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### Title

Nicotine levels in silicone wristband samplers worn by children exposed to secondhand smoke and electronic cigarette vapor are highly correlated with child's urinary cotinine.

### Permalink

<https://escholarship.org/uc/item/1pr071s2>

### Journal

Journal of exposure science & environmental epidemiology, 29(6)

### ISSN

1559-0631

### Authors

Quintana, Penelope JE  
Hoh, Eunha  
Dodder, Nathan G  
[et al.](#)

### Publication Date

2019-10-01

### DOI

10.1038/s41370-019-0116-7

Peer reviewed



file sent:

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Journal:	<i>Journal Of Exposure Science And Environmental Epidemiology</i>
Manuscript ID	JESEE-18-2883.R1
Manuscript Type:	Original Article
Date Submitted by the Author:	n/a
Complete List of Authors:	Quintana, Penelope; San Diego State University, School of Public Health Hoh, Eunha; San Diego State University, School of Public Health Dodder, Nathan; San Diego State University Research Foundation Matt, Georg; San Diego State University, Department of Psychology Zakarian, Joy; San Diego State University Research Foundation Anderson, Kim; Oregon State University College of Agricultural Sciences, Environmental and Molecular Toxicology Akins, Brittany; San Diego State University, School of Public Health Chu, Linda; San Diego State University, School of Public Health Hovell, Melbourne; San Diego State University, School of Public Health
Keywords:	child exposure/health, personal exposure, dermal exposure, biomonitoring

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5 Title:

6 Nicotine levels in silicone wristband samplers worn by children exposed to secondhand smoke  
7 and electronic cigarette vapor are highly correlated with child's urinary cotinine  
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10  
11 Authors

12 Quintana, Penelope JE, PhD, MPH\*

13 Hoh, Eunha, PhD

14 Dodder, Nathan G., PhD

15 Matt, Georg E., PhD

16 Zakarian, Joy M., MPH

17 Anderson, Kim A., PhD

18 Akins, Brittany, BS

19 Chu, Linda, BS

20 Hovell, Melbourne F., PhD, MPH

21  
22  
23  
24  
25  
26  
27 *\*corresponding author*  
28  
2930 **Corresponding author:**

31 Penelope JE (Jenny) Quintana, PhD, MPH

32 Professor and Graduate Advisor for MPH and MS

33 School of Public Health

34 San Diego State University

35 5500 Campanile Dr.

36 San Diego, CA 92182-4162

37 619-594-1688/ fax 619-594-6112

38 jquintan@sdsu.edu  
39  
40  
41  
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**Abstract**

Exposure assessment in children, especially young children, presents difficulties not found with adults. Simple silicone wristbands are passive samplers that have potential applicability in exposure studies of children. We investigated the performance of silicone wristbands as personal nicotine samplers in two wristbands worn by a child ( $n = 31$ ) for 7 days and for 2 days (worn day 5 to day 7). We compared levels of nicotine in wristbands with urinary cotinine, a metabolite of nicotine, measured in the child's urine obtained on day 7. Children were recruited who were exposed to contaminants in tobacco smoke and/or vapor from electronic nicotine delivery systems (ENDS; commonly known as electronic cigarettes or EC) as well as children who lived in nonsmoking homes. Caregivers were interviewed to obtain reported measures of the child's exposure. Analysis was by liquid chromatography with triple quadrupole mass spectrometry and isotope dilution (LC-MS/MS). The nicotine detected in the wristbands worn for 2 days was highly correlated with urinary cotinine concentration ( $df = 29$ ,  $r^2 = 0.741$ ,  $p < 0.001$ ), as was nicotine in wristbands worn for 7 days ( $df = 28$ ,  $r^2 = 0.804$ ,  $p < 0.001$ ). The 2-day and 7-day wristband nicotine amounts were also significantly correlated ( $df = 28$ ,  $r^2 = 0.852$ ,  $p < 0.001$ ). Silicone wristbands may be a useful tool for epidemiological and intervention studies of tobacco product exposure in children.

## Introduction

Exposure assessment is a necessary and vital part of environmental epidemiology studies, intervention studies and studies of changes in exposure over time. Exposure assessment in children, especially young children, presents difficulties not found in adult participants. Biological monitoring, while integrating exposure from all routes, presents challenges. Blood samples are difficult to collect unless a benefit to the child is perceived, such as a blood lead test. Urine samples, simple to collect in adults, are more difficult or potentially embarrassing for a child. Active air sampling is considered highly reliable and accurate for airborne exposures, but air samplers and pumps may be too heavy or restrictive for a young child. A new wave of simpler passive samplers that can be deployed with children include the silicone wristband (1), which is expected to have good acceptability, as these wristbands are already worn by many children by choice.

