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ORIGINAL ARTICLE

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Spectrophotometric assay of a wood preservative, didecyldimethylammonium chloride (DDAC), in aqueous solution

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Abstract A spectrophotometric assay based on the color reaction between didecyldimethylammonium chloride (DDAC) and 4-[4-(dipropylamino)phenylazo]-benzenesulfonic acid (propyl orange) was used for the determination of DDAC as a wood preservative. The assay was carried out using a propyl orange solution at pH 2.9. The visible absorbance spectrum of propyl orange showed an absorbance maximum at 510 nm, which decreased continuously with increasing DDAC concentration from 0 to 20 ppm. A linear correlation was observed at a DDAC concentration lower than 8 ppm. To apply this assay method to determine DDAC retention in treated wood, the influence of wood extractives on the assay was investigated. Wood extractives from sapwood and heartwood of Japanese cedar, Hinoki cypress, Japanese larch, and Western hemlock showed no influence on DDAC determination except in the case of heartwood from Japanese cedar and Hinoki cypress, which gave apparent DDAC concentrations higher than the actual values. However, it was also found that absorbance measurement at 477 nm solved this overestimation and gave precise values. It was concluded that this assay is a viable alternative to the current methods for DDAC determination.

Key words Didecyldimethylammonium chloride · Quaternary ammonium chloride · Propyl orange · Treated wood · Wood preservative

Introduction

Although wood is regarded as a carbon sink and a useful material for mitigating global warming,[1,2] it is also a source of carbon dioxide emission. In view of mitigating global warming, therefore, it is very important to prolong the service life of wooden materials and thus delay the emission of carbon dioxide gas. Therefore, extended service life is expected through preservative treatments.[3,4]

Among several types of preservatives, the ratio of waterborne copper-free preservatives has gradually increased in the last few decades in Japan.[5] Among the those preservatives, DDAC is the most commonly used by virtue of its colorless appearance, high penetrability, low odor, and zero metal content. The importance of these merits means that DDAC-treated timber will continue to be used in the future.

The performance of wood treated with DDAC has been studied by many laboratories, and the results reveal that the degree of protection achieved by DDAC depends on proper penetration and retention of the chemicals.[6–8] Therefore, the minimum penetration and retention are set for each use category.[9]

DDAC penetration is commonly determined by means of an easy color identification test using bromophenol blue[9]; on the other hand, analysis of DDAC retention is more difficult. In accordance with Japan Agricultural Standards (JAS)[9] and the American Wood Protection Association Standard (AWPAS),[10] DDAC is first extracted from treated wood using a solvent, and then analyzed to determine its concentration. Quantitative assay of DDAC in the solvent can be carried out in several ways. For example, according to the JAS method, DDAC coupled with Orange II, 4-(2-hydroxy-1-naphthylazo)benzenesulfonic acid, in a buffer solution at pH 3.5 is extracted using chloroform, and visible absorbance of the DDAC–Orange II complex is measured at 485 nm. The AWPAS prescribes using an HPLC system with a cation-exchange column. Although either of these methods can be used to determine DDAC concentration, several problems remain. For example, in the JAS method, a large amount of chloroform, which is listed as a toxic substance in Japan, is required to extract the DDAC–Orange II complex. The JAS method was also mentioned to lack reproducibility and stability.[11] In the AWPAS method, DDAC is analyzed by an ion-pair chromatography using an ion-exchange column. It is noted that the ion-pair chromatography requires a long time for column equilibration.[12] In other words, this method is not

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suitable to determine DDAC amounts of a few samples. Therefore, a new environmentally benign, rapid, and cost-effective technique is expected for the determination of DDAC retention.

DDAC is classified as a quaternary ammonium compound (QAC), which are compounds widely used in fabric softeners, mineral flotation agents, corrosion inhibitors, phase-transfer catalysts, and bioactive agents.[13] Consequently, numerous techniques have been developed for determination of these compounds.[14–22]

We experimentally applied one of those techniques to the determination of DDAC retention and found it to be successful. This report describes our study on a simple, eco-friendly, and cost-effective approach for DDAC determination.

Materials and methods

Materials

DDAC standard was kindly supplied by Mr. Hiroyuki Nagano, Xyence Co. Ltd. *N*,*N*-di-*n*-propylaniline was a first-grade reagent, obtained from Wako Pure Chemicals (Tokyo, Japan). All other chemicals were special-grade reagents (from Wako Pure Chemicals).

