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Occurrence of Benzotriazole-Based UV Absorbers in Japanese Household Dust

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Benzotriazole-based ultraviolet absorbers (BUVs) are widely used in polymers due to their high thermal and photo stability. However, their environmental persistence and bioaccumulation potential have raised concerns, and some have become subject to regulation. BUVs have been detected in water, air, road dust, biota, and indoor products such as plastics and paints. They have also been frequently found in household dust (HD), which accumulates semi-volatile organic compounds; however, no studies to date have investigated the presence of BUVs in Japanese HD. This study established an LC–MS/MS method with atmospheric pressure chemical ionization (APCI) to quantify eight BUVs in HD and evaluate their occurrence in Japanese residences. Here, eight BUVs were selected for analysis, including UV-320—classified as a Class I specified chemical substance under Japan's Chemical Substances Control Law—and UV-328, newly listed in 2024. Sample preparation was optimized by evaluating six solid-phase extraction methods, among which basic alumina-based cartridges demonstrated selective BUV retention. High recoveries ranging from 86.9 to 100% were achieved using a back-flush elution approach. Using the developed method, HD samples collected in 2023 and 2024 were analyzed. Six compounds, excluding UV-320 and UV-PS, were detected in 100% of the samples. Although the maximum and minimum concentrations varied considerably between the two years, the median values and detection frequencies showed generally consistent trends. These findings reveal the contamination status of BUVs in Japanese HD and demonstrate that the developed APCI–LC–MS/MS method is a reliable approach for indoor pollution monitoring.

Key words indoor environment, household dust, benzotriazole-based UV absorbers, LC-MS/MS, atmospheric pressure chemical ionization

INTRODUCTION

Ultraviolet (UV) absorbers have been synthesized since the 1950s and have primarily been used to protect plastic products from UV-induced degradation. Generally, plastic deterioration is initiated by alkyl radicals generated by exposure to heat or UV radiation, which then trigger autoxidation reactions that lead to the cleavage of polymer chains.[1)] UV absorbers function by absorbing UV light and photonic energy, thereby suppressing radical formation and preventing material degradation. A distinctive feature of these compounds is that their absorption wavelength range and relaxation mechanisms are largely determined by their molecular structures.[2)]

Benzotriazole-based UV absorbers (BUVs) efficiently absorb UV radiation in the 200–400 nm range and exhibit high thermal and photo stability.[3)] Owing to their strong affinity for various polymers, they are widely added to plastics, coatings, textiles, and other materials. The global market for UV absorbers was approximately USD 670 million in 2020 and is projected to reach around USD 920 million by 2027.[3)] Among these, BUVs are particularly common due to their ability to impart heat and light resistance to materials, and their demand is expected to increase further.

BUVs are widely used in industrial and commercial applications, which increases the likelihood of their release into the environment. Owing to their persistence and high bioaccumulation potential, several compounds have been designated as regulated substances.[4)] Their environmental occurrence has been reported in various matrices, including seawater, ambient air, and road dust.[4-6)] Moreover, their detection in aquatic organisms and human breast milk has raised increasing concern about their potential biological effects.[5,7)]

With respect to toxicity, several BUVs have demonstrated biological effects. For instance, UV-320, a representative BUV, showed a no-observed-adverse-effect level of 2.5 mg/kg/day in female rat studies, suggesting a potential for endocrine-disrupting activity.[6)] With UV-328, a lowest-observed-adverse-effect level (LOAEL) of ~52 mg/kg/day was reported in rat studies conducted via dietary administration.[8)] Given the structural diversity of BUVs, evaluating the toxicological profiles

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of other related compounds is essential as well.

The presence of BUVs in indoor air and household dust (HD) has been investigated in residential homes in countries such as the Philippines and Norway,[9,10] where multiple BUVs, including UV-320—designated as a Class I chemical substance under Japan's Chemical Substances Control Law (CSCL)—were frequently detected. HD has long been recognized as a reservoir for various chemicals, including plasticizers and insecticides, underscoring its role in accumulating environmental pollutants.[11,12] However, no survey of BUVs in HD has been conducted in Japan to date.

