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and the Environment
Ministry of Health, Welfare and Sport

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RIVM Letter report 2015-0144
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Synopsis

Electronic cigarettes, or e-cigarettes, vaporise a liquid that often contains nicotine and flavourants. Although E-cigarettes are less harmful than tobacco cigarettes, the e-cigarette vapour contains several ingredients and chemical impurities such as nicotine, propylene glycol, glycerol, aldehydes, nitrosamines and metals at concentrations that can be detrimental to health. Inhalation of these can lead to irritation of and/or damage to the respiratory tract, palpitations and an increased risk of developing cancer. These health effects are, however, less severe than those associated with smoking tobacco.

The Dutch National Institute for Public Health and the Environment (RIVM) undertook a survey of e-cigarette users (vapers), performed measurements and assessed the risks. The study was performed for the ministry of Health, Welfare and Sport (VWS) because of the substantial increase in e-cigarette users, and the uncertainties surrounding the health effects of e-cigarette use. In this study, the RIVM assessed the possible health risks associated with exposure to substances in e-cigarette vapour. In 2015, the RIVM will assess the possible health effects resulting from exposure to compounds present in exhaled vapour.

Findings regarding users
The survey of Dutch e-cigarette users showed that people use e-cigarettes on the assumption that it is less harmful than smoking conventional cigarettes and will help them to stop smoking. Of the many brands and models available, refillable e-cigarettes were the most popular. Virtually all users had smoked tobacco prior to using e-cigarettes and most were dual users, continuing to smoke tobacco in addition to using e-cigarettes. There were major behavioural differences amongst 'vapers', such as large variances in the number of puffs reported per day.

E-liquid and vapour composition
Considerable differences were observed in the composition of different kinds of e-liquid available in the Dutch market and that of the resulting vapour. In some cases, the amount of nicotine in the e-liquid did not match the declared amount on the packaging. The concentration of some compounds was found to be higher in the vapour than in the liquid. Aldehydes were formed when the e-liquids are heated, and metals were released from the atomiser. Propylene glycol and glycerol function as a 'carrier liquid' for nicotine and flavourants.

Keywords:e-cigarette, e-liquid, composition, exposure, risk assessment, health effects

Publiekssamenvatting

E-sigaretten, oftewel elektronische sigaretten, verdampen een vloeistof die meestal nicotine en een smaakstof bevat. De e-sigaret is weliswaar minder ongezond dan tabakssigaretten, maar de damp van e-sigaretten bevat een aantal ingrediënten en chemische onzuiverheden in hoeveelheden die schadelijk zijn voor de gezondheid. Het gaat onder andere om nicotine, propyleenglycol en glycerol en aldehydes, nitrosamines en metalen. Inhalatie hiervan kan leiden tot irritatie en schade aan de luchtwegen, hartkloppingen en een verhoogde kans op kanker. Deze gezondheidseffecten zijn wel veel minder ernstig dan die van tabak roken.

Dat blijkt uit onderzoek van het RIVM, waarvoor metingen zijn verricht, risicobeoordelingen zijn gedaan en gebruikers zijn geraadpleegd. Het onderzoek is in opdracht van VWS uitgevoerd vanwege de forse groei van het aantal e sigaretgebruikers en de onduidelijke gezondheidseffecten van het gebruik. Voor dit onderzoek is het risico voor gebruikers beoordeeld op basis van de stoffen in de damp. In 2015 gaat het RIVM de effecten van stoffen in uitgeademde damp op omstanders onderzoeken.

Bevindingen gebruikers
Uit het onderzoek blijkt dat mensen vooral e-sigaretten roken in de veronderstelling dat het minder schadelijk is voor de gezondheid dan een gewone sigaret en helpt om te stoppen met roken. Van de vele merken en modellen zijn navulbare e-sigaretten het meest populair. Vrijwel alle gebruikers rookten tabak voordat ze met de e-sigaret begonnen en de meesten gebruiken tabak naast hun e-sigaret. De ‘dampers’ verschillen sterk in hun dampgedrag, bijvoorbeeld in het aantal trekjes dat zij per dag gebruiken.

Samenstelling vloeistoffen en damp
De samenstelling van de vele soorten e-vloeistof op de Nederlandse markt en die van de resulterende damp blijken onderling sterk te verschillen. Soms komen de gevonden hoeveelheden nicotine in de vloeistof niet overeen met de gehalten die op de verpakking staan. Van sommige stoffen blijken de concentraties in de damp hoger te zijn dan in de vloeistof. Aldehydes ontstaan bij de opwarming van de vloeistoffen en metalen komen vrij uit de verdamper. Propyleenglycol en glycerol zijn ‘dragervloeistoffen’ voor nicotine en de smaakstoffen.

Kernwoorden: e-sigaretten, e-vloeistof, samenstelling, blootstelling, risicobeoordeling, gezondheidseffecten

Glossary of terms and abbreviations

DAD	diode array detector
GC	gas chromatography
HPLC	high-pressure liquid chromatography
ICP	inductively coupled plasma
LOD	limit of detection
LOQ	limit of quantification
mod	a customised e-cigarette assembled from separate components by the user
MoE	margin of exposure
MS	mass spectrometry
NIST	National Institute of Standards and Technology
NVWA	the Dutch Food and Consumer Product Safety Authority
PG	propylene glycol
PoD	point of departure
TBME	tert-butyl methyl ether
TSNA	tobacco-specific nitrosamines
vaporiser	part of a second-generation e-cigarette, which contains the liquid reservoir, heating element and mouthpiece
VOC	volatile organic compound

Contents

Summary

This report is a partial translation of RIVM report 2014-0143 and describes the most important results of a large-scale study of the composition of electronic cigarettes (e-cigarettes) and the health effects associated with their use.

An e-cigarette has a reservoir or cartridge filled with a carrier liquid, which contains flavourants such as tobacco flavour and menthol and usually also contains nicotine. The carrier liquid is evaporated by means of a battery-powered heating element. The use of e-cigarettes is increasing in the Netherlands. Many of the people taking up the habit are smokers who use e-cigarettes as well as, or instead of conventional cigarettes. However, it is not clear what the health effects of using e-cigarettes are, or how the effects compare with those of smoking tobacco. Although the presence of harmful substances in the liquid and the vapour has been investigated, most of the studies concerned were relatively small and did not include a risk assessment.

The research reported here began with a survey of e-cigarette users undertaken in collaboration with TNS NIPO and the Trimbos Institute. Respondents were asked which brands they used most and how they used e-cigarettes. In the next phase of the research, the chemical composition of the liquid and of the vapour was analysed. Then, drawing on the information gathered regarding the usage and composition of e-cigarettes, the health risks for users were estimated. The risk was also compared with the risks associated with smoking tobacco. Population-level effects, such as changes in the levels of tobacco product use, were not taken into consideration.

The market research involved a questionnaire-based survey of a representative group of 456 daily or weekly users of the e-cigarette in the Netherlands. The survey yielded data on the most widely used hardware and liquids and on the usage patterns of experienced e-cigarette-users. The findings indicate that almost all users are current or former tobacco smokers, who took up e-cigarettes in the belief that they would be less harmful and/or cheaper than tobacco and would help the user to stop smoking. By far the majority of current 'vapers' (as e-cigarette users are known) intend to go on using e-cigarettes for some time.

Usage habits – e.g. frequency of use – vary considerably. Daily users inhale from e-cigarettes several times a day, have longer vaping sessions (which involve *fewer* inhalations, however) and inhale a little more deeply than weekly users. Most of the scientific literature does not distinguish between different usage profiles, although a user's profile may affect the level of exposure.
It was found that numerous brands and models of e-cigarette are used, with refillable types being most popular. Three quarters of users refill their own e-cigarettes using proprietary liquids, most of which contain nicotine and tobacco flavour or menthol. Only 8 per cent use nicotine-free liquid; 42 per cent use low-strength liquids (up to 10 mg) and

45 per cent use high-strength liquids (11-23 mg). Extra-high-strength liquids (> 24 mg) are rarely used. Tobacco flavour is by far the most commonly chosen flavour, followed by menthol and fruit or (other) sweet flavours. Most users were using models that did not support battery voltage adjustment; the 10 per cent who used variable-voltage models mainly opted for an average voltage.

On the basis of their popularity with users, their flavours and their nicotine content, 183 e-liquids available on the Dutch market were selected for compositional analysis. All the tested e-liquids contained the carrier liquid propylene glycol (range 0-1.14 g/ml) and/or glycerol (range 0-1.16 g/ml). In two liquids, small quantities of diethylene glycol were detected, possibly indicating contamination of the carrier liquids. The nicotine content varied from 0 to 37.4 mg/ml; in fifteen of the 183 analysed e-liquids, the measured nicotine concentration differed from the supplier's stated value by more than 25 per cent. The latter observation may justify enforcement action with regard to the nicotine content of e-liquids, since the marketing of products whose composition is not as stated on the label is contrary to the Food and Commodities Act.

Formaldehyde was present in a measurable concentration in sixty-three of the 183 liquids, with the highest recorded concentration being 24 µg/ml. Measurable acetaldehyde concentrations were found in twelve liquids, the highest being 300 µg/ml. Acrolein was detected in four liquids, at a maximum concentration of 1.6 µg/ml. The flavourant diacetyl was present in thirty-four liquids, with the highest concentration found being 5591 µg/ml. Almost all samples contained other aldehydes and ketones besides the four substances mentioned above, sometimes in high concentrations, probably due to use as flavourants. For practical reasons, the presence of VOCs and TSNAs was investigated in a sample group of sixty liquids. Only two of the liquids were found to have a measurable concentration of VOCs: 9.5 µg/ml of benzene and 0.58 µg/ml toluene. In fifteen of the sixty tested liquids, a measurable quantity of one or more TSNAs was present, the highest concentration detected being 80 ng/ml. In addition, various metals were found in extremely varied concentrations.

