E-VAPOR SAFETY EVALUATION AND RISK ASSESSMENT: A LEGA ITALIANA ANTI FUMO (LIAF) REPORT

Electronic cigarettes (ECs) are the newest and most promising products for tobacco harm reduction (THR) [Polosa et al. 2013a]. They are electrically-driven devices consisting of the battery part (usually lithium battery), and an atomizer where liquid is stored and is aerosolized by applying energy and generating heat to a resistance encircling a wick. The liquid used mainly consists of propylene glycol, glycerol, distilled water, flavorings (that may or may not be approved for food use) and nicotine. Consumers (commonly called "vapers") may choose from several nicotine strengths, including non-nicotine liquids, and a countless list of flavors; this assortment is a characteristic feature that distinguishes ECs from any other THR products. Since their invention in 2003, there has been constant innovation and development of more efficient and appealing products. Currently, there are mainly 3 types of devices available [Dawkins, 2013a]: (1) First-generation devices, generally mimicking size and look of regular cigarettes and consisting of small lithium batteries and cartomizers (i.e. cartridges, which are usually prefilled with a liquid that bathes the atomizer). Batteries may be disposable (to be used once only) or rechargeable. (2) Second-generation devices, consisting mainly of higher-capacity lithium batteries and atomizers with the ability to refill them with liquid (sold in separate bottles). In the most recent atomizers you can simply change the atomizer head (resistance and wick) while keeping the body of the atomizer, thus reducing the operating costs. (3) Third-generation devices (also called "Mods" – from modifications), consisting of very large-capacity lithium batteries with integrated circuits that allow vapers to change the voltage or power (wattage) delivered to the atomizer.

Awareness and use (vaping) of ECs has increased exponentially in recent years. Data obtained from the HealthStyles survey showed that in the US awareness of ECs rose from 40.9% to 57.9% from 2010 to 2011, with ECs use rising from 3.3% to 6.2% over the same time period [King et al., 2013]. In the United Kingdom, ECs use in regular smokers increased from 2.7% in 2010 to 6.7% in 2012 [Dockrell et al., 2013]. Similar findings were obtained from the International Tobacco Control Four-Country Survey [Adkison et al., 2013]. A recent prospective study in Swiss army recruits showed that 12% of smokers who tried EC progressed to daily use [Douptcheva et al. 2013]. It must be noted that this increase in ECs use, has occurred despite the concerns raised by public health authorities about the safety and appropriateness of using these products as alternatives to smoking [National Association of Attorneys General, 2013; Food and Drug Administration, 2009; Mayers, 2009].

ECs popularity may be due to their ability to deal both with the physical (i.e. nicotine) and the behavioral component of smoking addiction. In particular, sensory stimulation [Rose and Levin, 1991] and simulation of smoking behavior and cigarette manipulation [Hajek et al., 1989] are important determinants of a product's effectiveness in reducing or completely substituting smoking. Whereas these features are generally absent in nicotine replacement therapies (NRTs) and oral medications for nicotine dependence, ECs are unique in that they provide rituals associated with smoking behavior (e.g. hand-to-mouth movement, visible "smoke" exhaled) and sensory stimulation associated with it [Farsalinos et al., 2013a]. This explains why these products can be effective in reducing consumption of tobacco smoking [Bullen et al., 2013; Caponnetto et al., 2013a; Polosa et al., 2011] and efficient as long-term substitutes of conventional cigarettes [Farsalinos et al., 2013a].

Currently available evidence indicates that electronic cigarettes are by far a less harmful alternative to smoking and significant health benefits are expected in smokers who switch from tobacco to electronic cigarettes. Here we present a summary of a total of 107 studies investigating existing laboratory and clinical research on the potential risks from electronic cigarette use, compared to the well-established devastating effects of smoking tobacco cigarettes.

Conventional cigarettes are the most common form of nicotine intake. Although the addictive potential of nicotine and related compounds is largely documented [Guillem et al. 2005], much less dissemination has been given to the notion that nicotine does not contribute to smoking-related disease. It is not classified as a carcinogen by the International Agency for Research on Cancer [WHO-IARC, 2004] and does not promote obstructive lung disease. It has been established that nicotine itself has minimal effects in initiating and promoting atherosclerotic heart disease [Ambrose and Barua, 2004]. It does not promote platelet aggregation [Zevin et al. 1998], does not affect coronary circulation [Nitenberg and Antony, 1999] and does not adversely alter the lipid profile [Ludviksdottir et al. 1999]. An observational study of more than 33,000 smokers found no evidence increased risk for myocardial infarction or acute stroke after NRT subscription, although follow-up was only 56 days [Hubbard et al. 2005]. Up to five years of nicotine gum use in the Lung Health Study was unrelated to cardiovascular diseases or other serious side effects [Murray et al. 1996]. A meta-analysis of 35 clinical trials found no evidence of cardiovascular or other life threatening adverse effects caused by nicotine intake

