and are assumed to be produced via 3$\beta$-hydroxy-pregn-5-en-20-one or progesterone derived from cholesterol or sitosterol [2]. Some $\Delta^{16}$-20-keto-pregnanes could be assumed to be formed in plants from co-occurring spirostanes [3, 4, 9] or furostane derivatives as well as spirosolane alkaloids [10, 11] in a manner similar to the corresponding Marker type chemical degradation. It is unknown if 3$\beta$-hydroxy-5$\alpha$-pregnan-16-one in *S.hainanense* is produced as a secondary product of one of the above degradation pathways or by a new biosynthetic route. However, the present findings show that the possibilities for pregnane biosynthesis in higher plants seem to be more various than assumed up to now [12].

## EXPERIMENTAL

UV spectra were measured in MeOH; ORD determinations were in MeOH; NMR spectra were taken in $CDCl_3$ with HMDS as internal standard.

*Isolation of 3$\beta$-hydroxy-5$\alpha$-pregnan-16-one.* Dried and powdered roots (100 g), collected near Hanoi, Vietnam, were extracted exhaustively with $CHCl_3$ in a Soxhlet apparatus, $CHCl_3$ was concd to 1/3 and extracted 3 × with petrol to remove pigments and lipids. Evapn of the $CHCl_3$ gave a residue which was chromatographed over $Al_2O_3$ (Woelm, neutral, grade I). The progress of the separation was followed by TLC on Si gel ($CHCl_3$–EtOH, 9:1). Elution with $CHCl_3$–EtOH (8:2) yielded 3$\beta$-hydroxy-5$\alpha$-pregnan-16-one. Needles ($Me_2CO$–$H_2O$), mp 153°, $[\alpha]_D^{26} - 47.3°$ ($c = 0.300$, in EtOH), $R_f$ 0.74, identical mmp, $R_f$, IR, ORD) with an authentic specimen [7].

Further elution with $CHCl_3$–EtOH (7:3) gave solasodenone as already described [8].

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# A NATURAL APOCAROTENOL FROM THE PEEL OF THE RIPE GOLDEN DELICIOUS APPLE

Jeana Gross* and Gert Eckhardt†

*Institute for Fruit and Vegetable Research, †Department of Organic Chemistry, Rheinische Friedrich Wilhelms University, Bonn, West Germany

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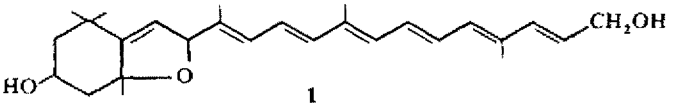
**Key Word Index**—Golden Delicious apple; natural apocarotenols.

## INTRODUCTION

In various fruits, polar UV fluorescent pigments with sharp fine structure, typical of short in-chain chromophores, were found. They were named according to their source, valenciaxanthin, valenciachrome, from Valencia and Navel orange peel [1, 2]. Persicaxanthin and persicachrome were detected in Rosaceae (peaches, apricots, prunes) [3, 5] and in other fruits with low carotenoid content (figs, blackberries, grapes) [6]. Their structure is still unknown [7].

In the pulp of avocado fruits, *Persea americana*, two new UV fluorescent apocarotenoids were identified by Gross *et al.* [8]. One of these was assigned the structure 5,8-epoxy-5,8-dihydro-10′-apo-$\beta$-carotene-3,10′-diol (1): The pigment is related to the natural apo-10′-violaxanthal (5,6-epoxy-3-hydroxy-5,6-dihydro-10′-apo-$\beta$-caroten-10′-al) found in Valencia orange peels [9]. This compound ($\lambda_{max}$ 440 nm) was reduced to the corresponding alcohol $\lambda_{max}$ 370, 392, 414 nm. On treatment with HCl the maxima shifted to 352, 371, 394 nm, through 5,8-isomerization of the 5,6-epoxide. Curls *in vitro* product appears to be identical with the apocarotenol (1) found in avocado.

While investigating the carotenoid changes during the ripening of Golden Delicious apples, we detected a similar pigment that appeared only in ripe fruit, and its structure was investigated.

