HETEROCYCLES, Vol. 102, No. 11, 2021, pp. 2168 - 2178. © 2021 The Japan Institute of Heterocyclic Chemistry
Received, 19th August, 2021, Accepted, 1st September, 2021, Published online, 7th September, 2021
DOI: 10.3987/COM-21-14541

DETECTION OF NEW 3,4-DIMETHYLPYRROLE DERIVATIVES UPON THE INCUBATION OF EXOGENOUS AMINES WITH EXTRACT OF ONION (*ALLIUM CEPA*) AND CRUDE ALLIINASE FROM GARLIC (*A. SATIVUM*)

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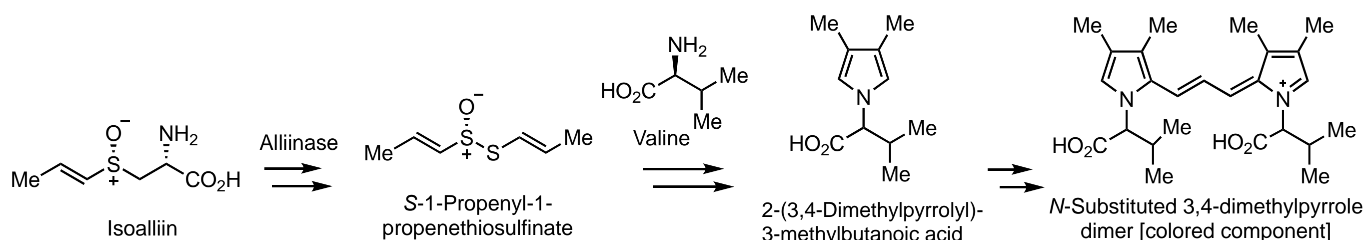
Abstract – Seven new *N*-substituted 3,4-dimethylpyrroles were obtained *in situ* through the reaction of an amine ((L)-phenylalanine, (L)-tryptophan, (L)-leucine, (L)-methionine, 1-phenethylamine, aniline or dodecylamine) and a mixture of unstable sulfur-containing compounds derived from the reaction of amino acids in *Allium cepa* and plant enzyme alliinase. In addition, a plausible reaction mechanism for the synthesis of *N*-substituted 3,4-dimethylpyrroles was displayed.

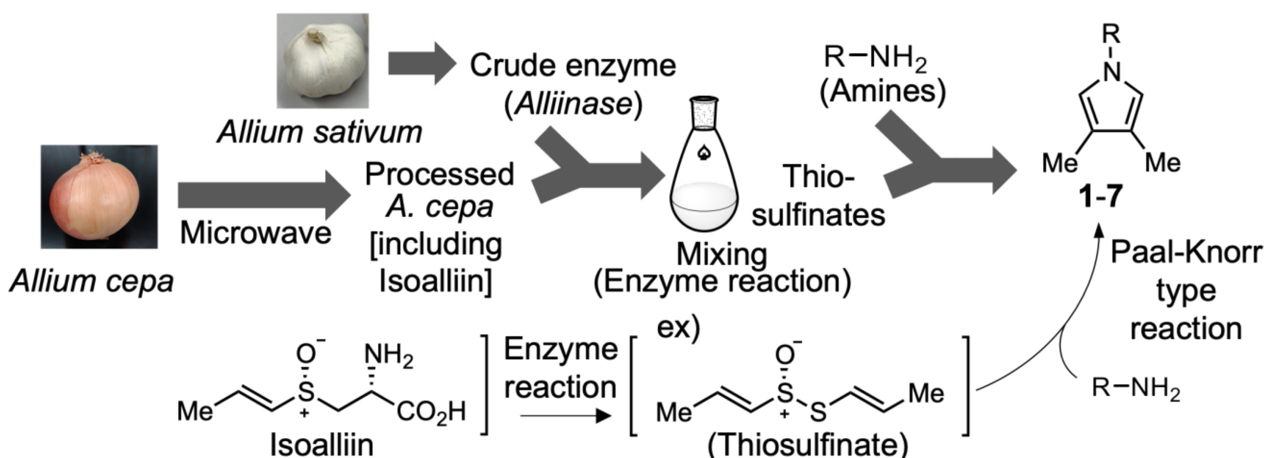
INTRODUCTION

Five-membered heterocycles such as pyrroles are important medicinal seeds.[1-6] For example, 2-formylpyrrole derivatives have several biological activities, such as anti-tuberculosis activity,[1] anti-oxidant activity,[3] and immunopotentiating activity.[5] Recently, we have reported the isolation and synthesis of heterocycles, including pyrroles and pyrrole dimers.[7-14] On the other hand, several *N*-substituted pyrroles are contained in *Allium* plants as colored components. Homogenized onion (bulb of *Allium cepa*) or garlic (bulb of *A. sativum*) is known to form colored components. For example, processed onion turns pink or red. Imai *et al.* and Kubec *et al.* identified *N*-substituted 3,4-dimethylpyrroles, such as 2-(3,4-dimethylpyrrolyl)-3-methylbutanoic acid and 2-(3,4-dimethyl-1*H*-pyrrolyl)propanoic acid, as pigment precursors, and revealed that the colored components are complex *N*-substituted pyrroles and their dimers (Scheme 1).[15-17]



