A NEW AMINO ACID, "PRECATORINE"

YIH-CHIH T'UNG, CHIEN-TIEN HSU, HUNG-TU LEE and HSUEN-SAW KUO

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The seeds of Abrus precatorius which grow wild in the southern part of Taiwan are claimed to be powerfully poisonous (1,2). Since 1932 the chemical components of Joquility seeds have been studied by many authors (3-7). Among the chemical components, two indole positive substances, i.e. abrine (α-N-monomethyl-L-tryptophan) and hypaphorine (the betaine of L-tryptophan), have been identified. Both are the derivatives of an important essential amino acid, L-tryptophan. For the presence of these two L-tryptophan derivatives, we believe that there is an enzyme system which converts abrine to hypaphorine by methylation, or in the reverse by demethylation, and there might be an intermediate substance, i.e. α-N, N-dimethyl-L-tryptophan occurring in Jequirity seed. Since our laboratory is especially interested in the studies of indole derivatives, the purpose of the investigation was to isolate this new amino acid, and to study its physiological functions.

In our laboratory, the new amino acid was crystallized and we named it "precatorine". In this paper, we shall describe the method of investigation of "precatorine" (α-N, N-dimethyl-L-tryptophan) from the alcohol soluble constituents of the seed. The method of identification and some properties of the new amino acid are also described.

EXPERIMENTAL AND RESULTS

After concentrating the alcohol extracts of the seed (ca. 2.5 kg.), crude abrine crystals were obtained and the syrupy mother liquor was diluted with a large amount of water. Using adsorption-dialysis technique (8), a cellophane bag (No. 4465-A4 dialyzer tubing cellulose, Arthus H. Thomas Company, Philadelphia 5, U.S.A., 3 cm × 240 cm) containing an aqueous slurry of Amberlite IR 120 (in the acid form) was immersed into extract. Stirring was continued for 48 hours at room temperature. The cellophane bag was then taken out and washed with distilled water to remove any adhering contaminants. Then the resin in the bag was poured into a column (4 cm, in diameter) and was washed with distilled water (ca. 5 l.) until the eluants were clear. The washing was repeated with 0.5 N ammonia and indole positive yellow eluant was collected. The ammoniacal eluants were concentrated under reduced pressure to dryness and the residue was extracted with methyl alcohol. The methanol extracts were passed through alumina column (2 cm × 25 cm. active alumina for absorption chromatography, Wako Pure Chemical Industries, L.T.D.) and eluted with methyl alcohol. After the first indole positive fraction had been collected it was concentrated under reduced pressure to one-third of its original amount. Then acetone was added. A large amount of needle-shaped crystals appeared, which decomposed at 142°C. The crystals gave no depression of m.p. with synthesized authentic trigonelline (9). Filtration and concentration of the filtrate gave rod-like crystals after standing 2 days in the refrigerator. Recrystallization from ethyl alcohol gave white-crystals of m.p. 261-262°C (decompose) which were identified to be α-N, N-dimethyl-L-tryptophan ("precatorine"). The yield was 1.0 g.

According to Barger (3), iodine and 10% sulfuric acid, when sprayed onto the reaction mixture, made it black, and the crystal solution was yellow. This indicates the presence of 2-N-monomethyl-3-indole-propionic acid.

The ultraviolet absorption spectrum of abrine and hypaphorine is consistent with that of the amino acid (10), and the ultraviolet absorption spectra of the three indole derivatives are similar. Three derivatives...
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The yield was 0.0022%. The analysis (anal. calcd. for C_{19}H_{29}O_{4}N_{3}) found:

- C: 67.22% (67.19%)
- H: 6.94% (6.98%)
- N: 12.06% (12.07%)

According to Van Romburgh and Barger, iodide of methyl-\(\alpha\)-trimethylamino-\(\beta\)-indole-propionate, m.p. 197-199°C, can be synthesized when tryptophan is treated with methyl iodide and sodium hydridoxide. Hypaphorine nitrate m.p. 215-219°C (decompose), can be isolated from the above iodide by hydrolysis with 1% aqueous sodium hydrosulphite and on acidification with concentrated nitric acid. Instead of tryptophan, using precatorine (or abrine) as starting material, iodide of methyl-\(\alpha\)-trimethylamino-\(\beta\)-indole-propionate and hypaphorine nitrate can also be obtained, which give no depression of m.p. with the respective products obtained in the experiment of Van Romburgh and Barger.

This confirms the presence of precatorine. 

