Characterization of toxic metal exposure from electronic cigarette use

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Conflict of Interest disclosure

The authors have no financial disclosure or conflict of interest with the material in the presentation.
Outline

• Introduction: Electronic cigarettes
  • How they work
  • Increase in teen use + market growth
• Why are we concerned?
  • What we know so far about composition
  • Focus on metals
• Research Findings
  • Daily E-cig user demographics
  • Metals in biospecimens of e-cig users
  • Metals in aerosol samples
• Risk Balance
• Future directions

Electronic cigarettes (e-cigarettes)

• Heat a liquid solution to produce an aerosol that is inhaled by the user
• Solutions (e-liquids) typically include
  • Nicotine, flavorings, glycerin and propylene glycol
• “Vaping” – Inhaling aerosol generated by an e-cigarette

How do they look like?

- 1st generation: Cig-a-likes
- 2nd generation: Pens (2010)
- 3rd generation: MODS (~2010)

Prevalence of e-cigarette use
- Increased since introduced in 2006
  - Adult Current use: 2.8% (6.9 million) (NHIS, 2017)
- Most popular tobacco product among youth, with current use

- High school students:
  - 2017: 11.7%
  - 2011: 1.5%

- Middle school students:
  - 2017: 3.3%
  - 2011: 0.6%

Image source: 2017 National Youth Tobacco Survey
**E-Cigarette Unit Sales by Manufacturer — US, 2013-2017**

![Graph showing e-cigarette unit sales by manufacturer from 2013 to 2017. The graph highlights the dominance of Juul Laboratories and other major brands like Altria (MarkTen, Green Smoke), British American Tobacco (Vuse), Imperial Tobacco (Blu), and Logic.](https://www.cdc.gov/tobacco/basic_information/e-cigarettes/pdfs/Electronic-Cigarettes-Infographic-p.pdf)


**Why are we concerned?**

Research is playing catch-up to fast market changes

- EVALI outbreak over the summer 2019 (THC / Vit E acet)
- little is known about long-term effects of e-cigarette use

Image source: https://www.cdc.gov/tobacco/basic_information/e-cigarettes/pdfs/Electronic-Cigarettes-Infographic-p.pdf*
Why Metals?

– E-liquid is in contact with heating coil
– Common heating coil alloys:
  § Nichrome: nickel-chromium alloy (80/20 %)
  § Kanthal: iron-chromium-aluminum alloy
  § Stainless steel: chromium (min 10.5%)-nickel-manganese-carbon alloy

![E-cigarette components]

Metal toxicity

• Decreased lung function, respiratory irritation, asthma (Ni, Cr (VI), Zn, Cu, Al)
• Type 1 Lung Carcinogens (Ni and Cr(VI))
• Neurotoxicants (Mn, Pb)
• Cardiovascular and kidney disease (Pb)
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• Why are we concerned?
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  • Focus on metals
• Research Findings
  • Daily E-cig user demographics, behaviors, device type
    • Who may be at higher risk of metal exposure?
  • Metals in biospecimens of e-cig users
  • Metals in aerosol samples
• Risk Balance
• Future directions

Research Study

• Maryland residents, ≥ 18 years
• December 2015 – October 2017
• E-cigarette users (n= 100)
  • Daily exclusive users for at least 6 weeks
  • If former smoker, quit at least 6 months
  • MODs/tanks
• Non-e-cigarette users (n= 50)
  • Friends/colleagues (1st strategy)
  • Similar characteristics (age, sex, race)
• Recruitment
  • Flyers posted in vape shops, universities
  • Ads in the local paper; Craigslist
  • E-flyers on social media
  • Vaping expos
Study Recruitment: Vape Expo

Maryland Vape Expo, April 2016

Study Recruitment: Vape Expo

Interview-based questionnaire

- 120 questions
- Demographics, tobacco use, self-reported health status
- E-cigarette users: device characteristics, use behaviors, reasons for use, intentions to quit