[Greenland et al. 1998]. Even in patients with established cardiovascular disease, nicotine use in the form of NRTs does not increase cardiovascular risk [Woolf et al. 2012; Benowitz and Gourlay, 1997]. It is anticipated that any product delivering nicotine without involving combustion, such as the EC, would confer significantly lower risk compared to conventional cigarettes and to other nicotine containing combustible products.

The importance of using nicotine in the long-term has been recognized several years ago by Russell [1991], indicating that the potential of nicotine delivery systems as long-term alternatives to tobacco should be explored in order to make the elimination of tobacco a realistic future target. However, current regulations restrict the long-term use of pharmaceutical or recreational nicotine products (such as snus) [Le Houezec et al. 2011]. Obviously, the addictive potential is an important factor in any decision to endorse nicotine administration; however, it should be considered a slight "collateral damage" with minimal impact in vapers' health compared to the tremendous benefit of eliminating all disease-related substances coming from tobacco smoking. In fact, smokers are already addicted to nicotine; therefore the use of a "cleaner" form of nicotine delivery would not represent any additional risk of addiction. Surveys have shown that ECs are used as long term substitutes to smoking [Dawkins et al. 2013b; Etter and Bullen, 2012]. Although consumers try to reduce nicotine use with ECs, many are unable to completely stop its intake, indicating an important role for nicotine in the ECs' effectiveness as a smoking substitute [Farsalinos et al. 2013a].

Nicotine overdose or intoxication is unlikely to occur with vaping, since the amount consumed [Farsalinos et al. 2013b] and absorbed [Nides et al. 2013; Dawkins and Corcoran, 2013c] is quite low. Moreover, although not yet proven, it is expected that vapers will self-titrate their nicotine intake in a similar way to tobacco cigarettes [Benowitz et al. 1998]. Last but not least, there is evidence suggesting that nicotine cannot be delivered as fast and effectively from ECs compared to tobacco cigarettes [Farsalinos et al. 2013c]. Therefore, it seems that ECs have a huge theoretical advantage in terms of health risks compared to conventional cigarette due to the absence of toxic chemicals that are generated in vast quantities by combustion. Furthermore, nicotine delivery by ECs is unlikely to represent a significant safety issue, particularly when considering that they are intended to replace tobacco cigarettes, the most efficient nicotine delivery product.

Findings on the safety/risk profile of ECs have just started to accumulate. Existing studies about the safety/risk profile of ECs can be divided into chemical, toxicological and clinical studies (Table 1).

Table 1. Types of studies performed to determine safety and to estimate risk from ECs use

Type of studies	Research subject	Advantages	Disadvantages
Chemical studies	Evaluate the chemical composition of liquids and/or aerosol. Examine environmental exposure (passive "vaping").	Easier and faster to perform. Less expensive. Could realistically be implemented for regulatory purposes.	Usually targeted on specific chemicals. Unknown effects of flavorings when inhaled. No validated protocols for vapor production. Provide no evidence about the end-results (effects) of use (besides by applying theoretical models).
Toxicological studies	Evaluate the effects on cell cultures or experimental animals.	Provide some information about the effects from use.	Difficult to interpret the results in terms of human in *vivo* effects. More expensive than chemical studies. Need to test aerosol and not liquid. Standards for exposure protocols have not been clearly defined.
Clinical studies	Studies on human *in vivo* effects.	Provide definite and objective evidence about the effects of use.	Difficult and expensive to perform. Long-term follow-up is needed due to the expected lag from initiation of use to possible development of any clinically evident disease. For now, limited to acute effects from use.

Chemical studies are relatively simple and can provide quick results but there are disadvantages. Research is usually focused on the known specific chemicals (generally those known to be toxic from studies of cigarette smoke) and fails to address unknown, potentially toxic contaminants that could be detect in the liquid or the emitted aerosol. Problems may also arise from the detection of the chemicals of flavors. Such substances, although approved for use in the food industry, have largely unknown effects when heated and inhaled; thus, information on the presence of such substances is difficult to be

interpreted in terms of in vivo effects. In fact, chemical studies do not provide any objective information about the effects of use; they can only be used to calculate the risk based on theoretical models and on already established safety levels determined by health authorities.

