After removal of the catalyst by filtration, 3 cc. of Ac_2O was added to the filtrate and the mixture was refluxed for 1.5 hrs. This reaction mixture was poured into water, decolorized, and neutralized with $NaHCO_3$, from which 0.2 g. of crystals were obtained. Recrystallization from MeOH-EtOH afforded colorless needles, m.p. 205~206°, of 1-methyl-5-acetamino-6-acetyloxyindole (Ⅶ). *Anal.* Calcd. for $C_{13}H_{14}O_3N_2$: C, 63.42; H, 5.69; N, 11.39. Found : C. 63.90; H, 5.52; N, 11.15.

(Ⅶ) was formed even on the use of optically active oxime. (Ⅶ) is not optically active.

iv) A mixture of 0.2 g. of (Ⅶ) obtained in iii) and 1 cc. of Ac_2O was refluxed for 1.5 hrs., cooled, and the mixture was poured into ice water to effect decomposition. The crude crystals that separated out were collected by suctional filtration and recrystallized from MeOH to 0.2 g. of scaly crystals, m.p. 169~171°, showing no depression on admixture with (Ⅵ) obtained in ii), proving it to be 1-methyl-3-acetyl-5-acetamino-6-acetoxyindole.

Summary

Adrenochrome possesses >C=O group in the 5- and 6-positions but no synthetic evidence has been offered on which of these >C=O groups had reacted with semicarbazide to form adrenochrome monosemicarbazone. Catalytic reduction of the monosemicarbazone with palladium-carbon catalyst results in absorption of 1 mole of hydrogen to form 1-methyl-5-semicarbazido-6-hydroxyindole (Ⅱ), whose methylation with dimethyl sulfate and sodium hydroxide affords 1-methyl-5-semicarbazido-6-methoxyindole (Ⅲ). Pyrolysis of (Ⅲ) by heating with sodium carbonate in glycerol at 185~195° ended in liberation of nitrogen and 1-methyl-6-methoxyindole (Ⅳ) was formed. On the other hand, methylation of 6-methoxyindole (Ⅴ) with methyl iodide in liquid ammonia with metallic sodium yielded (Ⅳ). This has proved that adrenochrome monosemicarbazone is 1-methyl-3-hydroxy-5-semicarbazono-6-oxo-2, 3, 5, 6-tetrahydroindole.

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47. **Jun-ichi Iwao** : Studies on Adrenochrome Derivatives. V.[1] Solubilization of Adrenochrome Monosemicarbazone and Its Derivatives. (1). Synthesis of 4β-Hydroxyethylsemicarbazones.

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It has already been shown that adrenochrome monosemicarbazone (I) is sparingly soluble in solvents, its solubility in water being 0.3 mg./cc. at 5° and 0.4 mg./cc. at 20°. The excellent hemostatic action of adrenochrome had been reported by Derouaux[2] but its actual medicinal usage had seemed difficult due to its instability. The more stable (I) is also known to possess excellent hemostatic effect but due to its difficulty of dissolving in water, preparation of higher concentrations had been impossible. In 1948, a patent was taken out for the molecular compound of sodium salicylate and (I),[3] and its specification revealed the fact that a mixture of 25 : 1 of salicylic acid and (I) dissolved in water up to 25 mg./cc. The Belgian Patent[4] seems to use sodium benzoate as the solubilization agent but no details are known.

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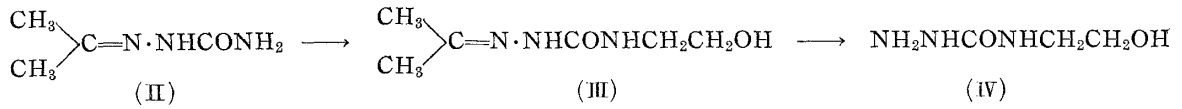
1) Part Ⅳ : J. Pharm. Soc. Japan, **76**, 814(1956)
2) C. Derouaux : Compt. rend. soc. biol., **131**, 830(1939)(C. A., **33**, 7887(1939)).
3) U. S. Pat. 2,581,850 (C. A., **46**, 2759(1952)).
4) Belgian Pat. 525,542 (through C. A., **48**, 14132(1954)).
5) a) Part Ⅱ. J. Pharm. Soc. Japan, **76**, 808(1956). b) Part Ⅲ. *Ibid.*, **76**, 811(1956).

