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Acute and chronic effects of vaping electronic devices on lung physiology and inflammation

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Abstract

The impact of e-cigarette use on the inflammatory state and function of the lungs is not well understood. Here we review the latest studies on the impact of short and long term e-cigarette aerosol inhalation on molecular pathways, cellular recruitment, gas exchange and airway physiology. Inflammatory cytokines IL-6 and IL-8 were increased by e-cigarette exposures, and a variety of immune cells were recruited to the parenchyma and airways across models. While there are few consistent signals across *in vitro*, *in vivo* and human studies, due to the multitude of different e-devices and the combination of chemicals within different aerosols generated, it is clear that use of e-cigarettes does alter the inflammatory state and function of the lungs with both acute and chronic use. This is evidenced by the multitude of inflammatory lung diseases already tied to e-cigarette use, but the causal chemicals are primarily remain at large.

Introduction

Cigarette smoking is well known to cause many detrimental effects across the human body. Because the lungs come in direct contact with conventional tobacco smoke, they are more affected than any other organ. Electronic cigarettes (e-cigarettes) were introduced to the international market in 2003, and have been aggressively advertised as a healthy alternative to conventional cigarettes and as smoking cessation aids, but without supportive data [1,2•]. E-cigarettes produce an aerosol, commonly called e-cigarette vapor, by heating and aerosolizing e-liquids containing nicotine, propylene glycol, glycerol, flavorants, and other chemicals. E-cigarette aerosols typically contain 40–120 chemicals, such that the cells of the lungs are regularly exposed to this milieu in the setting of chronic use.

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Conflict of interest statement

Nothing declared.

While cigarette smoking has continued to decline worldwide, e-cigarette use has increased rapidly. E-cigarettes are used by approximately 9% of the US population, with the highest rates in teenagers and young adults (16–28%) [3]. Rapid evolution of e-devices and their e-liquids have made it difficult to define their acute and chronic health effects [2•]. Moreover, the variety of experimental approaches used to assess their impact have resulted in different and sometimes opposing findings [2•,4]. It is important to mention that some of these studies have been performed using different e-devices and different e-liquid constituents (flavorants, vehicles and other additives) [2•]. With the epidemic of e-cigarette or vaping product use-associated lung injury (EVALI), that affected thousands of vapers, began in 2019 and is ongoing, the relevance of studying pulmonary effects of e-cigarettes has become even more urgent. In addition, the unknown inflammatory and physiologic effects of e-cigarettes in individuals with COPD, asthma, lung cancer and cystic fibrosis are of great concern. Here, we review the known acute and chronic effects of e-cigarette use on lung physiology and inflammation based on published, peer-reviewed data generated with *in vitro* and *in vivo* models, and with human subjects.

E-cigarette effects on lung inflammation

Acute

The majority of data regarding inflammatory effects of e-cigarettes and their components are from *in vitro* approaches, with a focus on the production and release of cytokines and chemokines upon exposure to e-cigarette vapor. Using the definition of e-cigarette exposures of 4 weeks or less as acute, we discuss recent findings in the field across experimental models.

In vitro and ex vivo studies—Data from *in vitro* and *ex vivo* cell cultures represent the bulk of studies to date assessing inflammatory responses to acute e-cigarette aerosol exposures (Table 1). These exposure models either incorporated the aerosols into the growth media as vapor extracts, direct addition of e-liquids to media, or exposure of cells via the air-liquid interface (Table 1). Most studies have used epithelial cells [5–11,12•,13,14•], although a few have evaluated endothelial cells [15•], leukocytes [16] and fibroblasts [5,17]. One of the most common cytokines assessed is IL-8 (an important neutrophil chemoattractant). Most studies found that e-cigarettes induce IL-8 expression in epithelial cells [5,9,11,13,14•,17,18], while a few showed no induction or downregulation [12•,19]. Moreover, IL-8 has been also shown to be induced in fibroblasts [17]. IL-6 is also frequently assessed, and has been found to be increased upon e-cigarette exposure in epithelial cells [6,8,10,17], fibroblasts [17] and alveolar macrophages [16]. Similarly, a few studies show decrease or downregulation of IL-6 [8,18,19]. While changes in powerful inflammatory cytokines IL-6 and IL-8 are most consistent with a significant impact of e-cigarette aerosols on lung inflammation, the lack of consistency across studies suggests that the exact type of e-device and e-liquid (including factors such as flavorants and nicotine concentration), and duration of exposure, may be playing key roles, leading to disparate findings when different devices and liquids are used.

Beyond IL-6 and IL-8, other important inflammatory mediators that have been shown to be impaired using *in vitro* and *ex vivo* approaches are: TNFα [8,16], TGF-β [12•], CXCL8 [6,8,16], MCP-1 [16], and MMP-9 [8,16], PGE_2 [11,18], *TIMP1* [8], *SLPI* [8], NFκB [8], PI3 [8], IL10 [8] and IL13 [8]. Relatedly, Chatterji *et al.* showed that human serum harvested 2 hours after subjects used nicotine-free e-cigarette for 3 min induced inflammation in human pulmonary microvascular endothelial cells [15•]. These findings show that acute exposures as short as 3 min alter the systemic inflammatory state in e-cigarette vapers. Across studies, flavoring and nicotine were found to play key roles in the inflammatory modulation caused by acute e-cigarette aerosol exposures [5,8,10,11,16].