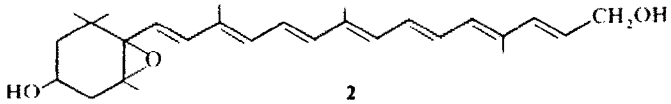


1

## RESULTS AND DISCUSSION

The pale yellow pigment constituted *ca* 2% of the total carotenoids of the apple peel (20 µg/g fr. wt). The pigment was detected in the most polar fraction obtained from CC on MgO–Hyflo Super Cel, which is the neoxanthin fraction. Upon TLC on Si gel G the pigment was found between luteoxanthin and neoxanthin. According to its electronic spectrum and its positive epoxide test, structure **2** was proposed: 5,6-epoxy-5,6-dihydro-10′-apo-β-carotene-3,10′-diol. This structure was confirmed by MS of the pigment, its 5,8-epoxide isomer and its diacetate.

These findings provide further evidence that this new class of apocarotenols are metabolites formed only in ripe fruit. These metabolites may be formed through the Glover–Redfearn degradation of the most abundant pigment present. Thus in avocado, chrysanthemaxanthin [10] yields pigment **1**; in apple, where at this stage of maturation violaxanthin is the main pigment, it yields pigment **2**.



**2**

Evidence that an enzymatic reduction occurs subsequently must still be produced.

## EXPERIMENTAL

The analytical methods were as reported previously [11]. Prior to extraction, a neutralizing agent was added. After extraction, saponification and removal of sterol, the extract was chromatographed on a column of MgO–Hyflo Super Cel (1:1). The column was developed stepwise with increasing amounts of $Me_2CO$ in petrol. The most polar fraction was separated by developing with $Me_2CO$–petrol–EtOH (9:88:3). This neoxanthin fraction was further separated by TLC on Si gel G developed with $Me_2CO$–petrol (2:3). The pale yellow pigment was situated between luteoxanthin and neoxanthin. Its electronic spectrum $\lambda_{max}$ 372, 394, 418 nm was indicative for a heptaene chromophore. This spectrum was identical with that of Curl's reduced violaxanthal. The HCl–epoxide test was positive, with a hypsochromic shift of 22 nm. The 5,8-furanoxide had the spectrum 353, 372, 395 nm identical with that of (**1**). For high resolution MS the samples were introduced via the probe inlet system, the source temp. varied between 200–250°, the ionizing voltage was 70 eV.

The elementary composition of **2** was $C_{27}H_{38}O_3$ (Found 410.2816; calc. 40.2821). Fragments at *m/e* 221 ($C_{14}H_{21}O_2$) and 181 ($C_{11}H_{17}O_2$) supported the presence of the 3-hydroxy-epoxide ring. Its diacetate gave the $M^+$ at *m/e* 494 ($C_{31}H_{42}O_5$) and the characteristic fragments for the end groups shifted to *m/e* 263 ($C_{16}H_{23}O_3$) and 223 ($C_{13}H_{19}O_3$). The 5,8-epoxide isomer (**1**) exhibited the same MS as **2**: *m/e* 410 ($C_{27}H_{38}O_3$, $M^+$), 330 ($[M-C_6H_8]^+$); 221 ($C_{14}H_{12}O_2$); 181 ($C_{11}H_{17}O_2$). This MS is identical with that of the pigment isolated from avocado pulp [8].

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# LAMBERTELLIN AND CHRYSOPHANOL FROM THE IMPERFECT FUNGUS *PSEUDOSPIROPES SIMPLEX*

GIJSBERTUS W. VAN EIJK and HENRY J. ROEYMANS

Centraalbureau voor Schimmelcultures, P.O. Box 273, Baarn, The Netherlands

(*Received* 10 *March* 1978)

**Key Word Index**—*Pseudospiropes simplex*; Hyphomycetes; fungi; lambertellin; 9-hydroxy-3-methylnaphtho[2,3-b]pyran-2,5,10-trione; chrysophanol; 1,8-dihydroxy-3-methylanthraquinone.

From the EtOAc extract of 25-day-old cultures of *Pseudospiropes simplex*, an orange-red crystalline substance was isolated by PLC which became violet in aq. $Na_2CO_3$. Its IR spectrum showed a band at 1620 $cm^{-1}$, characteristic of a hydrogen-bonded quinone group, in addition to absorptions at 1742 and 1660 $cm^{-1}$. The compound showed the UV absorption of a juglone and formed an acetate. High resolution MS gave the molecular formula $C_{14}H_8O_5$. A compound with similar physico-chemical properties is the pyranonaphthoquinone, lambertellin (**1**), a pigment hitherto only reported from the discomycete genus *Lambertella* Höhn