Scheme 1. *N*-Substituted 3,4-dimethylpyrroles from processed onion

The generation process was suggested to be triggered by the alliinase-mediated cleavage of cysteine sulfoxides such as isoalliin, the major component in *Allium* plants, which gave rise to various complex sulfur-containing compounds such as *S*-1-propenyl-1-propenethiosulfinate. Subsequently, some of those sulfur-containing compounds were suggested to react with amino acids in *Allium* plants to give *N*-substituted 3,4-dimethylpyrroles. Inspired by the reports of these biosynthetic processes, we devised a plan to synthesize non-natural *N*-substituted 3,4-dimethylpyrroles *in situ*. We tried to obtain thiosulfinates *in situ* by an alliinase-catalyzed decomposition reaction of cysteine sulfoxides contained in processed *A. cepa*, followed by a [3,3]-sigmatropic rearrangement of thiosulfinates and a nucleophilic reaction with an amine to obtain *N*-substituted 3,4-dimethylpyrroles (Scheme 2). In the present study, we discuss the detection of seven new *N*-substituted 3,4-dimethylpyrroles **1**–**7** (Scheme 2). We also present the chemical elucidation and the estimation of the generation process of **1**–**7**.

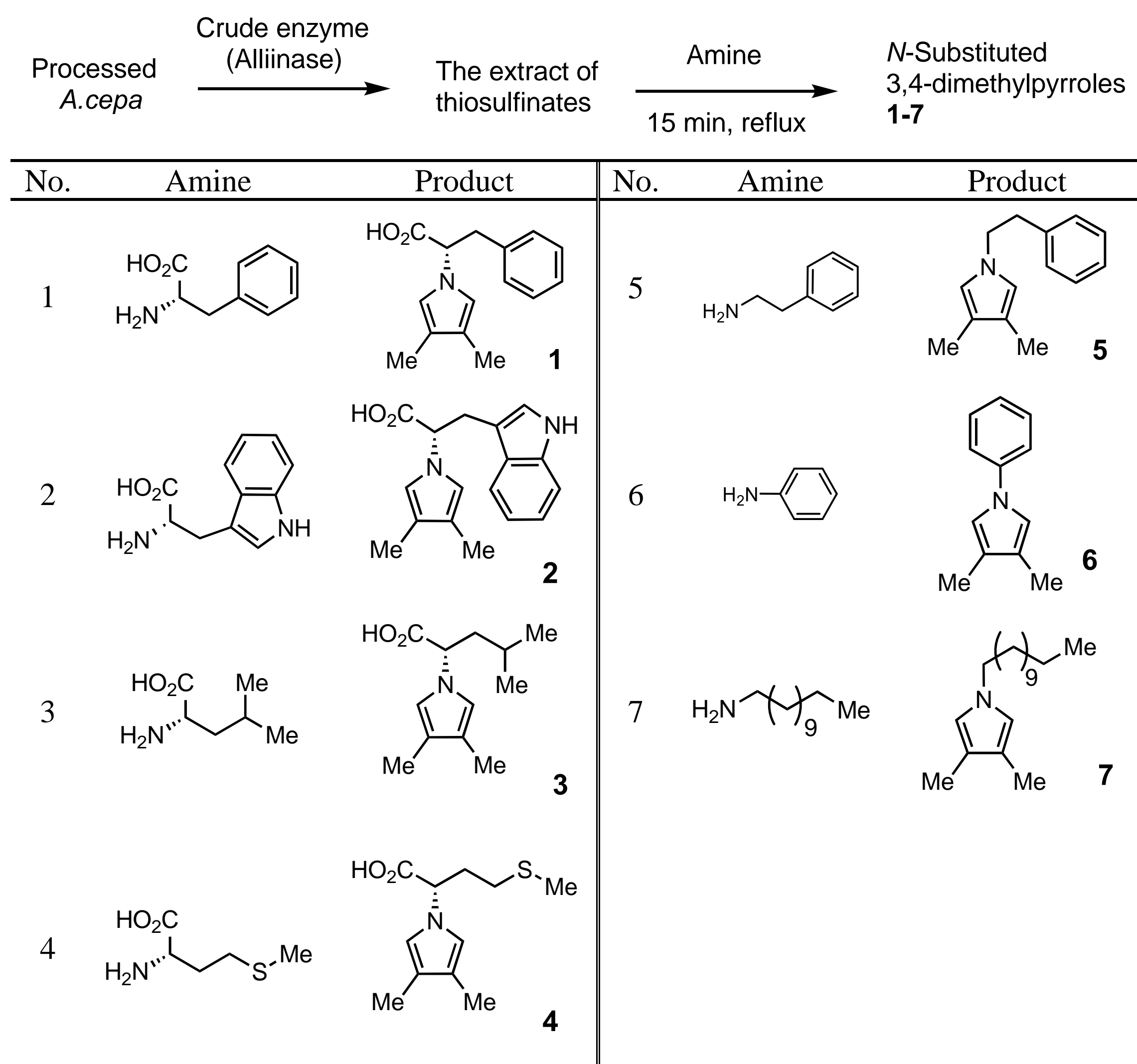


Scheme 2. Construction method of *N*-substituted 3,4-dimethylpyrroles

RESULTS AND DISCUSSION

To synthesize the objective pyrroles, we envisioned harnessing the crude fraction of cysteine sulfoxides from *Allium* plants as the starting material. Cysteine sulfoxide degrading enzyme in *A. cepa* bulb was inactivated by heating in a microwave oven (500 W). A blank experiment using the processed *A. cepa* was carried out. First, the incubation of processed *A. cepa* and crude alliinase, which was obtained from *A.*

