



A randomised, crossover study on an electronic vapour product, a nicotine inhalator and a conventional cigarette. Part B: Safety and subjective effects



Tanvir Walele ^{a, *}, Girish Sharma ^c, Rebecca Savioz ^d, Claire Martin ^d, Josie Williams ^b

^a Fontem Ventures, Barbara Strozilaan 101, 1083 HN Amsterdam, Netherlands

^b Imperial Tobacco, Product Science, 121 Winterstoke Road, Bristol, UK

^c Simbec Research, Merthyr Tydfil, CF48 4DR, UK

^d Clinopsis S.A., Jardins 6, Concise 1426, Switzerland

ARTICLE INFO

Article history:

Received 17 August 2015

Received in revised form

10 December 2015

Accepted 10 December 2015

Available online 15 December 2015

Keywords:

Adverse events

Withdrawal symptoms

Smoking urges

Electronic vapour product

Electronic cigarette

Nicotine inhalator

Cigarette

Clinical study

ABSTRACT

An Electronic Vapour Product (EVP) has been evaluated for short-term safety parameters and subjective effects in a 2-part study, in smokers. Part 1 compared the EVP with unflavoured (UF) and flavoured (FL) e-liquid at 2.0% nicotine to a conventional cigarette (CC; JPS Silver King Size, 0.6 mg) and a licensed nicotine inhalator (Nicorette[®], 15 mg). Part 2 assessed the effect of increasing concentrations of nicotine in the e-liquid used with the EVP (0%, 0.4%, 0.9%, 2.0%). The study was designed as a randomised, controlled, crossover trial. Outcomes included adverse events (AEs), vital signs, exhaled carbon monoxide (CO), clinical laboratory parameters, smoking urges and withdrawal symptoms. In both study parts, only mild non-serious AEs were reported. No major differences were observed in AEs between the EVPs and Nicorette[®]. Exhaled CO levels only increased for CC. All products appeared to decrease smoking urges and nicotine withdrawal symptom scores to a similar extent. The EVP had a similar short-term safety profile to Nicorette[®] and relieved smoking urges and nicotine withdrawal symptoms to a similar extent as Nicorette[®] and CC. Unlike nicotine replacement therapies, the EVP may offer an alternative for those finding it difficult to quit the behavioural and sensorial aspects of smoking.

© 2015 The Authors. Published by Elsevier Inc. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

1. Introduction

Electronic vapour products (EVPs), also known as “electronic cigarettes” are a relatively new class of products. Even though the majority of EVPs are marketed as consumer products, they are often reported to be used as a means to stop smoking conventional cigarettes (Berg et al., 2015; Dockrell et al., 2013; Etter and Bullen, 2011). Any claims to cessation or harm reduction must require a medicinal license (MHRA, 2015). The few short-term studies performed to date suggest that EVPs have the potential for being safer alternatives to conventional cigarettes (CC) and at the same time satisfy the ritualistic elements of smoking. For example, EVPs do not increase exhaled carbon monoxide (CO) levels or white blood cell count, and do not have immediate effects on myocardial and

lung functions (Farsalinos, 2012; Flouris et al., 2013, 2012; Vansickel et al., 2010; Vardavas et al., 2012). When smokers switch to use EVPs and are followed-up for prolonged periods, observations have included a progressive decrease in occurrences of adverse events (AEs) commonly reported by CC smokers, e.g. cough, dry mouth, shortness of breath, throat irritation and headache (Caponnetto et al., 2013; Farsalinos et al., 2014a; Polosa et al., 2014; van Staden et al., 2013). A higher frequency of mouth and throat irritation was observed in smokers switching to using a Nicorette[®] inhalator, compared to those using EVPs (Bullen et al., 2010). Few commercially available EVPs have been studied for their subjective effects such as the suppression of desire to smoke and tobacco or nicotine abstinence symptoms. Some studies have demonstrated that even with no nicotine present in EVP e-liquid, nicotine craving and withdrawal symptoms were alleviated albeit less compared to CCs (Bullen et al., 2010; Vansickel et al., 2010).

This study was conducted as part of a product stewardship evaluation of an EVP prototype. The evaluation of the product's plasma nicotine pharmacokinetic (PK) profile is reported elsewhere

Abbreviations: AE, Adverse event; CC, Conventional cigarettes; CO, Carbon monoxide; EVP, Electronic vapour product; NRT, Nicotine replacement therapy.

* Corresponding author.

E-mail address: tanvir.walele@fontemventures.com (T. Walele).

<http://dx.doi.org/10.1016/j.yrtph.2015.12.004>

0273-2300/© 2015 The Authors. Published by Elsevier Inc. This is an open access article under the CC BY license (<http://creativecommons.org/licenses/by/4.0/>).

