Table II—Percent of Radioactivity [a] Found in Each Phase following Folch Extraction of Tissue Radioactivity at 1 and 4 Days Postadministration of $^{125}$I-Labeled 19-Iodocholesterol

| Tissue | Day 1 | | | Day 4 | | |
|---|---|---|---|---|---|---|
| | Precipitate | Upper | Lower | Precipitate | Upper | Lower |
| Adrenal | 5.47 ± 2.36[b] | 0.40 ± 0.42[b] | 94.14 ± 2.48[b] | 9.02 ± 3.54[b] | 0.12 ± 0.14[b] | 90.87 ± 3.65[b] |
| Blood | 19.50 ± 4.74 | 13.87 ± 0.96 | 66.63 ± 5.21 | 29.00 ± 13.09 | 14.54 ± 4.98 | 56.47 ± 14.02 |
| Liver | 8.83 ± 2.00[b] | 13.60 ± 2.04[b] | 77.58 ± 2.29[b] | 12.53 ± 1.91 | 11.18 ± 2.37 | 76.30 ± 3.87 |
| Ovary | 7.89 ± 2.21 | 0.92 ± 0.53 | 91.19 ± 1.82 | 8.81 ± 3.71 | 0.35 ± 0.10 | 90.84 ± 3.70 |

[a] Each value represents the mean ± *SD* for five rats unless noted. [b] Four rats.

uptake of radioactivity by pretreatment with potassium iodide solution (1) and in Folch extraction studies where >98% of the thyroid radioactivity was associated with the precipitated fraction[7].

The TLC systems used gave good separation of the free and esterified sterol. No further identification of radioactive products was attempted. Therefore, the esterified sterol peak could be made up of 19-iodocholesterol esterified with different saturated and unsaturated fatty acids. Similarly, slight alterations in the sterol portion of the molecule also could go undetected. Thus, [$^{14}$C]cholesterol and $^{125}$I-labeled 19-iodocholesterol have the same $R_f$ value in these systems.

Despite these limitations, these studies support the view that 19-iodocholesterol mimics cholesterol with regard to esterification and storage and that this property is a factor in its accumulation and retention for long periods in the adrenal cortex and ovary. In addition to its use in adrenal imaging, 19-radioiodinated cholesterol or related compounds may have clinical utility for the diagnosis of ovarian disorders.

[7] R. E. Counsell, N. Korn, R. Langdon, and J. L. McGauley, unpublished data.

## REFERENCES


(1) R. E. Counsell, V. V. Ranade, R. J. Blair, W. H. Beierwaltes, and P. A. Weinhold, *Steroids*, **16**, 317 (1970).

(2) R. E. Counsell and N. Korn, in "Principles of Radiopharmacology," L. G. Columbetti, Ed., CRC, Boca Raton, Fla., 1979, p. 189.

(3) D. Schulster, S. Burstein, and B. A. Cooke, "Molecular Endocrinology of the Steroid Hormones," Wiley, New York, N.Y., 1976, pp. 167, 230.

(4) J. Folch, M. Lees, and G. H. Sloane-Stanley, *J. Biol. Chem.*, **226**, 497 (1957).


## ACKNOWLEDGMENTS


Supported in part by U.S. Public Health Service Grant CA-08349. The authors thank Searle Laboratories for the 19-iodocholesterol.


# Allyl Derivatives of Zearalenone


## M. T. SHIPCHANDLER



Received May 10, 1979, from the *Research & Development Division, International Minerals & Chemical Corporation, Terre Haute, IN 47808.* Accepted for publication June 21, 1979. Present address: Diagnostics Division, Abbott Laboratories, North Chicago, IL 60064.



**Abstract** □ The Claisen rearrangement of 4-*O*-allylzearalenone led to only one principal product, which was identified as 3-allylzearalenone with the aid of photochemical isomerization and PMR spectroscopy. This general method is useful for distinguishing 3-substituted zearalenone isomers from 5-substituted isomers. Hydrogenation of 3-allylzearalenone gave 3-(1-propyl)zearalanone.