An IKA Labortechnik A11 basic analytical mill was used to prepare wood powder from the sapwood and heartwood of commercial timber from Japanese cedar (*Cryptomeria japonica*), Hinoki cypress (*Chamaecyparis obtusa*), Japanese larch (*Larix kaempferi*), and Western hemlock (*Tsuga heterophylla*), respectively. Extraction solvent was prepared by adding formic acid to methanol until a pH meter (TOA; HM-30S) showed pH 5.0.

Preparation of assay solution

Propyl orange was synthesized from sodium sulfanilate, sodium nitrite, and *N*,*N*-di-*n*-propylaniline according to the method reported by Motomizu et al.[16] Propyl orange was crystallized twice from water and kept under dry, dark conditions. Synthesized propyl orange was checked by measuring nuclear magnetic resonance (NMR) spectra on a JEOL ALPHA-500 NMR spectrometer at room temperature in dimethyl sulfoxide-*d6*: 1*H*-NMR (ppm) δ: 0.90 (6H, *t*, J = 7.5 Hz), 1.57 (4H, m), 3.39 (4H, *t*, J = 7.5Hz), 6.87 (2H, *d*, J = 9.0Hz), 7.70 (2H, *d*, J = 8.5 Hz), 7.73 (2H, *d*, J = 8.5 Hz), 7.79 (2H, *d*, J = 9.0 Hz).

The assay solution was prepared by mixing about 7.2 mg propyl orange with 1 l 20 mM sodium monochloroacetate buffer adjusted to pH 2.9 and containing 0.02% Triton X-100.

Determination of DDAC concentration using propyl orange

DDAC standard containing 19.9% DDAC was diluted with the extraction solvent to make a standard solution containing 0–5000 ppm DDAC. Then, 20 µl standard solution was added to 1980 µl assay solution. The visible absorption spectrum of the mixed solution was recorded on a Shimadzu UV-2400 spectrometer.

Preparation of methanol solution containing wood extractives

Wood extractives were prepared from the abovementioned wood powders. The extraction procedure basically followed the method of AWPAS A16-08.[10] One gram of wood powder, weighing double that in the standard, was placed in a bottle with a screw cap. The wood powder was mixed with 20 ml extraction solvent in the bottle and placed in an ultrasonic bath for 3 h. The extraction solution was then allowed to cool and settle. Part of the supernatant solution was filtered with a membrane filter (DISMIC-25HP; ADVANTECH) for use in further studies.

Determination of DDAC concentration in the presence of wood extractives

Sample solution was prepared by mixing filtered supernatant containing wood extractives with the same volume of DDAC standard solution. Control solution was prepared by mixing the extraction solvent with the DDAC standard solution. Then, 20 µl sample solution or control solution was mixed with 1980 µl assay solution and subjected to visible absorbance measurement using the Shimadzu UV-2400 spectrometer.

Results and discussion

Determination of DDAC with propyl orange

Many of the QAC determination techniques have been improved as a result of the wide use of QACs for bioactive agents and other purposes.[13] In this study, an assay method using propyl orange was investigated to determine DDAC concentration.

The results revealed that propyl orange is useful for determining the concentration of QACs such as distearyldimethylammonium ion[21] and nCx-trimethylammonium ion ($x = 10–18$).[16] However, it had not yet been clarified whether propyl orange is useful for determining DDAC concentration. Therefore, the first step was to investigate the color reaction between propyl orange and DDAC.

The spectra change of the DDAC–propyl orange complex in the assay solution is shown in Fig. 1. Propyl orange has a visible absorbance maximum at 510 nm that decreased with increasing DDAC concentration. In contrast, a new peak appeared at 422 nm. An isosbestic point was observed at about 460 nm. These spectra are similar to that reported for QACs with longer dialkyl side chains. Motomizu et al. reported that propyl orange solution in an acidic buffer shows a large decrease in absorbance near

