Supporting Information

Detection of Highly Reactive Free Radicals in Electronic Cigarette Aerosols

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Materials and Methods

Electronic Cigarette (EC): For testing, popular brands of EC batteries (eGo-ce4 3.3 V and Tesla 3.0-6.0 V) and cartomizer (SmokTech: XXL, 1.5 ohms resistance, dual heating coils) were selected and obtained online (zainy.net).

Eliquids: The ingredient list for eliquids included propylene glycol(PG), glycerol (Gly), water and undisclosed flavorings. Three eliquids were selected for testing: 1. (eliq1): 36 mg/ml nicotine, menthol flavor, PG:Gly ratio 70:30 (US Menthol, Life Smoke); 2. (eliq2) 6 mg/ml nicotine, citrus flavor, PG:Gly ratio 10:90 (Citrus stream, Pitbull); 3. (eliq3) 36 mg/ml nicotine, tobacco flavor, PG:Gly ratio 60:40(US Camel, Life Smoke). Cartomizers were filled with 1.5 ml of eliquids prior to each experiment and were used for only one type of liquid. To test if eliquid solvents alone could yield radicals under these test conditions, solutions containing either 90% propylene glycol (Fisher Scientific) and 10% distilled water or 90% glycerol (Fisher Scientific) and 10% distilled water were used.

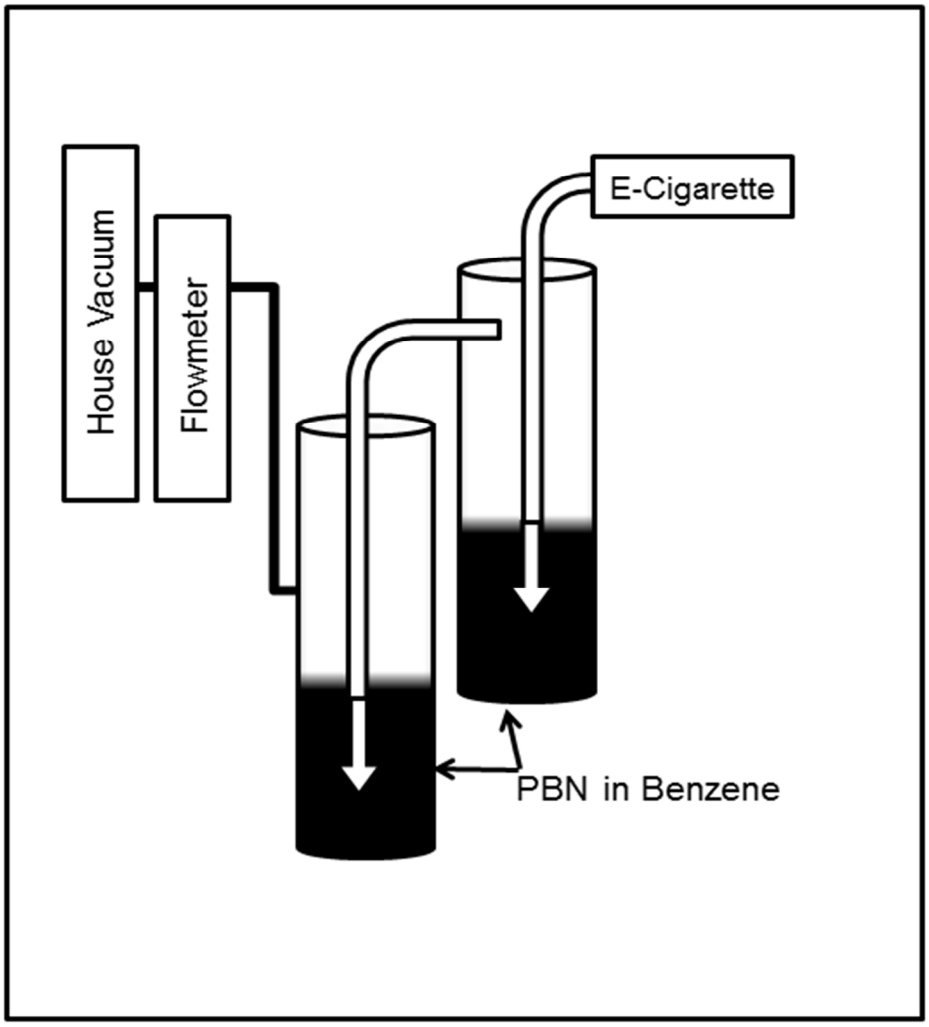
Generation of EC Aerosols: ECs were connected to house vacuum via a flow meter and aerosols were generated by manually turning on the fully charged EC battery at voltages adjusted to either 3.3 V or 5.0 V. Puffs were simulated by turning off/on the flow (vacuum) and voltage (battery) simultaneously. Topography parameters used were based on human usage conditions as described previously[1] and were as follows: puff duration, 5 sec; puff interval, 20 sec; flow rate, 500 ml/min: and number of puffs, 40 per experiment.

Spin Trapping of Free Radicals in EC Aerosols: The nitrone spin trap phenyl-N-tert-butylnitrone (PBN) has been used extensively for the detection of reactive radicals in cigarette smoke.[2] EC aerosols generated as described above were passed through two impingers containing a benzene solution

(Sigma-Aldrich) with the spin trap PBN (Sigma-Aldrich). The two 25 ml impingers were placed in series between the vacuum source and 20 cm downstream of the EC **(Figure S1)**. Each impinger contained 20 ml of 10 mM PBN in benzene. After each session, benzene was evaporated and the residue reconstituted in 1 ml benzene. EPR tubes (Sigma-Aldrich, 4 mm o.d.) made of standard high purity quartz cells were filled with 500 µl of the benzene solution and deoxygenated using the freeze-pump-thaw technique which was performed in a thick-walled Schlenk line (Chemglass).[3] Samples were subjected to a total of three freeze-pump-thaw cycles, and argon was used as the inert atmosphere after deoxygenation.

EPR measurements: EPR spectroscopy is a highly selective and sensitive method for detection of free radicals in cigarette smoke.[4] The EPR spectra derived from PBN-radical adducts were measured using a Bruker eScan R spectrometer (Bruker-Biospin, Billerica, MA) operating in X-band. The EPR parameters were as follows: microwave frequency, 9.7 GHz; modulation frequency, 86.0 kHz; microwave power, 6.00 mW; scan range, 60G; modulation amplitude, 1.10 G; sweep time, 41.94 s; time constant, 163.84 ms; and conversion time, 81.92 ms. All measurements were carried out at room temperature (21.5 ± 0.5^{o}C). Spin concentrations were determined by double integration of the first derivative EPR signal using WinSim2002 software (version 0.98, National Institute of Environmental Health Sciences, National Institutes of Health, USA), and comparing the integral with a standardized solution of 2,2,6,6-Tetramethyl-1-piperidinyloxy (TEMPO).

Figure S1. Schematic diagram for e-cigarette (EC) aerosol collection.



References:

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