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Greatly Enhanced Removal of Volatile Organic Carcinogens by a Genetically Modified Houseplant, Pothos Ivy (*Epipremnum aureum*) Expressing the Mammalian Cytochrome P450 *2e1* Gene

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Supporting Information

ABSTRACT: The indoor air in urban homes of developed countries is usually contaminated with significant levels of volatile organic carcinogens (VOCs), such as formaldehyde, benzene, and chloroform. There is a need for a practical, sustainable technology for the removal of VOCs in homes. Here we show that a detoxifying transgene, mammalian cytochrome P450 *2e1* can be expressed in a houseplant, *Epipremnum aureum*, pothos ivy, and that the resulting genetically modified plant has sufficient detoxifying activity against benzene and chloroform to suggest that biofilters using transgenic plants could remove VOCs from home air at useful rates.

INTRODUCTION

Household air is more polluted than office air and school air, and those who spend much of their time at home, such as children and home workers,[1] receive a proportionately higher dose of home air carcinogens[2] than the general population. Infants are particularly susceptible to indoor air pollution due to their low body weight and continuous exposure to indoor air. Loh et al.[3] ranked the cancer risks of indoor air volatile organic carcinogens (VOCs). The highest risk VOCs were benzene, formaldehyde, 1,3-butadiene, carbon tetrachloride, acetaldehyde, 1,4-dichlorobenzene (PDCB), naphthalene, perchloroethylene, chloroform, and ethylene dichloride. VOCs that exceeded acute exposure standards were acrolein and formaldehyde (during cooking)[4] and chloroform (during showering).[3]

Some sources of these chemicals can be eliminated or reduced. For example, PDCB could be greatly reduced by eliminating products containing it from the home. Formaldehyde in household air can be reduced by changing construction and upholstery material compositions, but formaldehyde is also emitted from other sources, including cooking,[4] which are not easily eliminated. Other carcinogens with multiple sources are more difficult to eliminate, such as benzene, which originates from fuel storage in attached garages, outside air, and environmental tobacco smoke.

Physical-chemical methods for VOC removal include adsorption on activated carbon, activated alumina, zeolites or other surfaces and photocatalytic oxidation.[5] Adsorption methods are not well suited for formaldehyde and other polar compounds. Low molecular weight compounds may be desorbed in competition with higher molecular weight pollutants. Adsorption methods are not destructive, and the sorbents must be periodically regenerated, usually remotely using energy intensive methods. Low temperature in-place methods achieved energy efficient regeneration but would require exterior ducting.[6] Oxidation methods use photocatalysized redox destruction of VOCs on catalytic materials, such as TiO_2. Photocatalytic oxidation methods result in complete mineralization of most pollutants, but they are ineffective with chlorinated VOCs such as chloroform. Further, photocatalytic oxidation methods may introduce ozone into the home air, and they are energy intensive.[5]

Indoor plants have been widely touted as having the ability to remove air pollutants from indoor air. This approach is known as the "green liver" concept and is a central idea of the field of phytoremediation, the use of plants to remove xenobiotic pollutants from the environment.[7] Early studies of air detoxification by household plants found that formaldehyde was removed from the air of chambers containing spider plants.[8,9] Other researchers reported that soil or water alone could explain the removal.[10] Subsequently, controlled pure culture plant experiments showed that plants can assimilate and metabolize formaldehyde from the air.[11−13] However, the formaldehyde uptake rate through the leaf surface of typical house plants appears to be insufficient to remove formaldehyde

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from a typical room without an excessive number of plants.[12,14] Several studies have found that common plants can remove VOCs such as formaldehyde and benzene from air, but those studies produced highly variable estimates of the rate that a particular plant species removes a given pollutant from air. The concentrations used in these tests were several orders of magnitude greater that those typical of home air (e.g., 1−7 μg m^{-3}).[15] For example, five different laboratories found that seven plant species removed benzene from the air at rates that varied by 7 orders of magnitude for the same plants.[16]

These conflicting data notwithstanding, plants do have many attractive features as a platform for metabolism of organic pollutants. Unlike most bacteria, cultivated plants have excess energy available to support cometabolic catalysis. Plants have high surface areas that facilitate mass transfer of trace gases from the air. Plants are self-sustaining and do not require the high maintenance typical of bacterial systems. There is certainty of the genetic and enzymatic composition of the cultivated plant compared to a soil bacterial community.