Statistical Analysis

- Linear regression to assess mean differences in use behaviors/device preferences by demographics
  - E-liquid consumed/week
  - Voltage
  - Nicotine content
  - Puff count/day
### Participant characteristics by vaping status

<table>
<thead>
<tr>
<th>General Characteristics</th>
<th>N Total (n = 150)</th>
<th>E-cig users (n = 100)</th>
<th>Non-users (n = 50)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, mean (SD)</td>
<td>150 30.1 (9.6)</td>
<td>30.3 (9.2)</td>
<td>29.7 (10.5)</td>
<td>0.7</td>
</tr>
<tr>
<td>Gender %</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>97 64</td>
<td>67</td>
<td>60</td>
<td>0.60</td>
</tr>
<tr>
<td>Female</td>
<td>59 36</td>
<td>33</td>
<td>40</td>
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</tr>
<tr>
<td>Education level %</td>
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<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>≤ High School</td>
<td>46 30.7</td>
<td>41</td>
<td>10</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>&gt; High School</td>
<td>104 69.3</td>
<td>59</td>
<td>90</td>
<td></td>
</tr>
<tr>
<td>Race %</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>124 82.7</td>
<td>87.0</td>
<td>74.0</td>
<td>0.05</td>
</tr>
<tr>
<td>Non-White</td>
<td>26 17.3</td>
<td>13.0</td>
<td>26.0</td>
<td></td>
</tr>
<tr>
<td>Employed %</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>99 66</td>
<td>75.0</td>
<td>48.0</td>
<td>0.001</td>
</tr>
<tr>
<td>No</td>
<td>51 34</td>
<td>25.0</td>
<td>52.0</td>
<td></td>
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<tr>
<td>Current Student</td>
<td>29 19.3</td>
<td>9.00</td>
<td>40.0</td>
<td></td>
</tr>
</tbody>
</table>

*Comparing sole e-cig users vs. non-users

### E-cigarette use characteristics

- 72% owned 1 or 2 devices
- Median: 200 puffs/day, 56% vaping throughout the day
- 41% first vape within 15 minutes of waking in the AM
- Voltage: average (range) = 4.21 V (2.12 – 12.5 V)
- E-liquid consumption: 53.3 ml/wk (5-240 ml/wk)
- Nicotine concentration: 5.3 mg/ml (0-24 mg/ml)
<table>
<thead>
<tr>
<th>Age (per year)</th>
<th>N</th>
<th>Voltage (volts)</th>
<th>N</th>
<th>Nicotine Use (mg/ml)</th>
<th>N</th>
<th>E-liquid/wk (ml)</th>
<th>N</th>
<th>Puff count/day</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>92</td>
<td>-0.01 (0.03, 0.02)</td>
<td>98</td>
<td>0.24 (0.12, 0.36)</td>
<td>98</td>
<td>-0.46 (-1.55, 0.63)</td>
<td>50</td>
<td>-25.1 (-49.9, -0.25)</td>
</tr>
<tr>
<td>Gender</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
<td>65</td>
<td>0.00 (ref)</td>
<td>66</td>
<td>0.00 (ref)</td>
<td>67</td>
<td>0.00 (ref)</td>
<td>35</td>
<td>0.00 (ref)</td>
</tr>
<tr>
<td>Female</td>
<td>27</td>
<td>-0.54 (-1.04, -0.03)</td>
<td>32</td>
<td>0.20 (1.95, 2.34)</td>
<td>31</td>
<td>-22.7 (-42.9, -2.46)</td>
<td>15</td>
<td>-132.2 (-49.9, -0.25)</td>
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<tr>
<td>Education</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>≤ HS</td>
<td>38</td>
<td>0.00 (ref)</td>
<td>41</td>
<td>0.00 (ref)</td>
<td>41</td>
<td>0.00 (ref)</td>
<td>19</td>
<td>0.00 (ref)</td>
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<tr>
<td>&gt; HS</td>
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<td>-0.02 (-2.09, 2.04)</td>
<td>57</td>
<td>-20.4 (-39.7, -1.09)</td>
<td>31</td>
<td>134.1 (-301, 570)</td>
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<tr>
<td>Previous smoker</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
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<tr>
<td>No</td>
<td>10</td>
<td>0.00 (ref)</td>
<td>11</td>
<td>0.00 (ref)</td>
<td>11</td>
<td>0.00 (ref)</td>
<td>6</td>
<td>0.00 (ref)</td>
</tr>
<tr>
<td>Yes</td>
<td>82</td>
<td>-1.31 (-2.10, -0.53)</td>
<td>87</td>
<td>-1.09 (-4.38, 2.19)</td>
<td>87</td>
<td>-3.10 (-33.9, 27.7)</td>
<td>44</td>
<td>247 (-423, 919)</td>
</tr>
</tbody>
</table>