sativum bulb, was conducted to determine whether cysteine sulfoxides in the processed *A. cepa* were a good substrate for alliinase. HPLC analysis confirmed that isoalliin, which is one of the cysteine sulfoxides, was consumed by the enzymatic reaction. In addition, the generation of complex compounds by the multi-reactions of thiosulfinates derived from cysteine sulfoxides was also confirmed by HPLC analysis. The enzyme-catalyzed reaction proceeded on a time-series basis, and cysteine sulfoxides in the processed *A. cepa* were confirmed to be a good substrate for the garlic-derived enzyme.

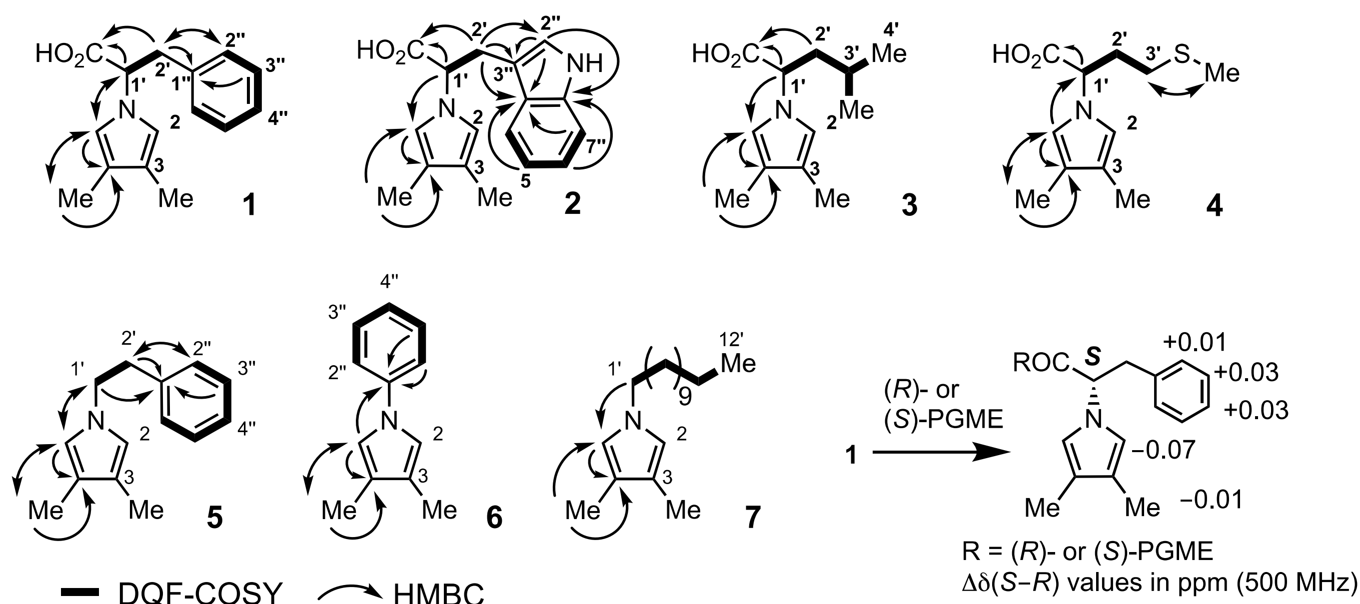


Scheme 3. Detection of *N*-substituted 3,4-dimethylpyrroles **1**–**7** *in situ*

Next, we tried to synthesize *N*-substituted 3,4-dimethylpyrroles from *A. cepa* cysteine sulfoxides by using a biochemical reaction. The processed *A. cepa* and crude alliinase were mixed, and plant pulp was removed by filtration. The filtrate was partitioned in a diethyl ether-H_2O mixture to extract the thiosulfinates into the diethyl ether fraction. The diethyl ether fraction was concentrated *in vacuo*. (L)-Phenylalanine was added immediately into an aqueous solution (potassium phosphate buffer [pH 5.5])

of the concentrated diethyl ether fraction, and the reaction mixture was refluxed at 100 °C for 15 min. As expected, the nucleophilic reaction proceeded, affording new compound **1** (Scheme 3).