(Walele et al., 2015). In this paper, the short-term health effects and the potential of the EVP for reducing smoking urges and withdrawal symptoms are described. The study consisted of two parts. Part 1 compared the EVP with an unflavoured (UF) and a flavoured (FL) e-liquid containing 2.0% nicotine to a nicotine replacement therapy (NRT) product and a commercially available CC. Part 2 investigated the effects of the EVP with unflavoured e-liquids containing increasing levels of nicotine (0%, 0.4%, 0.9% and 2.0%).

2. Materials and methods

2.1. Study design

This study was performed at a single clinical site (Simbec Research Ltd, Wales) in a confinement setting. A total of 24 healthy male subjects, recruited in the UK, participated in the study: 12 assigned to Part 1 and 12 to Part 2. Both study parts were designed as a randomised, controlled, four-way crossover trial. Part 1 was performed open-label and Part 2 was blinded. Following overnight abstinence from smoking or using EVPs, subjects used each different product for one daily use session.

The study was approved by the South East Wales Research Ethics Committees on 31 October 2013, and is registered at the US National Institutes of Health (ClinicalTrials.gov) #NCT02032212. All procedures were performed in accordance with the International Conference on Harmonisation (ICH) Harmonised Tripartite Guideline for Good Clinical Practice (GCP). The Medicines and Healthcare Products Regulatory Agency (MHRA)-UK granted Clinical Trials Authorisation (CTA) for the use of the NRT product in this study. All subjects signed an informed consent form prior to any study procedures being performed.

2.2. Study population

Detailed inclusion and exclusion criteria are presented in our paper reporting the plasma PK results (Walele et al., 2015). Subjects were 21–65 year old males and were confirmed smokers (5–30 cigarettes per day for at least one year). The subjects' smoking history was recorded using internal questionnaires and with the Fagerström Test for Nicotine Dependence (FTND) (Heatherton et al., 1991). Subjects were excluded if they were taking or receiving any form of NRT, snuff or chewing tobacco or if they intended to stop smoking.

2.3. Products used in this study

The EVP prototype used in this study was developed by Fontem Ventures B.V. It consisted of a rechargeable battery, an atomiser and a capsule containing e-liquid (Fig. 1). The capsules were replaceable and the battery and atomiser were reusable. The base components of the e-liquids used are propylene glycol (70–75% w/w), glycerol (18–20% w/w) and water (5% w/w). Two e-liquids were used in Part

1 of the study, which differed solely in their flavour content: an unflavoured base e-liquid with 2.0% nicotine (UF2.0%; 2.7 mg/capsule) and a flavoured (menthol) e-liquid with 2.0% nicotine (FL2.0%; 2.7 mg/capsule). In Part 2, four unflavoured e-liquids were used, which differed in their nicotine content: 0% nicotine (UF0%), 0.4% nicotine (UF0.4%; 0.4%; 0.54 mg/capsule), 0.9% nicotine (UF0.9%; 1.22 mg/capsule) and UF2.0%. The EVP with UF2.0%, FL2.0% and UF0.4% delivers mean amounts of 0.013, 0.007 and 0.002 mg nicotine per puff, respectively (internal data, generated under Health Canada Intense smoking regime). Nicotine delivery with UF0.9% was not measured.

In Part 1 of the study, the NRT Nicorette® Inhalator (15 mg nicotine, manufacturer Johnson & Johnson; coded NIC15) was used as a comparator product and a JPS Silver King Size CC (0.6 mg nicotine; manufacturer Imperial Tobacco Group) was used as a control.

2.4. Study interventions and schedule

Subjects were admitted to the study site on the morning of Day –2 (baseline) for confirmation of eligibility and training on using the EVP or NIC15. Smoking status was verified by measuring the urinary cotinine levels in a spot urine sample (NicAlert strip), the exhaled CO levels (measured with a portable Bedfont Micro + Smokerlyser device) and blood carboxyhemoglobin (COHb) levels (2 mL sample, from a forearm vein, in lithium heparin, measured with a blood gas analyser system). A safety assessment was performed on Day –2, which included vital signs (blood pressure, heart rate and oral body temperature in supine position), AEs, a physical exam, a lung function test (spirometry) and a 12-lead electrocardiogram (ECG). Blood (7.6 mL, from forearm vein) and urine samples were taken to measure haematology, clinical chemistry and urinalysis parameters, for standard clinical laboratory evaluations. From the time of admission, subjects were not permitted to use any EVP, NRT or CC other than that assigned by the study design and were not allowed to consume alcohol. Subjects remained in confinement until the end of the study period, on the morning of Day 5. On Day –1, the revised Minnesota Nicotine Withdrawal Scale questionnaire (MWS-R) was administered to document nicotine withdrawal symptoms (Hughes, 2007) and the Brief Questionnaire of Smoking Urges (QSU-Brief), to measure craving (Cox et al., 2001). On Day –1, subjects were randomly assigned to one of four pre-defined sequences of product use within their allocated study Part, in a 3:3:3:3 ratio.

