

**2-Methyl-3-*o*-hydroxymethylphenyl-4(3*H*)-quinazolinone, a Metabolite
of 2-Methyl-3-*o*-tolyl-4(3*H*)-quinazolinone in Rabbit**

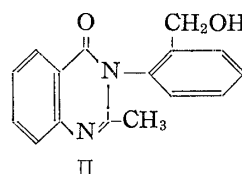
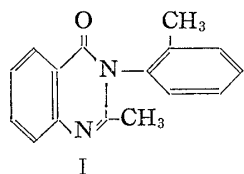
In the previous paper¹⁾ the physiological disposition and the metabolic fate of 2-methyl-3-*o*-tolyl-4(3*H*)-quinazolinone (MTQ) (I) have been reported. In the present communication the authors isolated a major MTQ metabolite (II) from the urine of rabbits each of which received a single oral dose (200 mg./kg. body weight) of MTQ and established the structure of II as 2-methyl-3-*o*-hydroxymethylphenyl-4(3*H*)-quinazolinone.

II was isolated as needles from the β -glucuronidase treated urine by continuous extraction with ether, followed by silica gel chromatography, m.p.*¹ 109~110° (from dil. methanol) (*Anal.* Calcd. for C₁₆H₁₄O₂N₂: C, 72.16; H, 5.30; N, 10.52. Found: C, 71.87; H, 5.59; N, 10.56). The structure of II was shown to be 2-methyl-3-*o*-hydroxymethylphenyl-4(3*H*)-quinazolinone based upon the following evidences:

(1) The presence of a quinazolinone ring in II was shown by ultraviolet spectrum similar to that of I in hydrochloric acid, sodium hydroxide and ethanol solutions [I; UV $\lambda_{\max}^{0.1N\text{HCl}}$ m μ (log ϵ): 234 (4.52), 270 (3.90). II; UV $\lambda_{\max}^{0.1N\text{HCl}}$ m μ (log ϵ): 234 (4.54), 274 (3.91)], and infrared spectrum (IR $\nu_{\max}^{\text{Nujol}}$ cm⁻¹: 1690, 1610, 1600), and positive Dragendorff test, negative Ehrlich test and no fluorescence.

(2) The presence of an alcoholic hydroxyl in II was identified by infrared spectrum (IR: $\nu_{\max}^{\text{Nujol}}$ 3300 cm⁻¹), nuclear magnetic resonance spectrum*² [I: 7.86, 7.78 (-CH₃), II: 7.77 (-CH₃), 6.73 (-OH), 5.55 (-CH₂-)], and positive Nessler test, negative phenol test and insolubility in dil. sodium hydroxide solution.

(3) Alkaline degradation products of II gave anthranilic acid (III) and *o*-aminobenzyl alcohol (IV). II was hydrolyzed in sodium hydroxide solution and the degradation products were characterized as III and IV by paper chromatography with the solvent systems of (a) BuOH-AcOH-H₂O (4:1:5), (b) BuOH-NH₄OH-H₂O (4:1:3), (c) EtOH-NH₄OH-H₂O (4:2:3) and (d) MeOH-AmOH-benzene-2*N* HCl (14:7:14:5). Two spots were detected with diazotized *p*-nitroaniline, Ehrlich reagents and fluorescence, and their R_f values were just the same as those of the authentic samples of III [(a) 0.85, (b) 0.23, (c) 0.74 and (d) 0.60] and IV [(a) 0.79, (b) 0.80, (c) 0.82 and (d) 0.22]. A part of the degradation products of II was subjected to steam distillation, and the distillate was extracted with ether, and the ultraviolet spectrum of dil. hydrochloric acid extract from the ether layer was similar to that of an authentic IV [UV $\lambda_{\max}^{\text{HCl}}$ m μ : 218, 257, 266 (shoulder)], whereas the spectrum from the degradation products of I was similar to that of *o*-toluidine (UV $\lambda_{\max}^{\text{HCl}}$ m μ : 218, 261, 268).