In vivo studies—The majority of *in vivo* e-cigarette studies have been carried out in rodents, specifically with C57BL/6 mice [9,12•,18,20,21]. Multiple immune cells have been found to be recruited to the lungs and airways upon acute e-cigarette exposure, including macrophages [9,20,21,22••,23], neutrophils [12•,18,22••,23], eosinophils [22••,23] and T cells [18,20,23]. Moreover, a recent study showed that exposure to e-cigarette aerosols downregulates innate immunity against viral pathogens in resident macrophages [24••]. In one model, an e-cigarette or vaping product use-associated lung injury (EVALI)-like condition was observed in rats exposed to e-cigarette aerosols for only for two hours, inducing alveolar wall thickening with foci of inflammation, red blood cell congestion, obliteration of alveolar spaces, and pneumonitis in some cases; bronchi showed accumulation of fibrin, inflammatory cells, and mucus plugs [25].

A Wide variety of cytokines and chemokines have been shown to be increased upon e-cigarette exposure, including Th2 cytokines: IL-3 [18], IL-4 [18], IL-5 [18,20], IL-9 [18,20], IL-13 [18], Th1 cytokines: IFN-γ [18,20], IL-2 [20], IL-12p70 [18], proinflammatory cytokines: IL-1α [18,20], IL-1β [12•,18,20,21], TNF-α [18,20], IL-6 [7,18,21], IL-17α [18], G-CSF [18], GM-CSF [18,20], chemokines: CCL4/MIP-1β [18,20], CCL11/eotaxin [18], inhibitory cytokines: IL-10 [18], growth factors: TGF-β1 [23], proteolytic enzymes that degrade extracellular matrix proteins: MMP2 [18,23], MMP8 [20] and MMP9 [20]. IL-1β [12•,18,20,21], IL-6 [7,18,21], TGF-β1 [23], IL-1α [18,20], MCP-1 [20], GM-CSF [20], IL-2 [20], IL-3 [18], IL-4 [18], IL-9 [18,20], IL-10 [18], IL-12p70 [18], IL-13 [18], IL-17α [18], IFNγ [18,20], RANTES [18,20], TNFα [18,20], MIP-1α [18], MIP-1β [18,20], IL-5 [18,20], KC [18], G-CSF [18], GM-CSF [18], eotaxin [18], MMP2 [18,23], MMP8 [20] and MMP9 [20]. Related to this, e-cigarettes have been shown to induce the pro-inflammatory NF-κB pathway activation in a dose-dependent fashion, with and without nicotine [12•]. In the context of pre-existing conditions, it has been shown that e-cigarette aerosol exposure can alter significantly airway inflammation in murine models of asthma [22••,23]. In addition, inflammatory responses driven by e-cigarettes seem to be strongly dependent on the sex of mice, and components of the e-liquid (including propylene glycol, flavorings and nicotine) [18,20,22••]. Thus, lung inflammation induced by e-cigarette aerosol inhalation in mice is variable across models and exposure systems. But the intensity of the signals seen, both in the cellular and molecular domains, again confirm that inhalation of these aerosols is not benign, and has direct effects on the inflammatory state of the lungs.

Human studies

A few studies have assessed acute effects of e-cigarette use in human subjects. A recent study suggested that cessation of vaping after 5 days of use can increase serum club cell protein-16 concentration in BAL, indicative of decreased lung inflammation [26]. Another study performed transcriptomic analyses on lung epithelial cell brushings and found no differences between subjects that vaped for 4 weeks versus control subjects. In addition, no significant differences in BAL inflammatory cell counts or cytokines were found [27]. Thus, it is not yet clear whether animal models accurately mimic the effects of e-cigarette use in humans.

Chronic

Data concerning the chronic effects of e-cigarettes on lung inflammation are lacking. There is a significant gap in knowledge about chronic effects of vaping and more studies are needed. Cigarette smoking is well known to have different effects on lung inflammation depending on the years to decades of use, and it is likely that other inhalants, such as e-cigarettes, will also induce changes that evolve over duration of use. Because human users vape a wide variety of e-cigarette devices and e-liquids, studies of chronic effects are so far limited to the few devices and e-liquids studied to date.