† Adjusted for age, gender, education level, race, and previous smoking status
Mean difference (95% CI) in e-cigarette use by demographics

<table>
<thead>
<tr>
<th>Age (per year)</th>
<th>N</th>
<th>Voltage (volts)</th>
<th>N</th>
<th>Nicotine Use (mg/ml)</th>
<th>N</th>
<th>E-liquid/wk (ml)</th>
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<td>98</td>
<td>-0.46</td>
<td>50</td>
<td>-25.1</td>
</tr>
<tr>
<td>0.00 (0.03, 0.02)</td>
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<td></td>
<td>0.12 (0.36)</td>
<td></td>
<td>0.55 (0.63)</td>
<td></td>
<td>(-49.9, -0.25)</td>
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</table>

<table>
<thead>
<tr>
<th>Gender</th>
<th>N</th>
<th>Voltage (volts)</th>
<th>N</th>
<th>Nicotine Use (mg/ml)</th>
<th>N</th>
<th>E-liquid/wk (ml)</th>
<th>N</th>
<th>Puff count/day</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male</td>
<td>65</td>
<td>0.00 (ref)</td>
<td>66</td>
<td>0.00 (ref)</td>
<td>67</td>
<td>0.00 (ref)</td>
<td>35</td>
<td>0.00 (ref)</td>
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<tr>
<td>0.00 (0.03, 0.03)</td>
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<td></td>
<td>0.20 (0.03)</td>
<td></td>
<td>-22.7 (15, 0.03)</td>
<td></td>
<td>(-49.9, -0.25)</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Education</th>
<th>N</th>
<th>Voltage (volts)</th>
<th>N</th>
<th>Nicotine Use (mg/ml)</th>
<th>N</th>
<th>E-liquid/wk (ml)</th>
<th>N</th>
<th>Puff count/day</th>
</tr>
</thead>
<tbody>
<tr>
<td>≤ HS</td>
<td>38</td>
<td>0.00 (ref)</td>
<td>41</td>
<td>0.00 (ref)</td>
<td>41</td>
<td>0.00 (ref)</td>
<td>19</td>
<td>0.00 (ref)</td>
</tr>
<tr>
<td>-0.60 (0.36)</td>
<td></td>
<td></td>
<td>-0.02 (0.24)</td>
<td></td>
<td>-20.4 (31, 134)</td>
<td></td>
<td>(-301, 570)</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Previous smoker</th>
<th>N</th>
<th>Voltage (volts)</th>
<th>N</th>
<th>Nicotine Use (mg/ml)</th>
<th>N</th>
<th>E-liquid/wk (ml)</th>
<th>N</th>
<th>Puff count/day</th>
</tr>
</thead>
<tbody>
<tr>
<td>No</td>
<td>10</td>
<td>0.00 (ref)</td>
<td>11</td>
<td>0.00 (ref)</td>
<td>11</td>
<td>0.00 (ref)</td>
<td>6</td>
<td>0.00 (ref)</td>
</tr>
<tr>
<td>-1.31 (0.53)</td>
<td></td>
<td></td>
<td>-1.09 (0.19)</td>
<td></td>
<td>-3.10 (4, 10)</td>
<td></td>
<td>(-423, 919)</td>
<td></td>
</tr>
</tbody>
</table>

† Adjusted for age, gender, education level, race, and previous smoking status.