The silicone wristband sampler is a commercially available low-cost silicone wristband, which can be of different sizes, and which must be carefully cleaned and prepared for use as a passive sampler (2). O'Connell et al. (1) demonstrated that silicone wristband samplers were able to detect an individual's exposure to a wide range of compounds, including polycyclic aromatic hydrocarbons (PAHs), pesticides, phthalates, industrial compounds, and other consumer products. In Peru, wristbands worn for 30+ days by occupationally pesticide-exposed and non-occupationally exposed community members detected a wide range of compounds, including PAHs and pesticides (3). Silicone wristbands have been used to estimate PAH exposures near fracking sites, with a higher level of the sum of PAHs found in participants' wristbands who lived near fracking sites compared to those living further away (4). In addition to the low cost, ease of use, and ability to detect a wide range of pollutants, the silicone wristband is especially applicable to global and consumer studies as it is lightweight and can be returned to the laboratory by mail

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3 at room temperature (5). Participants in O'Connell and colleague's study testing silicone  
4 wristbands as personal passive samplers reported no discomfort or work interference from the  
5 wristbands (1).  
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9 The silicone wristband has shown promise for personal exposure assessment when  
10 compared to personal exposures to PAHs assessed by more standard methods. PAHs exposure  
11 was assessed in pregnant women wearing silicone wristbands for 48 hours and levels in  
12 wristbands were compared to levels of PAHs in personal active air samples and PAH metabolites  
13 in urine (6). Correlations were more consistent between individual PAH levels detected in the  
14 wristbands and urinary metabolites than between the personal air samples and urinary  
15 metabolites (6). In children and adults, the wristbands were used to estimate exposure to flame  
16 retardants (7), and appeared to perform better than hand wipes (8). The silicone wristband was  
17 also used to estimate children's exposure to flame retardants in a study of behavioral effects (9).  
18 The sampler shows a significant correlation with urinary or serum concentrations in the same  
19 participant (8, 10). Nicotine was detected in a few wristbands in a study of roofers (1). Here we  
20 present data on the potential of the silicone wristband sampler to measure exposure to tobacco  
21 toxicants, specifically nicotine, in children.  
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37 Children are exposed to tobacco smoke pollution worldwide and this exposure is a leading  
38 cause of adverse health outcomes such as respiratory infections and asthma, among other  
39 problems. In the US, about 40% of children are exposed to secondhand smoke (SHS) (11).  
40 Secondhand smoke from cigarettes is the major source of exposure to tobacco toxicants but other  
41 sources of tobacco-related pollutant exposure such as hookah smoke and vapor from electronic  
42 nicotine delivery systems (ENDS, commonly known as electronic cigarettes, EC) are being  
43 increasingly recognized (12, 13).  
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51 Exposure to tobacco smoke pollutants can be assessed directly by examining  
52 physiological fluids (urine, saliva, and serum), hair, or nails for tobacco smoke constituents or  
53 their metabolites (14). Nicotine, cotinine (a metabolite of nicotine), thiocyanate,  
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3 carboxyhemoglobin, and protein or DNA adducts have been among those used as indicators (15).  
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5 The relationship between a biomarker and exposure is complex and varies as a function of  
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7 environmental and physiological factors. The degree of exposure is a function of the time an  
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9 individual spends in each setting and the concentration of tobacco-related constituents in that  
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11 environment. Factors that affect concentrations of tobacco smoke pollution near children include  
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13 the amount of tobacco products consumed, proximity to caregivers and visitors using the  
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15 products, as well as factors that are more common in low-income children such as a decreased  
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17 home volume, which increases exposure (16). Factors that affect a child's intake include size,  
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19 age and activity level (17). Cotinine, a metabolite of nicotine, is the most widely used biomarker  
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21 of secondhand smoke exposure (15). The average biological half-life of urinary cotinine is  
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23 approximately 15-19 hours in adult smokers and nonsmokers and up to 23-28 hours in newborns,  
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25 infants, and older children, making it a good indicator of nicotine exposure over the previous two  
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27 days (18, 19).  
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31 This study investigated whether nicotine measured in a silicone wristband worn by a child  
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33 on the wrist for a period of 7 days (wristband 1) and 2 days (wristband 2, worn in addition to  
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35 wristband 1 from day 5 to day 7, when both wristbands were removed) would correlate with  
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37 cotinine levels (ng/mL) in the child's urine obtained at the end of the wearing period. We  
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39 investigated 2 time periods for wearing the wristband (2 days and 7 days) in order to optimize the  
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41 time period required for wearing the sampler in future studies. Children were recruited who were  
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43 exposed to contaminants in conventional cigarettes (CC) and/or electronic cigarettes (EC), as  
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45 well as children who lived with nonsmoking caregivers. We hypothesized that nicotine levels (ng/  
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47 wristband) in silicone wristbands worn for 2 days would be significantly correlated with urine  
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49 cotinine levels (ng/ml) determined via a urine sample obtained from the same participant at the  
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51 end of the study period, and that due to the half-life of cotinine, the nicotine levels in the wristband  
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53 worn for 2 days would be more highly correlated with urine cotinine in the same individual than  
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55 would the nicotine levels in the wristband worn for 7 days.  
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## Materials and Methods:

### Study Overview

Permissions were obtained from the Human Research Protection Program (HRPP) at San Diego State University and informed consent and assent was obtained from participants. The research design of this pilot study was based on a quasi-experimental comparison of the levels of tobacco smoke and ENDS contaminants (nicotine) found in silicone wristbands worn for 7 days and 2 days compared to biomarkers of tobacco exposure (urinary cotinine) in children from several recruited groups as follows. Children ages 4–14 were recruited who lived with one or more adults who smoked CC or used EC indoors at home, or who lived with nonsmoking/non-EC users. Elementary to middle school aged children were chosen as they are more likely than older children to be with their caregivers. Following informed consent and assent, children wore wristbands for a period of 7 days and an additional wristband on the same wrist for the last two days. At the end of the 7-day period, caregivers were interviewed regarding the child's exposure to tobacco products and EC, and a urine sample was collected from the child. Wristbands were transported in a cooler with frozen ice packs and extracted and analyzed for nicotine, and urine was analyzed for cotinine using liquid chromatography with triple quadrupole mass spectrometry (LC-MS/MS). Study sampling was conducted from March to October, 2017.