Animal studies—Although some studies assessing chronic e-cigarette exposures in mice have found no changes in lung inflammation [14•,24••,28], other have shown increased dendritic cells, $CD4^+$ T cells and $CD19^+$ B cells, irrespective of the presence of flavoring [29]. Furthermore, it has also been shown that e-cigarette aerosols can impair inflammatory lipid mediators such as 2-arachidonoylglycerol (2-AG), an anti-inflammatory mediator and 12-hydroxyeico-satetraenoic acid (12-HETE), an inflammatory mediator, after chronic inhalation, regardless of the flavoring [29]. In challenge models, e-cigarettes were found to downregulate innate immunity against viral pathogens in resident macrophages, although mice had enhanced lung inflammation and tissue damage upon challenge with influenza [24••]. Chronic e-cigarette aerosol inhalation was found to enhance hyperresponsiveness to methacholine, regardless of the presence or absence of nicotine [28]. And a pregnancy study using maternal nicotine and nicotine-free e-cigarette aerosol exposures, mothers' lungs developed increased levels of proinflammatory cytokines IL-1β, IL-6, and TNF-α (regardless of presence of nicotine) and the adult offspring had increased TNF-α levels, whereas IL-1β was suppressed [30].

Human studies

A recent transcriptomic study from bronchoscopies found 181 transcripts that were related specifically to e-cig use as compared to smokers and non-smokers, which included genes involved in inflammation with significant differences between e-cig users and never-smokers for IL-1β, IL-6 and IFN-γ [31]. In addition, it has been determined that use of nicotine and flavor-containing e-cigarettes upregulates pro-inflammatory cytokines and inflammasome-related genes in bronchial epithelial cells, such as CCL5, CCR1, CXCL1, CXCL2, NOD2, and ASC [32]. In terms of cellular changes, one study found no differences in neutrophil and macrophage counts in the airways of e-cigarette users versus non-vapers [31]. However,

another study found high lipid-laden macrophages (LLM) in the airways of e-cigarette users, with no THC or conventional tobacco exposures. Furthermore, LLM were significantly associated with inflammatory cytokines IL-4 and IL-10 in e-cig users [33].

Case studies have associated e-cigarette use with chronic inflammation and have defined vaping as a risk factor for bronchiectasis, ground glass opacities, and nodule formation [34].

One series of three e-cigarette users hospitalized for acute shortness of breath found airway inflammation (mostly eosinophilic) and demonstrated a likely response to high-dose systemic corticosteroid treatment [35]. Over the last 10 years, e-cigarette use has been found to induce numerous inflammatory lung diseases. However, the driving chemicals and e-devices of these pathologic lung effects remain almost entirely unknown.

Physiology studies—While alterations in the inflammatory state of the lung due to e-cigarette use are important and are indicative of the lung diseases and susceptibility to clinical challenges to come, defining changes in the physiologic state of the lungs induced by inhalants also helps determine the long and short term pathologic effects. Only a small number of human and animal studies have been published in the last two years, since our comprehensive review of e-cigarette effects on lung and cardiac physiology [4]. These recent studies are reviewed here.

Human studies

Of the studies in human subjects, one focused on changes relative to conventional tobacco products and found that mean mid-expiratory phase of forced expiratory (FEF25%–75%) and blood pressure were no different in JUUL users versus conventional cigarette smokers [36] (Table 2). Chaumont *et al.* assessed acute effects of vaping and found transcutaneous partial pressure arterial O_2 ($TcpO_2$) to be decreased after acute nicotine and nicotine-free vaping, while transcutaneous partial pressure arterial CO_2 decreased after acute nicotine vaping alone. These findings are likely due to transient gas exchange disturbance induced by vaping. Baseline heart rate was also found to be decreased after short-term e-cigarette cessation, suggesting improvements in gas exchange [26]. Assessment of short-term respiratory responses after acute e-cigarettes use found decreased FeNO and airflow indices (PEF, MEF75), as well as increased exhaled airway temperature [37]. Together, these recent studies in human subjects consistently show an impact of short-term e-cigarette use on airway resistance and gas exchange.

Animal studies—Female C57BL/6 mice exposed daily for six weeks to VG-rich e-cigarette aerosols (70%/30% VG/PG), without nicotine or flavoring, were found to have increased populations of immune cells within the lung. This suggests that e-cigarette delivery vehicles alone influence the lung immunophenotype [29]. In contrast, exposure to e-cigarette aerosols containing chemicals to convey vanilla flavor led to physiologic changes, including higher tidal and minute volumes than air controls [29]. Consistently, across recent and past studies of e-cigarettes, different chemical compositions of the aerosols lead to variable effects on lung physiology, with both vehicle components (PG/VG), active components (nicotine), flavorants and contaminants leading to adverse effects on lung function.

Conclusions

More studies are desperately needed in define the inflammatory and physiologic effects of e-cigarette use on the lungs. In particular, the kinetics of lung inflammation upon exposure to e-cigarette aerosols remains unclear, with current data pointing to induction of inflammation during acute exposures, with evolution of inflammation over time such that acute patterns of lung inflammation and injury are vastly different than those seen with chronic exposure. While airway resistance, lung compliance and gas exchange have all been found to be adversely impacted by some, but not all, e-cigarette exposures, the exact chemicals causing the pathologic effects have not yet been defined. We can be clear with e-cigarette users, policy makers and regulatory bodies: e-cigarette use fundamentally alters the immunologic and physiologic state of the lungs and causes disease.

