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Ruyan® E-cigarette Bench-top tests

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Background

Electronic cigarettes, without tobacco, flame or smoke, claim to be cigarette substitutes and to deliver nicotine safely, without smoke toxicants. Are these claims justified?

Aims

To assess the Ruyan® e-cigarette and its mist for safety, emissions, and nicotine dose.

Participating laboratories, materials, methods

Environmental Science and Research, Porirua, NZ.

Hill Laboratories, NZ.

Hort Research, Hamilton, NZ.

Labstat International ULC, Canada.

Lincoln University, NZ.

National Radiation Laboratory of NZ.

Syft Technologies Ltd. Christchurch, NZ

Duke University Center for Nicotine and Smoking Cessation Research (CNSCR) Bioanalytical Lab. NC USA:

British American Tobacco, Group R&D, (Southampton, UK)

TEST MATERIALS

Ruyan in Beijing supplied V8 Classic e-cigarettes and 16 mg nicotine-labeled cartridges ex-factory to test laboratories, directly, or via distributors. Most were manufactured in 2008 and tested in 2008-9. Batteries were re-charged before testing, and fresh cartridges used. An ISO machine-smoked cigarette of 1 mg labeled tar yield provided smoke toxicants.[1]

PUFFING MACHINE SETTINGS

ISO mode *was* the default mode of one 35 ml puff every 60 seconds, of duration 2 seconds, no vent blocked.

Intense mode was 50 mL every 30 seconds, duration 2 seconds, no vent blocked.

Manual puffs. A gas-tight system of syringe with 3-way tap was used to enumerate puffs (35 mL) per cartridge.

Methods

ASSEMBLED E-CIGARETTE

Radiation. NRL of NZ measured the assembled e-cigarette for Pb210 gamma- emitting nucleotides.

Operating temperature. Using a 51-II Hand-held theromocouple, the heating coil, the vaporising contact, and the exiting aerosol were measured every 30 minutes for five hours of use.[1]

Pressure drop. Open pressure drop was measured (3 replicates) at a flow of 17.5 mL/second (based on ISO puff of 35 mL over 2 seconds), and compared with a cigarette.[1]

Battery. Power, current and voltage were measured using a Fluke 183 Digital Multimeter with two probe test leads, during actual puffing in ISO and intense mode on a single port Borgwaldt R58.02) puffing machine.[1]

CARTRIDGE LIQUID

Propyelene glycol, glycol and ethanol were tested by using GC against internal standards.[1,2]

TSNAs (Tobacco-specific nitrosamines) were analysed by LC-MS/MS at Labstat.[15]

Monoamine oxidase inhibition was tested by kynuramine substrate against a tobacco extract by ESR.[3]

PAHs 34 were tested by Hort Research using GCMS.[4,5]

Heavy metals were measured by ICP-MS at ESR. From cartridge liquid, ESR extracted nicotine into methanol using D_3 nicotine as internal standard, and analysed by single ion GCMS, with calibration based on nicotine bitartrate.[6]

The headspace above the liquid was scanned by HS-SPME and GCMS at Lincoln University,[14] and analysed by SIFT Ltd using SIFT-MS (selected ion flow technology), with some difficulty due to quantity of propylene glycol present.[16]

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2 Scientific Analysis of E-cigarettes by British American Tobacco Research &Development. November 2007.
3. Lewis A. Investigation into the effect of RUYAN cartridge exposure on Monoamine oxidase enzyme activity *in vitro*. ESR October 2007.
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5. Polycyclic aromatic hydrocarbons in Ruyan e-cigarettes. Hort Research. Analysed 17 March 2008.

MIST

Particle size distribution. BAT used a fast electrical mobility spectrometer (TSI 3090, MN,USA) measured Ruyan V8 mist in ISO mode. The TSI was unable to measure tobacco smoke, and so another fast electrical mobility spectrometer (Cambustion DMS-500, UK) was used instead. Duke CNSCR used the Marple Series 290 6-stage personal cascade impactor for e-cigarette mist.(Murugesan 2009 pers.comm).