On study Days 1, 2, 3, and 4, after overnight smoking abstinence, subjects used the allocated product for four product administrations at 1-h intervals (0hr, 1hr, 2hr and 3hr). Each administration consisted of 10 inhalations at 30 s intervals. Each inhalation was monitored, and subjects were instructed to take 4-s puffs for the EVP and NIC15, and 2-s puffs for the CC (an electronic tablet was used instructing subjects when to inhale and exhale). Vital signs were recorded approximately 30 min before the first product administration, and 30 min after the fourth one. Exhaled CO was measured 5 min before and 25 min after each product administration. Subjects filled the MWS-R and QSU-Brief questionnaires approximately 30 min after the third administration, at a similar timing as on Day –1. These assessments were done 30 min after the fourth administration because priority was given to PK sampling (Walele et al., 2015). AEs were monitored on each study day.

On Day 5, safety assessment parameters were checked, and subjects answered both the MWS-R and QSU-Brief questionnaires for the last time. Subjects were also provided full verbal smoking cessation advice by the investigator and were discharged from the clinic after all study assessments were performed.



Fig. 1. Schematic of the external appearance and parts of the tested EVP. From left to right, pieces are: the housing, which contains the battery and has a LED indicator on the side, the atomiser, the capsule containing the e-liquid and the mouthpiece.

2.5. Randomisation

Randomisation of subjects to one of the four product use sequences was performed according to randomisation codes produced using the PROC PLAN procedure of SAS[®] version 9.4.

2.6. Study outcome measures

Safety assessment measures included vital signs, 12-lead ECG, lung function tests, exhaled CO, monitoring of AEs and standard haematology, clinical biochemistry and urinalysis parameters (Table 1).

Withdrawal symptoms were evaluated with a modified MWS-R questionnaire (Hughes, 2007), to which only the 15 questions of the subject's part were completed. Subjects had to rate behaviours (e.g. angry, irritable, frustrated, depressed, restless, insomnia) from 0 (none) to 4 (severe). Craving was assessed with the QSU-Brief questionnaire. Subject had to rate 10 statements, such as "I have a desire for a cigarette right now", by a number ranging from 1 (strongly disagree) to 7 (strongly agree) (Cox et al., 2001).

2.7. Bioanalytical methods

COHb in whole blood samples was assessed with the Roche Cobas B221 Blood Gas Analyser System using a spectrophotometric method (Roche, 2009). Haematology samples were analysed using the Siemens Advia 2120[®] or Siemens Advia 120[®] using commercially available kits. Clinical biochemistry samples were analysed using the Roche Modular Analytics System[®] using commercially available kits. Urinalysis parameters were measured using the Siemens Clinitek 500 analyser.

2.8. Statistical methods

2.8.1. Sample size

The sample size was mainly determined for nicotine bioavailability comparisons (Walele et al., 2015) and was selected based on similar PK studies on similar products (Bullen et al., 2010; Dawkins and Corcoran, 2014; Farsalinos et al., 2014b) and guidance from competent authorities (EMA, 2010; HC, 2012). Twelve subjects per study part were considered sufficient in the crossover design, as all subjects used each of the four different products.

2.8.2. Safety parameters

In order to avoid an effect of multiple comparisons, no specific statistical analyses have been performed on the safety parameters. Descriptive statistics were calculated for continuous values and frequencies for categorical variables.

2.8.3. Withdrawal and craving parameters

For the MWS-R questionnaire, the sum of the scores of the first nine questions was calculated (core score), as well as the sum of all 15 questions (total score). For the QSU-Brief questionnaire, the sum of the scores of all 10 questions was calculated, as well as the sum of the scores for questions 1, 3, 6, 7 and 10 (Factor 1; desire and intention to smoke with smoking anticipated as pleasurable) and the sum of the scores for questions 2, 4, 5, 8 and 9 (Factor 2; anticipation of relief from negative effect and nicotine withdrawal). For both questionnaires, descriptive statistics (mean scores and standard deviations) were calculated.

3. Results

3.1. Subjects' characteristics

Subjects were screened from January 2014 to March 2014. All 24 enrolled subjects completed the study according to the protocol (see methods). There were no withdrawals. Detailed subjects' characteristics are provided elsewhere (Walele et al., 2015).

3.2. Safety

3.2.1. Adverse events

Table 2 summarises the AEs reported during both study parts. None of the subjects reported any moderate or serious AE and there were no AEs leading to study withdrawal. In Part 1, 5/12 subjects (41.7%) reported a total of 12 AEs, which were evaluated as mild. Given the overall low number of AEs, no clear product-trends were observed. In Part 2, 7/12 subjects (58.3%) reported a total of 13 AEs, all of which were evaluated as mild. As in Part 1, no clear product-related trend was observed, even though most AEs occurred with the products containing the highest nicotine concentrations. Cough, reported by two subjects using UF2.0% and one subject using UF0.9%, was the most common AE.