Here, a quantitative analytical method for BUVs in HD was developed using an LC-MS/MS. Eight BUVs were selected for analysis, focusing on those previously detected in HD overseas, including UV-320 and UV-328—newly added to the Class I Specified Chemical Substance list under the CSCL in 2024. A survey was then conducted to investigate their occurrence in residential HD in Japan.

This study aimed to establish a reliable LC–MS/MS method with atmospheric pressure chemical ionization (APCI) for the quantitative determination of eight BUVs in HD and to elucidate their contamination status in Japanese residential environments.

MATERIALS AND METHODS

Chemicals and Equipment The standard compounds used were UV-P, -PS, -234, -326, -327, -328, and -329 (Tokyo Chemical Industry, Tokyo, Japan, purity >98%) and UV-320 (AccuStandard Co., CT, USA, purity 100%). All solvents used were of residual pesticide/PCB testing and HPLC grade (FUJIFILM Wako Pure Chemical Co., Osaka, Japan). Solid-phase extraction (SPE) cartridges included Oasis HLB Plus, Sep-Pak Silica Plus, Sep-Pak Florisil Plus, Sep-Pak Alumina B Plus, Sep-Pak Alumina N Plus (Waters Corporation, Milford, USA), and Bond Elut PRS (bed mass 1 g, volume 6 mL; Agilent Technologies, Inc., Santa Clara, USA). The analytical instruments are summarized in Table 1.

Separation and Dynamic Range Evaluation for Eight BUVs by LC–MS/MS The separation of the eight BUVs was evaluated using LC–MS/MS. A simple gradient elution of water and methanol was applied, and both electrospray ionization (ESI) and APCI were examined as ionization modes. Calibration curves were prepared to determine the working concentration ranges for each ionization technique. The analytical conditions are summarized in Table 1.

Evaluation of Elution Patterns Each SPE cartridge was pre-conditioned with 10 mL of acetone followed by 10 mL of hexane. A 0.5 mL aliquot of a BUV mixed standard solution (1000 ng/mL in acetone) was transferred to a test tube, and the solvent was evaporated to dryness under a gentle nitrogen stream. The residue was reconstituted in hexane and loaded onto the SPE cartridge (500 ng per cartridge).

Elution tests were conducted as follows: 8 mL of hexane was eluted into a graduated test tube, followed by 8 mL of an acetone:hexane (1:9, v/v) mixture into a separate tube. The eluates were gently evaporated to dryness under nitrogen, redissolved in 10 mL of methanol, and analyzed by LC-MS/MS. For the Sep-Pak Alumina B cartridge, five different elution conditions were evaluated using a reverse connection (backflush) following the initial elution mixture and a 10 min drying period under nitrogen. Specifically, 8 mL of 2-propanol:water (7:3, v/v), 5 mL of 2-propanol:water (7:3, v/v), 5 mL of 2-propanol:water (6:4, v/v), 5 mL of 2-propanol:water (1:1, v/v), and 5 mL of water were each passed through separate SPE cartridges. The eluates were collected into graduated test tubes, adjusted to a final volume of 10 mL, and analyzed by LC-MS/MS in ESI mode.

Table 1. LC-MS/MS Analytical Conditions for Benzotriazole-Based Ultraviolet Absorbers (BUVs), Including Parameters for Both Electrospray Ionization (ESI) and Atmospheric Pressure Chemical Ionization (APCI) and Multiple Reaction Monitoring (MRM) Transitions.

Liquid Chromatograph			
Instrument	Nexera LC-40D_{XR}		
Column	Inertcore plus C18 2.1 x 100 mm, 2.6μm		
Gradient program	70%B(0 min)-70%B(6.5 min)-90%B(9.5 min)-93%B(14 min)-93%B(18 min)-100%B(18.01 min)-100%B(24 min)-70%B(24.01 min)-70%B(30 min)		
Mobile phase	A: water, B: methanol		
Flow rate	0.25 mL/min		
Injection volume	10 μL		
Column oven	40°C		
Mass Spectrometer			Monitor ion (*m/z*) [CE (V)]
Instrument	LCMS-8050		UV-P, 226>107 [-21]
Ionization	ESI (positive)	APCI (positive)	UV-PS, 268>212 [-21]
Interface temperature	300°C	350°C	UV-234, 448>370 [-23]
Desolvation temperature	525°C	600°C	UV-320, 324>268 [-22]
DL temperature	250°C	200°C	UV-326, 316>260 [-20]
Heat block temperature	400°C	200°C	UV-327, 358>302 [-24]
Nebulizing gas	3 L/min	3 L/min	UV-328, 352>282 [-24]
Heating gas	10 L/min	-	UV-329, 324>57 [-28]
Drying gas	10 L/min	5 L/min	