Self-reported health status

When using e-cigarettes, 27% reported

- Sore throat
- Runny nose
- Phlegm
- Coughing in AM + PM

Compared to non-users, e-cigarette users more likely to report symptoms of

- Wheezing and whistling in the chest (15% vs. 2%)
- Having hypertension (22% vs. 4%)
Summary Findings

Daily users: predominantly male, white, former smokers, owned 2 devices (MODs), vaped an average of 365 puffs/day, all throughout the day

*More intense use behaviors and device characteristics are seen in participants with certain demographic characteristics*

- Men
- Individuals of lower education

Outline

- Introduction: Electronic cigarettes
  - How they work
  - Increase in teen use
- Why are we concerned?
  - What we know so far about composition
  - Focus on metals
- Our Findings
  - Daily E-cig user demographics, behaviors, device type
    - **Metals in biospecimen of e-cig users**
      - Higher in e-cig users vs. non-users?
      - Specific use behaviors?
    - Metals in aerosol samples
- Risk Balance
- Future Directions
Sample Collection

- Collect aerosol and liquid from bottles and tanks of all participants
- Measure metal concentrations in e-liquid, aerosol, tank of MODS

Dispenser  Aerosol  Tank

• 30 to 50 puffs
• 1 L/min,
• 4 s per puff
• 30 s inter-puff time

Environmental Research 2016; 149:151-156

Sample Collection

- Collect biospecimens from all participants
- Measure metal biospecimens between users and non-users

Urine  Saliva  Exhaled Breath (EBC)

Statistical Analysis

Linear regression models on log-transformed metal levels to estimate Geometric mean ratios (GMR) to compare

1. E-cigarette users vs. non-users
2. E-cigarette use behaviors with biospecimens
3. Metal biospecimens with corresponding metals in e-liquid, aerosol, tank
### E-cigarette users vs. Non-users

<table>
<thead>
<tr>
<th>Metal</th>
<th>GMR (95%CI) Urine</th>
<th>GMR (95%CI) Saliva</th>
<th>GMR (95%CI) EBC</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>CHROMIUM</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-user</td>
<td>1.00 (ref)</td>
<td>1.00 (ref)</td>
<td>1.00 (ref)</td>
</tr>
<tr>
<td>E-cigarette user</td>
<td>1.96 (0.92, 4.17)</td>
<td>1.17 (0.55, 2.49)</td>
<td>1.06 (0.61, 1.86)</td>
</tr>
<tr>
<td>p-trend</td>
<td>0.08</td>
<td>0.673</td>
<td>0.831</td>
</tr>
<tr>
<td><strong>NICKEL</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-user</td>
<td>1.00 (ref)</td>
<td>1.00 (ref)</td>
<td>1.00 (ref)</td>
</tr>
<tr>
<td>E-cigarette user</td>
<td>1.30 (0.74, 2.29)</td>
<td>2.40 (0.86, 6.64)</td>
<td>3.12 (1.53, 6.35)</td>
</tr>
<tr>
<td>p-trend</td>
<td>0.362</td>
<td>0.092</td>
<td>0.092</td>
</tr>
<tr>
<td><strong>LEAD</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Non-user</td>
<td>1.00 (ref)</td>
<td>1.00 (ref)</td>
<td>1.00 (ref)</td>
</tr>
<tr>
<td>E-cigarette user</td>
<td>3.00 (1.66, 5.41)</td>
<td>1.62 (0.62, 4.24)</td>
<td>0.75 (0.28, 1.97)</td>
</tr>
<tr>
<td>p-trend</td>
<td>&lt;0.001</td>
<td>0.322</td>
<td>0.554</td>
</tr>
<tr>
<td><strong>MANGANESE</strong></td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Non-user</td>
<td>1.00 (ref)</td>
<td>1.00 (ref)</td>
<td>1.00 (ref)</td>
</tr>
<tr>
<td>E-cigarette user</td>
<td>1.19 (0.69, 2.06)</td>
<td>1.85 (0.34, 10.3)</td>
<td>2.29 (1.24, 4.21)</td>
</tr>
<tr>
<td>p-trend</td>
<td>0.533</td>
<td>0.477</td>
<td>0.008</td>
</tr>
</tbody>
</table>