The mammalian cytochrome P450 2E1 (2E1) oxidizes a wide range of important VOCs found in home air, such as benzene, chloroform, trichloroethylene, and carbon tetrachloride.[17] The *CYP2e1 (2e1)* gene has been introduced into several plants, including trees, resulting in significantly increased degradation of the VOCs.[17−20]

Plants have been genetically modified to overexpress native plant formaldehyde dehydrogenase activity, but the rate of formaldehyde removal was increased by only 25% over unmodified plants.[13] Expression in transformed tobacco plants of the transgene for formaldehyde dehydrogenase, *faldh*, from *Brevibacillus brevis* increased formaldehyde removal by 3-fold.[21] But, to date, no detoxifying genes have been expressed in houseplants.

Thus, our objective in this study was to increase the detoxification of indoor air by adding the ability to metabolize VOCs to a common houseplant by transgene modification. Our approach was to introduce the mammalian cytochrome P450 *2e1* gene into the common houseplant, pothos ivy (*Epipremnum aureum*). Pothos ivy has several advantages over other houseplants for this purpose: it is robust and grows well in low light and a method for the transformation of pothos ivy has been published.[22,23] Pothos ivy does not flower in indoor cultivation or outdoors in the U.S., which is an advantage for biosafety considerations regarding the release of the transgenes into the environment. In order to provide additional biosafety assurances, we added *egfp*, the gene for the enhanced green fluorescent protein,[24] EGFP, to the cassette of genes used to transform the plant.

MATERIALS AND METHODS

Preparation of Pothos Ivy. Golden pothos ivy plants, obtained from a retail horticulture store, were grown under 50 μE m^{-2} s^{-1} illumination with a 16 h day/8 h night cycle at 25 °C in a plant room. The stem fragments were excised, surface-sterilized with 15% sodium hypochlorite and then washed with sterile deionized water three times. The sterilized stem fragments were cultured on solid Murashige and Skoog's (MS) basic medium[25] in culture vessels. After 1−2 months culture under light, new leaves and roots developed from stems and these sterile plants were used for infection with engineered agrobacteria for genetic modification.

Vector Construction and Genetic Transformation. In order to genetically modify pothos ivy we constructed a genetic vector containing the transgenes *2e1*, *egfp*, and *hpt*, each flanked by promoter and terminator sequences suitable for pothos ivy. The *hpt* gene coded for hygromycin B phosphotransferase, which confers resistance to hygromycin. Hygromycin was used to select for transformed cells since it kills wild-type pothos. These three genes were integrated into a transformation vector ("binary vector") based on a system of cloning vectors called pSAT containing insertion sites for use with specific restriction enzymes.[26] Then the binary vector was introduced into the modified *Agrobacterium* strain EHA105, which was used to infect pothos ivy callus cultures.

The rabbit cytochrome P450 *2e1* gene was amplified by polymerase chain reaction (PCR) from the plasmid pSLD50−6 (the sequences of the primers are listed in Supporting Information (SI) Table S1), a kind gift from S. L. Doty (University of Washington) and double digested with restriction enzymes *Hind* III and *Kpn*I. Then *2e1* DNA was inserted into cloning vector pNSAT3a to produce pNSAT3a-2E1. After insertion into pNSAT3a, the *2e1* gene was integrated between promoter and terminator sequence to produce an expression cassette to drive the expression of *2e1* in plant cells. The *egfp* gene was cloned by PCR from vector pGH00.0126[28] and inserted into pNSAT6a as a *Hind* III-*Pst*I fragment to produce pNSAT6a-EGFP. The expression cassettes of *hpt*, *2e1*, and *egfp* genes were cut from pNSAT1a-HPT,[27] pNSAT3a-2E1, and pNSAT6a-EGFP vectors using restriction enzymes *Asc* I, I-*Ppo* I, and PI-*Psp* I separately and inserted into the pRCS2 binary vector to produce pRCS2−2E1-EGFP.

The binary vector pRCS2−2E1-EGFP was transferred into *Agrobacterium* strain EHA105 by the freeze−thaw method[29] and the resulting strain, EHA105 (pRCS2−2E1-EGFP) was grown in LB medium (lysogeny broth) with 50 mg L^{-1} rifampicin, 100 mg L^{-1} spectinomycin, and 300 mg L^{-1} streptomycin for infection of pothos ivy. EHA105 (pRCS2−2E1-EGFP) was initiated in 100 mL LB medium with rifampicin at 50 mg L^{-1}, spectinomycin at 100 mg L^{-1} and cultured overnight at 28 °C on a rotary shaker at 200 rpm. The bacteria were centrifuged at 4000 rpm for 10 min and resuspended in liquid E medium (MS medium with 2 mg L^{-1} thidiazuron (TDZ) and 0.2 mg L^{-1} 1-naphthaleneacetic acid (NAA)) with 100 μM acetosyringone (AS) and cultured under the same conditions until OD_{600} (absorbance of bacteria suspension at 600 nm) of 0.8−1.0 was reached.