Preparation of Washed HD HD collected using a vacuum cleaner was sieved using an electric sieve (MVS-1N, Taiyo Co., Ltd., Osaka, Japan), and the fraction <250 µm was used. A 100 mg portion of this HD was mixed with 3 mL of an acetone:methanol (1:1, v/v) solution and subjected to ultrasonic extraction for 15 min. After centrifugation (1,740 x *g* at 20°C for 5 min), the supernatant was discarded. This procedure was repeated three times. A final extraction was performed once using 3 mL of methanol under the same conditions. The resulting residue was dried in an oven at 75°C and used as washed HD.

Recovery Test Performed with Spiked to Washed HD A 0.05 mL aliquot of a BUV mixed standard solution (1000 ng/mL in acetone) was added to the washed HD using a micro syringe. After drying, the sample was extracted twice with 3 mL of acetone by ultrasonic treatment for 15 min. The supernatants obtained after centrifugation were collected into a concentrator tube and concentrated to 1 mL using a centrifugal evaporator. Subsequently, 9 mL of hexane was added, and the solution was loaded onto a Sep-Pak Alumina B cartridge pre-conditioned in advance. The cartridge was dried under a stream of nitrogen and then eluted in back-flush mode using 5 mL of the 2-propanol:water (6:4, v/v) mixture. The resulting eluate was analyzed. Recovery tests were performed in six replicates, and the eluates were analyzed and compared using both ESI and APCI ionization modes.

Determination of the Limits of Detection and Quantification The limits of detection (LOD) and quantification (LOQ) were determined by analyzing BUV mixed standard solutions at concentrations of 0.1 and 0.5 ng/mL in seven replicates. The LOD and LOQ were defined as 3σ and 10σ, respectively, based on the measurement standard deviation.

Field Survey in Residential Houses HD was collected by residents from individual households using vacuum cleaners, with samples accumulated over ~1 month. Sampling was conducted from October to December 2023 and from October to November 2024. The HD sample was sieved, and dust particles <250 µm were used. A 100 mg portion of the fractionated HD was subjected to ultrasonic extraction twice with 3 mL of acetone at 15 min. After centrifugation (1,740 x *g* at 20°C for 5 min), the combined supernatants were transferred to a concentration tube and concentrated to 1 mL using a centrifugal evaporator. Subsequently, 9 mL of hexane was added, and the mixture was loaded onto a pre-conditioned Sep-Pak Alumina B cartridge. After drying the SPE cartridge with nitrogen gas, back-flush elution was performed by reversing the cartridge and eluting with 5 mL of a 2-propanol:water mixture (6:4, v/v). The eluate was used as the analytical sample. For the field survey, extraction and purification procedures were performed in triplicate (*n* = 3) for each HD sample prior to analysis.