Full adjustment: Adjusted for age, sex, race, education, smoking status, and other sources of metal exposure

### Metal biospecimen by use behaviors

- **More e-liquid consumed/week (35-240 ml vs. <35 ml)**
  - 99% ↑ Cr and 247% Mn saliva levels

- **More frequent coil change (>2/month vs. ≤ 2/month)**
  - 84% ↑ Cr and 132% Ni saliva levels

- **Shorter time to first vape when waking (≤15 minutes)**
  - 67% ↑ Ni urine levels

- **Using Kanthal vs. titanium, stainless steel or nichrome coils**
  - 30-60% higher Cr, Ni, Mn saliva and EBC levels
Summary findings

1. Metal exposure among e-cigarette users is higher than that of non-users and non-smokers
   - Cr and Pb in urine
   - Ni and Mn in EBC

2. Certain e-cigarette device characteristics and use patterns determine levels of metal exposure
   - More e-liquid consumed/wk
   - Frequent coil change
   - Shorter time to first vape in AM
   - Coil (Kanthal)
   - Aerosol

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  - Metals in biomarkers of e-cig users
  - Metals in aerosol samples
    - Source of metal exposure during use?
    - Differ by device generation?
• Risk Balance
• Future Directions
Objectives

- Measure metal concentrations in e-liquid from dispenser, aerosol, tank of MOD e-cigarette devices

Sample Collection: MODs
Metals in e-liquid and aerosol

- Cr, Ni, Pb, other toxic metals present in pre-vaped liquid
- Found at higher concentrations after the liquid was heated in the aerosol generated
- Even higher concentrations in the liquid that remained in the tank of the device, in contact with the coil

Objectives

- Measure metal concentrations in e-liquid from dispenser, aerosol, tank of MOD e-cigarette devices
- Compare metal concentrations by device type
- Compare metal concentrations by brand and flavor (in PODs)
Sample collection: PODs

By brand (tobacco)

JUUL (n=18)
BO (n=6)
PHIX (n=6)
SUORIN (n=6)

By flavor

Virginia Tobacco (n=6)
Mango (n=6)
Mint (n=6)

N = 36 aerosol samples

Metal concentration by sample type (PODS)
Aerosol metal concentrations by JUUL flavor

Aerosol concentrations: MODs vs. PODs
Metal concentrations (mg/m$^3$) in collected aerosol samples

Health-based limits: 
- ATSDR MRL
- EPA NAAQS

$C_a = \text{mf(ug/kg)} \times \frac{m_{\text{tot}}}{V_{\text{air}}}$

$C_a$ = air concentration; mf = mass fraction; $m_{\text{tot}}$ = total mass collected; $V_{\text{air}}$ = volume of air

Metal biospecimen by Metals in E-liquid

Higher Ni urine and saliva levels were associated with higher aerosol Ni concentrations

Higher Cr saliva levels were associated with higher aerosol and tank Cr concentrations

No association between metal biospecimen and metals in e-liquid from the dispenser

Environmental Research, 2017 159: 313-320
Summary

- **MODs**: higher concentrations in the aerosol and tank samples versus the dispenser
  - Coil contact → e-liquid contamination

- **PODs**: higher metal concentrations in the POD liquid before heating vs. the aerosol
  - POD: Ni, Mn, Pb, Co, Al, Cu, Zn
  - Aerosol: Co

- **POD by brand**: showed high variability
  - PHIX: Co, Ni, Pb, As, Sn
  - Bo: Zn, Cu, Sb
  - Suorin: Cr, Mn, Fe
  - JUUL: Co

Very high concentrations of Cu, Pb, and Zn in both BO and PHIX

Summary

- **JUUL Flavors**:
  - higher metal conc. of As, Cu, Ni, Zn, in Tobacco compared to Mint and Mango
  - Mango → higher Co, similar Cr and Fe

