of problems in the synthesis, isolation, and characterization of I and II, and to call their attention to recently developed procedures for preparing I and II in greater than 98% chemical and radiochemical purity (*5,7*).

We have recently reported (*5*) a new route for the synthesis of I in greater than 98% chemical purity, the purity being established unambiguously by ^{13}C nuclear magnetic resonance (CMR). We then prepared II from I via the general route employed by the other groups (*7*). CMR and high-pressure liquid chromatography (HPLC) showed that this product contained not only the desired compound but at least 5 other impurities. HPLC was then employed to separate gram amounts of II, the purity of which (>98%) was established unequivocally by CMR.

Previous syntheses of II (*1,3,4*) employed preparative thin-layer chromatography (TLC) to obtain milligram quantities of II from a mixture containing both I and II. Kojima et al. (*1,3*) reported II as a glass, but Basmadjian et al. (*4*) as a low-melting solid. Furthermore, the proton nuclear magnetic (PMR) spectra of II reported by the two groups are significantly different. Our PMR data on II agree with those of Kojima et al. (*1,3*) but not with Basmadjian et al. (*4*). Moreover, the peak positions and intensities reported by Basmadjian et al. are not possible for steroids containing the C-20 cholestane side-chain (*7,8*). The PMR data reported for CH_2I in II by Basmadjian et al. are inconsistent with expectation (*9*). Since their data reported for CH_2I in II are almost identical with the CH_2I data for impure I (*6*), a possible explanation for the anomalous PMR data reported by Basmadjian et al. would be that although they have obtained II, it is contaminated by I and other unidentified compounds. This would not be surprising, since complete TLC separation of two compounds having a difference in RF of only 0.1, as in the case of I and II, can frequently be hard to achieve. For the preceding reasons, we believe that HPLC is the method of choice for purification of II.

In the PMR spectra of steroids, only methyl proton resonances and resonances of protons in or adjacent to functional groups can be assigned to specific protons (*9*). Most of the proton resonances, however (28 in the case of I, 29 in the case of II), form an overlapping, unresolved, and unassignable pattern that covers most of the right-hand half of the PMR spectrum. (See the PMR spectra of I and II in reference *7*.) Therefore, impurities that have proton peaks only in this region can go undetected in the PMR spectrum of a steroidal sample. For this reason, the PMR spectrum alone cannot unequivocally establish the purity of a steroid. On the other hand, inspection of the CMR spectra of I or II (*7*) shows 27 well-resolved and assignable peaks for the 27 carbons of the steroid. Therefore, impurity peaks (or their absence) are very readily recognizable from the CMR spectrum of a steroid. We believe, accordingly, that CMR is the method of choice for establishing the identity and purity of II. Since methods have been presented for the preparation of gram amounts of I (*5*) and II (*7*) of >98 mole % proven chemical purity and >99% radiochemical purity, the toxicity and radiopharmaceutical properties of the pure compounds can now be evaluated.

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Reply

Dr. Scott et al. prepared 6β-iodomethyl-19-norcholest-5(10)-en-3β-ol (II) by refluxing 19-iodocholest-5-en-3β-ol (I) in isopropanol. Although the procedure appears to provide adequate separation and identification by CMR and HPLP, the reaction time required to process the rearrangement (48 hr) and the use of isopropanol as a solvent are definite disadvantages, because long reaction time in the rearrangement of I to II in alcoholic solvents leads to the formation of solvolytic or damaged products in considerable amounts. Our result demonstrates that smooth conversion of I into II has been accomplished by the heating of I in acetonitrile without the appreciable formation of by-products, and a subsequent column chromatographic purification results in the isolation of pure II in a good yield (*1*). Thus, our previous procedure also appears sufficient to obtain pure II.

We have already reported that the available synthesis of pure I involves selective hydrolysis of 19-iodocholest-5-en-3β-ol acetate, which is produced in the reaction of sodium iodide with cholest-5-ene-3β,19-diol 19-p-toluenesulphonate 3-acetate in isopropanol, the purity being established unambiguously by PMR (*1*). We are now investigating the solvolytic behavior of 19-tosyloxycholesterol, I, and II, which results will be described in detail elsewhere. Lastly, we have also reported an alternative synthesis of II achieved through the use of 6β-p-toluenesulphonoxymethyl derivatives (*2*).

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Reply

The methods of synthesis of 6-iodomethylcholesterol reported by Scott et al. (*1*) were obvious, insignificant modifications (if any) that had been discussed by Kojima et al. and myself at the presentation of our respective findings at the 1975 Annual Meeting of the Society in Philadelphia.

Neither Kojima et al. (*2*) nor our group at Michigan (*3*) accepted the claim that the NMR scans proved unequivocally the purity of 6-iodomethylcholesterol we had discovered. Before and after our publications (*3,4*), we were working on different ways of identifying impurities and toward new synthetic methods for 6-iodomethylcholesterol that is now established.

There is no doubt in our minds that ^{13}C Nuclear Magnetic Resonance is probably the ultimate tool in establishing the purity of a compound, but at the time we isolated the 6-iodomethylcholesterol, we felt that the sample we had was pure enough to obtain an NMR, a mass spectrum, a melting point, and chromatographic data that warranted its identification and publication.

It is always pleasant to know that other researchers have continued to confirm the pioneering findings at Michigan and have worked diligently to improve our simple synthetic procedures to obtain purer end products.

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Gonadal Radiation Dose and its Genetic Significance in Radiation Therapy of Hyperthyroidism

In their recent paper (*1*) concerning radiation dose to the gonads resulting from the therapeutic dose of ^{131}I in hyperthyroidism, Robertson and Gorman have estimated the ovarian dose as 0.2 rad/mCi administered. They assumed a thyroidal uptake of 80% of the dose and average values for urinary excretion and release rate of thyroidal hormone of 7.2%/hr and 0.18%/hr, respectively.

We have used thermoluminescent dosimeters of LiF and Ca/Dy sulfate, attached to copper intrauterine contraceptive devices, to measure directly the dose to the uterus in a series of patients with Graves' disease. The dose-meters were inserted just before the administration of ^{131}I and were retrieved 1 month later. This method measures only the gamma-radiation dose to the uterus and neglects that resulting from beta particles.

The mean result obtained from our first seven observations was 0.145 (±0.10) rad/mCi administered.

The mean thyroidal uptake in our patients was 74 ± 7%. To compare our results with the calculations of Robertson and Gorman, we assume a gamma dose to the ovaries equal to the dose to the uterus. Furthermore, one must subtract the self irradiation by beta particles from the calculated dose, for this was not measured in our in vivo dosimetry. This component is 0.086 rad/mCi, and the calculated gamma dose to the ovaries is therefore 0/204 − 0.086 = 0.118 rad/mCi. This value is in fair agreement with our measured results, and we feel that our in vivo findings support the validity of the assumptions made by Robertson and Gorman in their calculations.

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Reply

We appreciate the comments by Philippon and Briere relating their measurements of the uterine dose to our calculations of the ovarian dose. Further measurements of this nature as a cross check on radiation dose calculations are to be encouraged.

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Chromatography of ^{99m}Tc Labeled Radiopharmaceuticals

The article by Colombetti, et al. (*1*) confirms our own findings with the MAC-1 kit in the testing of water soluble radiopharmaceuticals. The MAC-1 kit indicates high values