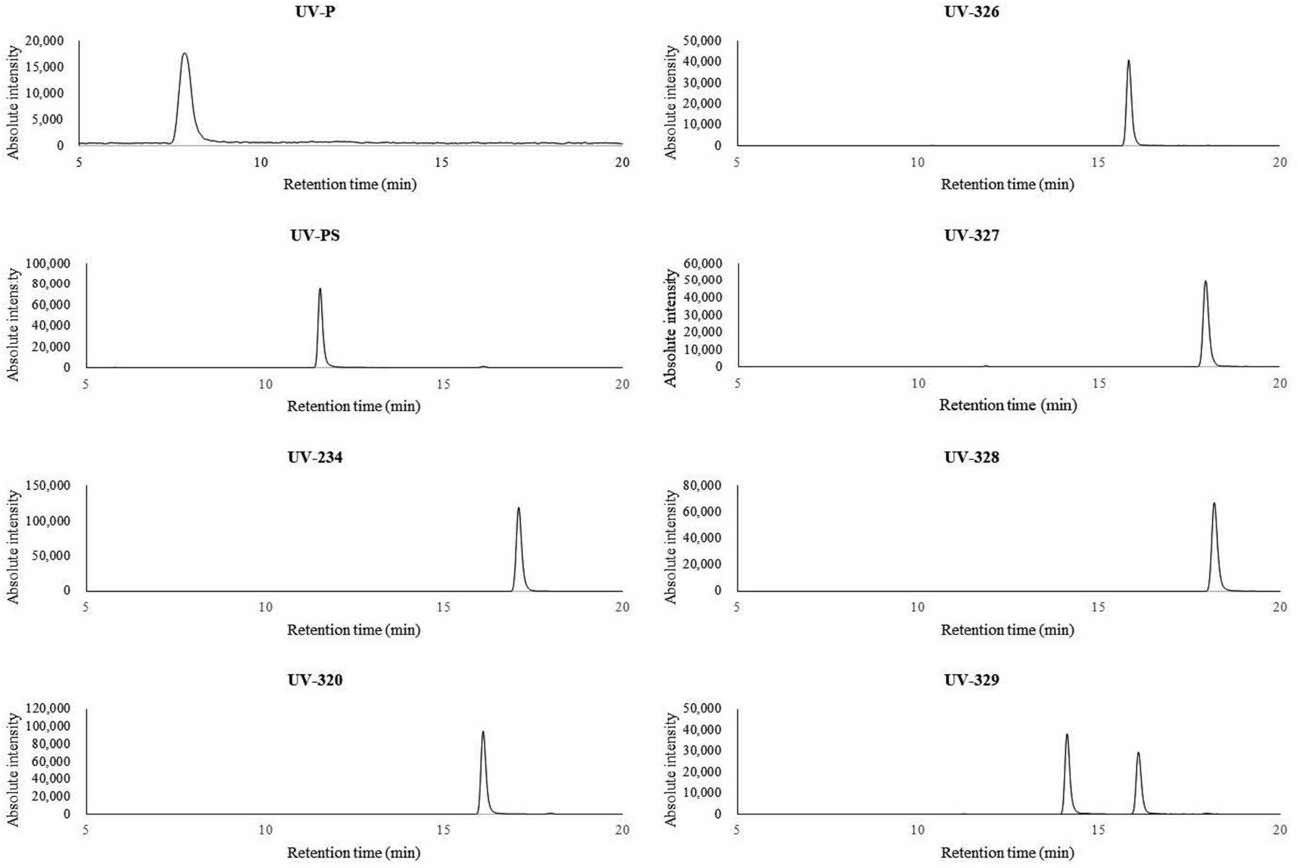

Fig. 1. Representative Multiple Reaction Monitoring Chromatograms of Eight Benzotriazole-Based Ultraviolet Absorbers (BUVs) Obtained under Optimized LC-MS/MS Conditions.

