Exposing the exposome to elucidate disease

Environmental factors, not genes constitute most disease risk. Myriad approaches are attempting to use the latest science and technology to more clearly reveal the complex mix of pollutants that contribute.

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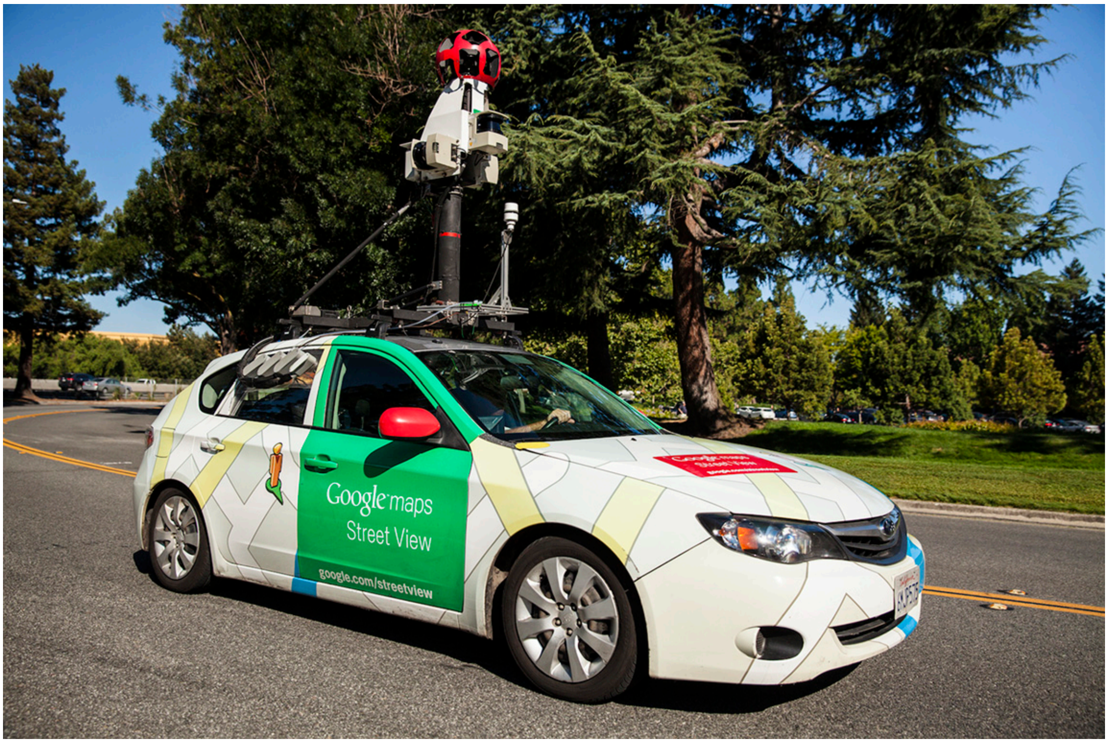
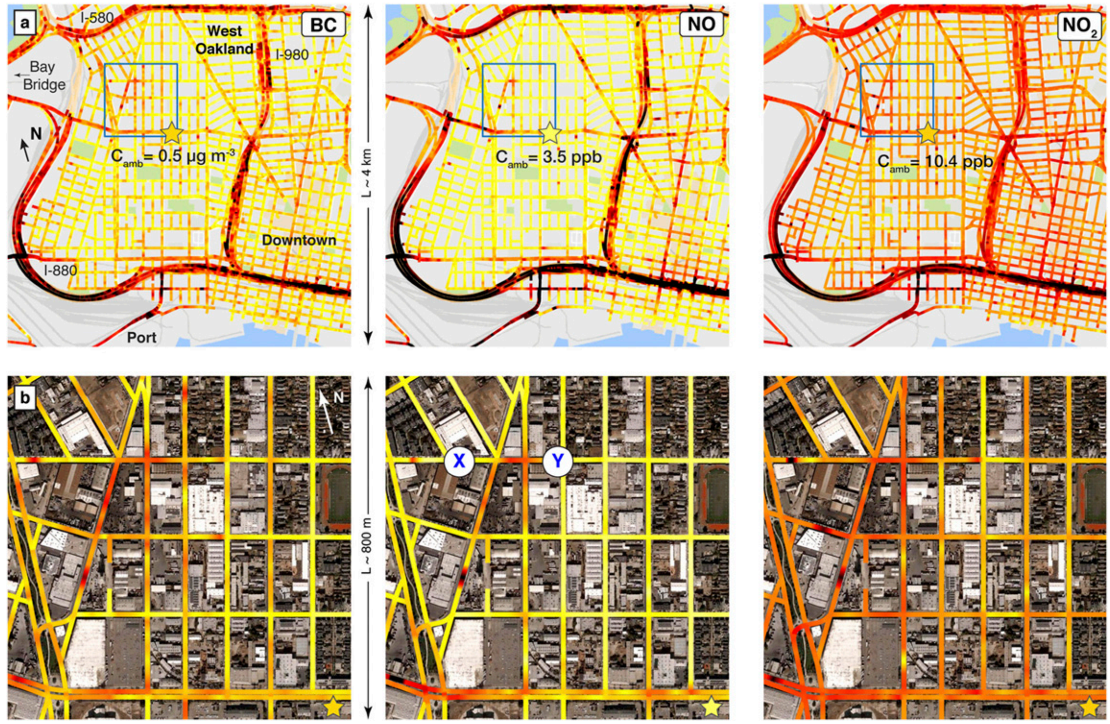
Google Street View cars traversing the roads of Oakland, CA, once captured a picture that goes beyond the usual map. On weekdays for 1 year starting in May 2015, two cars equipped with air pollution sensors drove city streets repeatedly. They produced a detailed view of air pollutant levels that differed greatly even within a given block (1). It's the sort of high-resolution detection that promises to greatly improve our understanding of how the environment affects our health. "Being able to understand how our exposures vary at the scales we live gives us a new, powerful tool to better understand health impacts and also be able to mitigate them," says Steven Hamburg, chief scientist at the Environmental Defense Fund.