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References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

• of special interest

•• of outstanding interest

1. Hon LI: Best Partners Worldwide Ltd Fontem Holdings 1 BV, assignee. Electronic Atomization Cigarette. 2003. https://patentsgooglecom/patent/US8393331B2/en. (Accessed 01 April 2020).
2 •. Bozier J, Chivers EK, Chapman DG, Larcombe AN, Bastian N, Masso-Silva JA et al. : The evolving landscape of electronic cigarettes: a systematic review of recent evidence. Chest 2020, 157:1362–1390 [PubMed: 32006591] This comprehensive review of e-cigarette effects on health covers multiple organ systems, acute and chronic effects, and tackles the difficult problem of comparing findings across different models.
3. Miech R, Johnston L, O'Malley PM, Bachman JG, Patrick ME: Trends in adolescent vaping, 2017–2019. N Engl J Med 2019, 381:1490–1491. [PubMed: 31532955]
4. Tsai M, Byun MK, Shin J, Crotty Alexander LE: Effects of e-cigarettes and vaping devices on cardiac and pulmonary physiology. J Physiol 2020, 598:5039–5062. [PubMed: 32975834]
5. Gerloff J, Sundar IK, Freter R, Sekera ER, Friedman AE, Robinson R et al. : Inflammatory response and barrier dysfunction by different e-cigarette flavoring chemicals identified by gas chromatography-mass spectrometry in e-liquids and e-vapors on human lung epithelial cells and fibroblasts. Appl In Vitro Toxicol 2017, 3:28–40.
6. Higham A, Bostock D, Booth G, Dungwa JV, Singh D: The effect of electronic cigarette and tobacco smoke exposure on COPD bronchial epithelial cell inflammatory responses. Int J Chron Obstruct Pulmon Dis 2018, 13:989–1000. [PubMed: 29615835]
7. Husari A, Shihadeh A, Talih S, Hashem Y, El Sabban M, Zaatari G: Acute exposure to electronic and combustible cigarette aerosols: effects in an animal model and in human alveolar cells. Nicotine Tob Res 2016, 18:613–619. [PubMed: 26272212]
8. Ganguly K, Nordström A, Thimraj TA, Rahman M, Ramström M, Sompa SI et al. : Addressing the challenges of E-cigarette safety profiling by assessment of pulmonary toxicological response in bronchial and alveolar mucosa models. Sci Rep 2020, 10:20460. [PubMed: 33235237]

9. Bahmed K, Lin CR, Simborio H, Karim L, Aksoy M, Kelsen S et al. : The role of DJ-1 in human primary alveolar type II cell injury induced by e-cigarette aerosol. Am J Physiol Lung Cell Mol Physiol 2019, 317:L475–L485. [PubMed: 31313616]

10. Gellatly S, Pavelka N, Crue T, Schweitzer KS, Day BJ, Min E et al. : Nicotine-free e-cigarette vapor exposure stimulates IL6 and mucin production in human primary small airway epithelial cells. J Inflamm Res 2020, 13:175–185. [PubMed: 32368126]

11. Muthumalage T, Lamb T, Friedman MR, Rahman I: E-cigarette flavored pods induce inflammation, epithelial barrier dysfunction, and DNA damage in lung epithelial cells and monocytes. Sci Rep 2019, 9:19035. [PubMed: 31836726]

12 •. Ma T, Wang X, Li L, Sun B, Zhu Y, Xia T: Electronic cigarette aerosols induce oxidative stress-dependent cell death and NF-κB mediated acute lung inflammation in mice. Arch Toxicol 2021, 95:195–205 [PubMed: 33159582] This work is the first to implicate activation of the powerful NFkB axis by e-cigarette aerosols. Multiple other studies have found cytotoxicity caused by aerosol exposures, and this work confirms those findings in mice.

13. Herr C, Tsitouras K, Niederstraßer J, Backes C, Beisswenger C, Dong L et al. : Cigarette smoke and electronic cigarettes differentially activate bronchial epithelial cells. Respir Res 2020, 21:67. [PubMed: 32164736]

14 •. Crotty Alexander LE, Drummond CA, Hepokoski M, Mathew D, Moshensky A, Willeford A et al. : Chronic inhalation of e-cigarette vapor containing nicotine disrupts airway barrier function and induces systemic inflammation and multiorgan fibrosis in mice. Am J Physiol Regul Integr Comp Physiol 2018, 314:R834–R847 [PubMed: 29384700] The *in vivo* work in mice was the first of its kind to find a powerful phenotype induced by e-cigarette aerosol inhalation — fibrosis in heart, kidney and liver tissues. In addition, the *ex vivo* work using primary human airway epithelial cells was the first to find increased permeability and increased IL-8 secretion in response to a few short exposures to the core ingredients within e-cigarette aerosols (PG, VG and nicotine).