3.2.2. Exhaled CO

In Part 1, exhaled CO levels increased with each CC administration, reaching a maximum value of 14.8 ppm 25 min after the 4th administration, compared to maximum values of 2.5 ppm following UF2.0%, 2.2 ppm after FL2.0% and 2.3 ppm after NIC15 (Fig. 2).

In Part 2, there were no dose-related effects on CO levels, as reflected by similar mean values of approximately 2–3 ppm observed following each administration of UF0%, UF0.4%, UF0.9% and UF2.0% (Fig. 3).

3.2.3. Other safety parameters

For both Part 1 and Part 2 of the study, there were no clinically

Table 1
Study outcome measures.

Outcome measure	Detail of measured parameters
Vital signs	Supine systolic and diastolic blood pressure, pulse rate and oral temperature
12-lead ECG	Heart rate, PR interval, QSR width, QT interval and QTcB interval
Lung function tests	lung function tests: FVC and % predicted FVC (FVC _{%pred.}), FEV ₁ and % predicted FEV ₁ (FEV _{1%pred.}), forced expiratory flow 25%–75% (FEF _{25–75}) and % predicted FEF _{25–75} (FEF _{25–75%pred.}), peak expiratory flow (PEF) and % predicted PEF (PEF _{%pred.}), FEV ₁ /FVC ratio and % predicted FEV ₁ /FVC ratio (FEV ₁ /FVC _{%pred.})
Exhaled CO	Exhaled CO
AEs	Monitoring of AEs along with severity grades and relationship to product. All AEs were coded using the Medical Dictionary for Regulatory Activities (MedDRA), version 16.1, 2013.
Haematology	Haemoglobin, haematocrit, mean cell volume, mean cell haemoglobin, mean cell haemoglobin concentration, red blood cells, white blood cells, neutrophils, lymphocytes, monocytes, eosinophils, basophils and platelets
Clinical biochemistry	Total protein, albumin, total bilirubin, inorganic phosphorus, alkaline phosphatase, alanine transaminase, aspartate transaminase, gamma glutamyl transferase, glucose, sodium, potassium, chloride, blood urea nitrogen, creatinine and calcium
Urinalysis	pH, protein, glucose, ketones, urobilinogen, blood and specific gravity

Table 2
Number of subjects (%) reporting AEs and number of AEs by relationship and by SOC for each product in Part 1 and Part 2.

Part 1					
	UF2.0%	FL2.0%	NIC15	CC	Overall
	(N = 12)	(N = 12)	(N = 12)	(N = 12)	(N = 12)
Number of AEs	2	3	7	0	12
Number (%) subjects with ≥ 1 AE:	1 (8.3%)	3 (25.0%)	3 (25.0%)	0	5 (41.7%)
Number (%) of subjects with AE/number of AEs by relationship					
Related	1 (8.3%)/2	2 (16.7%)/2	3 (25.0%)/7	0	4 (33.3%)/11
Unrelated	0	1 (8.3%)/1	0	0	1 (8.3%)/1
Number (%) of subjects with AE/number of AEs by SOC					
Gastrointestinal disorders					
Glossodynia (related)	0	0	1 (8.3%)/1	0	1 (8.3%)/1
Infection and infestation					
Nasopharyngitis (unrelated)	0	1 (8.3%)/1	0	0	1 (8.3%)/1
Respiratory, thoracic and mediastinal disorders:					
Cough (related)	1 (8.3%)/2	1 (8.3%)/1	2 (16.7%)/5	0	2 (16.7%)/8
Throat irritation (related)	0	1 (8.3%)/1	1 (8.3%)/1	0	2 (16.7%)/2
Part 2					
	UF2.0%	UF0.9%	UF0.4%	UF0%	Overall
	(N = 12)	(N = 12)	(N = 12)	(N = 12)	(N = 12)
Number of AEs	6	3	3	1	13
Number (%) subjects with ≥ 1 AE:	3 (25.0%)	3 (25.0%)	3 (25.0%)	1 (8.3%)	7 (58.3%)
Number (%) of subjects with AE/number of AEs by relationship:					
Related	2 (16.7%)/3	1 (8.3%)/1	0	0	3 (25.0%)/4
Unrelated	1 (8.3%)/3	2 (16.7%)/2	3 (25.0%)/3	1 (8.3%)/1	4 (33.3%)/9
Number (%) of subjects with AE/number of AEs by SOC					
Gastrointestinal disorders:					
Toothache (unrelated)	0	1 (8.3%)/1	0	0	1 (8.3%)/1
General disorders and administration site conditions:					
Fatigue (unrelated)	1 (8.3%)/1	0	1 (8.3%)/1	0	2 (16.7%)/2
Musculoskeletal and connective tissue disorders:					
Myalgia (unrelated)	0	1 (8.3%)/1	0	0	1 (8.3%)/1
Musculoskeletal and connective tissue disorders:					
Dizziness (unrelated)	1 (8.3%)/1	0	0	1 (8.3%)/1	2 (16.7%)/2
Headache (unrelated)	0	0	2 (16.7%)/2	0	2 (16.7%)/2
Paraesthesia (unrelated)	1 (8.3%)/1	0	0	0	1 (8.3%)/1
Respiratory, thoracic and mediastinal disorders:					
Cough (related)	2 (16.7%)/3	1 (8.3%)/1	0	0	3 (25.0%)/4