The following method for transformation of pothos ivy was adapted from that of Zhao et al.[22] Leaf discs and petiole segments from sterile pothos plants were immersed in the agrobacterium culture at 25 °C for 20 min and then transferred to double-layered filter paper moistened with liquid E medium with AS at 100 μM in Petri dish for 5-day coculture at 25 °C. The leaf discs and petiole fragments were washed with sterile water and transferred to E medium with 100 mg L^{-1} cefotaxime, 100 mg L^{-1} carbenicillin (PhytoTechnology Laboratories) and 20 mg L^{-1} hygromycin for screening. The explants were subcultured to fresh selection medium every 3 weeks.

After 2−3 months selection, the somatic embryos that developed from explants on selection medium were transferred to fresh medium for another month and then transferred to G medium (MS medium with 2 mg L^{-1} 6-benzylaminopurine (6-BA) and 0.2 mg L^{-1} NAA) with 100 mg L^{-1} cefotaxime, 100 mg L^{-1} carbenicillin and 20 mg L^{-1} hygromycin and cultured under light for regeneration. After culture for two months, the

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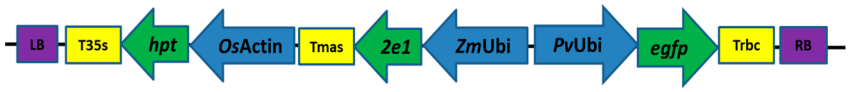

Figure 1. Structure of binary vector pRCS2−2E1-EGFP used to transform pothos ivy. T35s, terminator of CaMV 35s gene; *hpt*, hygromycin phosphotransferase gene, provides hygromycin resistance; *Os*Actin, promoter of actin gene of *Oryza sativa*; Tmas, terminator of mannopine synthase gene; *2e1*, cytochrome P450 2E1 gene from rabbit; *Zm*Ubi, promoter of ubiquitin of *Zea mays*; *Pv*Ubi, promoter of ubiquitin gene of *Panicum virgatum* (switchgrass); *egfp*, enhanced green fluorescent protein; Trbc, terminator of rubisco small subunit gene; LB, left border of T-DNA region; RB, right border of T-DNA region.