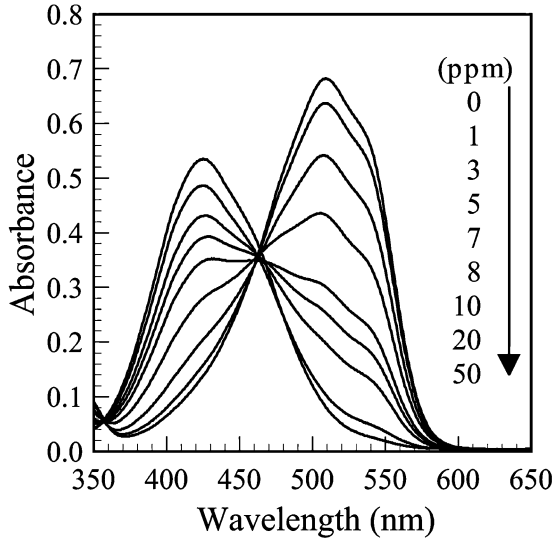

Fig. 1. Visible absorption spectra of didecyldimethylammonium chloride (DDAC)–propyl orange complex in an acidic buffer. DDAC concentration in assay solution varied from 0 to 50 ppm

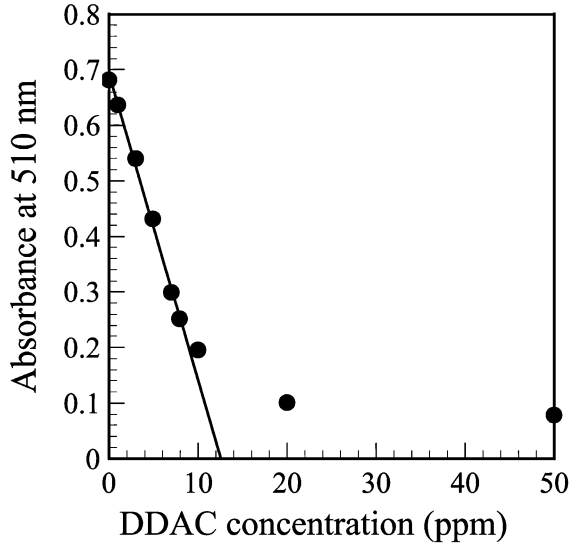

Fig. 2. Relationship between DDAC concentration and absorbance at 510 nm. The *line* indicates the linear least square fit of absorbance at 510 nm against DDAC concentration in assay solution from 0 to 8 ppm drawn by the following equation: Absorbance at 510 nm = −0.055 × DDAC concentration + 0.69

510 nm and an increase in absorbance near 400 nm upon reaction with octadecyltrimethylammonium ion or distearyldimethylammonium ion.[16,21] Our results showed good accordance with those findings.

To investigate the relationship, absorbance at 510 nm was plotted against DDAC concentration (Fig. 2). The absorbance decreased continuously with increasing DDAC concentration from 0 to 20 ppm. A linear correlation in the range lower than 8 ppm indicates that DDAC can be precisely determined by this method when its concentration is 0 to 8 ppm.

Reproducibility of this assay was also investigated. Standard solution containing 0 or 800 ppm DDAC was prepared and DDAC concentration was measured using the assay. As a result of five measurements, coefficients of variation were found to be 1.2% for the solution containing 0 ppm DDAC and 2.0% for that containing 800 ppm DDAC. The low value of variation coefficient indicates that this assay procedure has enough reproducibility for DDAC determination.

Application of propyl orange to determine DDAC retention in treated wood

As already described, propyl orange has proved useful for DDAC assay, especially for concentrations lower than 8 ppm. However, it was still unclear if DDAC determination using propyl orange is useful even in the presence of wood extractives. To estimate the influence of wood extractives on the assay, DDAC standard solution was mixed with wood extractives from four wood species and subjected to the DDAC assay.

DDAC extraction solvent were prepared according to the AWPAS because the extraction solvent described in the JAS for sawn timber contains hydrochloric acid and it was thought to affect the DDAC assay with propyl orange. As the AWPAS defines 0.5 g as the amount of wood powder placed in 20 ml extraction solvent, double this amount of wood powder was used in this experiment and mixed with the same volume of DDAC standard solution. DDAC concentration after mixing both solutions was set to 800 ppm, which is slightly higher than the value expected for treated Japanese cedar timber at the K3 level.[9]

Table 1 shows the visible absorbance at 510 nm in the presence or absence of wood extractives. Absorbance at 510 nm was hardly affected by the presence of wood extractives except in the case of heartwood from Japanese cedar and Hinoki cypress. It was concluded that this assay procedure can determine DDAC retention in the sapwood of Japanese cedar, Hinoki cypress, Japanese larch, and Western hemlock, and in the heartwood of Japanese larch and Western hemlock, within a few percent of experimental error.