Note. SOC, system organ class.

significant biochemistry, haematology, urinalysis or physical examination findings. At baseline and at Day 5 there were no clinically significant 12-lead ECG or lung function test findings, for any product in either study part. There were no product-related effects on vital signs during Part 1 of the study, as reflected by similar mean values for each parameter pre-administration (30 min before the first administration) and post-administration (30 min after the

4th administration) of UF2.0%, FL2.0%, NIC15 and CC (data not shown). There were no dose-related effects on vital signs during Part 2 of the study, as reflected by similar mean values for each parameter pre-administration and following administration of UF0%, UF0.4%, UF0.9% and UF2.0% (data not shown).

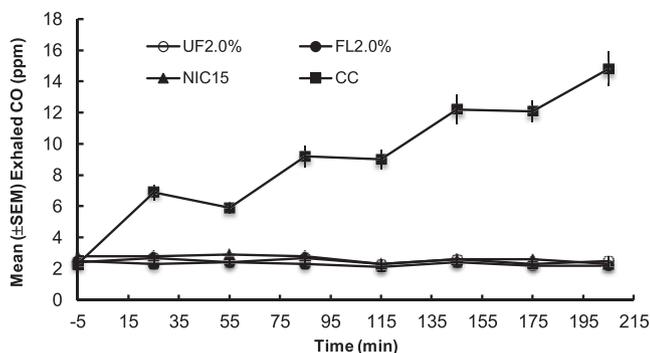


Fig. 2. Mean (\pm SEM) exhaled CO levels 5 min before and 25 min after each product administration. Twelve subjects received each product in a crossover design. CO, carbon monoxide; UF2.0%, unflavoured base e-liquid at 2.0% nicotine; FL2.0%, flavoured base e-liquid at 2.0% nicotine; NIC15, Nicorette[®] Inhalator 15 mg nicotine; SEM, standard error of the mean.

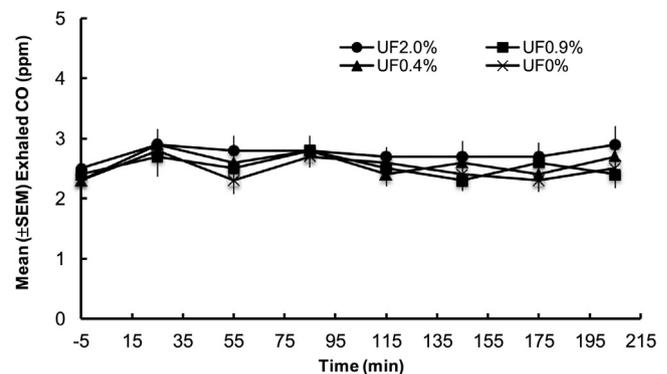


Fig. 3. Mean (\pm SEM) exhaled CO levels 5 min before and 25 min after each product administration. Twelve subjects received each product in a crossover design. CO, carbon monoxide; UF2.0%, unflavoured base e-liquid at 2.0% nicotine; UF0.9%, unflavoured base liquid at 0.9% nicotine; UF0.4%, unflavoured base liquid at 0.4% nicotine and UF0%, unflavoured base liquid at 0% nicotine; SEM, standard error of the mean.

3.3. Craving and withdrawal symptoms

3.3.1. MWS-R questionnaire

Mean total scores of the MWS-R questionnaire by product, for Part 1 and Part 2 of the study, are shown in Table 3.

In Part 1, the mean total score was 9.80 on Day –1 and slightly decreased to a range of 6.40–8.10 on Days 1–4, regardless of the administered product. The scores were similar for UF2.0%, FL2.0%, NIC15 and the CC. On Day 5, the total score increased slightly to reach 9.50. These results indicate that UF2.0%, FL2.0%, NIC15 and the CC induced a small reduction of nicotine withdrawal symptoms, and did so to a similar extent. Results from the core scores showed a similar trend (not shown). In Part 2, all four products decreased the mean total score from a level of 13.20 on Day –1 to a range of 7.30–9.00 on Days 1–4. UF0%, UF0.4%, UF0.9% and UF2.0% had similar mean total scores, indicating that the level of nicotine had no influence on withdrawal symptoms. On Day 5, the total score decreased slightly further to a mean level of 5.30. Core scores indicated similar trends (data not shown).