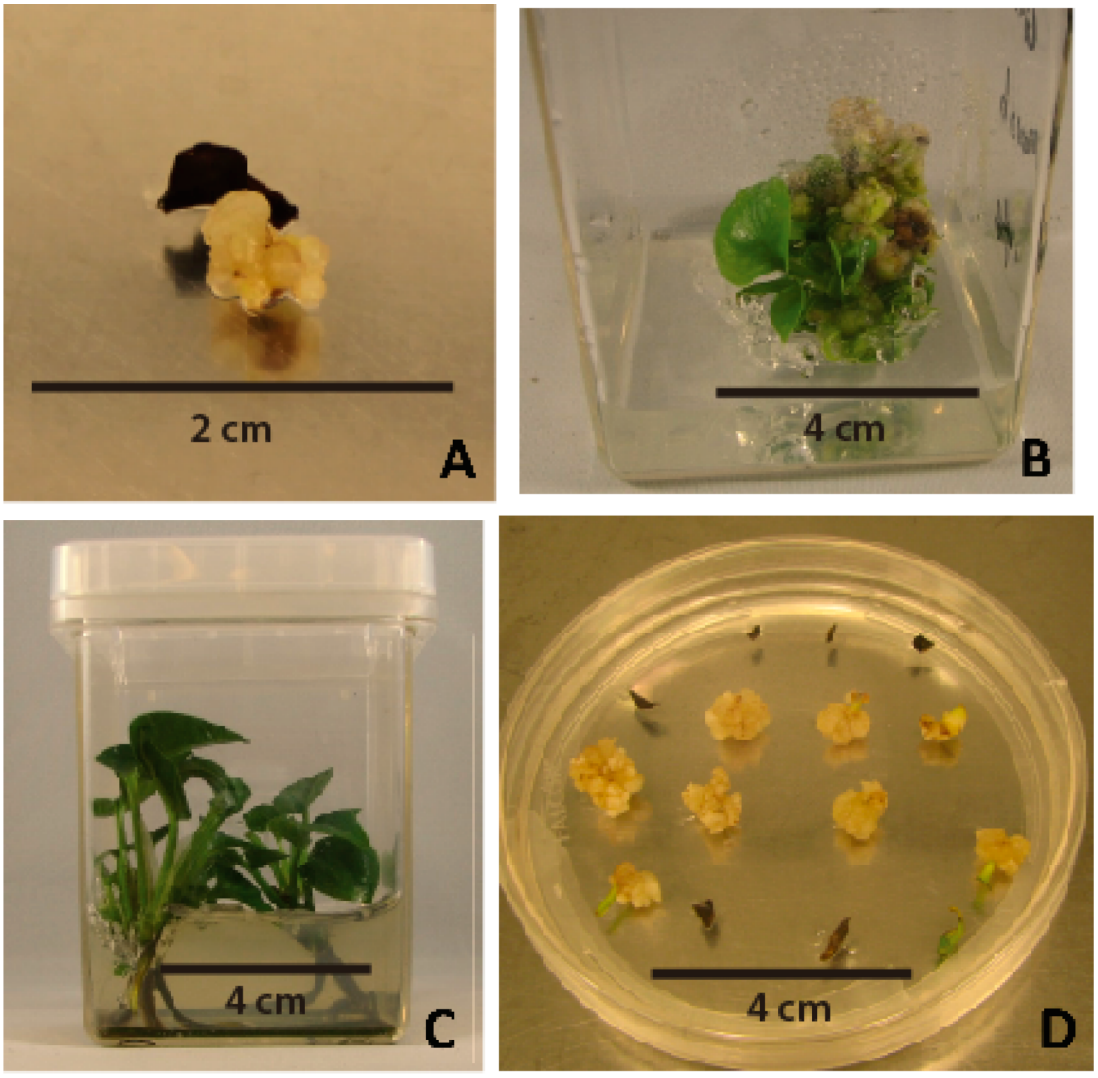

Figure 2. Genetic transformation of pothos ivy with *2e1* gene via *Agrobacterium* infection. Leaf discs and fragments of petiole of pothos ivy were infected with EHA105 harboring pRCS2−2E1-EGFP. The explants were cultured on somatic embryo induction medium with hygromycin at 20 mg L^{-1} for selection. (A) Callus developed from explants after 3−4 months screening. (B) Hygromycin resistant callus was transferred to regeneration medium supplied with hygromycin at 20 mg L^{-1} for 2−4 months to induce development of new plantlets. (C) Regenerated plants were transferred to MS medium with hygromycin at 20 mg L^{-1} for rooting and growth. (D) PCR- and RT-PCR-positive transformed plants were cultured on callus induction medium with hygromycin for callus induction and propagation.

regenerated plantlets were transferred to MS medium with 100 mg L^{-1} cefotaxime, 100 mg L^{-1} carbenicillin and 20 mg L^{-1} hygromycin for rooting and growth, which required two additional months of culture.

Molecular Analysis of Transformed Plants. For polymerase chain reaction (PCR) analysis, the DNeasy plant mini kit (Qiagen, Valencia, CA) was used to purify DNA from hygromycin-resistant plants. PCR reactions were carried out with primers specific to *2e1* and *egfp* cassettes (SI Table S1).

Total RNA was extracted from the leaves of plants using the RNeasy plant mini kit (Qiagen, Valencia, CA). For real-time quantitative RT-PCR analysis, 1 μg of total RNA was reverse transcribed to cDNA using M-MLV reverse transcriptase (Promega, Madison, WI). Real-time quantitative PCR was performed using the SensiFAST SYBR No-ROX kit (Bioline) on a fluorometric thermal cycler, Light Cycler (Roche), and data were analyzed with Light Cycler 3 software (Roche). The standard curve was constructed from the plasmid DNA of pRCS2-2E1-EGFP. The values of transcripts measured using

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RT-qPCR were normalized to the pothos ivy 5.8S gene and presented relative to the level of the transcript in clone VD1.

Benzene and Chloroform Uptake by Transformed Pothos Ivy. Sterile plantlets of pothos ivy clones (1 g) were incubated in 40 mL volatile organic analysis (VOA) vials (Fisher Scientific, 14−823-213), closed with septum valves (Mininert, Valco Instruments Co. Inc., 614163), and containing 5 mL half-strength Hoagland's solution (Caisson Laboratories, HOP01−10LT.1). Wild-type untransformed plantlets and no-plant controls were incubated in parallel with clone VD3 and each treatment was repeated in quadruplicate.

