td></tr>
</table>
ND, not detected; * LOD value not stated

Authors noted/concluded that:</td></tr>
</table>

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
		205,000 sq.ft exhibition/tradeshow, 13 ft ceiling height	were used, AirChek XR5000 & 2000, SKC Inc; Escort Elf, Zefon International Inc. PG was sampled according to NIOSH Method 5223 on XAD-7 OVS sorbent tubes (sampler flow rate, 800 or 1000 mL/min), using GC with a flame ionisation detector (FID) Nicotine was sampled according to NIOSH Method 2551 using XAD-4 sorbent tubes (sampler flow rate, 1000 mL/min), and analysed by GC-NPD. Formaldehyde and acetaldehyde were sampled according to EPA Compendium Method TO-11A, using Sep-Pak DNPH-silica plus short cartridges (sampler flow rate, 800 mL/min), and analysed using HPLC. Acrolein was sampled according to OSHA Method 52 using XAD-2 sorbent tubes (sampler flow rate, 100 mL/min), and analysed by GC-NPD.	PG concentrations were similar to those reported in other studies of E(N)NDS use. Levels were generally much higher than the ATSDR intermediate duration (> 14–364 days) inhalation MRL for PG of 28.01 µg/m^3. Authors felt that, given that E(N)NDS are emerging and that PG exposures measured in this study were much higher than the ATSDR intermediate MRL, further research is needed to determine if a new health guidance value for PG is warranted. Nicotine concentrations were similar to the range reported in the literature (< 0.01–7.00 µg/m^3). All nicotine concentrations were below the occupational OSHA PEL and NIOSH REL (0.5 mg/m^3). Levels of formaldehyde at crowded events with or without E(N)NDS exceeded the NIOSH REL 8-h TWA of 19.65 µg/m^3 for formaldehyde. Levels of acetaldehyde at crowded events with or without E(N)NDS were below the OSHA 8-h TWA PEL of 360 mg/m^3, but exceeded the EPA RfC of 9 µg/m^3. Authors concluded that the results of this study thus do not provide convincing evidence that second-hand E(N)NDS aerosol produced by human subjects in a natural environment contains elevated levels of formaldehyde or acetaldehyde. Finally, the authors commented that this study used sorbent tubes, which are only designed to capture chemicals in the gas phase, thus chemicals present in the particle phase may not have been measured.
Khachatoorian et al. (2018) Measured levels of nicotine, other alkaloids, and TSNAs deposited on materials in a shop adjacent to a vaping	Nicotine and related compounds	The test ('field') site was a basement floor of a 2-story shopping centre in a metropolitan area: Suite 1, an actively trading shop (37 m^2); Suite 2, a shop that was seldom used	Cotton towels, paper towels, terrycloth towels and 2 air filters (3M high performance 20×25×1 Filtrete air filter and Rabbit Air Classic BioGS Replacement HEPA filter) were placed in the field site for short-term or long-term exposures. Short-term exposure samples	LOQ values were: nicotine 2 ng/mL; cotinine 1 ng/mL, n-formylnornicotine 1 ng/mL, bipyridine 1 ng/mL, nicotelline 0.2 ng/mL, myosmine 1 ng/mL, N-nitrosoanatabine (NAB) 1 ng/mL, 4-(methylnitrosamino)−1-(3-pyridyl)−1-butanone (NNK) 0.1 ng/mL, N'-nitrosonornicotine (NNN) 0.1 ng/mL, N-nitrosoanatabine (NAT) 0.2 ng/mL, 1-(N-methyl-N-nitrosamino)−1-(3-pyridinyl)−4-butanal (NNA) 1 ng/mL. Nicotine, other alkaloids, and TSNAs were all detected in extracts