**Keyphrases** □ Zearalenone—allyl derivatives, synthesis □ Anabolic agents—zearalenone, synthesis of allyl derivatives


Zearalenone (I), a metabolite of *Gibberella zeae*, is produced *via* an industrial fermentation process (1). Several derivatives of I were synthesized to enhance its anabolic activity (2, 3). Zeranol (VI), a tetrahydro derivative, is used commercially as an anabolic agent in feedlot steers. The same fungus also produces four structurally related minor metabolites, one of which was characterized as 3-formylzearalenone (II) (4).

## DISCUSSION

Isomeric 3- and 5-substituted zearalenones are not distinguished readily from each other by standard spectroscopic methods. Bolliger and Tamm (4) formylated zearalenone, and extensive PMR spectral scrutiny of the resulting 3- and 5-formylzearalenones and their corresponding dimethyl ethers led to the unequivocal assignment of Structure II to the metabolite (4). In another instance, III, the Kolbe–Schmitt reaction product of zearalenone, was subjected to extensive degradation (5).

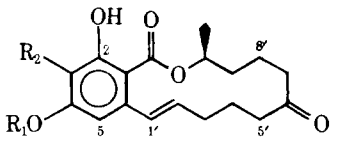
This report describes a simple photochemical method coupled with PMR spectroscopy for a facile structural assignment. 4-*O*-Allylzearalenone (IV) was subjected to the Claisen rearrangement at 190–195°, and the principal product, V (mp 116–118°), was isolated in a 48% yield. It was isomerized to 3-allyl-*cis*-zearalenone (VIII) *via* the photochemical method of Peters (6). In the comparative PMR spectra of V and VIII, the upfield shift (13 Hz) of the aromatic proton of C-5 was very distinct, while the shifts of the allyl group protons were practically unchanged (6). Such an upfield shift of the aromatic proton of C-3 is not expected to occur in 5-substituted derivatives. The Claisen product thus was identified as 3-allylzearalenone (V). Low-pressure hydrogenation of VI gave 3-(1-propyl)zearalanone (VII).

## EXPERIMENTAL[1]

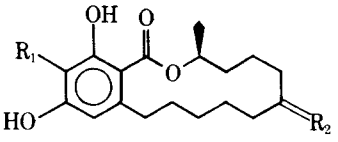
**4-*O*-Allylzearalenone (IV)**—Compound I (15.9 g, 0.05 mole), allyl bromide (24.2 g, 0.2 mole), and potassium carbonate (10.0 g, 0.07 mole) were stirred and refluxed in acetone (200 ml) for 8 hr. The filtered inorganic salts were washed well with more solvent. The combined washings and filtrate were evaporated under reduced pressure. The residue was crystallized from 2-propanol to give 10 g of IV, mp 123–125°. In the PMR spectrum, a peak at δ 12.00 indicated the presence of the chelated hydroxyl proton at the 2-position; the absence of a peak at about δ 8.00 of the hydroxyl proton at the 4-position indicated allylation of this site. With two aromatic protons at δ 6.38 and 6.48 ($J$ = 2.5 Hz), the rest of the spectrum was consistent with the assigned structure.

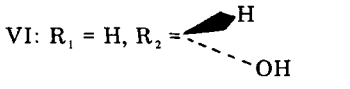
[1] Melting points (uncorrected) were measured with a Thomas-Hoover apparatus. PMR spectra were recorded on a Varian A 60-A instrument in deuterochloroform with tetramethylsilane as the internal standard. Elemental analyses were carried out by Microanalyses Inc.


0022-3549/80/0900-1017$01.00/0

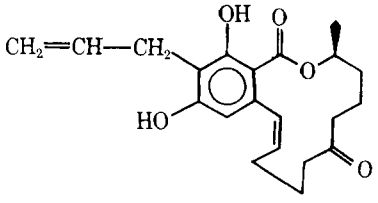


I: $R_1 = R_2 = H$
II: $R_1 = H$, $R_2 = CHO$
III: $R_1 = H$, $R_2 = COOH$
IV: $R_1 = CH_2CH{=}CH_2$, $R_2 = H$
V: $R_1 = H$, $R_2 = CH_2CH{=}CH_2$





VII: $R_1 = CH_2CH_2CH_3$, $R_2 = {=}O$



VIII

*Anal.*—Calc. for $C_{21}H_{26}O_5$: C, 70.37; H, 7.31. Found: C, 70.63; H, 7.13.

