EXPOSURE TO TOXIC VOLATILE ORGANIC CHEMICALS FROM E-CIGARETTE USE IN ADOLESCENTS

Mark L. Rubinstein, MD
Kevin Delucchi, Ph.D.
Neal L. Benowitz, M.D.
Daniel Ramo, PhD
Disclosures

• Funding: NIH/NIDA R21DA040718 & TRDRP P0504871
Background

• E-cigarettes are marketed to promote cessation or harm reduction in adults.
Background

- Marketing strategies clearly have a broader population in mind.
Background

• Marketing strategies clearly have a broader population in mind.
Background

• Marketing strategies clearly have a broader population in mind.
Background

• Social influences and marketing strategies for these products have clearly had an effect on children.
Reasons for increased use

- Peer influence
- Enticing flavors
- Extensive marketing of e-cigarettes as safer.

E-Cigarette Liquid Ingredients

VS

Traditional Cigarette Ingredients

- Propylene Glycol
- Vegetable Glycerin
- Nicotine Concentrate
- Artificial Flavoring
- 4000+ Known Carcinogens
- Cyanhydric Acid
- Dibenzacridine
- Vinyl Chloride
- Nicotine
- Cadmium
- Urethane
- Acetone
- Ammoniac
- Toluene
- Polonium 210
- DDT
- Naphthalene
- Pyrene
- Methanol
- Carbon Monoxide

- Propylene Glycol has undergone extensive testing and is widely used in a variety of consumer products, including food. It is also approved by US FDA for a variety of pharmaceutical formulations.
- Vegetable Glycerin is a plant-based carbohydrate. Its safety is pretty much unchallenged and studies demonstrate that it is non-carcinogenic. Likewise, both PG and VG are widely used in many industries, including pharmaceutical, cosmetic and food.
- Tobacco smoke contains 4000+ chemicals and compounds - at least 69 of these are cancer-causing.
Safety

• Despite claims, there is uncertainty about the safety of e-cigarettes.
• By aerosolizing nicotine rather than combusting tobacco, e-cigarettes do produce fewer toxins.
e-cigarettes contain additives and solvents, which can form carcinogenic compounds when heated.
Volatile Organic Compounds (VOC)

- When heated to vaporize:
  - propylene glycol can form the VOC propylene oxide (a carcinogen).¹
  - glycerol forms acrolein,² which has been associated with lung cancer.³

Toxicants in adults

• There is some controversy on how usage patterns may affect exposure

• Data from adults show that these toxicants can be detected in the urine of ENDS users.*

Adolescent Use

• Currently no data on toxicant exposure in adolescent users.
Adolescence onset is riskier

• Exposure to toxicants during adolescence may result in greater harm given their cumulative exposure compared to adult onset users.
Goals

• Using a cross-sectional design, we sought to assess the presence of certain VOC toxicants linked to adolescent e-cigarette use and examine how specific patterns of use, may influence exposure.
METHODS
Participants

- Adolescent (aged 13-18) e-cigarette users (use in past 30 days and least 10 lifetime use episodes) were recruited from the San Francisco Bay area using fliers and online advertising.
- E-cigarette users who only use their devices to vaporize marijuana were excluded.
Baseline Visit

- Baseline appointments were scheduled within 24 hours from last use of their e-cigarettes.
- Surveys were completed including questions about use behaviors.
- Urine was collected for the measurement of the tobacco-specific nitrosamine NNAL and levels of metabolites of 8 volatile organic compounds (VOCs).
- Participants received $30.
Controls

• Specimens were also collected from 20 age-matched controls attending pediatric clinics at a Bay area public hospital with undetectable cotinine and NNAL confirming no e-cigarette or nicotine use.
Categories of Participants

- Three categories were developed based on a combination of reported e-cigarette and cigarette use and urine NNAL.

1) E-cigarette-Only users
   - no traditional combustion cigarettes in past 30 days and NNAL <1 pg/mg creatinine

2) Dual Users
   - use traditional cigarettes in the past 30 days in addition to e-cigarettes and NNAL > 30 pg/mg creatinine

3) Controls
   - NNAL and cotinine below the limit of quantitation
**Exclusion from Analyses**

- We excluded from analyses participants who did not use e-cigarettes in the prior 24 hours.
- To create well-differentiated comparison groups, we also set an a priori exclusion from analyses participants with intermediate levels of NNAL (i.e., 1-29 pg/ml creatinine)
Measures to assess ENDS exposure

• Frequency of use:
  • days of use in past 30
  • number of sessions on each day in past week

• Flavor preferences

• Type of vape instrument (i.e. Mod, Juul, vape pen, or other)

• Nicotine content (if known)
Volatile Organic Compounds (VOCs)

• VOCs were measured in urine and expressed per mg of creatinine.
  1. Propylene oxide
  2. Acrolein
  3. Ethylene oxide
  4. Crotonaldehyde
  5. Acrylonitrile
  6. Benzene
  7. Acrylamide
  8. 1,3, butadiene
RESULTS
Participants