Laugesen [2009] performed the first studies evaluating the chemical composition of EC aerosol. The temperature of the resistance of the tested EC was 54^{o}C during activation, which is approximately 5-10% of the temperature of a burning tobacco cigarette. Toxic chemicals such as aldehydes, heavy metals, polycyclic aromatic hydrocarbons and phenols were not detected, with the exception of trivial amounts of mercury (0.17ng per EC). Laugesen evaluated emissions based on a toxicant emissions score and reported a score of zero in EC compared to a score of 100-134 for tobacco cigarettes (Figure 3). The US Food and Drug Administration also performed chemical analyses on 18 commercially available products in 2009 [Westenberg, 2009]. They detected the presence of tobacco-specific nitrosamines (TSNAs) but did not declare the levels found. Small amounts of diethylene glycol were also found in one sample, which was unlikely to cause any harm from normal use. Another study identified small amounts of amino-tandalafil and rimonambant in EC liquids [Hadwiger et al. 2010]. Subsequently, several laboratories performed similar tests, mostly on liquids, with Cahn and Siegel [2011] publishing a review on the chemical analyses of ECs and comparing the findings with tobacco cigarettes and other tobacco products. They reported that TSNAs, levels were similar to those measured in pharmaceutical NRTs. The authors concluded that, based on chemical analysis, ECs are by far less harmful compared to tobacco cigarettes. The most comprehensive study on TSNAs has been recently performed by a South Korean group, evaluating 105 liquids obtained from local retailers [Kim and Shin, 2013]. They found on average 12.99ng TSNAs per ml of liquid, with the amount of daily exposure to the users estimated to be similar to users of NRTs [Farsalinos et al. 2013d]. The estimated daily exposure to nitrosamines from tobacco cigarettes (average consumption of 15 cigarettes per day) is estimated to be up to 1800 times higher compared to EC use. Etter et al. [2013] evaluated the accuracy of nicotine labeling and the presence of nicotine impurities and degradation products in 20 EC liquid samples. They found that nicotine levels were 85-121% of what was labeled, while nicotine degradation products were present at levels of 0-4.4%. Although in some samples the levels were higher than those specified in European Pharmacopoeia, they are not expected to cause any measurable harm to users.

Besides the evaluation for the presence of TSNAs, analyses have been performed for the detection of carbonyl compounds. It is known that the thermal degradation of propylene glycol and glycerol can lead to the emission of toxic compounds such us aldehydes [Antal et al. 1985; Stein et al. 1983]. Goniewicz et al. [2013b] evaluated the emission of 15 carbonyls from 12 brands of ECs (mostly first-generation). In order to produce vapor, researchers used a smoking machine and followed a regime of 1.8-second puffs with a very short 10 seconds interpuff interval, which does not represent realistic use [Farsalinos et al. 2013b]; although the puff duration was low, interpuff interval was remarkably short, which could lead to overheating. Additionally, the same puff number was used in all devices tested, although there was a significant difference in the design and liquid content between devices. Despite these, out of 15 carbonyls, only 3 were detected (formaldehyde, acetaldehyde and acrolein); levels were 9-450 times lower compared to emissions from tobacco cigarette (derived from existing literature but not tested in the same experiment). Formaldehyde and acetaldehyde were also emitted from the nicotine inhalator, although at lower levels. Additionally, they examined for the presence of 11 volatile organic carbons and found only toluene (at levels from 0.2 μg to 6.3 μg per 150 puffs) and traces of xylene (from 0.1 μg to 0.2 μg per 150 puffs) in 10 of the samples; toluene levels were 120 times lower compared to tobacco cigarette (again, derived from existing literature but not tested in the same experiment).

Given that ECs have several metal parts in direct contact with the e-liquid, it is quite obvious to expect some contamination with metals in the vapor. Goniewicz et al. [2013b] examined samples for the presence of 12 metals and found nickel, cadmium and lead emitted; the levels of nickel were almost similar to those present in a pharmaceutical nicotine inhalator, while lead and cadmium were present at 2-3 times higher levels compared to the inhalator. Still, the absolute levels were very low (few nanograms per 150 puffs). Williams et al. [2013] focused their research on the presence of heavy metals and silicate particles emitted from ECs. They tested poor quality first-generation cartomisers and found several metals emitted in the aerosol of the EC, specifying that in some cases the levels were higher compared to conventional cigarettes. As mentioned earlier, it is not unusual find trace levels of metals in the vapor generated by these products under experimental conditions that bear little relevance with their normal use; however, it is unlikely that such small amounts pose a serious threat to users' health. Even if all aerosol was absorbed from the consumer (which is not the case since most aerosol is visibly exhaled), an average user would be exposed to 4-40 times lower amounts for most metals than