### Recruitment

Participants were recruited from past participants in a home air quality study who had agreed to be re-contacted about future study opportunities (n = 27), referrals from other study participants (n = 2), and advertisements on Craigslist (n = 1) and Facebook (n = 1). Both the child and the adult had to be willing to participate. Children were all nonsmokers and nonusers of CC or EC. We recruited children who lived with at least one adult who smoked a minimum of 7 cigarettes/week inside the home (n = 12), children who lived with at least one adult who used EC at least 4 days/week inside the home and used e-liquids with nicotine (n = 9), and children not exposed to nicotine products, who lived with adult nonsmokers and nonusers of EC who had a



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3 complete ban on smoking and EC use inside their home (n = 10). Final classification of exposure  
4 categories shown in Table 2 was based on the exposure of the child to CC and EC during the 7-  
5 day study period.  
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### 9 Samples Collection

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11 All samples and interview data were collected by pairs of research assistants during two  
12 visits to participants' homes with the adult and the child participant. Each visit took approximately  
13 45 minutes. At the first visit, research assistants confirmed eligibility and obtained informed  
14 consent, deployed the silicone wristbands, and left a urine collection cup. Homes were visited  
15 again one week later to retrieve the wristbands and urine sample and to conduct an in-person  
16 interview about smoking and EC use and the child's exposure during the past week.  
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24 **Home Interview.** Face-to-face interviews were conducted by trained research assistants to  
25 collect the following data: the child's exposure to CC and EC over the sampling week: number of  
26 cigarettes smoked and proportion of EC liquid used when the child was in the same indoor room  
27 at home; number of days the child was exposed to CC and EC by visitors inside the home, in  
28 cars, and indoors away from home; personal background including child's age, gender,  
29 race/ethnicity, and family income; and home characteristics including numbers of rooms and  
30 residents.  
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39 **(2) Daily monitoring of CC/EC use and exposure.** Caregivers were asked to report their  
40 daily use of CC/EC and the child's exposure to CC/EC during brief (5 minute) daily telephone  
41 calls with a research assistant.  
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45 **(3) Urine samples.** Single spot urine samples were collected from child participants using  
46 procedures from our previous studies (20, 21), for analysis of cotinine concentration (22). Children  
47 were asked to urinate into a standard urine collection cup, with their caregivers' assistance if  
48 needed. Caregivers were asked to have the child collect their sample on the morning of the  
49 second home visit. Due to variations in participants' and caregiver schedules, however, samples  
50 from ten children were collected in the afternoon and evening as late as 7:00 pm.  
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3       **(4) Silicone wristbands.** Cleaned prepared wristbands in Teflon bags were obtained from K.  
4 Anderson, developer of this technology (1, 2). Wristbands were stored and transported to  
5 participants' homes in clear glass jars with polytetrafluoroethylene (PTFE)-faced PE-lined caps,  
6 or 2.0 mil thick Teflon PFA bags (2-day wristband).  
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11       Each child participant received two wristbands at the initial home visit. One was placed on the  
12 arm at the home visit, to be worn for one week. The actual wearing time was from 6.0 days to 8.7  
13 days, with a median of 7.0 days. The research assistants also collected a wristband field blank  
14 for each participant, by transporting a wristband to the home, opening its container and replacing  
15 it in the container, and transporting back without being worn. Research assistants reminded  
16 caregivers by text message or telephone call to open the protective Teflon PFA bag and place  
17 the second wristband on the child's arm on the morning of day five, to be worn for the last two  
18 days. The actual wearing time for this second wristband was from 1.2 days to 2.5 days, with a  
19 median of 2.4 days. Participants were instructed to wear the wristbands at all times, including  
20 when sleeping, bathing, or swimming. We did not ask the caregiver about the frequency of bathing  
21 or swimming. Most (26/31) children received a 'small' size wristband (average of 3 bands: length  
22 17.9 cm, weight 3.7 g), and 5/31 (2 from CC, 2 from EC and 2 from NS groups) received a 'large'  
23 size wristband (average of 3 bands: length 20.2 cm, weight 4.2 g).  
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39       For verification of wearing, caregivers texted a picture of the wristbands on the child's wrist  
40 once a day. Sample wristbands and field blanks were transported back to the lab in individual  
41 borosilicate amber glass vials, with Thermoset lids lined with F217/PTFE, and stored at -20°C  
42 until analysis for nicotine concentration.  
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#### 47 Laboratory Analysis