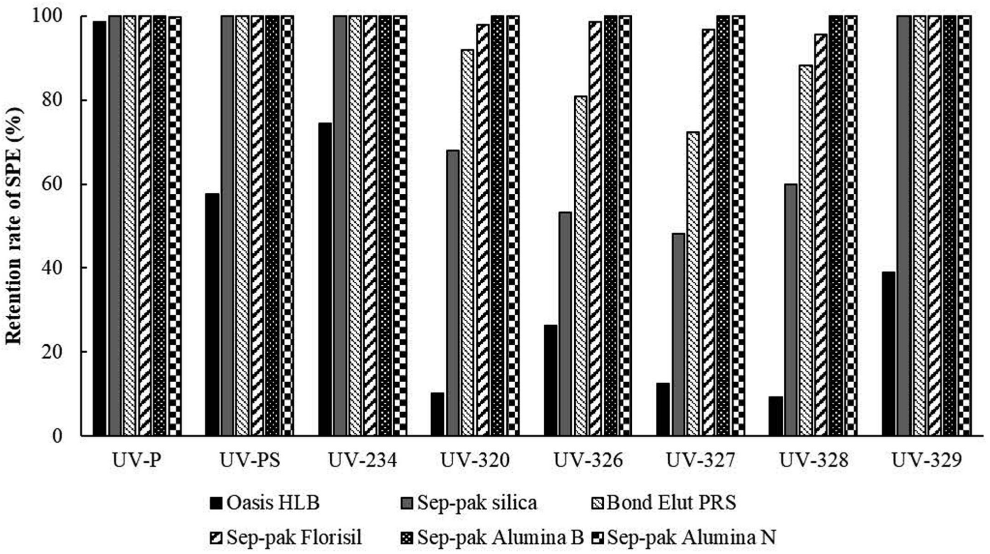

Fig. 2. Evaluation of BUVs Retention on Different Solid-Phase Extraction Cartridges Based on Elution with Hexane.

RESULTS AND DISCUSSION

Separation and Dynamic Range Evaluation for Eight BUVs by LC–MS/MS Analytical conditions for eight BUVs were first optimized using LC-MS/MS. A satisfactory chromatographic separation was achieved with a simple gradient elution of water and methanol (Fig. 1). Although overall separation was adequate, an overlap in precursor ions was observed between UV-320 and UV-329, both containing a *tert*-butyl group that generates a common fragment ion at *m/z* 57. As a result, two peaks appeared in the 324→57 transition. The retention time was verified using an authentic standard of UV-329, and the peak at ~14 min was assigned to UV-329 and used for quantification. Subsequently, both ESI and APCI were evaluated as ionization techniques. Based on calibration curves, the calibration ranges extended down to 0.01 or 0.05 ng/mL for ESI and to 0.1 ng/mL for APCI. Although ESI showed higher sensitivity at low concentrations, it tended to saturate at concentrations above 10 ng/mL for several compounds. In contrast, APCI enabled quantification up to 100 ng/mL, offering a broader dynamic range (Fig. S1).

Evaluation of Elution Patterns Because sample cleanup is essential for HD analysis, the elution patterns of various SPE cartridges were investigated using standard compounds under ESI conditions. As a result, three cartridges—Oasis HLB, Sep-Pak silica, and Bond Elut PRS—showed almost full elution with hexane alone, indicating insufficient retention and thus unsuitability for use here (Fig. 2). In contrast, florisil, alumina B, and alumina N retained the analytes under hexane conditions.

In a comparison between alumina B and florisil, alumina B retained the analytes even when eluted with the acetone:hexane (1:9, v/v) mixture, while florisil showed an elution efficiency of nearly 90% (Fig. S2). Furthermore, a comparison between alumina B and alumina N under the same solvent conditions revealed that alumina B exhibited stronger retention than alumina N (Fig. S2). These results indicate that alumina B has the potential to selectively retain BUVs.

Due to its strong retention capacity, alumina B yielded only 60–70% elution efficiency even when methanol-, water-, or acid-containing solvents were used. In particular, UV-P and UV-PS, which are less hydrophobic than other BUVs, showed elution efficiencies of only a few percent. To improve elution efficiency, a back-flush elution technique was employed by reversing the SPE cartridge and using a 2-propanol/water mixture as the eluent. This approach enabled efficient elution of the analytes with high yields. An optimization study on eluent volume revealed that 5 mL of the 2-propanol:water (6:4, v/v) mixture was optimal (Fig. 3). Given that the activity (retention strength) of alumina can be modulated stepwise by moisture content, the interaction between alumina and BUVs was presumed to be moderately weakened under these conditions.[13)]

Recovery Test Using Washed HD Samples and Ionization Mode Selection To further validate the analytical method, a recovery test was conducted using washed HD samples. The HD was washed with solvent to prepare blank HD, which served as a matrix for recovery evaluation. To confirm the effectiveness of the washing procedure in removing organic compounds, a GC-FID analysis was performed. The results verified that a substantial portion of the organic matter was successfully removed (Fig. S3).

