# Solid-Phase Exchange Radioiodination of Aryl Iodides. Facilitation by Ammonium Sulfate

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A mild and simple technique for the synthesis of aryl radioiodides of high specific activity involving a solid-phase exchange between no-carrier-added radioiodide and unactivated aryl iodides is described. Mildly acidic, oxidizing conditions, provided by the in situ thermal decomposition of ammonium sulfate, appear to be necessary for the success of the exchange. Typical isolated radiochemical yields of >70% have been obtained for a variety of aryl iodides, including various aralkyl amines, guanidines, carboxylic acids, and amino acids. Specific activities of up to 100 Ci/mmol of our model compound, (*m*-[$^{125}$I]iodobenzyl)guanidine, have been achieved. Preliminary experiments suggest that, with this technique, interhalogen exchange of aryl bromides with radioiodine is a feasible approach to the preparation of no-carrier-added aryl radioiodides.

## Introduction

Organic compounds containing radioiodine are valuable tools used in biochemistry, pharmacology, toxicology, and nuclear medicine.[2,3] In the latter application, compounds with very high specific activity (1–100 Ci/mmol) are often required. Aromatic compounds activated by electron-donating groups such as phenols and anilines are easily labeled by electrophilic radioiodination in the presence of radioiodide and iodine monochloride,[4] chloramine-T,[5] or "iodogen".[6] No such universal technique is available, however, for the high-specific-activity synthesis of radiolabeled aryl iodides not activated by electron-donating substituents.

Of the possible synthetic approaches to unactivated aryl radioiodides,[7-12] halogen exchange of aryl iodides with radioiodine was chosen for our purposes because (a) aryl iodides are generally stable and readily available, and (b) no separation of the aryl iodide substrate from the radioiodinated product following exchange would be required since they are chemically equivalent. Methods originally developed for the exchange labeling of *o*-iodohippuric acid (1), such as exchange under mildly oxidizing conditions[13]

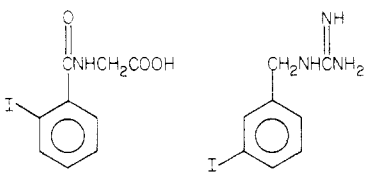


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Table I. Effect of Various Media on Radioiodide Exchange between Na$^{125}$I and mIBG in Solution[a]

| additive | initial pH | final pH | time, h | % exchange (TLC) |
|---|---|---|---|---|
| none | 9.1 | 8.6 | 5 | 0 |
| none | 3.1[b] | 3.1 | 6 | 0 |
| none[c] | | | 17 | trace |
| none[d] | | | 8 | 0 |
| $NaIO_3$ | 6.0[b] | 6.5 | 6 | 0 |
| $NaNO_2$ | 5.8[b] | 6.9 | 6 | trace |
| ICl[e] | 5.8[f] | 5.6 | 5 | trace |

[a] Conditions: mIBG sulfate (1.0 mg, 3.1 μmol), Na$^{125}$I (1.0 mCi), additive (0.5 μmol), in refluxing $H_2O$ (2 mL) unless otherwise noted. [b] pH adjusted with 0.01 N $H_2SO_4$. [c] Solvent: propylene glycol (2 mL) at 150 °C. [d] Solvent: molten acetamide (2 g) at 140 °C. [e] 7.5 μL of 0.0665 M ICl in glacial AcOH. [f] pH adjusted with 1 N NaOH.

Table II. Effect of Ammonium Sulfate on Solid-Phase Exchange between Na$^{125}$I and mIBG[a]

| ammonium sulfate, mg | initial pH | final pH | % exchange (TLC) | % yield (isolated) |
|---|---|---|---|---|
| 0.0 | 7.0 | 7.0 | 0 | 0 |
| 0.1 | 7.0 | 2.8 | 100 | 97 |
| 0.5 | 6.8 | 3.0 | 100 | 94 |
| 1.0 | 6.7 | 3.0 | 100 | 93 |
| 5.0 | 6.2 | 2.9 | 99 | 92 |
| 10.0 | 6.1 | 2.8 | 98 | 90 |

[a] Conditions: mIBG sulfate (1.0 mg, 3.1 μmol), Na$^{125}$I (1.0 mCi), initial volume ($H_2O$), 0.2 mL, solid phase, 140 °C, 30 min.

or in a molten state with radioiodide,[14] either fail altogether when applied to other iodo aromatic compounds or, in the case of the "melt" method, cannot be used for thermally unstable compounds. We report here a mild, solid-phase exchange technique that is generally applicable to the high-specific-activity radioiodination of a variety of unactivated aryl iodides.