In order to determine what other substances are present in the e-liquids, we analysed sixty of the 183 e-liquids by means of headspace GC-MS. Using this method, roughly a further 150 substances were detected, many of them flavourants.
For each category of substance, fifteen different e-liquids were vaporised using a vaping robot and a commercially available vaporiser**.** The carrier liquids glycerol and propylene glycol and nicotine were found to be almost completely converted to vapour. However, the concentrations of aldehydes and most metals were many times higher in the vapour than in the liquid. It seems likely that the aldehydes are formed by heating the carrier liquids and that metals emanate from the vaporiser itself. By comparing various types of vaporiser, it was established that the vaporiser has a major bearing on the concentrations of aldehydes in the vapour. Further research is needed to determine why that is the case and whether the choice of vaporiser also influences the concentrations of other substances in the vapour.

On the basis of the information on e-cigarette usage and composition, a risk assessment was made. First, an exposure scenario was developed, including details of the substance(s) to which users are exposed. Information was then sought regarding the relationship between exposure to each of the substances in question and potential adverse health effects. The selection of information took account of relevance to the exposure scenario in terms of, for example, duration, frequency and level of exposure. Using the selected information, the likelihood of the exposure scenario giving rise to health risks was estimated, allowing for the degree of consistency between the health effect information and the exposure scenario.
The substance-specific comparison indicated that the concentrations of most relevant substances in vapour from e-liquids are lower or much lower than that in tobacco smoke. Only the concentrations of the carrier liquids glycerol and propylene glycol are higher in vapour than in tobacco smoke. The main effects of those substances are damage to the respiratory tract and, in the case of propylene glycol, effects on leukocytes. In addition, the concentration of formaldehyde can be up to three times higher in e-cigarette vapour than in tobacco smoke from cigarettes. On the other hand, for example, the concentrations of carcinogenic tobacco-specific nitrosamines in tobacco smoke are up to four hundred times higher than in e-liquid vapour. Furthermore, tobacco smoke contains thousands of other substances, many of which are toxic. Consequently, the toxic substance-related health risks associated with the use of tobacco cigarettes are far greater than those associated with e-cigarettes.

Various health effects associated with tobacco use are not known to be linked to any individual substance present in e-liquid vapour. However, it should be noted that extensive epidemiological data on the health effects of tobacco use are available, while the availability of comparable data on the use of e-cigarettes or on individual vapour components is limited. The health risks associated with using tobacco cigarettes are considerably higher than those associated with e-cigarettes, assuming comparable usage patterns (a similar number of inhalations over a comparable period). The health risks are of course strongly dependent on individual vaping and smoking habits. Furthermore, the research reported here considered only the risk for the individual user. Effects at the population level, such as changes in the levels of e-cigarette and tobacco use, have not been considered. Population-level effects may occur as a result of, for example, former smokers continuing to vape and non-smokers taking up vaping in the belief that it is relatively harmless. E-cigarette use may also support continued nicotine addiction amongst smokers.

Nevertheless, daily use of e-cigarettes is not without health risks. Exposure to the polyols can damage the respiratory tract and influence the leukocyte pattern. Nicotine-containing e-liquids can also affect health in various ways. Furthermore, while the vapour concentrations of tobacco-specific nitrosamines are many times lower than in tobacco smoke, they are sufficiently high in some cases to give an elevated risk of tumour development. The vapour concentrations of aldehydes can also be sufficient to induce effects on the respiratory tract, although the concentrations in question are probably attributable to the heating

process rather than the properties of particular e-liquids. Health risks are liable to increase sharply at higher intensities of use. However, insufficient dose-response relationship data are available to support evaluation of the complex and fluctuating exposure patterns associated with e-cigarette use.
Finally, the concentrations of cadmium, lead, nickel and arsenic in smoke from tobacco cigarettes are (considerably) higher than those found in e-cigarette vapour, while the chromium concentrations are comparable. While the possibility of health effects at the measured metal concentrations cannot be excluded, it is safe to say that the risks are lower with vaping than with smoking.

1 Introduction

E-cigarette use in the Netherlands is increasing. Although in 2013 only 3 per cent of Dutch people above the age of fourteen reported using an e-cigarette at least once in the previous year, that represents a threefold rise in the space of a year. In other countries, such as the United Kingdom, usage is increasing even more quickly. The number of adult users in the UK is now estimated to be 2.1 million (ASH.), although the situation appears to have stabilised in 2014 (www.smokinginengland.info). The great majority of e-cigarette users are people who also smoke conventional cigarettes. In 2014, 20 per cent of British smokers were regular e-cigarette users as well.

Doubt remains regarding the exact risks and benefits of e-cigarette use (Callahan-Lyon 2014, Grana, Benowitz et al. 2014). A series of studies recently reported in *Tobacco Control* (May 2014, volume 23, supplement 2) underlined the uncertainty. There is ample evidence, however, that using e-cigarettes is less harmful than smoking tobacco products (Farsalinos and Polosa 2014, Hajek, Etter et al. 2014, McNeill, Etter et al. 2014). On the other hand, quality control during production can leave something to be desired and e-cigarettes are not as safe as nicotine-replacement products such as patches (DKFZ 2013). Various studies have shown that the correlation between the measured nicotine concentrations in refill cartridges and e-liquids and the concentrations stated on the labels is poor. Although there is no convincing evidence of non-smokers becoming addicted to nicotine as a result of using e-cigarettes, the possibility of the activity acting as a gateway to smoking, especially amongst young people, should not be overlooked. E-cigarette use by smokers may also have the effect of sustaining nicotine addiction.

Not enough is yet known about the hazardousness of using e-cigarettes ('vaping'), or about the amounts of the various toxic and other substances that enter the body as a result of the practice (Orr 2014). E-cigarettes do not burn like conventional cigarettes, but produce a vapour that contains flavourants, carrier substances such as (vegetable) glycerol and propylene glycol, often nicotine, heavy metals, particulates, known carcinogens (substances that cause cancer, such as the nitrosamines NNK and NNN, acetaldehyde, formaldehyde, acrolein) and other chemicals or contaminants produced during production and use (Grana, Benowitz et al. 2014, Hajek, Etter et al. 2014). Users report undesirable effects such as throat irritation, coughing, nausea and vomiting. The reported side-effects are generally mild and, although causal relationships between the reported problems and the use of e-cigarettes are likely, they have not been proven. Because e-cigarettes have not been on the market for very long, the long-term health effects of their use has not yet been properly determined. Furthermore, e-cigarette use is liable to involve the exposure of non-users to exhaled vapour, which can contain low concentrations of harmful substances (Schripp, Markewitz et al. 2013, DKFZ 2013).

In view of the rapid growth in the number of users and the absence of conclusive scientific evidence regarding the safety of e-cigarettes, the RIVM has undertaken the risk assessment of which the main results are reported here and, in more detail, in RIVM report 2014-0143 (Visser W 2015). In 2013, the RIVM published a factsheet on trends in e-cigarette use and the associated risks, which was compiled on the basis of literature research (RIVM 2014). The RIVM had previously made a risk assessment of the shisha pen. That risk assessment was based on information about the main substances present in shisha pen vapour, namely glycerol and propylene glycol (RIVM 2013). In 2014, an analysis of the composition of e-cigarettes on the Dutch market was performed and the findings used to estimate the risk, both in absolute terms and in comparison with tobacco smoking. The most important results are presented in this report, which is a partial translation of the RIVM report 2014-143.

1.1 Exposure and risks

This report describes a risk assessment of e-cigarettes, focusing on the composition of e-cigarette liquids, the vapour produced from them and the effects of that vapour on users. A quick scan of recent literature revealed that there remains a serious shortage of knowledge regarding the health effects of e-cigarette use and regarding the test models (analysis methods, conditions and parameters) (Cheng 2014). A series of studies was recently reported in *Tobacco Control* (May 2014, volume 23, supplement 2), summarising what is currently known about health effects, product properties, the topography of e-cigarette use, the chemical composition and nicotine transfer.

E-cigarettes may be divided into three basic types:

- First-generation e-cigarettes look most like conventional cigarettes. They are available for a few euros from drug stores, handed out at fairs, etc. E-cigarettes of this type are regarded as entry-level products. Some have rechargeable batteries and replaceable e-liquid cartridges, but many are disposable products.
- Second-generation e-cigarettes are more expensive and usually offer better perfomance. They have rechargeable batteries and user-refillable e-liquid reservoirs. Parts made by different manufacturers are interchangeable due to dimensional standardisation.
- Personalised vaporizers, also known as 'mods' are e-cigarettes assembled by users from separate components. They often have high-capacity, high-power batteries and large e-liquid reservoirs or 'tanks'. Such characteristics enable greater vapour production, which users regard as an advantage.

A survey carried out in January of 2014 (Zhu, Sun et al. 2014) put the number of brands available over the internet at 466. The researchers identified 7,764 unique flavours of e-liquid (now also known as 'e-juices'). It was noted that many supposedly new products consist of established hardware combined with a new e-juice. Such combinations are often marketed as completely new products, when in fact they may more accurately be described as creatively packaged new flavours.