Compound **1** was obtained as a colorless amorphous powder, and its molecular formula was determined to be $C_{15}H_{16}NO_2$ by ESIMS analysis and HR-ESIMS analysis. ^{1}H and ^{13}C NMR spectra ($CDCl_3$) indicated that compound **1** contains a pyrrole moiety and a phenylalanine moiety. The position of the phenylalanine moiety was characterized on the basis of an HMBC experiment in which long-range correlations were observed between 1'-H and 2, 5-C (Scheme 4). In addition, the absolute configuration at 1'-position in **1** was confirmed by applying the phenylglycine methyl ester (PGME) method[18] (Scheme 4). Treatment of **1** and (*S*)-phenylglycine methyl ester [(*S*)-PGME] with condensing agents yielded (*S*)-PGME amide (**1a**). On the other hand, (*R*)-PGME amide (**1b**) was obtained from **1** when (*R*)-PGME was used. As shown in Scheme 4, signals assigned to protons of the pyrrole ring in **1a** were observed at a higher field relative to those in **1b** [$\Delta\delta$: negative], whereas signals assigned to protons of the benzene ring in (*S*)-PGME (**1a**) were observed at a lower field relative to those in (*R*)-PGME (**1b**) [$\Delta\delta$: positive]. Thus, the absolute configuration at 1'-position in **1** was confirmed to be *S*, and the absolute configuration of the α-amino acid was found to be retained in this reaction. Consequently, the chemical structure of compound **1** was determined as (*S*)-2-(3,4-dimethyl-1*H*-pyrrol-1-yl)-3-phenylpropanoic acid.



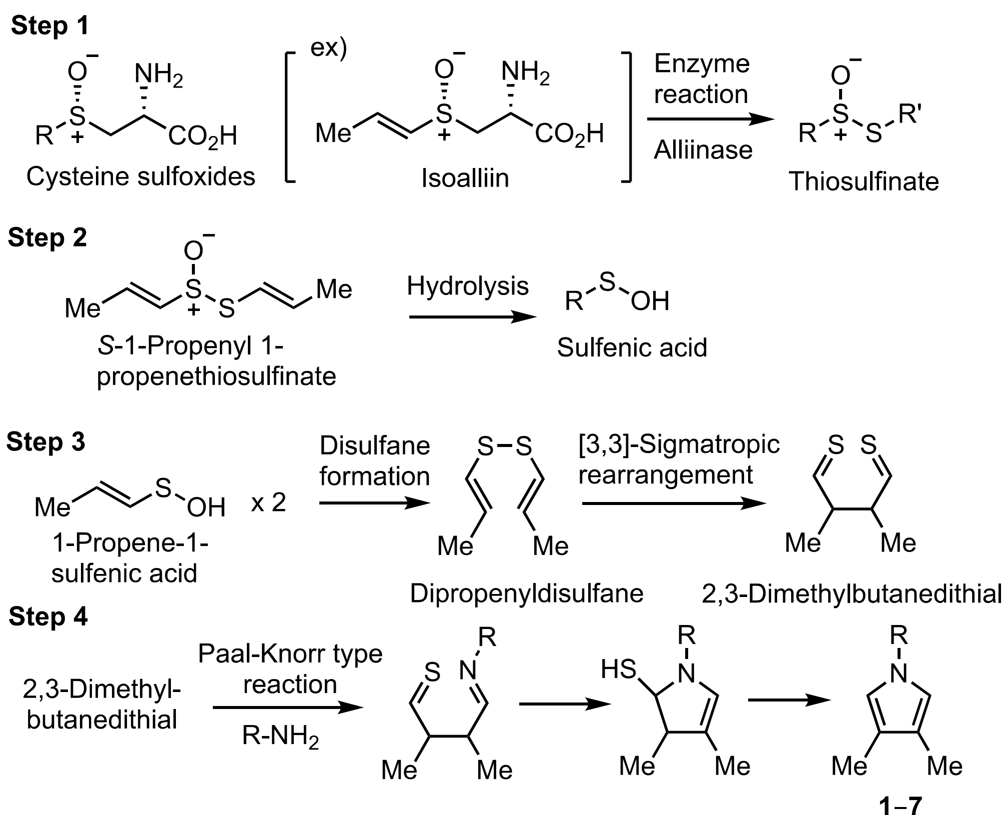
Scheme 4. Key correlations in 2D NMR measurements of compounds **1**–**7** and confirmation of absolute structure of **1**