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49       **Materials.** All solvents were of LC/MS grade. Chemical standards of nicotine, nicotine-*d*<sub>4</sub>,  
50 cotinine, and cotinine-*d*<sub>3</sub> were purchased from MilliporeSigma.  
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54       **(1) Wristband Nicotine.** The method closely followed the QuEChERS extraction procedure  
55 (23, 24) previously modified for nicotine analysis on surface wipes and in dust (25-27). Personnel  
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3 performing extraction and analysis were blinded as to exposure status of participants. All sample  
4 containers and laboratory tools (scissors, tweezers, pipet tips, syringes, and syringe filters) were  
5 rinsed with methanol prior to use. Laboratory personnel wore disposable caps and laboratory  
6 coats when processing samples. Each silicone wristband was cut into small pieces, and all pieces  
7 were placed in a 50 mL centrifuge tube and spiked with 15 ng of the internal standard nicotine- $d_4$ .  
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9 Four mL of 0.1% formic acid in water was added, and the samples were vortexed for 1 minute.  
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11 One mL of 1M KOH was added, and the samples were vortexed for 1 minute. Three mL of  
12 acetonitrile was added, and the samples were vortexed for 5 minutes. The wristband pieces were  
13 removed, 2 g magnesium sulfate and 0.5 g sodium chloride were added, and the samples  
14 vortexed for 1 minute. Samples were centrifuged at 3000 rpm ( $900 \times g$ ) for 5 minutes. One mL of  
15 the organic (top) layer was passed through a PTFE syringe filter (13 mm diameter, 0.2  $\mu\text{m}$  pore  
16 size). The final concentration of nicotine- $d_4$  was 5 ng/mL.  
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28 Nicotine was quantified by LC-MS/MS (Agilent 1200 Series LC system coupled to an Agilent  
29 6460 Triple Quadrupole system) operated in positive electrospray ionization (ESI+) mode. The  
30 injection volume was 1  $\mu\text{L}$ . The chromatographic separation was performed using an Agilent  
31 ZORBAX Hilic Plus column (dimensions of 2.1 $\times$ 50 mm, particle size 1.8  $\mu\text{m}$ ), with a 0.3 mL/min  
32 isocratic solvent program of 35% aqueous phase (5 mM ammonium acetate and 0.1% formic acid  
33 in water) and 65% organic phase (0.1% formic acid in acetonitrile). The multiple-reaction-  
34 monitoring (MRM) transitions were: 163.1  $\rightarrow$  130.1 (quantitative), 163.1  $\rightarrow$  132.1 (qualitative),  
35 and 163.1  $\rightarrow$  117.1 (qualitative) for nicotine; and 167.3  $\rightarrow$  136.1 (quantitative), 167.3  $\rightarrow$  134.1  
36 (qualitative), and 167.3  $\rightarrow$  121.1 (qualitative) for nicotine- $d_4$ . The dwell time was 200 ms for each  
37 transition, and the fragmentor voltage was 35. Collision energies were 12, 20, and 28 for the  
38 transitions with the highest, medium, and lowest product ion  $m/z$  values, respectively. The  
39 calibration curve consisted of 9 standard solutions ranging from 0.1 to 500 ng/mL of nicotine and  
40 each with 5 ng/mL of nicotine- $d_4$ . As per the method, quantitation was made based on 3 mL as  
41 the total volume. An acetonitrile instrumental blank was run between every standard and sample  
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3 injection to ensure non-significant carry-over between injections, defined as an injection blank  
4 response < 0.5% of the response of the corresponding sample. Results were reported as ng  
5 nicotine /wristband. The limit of quantification (LOQ) was 0.30 ng/wristband, and the estimated  
6 method detection (MDL) limit was 0.19 ng/wristband. The LOQ was defined as the lowest  
7 concentration that could be consistently measured without bias (28), which we also set as the  
8 concentration of the standard solution with the lowest value in the calibration curve. The estimated  
9 MDL was defined as the concentration corresponding to a signal/noise ratio of 5 (29).

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18 **(2) Urinary Cotinine.** Cotinine was analyzed using a modified version of a previously  
19 developed LC-MS/MS method (25, 26). Personnel performing extraction and analysis were  
20 blinded as to exposure status of participants. Two mL of urine sample was added to a 15 mL  
21 centrifuge tube, spiked with 10 ng of internal standard cotinine- $d_3$ , and mixed. Two mL of  
22 acetonitrile was added and the samples were vortexed for 1 minute. One g of salt mixture, 0.8 g  
23 magnesium sulfate and 0.2 g sodium chloride were added and the samples vortexed for 1 minute,  
24 then centrifuged at 3000 rpm ( $900 \times g$ ) for 5 minutes. One mL of the organic (top) layer was  
25 removed and added to a 2 mL vial containing the dispersive solid-phase-extraction mixture  
26 (dSPE, Agilent, QuEChERS Dispersive SPE AOAC method, containing 50 mg primary-secondary  
27 amine, 50 mg  $C_{18}$ , and 50 mg  $MgSO_4$ ). Samples were vortexed for 1 minute, then centrifuged at  
28 10,000 rpm ( $5600 \times g$ ) for 1 minute. The liquid layer was removed and passed through the syringe  
29 filter. Solutions were evaporated to approximately 100  $\mu$ L and reconstituted in 1 mL of 95% water,  
30 5% acetonitrile. The final concentration of cotinine- $d_3$  was 5 ng/mL. One laboratory blank was  
31 prepared with every sample preparation batch ( $n = 10$  samples).