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
shop.		(37 m^2); Suite 3, a vape shop in which active vaping occurred (28 m^2). The air ventilation system allowed air to be recirculated from Suites 2 and 3 back to Suite 1, with an air filter system in the return vent to suite 1. Tests were also conducted in the home of a nonsmoker and in another area of the shopping centre. Air flow through the site was reported as approximately 39.67–48.39 m^3/min, and there was an air filter in the return vent from the vaping shop to the actively trading shop where measurements were performed.	were collected after 1, 4, and 8 days. Long-term exposure samples were collected after 1, 2, and 3 months. Control fabrics (terrycloth towel) were exposed in a hallway on a separate HVAC system outside the field site for 1 and 3 days, and 1 week. Additional control samples of fabrics (terrycloth towels) were collected from a nonsmoker home in the same community after exposure for 1 and 4 days, and 1 week. Unexposed samples of each type of fabric and both air filters were also used as controls. Exhaled aerosol residue was extracted by incubation of samples in cell culture medium, then filtered through 0.22 µm filters. Nicotine, nicotine derivatives, and TSNAs were quantified by LC-MS/MS.	of cotton towels and paper towels from Suite 1 (actively trading shop). No results were presented for Suites 2 (seldom-used shop) or 3 (vape shop). Nicotine was reported to be the most abundantly detected compound, found on 1-day samples (example level, 154 ng/g fabric), and with concentration increasing with exposure time (maximum cited concentration, 23,260 ng/g fabric). Nicotine was also identified in test filters. Nicotine was mostly not detected in the home of a nonsmoker. Tobacco alkaloids were found most frequently in paper towel extracts. Most alkaloids were also identified in test filters. TSNAs were most abundant in paper towel extracts, with concentrations of NNK and NNN increasing over time, while NAT was only found in short-term paper towel samples. NNN and NNK were detected in 1 of the 2 test filters. Authors concluded that in a multi-tenant retail building, chemicals in E(N)NDS aerosol travelled from a vape shop into an adjacent business where they deposited forming E(N)NDS exhaled aerosol residue, and that regulatory agencies and tenants occupying such buildings should be aware of this potential environmental hazard.
Biomarkers of bystander exposure to E(N)NDS aerosols				
Flouris et al. (2012) Effect of exposure to CC smoke or E(N)NDS aerosol on complete blood count (CBC)	Biomarkers: white and red blood cell parameters (active and passive exposure)	Adult volunteers (male and female): 15 CC smokers (≥ 15 CC per day; average age 36.8 y); 15 never-smokers (average age 28.9 y)	3 experimental sessions per group, interspersed with 7-day washout, as follows: Smokers ('active' sessions): Control (AS_{CON}) – 'smoked' an unlit CC for 30 min; CC smoking (AS_{TOB}) – smoked 2	CC smokers AS_{CON} – no changes over baseline AS_{TOB} – increased WBC count, lymphocyte count, granulocyte count ($p < 0.05$ at 0 h and 1 h post exposure compared with baseline); no change in other variables $AS_{E\text{-}CIG}$ – no changes over baseline

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
parameters		CC: subject's own brand (CC smokers) or 'various popular brands' (passive exposure of never-smokers). E(N)NDS: Giant Nobacco G.P., Greece, with Nobacco USA Mix e-liquid ('tobacco taste; >60% PG, <5% linalool, <5% tobacco essence, <1% methyl vanilin, 11 mg/mL nicotine).	x CC within 30 min; E(N)NDS use ($AS_{E\text{-}CIG}$) - used. Never-smokers (60 m^3 chamber; 60-min sessions): Control (PS_{CON}) – room air; Passive CC smoke (PS_{TOB}) – tobacco smoke at bar/restaurant levels (23 ± 1 ppm CO); Passive E(N)NDS aerosol ($PS_{E\text{-}CIG}$) – 'bar/restaurant' levels of machine-generated E(N)NDS aerosol were pumped into the chamber at an air flow rate of 4 L/min (no further details given). Order of sessions was assigned randomly. Blood samples were collected before, immediately after (0 h), and 1 h after each session. Blood evaluations WBC count, red blood cell (RBC) count, haemoglobin, haematocrit, mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), MCH concentration, RBC distribution width, platelet count, mean platelet volume, platelet haematocrit, platelet distribution width.	Never-smokers PS_{CON} – no changes over baseline. PS_{TOB} – increased WBC count, lymphocyte count, granulocyte count ($p < 0.05$ at 0 h and 1 h post exposure compared with baseline); no change in other variables. $PS_{E\text{-}CIG}$ – no changes over baseline. Results were presented for WBC count, lymphocyte count, and granulocyte count only. There appears to be substantial variability in baseline values measured before the test sessions, both within and between groups (see Fig. 1 of the publication). Authors concluded that active and passive E(N)NDS exposures do not lead to alterations in CBC indices.
Flouris et al. (2013) Effect of exposure to CC smoke or E(N)NDS aerosol on serum cotinine and lung function parameters	Biomarkers of exposure and effect: Exhaled CO Exhaled NO Serum cotinine	Adult volunteers (male and female): 15 CC smokers (≥ 15 CC per day, 23.5–54 years); 15 never-smokers (18–57 years)	3 experimental sessions per group, interspersed with 7-day washout, as follows: Smokers ('active' sessions AS): Control (AS_{CON}) – 'smoked' an unlit CC for 30 min; CC smoking (AS_{TOB}) – smoked 2	Serum cotinine after test session AS_{TOB} 60.6 ± 34.3 ng/mL $AS_{E\text{-}CIG}$ 61.3 ± 36.6 ng/mL PS_{TOB} 2.4 ± 0.9 ng/mL $PS_{E\text{-}CIG}$ 2.6 ± 0.6 ng/mL