15 •. Chatterjee S, Tao J-Q, Johncola A, Guo W, Caporale A, Langham MC et al. : Acute exposure to e-cigarettes causes inflammation and pulmonary endothelial oxidative stress in nonsmoking, healthy young subjects. Am J Physiol Lung Cell Mol Physiol 2019, 317:L155–L166 [PubMed: 31042077] This work in human subjects confirmed what *in vivo* and *ex vivo* studies had been telling us for years, that even short-term use of e-cigarettes has a definable impact on the inflammatory state of the body. Additionally, these findings were seen with nicotine-free aerosols, confirming that nicotine is not the only concerning chemical within e-devices, and that the other constituents must be studied to fully understand the impact of e-cigarette use on health.

16. Scott A, Lugg ST, Aldridge K, Lewis KE, Bowden A, Mahida RY et al. : Pro-inflammatory effects of e-cigarette vapour condensate on human alveolar macrophages. Thorax 2018, 73:1161–1169. [PubMed: 30104262]

17. Lerner CA, Rutagarama P, Ahmad T, Sundar IK, Elder A, Rahman I: Electronic cigarette aerosols and copper nanoparticles induce mitochondrial stress and promote DNA fragmentation in lung fibroblasts. Biochem Biophys Res Commun 2016, 477:620–625. [PubMed: 27343559]

18. Wang Q, Khan NA, Muthumalage T, Lawyer GR, McDonough SR, Chuang TD et al. : Dysregulated repair and inflammatory responses by e-cigarette-derived inhaled nicotine and humectant propylene glycol in a sex-dependent manner in mouse lung. FASEB Bioadv 2019, 1:609–623. [PubMed: 31825014]

19. Czekala L, Simms L, Stevenson M, Tschierske N, Maione AG, Walele T: Toxicological comparison of cigarette smoke and e-cigarette aerosol using a 3D in vitro human respiratory model. Regul Toxicol Pharmacol 2019, 103:314–324. [PubMed: 30721718]

20. Wang Q, Sundar IK, Li D, Lucas JH, Muthumalage T, McDonough SR et al. : E-cigarette-induced pulmonary inflammation and dysregulated repair are mediated by nAChR α7 receptor: role of nAChR α7 in SARS-CoV-2 Covid-19 ACE2 receptor regulation. Respir Res 2020, 21:154. [PubMed: 32552811]

21. Glynos C, Bibli S-I, Pavlidou A, Katsaounou P, Kalomenidis I, Zakynthinos S et al. : Comparison of the effects of e-cigarette vapor vs cigarette smoke on lung function and inflammation in mice. Eur Resp J 2015, 46(Suppl. 59):OA279.

22 ••. Chapman DG, Casey DT, Ather JL, Aliyeva M, Daphtary N, Lahue KG et al. : The effect of flavored e-cigarettes on murine allergic airways disease. Sci Rep 2019, 9:13671 [PubMed: 31541174] Very few studies have been conducted using relevant clinical challenges. This work defined the impact of nicotine and several flavorants on the development and severity of asthma phenotypes in mice. The authors found that nicotine has specific anti-inflammatory effects, while different flavors impacted asthma pathogenesis in different ways.

23. Taha HR, Al-Sawalha NA, Alzoubi KH, Khabour OF: Effect of e-cigarette aerosol exposure on airway inflammation in a murine model of asthma. Inhal Toxicol 2020, 32:503–511. [PubMed: 33297792]

24 ••. Madison MC, Landers CT, Gu B-H, Chang C-Y, Tung H-Y, You R et al. : Electronic cigarettes disrupt lung lipid homeostasis and innate immunity independent of nicotine. J Clin Investig 2020, 129:4290–4304This work was highly timely as it showed that inhalation of e-cigarette aerosols by mice led to increased lung injury and dysfunction in the setting of a viral respiratory infection. Significantly, aerosols generated without nicotine in the e-liquids led to the same degree of severe lung inflammation with influenza infection, again confirming that the non-nicotine contents of e-cigarettes are impacting the immunophenpotype of the lung and are increasing the risk of adverse responses to common clinical challenges.

25. Kleinman MT, Arechavala RJ, Herman D, Shi J, Hasen I, Ting A et al. : E-cigarette or vaping product use-associated lung injury produced in an animal model from electronic cigarette vapor exposure without tetrahydrocannabinol or vitamin e oil. J Am Heart Assoc 2020, 9:e017368. [PubMed: 32896206]

26. Chaumont M, Tagliatti V, Channan EM, Colet JM, Bernard A, Morra S et al. : Short halt in vaping modifies cardiorespiratory parameters and urine metabolome: a randomized trial. Am J Physiol Lung Cell Mol Physiol 2020, 318:L331–L344. [PubMed: 31721596]

27. Song MA, Reisinger SA, Freudenheim JL, Brasky TM, Mathé EA, McElroy JP et al. : Effects of electronic cigarette constituents on the human lung: a pilot clinical trial. Cancer Prev Res (Phila) 2020, 13:145–152. [PubMed: 31619441]

28. Larcombe AN, Janka MA, Mullins BJ, Berry LJ, Bredin A, Franklin PJ: The effects of electronic cigarette aerosol exposure on inflammation and lung function in mice. Am J Physiol Lung Cell Mol Physiol 2017, 313:L67–L79. [PubMed: 28360111]

29. Szafran BN, Pinkston R, Perveen Z, Ross MK, Morgan T, Paulsen DB et al. : Electronic-cigarette vehicles and flavoring affect lung function and immune responses in a murine model. Int J Mol Sci 2020, 21.