- 386 adolescents were screened, 229 were found to be eligible and 180 agreed to participate.
- 29 admitted to not using an e-cigarette in the prior 24 hours and were excluded from analyses.
- 48 had levels of NNAL 1-29 ppm/mg creatinine, and per our a priori criteria, were excluded from analyses.
- The final sample consisted of 67 E-cigarette-Only users, 16 Dual Users, and 20 Controls.
## Demographics

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>E-Cigarette-Only (n=67)</th>
<th>Dual Users (n=16)</th>
<th>Controls (n=20)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age (years)(^1)</td>
<td>16.3 (1.2)</td>
<td>17.1 (.96)</td>
<td>16.0 (1.8)</td>
<td>.06</td>
</tr>
<tr>
<td>% Male(^2)</td>
<td>49 (73%)</td>
<td>12 (80%)</td>
<td>7 (35%)</td>
<td>&lt;.01</td>
</tr>
</tbody>
</table>

1. Comparing 3 groups on age (Anova)
2. comparing 3 groups on sex (chi-square)
# Demographics

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>E-Cigarette-Only (n=67)</th>
<th>Dual Users (n=16)</th>
<th>Controls (n=20)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Race/Ethnicity¹</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>White</td>
<td>36 (54%)</td>
<td>9 (67%)</td>
<td>0</td>
<td>.06</td>
</tr>
<tr>
<td>Asian/PI</td>
<td>12 (19%)</td>
<td>2 (12%)</td>
<td>2 (10%)</td>
<td></td>
</tr>
<tr>
<td>Mixed race</td>
<td>10 (15%)</td>
<td>3 (19%)</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Hispanic</td>
<td>7 (10%)</td>
<td>2 (12%)</td>
<td>18 (90%)</td>
<td></td>
</tr>
</tbody>
</table>

1. comparing 3 groups on sex (chi-square)
## Flavors

<table>
<thead>
<tr>
<th>E-cig characteristic</th>
<th>E-Cigarette-Only (n=67)</th>
<th>Dual Users (n=16)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Usual flavor</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>fruit</td>
<td>37 (55%)</td>
<td>10 (67%)</td>
<td>.42</td>
</tr>
<tr>
<td>candy</td>
<td>11 (16%)</td>
<td>2 (13%)</td>
<td>.77</td>
</tr>
<tr>
<td>menthol</td>
<td>12 (18%)</td>
<td>2 (13%)</td>
<td>.67</td>
</tr>
<tr>
<td>tobacco</td>
<td>5 (8%)</td>
<td>2 (13%)</td>
<td>.46</td>
</tr>
</tbody>
</table>
## Frequency of use

<table>
<thead>
<tr>
<th>E-cig characteristic</th>
<th>E-Cigarette-Only (n=67)</th>
<th>Dual Users (n=16)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hours from last use</td>
<td>1:58 (6:29)</td>
<td>2:02 (7:17)</td>
<td>.91</td>
</tr>
<tr>
<td>Days used in past 30</td>
<td>12.8 (8.9)</td>
<td>25.5 (6.6)</td>
<td>&lt;.001</td>
</tr>
<tr>
<td>Sessions per day</td>
<td>2.0 (3.6)</td>
<td>8.4 (11.6)</td>
<td>&lt;.05</td>
</tr>
</tbody>
</table>
## Type of Device

<table>
<thead>
<tr>
<th>E-cig characteristic</th>
<th>E-Cigarette-Only (n=67)</th>
<th>Dual Users (n=16)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Usual Device</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Vape pen</td>
<td>24 (36%)</td>
<td>6 (40%)</td>
<td></td>
</tr>
<tr>
<td>Mod</td>
<td>17 (25%)</td>
<td>4 (27%)</td>
<td>.82</td>
</tr>
<tr>
<td>Juul</td>
<td>18 (27%)</td>
<td>4 (27%)</td>
<td></td>
</tr>
<tr>
<td>Other/unsure</td>
<td>8 (12%)</td>
<td>1 (7%)</td>
<td></td>
</tr>
</tbody>
</table>
## Nicotine

<table>
<thead>
<tr>
<th>E-cig characteristic</th>
<th>E-Cigarette-Only (n=67)</th>
<th>Dual Users (n=16)</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>E-cigarettes contain nicotine</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Always</td>
<td>21 (31%)</td>
<td>9 (60%)</td>
<td></td>
</tr>
<tr>
<td>Sometimes</td>
<td>26 (39%)</td>
<td>6 (40%)</td>
<td>.06</td>
</tr>
<tr>
<td>Unsure</td>
<td>10 (15%)</td>
<td>0 (0%)</td>
<td></td>
</tr>
<tr>
<td>Never</td>
<td>10 (15%)</td>
<td>0 (0%)</td>
<td></td>
</tr>
</tbody>
</table>
# Toxicants (Median ng/mg of creatinine)