To investigate the influence of wood extractives from Japanese cedar and Hinoki cypress heartwood, the absorption spectrum of the DDAC–propyl orange complex was recorded in the presence or absence of wood extractives, and the differential spectrum was calculated from those spectra. As shown in Figs. 3 and 4, in the presence of these wood extractives, absorbance at 510 nm decreased and that at 425 nm increased. In addition to these peak height changes, isosbestic points were observed at around 477 nm. The isosbestic points indicate that absorbance at those wavelengths is not influenced by the wood extractives. Therefore, it is considered that the DDAC assay could be carried out at this wavelength even in the presence of wood extractives from Japanese cedar or Hinoki cypress heartwood. To investigate this hypothesis, absorbance at

Table 1. Effect of wood extractives on didecyldimethylammonium chloride (DDAC) assay at a wavelength of 510 nm (absorbance of control sample at each DDAC concentration = 100)

DDAC conc.[a]	Control	Japanese cedar		Hinoki cypress		Japanese larch		Western hemlock	
		Sapwood	Heartwood	Sapwood	Heartwood	Sapwood	Heartwood	Sapwood	Heartwood
0 ppm	100 (0.5)	100 (0.1)	96.7 (0.0)	100 (0.2)	97.7 (0.1)	100 (0.3)	100 (0.1)	100 (0.3)	101 (0.2)
8 ppm	100 (0.5)	100 (0.5)	85.0 (0.3)	95.8 (0.2)	89.9 (0.1)	96.6 (0.1)	98.1 (0.1)	102 (0.0)	101 (0.1)

Coefficient of variations are shown in parentheses (n = 3)
[a] DDAC concentration in assay solution

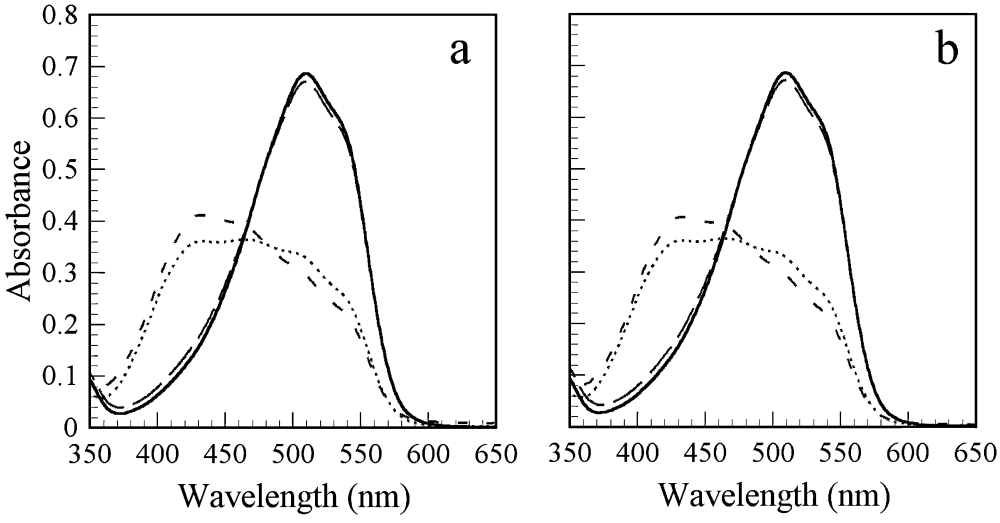

Fig. 3. Absorption spectra in the presence and absence of wood extractives: in Japanese cedar heartwood (**a**); in Hinoki cypress heartwood (**b**). *Unbroken lines*, DDAC 0 ppm without wood extractives; *long-dash lines*, DDAC 0 ppm with wood extractives; *short-dash lines*, DDAC 8 ppm without wood extractives; *dotted lines*, DDAC 8 ppm with wood extractives