**3-Allylzearalenone (V)**—Compound IV (5.0 g) was placed in an oil bath maintained at 190–195° for 5 hr. TLC examination revealed only one principal product. The melt was chromatographed over a silica gel column[2] (100 g, 28 × 3.3 cm) and eluted with benzene and chloroform mixtures of increasing polarity. The fractions containing the product were evaporated together, and V was crystallized from methanol–water to give 2.5 g of white crystals, mp 116–118°. In the PMR spectrum, only one aromatic proton appeared as a singlet at δ 6.48 and two peaks of phenolic proton resonances appeared at δ 12.00 and 8.64. The C-1′ olefinic proton appeared as a doublet at δ 7.12 ($J$ = 15 Hz).

*Anal.*—Calc. for $C_{21}H_{26}O_5$: C, 70.37; H, 7.31. Found: C, 70.47; H, 7.33.

**3-Allyl-*cis*-zearalenone (VIII)**—Compound V (1.3 g) was photoisomerized[3] in methanol (100 ml) for 60 hr according to the method of Peters (6). Evaporation gave a glassy substance (1.2 g), which could not be purified by crystallization. The peak ratio of the C-5 aromatic protons in the PMR spectrum (deuterium oxide was added to remove the interfering resonance of the C-4 phenolic proton) indicated a 90–95% conversion. The singlet resonance of the C-5 aromatic proton appeared at δ 6.26, and the doublet resonance of the C-1′ olefinic proton appeared at δ 6.65 ($J$ = 12 Hz). This observation was consistent with that of Peters (6).

**3-(1-Propyl)zearalanone (VII)**—Compound V (1.0 g) was hydrogenated under 50 psi in methanol (100 ml) for 3 hr in the presence of palladium-on-carbon (5%, 0.1 g). The catalyst was removed *via* filtration, and the filtrate was evaporated under reduced pressure. The residue was crystallized from methanol–water to yield 1.1 g of white crystals, mp 161–162.5°. There were no resonances of olefinic protons in the PMR spectrum. The singlet resonance of the C-5 aromatic proton appeared at δ 6.27, and the pseudotriplet resonance of the methyl protons (of the propyl group) appeared at δ 1.00 ($J$ = 6 Hz).

*Anal.*—Calc. for $C_{21}H_{30}O_5$: C, 69.58; H, 8.34. Found: C, 69.70; H, 8.40.

## REFERENCES


(1) P. H. Hidy, R. S. Baldwin, R. L. Greasham, C. L. Keith, and J. R. McMullen, in "Advances in Applied Microbiology," vol. 22, D. Perlman, Ed., Academic, New York, N.Y., 1977, p. 59.

(2) W. H. Urry, H. L. Wehrmeister, E. B. Hodge, and P. H. Hidy, *Tetrahedron Lett.*, **1966**, 3109.

(3) M. T. Shipchandler, *Heterocycles*, **3**, 471 (1975).

(4) G. Bolliger and C. Tamm, *Helv. Chim. Acta*, **55**, 3030 (1972).

(5) T. B. Windholz and R. D. Brown, *J. Org. Chem.*, **37**, 1647 (1972).

(6) C. A. Peters, *J. Med. Chem.*, **15**, 867 (1972).


## ACKNOWLEDGMENTS


The author thanks Mr. Marvin C. Bachman for continued interest in this research and Mr. R. P. Sidenbender for technical assistance.


[2] Baker No. 3404.

[3] The photoisomerization process does not need to be complete. For example, the shifts at δ 6.48 and 6.26 of the C-5 aromatic protons were distinct in the PMR spectrum of the reaction mixture containing approximately equal amounts of V and VIII.