Benzene gas was injected into the vials using gastight glass syringes to achieve a headspace concentration of 1850 ± 160 mg m^{-3}, taking into account gas liquid partitioning by Henry's Law. The vials were incubated for 9 days with rotary shaking at 80 rpm. The concentration of benzene was determined by manually injecting 100 μL of the headspace into a GC−FID (flame ionization detector) (PerkinElmer AutoSystem XL). Chromatographic parameters were: oven temperature 60 °C, injector temperature 250 °C, and detector temperature 250 °C, 1.33 mL min^{-1} nitrogen carrier gas, using a ResTek RTX-1 microcapillary column (ResTek, 10121).

Similarly, chloroform was introduced into VOA vials using gastight glass syringes from sealed aqueous dilutions of chloroform (Acros Organics, 423550010). Transformed plantlets, wild type (WT), and no plant controls were incubated in quadruplicate, and headspace samples taken for analysis of chloroform levels by gas chromatography with an electron capture detector (GC−ECD) (PerkinElmer AutoSystem XL) with a VOCOL capillary column 60 m × 0.53 mm (Sigma). Chromatographic parameters were detector temperature at 325 °C, nitrogen carrier gas at 1.76 mL min^{-1}, with a 100 mL min^{-1} split, the oven at 100 °C, and the injection port at 300 °C.

EGFP Fluorescence. The EGFP signal of epidermal cells of pothos leaf was observed by fluorescent microscopy using the LSM 5 PASCAL system (ZEISS). The EGFP signal was excited by blue light and a FITI filter was used to collect fluorescent light. Axiocam 503 mono camera and software ZEN 2.3 lite were used to capture pictures.

Data Analysis. Data were analyzed for statistical significance using ANOVA in Microsoft Excel software (Microsoft Excel 2016 MSO). When ANOVA analysis gave a significant difference, Fishers Least Significant Difference (LSD) method was performed to compare the means. Groupings differing by statistical significance ($p < 0.05$) are labeled by letters in the figures.

RESULTS

Vector Construction and Generation of Transgenic Pothos Ivy. The structure of the plasmid pRCS2−2E1-EGFP used to transform pothos ivy is shown in Figure 1. In order to achieve constant, high levels of expression, all of the transgenes were driven by constitutive monocot promoters. The hygromycin resistance gene, *hpt*, was driven by the actin promoter from rice (*Oryza sativa*),[30] the *2e1* gene was driven by the ubiquitin promoter of corn (*Zea mays*),[31] and the *egfp* gene was driven by the ubiquitin promoter from switchgrass (*Panicum virgatum*).[32]

The explants of pothos ivy were infected with EHA105 containing the vector pRCS2−2E1-EGFP and then screened on callus induction medium with hygromycin as selection agent for 2−3 months. Capitate somatic embryos developed from cut edges of leaf discs and petiole fragments (Figure 2A). During subsequent culture, calli formed at the base and more cluster somatic embryos developed from the calli. After 3−4 months culture, the hygromycin-resistant calli were transferred to regeneration medium for induction of plantlets. After another 2−3 months culture, plantlets developed with both shoots and roots from the somatic embryos. Some plantlets developed only with shoots (Figure 2B). These plants were transferred to MS medium with hygromycin for further growth and rooting (Figure 2C). The leaf discs of PCR and RT-qPCR positive lines were cultured on E medium with 15 mg L^{-1} hygromycin to induce somatic embryos for propagation while still under selection (Figure 2D).

Molecular Analysis to Confirm the Transformation of Hygromycin-Resistant Lines. PCR primed by primer pairs annealing to promoter and terminator regions of *2e1* and *egfp* cassettes confirmed the integration of target genes into the genome of pothos ivy (data not shown). To measure the transcript abundance of *2e1* and *egfp* genes RT-qPCR was performed for eight transgenic lines, VD1-VD8. The expression levels of *egfp* were lower than that of *2e1*, and were separated into two groups, with significant differences between VD3 and VD2 or VD7 ($p < 0.01$, Figure 3). The expression levels of the *2e1* gene between different transformed lines were different with high significance (p = 0.00001). The clonal lines VD3, VD7, and VD8 had much higher expression levels of *2e1* compared to other lines. None of the transformed clonal lines had observable changes in morphology or growth compared to wild types.