Mist - Selection of toxicants for testing

Over 60 toxicants were selected for testing, based on published priority lists of cigarette smoke toxicants:

9 toxicants recommended by WHO TobReg committee for mandatory lowering;[9]

36 smoke toxicants prioritised by toxicological risk assessment by Fowles & Dybing, including the 9 above;[10]

17 toxicants, additional to the above 45, routinely tested in British Columbia,[11] known loosely as the Hoffman analytes.

Analytical methods

Carbon monoxide *(CO):* An NDIR analyser was calibrated against standard CO/N_2 mixtures. 35 puffs were drawn in ISO mode from e-cigarette by single port smoking machine into a 3L Tedlar bag, then exhausted into analyser.[1] Two replicates.

Other smoke toxicants. BAT and Labstat used their own standard methods to extract, analyse and quantify each toxicant in mist and in cigarette smoke. Using two replicates and based on 50 e-cigarette machine puffs the toxicant in the mist was estimated as the amount expeced in the same number of puffs as were required to machine smoke the tobacco cigarette control.

Nicotine: 50 puffs of 35 mL mist each were collected on a single Cambridge filter pad,[1,2] kept refrigerated until the nicotine was extracted with NaOH and analysed by GCMS. (Use of liquid impinger bottles resulted in analyte loss.)[1,2, 7]

6. Fitzmaurice P. Heavy metal testing of Ruyan cartridge liquid. Inductively coupled plasma mass spectrometry (ICP-MS) Environmental Science and Research (ESR) Porirua 2008.
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9 Burns DM, Dybing E, Gray N, Hecht S, et al. Mandated lowering of toxicants in cigarette smoke: a description of the World Health Organization TobReg Proposal. *Tobacco Control* 2008; 17:132-41.
10. Fowles J, Dybing E. Application of toxicological risk assessment principles to the chemical constituents of cigarette smoke. *Tobacco Control* 2003; 12: 424-430.

Results

THE ASSEMBLED E-CIGARETTE

Radiation. NRLNZ found no gamma-emitting nucleotides.

Operating temperature, at the heating coil averaged 54˚C, at the point of vapourisation, 29.9˚C, and 23.1˚C in the exiting aerosol.[1]

Pressure drop (PD) was greater for the e-cigarette (152 mm WG) than for an unlit conventional cigarette (80-120 mmWG). However in use, with either product, the pressure drop could be 50 mm WG higher.[1]

Battery. The lithium-ion battery was rated by Ruyan to last 1300 puffs – we tested it for 300 puffs only. Per puff it delivered approximately 0.1 mW of power. However at 17 months, towards the end of a battery's shelf life the power delivered faded after 6 puffs. Intense puffing consumed more power.[1]

THE CARTRIDGE LIQUID

The cartridge (labeled 16 mg), contained 1.06 g of liquid, composed of 1.4% nicotine, (13 mg[1] to 14 mg[8]) 90% propylene glycol, and 0.1% glycerol, water 8.8%.

Tobacco specific nitrosamines totalled 8 ng per g.[2]. There was no monoamine oxidase inhibitor activity, as can be detected in tobacco smoke extract.[3]

Of 35 PAHs (polycyclic aromatic hydrocarbons) tested in Ruyan® cartridges in 2008, the carcinogen, benzo-alphapyrene was not detected.[4]. Of the remainder, four carcinogens were detected, in cartridge liquid but these were not human carcinogens, and have no human cancer potency ratings.[5] The carcinogenic heavy metals, including As, Cd, Chr, Ni, Pb were not detected – at the limit of 0.2 micrograms per gram.[6]

Other compounds preent in the liquid included toluene, ethanol, bipyridine, propan-1-ol, bipyridine, tripropylene glycol and beta nicotyrine found in trace quantities.[2]

Headspace above cartridge liquid

Acetaldehyde was detected at 5 ppm, and acrolein at 0.3 ppm.[15] Other cigarette toxicants found to be absent or not detected at 0.01 parts per million or more, were acrylonitrile, benzene, 1,3-butadiene, m-o- and p-cresols, ethylene oxide, HCN, styrene, and xylenes.[16]