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td>Lung function parameters

(active and passive exposure)</td><td>CC: subject's own brand (CC smokers) or 'various popular brands' (passive exposure of never-smokers).

E(N)NDS: Giant Nobacco G.P., Greece device, with Nobacco USA Mix e-liquid ('tobacco taste; >60% PG, <5% linalool, <5% tobacco essence, <1% methyl vanilyn, 11 mg/mL nicotine)</td><td>x CC within 30 min;
E(N)NDS use ($AS_{E\text{-}CIG}$) – used E(N)NDS for 30 min (median 11 puffs, range 3–14 puffs)

<u>Never-smokers</u> (60 m^3 chamber; 60-min sessions):
Control (PS_{CON}) – room air;
Passive CC smoke (PS_{TOB}) – tobacco smoke at bar/ restaurant levels (23 ± 1 ppm CO)
Passive E(N)NDS aerosol ($PS_{E\text{-}CIG}$) – 'bar/restaurant' levels of E(N)NDS aerosol;
Passive air was pumped into the chamber at an air flow rate of 4 L/min.

Serum cotinine levels, lung function parameters, exhaled CO and NO were assessed.</td><td><u>Other parameters</u>
FEV_1/FVC was significantly reduced after $AS_{TOB.}$ Exhaled CO was significantly increased after AS_{TOB} and $PS_{TOB.}$ These parameters were not significantly changed after either $AS_{E\text{-}CIG}$ or $PS_{E\text{-}CIG}$.

Authors concluded that E(N)NDS (containing nicotine) produce a similar 'nicotinergic impact' but smaller changes on lung function than CC.</td></tr>
<tr><td>Ballbe et al. (2014)

Evaluation of passive exposure to nicotine in the home from CC smoking or E(N)NDS use</td><td>Nicotine in ambient air

Biomarkers of (passive) nicotine exposure: salivary and urinary cotinine</td><td>Nonsmokers/non-E(N)NDS users living in a home environment where CC smoking (n=25), or E(N)NDS use (n=5), or neither (n=24) took place (non-users exposed ≥ 2 h per day)

All E(N)NDS were tank systems with PG-based liquids

Barcelona, Spain (2011-2012)</td><td>Nicotine was sampled for a period of 1 week in the main family room using a filter hanging from the ceiling (flow rate, 24 x 10^{-6} m^3/min); analysis by GC-MS

At the end of the 1-week period, saliva and urine samples were collected from nonsmoker/non-E(N)NDS-user volunteers and analysed for cotinine levels; a secondhand smoke questionnaire was administered</td><td>The LOQ for nicotine was 5 ng/filter (equivalent to 0.02 µg/m^3 air concentration over 1 week)