30. Chen H, Li G, Chan YL, Chapman DG, Sukjamnong S, Nguyen T et al. : Maternal e-cigarette exposure in mice alters DNA methylation and lung cytokine expression in offspring. Subst Use Misuse 2018, 53:734–743 10.1080/10826084.2017.1363237 Epub 2017 Sep 29. [PubMed: 28960130]

31. Song MA, Freudenheim JL, Brasky TM, Mathe EA, McElroy JP, Nickerson QA et al. : Biomarkers of exposure and effect in the lungs of smokers, nonsmokers, and electronic cigarette users. Cancer Epidemiol Biomarkers Prev 2020, 29:443–451. [PubMed: 31848205]

32. Lee AC, Chakladar J, Li WT, Chen C, Chang EY, Wang-Rodriguez J et al. : Tobacco, but not nicotine and flavor-less electronic cigarettes, induces ACE2 and immune dysregulation. Int J Mol Sci 2020, 21.

33. Shields PG, Song MA, Freudenheim JL, Brasky TM, McElroy JP, Reisinger SA et al. : Lipid laden macrophages and electronic cigarettes in healthy adults. EBioMedicine 2020, 60:102982. [PubMed: 32919101]

34. Mull ES, Shell R, Adler B, Holtzlander M: Bronchiectasis associated with electronic cigarette use: a case series. Pediatr Pulmonol 2020, 55:3443–3449. [PubMed: 32902877]

35. Suhling H, Welte T, Fuehner T: Three patients with acute pulmonary damage following the use of e-cigarettes—a case series. Dtsch Arztebl Int 2020, 117:177–182. [PubMed: 32327029]

36. Pulvers K, Nollen NL, Rice M, Schmid CH, Qu K, Benowitz NL et al. : Effect of pod e-cigarettes vs cigarettes on carcinogen exposure among African American and Latinx smokers: a randomized clinical trial. JAMA Netw Open 2020, 3:e2026324–e. [PubMed: 33206193]

37. Brożek GM, Jankowski M, Zejda JE: Acute respiratory responses to the use of e-cigarette: an intervention study. Sci Rep 2019, 9:6844. [PubMed: 31048778]

Table 1

E-cigarette effects on lung inflammation

Experimental Approach	Species	Nicotine	Flavored	Exposure time	Key findings	Ref
In vitro	Human	Yes	No	15 min daily × 2–5 days	E-cigarette-exposed normal human bronchial epithelial cells (NHBEs) secreted greater IL-8 in response to bacterial infection	[14•]
In vitro	Human	Yes	Yes	10 min	Exposure at air-liquid interface (ALI) led to increased IL-8 and IL-6 expression in human lung fibroblasts (HFL-1)	[17]
In vitro	Human	Yes	No	15 min	Exposure induced IL-8 secretion from Calu-3 cells but had no effect on NCI-H292 or pHBE cells.	[13]
In vitro	Human	Yes	No	15 min	• IL-6 levels decreased significantly in the PG with nicotine and PG alone groups compared to healthy controls. However, IL-6 levels significantly increased in the COPD group exposed to inhaled e-cig aerosols containing PG with nicotine. • Acute exposure to inhaled e-cig aerosols containing PG with or without nicotine induced a significant increase in IL-8 levels in the healthy donor group, but showed no significant difference in the COPD group. • E-cig aerosol containing PG with nicotine suppressed the release of PGE2 in healthy donors	[18]
In vitro	Human	No	No	2 hours	Nicotine-free e-cig aerosol induced oxidative stress and inflammation (assessed by ICAM-1 expression and ROS generation) in human pulmonary microvascular endothelial cells (HPMVEC)	[15•]
In vitro	Human	Yes	No	24 hours	E-cig aerosol exposure increased TGF-β and decreased IL-8 in BEAS-2B cells	[12•]
In vitro	Human	No	Yes	24 hours	• Flavorings (acetoin, diacetyl, ortho-vanillin and maltol) induced IL-8 release in Beas2B cells • Acetoin-treated and pentanedione-treated HFL-1 cells induced IL-8 release • Acetoin and maltol were more potent inducers of IL-8 release	[5]
In vitro	Human	Yes	Yes	24 hours	E-cig extracts caused release of IL-6 and CXCL8 from Calu-3 cells and dampened poly I:C-stimulated CXCL10 release	[6]
In vitro	Human	Yes	Yes	6 × 15 min/24 hours	Using human bronchial and alveolar lung mucosa models, significant changes in inflammatory mediators (TNF, CXCL8, GSTA1, HMOX1, SOD3, MMP9, TIMP1, SLPI, NFKB1 and DEFB4A, PI3, IL1B, IL6, IL10 and IL13) were fund in response to e-cig aerosol exposures. Differences depended on flavors, nicotine content, and model (bronchial or alveolar).	[8]
In vitro	Human	Yes	Yes	3 × 30 min 24 hours	Lung epithelial cells (16-HBE, BEAS-2B) exposed to various pod aerosols (primarily JUUL) resulted in increased inflammatory mediators, such as IL-8 and PGE2	[11]
In vitro	Human	Yes	Yes	80, 240 or 400 puffs	Tissues exposed to up to 400 puffs of e-cigarette aerosol with or without blueberry flavor did not differ in IL-6 and IL-8 production as compared to air-exposed tissues in any of the measured endpoints.	[19]
In vitro/in vivo	Human/ Mouse	Yes	Yes	3 days	E-cig exposure led to increased IL-1β	[7]
Ex vivo	Human	Yes	No	24 hours	ECVC/nfECVC increased ROS production but only ECVC induced IL-6, TNFα, CXCL-8, MCP-1 and MMP-9 in alveolar macrophages.	[16]