(Orr 2014) emphasised that the design or model of e-cigarette used is an important determinant of the toxic substances produced and their concentrations in the aerosol. The design influences pressure drop, airflow, aerosol particle size, substance absorption, etc. The huge variety of models available therefore implies major differences in toxic profile, associated with both the hardware and the composition of the e-liquid. By way of example, Orr points out that non-pharmaceutical nicotine contains higher levels of tobacco-specific nitrosamines (TSNA) and other contaminants than pharmaceutical nicotine, although most published data indicate that TSNA levels in e-cigarette cartridges are very low. Without clarity as to which aspects of e-cigarette design have the greatest bearing on the production of toxic substances, it remains difficult to extrapolate toxicity findings from one model to another. A further complication is the lack of consensus as to which toxic and other substances or biomarkers are most indicative of the hazardousness of an e-cigarette.

The materials from which an e-cigarette is made (e.g. metals, rubber, ceramics) can influence the processes that occur during e-cigarette use (particularly the heating of the liquid to create the aerosol), with possible adverse health implications (Brown and Cheng 2014).

Experienced vapers can have plasma nicotine levels as high as those found in smokers (Schroeder and Hoffman 2014). However, the nicotine 'yield' of an e-cigarette (the amount of nicotine released and available for consumption by inhalation) is lower than that of a conventional cigarette. In addition, behavioural factors (the manner of inhalation) can have a major bearing on how much of the available nicotine actually finds its way into in the bloodstream.

- Where ordinary tobacco is concerned, the pharmacokinetics of nicotine are linked to the manner of administration, the pH of the tobacco, the pH of the smoke, the interval between the inhalations, the duration of individual inhalations, etc. It was once established that the nicotine yield of Marlboro cigarettes was 152-193 µg nicotine/100 ml inhaled (Trehya ML 2011). When tested in smoking machines, the nicotine yields of conventional cigarettes are found to vary between 0.5 and 1.5 mg/cigarette (Schroeder and Hoffman 2014).
- The nicotine yield of an e-cigarette is much lower than that of a conventional cigarette (varying greatly, from 0-43 µg nicotine/100 ml inhaled; Trehya *et al* 2011; 83 µg nicotine/100 ml inhaled with a 18 mg/ml nicotine cartridge (Goniewicz, Lingas et al. 2013), converted by (Schroeder and Hoffman 2014). However, the use of smoking machines to study e-cigarettes requires different assumptions and methods from those used in the study of conventional cigarettes; consequently, nicotine yield data for e-cigarettes are not directly comparable with data for conventional cigarettes.
- In an e-cigarette, the temperature has a direct effect on the nicotine yield in the vapour (higher temperatures lead to increased aerosol formation). Features of an e-cigarette's design (rechargeable battery, tank, variable voltage) can also influence the nicotine yield. The vaper's level of experience has an important bearing on the plasma nicotine concentration attained.

In addition to the composition of the liquid, the design of the e-cigarette and how dirty or worn it is are also significant determinants of the presence of toxic and other substances in the aerosol (Schroeder and Hoffman 2014).

- (Farsalinos, Spyrou et al. 2014) demonstrated that the nicotine yield of first-generation e-cigarettes is lower than that of later models.
    - People who vaped under experimental conditions for one hour using a new model of e-cigarette filled with liquid containing 18 mg/ml nicotine were found to attain 35-72 per cent higher plasma nicotine levels than those obtained by vapers using old models filled with the same liquid.
    - After five minutes' use, the e-cigarettes had yielded between a quarter and a third of the nicotine yielded by a conventional cigarette (five minutes being the average time needed to smoke a conventional cigarette).
    - To reach the same nicotine level as that attained by smoking a conventional cigarette, experienced vapers needed to use a new high-voltage model for thirty-five minutes; after sixty-five minutes of vaping with a first-generation model containing a liquid with a nicotine concentration of 18 mg/ml, plasma nicotine concentrations had not reached the level associated with smoking a conventional cigarette for five minutes.

(Farsalinos, Romagna et al. 2013) undertook an internet survey of e-liquid flavour preferences amongst 4,618 vapers (recruited via popular e-cigarette websites). Tobacco flavours were found to appeal most to new vapers, while fruit flavours proved most popular in this survey of predominantly experienced e-cigarette users (average twelve months' experience). On average, such vapers used three different flavours, often using more than one on the same day. Respondents said that varying flavours catered for personal taste and was very important in helping them to cut down on or stop smoking.
From the general literature scan, it is clear that there is no consensus regarding many of the possible input parameters for analysing the risks of e-cigarette use.

1.2 This report

The first part of the risk analysis presented in this report describes a questionnaire-based market survey of 456 daily or weekly e-cigarette users in the Netherlands. Respondents were asked about the products they use, how they use them and the circumstances under which they do so. The market research was undertaken in collaboration with TNS NIPO and the Trimbos Institute. It yielded data on the most widely used hardware and liquids and the usage patterns of experienced e-cigarette users. Those data served as input for the remaining phases of the risk assessment. The subsequent sections of the report describe the results of the e-liquid and vapour analyses and the assessment of the risk to the user. The composition of 183 popular e-liquids, including nicotine-containing and nicotine-free products and products of various flavours, was analysed. Using a vaping robot and various types of commercially available vaporisers, some of the e-liquids were heated to form vapour,

which was collected and tested. The usage and composition data were then used to estimate the risk to users. The quantities of the relevant substances to which consumers are exposed were compared with the quantities known to have adverse health effects. The risks of e-cigarette use were then compared with the risks of tobacco smoking. The risk estimates presented here relate only to the individual user. Effects at the population level, such as changes in the levels of e-cigarette and tobacco use, have not been considered.
The main results of the conducted studies are included in this report. More detailed information can be found in RIVM report 2014-0143 (Visser W 2015).

2 Market research

This section of the report summarises the main results of the market research.

2.1 Research questions

The market research focused on two questions:

1. What models and flavours of e-cigarette and e-liquid are used by regular e-cigarette users in the Netherlands?
2. What patterns of use do regular users display, what is the duration of their exposure, and what is the 'topography' of their use (i.e. the average number, depth and duration of inhalations)?

2.2 Results

The 456 respondents included slightly more women (59 per cent) than men. The majority were between thirty-five and sixty-four years old. Almost all respondents were smokers (83 per cent) or ex-smokers (16 per cent) (see figure 2-1). At the time they completed the questionnaire, fifty respondents had stopped using e-cigarettes (ex-vapers).

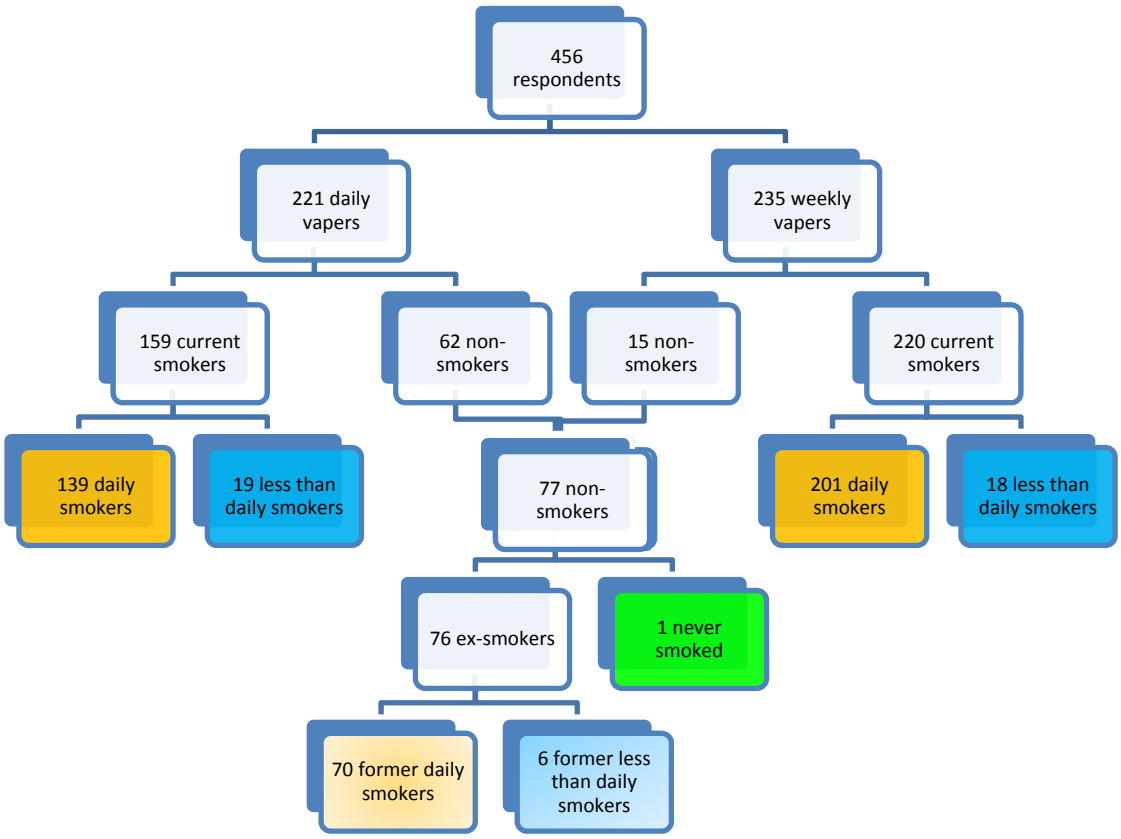

Figure 2-1: breakdown of vaping and smoking habits amongst respondents. Due to weighting and rounding, some of the stated values do not equal the sum of the values in boxes below.

Almost all the current and former e-cigarette users were tobacco smokers before they started vaping. Eighty-two of the respondents have used both conventional tobacco products and e-cigarettes during the same period ('dual use'). Of the dual users who are daily vapers, 56 per cent use e-cigarettes more than conventional cigarettes. By contrast, nearly three quarters of weekly vapers use tobacco products more than e-cigarettes. Roughly one in six use e-cigarettes and tobacco products about equally often, or have variable habits, sometimes vaping more than smoking and sometimes the other way around. The number of ex-vapers who responded was too small to support conclusions. Some 95 per cent of respondents had been vaping for at least two years. Just over half had been vaping for less than six months.