Approximately 70 to 90% of disease risks are likely attributable to differences in environments (2), as suggested by studies of twins and research tracking groups that move from nations of low to high disease risk (3). In 2005, Christopher Wild, now director of the International Agency for Research on Cancer, coined the term "exposome" to describe the full suite of environmental exposures that a human experiences throughout life,



Google isn't only driving through neighborhoods to update its maps. This car, equipped with an air pollutant sensor, traveled through the streets of Oakland in 2014 and 2015 to get detailed readings of nitric oxide, nitrogen dioxide, and black carbon particles. Image courtesy of Aclima/Google.



In Oakland, air pollutant levels of black carbon (BC), nitric oxide (NO), and nitrogen dioxide (NO_2) can vary greatly within a single city block, even as the patterns of pollution remain stable throughout the year. Redder shading indicates higher concentrations of pollutants. The area within the blue squares (*Top*) is shown in detail in the aerial photographs (*Bottom*). Reproduced with permission from ref. 1 © (2017) American Chemical Society; images © 2016 Google, map data © 2016 Google.

starting at conception (4). These exposures include everything from infections, lifestyle, and stress to radiation, drugs, and pollution, which can all translate into chemical changes within the body that affect health.

Researchers have measured environmental exposures for decades. But new technologies are allowing Hamburg and others to capture an increasingly detailed view of those exposures at many scales—from satellites that sense chemicals across the United States, to biomarker studies that search for indicators of exposure within the body itself. Some studies focus on a single chemical, whereas others seek to reveal a bigger portion of the exposome, detecting exposures to hundreds of chemicals at once. In both fine and broad strokes, these studies bring researchers nearer to pinpointing the roots of disease. But each individual's collection of environmental exposures is diverse, with myriad interacting and changing components, whose direct influences on health are often unclear.