Experimental Approach	Species	Nicotine	Flavored	Exposure time	Key findings	Ref
Ex vivo	Human	Yes	No	24 hours	Human alveolar type II cell exposure to e-cigarette aerosol increased IL-8 levels	[9]
Ex vivo	Human	Yes	No	24 hours	Nicotine-free e-vapor and nicotine containing e-vapor significantly increased IL-6	[10]
In vivo	Mouse	Yes	Yes	3 days	Increased IL-6 in BAL	[7]
In vivo	Mouse	Yes	No	3 days	Increased macrophage recruitment into the lungs	[9]
In vivo	Rat	Yes	No	2 hours	Induction of E-cigarette or vaping product use-associated lung injury (EVALI)-like condition acutely after use of a nichrome heating element at high power, without the use of tetrahydrocannabinol, vitamin E, or nicotine.	[22••]
In vivo	Mouse	Yes	No	30 days	Subacute e-cig exposure increased inflammatory cellular influx of macrophages and T-lymphocytes including increased pro-inflammatory cytokines in BALF	[20]
In vivo	Mouse	Yes	Yes	3 days and 4 weeks	E-cig aerosols, especially those containing PG:VG, nicotine and flavor, increased BALF cellularity. IL-6 and IL-1β were increased at 3 days (only in PG:VG-N + F), but not at 4 weeks. In many cases, the added flavor in e-cigs exacerbated the detrimental effects of e-cigs.	[21]
In vivo	Mouse	Yes	No	4 weeks	Exposure to E-Cig aerosol significantly increased the number of neutrophils, eosinophils, macrophages and lymphocytes in BALF and reduce levels of TGF-β1 and MMP-2 in lung homogenates.	[24••]
In vivo	Mouse	Yes	Yes	0–18 days	Flavored e-cigarettes without nicotine had significant but heterogeneous effects on features of allergic airways disease. This suggests that some flavored e-cigarettes may alter asthma pathophysiology even when used without nicotine	[23]
In vivo	Mouse	Yes		4 weeks, 3–4 months	Mice exposed for four weeks to ENDS vapor exposure downregulated innate immunity against viral pathogens in resident macrophages. At eight weeks ENDS exposure alters immune responses and recovery from influenza A infection. Although, at four months exposure to ENDS does not induce inflammation in the lung but had decreased production of surfactant proteins in airways	[25]
In vivo	Human	Yes	No	5 days	Cessation of vaping after five days of use increased serum club cell protein-16 concentration, an indicative of decrease in lung inflammation	[26]
In vivo	Human	No	No	4 weeks	Transcriptomic analysis from lung epithelial cell brushings showed no significant changes between subjects that vaped for four weeks versus non-vaper controls. In addition, no significant differences in changes of BAL inflammatory cell counts or cytokines were found either	[27]
In vivo	Mouse	Yes	Yes	8 weeks	Mice exposed to e-cigarette aerosol did not have increased inflammation, although mice exposed to glycerin-based e-cigarette aerosols were hyperresponsive to methacholine regardless of the presence or absence of nicotine	[28]
In vivo	Mouse	Yes	Yes	6 weeks	Increased number of dendritic cells, $CD4^+$ T cells and $CD19^+$ B cells, and impaired lipid inflammatory mediators in the VG/PG-exposed group compared to air, irrespective of the presence of vanilla flavoring.	[29]
In vivo	Mouse	Yes	No	6 months	Mice that inhaled e-cigarette vapor no pulmonary inflammation, emphysema or fibrosis relative to Air controls.	[14•]
In vivo	Mouse	Yes	No	6 weeks dams 13 weeks offspring	Pregnant mice were exposed to e-cigarette (with and without nicotine) vapor and their lungs had increased proinflammatory cytokines, such as IL-1β, IL-6, and TNF-α. In adult offspring (after 13 weeks), TNF-α protein levels were increased, whereas IL-1β was suppressed.	[30]
In vivo	Human	Yes	Non-specified	Non-specified	Bronchiectasis was associated with the chronic use of electronic cigarette	[33]
In vivo	Human	Yes	Non-specified	1–3 years	Three patients hospitalized for acute shortness of breath were associated with airways inflammation (mostly eosinophilic) and they recovered with high-dose systemic corticosteroid treatment	[34]