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47 Cotinine was quantified on the same LC-MS/MS instrumentation as nicotine and operated in  
48 positive electrospray ionization (ESI+) mode. The injection volume was 15  $\mu$ L. The  
49 chromatographic separation was performed using an Agilent Poroshell 120 SB- $C_{18}$  column  
50 (dimensions of 2.1 $\times$ 50 mm, particle size 2.7  $\mu$ m), with a 0.4 mL/min gradient solvent program  
51 consisting of aqueous phase A (5 mM ammonium acetate) and organic phase B (acetonitrile).  
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3 The gradient conditions at each run time,  $t$ , were: 95% A, 5% B at  $t = 0$  min; 95% A, 5% B at  $t =$   
4 0.3 min; 70% A, 30% B at  $t = 4$  min; 70% A, 30% B at  $t = 4.5$  min; 95% A, 5% B at  $t = 5.5$  min;  
5 and run end at  $t = 12$  min. The multiple-reaction-monitoring (MRM) transitions were: 177.0  $\rightarrow$  98.0  
6 (quantitative) and 177.0  $\rightarrow$  80.0 (qualitative) for cotinine; and 180.0  $\rightarrow$  101.0 (quantitative) and  
7 180.0  $\rightarrow$  80.0 (qualitative) for cotinine- $d_3$ . Dwell time was 200 ms for each transition, fragmentor  
8 voltage was 90, and collision energy was 25. The calibration curve consisted of 7 matrix-matched  
9 standard solutions ranging from 0.1 to 25 ng/mL of cotinine and each with 5 ng/mL of cotinine- $d_3$ .  
10 The matrix-match was prepared by processing synthetic urine (Surine Negative Urine Control,  
11 MilliporeSigma) through the urinary cotinine extraction procedure. The LOQ was 0.10 ng/mL  
12 urine, and the estimated MDL was 0.033 ng/mL urine.  
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#### 24 Statistical Analysis

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26 All wristband samples had detectable values. Nicotine was present in 4 of the 31 field  
27 blank samples at concentrations of 7.8, 8.0, 9.4 and 9.5 ng/wristband or 0.5% to 1.8% of the levels  
28 in the respective exposure samples. All of the field blanks with detectable nicotine levels were  
29 from homes with active conventional cigarette smokers, a trend we have reported in previous  
30 surface sampling studies of second and thirdhand smoke contamination of homes (30) . We  
31 subtracted field blank nicotine levels before reporting final results. One cotinine sample had a  
32 non-detect value and was replaced with one half the MDL (0.05 ng/mL). Cotinine was not detected  
33 in the laboratory blanks. Nicotine and cotinine concentrations were  $\log_{10}(x+1)$  transformed before  
34 statistical analysis. Descriptive statistics were generated in SPSS v25. Differences between 2-  
35 day and 7-day nicotine concentrations were determined with the paired t-test on log-transformed  
36 values, and differences in correlations were determined using the z-test for the difference between  
37 two dependent correlations in STATA v15. Figures were made using the R (31) package ggplot2  
38 (32).  
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#### 53 **Results**

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3 All 31 children provided 2-day wristbands and 30 children provided 7-day wristbands, and  
4 a urine sample was obtained from each child. Three caregivers reported minor problems with  
5 wearing the wristbands: One reported that the child lost the 7-day wristband, and another reported  
6 that the 7-day wristband fell off when the child was sleeping and was in the bed covers away from  
7 the child for 420 minutes before it was found. One caregiver reported that the child's wristbands  
8 fell off when playing and were off for 185 minutes. Table 1 reports demographics and tobacco-  
9 related product exposure for each child. Approximately one half of participants reported exposure  
10 to tobacco-related products inside the home, defined as the child was present in the same indoor  
11 room when CC smoking or EC use occurred. Children's exposure category was based on  
12 caregivers' reports of exposure only during the week the wristband was worn, so the number of  
13 children in each exposure category (Table 1) varies from the number in each recruitment category  
14 (Methods). Nicotine was detected in all wristbands and cotinine was detected in all but one urine  
15 sample. Levels of nicotine in the two wristband and cotinine in urine from the same child ranged  
16 over several orders of magnitude (Table 2). A strong correlation was observed between exposure  
17 to nicotine as assessed by levels of urine cotinine and exposure to nicotine as assessed by  
18 nicotine detected in the wristband (Figure 1, log-log scale). The correlation between urinary  
19 cotinine and nicotine in the 2-day wristband was  $r^2 = 0.741$ ,  $p < 0.001$ , (left panel of Figure 1), and  
20 the correlation between urine cotinine and nicotine in the 7-day wristband was  $r^2 = 0.804$ ,  $p < 0.001$ ,  
21 (right panel, Figure 1). Spearman's correlations for untransformed data were  $\rho = 0.887$ ,  $p < 0.01$ ,  
22 and  $\rho = 0.921$ ,  $p < 0.01$ , respectively. The correlations between 7-day wristband nicotine and  
23 urine cotinine were slightly greater but did not significantly differ from that between the 2-day  
24 wristband nicotine and urine cotinine (2-tailed p-value = 0.2241). Nicotine levels in the 7-day  
25 wristbands were slightly but significantly higher than those in the 2-day wristbands ( $p < 0.01$ ,  
26 geometric mean 127.6 ng/wristband vs. 92.1 ng/wristband). We found a high correlation between  
27 levels of nicotine in the wristband worn for 2 days and the wristband worn for 7 days ( $r^2 = 0.852$ ,  
28  $p < 0.001$ , Figure 2). For untransformed data,  $\rho = 0.930$ ,  $p < 0.01$ .

## Discussion

Silicone wristbands show promise for exposure assessment of nicotine exposure in children, given the 100% nicotine detection rate and high dynamic range of nicotine in the wristbands, and the high correlation with urine cotinine, a validated marker of exposure to nicotine. This adds to the current literature on compounds for which the silicone wristbands have been assessed in relation to a level of a biomarker in the same person. Dixon et al. (6) found generally lower associations than we report here, between urinary levels of PAHs metabolites and wristband levels of the parent PAH compound in a sample of 22 pregnant women, though they reported a greater percent of non-detects in their samples, which could influence the strength of associations.