Measured nicotine levels are shown in the table below:
<table>
<tr><th></th><th></th><th>Air nicotine (µg/m^3)</th><th>Non-user salivary cotinine (ng/mL)</th><th>Non-user urinary cotinine (ng/mL)</th></tr>
<tr><td rowspan="2">Control home (n=24)</td><td>Median (IQR)</td><td>0.01 [0.01; 0.05]</td><td>0.05 [0.05; 0.13]</td><td>0.72 [0.55; 1.09]</td></tr>
<tr><td>GM (GSD)</td><td>0.02 (3.51)</td><td>0.07 (1.79)</td><td>0.70 (1.76)</td></tr>
<tr><td rowspan="2">E(N)NDS-use home (n=5)</td><td>Median (IQR)</td><td>0.11 [0.06; 0.32]</td><td>0.24 [0.15; 0.31]</td><td>2.64 [0.70; 4.04]</td></tr>
<tr><td>GM (GSD)</td><td>0.13 (2.4)</td><td>0.19 (2.17)</td><td>1.75 (2.67)</td></tr>
<tr><td rowspan="2">CC-smoker home (n=25)</td><td>Median (IQR)</td><td>1.03 [0.21; 1.99]</td><td>0.32 [0.23; 0.63]</td><td>2.58 [1.13; 4.85]</td></tr>
<tr><td>GM (GSD)</td><td>0.74 (4.05)</td><td>0.38 (2.34)</td><td>2.46 (2.67)</td></tr>
</table></td></tr>
</table>

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr><td></td><td></td><td></td><td></td><td>Air nicotine, salivary cotinine, and urinary cotinine levels were highly correlated ($p < 0.001$).
There was a statistically significant difference of all 3 measurements according to type of home (ANOVA < 0.001).
There was a statistically significant difference in measured values for:
Control home vs. E(N)NDS home (for nicotine, salivary cotinine, and urinary cotinine);
Control home vs. CC home (for nicotine, salivary cotinine, and urinary cotinine);
E(N)NDS home vs. CC home (for nicotine only)

Authors concluded that nonsmokers passively exposed to E(N)NDS emissions absorb nicotine.</td></tr>
<tr><td>Melstrom et al. (2018)

Measurement of cotinine levels in E(N)NDS non-users with second-hand exposure to nicotine-containing E(N)NDS</td><td>Cotinine (saliva, serum, urine)</td><td>Disposable E(N)NDS (choice of blu or Fling) (12–20.5 mg/mL nicotine)

iTaste variable-voltage tank-style E(N)NDS (14.5–15.5 mg/mL nicotine)</td><td>52.6 m^3 room. Mean air exchange rate, approximately 5/h.

2 x 2-h E(N)NDS use test sessions: 3 experienced E(N)NDS users used the test product ad libitum while 6 non-users were also present in the room.

Blood, urine, saliva samples collected from non-users before and during 6 h after passive exposures.

Analysis of cotinine in serum, saliva, and urine by CDC standard methods.</td><td>[Air nicotine levels measured at mouth-level of non-users are summarised in Melstrom et al. (2017)].

Estimated total mass of nicotine consumed during a session.

Disposable E(N)NDS: 14.3 mg
Tank-style E(N)NDS: 26.2 mg

Baseline cotinine levels showed substantial variation between non-users. In general, for non-users with low baseline cotinine, levels increased after second-hand E(N)NDS exposure, but for non-users with higher baseline cotinine levels, values did show further increases after second-hand E(N)NDS exposure.

Median changes in cotinine levels are shown in the table below:
<table>
<tr><td></td><td colspan="3">Median change in cotinine</td></tr>
<tr><td></td><td>Serum (ng/mL)</td><td>Saliva (ng/mL)</td><td>Urine (ng/mg creatinine)</td></tr>
<tr><td>Disposable</td><td>0.007</td><td>0.033</td><td>0.316</td></tr>
<tr><td>Tank-style</td><td>0.041</td><td>0.060</td><td>0.948</td></tr>
</table>
Authors concluded that non-users can systemically absorb nicotine following acute exposure to second-hand E(N)NDS aerosol.</td></tr>
</table>

<table>
<tr><th>Authors / study</th><th>Compound / species / index measured</th><th>Test materials / subjects / setting</th><th>Experimental methods</th><th>Results / author comments and conclusions</th></tr>
<tr>
<td>Martinez-Sanchez et al. (2015)

Evaluation of levels of the NNK metabolite, NNAL (4-(methylnitrosamino)-1-(3-pyridyl)-1-butanol) in urine of subjects exposed to firsthand or to secondhand E(N)NDS aerosol</td>
<td>NNAL</td>
<td>Non-users of tobacco products living in a home environment where CC smoking (n=25), or E(N)NDS use (n=6), or neither (n=24) took place (non-users exposed ≥ 2 h per day). Non-users had self-reported lack of exposure to these products in other life settings. Details of ventilation in the households were not reported.