- **Aerosol MODs vs. PODs**:
  - similar aerosol concentrations (high variability)
  - MOD slightly higher Fe, Pb, and Sb
  - PODs higher Co, Ni, and Zn
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  - Metals in aerosol samples
- **Risk Balance**
  - Future directions

Difficult balance between the potential pros and cons of e-cigarettes.
The Public Health Perspective

Pros

- Health gain for smokers who quit via EC
- Health gain for smokers who substantially reduce smoking by use of EC
- Health gain for reluctant smokers who switch to EC

Cons

- Re-normalization of smoking in society
- Increased risk for smokers who switch to EC instead of stopping smoking
- Increased risk for smokers who become dual users
- Increased risk for ex-smokers who start using EC
- Increased risk for never smokers using EC

Source: Dr. Jonathan Samet, MD
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Future directions

1. Biospecimen analysis with POD users

2. Exposure assessment with disposable PODs
Future directions

3. Pulmonary health measures in assoc. with e-cig use

4. Link to COVID?

“Several factors that led to the progression of COVID-19 pneumonia were identified, including age, history of smoking, maximum body temperature on admission, respiratory failure, albumin, C-reactive protein.”

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EVALI outbreak

E-cigarette, or Vaping, product–use Associated Lung Injury

**CDC Report as of January 7, 2020**

- **2,602** hospitalized cases of EVALI or deaths from all 50 states, DC, PR, VI
  - Increase of 41 cases since December 27
- A total of **57** EVALI deaths
- CDC completed analysis of 71 bronchoalveolar lavage samples from 54 EVALI patients.
  - Almost all are positive for vitamin E acetate.

→ **little is known about long-term effects of e-cigarette use**
Cessation

• E-cigarette use patterns are highly variable over a one-year period

• PATH - Dual users from Wave 1 (2013/14)
  – 44.3% maintained dual use in Wave 2 (2014/15)
  – 43.5% quit vaping, still smoking
  – 12.1% quit smoking
    • 5.1% quit smoking, still vaping
    • 7% quit both


FAQ: how do e-cigs compare to cigarettes? Do they reduce risk?

For young vapers that have never smoked (and probably never would have) a better comparison is ambient air

  – For adolescents, vaping represents an increase in risk
  – Recent study found increase risk of becoming smokers ¹

JUUL is marketing as delivering the same nicotine dose as cigarettes

  – Extensive research has shown the adverse effects of nicotine on developing brains, and nicotine exposure during adolescence is likely to adversely affect cognitive function and development.

We only evaluated metals of e-cigs, not cigarettes.

  – Extensive literature on metals from cigarette smoke

It is hard to directly compare e-cigarettes to combustible cigarettes because cigarette emissions are typically reported “per cigarette”.

  – 15 e-cig puffs ➔ equivalent to one cigarette
  – Emission rates were similar between published cigarette conc. and e-cigarettes for chromium, nickel, zinc, (all toxic to the lungs) and lead.
  – We found lower concentrations in e-cigarettes for cadmium and arsenic.

¹ Dunbar et al., N&TR Oct 2 2018; https://doi.org/10.1093/ntr/nty179
Aerosol vs. Cigarette smoke

• Mainstream smoke of conventional cigarettes (Pappas et al. 2014)
  - Cd (<5.0 - 80 ng/cigarette)
  - Pb (<5.0 – 23 ng/cigarette)
  - Other metals (As, Co, Cr, Mn, Ni)
    - Undetectable (Ni, Cr)

• Closed system devices (Blu *cig-a-likes*)
  - Cu levels were 6.1 times higher (Lerner et al. 2015)

• Open system devices
  - Higher Cr and Ni levels in e-cigarettes
  - Similar Pb and Zn levels (Olmedo et al. 2016)

Key Points

• Introduction: Electronic cigarettes
  • How they work
  • Increase in teen use
• Why are we concerned?
  • What we know so far about composition
  • Focus on metals