The adrenochrome analogs of 5,6-dioxo-2,3,5,6-tetrahydroindole type,[5a] *o*-benzoquinone type,[5b] and 6,7-dioxo-1,2,3,4,6,7-hexahydroquinoline type[1] were synthesized in order to examine the relationship between the chemical structure and pharmacological effect of adrenochrome and to obtain water-soluble compounds. However, all the compounds were found to be just as or more sparingly soluble than adrenochrome monosemicarbazone so that their pharmacological effect was not examined. Sobotka[6] prepared a water-soluble, betaine-type hydrazone by the application of the Girard reagent to adrenochrome but the compound could not be used due to its instability. The other Belgian Patent[7] describes a stable, soluble adrenochrome derivatives but they do not seem to have been put to actual usage.

Urea is generally used as the solubilizing agent but it is ineffective with adrenochrome monosemicarbazone. β-Hydroxyethylurea, possessing a hydrophilic $HOCH_2CH_2$- group was also ineffective. However, when 4-β-hydroxyethylsemicarbazide, hitherto unknown substance, was prepared and condensed with adrenochrome to introduce β-hydroxyethylurea group into the molecule, the solubility of the resultant compound in water was found to have been much improved. The sparingly soluble compounds mentioned above[1,5a] were derived to this type of semicarbazone and showed a fairly good solubility in water.

4-β-Hydroxyethylsemicarbazide (IV) was prepared by the usual method[8] by heating acetone semicarbazone (II) and ethanolamine in toluene for a long period of time, or heating for 1 hour at 180°, to obtain acetone 4-β-hydroxyethylsemicarbazone (III) and its hydrolysis with acid.



The reaction of 4-β-hydroxyethylsemicarbazide (IV) thereby obtained with the compound of 5,6-dioxo-2,3,5,6-tetrahydroindole type[5a] or with 6,7-dioxo-1,2,3,4,6,7-hexahydroquinoline type afforded, in the majority, yellowish orange or red crystals, though some were syrupy and crystallized on standing. The ultraviolet spectra of 1-methyl-3-hydroxy-5-(4'-β-hydroxyethyl)semicarbazono-6-oxo-2,3,5,6-tetrahydroindole (V) showed $\lambda_{max}^{H_2O(pH\,2.6)}$ 360 mμ ($\log\epsilon$ 5.38) and $\lambda_{max}^{H_2O(pH\,11.4)}$ 445 mμ ($\log\epsilon$ 5.37), and of 1-β-hydroxyethyl-3-hydroxy-5-(4'-β-hydroethyl)semicarbazono-6-oxo-2, 3, 5, 6-tetrahydroindole,(VI) $\lambda_{max}^{H_2O(pH\,2.8)}$ 365 mμ ($\log\epsilon$ 5.40) and $\lambda_{max}^{H_2O(pH\,11.5)}$ 462 mμ ($\log\epsilon$ 5.46). Such

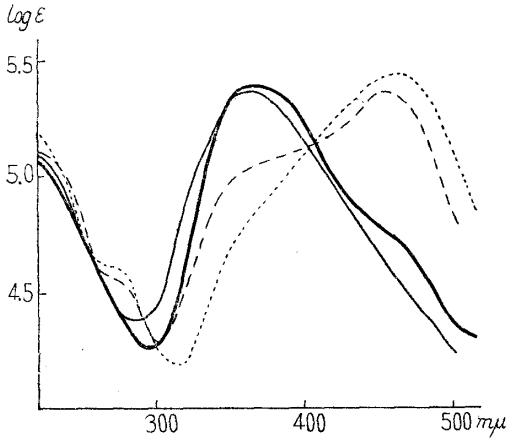
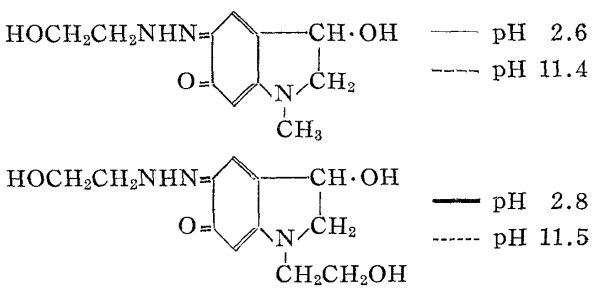


Fig. 1. Ultraviolet Absorption Spectra



6) H. Sobotka, J. Austin : J. Am. Chem. Soc., **73**, 3077(1951).
7) Belgian Pat. 510,295 (through C.A., **48**, 3397(1954)).
8) F. Wilson, I. Hopper, A. Crawford : J. Chem. Soc., **1922**, 866.

changes of ultraviolet absorption are the same as in adrenochrome monosemicarbazone and the derivatives reported earlier[1,5] (Fig. 1).

