V bei höherem Feld als die des Pterosin A-Acetats liegen. (siehe Tabelle). Die Methoxygruppe muss daher an das C(β) gebunden sein. Diese Befunden, zusammen mit den CD-Daten von V ($[\theta]^{25}$: +1570 (335 nm, $CHCl_3$)), weisen für dieses neue Pterosin III, das wir als Pterosin V bezeichnen, die Struktur (2*S*)-2,5,7-Trimethyl-2-hydroxymethyl-6-(β-methoxyäthyl)-indan-1-on auf.

Ausserdem aus der $CHCl_3$-Phase konnten wir durch eine GC-MS-Analyse Pterosin K (VI)[5] und F (VII)[6] identifizieren.

Danksagung Wir haben zu danken Herrn Dr. S. Natori, National Institute of Hygienic Sciences, für die freundliche Überlassung der Vergleichssubstanzen. Ebenso sind wir Herrn R. Ito für die Hilfe bei der Suche und Sammlung des Pflanzenmaterials und Herrn Dr. T. Yamagishi, Hokkaido Institute of Public Health, für die Aufnahmen der GC-MS-Spektren sehr verbunden.

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5) M. Fukuoka, M. Kuroyanagi, M. Toyama, K. Yoshihira, und S. Natori, *Chem. Pharm. Bull.* (Tokyo), **20**, 2282 (1972).
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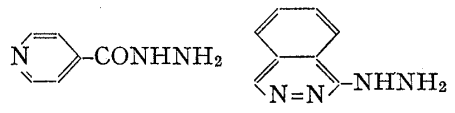
[Chem. Pharm. Bull. 23(7)1632—1634(1975)]

UDC 547.475.2.04 : 543.422

The Formation of Free Radical Intermediates from Vitamin C and Isoniazid or Apresoline

L-Ascorbic acid and isoniazid or apresoline were found to produce stable free radical intermediates when coexisted in alkaline aqueous solutions at room temperature. The electron spin resonance parameters and a possible skeletal structure of the radical species were presented.

In this communication we shall briefly report the reactions between L-ascorbic acid and isonicotinic acid hydrazide (isoniazid) or 1-hydrazinophthalazine (apresoline) which lead to the formation of free radical intermediates in alkaline aqueous solutions. Since it has previously been found[1,2] that L-ascorbic acid reacts with hydrazine and substituted hydrazines in aerobic, alkaline solutions to give rise to some stable radical intermediates, we anticipated that similar reactions may also take place with isoniazid (I) or apresoline (II) which is frequently used as a tuberculostatic or antihypertensive drug. Investiga-



I II

1) L. Burlamacchi, P. Sarti-Fantoni, and E. Tiezzi, *Tetrahedron Letters*, **1969**, 5005.
2) H. Utsumi, Y. Kirino, and T. Kwan, *Chem. Pharm. Bull.* (Tokyo), **22**, 1417 (1974); *idem*, *ibid.*, **27**, 1516 (1975).

tions were therefore carried out and led to essentially the same results as already reported,[2] *i.e.* stable radical intermediates were formed by the reaction between L-ascorbic acid and isoniazid or apresoline.

A typical reaction of L-ascorbic acid with isoniazid or apresoline was carried out as follows; a 150 mg isoniazid or 30 mg apresoline was added to a 2 ml aqueous solution of L-ascorbic; acid (0.2M). The resultant solution was acidic; either isonizid or apresoline alone was neutral in aqueous solutions. The solution was stable over several hours and pale yellow in the presence of air at room temperature. However, it became gradually orange when initially alkalified. After several hours, a 50 μl of the orange-colored solution was put into a quartz tube and then subjected to electron spin resonance (ESR) measurements at room temperature. The spectra obtained with isoniazid or apresoline are respectively illustrated in Fig. 1 and Fig. 2.

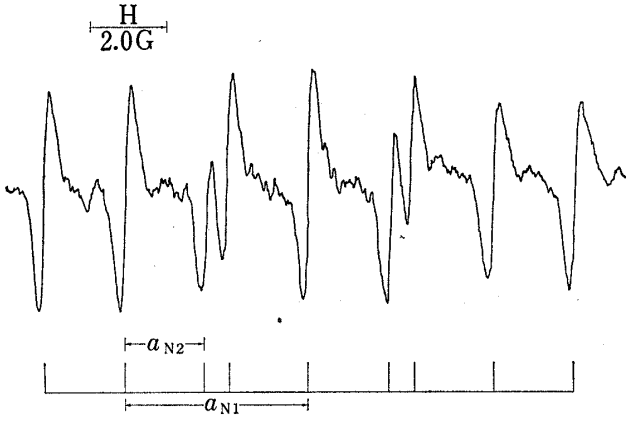


Fig. 1. The ESR Spectrum of Free Radical Intermediate formed during the Reaction of L-Acsorbic acid with Isoniazid at pH 8.5 (a_{N1}=4.81 G, a_{N2}=2.12 G)