the maximum daily dose allowance from impurities in medicinal products [US Pharmacopeia, 2013]. Silicate particles were also found in the EC aerosol. Such particles are coming from the wick material, however the authors did not clarify whether crystalline silica oxide particles were found, which are responsible for respiratory disease. In total, the number of microparticles (< 1000nm) estimated to be inhaled by EC users from 10 puffs were 880 times lower compared to one tobacco cigarette. Similar finding concerning microparticles were reported by Pellegrino et al. [2012] who found that, for each particulate matter fraction, conventional cigarettes produced hundreds of times higher amount of particles compared to the EC tested.

Burstyn [2013] has recently reviewed current data on chemistry of aerosols and liquids of ECs (including reports which were not peer-reviewed) and estimated the risk to consumers based on workplace exposure standards (i.e. Threshold Limit Values-TLVs). After reviewing all available evidence, the author concluded that there is no evidence that vaping produces inhalable exposures to contaminants of aerosol that would warrant health concerns. He added that surveillance of use is recommended due to the high levels of propylene glycol and glycerol inhaled (which are not considered contaminants but ingredients of the EC liquid). There are limited data on the chronic inhalation of these chemicals by humans, although there is some evidence from toxicological studies (which are discussed later in the manuscript).

In conclusion, chemical studies have found that exposure to toxic chemicals from ECs is by far lower compared to tobacco cigarettes. Obviously, surveillance of use is warranted in order to objectively evaluate the *in vivo* effects and because the effects of inhaling flavoring substances approved for food use are largely unknown.

The cytotoxicity approach has also its flaws. Findings cannot be directly applied to the *in vivo* situation and there is always the risk to over- (as well as under-) estimate interpretation of the toxic effects in these investigational models. An ample degree of results variability is to be expected from different cell lines and, sometimes, also within the same cell line. Comparing the potential cytotoxicity effects of ECs vapor with those resulting from the exposure of cigarette smoke should be mandatory, but standards for vapor production and exposure protocols have not been clearly defined.

Bahl et al. [2012] performed cytotoxicity tests on 36 EC liquids, in human embryonic stem cells, mouse neural stem cells and human pulmonary fibroblasts and found that stem cells were more sensitive to the effects of the liquids, with 15 samples being moderately cytotoxic and 12 samples being highly cytotoxic. Propylene glycol and glycerol were not cytotoxic, but a correlation between cytotoxicity and number and height of the flavoring peaks in high-performance liquid chromatography was noted. Investigations were just restricted to the effect of EC liquids and not to their vapors, thus limiting the importance of the study findings; this is not a trivial issue considering that the intended use of these products is by inhalation only and that it is unlikely that flavoring substances in the EC liquids will be still present in the aerosol in the same amount, due to differences in evaporation temperature [Romagna et al., 2013]. Regrettably, a set of experiments with cigarette smoke extracts as comparator was not included. Of note, the authors emphasized that the study could have underestimated the cytotoxicity by 100 times because when they added the EC liquids in the cell medium final concentration was 1%. However, cells were cultured for 48h with continuous exposure to the liquid, while in real use the lungs come in contact with aerosol instead of liquid, the contact lasts for 1-2 seconds per puff and most of the aerosol is visibly exhaled. Finally, Cinnamon Ceylon, the liquid found to be mostly cytotoxic in this study, was not a refill liquid but concentrated flavor which is not used for EC use unless it is diluted to 3-5%.

Romagna et al. [2013] performed the first cytotoxicity study of EC vapor on fibroblast cells. They used a standardized ISO 10993-5 protocol, which is used for regulatory purposes of medical devices and products. They tested the vapor of 21 liquid samples containing the same amount of nicotine (9 mg/ml), generated by a commercially-available EC device. Cells were incubated for 24 hours with each of these vapors and with smoke from a conventional cigarette. Only 1 sample was found to be marginally cytotoxic, whereas cigarette smoke was highly cytotoxic (approximately 795% more cytotoxic), even when the extract was diluted up to 25% of the original concentration.