6 E(N)NDS users in the households described above.

E(N)NDS types (e-liquids) used in the 6 homes were as follows:

1. Cigalike (menthol flavour, 11 mg/mL nicotine)

2. e-Go (Marlboro flavour, 24 mg/mL nicotine)

3. e-Go (menthol flavour, 18 mg/mL nicotine)</td>
<td>Urine samples were obtained from and analysed by LC-MS/MS for NNAL concentration in pg/mL, adjusted for creatinine excretion (LOQ, 0.25 pg/mg in 5 mL urine for 1 mg/mL creatinine excretion)</td>
<td>Percentage of non-user urine samples by household type with quantifiable level of NNAL

Control homes: 29.2%
E(N)NDS homes: 66.7%
CC homes: 76%

Median (IQR) urinary NNAL concentrations (pg/mL) by household type:

Control homes (non-users): 0.33 (0.16–0.51)
E(N)NDS homes (non-users): 0.55 (0.26–2.94)
CC homes (non-users): 0.46 (0.29–1.11)

Data for individual E(N)NDS use households:

<table>
<tr><th></th><th colspan="6">Household</th></tr>
<tr><th></th><th>1</th><th>2</th><th>3</th><th>4</th><th>5</th><th>6</th></tr>
<tr><td>Estimated nicotine consumed per day by E(N)NDS user (mg)</td><td>62.7</td><td>48</td><td>180</td><td>27</td><td>19.2</td><td>12.85</td></tr>
<tr><td>E(N)NDS user urinary NNAL concentration*, (pg/mL)</td><td>0.37</td><td>0.33</td><td>6.1</td><td>0.42</td><td>5.3</td><td>9.4</td></tr>
<tr><td>non-E(N)NDS-user urinary NNAL concentration*, (pg/mL)</td><td>< 0.56</td><td>< 0.44</td><td>3.0</td><td>0.44</td><td>0.67</td><td>2.9</td></tr>
</table>
*Adjusted for urinary creatinine

Spearman’s correlation of NNAL in urine of E(N)NDS user and non-user in the same household was 0.943 (p = 0.005).

Authors concluded that NNAL was detected in urine samples from people exposed to second-hand aerosol from E(N)NDS, with the suggestion that the findings should be evaluated in studies with larger sample sizes.</td>
</tr>
</table>

Authors / study	Compound / species / index measured	Test materials / subjects / setting	Experimental methods	Results / author comments and conclusions
		4. e-Go (Keen Tobacco flavour, 9 mg/mL nicotine) 5. e-Go (mint flavour, 6 mg/mL nicotine) 6. e-Go (mint flavour, 6 mg/mL nicotine) Details of CC products used were not described. 2012, Barcelona, Spain		

COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT (COT)
Potential toxicological risks from electronic nicotine (and non-nicotine) delivery systems (E(N)NDS – e-cigarettes). Paper 9: Bystander exposure.

US EPA (2008). Reregistration Eligibility Decision (RED) Document for Nicotine (https://archive.epa.gov/pesticides/reregistration/web/pdf/nicotine_red.pdf, accessed 04/03/2019).

COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT (COT)
Potential toxicological risks from electronic nicotine (and non-nicotine) delivery systems (E(N)NDS – e-cigarettes). Paper 9: Bystander exposure.

EFSA STATEMENT. Potential risks for public health due to the presence of nicotine in wild mushrooms. Issued on 07 May 2009) (https://www.efsa.europa.eu/en/efsajournal/pub/rn-286, accessed 04/03/2019).