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Ruyan® E-cigarette Bench-top tests continued

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Results

THE MIST

Particle size distribution. Particle size was approximately 0.04 microns (count median diameter or CMD).[1] A second laboratory confirmed that CMD was below their cutoff of 0.1 micron.[7] CMD of tobacco smoke was much greater, at 0.15 to 0.25 microns, though measured on a different instrument.[1]

Chemical composition. The Ruyan V8 yielded >300 (35 mL) puffs of mist: 82% PG, 15% water, 1% free-based nicotine, 2% particulates and flavours.[1] Mean puff weight was 0.88 mg.[1]

Toxicology and safety of mist. Of 50+ priority-listed toxicants tested and found present in cigarette smoke, none was found in Ruyan V8 e-cigarette mist.[1 12 17]

Toxicants tested for in Ruyan e-cigarette mist

Not detectable (52)

Aldehydes Acrolein was tested and found absent.[17] Acetaldehyde was found to be present, but with the method used (thermal desorption tubes) could be artefact from ethanol.

Volatiles: Acrylonitrile, Ammonia, Benzene, Carbon monoxide, Isoprene, 1,3-butadiene, toluene, vinyl choride;

Metals: Arsenic, beryllium, cadmium, chromium, lead, nickel, selenium.

Miscellaneous. Acetamide, benzalphapyrene, hydrogen cyanide, pyridine, quinoline, styrene

PAHs and azarenes. Benzo(a)anthracene, Chrysene, Benzo(k)fluoranthene, Benzo(a)pyreneIndeno(1,2,3,-cd)pyrene, 5-methylchrysene, Benzo(b)fluoranthene, Benzo(j)fluoranthene, Dibenz(a,h)acridine, Dibenz(a,j)acridine, 7H-Dibenzo(c,g)carbozole, Dibenz(a,h)anthracene, Dibenz(a,i)pyrene.

Phenols: Catechol, hydroquinone, phenol, resorcinol, m-,o- and p-cresols.

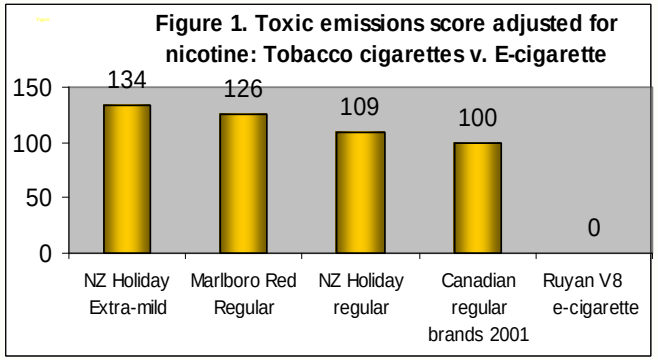
TSNAs: NAB, NAT, NNK, NNN, NDMA, NPYR, NEMA, NDEA, NDPA, NDBA, NPIP.

Detected: (1)

Mercury was detected in trace quantity of 0.17 ng per e-cigarette, which was just above the reporting limit of 0.13 ng, and within the reported 38% coefficient of variation.[1]

Not tested: (14)

Acetaldehyde, formaldehyde, 3 aldehydes + Me Et Ketone: delayed by world shortage of acetonitrile reagent; 1- and 2- amino-naphthalenes, and 3, and 4-aminobiphenyls; chlorinated dioxins and furans; hydrazine; nitrous oxide and other oxides of nitrogen; and urethane.