Pollutants, Block by Block

A couple of decades ago, Hamburg often sat on his porch in Providence, RI, wondering how the power plant down the street affected the health of his then young daughter. The plant burned Number 6 oil, a fuel known to release air pollutants. The Environmental Protection Agency and other regulatory agencies maintain air quality monitors across the United States. But these stationary sensors are too dispersed to provide data on differences within communities that might affect individuals. "What was the impact on the neighborhood?" Hamburg still wonders. "We didn't see black smoke, but we didn't know."

The plant has since switched to natural gas. But Hamburg still wants to know which chemicals people are exposed to, in what quantities, and from what sources. His goal for his Google Street View study was to begin to answer that question for the people of Oakland.

Industrial areas and warehouses sit beside residential homes in many parts of the city, and three major interstate highways and a container port surround West Oakland. But until pollution levels are directly measured on the scale at which people live, it's difficult to know where pollution hotspots really lie. "In some cases, it's going to be the big plant or a big highway," says Hamburg. "In other cases, it could be more localized sources." Boilers in homes, malfunctioning automotive engines, or poorly timed traffic lights that leave cars idling could all cause pollution.

Hamburg teamed up with Google Earth Outreach, researchers from the University of Texas at Austin and other institutions, and Aclima, Inc., an environmental-sensor technology company whose platform measured and logged nitric oxide, nitrogen dioxide, and black carbon particles from atop the cars every second along the routes.

The team found higher levels of these pollutants along busy streets and highways. But it also found localized hotspots. Even within a city block, pollutant concentrations could vary by more than five times, and the patterns remained stable throughout the year. Researchers searched the accompanying Google

Street View images for localized pollutant sources and saw many candidates—an intersection with heavy truck traffic, a car dealership offering a smog-check service, a major intersection with nearby parking lots, and a drive-through restaurant.

The researchers shared their findings with community groups such as the West Oakland Environmental Indicators Project (WOEIP). Margaret Gordon, West Oakland resident and cofounder of WOEIP, says her group has taken their own air quality measurements using hand-held sensors provided by Intel. The Google Street View data agreed with their findings but were more comprehensive and, according to Gordon, gave them more leverage. WOEIP is using the pollution map to lobby city governments, hoping that they'll require businesses to mitigate pollutants or move away from residential areas.

Under a new California bill known as AB 617, air quality regulatory agencies will now be monitoring air quality at a more localized rather than regional scale, in direct collaboration with community organizations such as the WOEIP. The Bay Area Air Quality Management District (BAAQMD), which regulates stationary sources of air pollution in the counties surrounding San Francisco Bay, is working with the WOEIP on an emissions-reduction plan for the West Oakland community. Their steering committee met for the first time in July 2018. "What the Google-Aclima study does is allow us to focus on potential hotspots that the study outlined," says Eric Stevenson, director of the BAAQMD's Division of Meteorology and Measurement. If it turns out that there is, for example, a major distribution center at one of the hotspots, "we can take action to try to modify truck routes or work with the facility to modify operating hours or things of that nature that will help reduce emissions in that area," says Stevenson.

Wristbands and Satellites

As researchers try to paint a more complete picture of the chemicals people encounter, various other efforts have started to collect exposure data at different spatial scales. Looking to home in on exposures in individuals, Kim Anderson, a professor of environmental and molecular toxicology at Oregon State University, has been using silicone wristbands, such as those that the Livestrong movement popularized, to sample chemicals in wearers' environments since 2014 (5). Silicone absorbs organic chemicals much like fat does, so chemicals passively enter the wristbands as wearers go about their days. Anderson's team then extracts the chemicals and identifies them using gas chromatography–mass spectrometry, and they can screen for more than 1,500 organic chemicals at once (6).