In our data, the silicone wristband worn for 2 days was highly correlated with that worn for 7 days for an overlapping time period (Figure 2). The nicotine levels were higher in the 7-day wristband, but only slightly (an average of 38.5% higher, rather than the 350% higher expected if exposure were cumulative over 7 days vs. 2 days). It is not known whether the wristband becomes close to steady-state in nicotine concentration in 2 days, becomes saturated, or if there is some other explanation for this finding, such as transfer from one wristband to another. Controlled chamber studies regarding uptake, equilibrium etc. for nicotine under varying concentrations are desirable to investigate these issues. In addition, methodological studies of the reproducibility and related issues and details such as whether to wash the wristbands are needed. Anderson et al. reported that when air concentrations of PAHs were compared with silicone wristband concentrations, the 24- hour concentrations of low molecular weight PAHs (naphthalene, 2-methylnaphthalene, 1-methylnaphthalene, and acenaphthene) were equal to concentration in wristbands worn for 7 days (28), which the authors ascribe to being in equilibrium after 24 hours. It should be noted that the log  $K_{oa}$  (octanol:air partition coefficient) of nicotine is estimated to be 8.081 (EPA EPI Suite™ <https://www.epa.gov/tsca-screening-tools/download-epi-suite-estimation->

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3 [program-interface-v411](#)) which is similar to the  $K_{oa}$  of the PAHs estimated to be not in equilibrium  
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5 between wristband and air at the end of a 7-day period (2).  
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7 Other routes of contamination of the wristband in addition to air exposures include dermal  
8 contact or contact with nicotine contaminated surfaces and house dust. It should be noted that  
9 use of the cotinine biomarker measures exposure through all routes (inhalation, ingestion, and  
10 dermal). Future investigations would include comparison of personal air levels of nicotine with  
11 silicone wristband levels of nicotine in the same person to help understand routes of exposure  
12 determined by the wristband, which might vary by exposure source. In a recent study of pesticide  
13 residues in silicone wristbands, the authors suggested that some of the pesticides could have  
14 come from diet, possibly through sweat excretion (33). Nicotine is found in sweat (34) and  
15 emanates from skin (35), so sweat could be a source of nicotine in these wristbands as well.  
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26 The silicone wristbands are also sensitive to low levels of nicotine exposure. In Figure 2,  
27 several children classified as not exposed to tobacco products indoors have higher urine cotinine  
28 and also higher wristband nicotine levels than others classified as not exposed (Figure 2). This  
29 implies that the wristband sampler does indeed detect increased exposure to nicotine even at low  
30 levels, relying on cotinine as the 'gold standard'. Children could have been exposed to outdoor  
31 electronic cigarette vapor or secondhand smoke, or indoor thirdhand smoke (21), as the  
32 classification of 'exposed' in our study refers to indoor exposure to active smoking/vaping only.  
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41 Our results indicate that a 2-day deployment of a silicone wristband for detection of a  
42 child's nicotine exposure tracks closely with exposure estimates from urine cotinine, although  
43 shorter time periods such as 1 day were not tested. Our results also imply that if the silicone  
44 sampler is left for a period of a week for practical reasons, the exposure estimation would not be  
45 substantially altered for exposures similar to the children in this study. Little is known about the  
46 optimal period of deployment for wristbands to detect nicotine or other substances. Longer  
47 periods of time could detect more intermittent exposures, but losses could also occur. Time  
48 periods of deployment in other studies range from 8 and 40 hours in a pilot study of occupational  
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3 exposure to PAHs (1), for environmental PAH exposure, 24 hours and 7 days (2), 48 hours in  
4 pregnant women (6) and as long as 21 days (4) and 1 month in adults (1, 4). For pesticides,  
5 periods of time the wristband was worn include 5 days (33), 2 periods up to 5 days (94% 4 to 5  
6 days) (5), 7 – 14 days (36) or a month (3), and for flame retardants 5 days (8), and 7 days (7, 9,  
7 10). Shorter deployments may have practical advantages, as a longer deployment allows for more  
8 time for sampler loss or misplacement. In our study, one of the 7-day wristbands was lost and two  
9 wristbands were misplaced for time periods of hours. Clearly, how long the wristband is worn will  
10 vary with study design, exposure patterns (e.g., continuous vs. intermittent) and practical issues,  
11 but our data indicate that for nicotine the deployment time may be flexible.

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22 The exposure measured here was to nicotine in tobacco products. This may arise from  
23 secondhand smoke from conventional cigarettes or secondhand vapor from electronic cigarettes,  
24 as well as potentially from nicotine exposure to thirdhand smoke through ingestion, dermal and  
25 inhalation routes (21, 26). It is possible that additional chemical compounds associated with  
26 tobacco measured in the wristband in a future study might distinguish EC exposure from CC  
27 exposure (see Figure 2). One example might be the measurement of carcinogenic tobacco-  
28 specific nitrosamines (TSNAs), which are associated with secondhand smoke from CC but are  
29 absent or in lower concentrations in EC vapor (37). These TSNAs are present in lower  
30 concentrations than nicotine and vary in their chemical properties, so it is not known how long a  
31 wristband would have to be worn to reliably detect them.