3.3.2. QSU-Brief questionnaire

Mean total scores of the QSU-Brief questionnaire are summarised in Table 3. During Days 1–4 of Part 1, total scores were similar for UF2.0%, FL2.0%, NIC15 and the CC, and ranged from 22.80 to 25.60. They were lower than the total score of 38.00 on Day –1 and that of 31.40 on Day 5, which indicates that all four products reduced urge to smoke to a similar extent. The pattern was similar for Factor 1 and Factor 2 scores (data not shown). In Part 2, the total mean score reached 41.90 on Day –1, and on Days 1–4 was reduced to mean scores ranging from 31.50 to 34.90, regardless of the administered product. The total score increased to a mean of 33.00 on Day 5. These results indicate that all four products could slightly reduce craving. Since the study was not powered to perform statistical analyses on the secondary outcomes, no firm conclusions can be drawn on the influence of nicotine level on the questionnaire scores and that there was no influence of the nicotine level. Factor 1 and Factor 2 scores followed a similar trend (data not shown).

As the Day 5 questionnaires were performed at an earlier time of the day than those of Days –1 to 4, the results from Day 5, although presented here, have not been taken into account for the interpretation.

4. Discussion

Here we present the safety data of a clinical study performed on

an EVP prototype, along with data on smoking urges and nicotine withdrawal symptoms. We have previously shown that the plasma nicotine pharmacokinetics of the EVP with 2% nicotine e-liquids is similar to the Nicorette® inhalator (Walele et al., 2015). Overall, the EVP containing doses of nicotine ranging from 0 to 2% with and without flavourings was well tolerated and was observed to decrease smoking urges and tobacco withdrawal symptoms in smokers during a short-term use. At study completion, there were no clinically significant findings in 12-lead ECG and in lung function test results for all subjects. Regarding exhaled CO, increases were only observed in subjects using the CC. Exhaled CO levels did not change from baseline levels following four administrations of EVPs and NIC15. This finding is in agreement with other published studies where smokers used an EVP in short, controlled sessions (Flouris et al., 2013; Vansickel et al., 2010).

In both study parts, the use of the EVP was associated with very few AEs, all of which were mild. The two most common AEs, cough and throat irritation, have also been reported elsewhere as side effects of EVPs, both following acute exposure and with regular use in real-life settings (Bullen et al., 2010; Caponnetto et al., 2013; Dawkins and Corcoran, 2014; Farsalinos et al., 2014a; Polosa et al., 2014). Throat irritation was also reported as one of the most common AEs when using a new nicotine inhaler (Moyses et al., 2015). In the first part of our study, both the flavoured and the unflavoured EVPs had an AE profile similar to NIC15. Bullen et al., however, found a significantly higher incidence of mouth and throat irritation in subjects using a 10 mg Nicorette® inhalator, compared to a 0 mg and a 16 mg EVP (approx. 1.6% nicotine) (Bullen et al., 2010). In that study, subjects used the products for a whole day, whereas in our study, the product was used for four administrations, 1 h apart. None of the subjects in our study reported AEs when using the CC. As subjects were regular, confirmed smokers of CC it is anticipated that they were used to the effects of smoking a CC.

The different nicotine levels used had no impact on the incidence and nature of AEs, except for mild cough, observed to occur only following the use of the two highest nicotine strengths. As the incidences were low and no statistical analysis was performed, a clear effect of nicotine concentration on cough cannot be concluded. Bullen et al. did not find any difference in reported AEs between a 0 mg and a 16 mg electronic cigarette, except for mouth and throat irritation, which was more common with the 16 mg nicotine EVP (Bullen et al., 2010).

None of the EVPs used in this study had any measureable effect on blood pressure or pulse rate. In Part 1 of our study, CC smoking did not impact pulse rate or blood pressure. As these parameters

Table 3
Summary of MWS-R and QSU-Brief questionnaires scores.

		MWS-R total	QSU-Brief total
Part 1			
Baseline (Day –1)		9.80 (7.03)	38.00 (9.77)
Days 1–4	UF2.0%	6.40 (6.52)	24.80 (11.50)
	FL2.0%	6.80 (7.32)	25.00 (12.93)
	NIC15	7.50 (7.08)	25.60 (12.03)
	CC	8.10 (8.93)	22.80 (14.19)
End of study (Day 5)		9.50 (8.47)	31.40 (12.22)
Part 2			
Baseline (Day –1)		13.20 (11.54)	41.90 (12.92)
Days 1–4	UF0%	7.30 (6.48)	34.90 (19.67)
	UF0.4%	8.50 (6.53)	34.80 (15.05)
	UF0.9%	9.00 (9.77)	31.90 (19.60)
	UF2.0%	7.80 (5.89)	31.50 (18.06)
End of study (Day 5)		5.30 (4.86)	33.00 (19.48)

All values are mean (standard deviation). For Study Days 1–4, mean scores were calculated over the four days, for each product. Subjects did not use any product on Day 5, before answering the questionnaires.

