combined amino acid.[6]

After completion of the manuscript, it was learned that Virtanen and Mattila[7] had isolated the same peptide from garlic.

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Metabolism of S-(2-Carboxypropyl)glutathione in Rabbit

Isolation of S-(2-carboxypropyl)glutathione (I) from garlic was reported in previous paper.[1] The constituent sulfur-containing amino acid, S-(2-carboxypropyl)cysteine (II), was also found in human urine by Mizuhara and Oomori[2] independently almost at the same time. Recently, the present author[3] found an active incorporation of valine-[^{14}C] into (I) in excised root of garlic and presumed a close metabolic relation between S-2-carboxypropyl group in (I) and intermediary metabolite of valine such as methacrylic acid or the coenzyme-A derivative.

As suggested by Bray and Franklin,[4] S-substituted N-acetylcysteine, mercapturic acid excreted by animal, may be formed by the conjugation of an intermediary metabolite of substrate administered with thiol group in glutathione, giving a compound of type (I) and followed by the splitting off of peptide bonds and by N-acetylation. These facts prompted examination of mammalian metabolism of (I). Its results clearly indicated that formation of (II) and N-acetyl-S-(2-carboxypropyl)cysteine (III) from (I) *in vivo.* (III) thus formed and isolated in crystalline state is a new mercapturic acid which has not previously been reported, as far as is known.

A female rabbit, weighing 1.85kg., was injected intravenously with 185 mg. of (I). At various intervals after the administration of (I), specimens of the urine were collected by catheterization during a period of 30 hours. (II) and (III) in urine were separated from each other by passing through a column of Amberlite IR-120 (H form). Both fractions, the unadsorbed filtrate and the 4% NH_4OH eluate, were separately evaporated to dryness and a part of the resulting residues was subjected to two dimensional paper chromatography with the solvent systems of (a) BuOH-AcOH-H_2O (5:1:4) and (b) PhOH-0.08% NH_4OH (4:1). The unadsorbed fraction contained a Ninhydrin-negative and chloroplatinate-positive component, the Rf values of which were quite identical with those of synthesized sample of (III): Rf 0.77 (a) and 0.53 (b). The ammonia-eluate fraction also contained another chloroplatinate-positive component which colored grayish blue to Ninhydrin. The Rf values were just the same as those of authentic (II): Rf 0.36 (a) and 0.41 (b).

As shown in Table I, spots corresponding to (II) and (III) appeared only in the urine collected during the first 9 hours after administration of (I), and (II) was always dominant. As a control the urine before the administration of (I) was analyzed by the same method.

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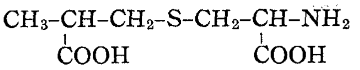
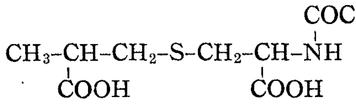
Table I. Urinary Metabolites after Administration of S-(2-Carboxypropyl)glutathione

Compound	Time after injection (hrs.)				
	Control	0∼4	4∼9	9∼24	24∼30
Fraction adsorbed on Amberlite IR-120					
 $CH_3\text{-}\underset{COOH}{CH}\text{-}CH_2\text{-}S\text{-}CH_2\text{-}\underset{COOH}{CH}\text{-}NH_2$	−	++	++	−	−
Fraction not adsorbed on Amberlite IR-120					
 $CH_3\text{-}\underset{COOH}{CH}\text{-}CH_2\text{-}S\text{-}CH_2\text{-}\underset{COOH}{CH}\text{-}\overset{COCH_3}{NH}$	−	+	+	−	−

To isolate both the metabolites in a crystalline form, main part of each fraction obtained from the urine during the first 9 hours was treated with decolorizing charcoal to remove colored impurities and the resulting colorless solutions, after being evaporated to a small volume, were submited to paper chromatography in preparative scale, with the solvent system (a). By eluting each zone with water and recrystallizing the residual substance of the eluate from hydrous ethanol, 20 mg. of (II) and 5 mg. of (III) were obtained as fine needles from 80 ml. of the urine. Rf values of crystalline (II) and (III) so isolated were quite identical with those of synthesized samples of (II) and (III). On hydrolysis with 6*N* HCl, (III) gave (II), which in turn gave alanine and isobutyric acid on desulfurization with Raney-Ni. From these facts it is certain that (II) is S-(2-carboxypropyl)-cysteine and (III) is N-acetyl-S-(2-carboxypropyl)cysteine.

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N-Oxidation of 3-Aminopyridazine Derivatives

Following synthetic studies on 3,6-disubstituted 4-nitropyridazine 1-oxides,[1] N-oxidation of 3-aminopyridazine (Ia) and its derivatives was examined.

First, (Ia) and 3-acetamidopyridazine[2] (Ib) were oxidized with ether solution of monoperphthalic acid in a usual way. (Ia) gave no crystalline product but formed a resinous substance. From (Ib), 3-acetamidopyridazine 2-oxide (IIb) was obtained as colorless needles, m.p. 199∼201°, and its 1-oxide (IIIb) as colorless needles, m.p. 259° (decomp.), in 82% and 2% yields respectively (*Anal.* Calcd. for $C_6H_7O_2N_3$: C, 47.06; H, 4.60; N, 27.44. Found (for (IIb)) : C, 47.00; H, 4.24; N, 27.73. Found (for (IIIb)) : C, 47.27; H, 4.83; N,

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