"What we see in our studies is there is a vast difference for individuals even within the same community," says Anderson. Her team found, for example, that each of 35 farmers in Diender, Senegal, who wore the wristbands for up to 5 days, had their own unique pesticide exposure profiles (7). When she asked participants to wear the wristbands for another period of up to 5 days, their individual results remained the same. The farmers were exposed to a wide range of pesticides, including dichlorodiphenyltrichloroethane (commonly known as DDT), cypermethrin, and deltamethrin. The study picked up 6 pesticides that the farmers reported using, as well as an additional 19 pesticides that they were unknowingly handling.

Looking to take advantage of a more expansive view, other researchers are using satellites to remotely sense chemicals via spectrophotometry. In a study published last year, researchers harnessed data collected by the Ozone Monitoring Instrument satellite sensor, part of NASA's Aura mission, every day over a 12-year period to map concentrations of formaldehyde, a carcinogen, across the contiguous United States (8). Although the satellite sampled vertical columns of approximately 13 × 24 kilometers, the researchers deduced formaldehyde levels at a finer resolution—5 × 5 kilometers—by analyzing the spaces where column measurements overlapped in subsequent readings. "We see the highest levels over the southeastern US," says lead author Lei Zhu, a postdoctoral fellow at Harvard University's Atmospheric Chemistry Modeling Group.

But the culprit doesn't appear to be manmade. "You have a lot of trees over there, especially the broadleaf trees—the main outdoor source of formaldehyde in the US," Zhu explains. The team then inferred the cancer risk associated with outdoor formaldehyde exposure across the United States by combining their findings with the same unit risk estimate for formaldehyde inhalation used by the Environmental Protection Agency.

Soon, researchers exploring pollutant exposure will have even finer-scale satellite data to draw on. As early as next year, NASA and the Smithsonian Astrophysical Observatory will launch a new instrument known as TEMPO (Tropospheric Emissions: Monitoring Pollution) that will capture air pollutant data hourly at an unprecedented spatial resolution of several square kilometers (9). Zhu notes that researchers harnessing data from this new satellite could use the same oversampling technique that his team used to further drill down in scale.

Hoping for even better exposure resolution, some work aims to identify biomarkers in biological samples that indicate a response to exposure (10). By analyzing blood or urine samples, for example, researchers can identify chemicals resulting from current exposures. Meanwhile, others are also exploring the biological signatures of past exposures, such as modified proteins that can indicate exposure to a chemical that occurred up to approximately 2 months before (11).

Through the Children's Health Exposure Analysis Resource (CHEAR), for example, the National Institute of Environmental Health Sciences (NIEHS) is working to reveal the associations between environmental exposures and the health and development of children. Some CHEAR-supported researchers are searching for links between biomarkers of exposure and health outcomes. One recent study of 439 pregnant women showed an association between biomarkers in urine that indicate exposure to phenols and parabens and altered thyroid hormone levels (12).

"We are not going to be able to measure everything, always, everywhere, in everybody," says David Balshaw, chief of the Exposure, Response, and Technology Branch at NIEHS. "But expanding our ability to measure lots of things very frequently, over time and over space—that is something that we can do and every day we are improving."

From Exposure to Risk

The challenge of measuring exposures looms large, in part, because of the sheer volume of unknown chemicals. Traditionally, researchers test for one or a few chemicals in an environmental or biological sample by comparing the sample to an analytical standard, a pure form of a chemical with known properties. But requiring a standard means ignoring almost all of chemical space, says Justin Teeguarden, the chief scientist for exposure science at the Pacific Northwest National Laboratory (PNNL), noting past research (13). A team of researchers at PNNL is now using supercomputing and computational chemistry to predict the properties of chemicals, allowing researchers to identify them without analytical standards. They can predict, for example, properties such as infrared and nuclear magnetic resonance spectra, which researchers can measure in real environmental samples using common laboratory instruments.