Furthermore, we tried to synthesize various *N*-substituted 3,4-dimethylpyrroles using (L)-tryptophan, (L)-leucine, or (L)-methionine instead of (L)-phenylalanine as the nucleophilic agent. In the same manner as

the synthesis of **1**, treatment of the diethyl ether fraction containing thiosulfinates with (L)-tryptophan, (L)-leucine, or (L)-methionine produced new compounds **2, 3** and **4**, respectively (Scheme 4). We also tried using primary amines, such as 1-phenethylamine, aniline or dodecylamine, instead of (L)-phenylalanine as the nucleophilic agent. As a result, target compounds **5**–**7** were obtained in similar yields to **1**. The chemical structures of **2**–**7** were determined by detailed analysis of ^{1}H and ^{13}C NMR spectra and 2D NMR (DQF-COSY and HMBC) spectra. Correlations between the pyrrole moiety and the side chain moiety of compounds **2**–**7** were confirmed on the basis of HMBC experiments. In addition, when other solvents, such as H_2O [pH 7.0], acetone, and diethyl ether, were used in this nucleophilic reaction, the yields of compound **5** were equivalent in all cases. These results suggest that the nucleophilicity of the nucleophilic agent and the polarity of the solvent hardly affect the progress of the reaction.

A plausible reaction mechanism for the synthesis of *N*-substituted 3,4-dimethylpyrroles **1**–**7** is shown in Scheme 5. First, thiosulfinates such as *S*-1-propenyl 1-propenethiosulfinate are generated from cysteine sulfoxides such as isoalliin in a reaction catalyzed by alliinase [Step 1]. Second, the thiosulfinates are decomposed into very unstable intermediates such as allylsulfenic acid by hydrolysis [Step 2], whereas some thiosulfinates are transformed into disulfanes. The generated disulfanes are presumed to produce 1,4-dithioketone intermediates by a [3,3]-sigmatropic rearrangement [Step 3]. This can be supported by the fact that the extract and distillates from *Allium* sp. contain di(1-propenyl)disulfane,[19,20] which was converted to 3,4-dimethylthiophene under heating.[21,22] In addition, LCMS analysis of the alliinase treated *A. cepa* extract showed *m/z* 147 (M+H), suggesting the presence of di(1-propenyl)disulfane. Finally, the pyrrole skeleton is formed by the Paal-Knorr type reaction between disulfane and amine [Step 4],[23] and compounds **1**–**7** are obtained.

In conclusion, we have detected seven unnatural *N*-substituted 3,4-dimethylpyrroles **1**–**7** *in situ* by using complex reactions in plant biochemistry and organic chemistry. Although the yields of the objective 3,4-dimethylpyrroles need to be improved, this synthetic strategy using *A. cepa* components and an *A. sativum* enzyme is important not only for the development of medicinal seeds, but also as an example of ‘green and sustainable chemistry’. Further investigations of the construction of various 3,4-dimethylpyrroles and their biological effects should be carried out.



Scheme 5. Proposed synthetic pathway for *N*-substituted 3,4-dimethylpyrroles **1**–**7**

EXPERIMENTAL

General experimental procedures. The following instruments were used to obtain physical data: specific rotation, Horiba SEPA-300 digital polarimeter (l = 5 cm); IR spectra, JASCO FT/IR-4600 Fourier Transform Infrared Spectrometer; ESIMS, Agilent Technologies Quadrupole LC/MS 6130; HR-ESI-MS, SHIMADZU LCMS-IT-TOF; EIMS and HREIMS, JEOL JMS-GCMATE mass spectrometer; ^{1}H NMR spectra, JEOL JNM-ECA 600 (600 MHz) spectrometer; ^{13}C NMR spectra, JEOL JNM-ECA 600 (150 MHz) spectrometer with tetramethylsilane as the internal standard; MPLC, a YMC (Kyoto, Japan) Multiple Preparative HPLC LC-Forte/R. YMC-DispoPack AT SIL-25 (12 g) column was used for analytical and preparative purposes. The following materials were used for chromatography: normal-phase silica gel column chromatography (CC), silica gel BW-200 (Fuji Silysia Chemical, Ltd., 150–350 mesh); reversed-phase silica gel CC, Chromatorex ODS DM1020T (Fuji Silysia Chemical, Ltd., 100–200 mesh); TLC, precoated TLC plates with silica gel 60F_{254} (Merck, 0.25 mm) (normal phase) and silica gel RP-18 F_{254S} (Merck, 0.25 mm) (reversed phase); reversed-phase HPTLC, precoated TLC plates with silica gel RP-18 WF_{254S} (Merck, 0.25 mm). Detection of compounds was achieved by UV irradiation and by spraying with 1% $Ce(SO_4)_2$–10% aqueous H_2SO_4 followed by heating.