were measured 30 min after the last administration, transient effects may have been missed. Indeed, in the study performed by Vansickel et al., (Vansickel et al., 2010) heart rate was monitored as from 5 min after the onset of product use, and subjects smoking a CC had a sharp increase in heart rate at 5 min, followed by a decrease to reach rates close to baseline at 30 min. Farsalinos et al. also observed a significant increase in diastolic blood pressure in EVP users, 5 min after the use session. Increased heart rate and blood pressure are known physiological effects of nicotine (Benowitz, 1997). In our study, the use of UF2.0% produced a maximum mean plasma nicotine level (C_{\max}) of 3.6 ng/mL, 9 min after the fourth product administration. FL2.0% produced a C_{\max} of 2.5 ng/mL, reached after 10 min (Walele et al., 2015). In comparison, the CC produced a C_{\max} of 21.2 ng/ml reached in 3 min and NIC15 a C_{\max} of 2.5 ng/ml, in 13 min. Consequently, in our study, the plasma nicotine levels may have been too low to trigger heart rate and blood pressure increases. According to recent studies, heart rate increases were observed in experienced EVP users who also displayed significant increases in plasma nicotine levels (Spindle et al., 2015; Vansickel and Eissenberg, 2013).

Despite not reaching high nicotine levels in blood (the EVP with 0%, 0.4%, 0.9% and 2.0% nicotine produced C_{\max} values of 0.6, 1.0, 1.9 and 3.6 ng/mL, respectively), nicotine craving and withdrawal symptoms slightly decreased during the EVP use sessions compared to baseline. All EVPs, including UF0%, decreased both MWS-R and QSU-Brief scores to a similar extent when compared to the CC and NIC15. This is in agreement with findings from others (Bullen et al., 2010; Vansickel et al., 2010). A recent study using a novel nicotine inhaler showed a slight but significant reduction in craving and smoking urges when compared to a 10 mg Nicorette inhalator (Moyses et al., 2015). The inhaler was shown to result in lower venous plasma C_{\max} (<4 ng/mL) than use of a Nicorette product (6–8 ng/mL). The time to C_{\max} however was shorter, suggestive of pulmonary rather than oral absorption of nicotine, as is the case for Nicorette. Corresponding data following use of a CC was not reported in that study so no further conclusions can be drawn.

The results in this study suggest that the behavioural aspects of EVP use may contribute to the reduction of both smoking urges and withdrawal symptoms as supported by the observation that the use of UF0% also produced small reductions in these scores. This observation may also partly be due to a psychological relief to use any inhalable product, as subjects, who were confirmed smokers of CC, were not allowed to use any product between each product use session. This artificial situation resulted from study design constraints needed for the nicotine PK measurements (Walele et al., 2015). Any conclusions on subjective effects drawn from our study should therefore be applied to real-life settings with care.

To conclude, the EVPs tested in this study, with different nicotine levels, with or without flavours, had a similar short-term safety profile to an MHRA approved NRT product. Users of all EVPs, regardless of their nicotine content, reported a decrease in nicotine craving and withdrawal symptoms, which was, in the short-term, similar to the decrease observed with a CC. The EVP studied here may therefore offer a viable alternative to CCs for those finding it difficult to quit the behavioural and sensorial aspects of smoking. These findings indicate that EVP use has the potential as an aid for smoking reduction or cessation if it can meet the standards of efficacy, safety and quality set by relevant regulatory authorities such as the MHRA. Further work should however be carried out on the use of the EVP in real-life settings.

Funding

This work was funded and supported by Fontem Ventures. Imperial Tobacco Group is the parent company of Fontem Ventures

B.V., the manufacturer of the EVP prototype used in this study.

Declaration of interests

Tanvir Walele is an employee of Fontem Ventures B.V. and Josie Williams is an employee of Imperial Tobacco Group. Girish Sharma, Rebecca Savioz and Claire Martin received personal fees from Fontem Ventures B.V.

Acknowledgements

We would like to thank Simbec Research for conducting the clinical trial and statistical evaluation. We would also like to thank Dr Constance Hoefer and Dirk Lindner for their contributions to study design and statistical analysis.

Transparency document

Transparency document related to this article can be found online at <http://dx.doi.org/10.1016/j.yrtph.2015.12.004>.