*¹ Melting point was determined on Kofler-Block "Leitz Microscope Heating Stage 350" and is uncorrected.

*² Nuclear magnetic resonance spectra (Japan Electron Optics Lab. Co., Ltd. 3H-60 NMR spectrometer) were measured at 60 Mc./sec. for CHCl₃ solution and calibrated against internal Me₄Si. Chemical shifts are given in τ -values.

1) M. Akagi, Y. Oketani, M. Takada, T. Suga: This Bulletin, 11, 321 (1963).

The authors would like to express their deep gratitude Mr. S. Shimokawa (Material Science Lab., Hokkaido University) for measurement of nuclear magnetic resonance spectra.

Faculty of Pharmaceutical Sciences,
School of Medicine, Hokkaido University,
Nishi-5-chome, Kita-12-jo, Sapporo.

Masuo Akagi (赤木満洲雄)
Yoneshiro Oketani (桶谷米四郎)
Sumitaka Yamane (山根純卓)

Received June 20, 1963

[Chem. Pharm. Bull.]
11 (9) 1217 ~ 1218

UDC 582.736 : 547.586.2

Isolation of 2-Phenylacetamide, Indole-3-acetamide, and Indole-3-carboxaldehyde from Etiolated Seedling of *Phaseolus*

Indole-3-acetic acid has been recognized as a naturally occurring plant growth hormone and a few other plant growth substances, such as indole-3-acetonitrile, ethyl indole-3-acetate, and their related compounds such as indole-3-carboxaldehyde and indole-3-carboxylic acid, have been obtained in a crystalline form.

Although many papers have appeared reporting the presence of other growth promoting substances in plants, evidence of their presence had only been proved by paper chromatography.

One of the present authors, Isogai¹⁾ had reported the presence of the several growth controlling substances in neutral and acidic fractions of aqueous extract of the whole plant of Moyashi.*¹ Now some of the active substances contained in the neutral fraction have been isolated in crystalline form.

Moyashi (900 kg. of fresh weight) was extracted with water at 56° and the extract was treated with charcoal. The active material adsorbed on charcoal was eluted with ammoniacal methanol. The solvent was evaporated from the eluate *in vacuo* and the residue was separated into neutral and acidic fractions, affording 24 g. of active neutral fraction.

This neutral fraction was submitted to column chromatography over activated alumina and at present four kinds of crystals were obtained, two of which being active as growth promoter and the other two inactive by means of Avena straight growth test. The two active crystals were identified as indole-3-acetamide, m.p. 151°, *Anal.* Calcd. for C₁₀H₁₀ON₂: C, 68.95; H, 5.79; N, 16.08. Found: C, 68.95; H, 5.89; N, 15.79, (yield, 600 mg.) and 2-phenylacetamide, m.p. 156°, *Anal.* Calcd. for C₈H₉ON: C, 71.09; H, 6.71; N, 10.36; O, 11.84. Found: C, 71.16; H, 6.71; N, 9.81; O, 11.58, (yield, 90 mg.) and one of the inactive crystals as indole-3-carboxaldehyde, m.p. 189~190°, *Anal.* Calcd. for C₉H₇ON: C, 74.47; H, 4.86; N, 9.65. Found: C, 74.59; H, 4.86; N, 9.63, (yield 10 mg.).

These products were identified by admixture with authentic samples and also by comparing their infrared and ultraviolet spectra.

The studies on the unidentified active substances and the mode of action of indole-3-acetamide and 2-phenylacetamide are now in progress.

*¹ Japanese name "Moyashi" is given for etiolated seedlings of *Phaseolus mungo* Linne, sprouted at factory and commercially sold in greengrocers as a sort of vegetable in Japan.

1) Y. Isogai: Sci. Pap. Coll. Gen. Educ., Univ. Tokyo, 10, 73 (1960).