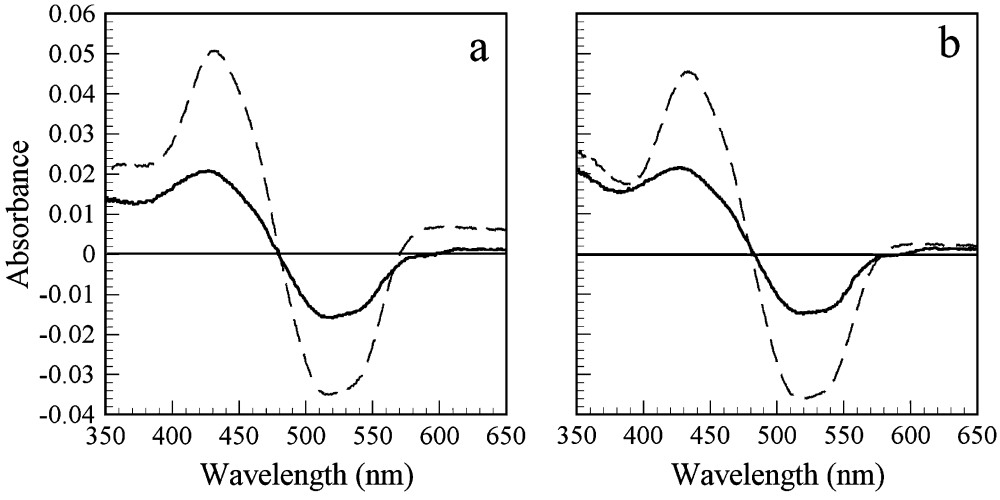

Fig. 4. Differential spectra of DDAC–propyl orange complex in presence and absence of wood extractives. Differences in visible absorption of DDAC–propyl orange complex in presence and absence of wood extractives were plotted: Japanese cedar heartwood (**a**); Hinoki cypress heartwood (**b**). *Unbroken lines*, with DDAC 0 ppm; *dashed lines*, with DDAC 8 ppm

477 nm was plotted against DDAC concentration in both the presence and absence of the wood extractives. The results (Table 2) clearly indicate that visible absorbance at this wavelength was not affected by the presence of wood extractives from heartwood and sapwood of Japanese cedar or Hinoki cypress. Because linear correlation is found between DDAC concentration and the absorbance at 477 nm in the absence of wood extractives (Fig. 5), DDAC concentration can be precisely determined by measuring the visible absorbance at this wavelength even in the presence of wood extractives from these wood species.

It is not certain if the wood extractives from other species would affect the results of this assay. However, those species must be precisely determined after cleanup procedures such as solid-phase extraction.[22] In conclusion, the assay method using propyl orange is a viable alternative to conventional methods such as JAS[9] or AWPAS[10] and has the advantage of delivering simplicity and cost-effectiveness, in harmony with humans and the environment.

Table 2. Effect of wood extractives on DDAC assay at a wavelength of 477 nm (absorbance of control sample at each DDAC concentration = 100)

DDAC conc.[a]	Control	Japanese cedar		Hinoki cypress	
		Sapwood	Heartwood	Sapwood	Heartwood
0 ppm	100 (0.2)	100 (0.1)	99.5 (0.1)	100 (0.2)	97.7 (0.1)
4 ppm	100 (0.2)	100 (0.1)	98.3 (0.2)	98.4 (0.1)	99.2 (0.1)
8 ppm	100 (0.1)	99.9 (0.3)	98.5 (0.3)	99.6 (0.2)	102 (0.3)

Coefficient of variations are shown in parentheses ($n = 3$).
[a] DDAC concentration in assay solution

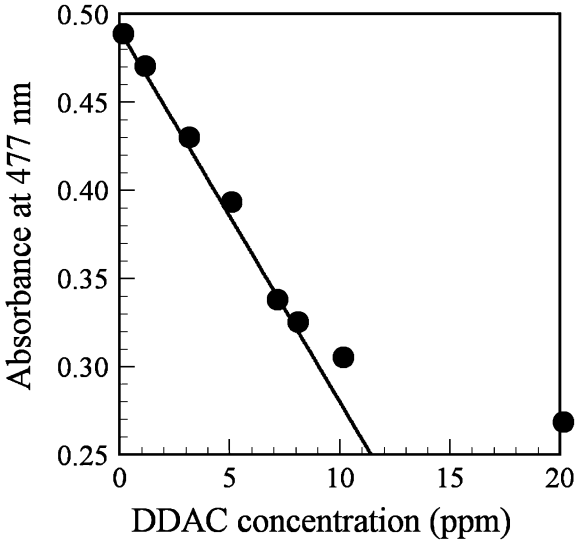

Fig. 5. Relationship between DDAC concentration and absorbance at 477 nm in the absence of wood extractives. The *line* indicates the linear least square fit of absorbance at 477 nm against DDAC concentration in an assay solution from 0 to 8 ppm drawn by the following equation: Absorbance at 477 nm = –0.021 × DDAC concentration + 0.49

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