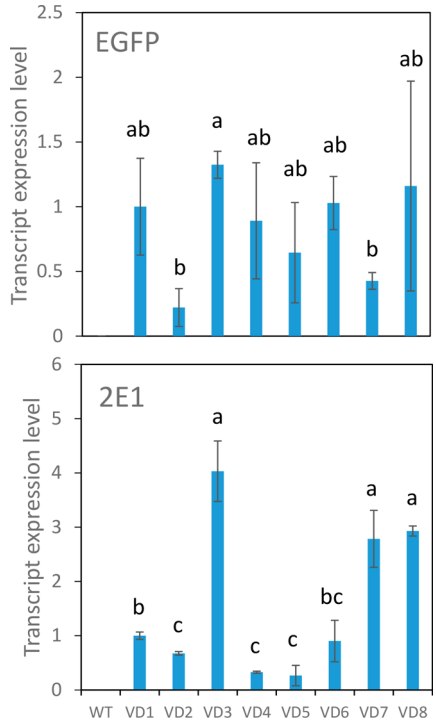

Figure 3. Transcript abundance measured using quantitative RT-PCR on pothos ivy lines transformed with *2e1* and *egfp* genes. The *y*-axis shows values that were normalized to the pothos ivy 5.8s rRNA gene and relative to the level of VD1 ($n = 3 \pm$ SE). Letters indicate means that were not significantly different ($p = 0.05$, ANOVA).

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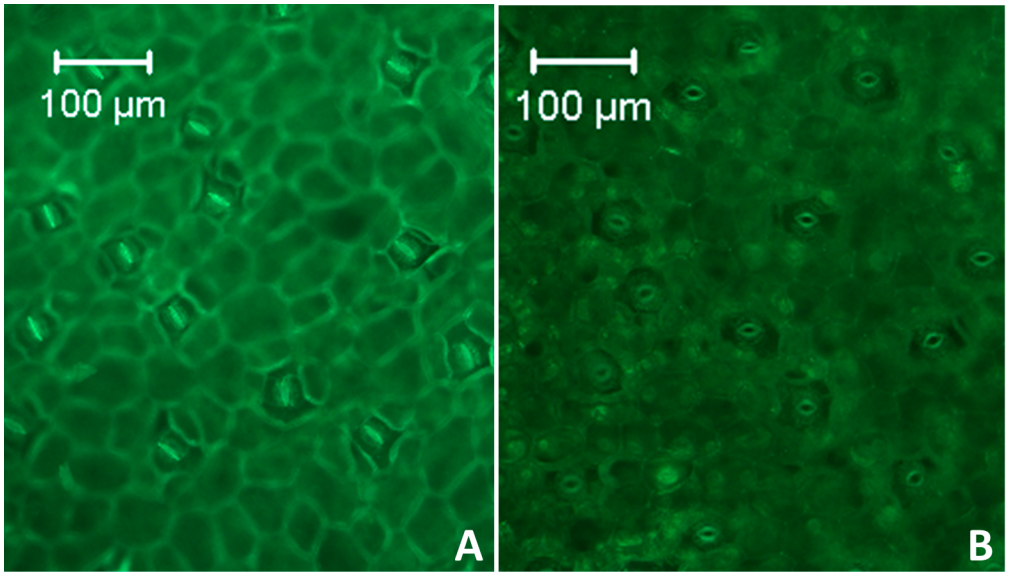

Figure 4. Observation of EGFP signal in the epidermal cells of pothos ivy clone VD3 using fluorescence microscopy. The green fluorescence signal of EGFP was observed in cytosol in the epidermal cells of leaf of pothos ivy clone VD3 (A). The emissions in the wild-type pothos ivy (B) were due to autofluorescence.

EGFP Observation. Using a fluorescent microscope, EGFP fluorescence was observed near the plasma membrane and around the nucleus (Figure 4A) due to the presence of vacuoles in the epidermal cells of pothos ivy leaf. The emissions were marginally greater than emissions from the wild type, but weak. The wild-type cells were weakly autofluorescent generally, but not specifically from the cytosol. Green fluorescence was not visible to the eye in the transformed pothos ivy with hand-held UV lamp illumination.