The main reason given for using e-cigarettes was that they were believed to be less harmful than conventional cigarettes. For nearly half of respondents, the (lower) price is an important motivator and 43 per cent expect e-cigarettes to help them stop smoking. Few people said

that they like the taste of e-cigarettes, or regard them as 'cool' or attractive.

Most vapers use e-cigarettes while alone. The percentage of e-cigarette users that vape in company of others is significantly higher amongst current weekly vapers (15 per cent) than amongst current daily vapers (5 per cent). Amongst ex-vapers, there is little difference. The most common places for vaping are at home and in the car. Vapers who use e-cigarettes less than daily more frequently indicated mainly vaping away from home (when visiting friends, in a restaurant or café).

Brands and models
Respondents had the opportunity both to select named brands and models and to enter other brands and models in a free-input field. The responses indicated that a large number of different products are in use. The main conclusions are:

- The most popular models are the second-generation eGo and EVOD, which feature interchangeable components.
- Daily vapers are more likely to use second-generation models, while weekly vapers tend to use first-generation entry-level models, which look more like conventional cigarettes.
- Notably, weekly vapers significantly more often indicated not to know what brand or model they use.

Filling and battery
Amongst current users, 86 per cent refill their own e-cigarettes, mostly with proprietary liquid. A similar percentage of ex-vapers (82 per cent) used to refill their e-cigarettes.

The liquids used are predominantly low-strength nicotine products: the most common strength used is 11-15 mg and 42 per cent of respondents preferred a very low strength (up to 10 mg). Only 8 per cent of current and ex-vapers use nicotine-free liquid. Nicotine strengths of 24 mg or higher are rarely used.

Users' liquid flavour preferences were determined by means of an open question. The responses were grouped into six categories: tobacco flavour (58 per cent), menthol (16 per cent), fruit (11 per cent), sweet (8 per cent), other (4 per cent) and no flavour (3 per cent)

Some models of e-cigarette feature variable-voltage batteries, enabling the user to regulate how much the e-liquid is heated. Because this can have implications for the composition of the vapour, it was important to determine whether users make use of this feature and, if so, what voltage they select. Some 10 per cent of current and ex-vapers reported using a model with a variable-voltage battery. Three quarters of those users said they used a medium voltage, while 10 per cent opted for a low-voltage setting and 10 per cent for a high-voltage setting.

Usage patterns
Daily and weekly vapers differ in the number of times per day that they use e-cigarettes.

- Two thirds of weekly vapers vape one to five times per usage day.

- Daily vapers vape more frequently on each day
- On average, weekly vapers vape 6.9 times per usage day, while daily vapers do so 16.4 times per day.
- No differences were found between current vapers and ex-vapers.

Weekly vapers inhale slightly more often per vaping session (7.6 times) than daily vapers (6.9 times). The difference is statistically significant, but small. On average, ex-vapers used to inhale less often: ex-daily vapers 4.2 times and ex-weekly vapers 6.1 times. In terms of average vaping session duration, no significant differences were found between weekly and daily vapers or between current vapers and ex-vapers.

On a scale of one to ten (where 'one' represents taking the vapour into the mouth only and 'ten' represents inhaling it deeply into the lungs), daily vapers are a little more likely to inhale deeply (average score 7.3) than weekly vapers (average 6.7). Amongst both daily vapers and weekly vapers, roughly half inhaled about as deeply when vaping as when smoking a conventional cigarette. A third of respondents in each group said that they believed they inhaled conventional cigarette smoke more deeply.

According to almost half of respondents, the duration of each inhalation is slightly longer with an e-cigarette than with a conventional cigarette. On the other hand, half of respondents said that the average session length was longer when smoking a conventional cigarette than when vaping an e-cigarette. That is consistent with one of the comments given in the questionnaire responses, namely that "it's easier to put down an e-cigarette; you always finish a conventional cigarette".

Expectations and stopping vaping

Amongst current e-cigarette users, 42 per cent are satisfied with the product and have no plans to stop vaping. A similar percentage (44 per cent) don't plan to stop soon but do ultimately intend to stop vaping. Seven per cent plan to stop in the near future. Of the latter group, three quarters want to stop smoking *and* vaping and an eighth expect to take up smoking again. None of the respondents said that they planned to switch to another nicotine replacement product, such as patches or gum.

Nearly half said e-cigarettes had not helped them stop smoking. A quarter were stopping vaping because they didn't like it and 22 per cent had a pragmatic or other reason.

2.3 Conclusions

The market research reported here was designed to yield data on a number of usage parameters to serve as input for an analysis of the risks of e-cigarette use. For pragmatic reasons, a questionnaire-based survey was performed on a representative group of experienced e-cigarette users in the Netherlands. Such research relies on self-reporting and that is one of the main limitations. A further complicating factor is that the range of e-cigarettes and e-liquids available is very wide and subject to rapid change. As a result, the data obtained will become outdated within a relatively short space of time. Despite those

limitations, the market research proved to be an expeditious means of obtaining an overview of e-cigarette usage patterns in early 2014.
The main conclusions are:

- Almost all e-cigarette users also smoke tobacco or used to do so; nearly half are seriously nicotine dependent, as indicated by the fact that they light a cigarette within half an hour of getting up in the morning.
- The three main reasons for using e-cigarettes are that they are assumed to be less harmful, that they are cheaper than tobacco and to help the user stop smoking.
- E-cigarettes are used almost everywhere: at home, in the car, when visiting friends or relatives, in the street or the countryside, at work, when out for the evening, etc.
- Many different brands and models are used, but second-generation products are the most popular. Daily and weekly smokers were found to differ in their model preferences: daily smokers have more specific requirements and wishes.
- Three quarters of experienced users refill their e-cigarettes themselves, using proprietary liquids; 10 per cent prepare their own liquids.
- Only 8 per cent use nicotine-free liquid; 42 per cent use low-strength liquids (up to 10 mg) and 45 per cent use high-strength liquids (11-23 mg). Extra-high-strength liquids (> 24 mg) are rarely used.
- Tobacco flavour is by far the most popular flavour, followed by menthol. A fifth of vapers prefer fruit or (other) sweet flavours.
- Most users do not use models with variable-voltage batteries; the 10 per cent who do use such models mainly select a medium voltage.
- There are clear differences between daily and weekly vapers in terms of the degree of exposure to the vapour that need to be taken into account in the risk analysis. Daily users vape more frequently per day and inhale slightly deeper than weekly vapers. Most scientific literature on this subject makes no distinction between different vaping profiles, although there may be implications for the level of exposure.
- The great majority of current vapers intend to continue using e-cigarettes for some time.

3 Composition of e-liquids

3.1 Introduction

The composition of 183 e-liquids currently available on the Dutch market was investigated. The findings are summarised in this section. A more detailed description of the results and methods is provided in RIVM report 2014-143 (in Dutch) (Visser W 2015).

3.2 Results and discussion

The concentrations of the following substances were determined in the e-liquids:

- Polyols such as propylene glycol and glycerol (the carrier liquid)
- Nicotine
- Aldehydes and ketones
- Volatile organic compounds (VOCs)
- Tobacco-specific nitrosamines (NNN, NAT, NAB and NNK)
- Metals

The results of the tests are summarised in table 3-1. The full results are included in the appendix (www.rivm.nl/bibliotheek/rapporten/2015-0144_data.xlsx). Additional tests were performed to establish what other substances were present in the e-liquids. The results of those tests are also included in the appendix.

Table 3-1: Summary of e-liquid analysis results. The values under 'range' are the lowest and highest measured values. For the calculation of the median, all samples were included (including samples for which the measured concentration was below the detection limit. n=60 for the nitrosamines, n=183 for the other values). LOQ stands for 'limit of quantification'.

	number	**range**			
	>LOQ	min	max	**median**	**unit**
carrier liquid and nicotine					
nicotine	140	< 0.01	38	12	mg/ml
propylene glycol	177	< 0.005	1.14	0.73	g/ml
glycerol	176	< 0.002	1.16	0.41	g/ml
di-ethylene glycol	2	< 0.07	3.6	< 0.07	mg/ml
tri-ethylene glycol	1	< 0.2	24	< 0.2	mg/ml
aldehydes					
formaldehyde	63	<1	24	<1	µg/ml
acetaldehyde	12	<10	300	<10	µg/ml
acrolein	4	<1	1.6	<1	µg/ml
diacetyl	34	<100	5591	<100	µg/ml
nitrosamines					
NNN	2	<1	49	<1	ng/ml

NAT	5	<1	35	<1	ng/ml
NAB	7	<1	9	<1	ng/ml
NNK	13	<1	42	<1	ng/ml
metals					
vanadium	6	<5.0	60	<5.0	ng/ml
chromium	77	<5.0	2243	<5.0	ng/ml
manganese	71	<5.0	7613	<5.0	ng/ml
cobalt	8	<5.0	482	<5.0	ng/ml
nickel	27	<10	225900	<10	ng/ml
copper	80	<5.0	45540	<5.0	ng/ml
zinc	155	<10	55295	28	ng/ml
arsenic	77	<5.0	35	<5.0	ng/ml
molybdenum	16	<5.0	53	<5.0	ng/ml
cadmium	6	<1.0	81	<1.0	ng/ml
tin	29	<5.0	2E+06	<5.0	ng/ml
lead	16	<5.0	4931	<5.0	ng/ml
uranium	2	<1.0	2.2	<1.0	ng/ml

3.2.1 Comparison of actual and labelled nicotine concentrations

The amount of active ingredient in a pharmaceutical product is not allowed to deviate by more than 10 per cent from the declared value. Given the hazardous nature of nicotine, it is the therefore striking that in sixty-seven (37 per cent) of the 183 e-liquids, the measured nicotine concentration was more than 10 per cent different from the value stated on the packaging. In most cases, the measured nicotine concentration was lower than the declared concentration, but there were a few exceptions. For example, nicotine was present in three e-liquids labelled as nicotine-free. In April 2014, the NVWA (the Dutch Food and Consumer Product Safety Authority) reported on the chemical product safety requirements of e-cigarettes and shisha pen refill packaging (NVWA 2014). The measured nicotine concentrations in the 206 e-liquids tested by the NVWA ranged between 0 and 35 mg/ml. In seven products, the measured nicotine level was 10 per cent higher than the stated level. The biggest divergence involved a liquid labelled as containing 0 mg/ml, which actually contained 17 mg/ml. The factsheet does not indicate whether any products were found of which the measured nicotine concentrations were significantly lower than their stated concentrations.