After validating the blank matrix, standard compounds were spiked into the washed HD, then extracted and purified. The resulting samples were analyzed using both ESI and APCI to compare quantification performance. The recovery rates obtained with APCI were favorable, ranging from 83.9 to 105% for all BUVs. In contrast, ESI yielded lower and more variable recoveries, ranging from 22.8 to 77.1%. ESI is susceptible to ion suppression depending on the analyte used during LC-MS/MS analysis.[14)] Here, the results suggest that ion suppression caused by matrix components may have affected ESI performance (Fig. 4). Based on these findings, APCI was selected as the ionization method for the field survey.

Calculation of LOD and LOQ under APCI Conditions

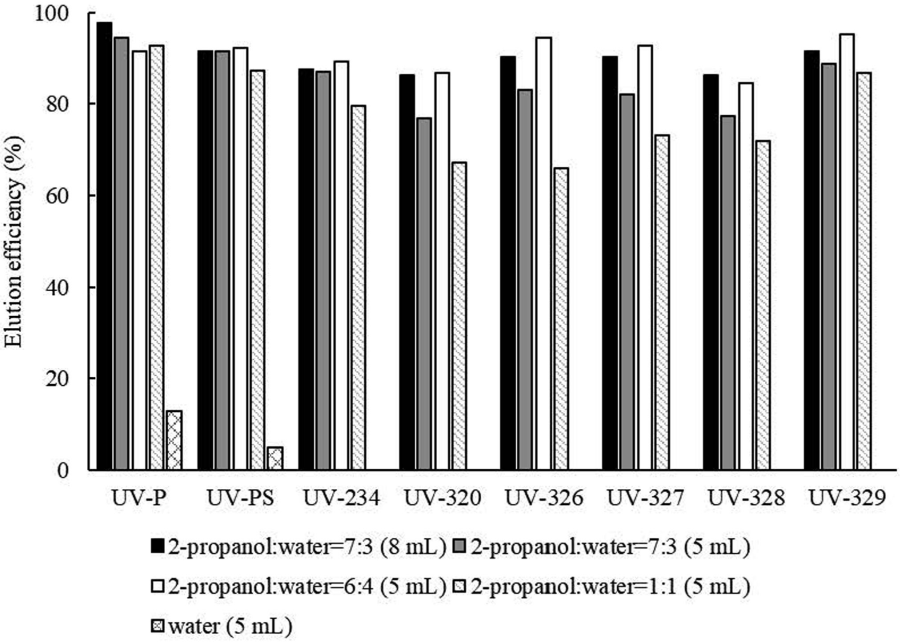

Fig. 3. Recovery of BUVs from Sep-Pak Alumina B Using Back-Flush Elution with Various Solvents.

The LOD and LOQ were calculated under optimized analytical conditions using APCI, based on seven replicate analyses of a mixed standard solution of BUVs. The resulting values were converted from ng per 0.1 g—assuming the sample mass used in the field survey—to units of ng/g (Table 2), yielding LOQs ranging from 5.18 to 14.3 ng/g across the eight BUVs. BUV concentrations in HD vary widely, with median values ranging from 3 to 84 ng/g.9) The findings indicate that the LOQs obtained here are sufficient for quantification in real samples.

Field Survey of BUVs in Residential HD A field survey of HD was conducted in residences in Tokyo and Kanagawa, covering 20 households in 2023 and 11 households in 2024. In total, samples were collected from 31 households across the two years. Of the eight BUVs investigated in HD, six were detected in 100% of the samples, except for UV-PS and UV-320 (Fig. 5, Table 3). This detection pattern generally aligns with those in previous findings in residential HD, 9, 10) although UV-320 represents a notable exception. In Japan, UV-320 was designated a CSCL in 2007, leading to a ban on its manufacture, use, and importation. Accordingly, the low detection frequency of UV-320 observed here is consistent with expectations based on these regulatory actions.

A comparison of the overall data from 2023 and 2024 revealed that although the maximum and minimum concentrations varied considerably between the two years, the median values and detection frequencies remained generally consistent (Table 3). Although the maximum concentrations of UV-327 differed between 2023 and 2024, the similarity in median values and detection frequencies across all samples suggests that the observed interannual difference is attributable to household-specific factors. This finding suggests that BUVs may persist as indoor contaminants in Japanese households. Among the BUVs analyzed, UV-328 has recently been added to Annex A of the Stockholm Convention and has also been classified as a Class I specified chemical substance under the CSCL in Japan. Therefore, continued monitoring of BUVs in indoor HD is necessary to track their presence and potential regulatory impacts.