The same group also investigated the cytotoxic potential of 20 EC liquid samples in cardiomyoblasts [Farsalinos et al. 2013e]. Vapor was produced by using a commercially-available EC device. Samples contained a wide range of nicotine concentrations. A base liquid mixture of propylene glycol and glycerol (no nicotine and no flavorings) was also included as additional experimental control. Four of the samples examined were made by using cured tobacco leaves in a steeping process, allowing them to impregnate a mixture of propylene glycol and glycerol for several days before being filtered and

bottled for use. Of note, this was the first study which evaluated a limited number of samples with an EC device delivering higher voltage and energy to the atomizer (third-generation device). In total, 4 samples were found cytotoxic; 3 of them were liquids made by using cured tobacco leaves, with cytotoxicity observed at both 100% and 50% extract concentration, while 1 sample (cinnamon flavor) was marginally cytotoxic at 100% extract concentration only. In comparison, smoke from 3 tobacco cigarettes was highly cytotoxic, with toxicity observed even when the extract was diluted to 12.5%. The samples made with tobacco leaves were 3 times less cytotoxic compared to cigarette smoke; this was probably due to the absence of combustion and the significantly lower temperature of evaporation in EC use. Concerning high-voltage EC use, the authors found slightly reduced cell viability without any of the samples being cytotoxic according to the ISO 10993-5 definition. Finally, no association between cell survival and the amount of nicotine present in the liquids was noted.

A recent study evaluated in more details the cytotoxic potential of 8 cinnamon-flavored EC liquids in human embryonic stem cells and human pulmonary fibroblasts [Behar et al. 2013]., The authors found that the flavoring substance predominantly present was cinnamaldehyde, which is approved for food use. They observed significant cytotoxic effects, mostly on stem cells but also on fibroblasts, with cytotoxicity associated with the amount of cinnamaldehyde present in the liquid. However, major methodological issues arose from this study. Once again, cytotoxicity was just restricted to EC liquids and not to their vapors. Moreover, the authors mentioned that the amount of cinnamaldehyde differed between liquids by up to 100 times, and this raises the suspicion of testing concentrated flavour rather than refills. By searching through the internet and contacting manufacturers, based on the names of samples and suppliers mentioned in the manuscript, it was found that at least 4 of their samples were not refills but concentrated flavors. Surprisingly, the levels of cinnamaldehyde found cytotoxic were about 400 times lower than those currently approved for use [Environmental Protection Agency, 2000].

Few animal studies have been performed to evaluate the potential harm of humectants in EC liquids (i.e. propylene glycol and glycerol) when given by inhalation. Robertson et al. [1947] tested the effects inhaling propylene glycol vapor for several months in primates and found no evidence of toxicity on any organ (including the lungs) after post-mortem examination of the animals. Similar observations were made in a recent study in rats and dogs [Werley et al. 2011]. Concerns have been raised in human use, based on studies of people exposed to theatrical fog [Varughese et al. 2005; American Chemistry

Council, 2003] or propylene glycol used in the aviation industry [Wieslander et al. 2001]. Irritation of the respiratory tract was found, but no permanent lung injury or other long-term health implications. It should be reminded that in these circumstances non-pharmaceutical purity propylene glycol is used and in some cases oils are added, making it difficult to interpret the results in the context of EC use. Evidence for the potential harm of inhaled glycerol is sparse. A study in Sprague-Dawley rats found minimal to mild squamous metaplasia of the epiglottis epithelium in the high-dose group only, without any changes observed in lungs or other organs [Renne et al. 1992]. No comparative set of experiments with cigarette smoke were included, but it is well known that exposure to tobacco smoke in similar animal models leads to dramatic changes in the lungs, liver and kidneys [Czekai et al. 2002].

In summary, toxicological studies have shown significantly lower adverse effects of EC vapor compared to cigarette smoke. Characteristically, the studies performed by using the liquids in their original liquid form have found less favorable results; however, no comparison with tobacco smoke was performed in any of these studies, and they cannot be considered relevant to EC use since the samples were not tested in the form consumed by vapers.

Clinical trials can be very informative, but they require monitoring of hundreds of users for many years to adequately explore the safety/risk profile of the products under investigation. Research surveys of EC users, on the other hand, can quickly provide information about potential harm of these products and are much cheaper to run. However, self reported data, highly self-selected study populations, and the cross-sectional design are some of the most common limitations of research surveys. Taken together, findings from surveys and follow-up studies of vapers have shown that EC use is relatively safe.