Benzene Uptake by Transformed Pothos Ivy. To determine the ability of *2e1-egfp* transformed pothos ivy to take up benzene, we incubated plants in closed vials with the VOC. Benzene (144 μg) was injected into 40 mL VOA vials containing transformed and wild-type pothos ivy to achieve a final headspace concentration of 2500 mg m^{-3} benzene. After 3 days culture, the benzene concentration in vials with VD3 plants had fallen dramatically (Figure 5). After 8 days, the benzene concentration in no-plant vials had fallen by about 10%. The benzene concentrations in the vials containing VD3 plants were significantly different compared to vials containing wild-type plants after 3 days culture (p = 0.039), p = 0.012 at day 4, and p = 0.0008 at day 8.

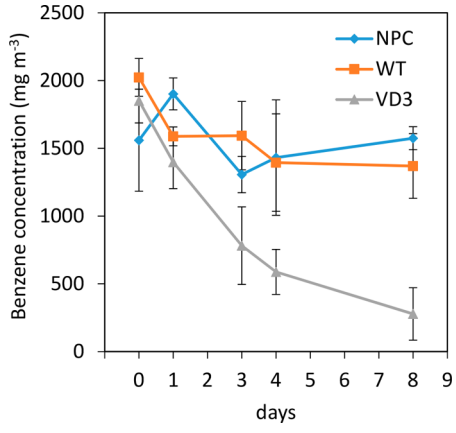

Figure 5. Uptake of benzene by *2e1-egfp* − transformed pothos ivy grown in liquid culture. The concentration of benzene in headspace during eight-day culture of VD3 (2e1), wild-type plants (WT), and no-plant controls (NPC). N = 4. Averages ± SE.

The time course of the benzene concentration in the vials with transformed pothos ivy clone VD3 was plotted on semilogarithmic axes and fit by linear regression with a first-order rate constant equal to (−0.249 d^{-1}, SI Figure S1), or −0.115 d^{-1} (g fresh biomass)$^{-1}$ (SI Table S2). Since small pothos plants have 29 cm^2 leaf area (g fresh biomass)$^{-1}$, this kinetic constant is equivalent to −39.8 d^{-1} (m^2 leaf area)$^{-1}$, normalized to leaf area. The slope of the best linear fit to the semilogarithmic plot of the time course of benzene concentration for wild-type plants (−0.044 d^{-1}, SI Figure S2) was significantly different from zero (p = 0.015), suggesting that the wild-type plants did take up some benzene. The wild-type pothos took up benzene at a first-order rate normalized to biomass equivalent to- 0.024 d^{-1} (g biomass)$^{-1}$, or −8.5 d^{-1} (m^2 leaf area)$^{-1}$, normalized to leaf area. The normalized rate constant for uptake of benzene uptake by transformed clone VD3 was 4.7 times that of the wild-type.

Chloroform Uptake by Transformed Pothos Ivy. The concentration of chloroform in the headspace of vials incubated with VD3 plants fell rapidly, while chloroform concentrations in incubations with wild-type plantlets and no-plant controls did not change significantly (Figure 6). The concentration of chloroform decreased by 82% during the first 3 days in the vials containing clone VD3 plants and chloroform was barely detectable after 6 days. Linear regression of the semilogarithmic plot of the chloroform data yielded a first-order degradation constant equal to −0.549 d^{-1} (SI Figure S3). The slope of the best linear fit to the semilogarithmic plot of the time course of chloroform concentration for wild-type plants (SI Figure S4) was not significantly different from zero (p = 0.22), suggesting that the wild-type plants did not take up chloroform. The rate constant for the VD3 transformed pothos ivy normalized to biomass was 0.552 d^{-1} (g fresh biomass)$^{-1}$, equivalent to 180 d^{-1} (m^2 leaf area)$^{-1}$, normalized to leaf area (SI Table S3).

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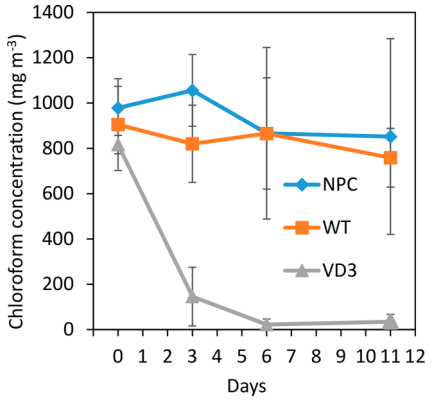

Figure 6. Uptake of chloroform by 2e1-egfp−transformed pothos ivy grown in liquid culture. The concentration of chloroform in headspace during 11-day culture of VD3, wild-type plants (WT), and no-plant controls (NPC). $N = 4 \pm$ SE.