The reactions were:

![Chemical Structure](image)

**TRYPTOPHAN**

**ABRINE**

**PRECATORINE**

**IODIDE OF METHYL-\(\alpha\)-TRIMETHYLAMINO-\(\beta\)-INDOLE PROPIONATE**

**HYPAHPORINE NITRATE**

It was found that precatorine is soluble in water, methyl alcohol, dil. acids and alkalies; slightly soluble in ethyl alcohol and insoluble in nonpolar solvents. It gave positive color reaction with Dragendorff's reagent, Ehrlich's reagent and was slightly positive to ninhydrin reaction. On polyamide thin layer chromatogram it gave a blue spot when sprayed with a mixture of equal volume of 1% aqueous ferric chloride solution and 1% aqueous potassium ferricyanide solution. It possessed the \(R_f\) value of 0.72 (Table 1) when developed in chloroform: ethylacetate: acetic acid=7:2:1

\[ \text{The ultraviolet absorption} \]

maximum absorption at 220 and 280 m\(\mu\) and infrared absorption of precatorine are shown in Fig. 1, 2 respectively. Specific rotation \([\alpha]_{D}^{25^\circ} = +133^\circ\).

Three derivatives of precatorine such as hydrochloride, picrate and chloroplatinate were also synthesized. The m.p. of these three derivatives were 244-245°C (decompose), 72-75°C and 196-196.5°C respectively.

**DISCUSSION**

Although many non-nitrogenous constituents, such as abralin, abranin chloride, a compound having molecular formula \(C_{19}H_{29}O_{4}\), a flavon with m.p. 218-9°C, gallic acid etc., have been reported in the seed of Abrus precatorius, precatorine is a nitrogen containing constituent of this seed other than abrine, hypaphorine and trigonelline. It is also interesting that all three kinds of \(N\)-methyl derivatives of \(L\)-tryptophan are present together in the seed. We must believe that there is an enzyme system that catalyze the methylation of...
Table 1: Some Physical and Chemical Properties of L-Tryptophan, L-Abrine, L-Precatorine and L-Hypaphorine

<table>
<thead>
<tr>
<th>Properties</th>
<th>Substance</th>
<th>Tryptophan</th>
<th>Abrine</th>
<th>Precatorine</th>
<th>Hypaphorine</th>
</tr>
</thead>
<tbody>
<tr>
<td>m. p.</td>
<td></td>
<td>298°C (decomp.)</td>
<td>295°C (decomp.)</td>
<td>261-2°C (decomp.)</td>
<td>237°C (decomp.)</td>
</tr>
<tr>
<td>Rf value on polyamide TLC</td>
<td>0.24</td>
<td>0.52</td>
<td>0.72</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ninhydrin reaction</td>
<td>positive</td>
<td>slightly positive</td>
<td>slightly positive</td>
<td>slightly positive</td>
<td></td>
</tr>
<tr>
<td>FeCl₃+K₃Fe(CN)₆ test</td>
<td>+ (blue)</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Ehrlich's reagent</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Dragendorf's reagent</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td></td>
</tr>
<tr>
<td>Citric acid-acetic anhydride reagent</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td></td>
</tr>
</tbody>
</table>

* This reagent is specific for tertiary amine.

Fig. 1. Ultraviolet absorption spectra

Abrine to hypaphorine or in the reverse by demethylation during the growth of the seed. Abrine has been interested by many investigators for its nutritive value and enzymatic studies. Decarboxylation of abrine produced N-methyltryptamine which is an oxytocic active amine and also possesses many physiological functions.

Hypaphorine in frog. The precatorine are the decarboxylation now. N, N-dimethyl-L-tryptophan by us.

From these facts, Abrine precatorine technique and can obtain some physical-N, N-dimethyl-L-tryptophan, "Precatorine". The some physical-N, N-dimethyl-L-tryptophan amino acid.

(3) Hoshi, J., 1932.
(4) Ghata, J., 1932.
Hypaphorine has a hyperexcitability on the frog\cite{20}. The biochemical studies of precatorine are being undertaken in our laboratory now. The physiological function of the decarboxylation product of precatorine—N, N-dimethyltryptamine is also interested by us.

**SUMMARY**

From the alcohol extracts of the seeds of Abrus precatorius, using adsorption-dialysis technique and alumina chromatography, we can obtain a new amino acid—a-N, N-dimethyl-L-tryptophan. We named it "precatorine". The method of identification and some physicochemical properties of this new amino acid are described in this paper.

**REFERENCES**


(4) GHATAK N. and KAUL P.: J. Indian

Chem. Soc. 9; 383, 1932.

(5) GHATAK N.: Current Sci. 2; 380, 1934.


**A New Amino Acid, "Precatorine"**

By the treatment of a crude extract, the amino acid "Precatorine" can be isolated. The amino acid possesses a unique property of activating the enzyme of the mammalian system. The physiological function of this amino acid is being investigated in our laboratory now. The method of identification and some physicochemical properties of this new amino acid are described in this paper.

**REFERENCES**


(4) GHATAK N. and KAUL P.: J. Indian

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