In this study, the margin of exposure (MOE) was calculated based on the maximum concentrations of UV-328 observed in the field surveys conducted in 2023 and 2024. The reported LOAEL of 52 mg/kg/day was used for MOE calculation.8) The HD ingestion rates were assumed to be 0.1 g/day for children and 0.05 g/day for adults, based on values reported in the literature.15) Body weights of 15 kg for children and 60 kg for adults were applied. As a result, the MOE values for 2023 were calculated to be 5.65×10^6 for children and 5.27×10^7 for adults, while those for 2024 were 4.92×10^6 for children and 3.94×10^7 for adults.

The calculated MOE values were evaluated by comparison with the product of uncertainty factors (UFs), following the approach proposed by the National Institute of Technology and Evaluation (NITE) and European Food Safety Authority (EFSA). Because MOE values do not inherently account for uncertainties such as interspecies differences and human variability, MOE values exceeding the corresponding UFs are interpreted as indicating low concern at a screening level.16) In this study, a UFs value of 1000 was applied, considering factors of 10 for interspecies differences, 10 for human variability, and 10 for the use of a LOAEL instead of a no-observed-adverse-effect level (NOAEL).17, 18) The MOE values obtained for UV-328 substantially exceeded the UFs value of 1000, suggesting that exposure to UV-328 via oral ingestion of HD is of low concern at a screening level.

Although this study clarified the occurrence of BUVs in HD, sampling was restricted to a specific region and period, without incorporating seasonal or regional variability. Future studies covering wider areas, longer sampling periods, and related additional analytes will be essential to elucidate BUV sources and exposure pathways in indoor environments.

Conclusion An analytical method for eight BUVs in HD using LC-MS/MS, along with a purification protocol, was

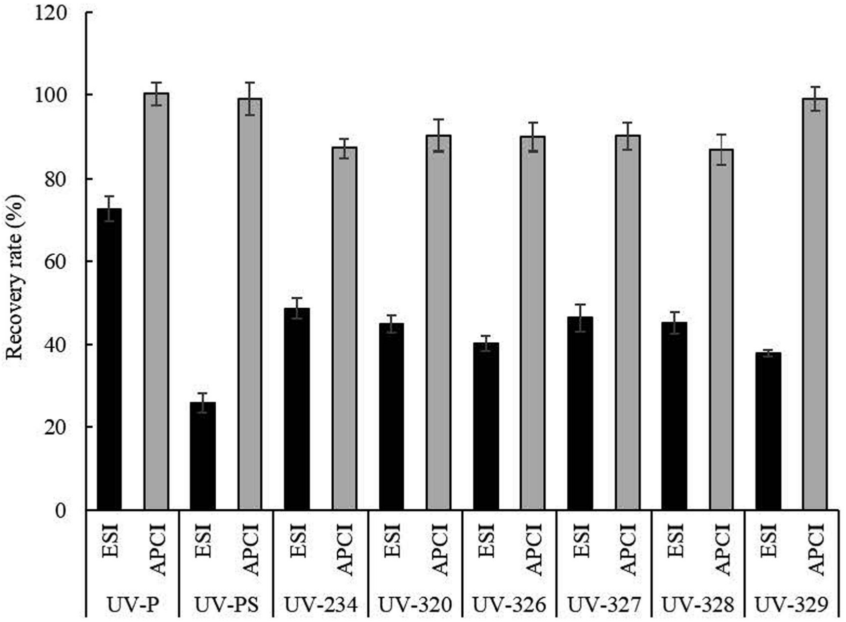

Fig. 4. BUVs Recovery Rates from Washed Household Dust (*n* = 6) Using Electrospray Ionization (Black Bars) and Atmospheric Pressure Chemical Ionization (Gray Bars).