DISCUSSION

VOCs in indoor air pose significant cancer risks to vulnerable populations, such as children, yet there are no practical, sustainable technologies available for their removal in the home. Physical-chemical methods based on sorbents and oxidation methods are energy intensive and of limited use for the removal of formaldehyde and chloroform, respectively. Various houseplants have been touted as having the ability to remove VOCs from air, but plant uptake rates vary exponentially from one study to another. Many studies appear to be affected by artifactual enhancement of soil bacterial activities by high VOC concentrations.[16] In this study we also used high VOC concentrations to facilitate analysis by hand injection of headspace samples onto GC−FID in the case of benzene, but we performed the assays in axenic conditions, without bacterial activity. As can be seen in Figures 5 and 6 there was little or no loss of benzene or chloroform in the vials containing wild-type pothos ivy, while most of the benzene and all of the chloroform was removed in 6 days in the vials with 2E1-expressing clone VD3. These results show the effectiveness of genetically modified pothos for VOC removal compared to wild-type pothos.

As we have shown for 2e1-transformed tobacco, other VOC substrates of 2E1 may also be removed by the transformed ivy.[33] Future work will determine whether other indoor air pollutants that are known to be substrates of 2E1 in mammalian cultures, such as PDCB, toluene, naphthalene, and methyl chloroform, are removed by *2e1*-transformed pothos.

Expression of green fluorescent protein was intended as a visible indication that the pothos ivy was transformed, but fluorescence of transformed clone VD3 was too weak to be visible without microscopy. Other variants of GFP, such as mGFP-ER[34] and the use of a stronger monocot promoter may provide stronger fluorescence.

Additional improvements for removal of VOCs from home air using transgenic houseplants could be made by combining expression of *2e1* with other detoxifying genes. Formaldehyde, the other VOC that poses most risk in home air will be of prime interest. Overexpression of *faldh* gene from *Brevibacillus brevis* in tobacco conferred plants a high tolerance to HCHO and increased the ability to take up formaldehyde 2−3 times faster than wild-type plants.[21] The *faldh* gene could be stacked with *2e1* and other detoxifying genes in vectors that are used to genetically modify pothos ivy and other houseplants, resulting in plants that could degrade most of the important indoor air VOCs.

Since *2e1* gene expression in the transformed pothos ivy is under constitutive promoters the level of 2E1 expression is expected to be independent of benzene or chloroform concentration. Therefore, the kinetic parameters of pollutant degradation are expected to be invariant with pollutant concentration. However, we have not confirmed this assumption empirically.

We calculated the performance of an enclosed, forced-air biofilter (see SI) using the same first-order degradation constant observed in the batch experiments with chloroform, 0.52 d^{-1} (g biomass)$^{-1}$. For the case of a completely mixed biofilter with a volume of 0.7 m^3 and an airflow rate of 300 m^3 h^{-1}, 10 kg of pothos ivy clone VD3 could remove 34% of the chloroform in one pass. This hypothetical biofilter would have a clean air delivery rate (CADR) of 100 m^3 h^{-1}, comparable to CADRs of current commercial home particulate filters.[35] This calculation, while tentative, suggests that genetically modified plants may have practical utility for sustainable phytoremediation of home air.

Compared to current chemical/physical methods for removal of VOCs from indoor air biofilters using transgenic plants offer the advantages of low energy use and decreased need for maintenance. All of the removal methods require a means for moving the air through the apparatus, but adsorptive methods also require significant energy expenditure to regenerate the media and photooxidative methods require high energy inputs to oxidize the pollutants, making those methods less sustainable. Transgenic phytoremediation requires very little additional energy beyond that required for air movement. Pothos ivy is well adapted to medium- and low-light levels so artificial lighting would usually not be required, giving phytoremediation an intrinsic sustainability advantage.

More work is needed to confirm these findings and to establish the practical usefulness of transgenic biofilters. It is necessary to determine the removal rates at low concentrations of indoor air pollutants, the effectiveness of the formaldehyde dehydrogenase gene expressed in pothos, the effects of light and dark and photoperiod on removal, the effects of increased mixing and air flow rate in the biofilter, and whether increased VOC removal efficiencies can be achieved through biological manipulations such as increased transgene copy numbers.

ASSOCIATED CONTENT

Supporting Information

The Supporting Information is available free of charge on the ACS Publications website at DOI: 10.1021/acs.est.8b04811.

The biofilter model, four figures and three tables (PDF)

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Notes

The authors declare no competing financial interest.

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