Even once researchers better understand exposure, they must tackle the tough task of determining how chemicals affect health. Members of Hamburg's team recently collaborated with researchers from Kaiser Permanente Northern California to link the Google Street View pollutant data with Oakland residents' health records. In May 2018, the team reported that, for elderly citizens, there was a correlation between higher air pollutant levels at residences and an increased risk of cardiovascular events (14). Many previous studies found similar results, says senior author, epidemiologist Stephen Van Den Eeden, a researcher at Kaiser Permanente Northern California. But his team required far fewer participants. "Even though we had roughly 41,000 individuals, many studies used hundreds of thousands or even millions," he says. The Google Street View data get "much closer to what people really experience at their homes," he says. By zeroing in on exposure levels, their team found answers with more modest costs and fewer logistical challenges than studies that assemble a massive number of participants. But although this study demonstrates a clear correlation, it, like similar epidemiological studies, does not demonstrate direct health effects of the pollutants.

Looking to come closer to connecting external exposure readings with internal consequences, Anderson's team asked 22 pregnant women to wear silicone wristbands for 48 hours. The team then compared the polycyclic aromatic hydrocarbons sampled by the wristbands with metabolites in the women's urine that the body produces in an attempt to detoxify these chemicals. In May 2018, the team reported correlations between exposure data and these metabolite biomarkers (15). But even in this case, the exact risk these chemicals pose to the participants' health is unclear. "The brass ring is health outcomes," says Anderson. "The urine doesn't tell you that you have a disease end point, but it does tell you that you've had an exposure."

Despite the challenges, researchers continue to accumulate increasingly detailed data on environmental exposure. Hamburg's team recently finished collecting Google Street View pollution data in Houston that they will once again use to produce a pollution map of the city. And in June 2018, they announced that London is next. "We believe firmly," says Hamburg, "that making the invisible visible empowers people."

1 Apte JS, et al. (2017) High-resolution air pollution mapping with Google Street View cars: Exploiting big data. *Environ Sci Technol* 51:6999–7008.
2 Rappaport SM, Smith MT (2010) Epidemiology. Environment and disease risks. *Science* 330:460–461.
3 Willett WC (2002) Balancing life-style and genomics research for disease prevention. *Science* 296:695–698.
4 Wild CP (2005) Complementing the genome with an "exposome": The outstanding challenge of environmental exposure measurement in molecular epidemiology. *Cancer Epidemiol Biomarkers Prev* 14:1847–1850.
5 O'Connell SG, Kincl LD, Anderson KA (2014) Silicone wristbands as personal passive samplers. *Environ Sci Technol* 48:3327–3335.
6 Bergmann AJ, Points GL, Scott RP, Wilson G, Anderson KA (2018) Development of quantitative screen for 1550 chemicals with GC-MS. *Anal Bioanal Chem* 410:3101–3110.
7 Donald CE, et al. (2016) Silicone wristbands detect individuals' pesticide exposures in West Africa. *R Soc Open Sci* 3:160433.
8 Zhu L, et al. (2017) Formaldehyde (HCHO) as a hazardous air pollutant: Mapping surface air concentrations from satellite and inferring cancer risks in the United States. *Environ Sci Technol* 51:5650–5657.
9 Smithsonian Astrophysical Observatory (2015) TEMPO Mission Overview. Available at tempo.si.edu/overview.html. Accessed October 8, 2018.
10 Dennis KK, et al. (2016) The importance of the biological impact of exposure to the concept of the exposome. *Environ Health Perspect* 124:1504–1510.
11 Grigoryan H, et al. (2016) Adductomics pipeline for untargeted analysis of modifications to Cys34 of human serum albumin. *Anal Chem* 88:10504–10512.
12 Aker AM, et al. (2018) Associations between maternal phenol and paraben urinary biomarkers and maternal hormones during pregnancy: A repeated measures study. *Environ Int* 113:341–349.
13 Dobson CM (2004) Chemical space and biology. *Nature* 432:824–828.
14 Alexeeff SE, et al. (2018) High-resolution mapping of traffic related air pollution with Google street view cars and incidence of cardiovascular events within neighborhoods in Oakland, CA. *Environ Health* 17:38.
15 Dixon HM, et al. (2018) Silicone wristbands compared with traditional polycyclic aromatic hydrocarbon exposure assessment methods. *Anal Bioanal Chem* 410:3059–3071.