Experimental Approach	Species	Nicotine	Flavored	Exposure time	Key findings	Ref
In vivo	Human	Yes	Non-specified	>1 year	High lipid-laden macrophages (LLM) were found in the lungs (bronchial alveolar fluids BAL) of half of the e-cig users analyzed. LLM were not related to THC exposure or smoking history. LLM were significantly associated with inflammatory cytokines IL-4 and IL-10 in e-cig users	[35]
In vivo	Human	Yes	Yes	2.7 years (average)	Bronchoscopies showed that e-cigarette users had similar macrophages and neutrophil counts than never-smokers. There were significant differences between e-cig users and never-smokers for IL-1β, IL-6 and IFN-γ. Study found 181 transcripts that were related specifically to e-cig use	[31]
In vivo	Human	Yes	Yes	>1 month	bronchial epithelial cells of vapers using nicotine and flavor-containing e-cigs led to upregulation of pro-inflammatory cytokines and inflammasome-related genes, including CCL5, CCR1, CXCL1, CXCL2, N0D2, and ASC	[32]

Table 2

E-cigarette effects on lung physiology

Experimental Approach	Species	Nicotine	Flavored	Subjects	Exposure Time	Key findings	Limitations	Ref.
Unblinded randomized interventional trial	Human	Yes (5%)	Yes (JUUL flavor of choice)	E-cig users (n = 126) Tobacco smokers (n = 61) Age: 43 ± 12 40% women	6 weeks	• The e-cigarette group had significantly greater reductions in urine • NNAL (a pulmonary tobacco-specific carcinogen), eCO, respiratory symptoms, and number of cigarettes smoked in the past seven days among those still smoking than the cigarettes as usual group. • Lung function and BP were similar in the two groups. • Cotinine was not significantly different at week six.	The six-week study period is insufficient to understand long-term effects of e-cigs.	[36]
In vivo	Mouse	No	Yes (vanilla)	Six-week old C57BL/6 female mice. n = 11–12/ group	2 hours/d for six weeks 70%30% VG/PG, and 70%30% VG/PG + vanilla flavoring	• Immunophenotyping of lung immune cells revealed an increased number of dendritic cells, CD4+ T cells, and CD19+ B cells in the VG/PG-exposed group compared to air, irrespective of the presence of vanilla flavoring. • Quantification of bioactive lung lipids demonstrated a >3-fold increase of 2-AG, an anti-inflammatory mediator, and a 2-fold increase of 12- • HETE, another inflammatory mediator, following VG/PG exposure, with or without vanilla flavoring. Take away: VG and PG disrupt immune homeostasis.	No difference in gene expression at six weeks in this study, contrary to the difference at 16 weeks in Madison *et al.* [9]	[29]
Randomized, investigator-blinded, three-period crossover study	Human	Yes	Yes	E-cig users (n = 30) whom were former tobacco smokers Age: 38 ± 2 years 100% male	**1** Vaping of nicotine e-cigs for 5d **2** Nicotine-free-vaping for 5d	• Compared with nicotine-free-sessions and nicotine-free-sessions, a specific metabolomic signature characterized the stop-session. • Baseline serum club cell protein-16 was higher during the stop-session • Heart rate was elevated in nicotine-vaping session	• No monitoring of vaping during the 5d before experimental sessions. • This study enrolled only male participants.	[26]

Experimental Approach	Species	Nicotine	Flavored	Subjects	Exposure Time	Key findings	Limitations	Ref.
					3 Cessation of vaping for 5d	• Compared with acute sham-vaping in the stop-session, acute nicotine-vaping and acute nicotine-free vaping slightly decreased skin oxygen tension. • FEF-25% was higher in the stop-session compared to the nicotine-free-session. Take away: Short-term e-cigarette cessation decreased baseline heart rate and increased CC16 and FEF-25%, suggesting slight improvement of airway status.		
Interventional cohort study	Human	Yes	Yes	Cigarette smokers, e-cigarette users, dual users and controls Age: 21.7 ± 2 years n = 30/group	E-cig use with everyday habits for 5 min. Measurements done at 1 and 30 min following exposure.	• Compared with controls, lower FeNO were found in cigarette smokers and dual users. • CO concentrations were lower in controls relative to inhalant users. • Smokers and dual users had decreased PEF and MEF75. • E-cigarette use was associated with decreased FeNO and airflow indices (PEF, MEF75), but increased airway temperature.	Between-subject differences in device used in the experiment might have affected the group results. There were small numbers of subjects in each arm.	[37]

*Studies where findings were likely to be biased by funding and authors had significant conflicts of interest. eCO: exhaled carbon monoxide.