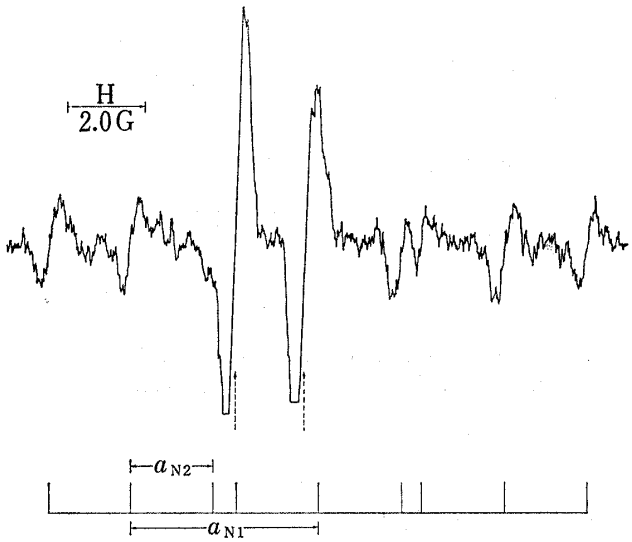


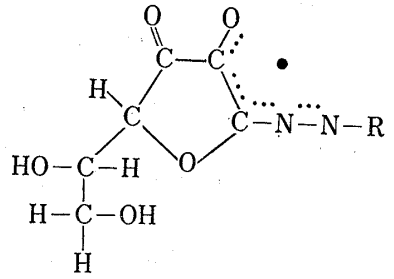
Fig. 2. The ESR Spectrum of Free Radical Intermediate formed during the Reaction of L-Ascorbic acid with Apresoline at pH 8.0 (a_{N1}=4.90 G, a_{N2}=2.17 G; ····→ : L-ascorbic acid radical)

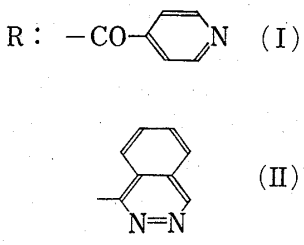
The spectrum shown in Fig. 1 consists of nine resonance lines and is nearly identical with that obtained from the reaction between L-ascorbic acid and hydrazine[1,2] which has been interpreted as due to hyperfine interaction of unpaired electron with two nonequivalent nitrogen atoms (I=1).

When L-ascorbic acid was added in excess over isoniazid or apresoline, the spectrum of the colored solution appeared to be accompanied by extra lines, for example, as that given in Fig. 2 (indicated by the dotted arrows). The extra signal was substantially absent provided that the solution was not in excess of L-ascorbic acid. The signal agreed with the major resonance lines of L-ascorbic acid radical.[3] Thus, it can be considered that L-ascorbic acid reacts with apresoline as well as with isoniazid to give rise to stable radical intermediates, the spectrum of which is characterized with nine resonance lines as indicated in Fig. 1.

The nitrogen hyperfine constants were a_{N1}=4.81G and a_{N2}=2.12G for the radical intermediate generated from L-ascorbic acid and isoniazid, and a_{N1}=4.90G and a_{N2}=2.17G for L-ascorbic acid and apresoline. The *g*-values were estimated to be close to 2.0045 for both radical species. In view of their similarities in the ESR parameters to the radical from L-ascorbic acid and hydrazine (a_{N1}=4.90G, a_{N2}=2.18G, g=2.0044), the skeletal structure is assumed to be identical with each other and represented as below.

3) Y. Kirino and T. Kwan, *Chem. Pharm. Bull.* (Tokyo), **20**, 2651 (1972).





The free radical intermediates were fairly stable (over days) in the aqueous solution, but might react depending on environmental conditions. The hydrazine analogs are frequently used as the tuberculostatic or antihypertensive drugs,[4] so that their reactions with vitamin C might involve some biological significance.

Acknowledgement The authors wish to thank Professor K. Takagi for his advice on the present work, and Takeda Chemical Industries, Ltd. for supply of purified L-ascorbic acid.

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Received April 30, 1975

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[Chem. Pharm. Bull. 23(7)1634—1637(1975)]

UDC 547.92'457.1.02 : 581.192

Chemische Untersuchungen der Inhaltsstoffe von *Pteris inaequalis* BAKER var. *aequata* (MIQ.) TAGAWA[1]

Im Zusammenhang mit der chemotaxonomischen Studien der Gattung *Pteris* und der verwandten Gattungen (*Pteridaceae*) wurden aus *Pteris inaequalis* BAKER var. *aequata* (MIQ.) TAGAWA Pterosin C, Acetylpterosin C, 5-Hydroxymethyl-2-Furfural, 2-Desoxy-D-Glukose (I), 3,6-Anhydro-2-desoxy-D-Glukose (II) und 2-Desoxy-D-Glukose-3-Monomethyläther (III) sowie ein Steringlykosid (IV) isoliert und identifiziert. Von ihnen sind II, III, und IV bisher noch nie in der Natur aufgefunden.

Nach Untersuchungen von Kobayashi und Mitarbb.[2] enthalten die Methanol-Extrakte von *Pteris inaequalis* BAKER var. *aequata* (MIQ.) TAGAWA (Jap. Name: Ōbanohachijoshida) neben dem bekannten Indan-1-on-Derivat, Pterosin B[3] zwei antibiotisch wirksame Komponenten, deren Strukturen noch nicht erklärt worden sind.

In Fortsetzung unserer chemischen und chemotaxonomischen Untersuchungen der Gattung *Pteris* und der verwandten Gattungen (*Pteridaceae*),[1] wurden dieselben Pflanzen (Fundort: Owase/Mie-Präfektur, Sammelzeit: Juli, 1973) erneut bearbeitet. Die oberiridischen

1) Chemische und chemotaxonomische Untersuchungen der Gattung *Pteris* und der verwandten Gattungen (*Pteridaceae*) VIII Mitteil, VII Mitteil.: T. Murakami, K. Owashi, N. Tanaka, T. Satake, und C.-M. Chen, *Chem. Pharm. Bull.* (Tokyo), **23**, 1630 (1975).
2) T. Kobayashi, K. Koshimizu, T. Mitsui, H. Egawa, und H. Shinkai, Abstracts of Papers, Annual Meeting of the Agricultural Chemical Society of Japan, Tokyo, 1973, s. 251.
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