Ten of the 183 liquids that we tested contained nicotine at a concentration exceeding 20 mg/ml (including six with a concentration exceeding 25 mg/ml), which is higher than the maximum permitted under the revised Tobacco Products Directive. Nine of the 206 liquids tested by the NVWA had concentrations exceeding 20 mg/ml.

3.3 Conclusions

The nicotine concentrations of 37 per cent of e-liquids were found to deviate by more than 10 per cent from the concentrations stated on the packaging or indicated by the retailer. In most cases, the actual nicotine concentration was lower than the stated concentration, but in a few cases considerable amounts of nicotine were present in e-liquids labelled as nicotine free.

In addition to the ingredients used by the manufacturer, e-liquids contained substances that were present as contaminants in the raw materials. In the context of this study, any substance other than nicotine, propylene glycol, glycerol and aromatic substances and flavourants is regarded as a contaminant. The concentrations of contaminants differs considerably between e-liquids. A small proportion of liquids contain diethylene glycol, benzene, toluene or TSNAs, but those substances were not demonstrably present in the great majority of liquids. Of the tested liquids, 45 per cent were found to contain measurable concentrations of short-chain aldehydes or ketones, and all the liquids had measurable quantities of one or more metals.
Many substances will pass into in the vapour unchanged, while others will decompose under the influence of heat during vaping. The following section describes the results of our analysis of the vapour composition.

4 Composition of the vapour

4.1 Introduction

The main results of the assays performed to determine the composition of e-cigarette vapour are summarised in this section. A more detailed description of the results and methods is provided in RIVM report 2014-143 (in Dutch) (Visser W 2015).

A vaping robot was used in order to sample the vapour in a reproducible manner. The influence of various factors was investigated, including the composition of the e-liquid, the model of e-cigarette, the voltage of the battery and the topography.

4.2 Result summary and discussion

Naturally, the composition of the e-liquid used has a major bearing on the composition of the vapour. The vapour from a number of different e-liquids was therefore analysed. The concentrations of the following substances in the vapour were measured:

- Propylene glycol and glycerol (the main ingredients of the carrier liquid)
- Nicotine
- Aldehydes and ketones
- Volatile organic compounds (VOCs)
- Tobacco-specific nitrosamines (NNN, NAT, NAB and NNK)
- Metals

The results are summarised in table 4-1. The full results are included in the appendix (www.rivm.nl/bibliotheek/rapporten/2015-0144_data.xlsx).

Table 4-1: Summary of the analysis of carrier liquid components and nicotine concentrations in the vapour. The values under 'range' are the lowest and highest measured values. For the calculation of the median, all samples were included (including samples for which the measured concentration was below the detection limit. n=12 for the nitrosamines, n=17 for the other values). LOQ stands for 'limit of quantification'.

	number	**range**			
	>LOQ	min	max	**median**	**unit**
carrier liquid and nicotine					
nicotine	14	0.001	0.142	0.051	mg/puff
propylene glycol	16	< 0.05	6.8	2.8	mg/puff
glycerol	17	< 0.02	5.0	2.7	mg/puff
di-ethylene glycol	2	< 0.6	18.0	< 0.6	µg/puff
tri-ethylene glycol	2	< 1.6	93.0	< 1.6	µg/puff
aldehydes					
formaldehyde	11	<0.2	33	0.2	µg/puff
acetaldehyde	1	<2	4.7	<2	µg/puff

acrolein	2	<0.2	3.3	<0.2	µg/puff
diacetyl	2	<10	16	<10	µg/puff
nitrosamines					
NNN	1	< 0.6	269	< 0.6	pg/puff
NAT	6	< 0.6	85	0.3	pg/puff
NAB	2	< 0.6	10	< 0.6	pg/puff
NNK	9	< 0.6	122	4.0	pg/puff
metals					
vanadium	3	< 0.05	0.11	< 0.05	ng/puff
chromium	16	< 0.05	9.3	6.7	ng/puff
manganese	7	< 0.05	0.47	< 0.05	ng/puff
cobalt	7	< 0.05	0.58	< 0.05	ng/puff
nickel	7	< 0.1	6.4	< 0.1	ng/puff
copper	17	0.38	24	2.1	ng/puff
zinc	17	2.7	67	17	ng/puff
arsenic	0	< 0.05	< 0.05	< 0.05	ng/puff
molybdenum	4	< 0.05	1.3	< 0.05	ng/puff
cadmium	10	< 0.01	0.10	0.01	ng/puff
tin	17	0.72	86	1.1	ng/puff
lead	17	0.16	2.1	0.59	ng/puff
uranium	0	< 0.01	< 0.01	< 0.01	ng/puff

4.2.1 *Influence of e-cigarette brand/model on concentrations of short-chain aliphatic aldehydes and ketones in the vapour*

Because aldehydes and ketones may be formed by the decomposition of carrier liquid constituents under the influence of heat, the influence of vaporiser brand and model on the formation of aldehydes was investigated. The tests revealed that the aldehyde concentrations in the vapour produced by various vaporisers filled with the same e-liquid differed considerably. Two vaporisers made by different manufacturers but filled with the same e-liquid were found to differ by a factor of a hundred. Two seemingly identical vaporisers made by the same manufacturer yielded vapour formaldehyde concentrations that differed by a factor of twenty-five. The variation in aldehyde concentrations is therefore almost entirely attributable to the vaporiser. The type of e-liquid and the aldehyde concentration in the e-liquid play no significant role.

4.3 Conclusions

The experimental approach allowed measurements of e-cigarette vapour composition. The concentrations of the various substances in the vapour appear to be determined principally by the e-liquid used, the model of e-cigarette used and of the behaviour of the user.
Carrier liquids and nicotine were almost completely vaporised, and their concentrations in the vapour are therefore determined nearly entirely by the power output of the vaporiser and the behaviour of the user.
Similarly, the harmful substances diacetyl, benzene and toluene were present in the vapour only if they were also present in the e-liquid.
However, our results show that short-chain aldehydes and ketones present in the vapour do not originate from the e-liquid, but are formed

during vaporisation. Propylene glycol and glycerol may partially decompose when heated, but the precise mechanism remains unresolved. The concentrations of those substances in the vapour varied greatly. Two apparently identical vaporisers made by the same manufacturer and filled with the same e-liquid yielded vapour formaldehyde concentrations that differed by a factor of more than twenty-five.

5 E-cigarette risk assessment

The main results and conclusions of the risk assessment of e-cigarette use are summarised in this section. A more detailed description of the results and methods is provided in RIVM report 2014-143 (in Dutch) (Visser W 2015).

5.1 Introduction

The risk assessment presented in this section is concerned with e-cigarette use and e-liquid composition in the Netherlands, as described in the previous sections. From the information in those sections, it will be apparent that exposure to substances due to e-cigarette use is a dynamic process, which is influenced by a wide variety of factors and liable to sizeable inter-individual variation. Significant factors include the concentrations in the inhaled vapour, the duration of exposure, the frequency of exposure events (vaping sessions) and the frequency of inhalation during vaping sessions.
Because of the extremely variable individual differences in the level of exposure, our risk assessment is based upon three pre-defined exposure scenarios for daily users. First, an assessment was made of the potential risk for an intensive e-cigarette user ('heavy vaper'). The rationale being that, if the assessment indicated that vaping entailed no health risk for a heavy user, no risk assessment would be necessary for other user groups. If, on the other hand, it appeared that the possibility of health effects for heavy vapers could not be excluded, we would proceed to assess the potential risks for less intensive users.
If inhaled, many substances are liable to irritate and possibly damage the respiratory tract. That is the case with many of the substances found in e-cigarette vapour, such as aldehydes, propylene glycol and glycerol (polyols). As well as affecting the respiratory tract, an inhaled substance may adversely affect health after absorption by the body. These health effects are known as systemic effects. Where possible, the risks of systemic effects are assessed on the basis of information from studies involving inhalatory exposure. Where no good inhalatory exposure data are available, data from studies involving other routes of exposure (e.g. oral ingestion) may under certain conditions be used. In such circumstances, the best possible account is taken of the differences between the exposure routes, e.g. with regard to the level and speed of the substance's absorption by the body.

5.2 Methodology

The exposure scenario for e-cigarette use is complex and shaped by numerous factors, of which some depend considerably on the individual user's vaping habits. The vaping topography information used for our assessment is derived from scientific literature (Farsalinos, Romagna et al. 2013, Goniewicz, Kuma et al. 2013, Hua, Yip et al. 2013).
The market survey (chapter 2) yielded information on the frequency and nature of e-cigarette use. On the basis of those data, three types of *daily* user have been defined: a 'light' vaper, an 'average' vaper and a 'heavy' vaper.