Polosa et al. [2013b] followed up smokers for 24 months, after a 6-month period of intervention during which ECs were given. Only mild symptoms such as mouth and throat irritation and dry cough were observed. Farsalinos et al [2013a] retrospectively evaluated a group of 111 EC users who had completely quit smoking and were daily EC users for a median period of 8 months. Throat irritation and cough were the most commonly reported side effects. Similar findings have been observed in surveys [Dawkins et al., 2013b; Etter et al. 2011]. However, it is expected that dedicated users who have more positive experience and less side effects compared to the general population participate in

such studies, therefore interpretation should be done with caution. The only two existing randomized controlled trials have also included detailed ECs safety analysis. The ECLAT study [Caponnetto et al. 2013a], a three-arms, controlled, randomized, clinical trial designed to compare efficacy and safety of a first-generation device with either 7.2 mg, or 5.4 mg, or 0 mg nicotine cartridges, reported clinically significant progressive health improvements already by week-2 of continuous use of a first-generation device and no serious adverse events (i.e. major depression, abnormal behavior or any event requiring unscheduled visit to the family practitioner or hospitalization) occurred during the study. The ASCEND study [Bullen et al. 2013], a three-arms, controlled, randomized, clinical trial designed to compare efficacy and safety of a first-generation device (with or without nicotine) to nicotine patch, reported no serious adverse events in any of the three study groups.

Few clinical studies have been performed to evaluate the short-term in vivo effects of EC use in current or former smokers. Vardavas et al. [2012] evaluated the acute effects of using an EC for 5 minutes on respiratory function. Although they did not report the results of commonly-used spirometry parameters, they found that a sensitive measure of airways resistance and nitric oxide levels in exhaled breath were adversely affected. Similar elevations in respiratory resistance were reported by another research group [Palamidas et al. 2013; Gennimata et al, 2012], who also documented some bizarre elevation in exhaled carbon monoxide levels after EC use; this finding has been challenged by several other studies [Farsalinos et al. 2013f; Nides et al. 2013; Van Stamer et al. 2013]. Schober et al. [2013] found that EC use led to elevated exhaled nitric oxide, contradicting the findings from Vardavas et al. Characteristically, none of the above studies performed any comparative tests after smoking tobacco cigarettes. Flouris et al. [2013] found that only smoking had an acute adverse effect on respiratory function; no difference was observed after the group of smokers was exposed to active or passive EC use.

Two studies have evaluated the short-term effects of ECs on the cardiovascular system. Farsalinos et al. [2012] evaluated the acute effects of using ECs with an 11mg/ml nicotine-containing liquid on hemodynamics and left ventricular function, in comparison with the effects of cigarette smoking. They found that EC use resulted in a slight elevation in diastolic blood pressure while after smoking both systolic and diastolic blood pressure and heart rate were significantly elevated. Obviously, this was due to the relatively low nicotine-content of the EC (which is considered medium-strength). Diastolic dysfunction was observed in smokers after smoking, which was in line with findings from previous

studies. However, no adverse effects were observed in EC users after using the device ad lib for 7 minutes. Another study by the same group [Farsalinos et al. 2013f] evaluated the acute effects of EC use on coronary flow. In particular, they measured flow velocity reserve of the left anterior descending coronary artery by echocardiography after intravenous infusion of adenosine, representing the maximal ability of the artery to deliver blood to the myocardium. Smoking was associated with a decline in flow velocity reserve by 16% and an elevation in resistance to flow by 19%. On the contrary, no difference was observed in any of these parameters after using the EC. Blood carboxyhemoglobin levels were also measured in participants; baseline values were significantly higher in smokers compared to vapers, were further elevated after smoking but were not altered after EC use. Similar observations for carboxyhemoglobin levels were observed by Van Staden et al. [2013]. Finally, a case report associated with a smoker suffering from chronic idiopathic neutrophilia was published. According to that report [Farsalinos and Romagna, 2013], switching from smoking to EC use led to reversal of the condition after 6 months. Additionally, C-reactive protein levels, which were consistently elevated for more than 6 years, decreased to normal levels.

One study evaluated the acute effects of tobacco and EC use on white blood cell (WBC) count [Flouris et al. 2012]. Smoking one tobacco cigarette caused an immediate elevation in WBCs, neutrophils and lymphocytes, indicating acute inflammatory distress. On the contrary, no differences were observed after using ECs.

In summary, clinical studies evaluating the effects of short-term EC use on selected cardiovascular and respiratory functional outcomes have shown that even if some harmful effects of vaping are reported, these are considerably milder compared to smoking conventional cigarettes.