## Results

The exchange method reported here is one which evolved during development of (*m*-iodobenzyl)guanidine

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Table III. Effect of Additives on Solid-Phase Radioiodine Exchange between $Na^{125}I$ and mIBG[a]

| additive | amount, μmol | initial pH | final pH | time, min | % exchange (TLC) |
|---|---|---|---|---|---|
| none | | 7.0 | 7.0 | 120 | 0 |
| none | | 7.0 | 7.0 | 15[b] | 0 |
| $(NH_4)_2SO_4$ | 7.6 | 6.8 | 3.0 | 15 | 94 |
| $(NH_4)_2SO_4$[c] | 7.6 | 6.8 | 3.1 | 30 | 7 |
| $H_2SO_4$ | 0.8[d] | 2.5 | 2.5 | 15 | 82[e] |
| $HO_2CCO_2H$ | 0.05[f] | 3.3 | 3.3 | 15 | trace |
| | | 3.3 | 3.4 | 120 | 25 |
| $(NH_4)_2SO_4$ + $SnSO_4$ | 7.6<br>2.3 | 2.8 | 2.5 | 30 | 21 |
| $NaIO_3$ | 0.5 | 6.3[g] | 8.0 | 60 | trace |
| $NaNO_2$ | 0.5 | 6.5[g] | 9.0 | 60 | trace |
| ICl | 0.5[h] | 5.6[i] | 6.7 | 30 | trace[j] |

[a] Conditions: mIBG sulfate (1.0 mg, 3.1 μmol), $Na^{125}I$ (1.0 mCi), initial volume ($H_2O$), 0.2–0.3 mL, solid phase, 140 °C. [b] Heated to melting at 170 °C. [c] Carried out under $N_2$ flow of 5 cm³/min. [d] 0.1 mL of 0.16 N $H_2SO_4$. [e] Not including 25% loss of intital activity as volatile radioiodine. [f] 0.1 mL of 0.01 N $HO_2CCO_2H$. [g] pH adjusted with 0.01 N $H_2SO_4$. [h] 7.5 μL of 0.0665 M ICl in glacial AcOH. [i] pH adjusted with 1 N NaOH. [j] Not including 78% loss of initial activity as volatile radioiodine.

(m-IBG, **2**) as an adrenomedullary imaging agent[15] and, consequently, experiments described herein utilize mIBG (as the sulfate salt) as the model compound. mIBG sulfate has a melting point of 167–168 °C, is moderately soluble in aqueous systems, and is quite stable to heat and hydrolytic conditions.

Results from attempts to exchange label mIBG with $Na^{125}I$ under a variety of previously established conditions are listed in Table I. As indicated, the exchange was unsuccessful in refluxing aqueous solution at both acidic and basic pH as well as in the presence of various oxidizing agents. Similarly, attempts at higher temperatures in propylene glycol[16] or molten acetamide[17] did not result in appreciable exchange. Also unsuccessful was the attempted exchange under "melt" conditions at 170 °C (see Table III).

During initial exploratory experiments to develop a method for radiolabeling mIBG and related (iodoaralkyl)guanidines, it was serendipitiously observed that exchange with $Na^{125}I$ in the absence of solvent was dramatically facilitated by various alkyl and aralkyl ammonium sulfates and bisulfates and, most conveniently, by ammonium sulfate.[18] Table II summarizes the results of an experiment carried out to determine the optimal amount of ammonium sulfate required for the solid-phase exchange of m-IBG with $Na^{125}I$. The exchange failed completely in the absence of ammonium sulfate. In contrast, when carried out with varying amounts of ammonium sulfate (0.1–10.0 mg), the exchange was very efficient and, under the conditions employed, unrelated to the amount of ammonium sulfate present.

A possible relationship between the extent of exchange and the pH lowering that occurs during the course of the reaction is noteworthy. All reactions done in the presence of ammonium sulfate resulted in virtual quantitative exchange (by TLC) after only 30 min at 140 °C. During this 30-min period, the pH dropped from 6–7 for the initial aqueous solution to ca. 3 after heating to dryness and redissolving the reaction mixture in water. Concurrently, the condensate had a pH of 8.5–9.5, owing to the presence of ammonia (from the thermal decomposition of ammonium sulfate[19]). A similar pH lowering was not observed in the absence of ammonium sulfate nor, as previously mentioned, was any exchange detected. In the ammonium sulfate facilitated exchanges, the loss of volatile radioiodine, reflected in the isolated yields and consistent with in situ iodide oxidation, was always less than 10% but increased slightly with lower initial pH.

Solid-phase exchange of mIBG with radioiodine was carried out in the presence of various additives to determine if the reaction is facilitated, as suggested by the experiments with ammonium sulfate, by either an acidic or oxidizing medium, or both. As Table III indicates, when the exchange was carried out under acidic, reducing conditions (i.e., in the presence of oxalic acid, a reducing acid, or in the presence of ammonium sulfate with stannous sulfate as a reducing agent), the radioiodine incorporation into mIBG was well below that observed in the presence of ammonium sulfate alone. A similar effect was obtained when oxygen was excluded during exchange with ammonium sulfate by running the reaction under a constant nitrogen flow. These observations demonstrate the requirement for at least a mildly oxidizing environment. Similarly, the exchange did not proceed in the presence of such oxidizing agents as sodium iodate, sodium nitrite, and iodine monochloride, at a near neutral pH of ca. 6, suggesting that acidic conditions, in addition to mildly oxidizing conditions, are necessary for the success of the exchange.