That led to definition of the following three exposure scenarios:

- Light vaper: fifteen inhalations per day, with a total daily vaping duration of sixty minutes
- Average vaper: sixty inhalations per day, with a total daily vaping duration of 120 minutes
- Heavy vaper: five hundred inhalations per day, with a total daily vaping duration of 240 minutes

The risk assessment is based primarily on the vapour concentration data for two e-liquids (no. 6 and no. 172), because in the case of those two e-liquids vapour concentrations were established for all substance groups (polyols, nicotine, aldehydes/ketones, tobacco-specific nitrosamines and volatile organic compounds). Where vapour concentrations in other e-liquids were higher, that has been taken into account in the risk assessment.

5.2.1 Discussion and conclusions

The vapour from two e-liquids (no. 6 and no. 172) underwent all the analyses, providing an overall picture of the risks associated with using these e-liquids.

E-liquid no. 6

Vaping e-liquid no. 6 results in exposure to polyols (propylene glycol, glycerol, diethylene glycol and triethylene glycol), nicotine and tobacco-specific nitrosamines (NNK, NAT). Aldehydes (formaldehyde and acrolein) were also detected in the vapour, but those compounds were produced during heating of the e-liquid. Hence, the aldehyde concentrations in the vapour were probably attributable to factors other than the nature of the e-liquid used and not therefore associable with the specific e-liquid. Exposure to the polyols in question brings a high risk of damage to the respiratory tract in heavy vapers; that risk cannot be excluded in light and average vapers. The possibility of heavy vapers experiencing systemic effects (reduced lymphocyte count) as a result of exposure to propylene glycol cannot be excluded. In heavy vapers, exposure to nicotine may induce effects on the respiratory tract and possibly systemic effects (effects on the cardiovascular system, fertility, a developing foetus). Such risks cannot be properly assessed due to a lack of appropriate data. Consequently, definitive conclusions cannot be drawn regarding light and average vapers. Vaping this liquid also involves exposure to the tobacco-specific nitrosamines NNK and NAT. In heavy vapers, that exposure will increase the risk of tumour development in the respiratory tract; in light and average vapers, the additional tumour risk may be considered negligible.

E-liquid no. 172

Vaping e-liquid no. 172 results in exposure to polyols (propylene glycol, glycerol) and nicotine. Formaldehyde was also detected in the vapour, but its presence is not specifically associated with this e-liquid. The concentration of propylene glycol in the vapour was roughly four to five times higher than with e-liquid no. 6, while the vapour concentration of glycerol was four to five times lower. In heavy vapers, daily use brings a high risk of damage to the respiratory epithelium; in light and average vapers, that risk cannot be excluded. The level of exposure to propylene glycol is sufficient to pose a risk of systemic effects (reduced lymphocyte

count) in heavy vapers. That risk cannot be excluded in average vapers. The nicotine concentration in the vapour from e-liquid no. 172 was two to three times lower than that in the vapour from e-liquid no. 6. It is concluded that, in heavy vapers, use of e-liquid no. 172 is liable to result in effects on the respiratory tract and systemic effects (effects on the cardiovascular system, fertility, a developing foetus). Such risks cannot be properly assessed due to a lack of appropriate data. As with the use of e-liquid no. 6, definitive conclusions cannot be drawn regarding the effects of e-liquid no. 172 in light and average vapers.

Overall analysis

The vapour from two e-liquids (no. 6 and no. 172) underwent all the analyses, providing an overall picture of the risks associated with using e-liquids in these two cases.

With both e-liquid no. 6 and e-liquid no. 172, exposure to the measured vapour concentrations of propylene glycol and glycerol involves a risk of effects on the respiratory tract. With the other analysed e-liquids, the propylene glycol and glycerol vapour concentrations were higher than with e-liquids no. 6 and no. 172; only with e-liquid no. 90 was the propylene glycol concentration in the vapour below the detection threshold. The propylene glycol concentrations were up to five times higher than with e-liquid no. 172 and the glycerol concentrations were up to five times higher than with e-liquid no. 6. The use of the other e-liquids may therefore pose a greater risk of damage to the respiratory epithelium than the use of e-liquid no. 6 or e-liquid no. 172. It is likely that the mechanism by which the various polyols damage the respiratory epithelium is the same in all cases. Account therefore needs to be taken of the cumulative effect of combined exposure to several polyols, as present in e-liquid vapour. Each substance will contribute to the risk of damage to the respiratory epithelium, so that the overall risk from exposure to an e-liquid vapour is significantly greater than consideration of the individual components would suggest. Propylene glycol can also induce a reduction in the lymphocyte count. Ethylene glycol was detected in one of the seventeen tested e-liquids (e-liquid no. 152). Use of that e-liquid may therefore pose a risk of systemic effects (renal damage) for heavy vapers. In average vapers using this e-liquid, that risk cannot be excluded, while in light vapers the risk is not present.

The nicotine concentration in the vapour from the other e-liquids was in most cases lower than or similar to that in the vapour from e-liquid no. 6. With two e-liquids, however, the nicotine concentrations were up to 1.4 times higher.

Diacetyl was detected in the vapour from two e-liquids. In the two cases concerned, the concentrations were sufficiently high to cause respiratory tract damage in heavy vapers. Such an effect cannot be excluded in average and light vapers. No risk of systemic effects appeared to be present. Vaping-related exposure to the aldehydes formaldehyde, acetaldehyde and acrolein cannot be associated with the use of specific e-liquids. Those aldehydes are not present in e-liquids, or are present only in very low concentrations; their presence in e-cigarette vapour is mainly a consequence of aldehyde formation during heating.

Formaldehyde was often detected in e-liquid vapour, while acrolein and acetaldehyde were occasionally detected. Formaldehyde and acrolein were present in concentrations sufficient for potential damage to the

respiratory tract. Formaldehyde-induced damage to the respiratory epithelium can be a precursor to tumour formation. In a few cases, the formaldehyde concentrations were sufficient to create a risk of tumour development in the respiratory tract. If the vapour contains not only formaldehyde but also acetaldehyde, acrolein and diacetyl in concentrations sufficient to damage the respiratory tract, the risk of formaldehyde-related tumours will be increased.
Where tobacco-specific nitrosamines are concerned, only the use of e-liquid no. 157 entails a considerably higher risk of respiratory tract tumour development than the use of e-liquid no. 6. Data on the vapour from eleven other analysed e-liquids indicate that use of the liquids in question is associated with a risk of tumour development that is lower than or similar to that associated with the use of e-liquid no. 6.
It should be noted that the risk assessment assumes daily exposure to vapour of a fixed composition. If the composition of the e-liquid used varies substantially, the composition of the vapour will of course vary too.

5.2.2 Comparison with the risk from conventional cigarette use

The potential health risks for e-cigarette users were compared with risks to which conventional cigarette users are exposed. Comparison was made in two ways.
First, the concentrations of substances detected in e-cigarette vapour were compared with the corresponding concentrations in tobacco smoke. Second, a comparison was made between the effects of conventional cigarette smoking described in the literature and the effects described for individual substances present in e-cigarette vapour.

5.2.2.1 Comparison on the basis of exposure scenarios

Concentration comparisons were made for those substances that are found both in e-cigarette vapour and in conventional cigarette smoke. The comparisons were based on the highest measured and/or reported concentrations. The substances in question were glycerol and propylene glycol, tobacco-specific nitrosamines, aldehydes, volatile organic compounds and the metals cadmium and lead. Where other substances were concerned, insufficient suitable data were available to support the comparison of e-cigarette vapour concentrations with conventional cigarette smoke concentrations. Information on the substance-specific concentrations in conventional cigarette smoke was derived from the scientific literature. In a few cases, the information is as previously used for the risk assessment of tobacco additives (Counts, Morton et al. 2005, Bos, Hernández et al. 2012).
The comparison showed that the concentration of glycerol in e-cigarette vapour is up to seventeen times higher than that in conventional cigarette smoke, while the concentration of propylene glycol is up to twenty-five times higher. Where tobacco-specific nitrosamines are concerned, the concentrations in e-cigarette vapour are up to four hundred times lower than those in conventional cigarette smoke. The e-cigarette vapour concentration of formaldehyde is up to three times higher than that in conventional cigarette smoke, while the acetaldehyde and acrolein concentrations in conventional cigarette smoke are, respectively, thirty-five times and four times higher than those in e-cigarette vapour. The concentrations of the volatile organic compounds benzene and toluene in e-cigarette vapour are, respectively, forty times

and 1,500 times lower than the concentrations in conventional cigarette smoke. Where the metals cadmium and lead are concerned, exposure associated with e-cigarette use is, respectively, 155 times and 3.5 times lower than the exposure associated with conventional cigarette use. Generally speaking, the comparison shows that the substance-specific vapour concentrations of the aldehydes acrolein and acetaldehyde, tobacco-specific nitrosamines, volatile organic compounds and certain metals in e-cigarette vapour are lower or much lower than the concentrations in conventional cigarette smoke. The situation is reversed, however, where propylene glycol, glycerol and formaldehyde are concerned.