Plant material. Fresh yellow onion (*Allium cepa* bulb) and garlic (*A. sativum* bulb) were obtained as commercial products in Kyoto, Japan in 2018–2021. The plants were identified by the author (S. Nakamura).

Measurement of dry weight loss of *A. cepa*.
A flat weighing bottle was dried for 1 h at 110 °C in a drying oven. After drying, it was transferred to a desiccator and allowed to cool for 30 min. After cooling, the weighing bottle was weighed, a portion of *A. cepa* (Weight A: 4.4452 g, 4.2056 g, 4.7513g) was placed in the weighing bottle respectively, and the whole was weighed again. The weighed *A. cepa* sample was heated in a 500 W microwave oven for 30 s and then transferred to a drying oven set at 110 °C for 5 h. After drying, the *A. cepa* sample was placed in a desiccator and allowed to cool for 30 min. After cooling, dry weight loss {(1 – [Weight B] / [Weight A])*100: 91%, 91%, 91%} was determined by measuring the weight (Weight B: 0.4006 g, 0.3736 g, 0.4471 g) of the sample, respectively. From these results, the dry weight of the dry *A. cepa* was calculated by considering the dry weight loss (91%) of fresh *A. cepa*.

Preparation of crude alliinase from *A. sativum*.
Crude alliinase was obtained following the method reported by Imai et al.[15,16] Fresh whole bulbs of *A. sativum* (233.35 g) were homogenized in distilled water (466 mL) by a hand mixer. The pulp was removed by filtration. Then, 1.0 M HCl aqueous solution was added to the filtrate until the pH reached 4.0. The solution was cooled at 4 °C for 1 h. The precipitate was collected by centrifugation (3000 *g*, 5 min, 4 °C) and redissolved in 0.05 M potassium phosphate buffer (60 mL, pH 6.5) containing 10% glycerol and 20 μM pyridoxal-5'-phosphate.

Synthesis of pyrrole derivative 1 from *A. cepa*.
Whole fresh bulbs of *A. cepa* (276.68 g) were heated in a 500 W microwave oven (6 min). [Heating time was 1 min for 50 g of bulbs]. The processed *A. cepa* was homogenized with crude alliinase (10 mL) by a hand mixer and extracted with Et_2O (150 mL). The pulp was removed by filtration. The filtrate was extracted with Et_2O (70 mL × 2). The Et_2O extract was dried over Na_2SO_4, filtered, and concentrated *in vacuo*. Phenylalanine (484.3 mg, 3 mmol, excess amount) was added to the extract, and the reaction mixture was refluxed in potassium phosphate buffer (pH 5.5, 20 mL) at 100 °C for 15 min. The reaction mixture was purified by normal phase medium pressure liquid chromatography {MPLC, mobile phase: EtOAc-MeOH (100:0→0:100 gradient 20 min, v/v) [YMC-DispoPackAT SIL-25 (12 g)]} to obtain pyrrole derivative **1** (5.4 mg, 0.022 mmol).

(*S*)-2-(3,4-Dimethyl-1*H*-pyrrol-1-yl)-3-phenylpropanoic acid (1): 5.4 mg from fresh *A. cepa* (276.68 g), dry *A. cepa* (24.90 g), pale yellow amorphous powder; ^{1}H NMR (500 MHz, $CDCl_3$) δ_H and ^{13}C NMR (125 MHz, $CDCl_3$) δ_C: see Table 2; IR (ATR) cm^{-1}: 1674, 1448, 1417; HR-ESI-MS *m/z*: 242.1187 $[M-H]^-$ (Calcd for $C_{15}H_{16}NO_2$: 242.1189); ESI-MS *m/z*: 282 $[M+K]^+$, 266 $[M+Na]^+$, 244 $[M+H]^+$. $[\alpha]_D^{21}$ −49.3 (*c* = 1.0, MeOH).

(*S*)-2-(3,4-Dimethyl-1*H*-pyrrol-1-yl)-3-(1*H*-indol-3-yl)propanoic acid (2): 18.7 mg from fresh *A. cepa* (275.11 g), dry *A. cepa* (24.76 g), blue amorphous powder; ^{1}H NMR (400 MHz, $CDCl_3$) δ_H and ^{13}C NMR (100 MHz, $CDCl_3$) δ_C: see Table 2; IR (ATR) cm^{-1}: 1646, 1449, 1418; HR-ESI-MS *m/z*: 283.1441 $[M+H]^+$ (Calcd for $C_{17}H_{19}N_2O_2$: 283.1444); ESI-MS *m/z*: 321 $[M+K]^+$, 305 $[M+Na]^+$, 283 $[M+H]^+$. $[\alpha]_D^{21}$ -3.0 (*c* = 1.0, acetone).