The solubility of (V) in water is 1 mg./cc. at 0°, showing an increase of approximately 3.5 times that of adrenochrome monosemicarbazone. Pharmacological effect of these compounds will be reported at a later date.

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Experimental

Acetone 4-β-Hydroxyethylsemicarbazone (III)—A mixture of 5.7 g. of acetone semicarbazone, 5 g. of ethanolamine, and 15 cc. dehyd. toluene was refluxed until the evolution of NH_3 gas ceased (ca. 13 hrs.). After cool, the oil that separated was collected and allowed to stand with an addition of ether, by which the oil solidified. Recrystallization from EtOH-ether mixture afforded 2.2 g. of colorless scales, m.p. 94~96°. *Anal.* Calcd. for $C_6H_{13}O_2N_3$: C, 45.27; H, 8.23; N, 26.40. Found: C, 45.15; H, 8.11; N, 26.44.

4-β-Hydroxyethylsemicarbazide (IV) Hydrochloride—i) A mixture of 1 g. of (III) and 10 cc. of 10% HCl was heated for 2 hrs., evaporated to dryness under a reduced pressure, EtOH was added to the residue, again evaporated to dryness, and the residue was recrystallized from EtOH to plate crystals, m.p. 130~133°. *Anal.* Calcd. for $C_3H_9O_2N_3 \cdot HCl \cdot \frac{1}{2}H_2O$: C, 21.89; H, 6.08; N, 25.53. Found: C, 21.70; H, 6.46; N, 25.44.

ii) After heating 35 g. of ethanolamine to 180°, 54 g. of acetone semicarbazone was added and the mixture was maintained at that temperature until complete solution took place, during which evolution of NH_3 gas occurred. To this was added 200 cc. of 10% HCl, warmed on a water bath for about 10 mins., and the solution was evaporated to dryness on a water bath under a reduced pressure. Recrystallization of the residue from 100 cc. of EtOH afforded 46.5 g. of crude crystals which were used, *per se*, for subsequent reactions.

1-Methyl-3-hydroxy-5-(4′-β-hydroxyethylsemicarbazono)-6-oxo-2, 3, 5, 6-tetrahydroindole (V)—To a solution of 3.0 g. of adrenochrome dissolved in 10 cc. of water, a solution of 4.5 g. of (IV) hydrochloride, 4.5 g. of $AcONa \cdot 3H_2O$, and 5 cc. of water was added, by which red crystals began to precipitate out after some time. Recrystallization from hydr. MeOH afforded 3.5 g. of red needles, m.p. 208°(decomp.). *Anal.* Calcd. for $C_{12}H_{16}O_4N_4$: C, 51.42; H, 5.75; N, 19.99. Found: C, 51.57; H, 5.89; N, 20.33.

1-β-Hydroxyethyl-3-hydroxy-5-(4′-β-hydroxyethylsemicarbazono)-6-oxo-2, 3, 5, 6-tetrahydroindole (VI)—To a solution of 10 g. of syrupy hydrochloride of α-(ω-hydroxyethylaminomethyl)protocatechuyl alcohol[5a] dissolved in 100 cc. MeOH, 40 g. of freshly prepared Ag_2O was added and oxidized at room temperature, by which the solution turned dusky reddish violet. Ag_2O was removed by filtration and 12.5 g. of (IV) hydrochloride, 12.5 g. of $AcONa \cdot 3H_2O$, and 30 cc. of water were added to the filtrate. After allowing the mixture to stand for some time, it was evaporated to dryness under a reduced pressure, a small amount of EtOH added, and allowed to stand over night. The crude crystals (10 g.) that separated out were recrystallized from MeOH to yellowish orange crystals, m.p. 197~199°(decomp.). *Anal.* Calcd. for $C_{13}H_{18}O_5N_4$: C, 50.31; H, 5.85; N, 18.06. Found: C, 50.43; H, 5.88; N, 18.10.

1-Methyl-5-(4′-β-hydroxyethylsemicarbazono)-6-oxo-2,3,5,6-tetrahydroindole (VII)—A solution of 4 g. of epinine hydrobromide dissolved in 20 cc. of water was oxidized by the addition of a solution of 18.8 g. of $K_3Fe(CN)_6$ and 6 g. $NaHCO_3$ dissolved in 45 cc. of water, under cooling, by which the solution turned dusky red, forming epinochrome. To this were added 4.4 g. of (IV) hydrochloride, 4.4 g. $AcONa \cdot 3H_2O$, and 15 cc. water, and the brownish yellow crystals that gradually separated were recrystallized from MeOH to 1.2 g. of yellow needles, m.p. 198°(decomp.). *Anal.* Calcd. for $C_{12}H_{16}O_3N_4$: C, 54.55; H, 6.10; N, 21.20. Found: C, 54.47; H, 5.60; N, 21.43.