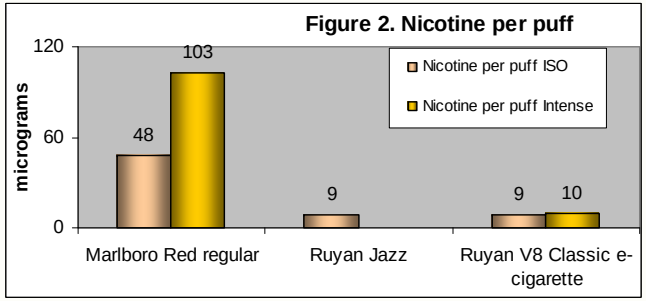


In figure 1, the toxicant emissions score adjusted for nicotine is a relative score comparing different cigarette brands, with Canadian brands scored at 100.[17]

NICOTINE CHEMISTRY

Free base nicotine The nicotine in the V8 inhaler condensate was checked against the UK governmental standard for nicotine. It is in the form of the S-stereo-isomer, and is free-base nicotine.[15] Loss of nicotine dissolved in PG from oxidation by ambient air reaching the cartridge liquid after the cartridge is opened and assembled into the e-cigarette is a theoretical possibility, which we have not studied. Visible mist is a pre-condition for adequate vaporisation of nicotine, but visible mist, as in the 0 mg nicotine cigarette, does not imply nicotine is present.

Nicotine delivery per puff A 35 mL puff from the Ruyan® V8 delivers only 10% of the nicotine obtained from a similar puff of a Marlboro regular cigarette. Deeper 50 mL puffs from the Ruyan V8 delivers only slightly more nicotine.[1]



Note: In Figure 2, a human-smoked Marlboro cigarette is estimated to produce 15 puffs.
Source: BAT Group R&D, November 2007.[2] BAT Group R&D, 15 April, 2009.[1]

Site of nicotine absorption No deposition of aerosol nicotine occurred on pulling mist through six levels of the diminishing apertures of a cascade impactor, indicating very small particle size and little or no absorption from lung.[7]

Discussion

Main finding Testing for over 50 cigarette key smoke toxicants found none in any but trace quantity, in Ruyan V8 mist.

Safety of e-cigarettes as a product class Safety results refer to the Ruyan® V8 Classic. However the low operating temperature (54˚C) of the atomiser, that is 5 to 10% of the temperature of a burning cigarette, suggests e-cigarettes as a class are unlikely to emit cigarette toxicants in their mist.

Nicotine dose (Figure 2) An e-cigarette user will need to take more puffs more often, and deeper puffs confer little advantage for V8 users. Six puffs every 5 minutes would deliver the same dose of nicotine delivered by shallow inhaling of 10 (35 mL) puffs) from one tobacco cigarette every hour. This however will not achieve the high immediate nicotine boost which many smokers crave.

Nicotine overdose is unlikely, even though nicotine delivery may vary considerably between brands.

Nicotine absorption site The nicotine dose and particle size are too small to ensure either deposition in the alveoli or bronchioles or rapid nicotine absorption as in cigarette smoking.

Limitations of study The results apply only for the products tested. Extrapolation to all product sold assumes production only from internationally-certified good manufacturing sites, and trademark enforcement.

Conclusion

The Ruyan® V8 nicotine e-cigarette does not deliver smoke or smoke toxicants. Consequently, the modest reductions recommended in 2008 by WHO's Tobacco Regulation committee for 9 major toxicants in cigarette smoke (in line with Articles 9 and 10 of the WHO FCTC World Health Organization Framework Convention Tobacco Control treaty), are already far exceeded and obtainable now by smokers switching to the Ruyan® e-cigarette.

Absolute safety does not exist for any drug, but e-cigarette emissions, relative to tobacco smoke, are likely to be 100 to 1000 times safer. On the evidence to date, the e-cigarette appears to be akin to a medicinal nicotine inhalator in the safety of its emissions, and nicotine dose.

E-cigarettes are cigarette substitutes. If they can wrest market share from cigarettes, they can improve smoker and population health. They may also have a more up-market role as medicinal nicotine inhaler to aid quitting. Further trials of acceptability, addiction potential, clinical safety, and quitting efficacy are needed.

Funding and acknowledgements

Hon Lik, of Ruyan Group is the inventor of this product, and Ruyan Group (Holdings) Ltd Beijing funded Health NZ to carry out initial tests. Duke University, (NC, USA) and British American Tobacco, Group R&D (UK), kindly supplied further results at no cost.

Competing interests

Neither the author, or his company, has any financial interest in Ruyan or any other manufacturer.