Table 1. ^{1}H NMR and ^{13}C NMR (**1**, **2**, **4**: $CDCl_3$, **3**: CD_3OD) data for compounds **1**–**4**

	1		**2**		**3**		**4**	
Position	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C	δ_H	δ_C
2,5	6.30 (1H, s)	118.3	6.46 (1H, s)	117.9	6.41 (1H, s)	118.4	6.36 (1H, s)	118.1
3,4		117.7		118.1		116.2		117.7
3,4-Me	1.91 (3H, s)	10.0	1.94 (3H, s)	10.1	1.92 (3H, s)	10.2	1.96 (3H, s)	11.2
1'	4.42 (1H, dd, *J* = 10.0, 4.5)	65.1	4.59 (1H, t-like, *J* = 6.8)	62.9	4.48 (1H, dd, *J* = 10.6, 5.3)	61.1	4.49 (1H, dd-like, *J* = 7.5)	61.5
2'	3.12 (1H, dd, *J* = 14.5, 10.0) 3.39 (1H, dd, *J* = 14.5, 4.5)	39.3	3.24 (1H, m) 3.54 (1H, m)	28.6	1.76 (2H, m)	42.6	2.10 (1H, m) 2.38 (1H, m)	31.9
3'					1.58 (1H, m)	25.8	2.20 (1H, m) 2.38 (1H, m)	30.8
4'					0.86 (3H, d, *J* = 6.7)	23.3	0.86 (3H, d, *J* = 6.7)	22.0
1'-CO_2H		176.2		175.4		175.6		176.3
1"		138.1						
2",6"	6.97 (1H, d, *J* = 6.5)	128.8						
3",5"	7.19 (1H, m)	128.3						
2"			6.61 (1H, s)	127.1				
3"				110.5				
3"a				123.0				
4"			7.52 (1H, d, *J* = 7.6)	118.2				
5"	7.19 (1H, m)	128.3	7.06 (1H, t, *J* = 7.6)	119.2				
6"	6.97 (1H, d, *J* = 6.5)	128.8	7.12 (1H, t, *J* = 7.6)	121.8				
7"			7.25 (1H, d, *J* = 7.6)	111.2				
7"a				135.8				
3'-Me					0.90 (3H, *J* = 6.7)	21.8		
S-Me							2.06 (3H, s)	15.3

(*S*)-2-(3,4-Dimethyl-1*H*-pyrrol-1-yl)-4-methylpentanoic acid (3): 27.3 mg from fresh *A. cepa* (279.97 g), dry *A. cepa* (25.20 g), colorless amorphous powder; ^{1}H NMR (400 MHz, CD_3OD) and ^{13}C NMR (100 MHz, CD_3OD) δ_C: see Table 2; IR (ATR) cm^{-1}: 1714, 1651, 1600, 1513, 1503, 1463, 1449, 1392; HR-EIMS *m/z*: 209.1413 [M] (Calcd for $C_{12}H_{19}NO_2$: 209.1416); EIMS *m/z*: 209 [M]. $[\alpha]_D^{21}$ +17.0 (*c* = 1.0, MeOH).

(*S*)-2-(3,4-Dimethyl-1*H*-pyrrol-1-yl)-4-(methylthio)butanoic acid (4): 14.1 mg from fresh *A. cepa* (241.54 g), dry *A. cepa* (21.74 g), yellow amorphous powder; ^{1}H NMR (500 MHz, $CDCl_3$) δ_H and ^{13}C NMR (125 MHz, $CDCl_3$) δ_C: see Table 2; IR (ATR) cm^{-1}: 1645, 1448, 1397, 1363; HR-ESI-MS *m/z*: 226.0907 [M-H]$^-$ (Calcd for $C_{11}H_{16}NO_2S$: 226.0909); ESI-MS *m/z*: 226 [M-H]$^-$. $[\alpha]_D^{21}$ -18.6 (*c* = 1.0, acetone).

3,4-Dimethyl-1-phenethyl-1*H*-pyrrole (5): 13.0 mg from fresh *A. cepa* (303.12 g), dry *A. cepa* (26.92 g), colorless amorphous powder; ^{1}H NMR (400 MHz, $CDCl_3$) δ_H and ^{13}C NMR (100 MHz, $CDCl_3$) δ_C: see Table 1; IR (ATR) cm^{-1}: 1603, 1535, 1496, 1454; HR-EIMS *m/z*: 119.1360 [M] (Calcd for $C_{14}H_{17}N$: 119.1361); EIMS *m/z*: 199 [M].

3,4-Dimethyl-1-phenyl-1*H*-pyrrole (6): 9.6 mg from fresh *A. cepa* (245.0 g), dry *A. cepa* (22.05 g), colorless amorphous powder; ^{1}H NMR (600 MHz, $CDCl_3$) δ_H and ^{13}C NMR (150 MHz, $CDCl_3$) δ_C: see Table 1; IR (ATR) cm^{-1}: 1601, 1529, 1499, 1443, 1399; HR-EIMS *m/z*: 171.1050 [M] (Calcd for $C_{12}H_{13}N$: 171.1048); EIMS *m/z*: 171 [M].