References

- Benowitz, N.L., 1997. The role of nicotine in smoking-related cardiovascular disease. *Prev. Med.* 26, 412–417.
- Berg, C.J., et al., 2015. Cigarette users' interest in using or switching to electronic nicotine delivery systems or smokeless tobacco for harm reduction, cessation, or novelty: a cross-sectional survey of US adults. *Nicotine Tob. Res.* 17, 245–255.
- Bullen, C., et al., 2010. Effect of an electronic nicotine delivery device (e cigarette) on desire to smoke and withdrawal, user preferences and nicotine delivery: randomised cross-over trial. *Tob. Control* 19, 98–103.
- Caponnetto, P., et al., 2013. Efficiency and safety of an eElectronic cigAreTte (ECLAT) as tobacco cigarettes substitute: a prospective 12-month randomized control design study. *PLoS One* 8, e66317.
- Cox, L.S., et al., 2001. Evaluation of the brief questionnaire of smoking urges (QSU-brief) in laboratory and clinical settings. *Nicotine Tob. Res.* 3, 7–16.
- Dawkins, L., Corcoran, O., 2014. Acute electronic cigarette use: nicotine delivery and subjective effects in regular users. *Psychopharmacol. Berl.* 231, 401–407.
- Dockrell, M., et al., 2013. E-Cigarettes: prevalence and attitudes in Great Britain. *Nicotine Tob. Res.* 15, 1737–1744.
- EMA, European Medicines Agency, 2010. Committee for Medicinal Products for Human Use (CHMP). Guideline on the Investigation of Bioequivalence. http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2010/01/WC500070039.pdf. Last accessed 23.10.2015.
- Etter, J.F., Bullen, C., 2011. Electronic cigarette: users profile, utilization, satisfaction and perceived efficacy. *Addiction* 106, 2017–2028.
- Farsalinos, K.E., 2012. Acute effects of using an electronic nicotine-delivery device (e-cigarette) on myocardial function: comparison with the effects of regular cigarettes. *ESC Congress, Munich*.
- Farsalinos, K.E., et al., 2014a. Characteristics, perceived side effects and benefits of electronic cigarette use: a worldwide survey of more than 19,000 consumers. *Int. J. Environ. Res. Public Health* 11, 4356–4373.
- Farsalinos, K.E., et al., 2014b. Nicotine absorption from electronic cigarette use: comparison between first and new-generation devices. *Sci. Rep.* 4, 4133.
- Flouris, A.D., et al., 2013. Acute impact of active and passive electronic cigarette smoking on serum cotinine and lung function. *Inhal. Toxicol.* 25, 91–101.
- Flouris, A.D., et al., 2012. Acute effects of electronic and tobacco cigarette smoking on complete blood count. *Food Chem. Toxicol.* 50, 3600–3603.
- HC, Health Canada, 2012. Guidance Document. Conduct and Analysis of Comparative Bioavailability Studies. http://www.hc-sc.gc.ca/dhp-mps/alt_formats/pdf/prodpharma/applic-demande/guide-ld/bio/gd_cbs_etc_ld-eng.pdf. Last accessed 23.10.15.
- Heatherington, T.F., et al., 1991. The Fagerstrom test for nicotine dependence: a revision of the Fagerstrom tolerance questionnaire. *Br. J. Addict.* 86, 1119–1127.
- Hughes, J.R., 2007. Measurement of the effects of abstinence from tobacco: a qualitative review. *Psychol. Addict. Behav.* 21, 127–137.
- MHRA, 2015. Licensing Procedure for Electronic Cigarettes and Other Nicotine Containing Products (NCPs) as Medicines. <http://www.mhra.gov.uk/home/groups/comms-ic/documents/websiteresources/con297583.pdf>. Last accessed 23.10.15.
- Moyses, C., et al., 2015. Evaluation of a novel nicotine inhaler device: part 2-effect on craving and smoking urges. *Nicotine Tob. Res.* 17, 26–33.
- Polosa, R., et al., 2014. Effectiveness and tolerability of electronic cigarette in real-life: a 24-month prospective observational study. *Intern. Emerg. Med.* 9, 537–546.
- Roche, 2009. Cobas b 221 blood gas analyser system. Instructions for use. Roche Reference Number 03261395001. Roche Diagnostics, Charles Avenue, Burgess Hill, West Sussex, UK.

- Spindle, T.R., et al., 2015. Preliminary results of an examination of electronic cigarette user puff topography: the effect of a mouthpiece-based topography measurement device on plasma nicotine and subjective effects. *Nicotine Tob. Res.* 17, 142–149.
- van Staden, S.R., et al., 2013. Carboxyhaemoglobin levels, health and lifestyle perceptions in smokers converting from tobacco cigarettes to electronic cigarettes. *S Afr. Med. J.* 103, 865–868.
- Vansickel, A.R., et al., 2010. A clinical laboratory model for evaluating the acute effects of electronic “cigarettes”: nicotine delivery profile and cardiovascular and subjective effects. *Cancer Epidemiol. Biomarkers Prev.* 19, 1945–1953.
- Vansickel, A.R., Eissenberg, T., 2013. Electronic cigarettes: effective nicotine delivery after acute administration. *Nicotine Tob. Res.* 15, 267–270.
- Vardavas, C.I., et al., 2012. Short-term pulmonary effects of using an electronic cigarette: impact on respiratory flow resistance, impedance, and exhaled nitric oxide. *Chest* 141, 1400–1406.
- Walele, T., et al., 2016. A randomised, crossover study on an electronic vapour product, a nicotine inhalator and a conventional cigarette. Part A Pharmacokinetics Regul. Toxicol. Pharmacol. 74, 187–192.