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43 There could be reasons why data points fall outside of the general association between  
44 the 2-day wristband nicotine and the child's urinary cotinine. In one case, the urine cotinine was  
45 high but the nicotine in the wristband was low (Figure 1, left upper quadrant). One possible  
46 explanation is that a high exposure to nicotine occurred just prior to the 2-day wearing period, and  
47 so was not captured by the wristband, but the exposure increased the level of cotinine in the urine.  
48 Another explanation is that the 2-day wristband was not worn or was covered part of the time  
49 even though not reported as such. The same participant had a urine cotinine value that was in  
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3 line with the 7-day wristband nicotine value (Figure 1, right panel). We did have verification  
4 procedures in place, including daily texts of photos, but relied on caregiver report. A third  
5 explanation is that the nicotine exposure occurred through a route not detected by the wristband,  
6 such as ingestion of dust.  
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11 In conclusion, the silicone wristband may be a simple sampler useful for assessing  
12 environmental exposure in children to secondhand smoke and tobacco-related products. Further  
13 development and testing are needed to determine the range of exposures detected, variation from  
14 wristband to wristband worn by the same child for the same time period, and practical details,  
15 such as how the samplers perform for nicotine analysis when mailed back by participants. In  
16 addition, the exposure route determined by the silicone sampler could be investigated through  
17 comparisons of air or sweat levels of nicotine with wristband levels. Further investigation should  
18 also take place into tobacco-related compounds that might be analyzed to determine the type of  
19 tobacco product exposure (e.g., conventional vs. electronic cigarettes) being measured by the  
20 wristband.  
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### Acknowledgements

The authors express their gratitude to Christine Batikian, MPH, Viridiana Mendoza, Samuel Padilla, and Madeleine Warman for assisting with data collection, and Mansi Vyas for sample preparation.

This research was supported by funds from the California Tobacco Related Disease Research Grants Program Office of the University of California, Grant Number 25IP-0023 (P. Quintana, Principal Investigator).

**Conflict of Interest**

KAA discloses a financial interest in MyExposome that is marketing products related to the research being reported. The terms of this arrangement have been reviewed and approved by Oregon State University in accordance with its policy on research conflict of interest. The other authors declare no conflict of interest.

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## **List of Figures**

**Figure 1.** Nicotine (ng/wristband) in silicone wristbands worn by a child for a) 2 days (DF = 29,  $r^2 = 0.741$ , F-statistic = 85, and  $p$ -value < 0.001) and b) 7 days (DF = 28,  $r^2 = 0.804$ , F-statistic = 120, and  $p$ -value < 0.001) compared to cotinine (ng/mL) levels in the child's urine (n = 31 and n = 30, respectively). Shaded area is the 95% confidence interval.

**Figure 2.** Nicotine (ng/wristband) levels in silicone wristbands worn by a child for 2 days compared to nicotine levels in wristband worn for 7 days (overlap for days 6 and 7) (DF = 28,  $r^2 = 0.852$ , F-statistic = 161, and  $p$ -value < 0.001). Shaded area is the 95% confidence interval.

*Abbreviations: CC—child exposed to conventional cigarettes inside the home, EC—child exposed to electronic cigarettes inside the home, CC + EC—child exposed to conventional cigarettes and electronic cigarettes inside the home, EC + outdoor CC— child exposed to electronic cigarettes inside the home and conventional cigarettes outdoors, NS—child not exposed to conventional cigarettes or electronic cigarettes inside or outside the home. All categories are based on caregiver self-report.*



Table 1. Child participant demographics, home characteristics, and exposure to nicotine products.

Child Study Participants (n = 31)	n (%)	25 <sup>th</sup> – 50 <sup>th</sup> – 75 <sup>th</sup> Centile
Age (years)		5 – 8 – 10
Gender (% female)	54.8	
Race/ethnicity (%)		
Multiracial and other	38.7	
Latino/Hispanic	29.0	
Caucasian/White	19.4	
African American/Black	12.9	
Annual family income (1,000 \$)		20 to 30 – 30 to 40 – 70 to 80
<b>Homes (n = 31)</b>		
Number of occupants		4 – 4 – 6
Number of rooms		6 – 7 – 8
Years living in residence (n = 29)		1.4 – 3.7 – 7.0
<b>Exposure to Nicotine Products *</b>		
Child exposed indoors <u>inside</u> the home to: (n = 30**) <ul style="list-style-type: none"> <li>Conventional cigarettes (CC)</li> <li>Electronic cigarettes (EC)</li> <li>Both CC and EC</li> <li>Neither CC or EC</li> </ul>	<ul style="list-style-type: none"> <li>7 (23.3)</li> <li>8 (26.7)</li> <li>1 (3.3)</li> <li>14 (46.7)</li> </ul>	
Child exposed in a car or indoors <u>away</u> from the home to: (n = 31) <ul style="list-style-type: none"> <li>Conventional cigarettes (CC)</li> <li>Electronic cigarettes (EC)</li> <li>Both CC and EC</li> <li>Neither CC or EC</li> </ul>	<ul style="list-style-type: none"> <li>7 (22.6)</li> <li>4 (12.9)</li> <li>1 (3.2)</li> <li>19 (61.3)</li> </ul>	
Child not exposed to CC or EC indoors or in a car (n = 31)	12 (38.7)	

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4 \* Note. "Exposure" is defined as the child was in the same indoor room or car when any part of  
5 a CC was smoked or EC was used in the past 7 days.