5.2.2.2 Comparison on the basis of current, known health risks of conventional cigarette smoking

The health risks associated with conventional cigarettes were additionally specified in a manner that permitted comparison with the health risks associated with e-cigarettes. To that end, a literature research was undertaken to identify the current, known health risks of conventional cigarette smoking. The following information was used:

- Information on the occurrence of certain health effects (cancer, cardiovascular effects, effects on the respiratory epithelium) in relation to smoking habits
- Substance-specific information regarding individual components of tobacco smoke that can also be found in e-cigarette vapour, and the possible relationship with the occurrence of certain health effects

General effects

Scientific knowledge regarding the relationship between tobacco smoking and cancer in humans is based primarily on data from epidemiological studies. There is convincing evidence of a causal relationship between tobacco smoking and a wide variety of cancers, including cancer of the lungs, oral and nasal cavities, (naso-, oro-, hypo-) pharynx, larynx, oesophagus, stomach, pancreas, colorectal passage, liver, kidneys, ureter and bladder, cervix and ovaries, and myeloid leukaemia (IARC 2012). A positive association has also been demonstrated between tobacco smoking and breast cancer in women. There is evidence that (postmenopausal) endometrial cancer and thyroid cancer are not linked to smoking (IARC 2012). Smoking is also known to increase the risk of other conditions, such as cardiovascular disease (RIVM 2014). Furthermore, smoking affects fertility in men and women, and smoking during pregnancy is one of the biggest preventable causes of problems such as low birthweight, premature birth, neonatal morbidity and mortality (Kramer 1987, Cliver, Goldenberg et al. 1995, WHO 2013).

Substance-specific effects

No epidemiological research data are currently available regarding the effects of e-cigarette use. Therefore, with a view to obtaining insight into possible relationships between e-cigarette use and the occurrence of certain health effects (including effects not detectable in animal experiments), a search was made for epidemiological research data relating specifically to particular substances found both in e-cigarette vapour and in conventional cigarette smoke. As far as possible,

evaluation reports by (inter)nationally recognised authorities (e.g. US EPA, AEGL, ATSDR, WHO, Health Council) were utilised.
The search revealed that very little additional information is available regarding effects other than those previously described. One patient-control study pointed to an elevated risk of leukaemia in farmers who used nicotine-based pesticides. However, the Dutch Health Council considers that the number of subjects (patients and control subjects) involved in the study was too small to support reliable conclusions regarding a possible link between working with nicotine (as a pesticide/crop protector) and the occurrence of leukaemia (Gezondheidsraad 2005). A recent study found that exposure to acrolein (as determined by the measurement of urine concentrations of acrolein's principal metabolite) was associated with parameters for cardiovascular effects, such as the activation of blood platelets and suppression of circulating angiogenic cells (DeJarnett, Conklin et al. 2014).

5.3 Discussion and conclusions

The substance-specific comparison indicated that the concentrations of a number of substances detected in e-liquid vapour are lower or much lower than the concentrations in tobacco smoke. By contrast, the concentrations of glycerol and propylene glycol are higher in e-cigarette vapour than in tobacco smoke. The main effects of the latter two substances are damage to the respiratory epithelium and (where propylene glycol is concerned) a reduced lymphocyte count. In addition, the formaldehyde concentration in e-cigarette vapour can be up to three times higher than that in conventional cigarette smoke. On the other hand, the concentrations of the carcinogenic tobacco-specific nitrosamines in tobacco smoke are up to four hundred times higher than those in e-liquid vapour. Furthermore, tobacco smoke contains thousands of other substances, many of which are toxic (including PAHs, butadiene and benzene).
Various health effects associated with tobacco use have not been described in connection with individual compounds present in e-liquid vapour vapour (a comparison is described in detail in RIVM report 2014-0143, section 10.3.2.2 (Visser W 2015)). It should be noted, however, that extensive epidemiological data on the health effects of tobacco use have been published, whereas little or no comparable information is available regarding e-cigarette use or the individual compounds associated with e-cigarette use.
It may be concluded that the health risks associated with smoking conventional cigarettes are considerably higher than those associated with using e-cigarettes, based on the findings described in RIVM report 2014-143 (Visser W 2015) (subsections 10.3.2.1 and 10.3.2.2). That conclusion assumes comparable usage patterns (a similar number of inhalations over a comparable period). The health risks are strongly dependent on individual vaping and smoking habits.
Nevertheless, daily use of e-cigarettes is not without health risks. Exposure to the polyols can damage the respiratory epithelium and reduce the lymphocyte count. Nicotine-containing e-liquids can also affect health in various ways. Furthermore, while the vapour concentrations of tobacco-specific nitrosamines are many times lower than in tobacco smoke, they are sufficiently high in some cases to give an elevated risk of tumour development. The vapour concentrations of

aldehydes can also be sufficient to induce effects on the respiratory tract, although the concentrations in question are probably attributable to the heating process rather than the properties of particular e-liquids. The level of risk and the seriousness of the potential effects depend considerably on the e-cigarette usage pattern. Health risks are liable to increase sharply at higher intensities of use. However, insufficient dose-response relationship data are available to support evaluation of the complex and fluctuating exposure patterns associated with e-cigarette use.

6 Conclusions

The market research reported here was intended to yield data on a number of usage parameters to serve as input for an analysis of the risks of e-cigarette use. For pragmatic reasons, the research involved a questionnaire-based survey of a representative group of experienced e-cigarette users in the Netherlands. Such research relies on self-reporting and that is one of its main limitations. A further complicating factor is that the range of e-cigarettes and e-liquids available is very wide and subject to rapid change. As a result, the data obtained may become outdated within a relatively short space of time. Despite those limitations, the market research proved to be an expeditious means of obtaining an overview of e-cigarette usage patterns in early 2014.

The main conclusions of the market research are:

- Almost all e-cigarette users also smoke tobacco or used to do so; nearly half are seriously nicotine dependent and need to light a cigarette within half an hour of getting up in the morning.
- The three main reasons for using e-cigarettes are that they are believed to be less harmful, that they are cheaper than tobacco and that they are expected to help the user stop smoking.
- E-cigarettes are used almost everywhere: at home, in the car, when visiting, in the street or the countryside, at work, when out for the evening, etc.
- Many different brands and models are used, but second-generation products are the most popular. Daily and weekly smokers differ in their model preferences: daily smokers have more specific requirements and wishes.
- Three quarters of experienced users refill their e-cigarettes themselves, using proprietary liquids; 10 per cent prepare their own liquids.
- Only 8 per cent use nicotine-free liquid; 42 per cent use low-strength liquids (up to 10 mg) and 45 per cent use high-strength liquids (11-23 mg). Extra-high-strength liquids (> 24 mg) are rarely used.
- Tobacco flavour is by far the most popular flavour, followed by menthol. A fifth of vapers prefer fruit or (other) sweet flavours.
- Most users do not use models with variable-voltage batteries; the 10 per cent who do use such models mainly select a medium voltage.
- There are clear differences between daily and weekly vapers in terms of the degree of exposure to the vapour. Those differences need to be taken into account in the risk analysis. Daily users vape more frequently per usage day, have longer vaping sessions (but during which they inhale *less* often) and inhale slightly deeper than weekly vapers. Most scientific literature on this subject makes no distinction between different vaping profiles, although there may be implications for the level of exposure.
- The great majority of current vapers intend to go on using e-cigarettes for some time.

Compositional analysis revealed that the nicotine concentrations of 36 per cent of e-liquids deviated by more than 10 per cent from the concentrations stated on the packaging. In most cases, the actual nicotine concentration was lower than the stated concentration, but in a few cases considerable amounts of nicotine were present in e-liquids labelled as nicotine free.
In addition to the ingredients used by the manufacturer, e-liquids contain substances that were present as contaminants in the raw materials. In the context of this study, any substance other than nicotine, propylene glycol, glycerol and aromatic substances and flavourants was regarded as a contaminant. E-liquids differ considerably in terms of measured contaminant concentrations. A small proportion of liquids contain diethylene glycol, benzene, toluene or TSNAs, but those substances were not demonstrably present in the great majority of liquids. Of the tested liquids, 45 per cent were found to contain measurable concentrations of short-chain aldehydes or ketones, and all the liquids had measurable quantities of one or more metals.
In order to determine what other substances are present in the e-liquids, some e-liquids were analysed by means of headspace GC-MS. Most of the other substances thus detected were very probably known flavourants and aromatics. Further research is required, however, to determine the identities and concentrations of the components in question with certainty.

The design of the experimental study supported effective measurement of e-cigarette vapour composition. The concentrations of the various substances in the vapour appear to be determined principally by the e-liquid used, the model of e-cigarette used and of the behaviour of the user.
Carrier liquids and nicotine were almost completely vaporised, and their concentrations in the vapour were consequently determined nearly entirely by the capacity of the vaporiser and the behaviour of the user. Similarly, the harmful substances diacetyl, benzene and toluene were present in the vapour only if they were also present in the e-liquid. However, our results show that short-chain aldehydes and ketones present in the vapour do not originate directly from the e-liquid, but are formed during vaporisation. Propylene glycol and glycerol may partially decompose under the influence of heat, but the precise mechanism is not known. The concentrations of those substances in the vapour varied greatly and were strongly dependent on the particular e-cigarette used. Two apparently identical vaporisers made by the same manufacturer and filled with the same e-liquid yielded vapour formaldehyde concentrations that differed by a factor of more than twenty-five.