<table>
<thead>
<tr>
<th>Toxin</th>
<th>Biomarker</th>
<th>control</th>
<th>e-cig only</th>
<th>Dual users</th>
</tr>
</thead>
<tbody>
<tr>
<td>Benzene</td>
<td>PMA</td>
<td>0</td>
<td>0</td>
<td>.2**</td>
</tr>
<tr>
<td>1,3-Butadiene</td>
<td>MHBMA</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Ethylene Oxide</td>
<td>HEMA</td>
<td>1.3</td>
<td>.5</td>
<td>1.0</td>
</tr>
<tr>
<td>Acrylonitrile</td>
<td>CNEMA</td>
<td>0**</td>
<td>1.3</td>
<td>59.4**</td>
</tr>
<tr>
<td>Acrolein</td>
<td>3-HPMA</td>
<td>192.8*</td>
<td>254.3</td>
<td>439.7*</td>
</tr>
<tr>
<td>Propylene Oxide</td>
<td>2-HPMA</td>
<td>15.2**</td>
<td>28.8</td>
<td>40.2</td>
</tr>
<tr>
<td>Acrylamide</td>
<td>AAMA</td>
<td>34.5**</td>
<td>67.3</td>
<td>235.6**</td>
</tr>
<tr>
<td>Crotonaldehyde</td>
<td>HPMMA</td>
<td>100.4*</td>
<td>148.7</td>
<td>185.4</td>
</tr>
</tbody>
</table>

*P<.05  **p<.001 e-cig-only as comparison group using regression models of log-transformed values including planned covariates (sex, race/ethnicity)
Significant VOC exposure in E-cigarette Only vs. Controls and E-cigarette Only vs. Dual Users.

Tests based on regression models of shifted log-transformed values including planned covariates (sex, race/ethnicity) with contrasts between E-Cigarette-Only users versus Controls and E-Cigarette-Only users versus Dual Users.

Note: All comparisons are made with E-Cigarette-Only (eCig-Only) Users as comparison group.

* $p<.05$
** $p<.001$
Acrylamide

AAMA log

Group

Controls  eCig-Only Users  Dual Users

* p<.05  ** p<.001
Propylene Oxide

** p<.001
Acrylonitrile

**p < .001**
* $p<.05$
ASSOCIATIONS BETWEEN VOCS AND E-CIGARETTE USE AMONG E-CIGARETTE ONLY USERS
Frequency

• The average number of sessions of e-cigarette use per day was associated with increased levels of:

• **CNEMA** *(Acrylonitrile*; *r* = .36, *p* = .003).

*Class 2B carcinogen*
Nicotine

• Compared to those who “never” used nicotine in their e-cigarettes, participants using nicotine “all” or “some” of the time had significantly higher median levels of:
  • **CNEMA** *(Acrylonitrile; 1.50 vs. .88 ng/ml creatinine, \( p = .05 \))
  • **AAMA** *(Acrylamide*; 71.5 vs. 60.4 ng/ml creatinine, \( p = .05 \)).

*Studies in rodent models have found that acrylamide exposure increases the risk for several types of cancer.*
Product type

• There were no differences in levels of the five significant VOCs based on type of product used.
  • F’s ranged from .51 to 2.3; $p$ values ranged from .09 to .67.
Flavors

• Participants who used fruit flavors had higher CNEMA (Acrylonitrile) levels.
  • 10.4 ng/ml vs. 2.1 ng/ml, $p=0.03$

• There were no differences in VOC levels among those who used other flavors.
Summary

• Adolescent **E-Cigarette-Only users** had levels of five VOC toxicants detected in quantities up to 3 times greater than in **controls**.

• **Dual Users** had levels of VOCs up to 3 times higher than E-Cigarette-Only users.
Conclusions

• While e-cigarette vapor may be less dangerous than combustion cigarettes, with lower overall exposure to VOC toxicants, our findings challenge the idea that e-cigarette vapor is safe.

• Many of the VOCs we identified among e-cigarette users are carcinogenic and were present whether or not the product contained nicotine.

• As with traditional cigarettes, messaging to teens must include warnings about the potential risk from toxic exposure to carcinogenic compounds generated by these products.
THANKS
Marijuana

• In all models, group membership (e-Cig Only, Dual User, Control) remained a statically significant predictor of VOC value independent of marijuana use frequency.
## Toxicants (Median ng/mg of creatinine)

<table>
<thead>
<tr>
<th>Toxin</th>
<th>control</th>
<th>e-cig only</th>
<th>Dual users</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acrylonitrile</td>
<td>0**</td>
<td>1.3</td>
<td>59.4**</td>
</tr>
<tr>
<td>Acrolein</td>
<td>192.8*</td>
<td>254.3</td>
<td>439.7*</td>
</tr>
<tr>
<td>Propylene Oxide</td>
<td>15.2**</td>
<td>28.8</td>
<td>40.2</td>
</tr>
<tr>
<td>Acrylamide</td>
<td>34.5**</td>
<td>67.3</td>
<td>235.6**</td>
</tr>
<tr>
<td>Crotonaldehyde</td>
<td>100.4*</td>
<td>148.7</td>
<td>185.4</td>
</tr>
</tbody>
</table>

*P<.05  ** p<.001 e-cig-only as comparison group using regression models of log-transformed values including planned covariates (sex, race/ethnicity)