6 \*\* One caregiver did not know the smoking behavior of visitors to the home during the study  
7 period.  
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Table 2. Descriptive statistics for the levels of nicotine in the silicone wristbands (ng/wristband) worn for 7 days and 2 days (the last 2 days of the 7 day period) and the urine cotinine values measured in the same participant at the end of the wearing period.

	<b>2-Day Exposure, Nicotine (ng/wristband)</b>	<b>7-Day Exposure, Nicotine (ng/wristband)</b>	<b>Urinary Cotinine (ng/mL)</b>
<b><i>n</i></b>	31	30	31
<b>Mean</b>	301	375	2.01
<b>Geometric Mean</b>	99	129	1.22
<b>Median</b>	100	148	0.88
<b>Minimum</b>	10	11	non-detect
<b>25th Percentile</b>	17	35	0.18
<b>75th Percentile</b>	257	376	2.83
<b>Maximum</b>	2388	1893	11.54

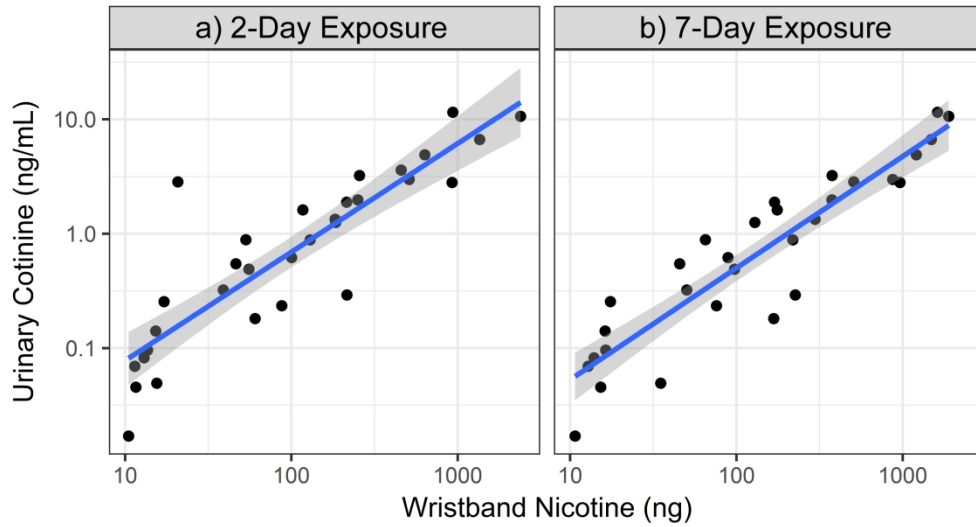


Figure 1. Nicotine (ng/wristband) in silicone wristbands worn by a child for a) 2 days (DF = 29,  $r^2 = 0.741$ , F-statistic = 85, and p-value < 0.001) and b) 7 days (DF = 28,  $r^2 = 0.804$ , F-statistic = 120, and p-value < 0.001) compared to cotinine (ng/mL) levels in the child's urine (n = 31 and n = 30, respectively). Shaded area is the 95% confidence interval

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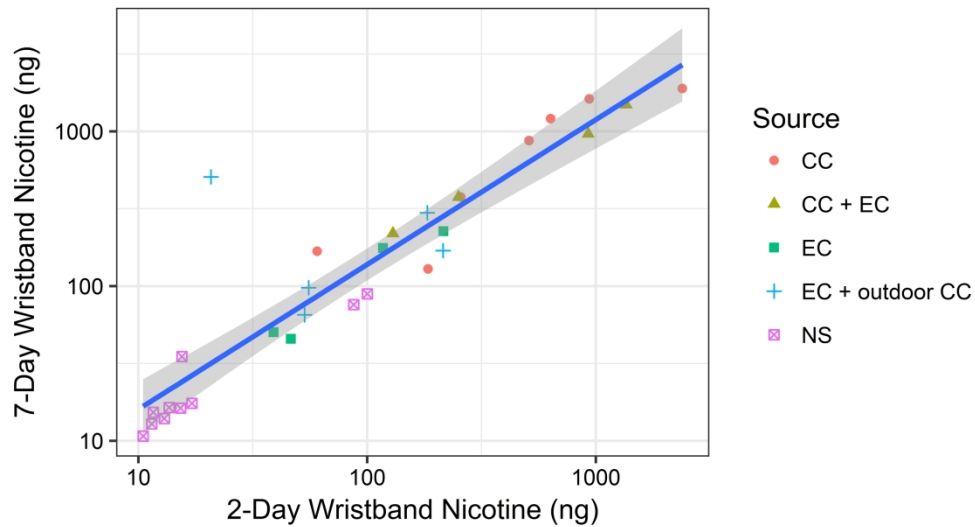


Figure 2. Nicotine (ng/wristband) levels in silicone wristbands worn by a child for 2 days compared to nicotine levels in wristband worn for 7 days (overlap for days 6 and 7) (DF = 28,  $r^2 = 0.852$ , F-statistic = 161, and p-value < 0.001). Shaded area is the 95% confidence interval. Abbreviations: CC—child exposed to conventional cigarettes inside the home, EC—child exposed to electronic cigarettes inside the home, CC + EC—child exposed to conventional cigarettes and electronic cigarettes inside the home, EC + outdoor CC—child exposed to electronic cigarettes inside the home and conventional cigarettes outdoors, NS—child not exposed to conventional cigarettes or electronic cigarettes inside or outside the home. All categories are based on caregiver self-report.

139x76mm (600 x 600 DPI)