The substance-specific comparison indicated that the concentrations of a number of substances detected in e-liquid vapour are lower or much lower than the concentrations in tobacco smoke. By contrast, the concentrations of the carrier substances glycerol and propylene glycol are higher in e-cigarette vapour than in tobacco smoke. The main effects of the latter two substances are damage to the respiratory epithelium and (where propylene glycol is concerned) a reduced lymphocyte count. In addition, the formaldehyde concentration in e-cigarette vapour can be up to three times higher than that in conventional cigarette smoke. On the other hand, the concentrations of

the carcinogenic tobacco-specific nitrosamines in tobacco smoke are up to four hundred times higher than those in e-liquid vapour. Furthermore, tobacco smoke contains thousands of other substances, many of which are toxic (including PAHs, butadiene and benzene).
Various health effects associated with tobacco use have not been described in connection with individual compounds present in e-liquid vapour. It should be noted, however, that extensive epidemiological data on the health effects of tobacco use have been published, whereas little or no comparable information is available regarding e-cigarette use or the individual compounds associated with e-cigarette use. It may be concluded that the health risks associated with smoking conventional cigarettes are considerably higher than those associated with using e-cigarettes. That conclusion assumes comparable usage patterns (a similar number of inhalations over a comparable period). The health risks are strongly dependent on individual vaping and smoking habits. Nevertheless, daily use of e-cigarettes is not without health risks. Exposure to the polyols can damage the respiratory epithelium and reduce the lymphocyte count. Nicotine-containing e-liquids can also affect health in various ways. Furthermore, while the vapour concentrations of tobacco-specific nitrosamines are many times lower than in tobacco smoke, they are sufficiently high in some cases to give an elevated risk of tumour development. The vapour concentrations of aldehydes can also be sufficient to induce effects on the respiratory tract, although the concentrations in question are probably attributable to the heating process rather than the properties of particular e-liquids. The level of risk and the seriousness of the potential effects depend considerably on the e-cigarette usage pattern. Health risks are liable to increase sharply at higher intensities of use. However, insufficient dose-response relationship data are available to support evaluation of the complex and fluctuating exposure patterns associated with e-cigarette use.
Thirteen different metals were detected in e-cigarette vapour. Like the aldehydes, the metals are not present in the e-liquid, but originate from the heating element. In the available time, detailed assessment of the risks associated with the detected metals was not possible. Nevertheless, where five metals were concerned, adequate information was available to support comparison of the initial alveolar concentration associated with vaping with that associated with smoking. For the purpose of the comparison, it was assumed that vaping behaviour and smoking behaviour were comparable (e.g. in terms of the number of inhalations per day and the duration of exposure). The conclusion was that smoking was associated with higher or much higher concentrations of cadmium, lead, nickel and arsenic than vaping; the chromium concentrations were comparable. While the possibility of health effects at the measured metal concentrations cannot be excluded, it is safe to say that the risks are lower with vaping than with smoking. A more precise assessment of the possible risks posed by metals in e-cigarette vapour will require further research.
In a previous risk assessment of the shisha pen, a limited analysis involving the measurement of glycerol and propylene glycol concentrations in shisha pen vapour led to the conclusion that a single inhalation of shisha pen vapour was potentially sufficient to cause irritation of the respiratory tract (RIVM 2013). The concentrations of glycerol and propylene glycol in shisha pen vapour determined in the

context of that analysis were similar to those found in e-cigarette vapour. On the basis of the more detailed risk assessment of e-cigarette use presented in this report, it may be assumed that exposure to glycerol and propylene glycol in the context of shisha pen use has the potential to pose risks similar to those associated with e-cigarette use. However, the actual risk depends greatly on the usage pattern (user behaviour) and the degree of heating.

7 References

ASH. "Use of electronic cigarettes in Great Britain. Factsheet. ." http://www.ash.org.uk/files/documents/ASH_891.pdf.

Bos, P. M. J., et al. (2012). "Risk Assessment of Tobacco Additives and Smoke Components. RIVM letter rapport 340031001/2012. Beschikbaar via: http://www.rivm.nl/en/Library/Scientific/Reports/2012/oktober/Risk_assessment_of_tobacco_additives_and_smoke_components_a_method_proposal."

Brown, C. J. and J. M. Cheng (2014). "Electronic cigarettes: product characterisation and design considerations." Tob Control **23 Suppl 2**: ii4-10.

Callahan-Lyon, P. (2014). "Electronic cigarettes: human health effects." Tob Control **23 Suppl 2**: ii36-40.

Cheng, T. (2014). "Chemical evaluation of electronic cigarettes." Tob Control **23 Suppl 2**: ii11-17.

Cliver, S. P., et al. (1995). "The effect of cigarette smoking on neonatal anthropometric measurements." Obstet Gynecol **85**(4): 625-630.

Counts, M. E., et al. (2005). "Smoke composition and predicting relationships for international commercial cigarettes smoked with three machine-smoking conditions." Regul Toxicol Pharmacol **41**(3): 185-227.

DeJarnett, N., et al. (2014). "Acrolein exposure is associated with increased cardiovascular disease risk." J Am Heart Assoc **3**(4).

DKFZ (2013). "Electronic Cigarettes – An Overview. ." Heidelberg: DKFZ (German Cancer Research Center).

Farsalinos, K. E. and R. Polosa (2014). "Safety evaluation and risk assessment of electronic cigarettes as tobacco cigarette substitutes: a systematic review." Ther Adv Drug Saf **5**(2): 67-86.

Farsalinos, K. E., et al. (2013). "Impact of flavour variability on electronic cigarette use experience: an internet survey." Int J Environ Res Public Health **10**(12): 7272-7282.

Farsalinos, K. E., et al. (2013). "Evaluation of electronic cigarette use (vaping) topography and estimation of liquid consumption: implications for research protocol standards definition and for public health authorities' regulation." Int J Environ Res Public Health **10**(6): 2500-2514.

Farsalinos, K. E., et al. (2014). "Nicotine absorption from electronic cigarette use: comparison between first and new-generation devices." Sci Rep **4**: 4133.

Gezondheidsraad (2005). "Nicotine (CAS no: 54-11-5). Health-based Reassessment of Administrative Occupational Exposure Limits (Revised version). Committee on Updating of Occupational Exposure Limits, a committee of the Health Council of the Netherlands. No. 2000/15OSH/105(R). http://www.gezondheidsraad.nl/sites/default/files/0015105OSHR.PDF ".

Goniewicz, M. L., et al. (2013). "Nicotine levels in electronic cigarettes." Nicotine Tob Res **15**(1): 158-166.

Goniewicz, M. L., et al. (2013). "Patterns of electronic cigarette use and user beliefs about their safety and benefits: an internet survey." Drug Alcohol Rev **32**(2): 133-140.

Grana, R., et al. (2014). "E-cigarettes: a scientific review." Circulation **129**(19): 1972-1986.

Hajek, P., et al. (2014). "Electronic cigarettes: review of use, content, safety, effects on smokers and potential for harm and benefit." Addiction **109**(11): 1801-1810.

Hua, M., et al. (2013). "Mining data on usage of electronic nicotine delivery systems (ENDS) from YouTube videos." Tob Control **22**(2): 103-106.

IARC (2012). "Personal habits and indoor combustions. Volume 100 E. A review of human carcinogens. International Agency for Research on Cancer, Lyon, France. http://monographs.iarc.fr/ENG/Monographs/vol100E/mono100E.pdf."

Kramer, M. S. (1987). "Intrauterine growth and gestational duration determinants." Pediatrics **80**(4): 502-511.

McNeill, A., et al. (2014). "A critique of a World Health Organization-commissioned report and associated paper on electronic cigarettes." Addiction.

NVWA (2014). "Marktverkenning navulverpakkingen voor e-sigaretten en
shishapennen."
http://www.vwa.nl/onderwerpen/consumentenartikelen/dossier/e-sigaret/nieuwsoverzicht/nieuwsbericht/2048401/gevaarsymbool-giftig-ontbreekt-op-veel-navulverpakkingen-e-sigaret-met-nicotin.

Orr, M. S. (2014). "Electronic cigarettes in the USA: a summary of available toxicology data and suggestions for the future." Tob Control **23 Suppl 2**: ii18-22.

RIVM (2013). "Risicoschatting van de shisha-pen. Factsheet. Rijksinstituut voor Volksgezondheid en Milieu, Bilthoven, Nederland.".

RIVM (2014). "E-sigaretten Factsheet."
http://www.rivm.nl/dsresource?type=pdf&disposition=inline&objectid=rivmp:242776&versionid=&subobjectname=.

RIVM (2014). "Volksgezondheid Toekomst Verkenning, Nationaal Kompas Volksgezondheid. Bilthoven: RIVM, http://www.nationaalkompas.nl, Nationaal Kompas Volksgezondheid, versie 4.17, 23 juni 2014."

Schripp, T., et al. (2013). "Does e-cigarette consumption cause passive vaping?" Indoor Air **23**(1): 25-31.

Schroeder, M. J. and A. C. Hoffman (2014). "Electronic cigarettes and nicotine clinical pharmacology." Tob Control **23 Suppl 2**: ii30-35.

Trehya ML, Y. W., . Hadwigera ME, Moorea TW, Allgirea JF, Woodruffa JT, Ahadia SS, Blacka JC, Westenbergera B (2011). "Analysis of electronic cigarette cartridges, refill solutions, and smoke for nicotine and nicotine related impurities." Journal of Liquid Chromatography & Related Technologies **34**: 16.

Visser W, G. L., Klerx W, Hernandez L, Stephens E, Croes E, Schwillens P, Cremers H, Bos P, Talhout R (2015). RIVM rapport 2014-0143: De gezondheidsrisico's van het gebruik van e-sigaretten, http://www.rivm.nl/bibliotheek/rapporten/2014-0143.pdf

WHO (2013). "WHO recommendations for the prevention and management of tobacco use and second-hand smoke exposure in pregnancy. World Health Organization, Geneva, Switzerland http://www.who.int/tobacco/publications/pregnancy/guidelinestobaccosmokeexposure/en/."

Zhu, S. H., et al. (2014). "Four hundred and sixty brands of e-cigarettes and counting: implications for product regulation." Tob Control **23 Suppl 3**: iii3-9.

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