1-Methyl-2-ethoxycarbonyl-5-(4′-β-hydroxyethylsemicarbazono)-6-oxo-2, 3, 5, 6-tetrahydroindole (VIII)—A solution of 1.7 g. of ethyl β-3,4-dihydroxyphenyl-α-methylaminopropionate dissolved in 25 cc. of water containing a small amount of MeOH was oxidized by the addition of 9.2 g. $K_3Fe(CN)_6$ and 3.1 g. $NaHCO_3$ dissolved in 22 cc. of water, by which the solution turned dusky red. To this solution were added 1.2 g. of (IV) hydrochloride, 1.2 g. $AcONa \cdot 3H_2O$, and 10 cc. of water and the mixture was allowed to stand, though it separated some resinous matter. The mixture gradually separated an oily substance which later solidified and its recrystallization from EtOH

afforded 1.3 g. of yellow needles, m.p. 165~167°. *Anal.* Calcd. for $C_{15}H_{20}O_5N_4$: C, 53.56; H, 5.99; N, 16.66. Found: C, 53,14; H, 5.64; N, 16.78.

1-Methyl-6-(4′-β-hydroxyethylsemicarbazono)-7-oxo-1, 2, 3, 4, 6, 7-hexahydroquinoline (IX)—Oxidation of a solution of 4.9 g. of 1-methyl-6,7-dihydroxy-1,2,3,4-tetrahydroquinoline hydrobromide dissolved in 200 cc. of MeOH with 30 g. of freshly prepared Ag_2O resulted in dusky reddish violet solution. After filtering off Ag_2O from this oxidized solution, 6.0 g. of (IV) hydrochloride, 6.0 g. $AcONa \cdot 3H_2O$, and 24 cc. water were added to the filtrate and the solution was evaporated to dryness under a reduced pressure. MeOH was added to the residue, insoluble matter was removed, the solution was again evaporated to dryness, and acetone-ether mixture was added to the residue by which it crystallized. Recrystallization from EtOH afforded 2.8 g. of red needles, m.p. 186~187°(decomp.). *Anal.* Calcd. for $C_{13}H_{18}O_3N_4$: C, 56.10; H, 6.52; N, 20.13; Found: C, 56.73; H, 6.81; N, 20.27.

1-Methyl-4-hydroxy-6-(4′-β-hydroxyethylsemicarbazono)-7-oxo-1,2,3,4,6,7-hexahydroquinoline (X)—To α-(2-Methylaminoethyl)-protocatechuyl alcohol hydrobromide, obtained by the catalytic reduction of 3 g. of ω-methylamino-3,4-dihydroxypropiophenone hydrobromide as described in Part IV of this series,[1] contained in EtOH solution, 15 g. of freshly prepared Ag_2O was added to effect oxidation. Ag_2O was filtered off from the dusky reddish violet oxidized solution and 4 g. of (IV) hydrochloride, 4 g. $AcONa \cdot 3H_2O$, and 15 cc. water were added to the filtrate. This solution was evaporated to dryness, inorganic matter was removed by the addition of MeOH, and MeOH was evaporated. The residue crystallized (0.4 g.) and this was recrystallized from MeOH to red prisms, m.p. 194°(decomp.). *Anal.* Calcd. for $C_{13}H_{18}O_4N_4 \cdot \frac{1}{2}H_2O$: C, 51.45; H, 6.32; N, 18.48. Found: C, 51.88; H, 6.58; N, 18.68.

Summary

Adrenochrome monosemicarbazone is sparingly soluble in water (0.3 mg./cc. at 0°), that there is some difficulty in preparing highly concentrated solutions. Urea is generally used as the solubilization agent but this substance was found to be ineffective in this case. β-Hydroxyethylurea, introduced with the soluble hydroxyethyl group, was also ineffective. However, adrenochrome 4-β-hydroxyethylsemicarbazone was found to be 3.5 times more soluble than the ordinary monosemicarbazone. 4-β-Hydroxyethylsemicarbazones were prepared of three compounds of 5,6-dioxo-2,3,5,6-tetrahydroindole type[5a] and two compounds of 6,7-dioxo-1,2,3,4,6,7-hexahydroquinoline type[1] and they were all found to be far more easily soluble than the monosemicarbazones. 4-β-Hydroxyethylsemicarbazide was prepared by heating acetone semicarbazone and monoethanolamine at 180° to form acetone 4-β-hydroxyethylsemicarbazone and its hydrolysis with mineral acid.

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