Table 2. The Limits of Detection (LOD) and Quantification (LOQ) for BUVs Determined Here (*n* = 7)

ng/g	UV-P	UV-PS	UV-234	UV-320	UV-326	UV-327	UV-328	UV-329
LOD	4.29	1.57	1.80	3.09	2.12	1.55	1.95	1.65
LOQ	14.32	5.23	5.98	10.31	7.05	5.18	6.52	5.50

Table 3. BUVs Concentrations Detected in Household Dust Collected from September to December 2023 and September to October 2024.

ng/g	UV-P		UV-PS		UV-234		UV-320	
Year	2023	2024	2023	2024	2023	2024	2023	2024
Max	1078	3706	Tr	31.3	2588	3021	Tr	Tr
Median	370	450	Tr	15.7	350	307	Tr	Tr
Min	85.8	195	Tr	3.56	49.1	80.2	Tr	Tr
Detection frequency (%)	100	100	8.33	18.2	100	100	26.7	18.2
Ref 9 (Philippines, Survey year 2008: Min-Max (DF%))	-		-		ND-817 (94%)		ND-25 (82%)	
Ref 10 (Norway, Survey year 2018: Min-Max (DF%))	-		-		-		<1-475 (96%)	

ng/g	UV-326		UV-327		UV-328		UV-329	
Year	2023	2024	2023	2024	2023	2024	2023	2024
Max	10582	4548	7458	448	1381	1584	5810	5758
Median	780	1347	27.6	19.0	126	164	281	399
Min	250	555	5.74	8.30	28.9	59.3	40.8	58.9
Detection frequency (%)	100	100	100	100	100	100	100	100
Ref 9 (Philippines, Survey year 2008: Min-Max (DF%))	ND-275 (88%)		ND-73 (88%)		ND-304 (82%)		-	
Ref 10 (Norway, Survey year 2018: Min-Max (DF%))	24-1600 (100%)		<1-800 (96%)		1-18000 (100%)		7.8-8250 (100%)	

Note: Tr: trace, ND: not detected, DF: detection frequency

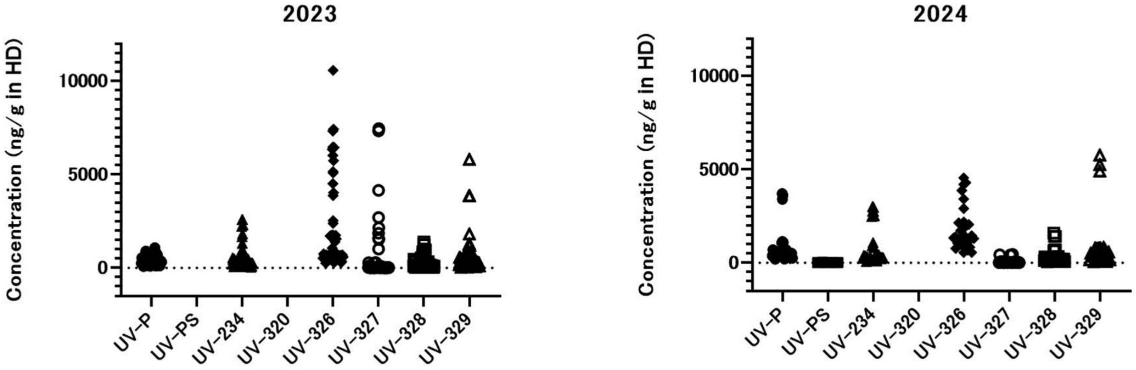

Fig. 5. Scatter Plots Showing BUVs Concentrations Detected in Household Dust Samples Collected in 2023 and 2024.

developed. APCI was selected as the ionization technique due to its broader dynamic range and more stable recoveries compared to those of ESI. For purification, Sep-Pak Alumina B demonstrated strong BUV retention, and quantitative elution was successfully achieved via a back-flush technique. A field survey applying the developed method revealed that BUVs are likely ubiquitous in HD from residential houses in Japan. These findings provide an analytical foundation for assessing indoor exposure to semi-volatile organic pollutants and contribute to the improvement of risk evaluation in indoor environments. To better understand the distribution and potential risks of BUVs in indoor environments, conducting extensive monitoring surveys using the excellent testing methods developed here is necessary.

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Conflict of interest The authors declare no conflict of interest.

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