Table 2. ^{1}H NMR and ^{13}C NMR ($CDCl_3$) data for compounds **5**–**7**

5			**6**			**7**		
Position	δ_H	δ_C	Position	δ_H	δ_C	Position	δ_H	δ_C
2	6.36 (1H, s)	118.4	2	6.84 (1H, s)	116.7	2	6.35 (1H, s)	118.3
3,4		117.9	3,4		120.8	3,4		117.5
5	6.36 (1H, s)	118.4	5	6.84 (1H, s)	116.7	5	6.35 (1H, s)	118.3
3,4-Me	1.99 (3H, s)	10.1	3,4-Me	2.08 (3H, s)	10.2	3,4-Me	2.00 (3H, s)	10.0
1'	3.95 (2H, t, *J* = 7.9)	51.0	1'		140.7	1'	3.71 (2H, t, *J* = 7.2)	49.3
2'	3.00 (2H, t, *J* = 7.9)	38.6	2',6'	7.31 (1H, d, *J* = 7.6)	119.5	2'	1.69 (2H, m)	31.7
1"		138.8	3',5'	7.37 (1H, t, *J* = 7.6)	129.4	3'-9'	1.25 (2H, m)	29.7
2",6"	7.13 (1H, d-like, *J* = 7.4)	128.8	4'	7.26 (1H, t, *J* = 7.6)	124.6	10'	1.25 (2H, m)	31.9
3",5"	7.29 (1H, t-like, *J* = 7.4)	128.6				11'	1.25 (2H, m)	22.7
4"	7.22 (1H, t-like, *J* = 7.4)	126.6				12'	0.88 (3H, t, *J* = 7.2)	14.1

1-Dodecyl-3,4-dimethyl-1*H*-pyrrole (7): 8.8 mg from fresh *A. cepa* (299.12 g), dry *A. cepa* (27.28 g), colorless amorphous powder; ^{1}H NMR (600 MHz, $CDCl_3$) δ_H and ^{13}C NMR (150 MHz, $CDCl_3$) δ_C: see Table 1; IR (ATR) cm^{-1}: 1462, 1399; HR-EIMS *m/z*: 263.2614 [M] (Calcd for $C_{18}H_{33}N$: 263.2613); EIMS *m/z*: 263 [M].

Preparation of (*S*)- and (*R*)-phenylglycine methyl ester (PGME) amides 1a and 1b

To a solution of compound **1** (2.2 mg, 0.00090 mmol) in DMF (0.5 mL) were added (*S*)-PGME (2.2 mg, 0.011 mmol), PyBOP® (7.1 mg, 0.014 mmol), 1-hydroxybenzotriazole (HOBT, 2.1 mg, 0.014 mmol), and *N*-methylmorpholine (3.1 μL, 0.033 mmol) at 0 °C. After the reaction mixture was stirred at room temperature for 3 h, EtOAc (20 mL) was added, and the resulting diluted solution was washed with 5% HCl aqueous solution, saturated sodium dicarbonate solution, and brine. The organic phase was dried (Na_2SO_4) and concentrated to give (*S*)-PGME amide **1a**. (*R*)-PGME amide **1b** was also obtained using the same method.

(*S*)-PGME amide **1a**: ^{1}H NMR ($CDCl_3$) δ: 1.97 (6H, s, 3-CH_3, 4-CH_3) , 3.13 (1H, dd-like, J = 14.5, 10.0, H-2'), 3.56 (1H, dd, *J* = 14.5, 5.0, H-2'), 4.58 (1H, dd, *J* = 10.0, 5.0, H-1'), 6.38 (2H, s, H-2, H-5), 7.05 (2H, d-like, *J* = 7.0, H-2'', H-6''), 7.20 (1H, t-like, *J* = 7.0, H-4''), 7.21 (2H, t-like, *J* = 7.0, H-3'', H-5'').

(*R*)-PGME amide **1b**: ^{1}H NMR ($CDCl_3$) δ: 1.98 (6H, s, 3-CH_3, 4-CH_3) , 3.18 (1H, dd-like, *J* = 14.5, 10.0, H-2'), 3.58 (1H, dd, *J* = 14.5, 5.0, H-2'), 4.53 (1H, dd, *J* = 10.0, 5.0, H-1'), 6.45 (2H, s, H-2, H-5), 7.04 (2H, d-like, *J* = 7.0, H-2'', H-6''), 7.16 (1H, t-like, *J* = 7.0, H-4''), 7.18 (2H, t-like, *J* = 7.0, H-3'', H-5'').

ACKNOWLEDGEMENT

This work was supported by JSPS KAKENHI grant number 20K07109 (to S. Nakamura).

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