The data represented in Table III also demonstrate that, in addition to ammonium sulfate, the exchange proceeds quite well in the presence of sulfuric acid. However, in contrast to the minimal loss of volatile radioiodine with ammonium sulfate, 25% of the initial radioactivity was lost after only 15 min at 140 °C in the presence of sulfuric acid.

The results of experiments summarized in Table IV emphasize the inhibitory effect of water on the ammonium sulfate facilitated exchange of mIBG. Again, exchange was quantitative after only 30 min at 140 °C when carried out in an "open" system allowing escape of the water initially present as well as the ammonia from the thermal decomposition of ammonium sulfate. Under reflux conditions, or at higher temperatures in a septum-sealed V-vial containing water, the facilitative effect of the ammonium sulfate on the exchange was not observed. However, failure of the exchange under these conditions could be due either to an inhibitory effect of the water or to inhibition caused by the prevention of ammonia evolution from ammonium sulfate decomposition. The last entry in Table IV distinguishes between these two possibilities. In this experiment, a solution of mIBG and ammonium sulfate was

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Table IV. Effect of Water on Solid-Phase Exchange between $Na^{125}I$ and mIBG[a]

| conditions | initial pH | final pH | time, h | % exchange (TLC) |
|---|---|---|---|---|
| solid phase[b] | 6.7 | 3.0 | 0.5 | 100 |
| refluxing $H_2O$ (2 mL) | 7.1 | 7.0 | 6.0 | 0 |
| sealed vial with $H_2O$ (0.5 mL) | 6.7 | 6.7 | 2.0 | 9 |
| sealed vial with $H_2O$ (0.05 mL)[c] | | | 0.5 | 15 |

[a] Conditions: mIBG sulfate (1.0 mg, 3.1 μmol), $Na^{125}I$ (1.0 mCi), $(NH_4)_2SO_4$ (1.0 mg, 7.6 μmol), oil bath temperature 140 °C. [b] Initial volume ($H_2O$) 0.2 mL. [c] mIBG sulfate + $(NH_4)_2SO_4$ in 0.2 mL of $H_2O$ heated to dryness for 30 min at 140 °C in an open vial before $Na^{125}I$ addition in 0.05 mL of $H_2O$.

heated to dryness for 30 min at 140 °C in an open 5-mL serum vial—conditions allowing the escape of the ammonia produced. After cooling and addition of an aqueous solution (50 μL) of $Na^{125}I$, the vial was sealed with a Teflon-lined septum and heated at 140 °C for an additional 30 min. Even though most of the added water had condensed on the septum, giving a dry appearance to the reaction mixture, only 15% exchange was obtained. A vial similarly treated in which the water was allowed to escape during heating with $Na^{125}I$ resulted in quantitative exchange. This suggests that the exchange does indeed occur in the solid phase and the presence of water, even in minute amounts, inhibits the reaction.

In order to investigate possible involvement of catalysis by the surface of the glass reaction vessel or by trace metals adhering to the surface, we carried out the solid-phase exchange with mIBG, $Na^{125}I$, and ammonium sulfate in vials pretreated by different methods. These pretreatments included the following: (a) rinsing with $H_2O$, (b) rinsing with dichromate cleaning solution followed by $H_2O$, (c) rinsing with 50% KOH followed by $H_2O$, 50% $H_2SO_4$, $H_2O$, and (d) silylation overnight with 5% hexamethyldisilazane in toluene followed by rinsing with toluene, $CH_3OH$, and $H_2O$. After 15 min at 140 °C, incorporation of radioiodine into mIBG in each case was >95%, indicating that neither glass nor trace metal catalysis plays a major role in the exchange.

Table V lists many of the iodoaromatic compounds that have been radiolabeled by the ammonium sulfate facilitated exchange developed during the course of this study. Even though many of the examples listed were carried out before conditions for the procedure were optimized, isolated radiochemical yields typically >70% were obtained for a variety of compounds. The isomeric identity of the radiolabeled compound following exchange was determined by HPLC analysis for several of the examples listed. In all cases examined, the isomeric location of the radioiodine was identical with that of the iodine in the cold compound used in the exchange. The specific activity values given in the table are those obtained in typical preparative procedures and in no way represent the maximum attainable by this technique. Indeed, in at least five separate experiments using only 25 μg of mIBG sulfate, exchange in the presence of 5–10 mCi of $Na^{125}I$ and 4 mg of ammonium sulfate, labeled compound was isolated in 40–84% radiochemical yield with specific activities of 30–100 Ci/mmol.

Among the compounds listed in Table V, no major differences in the rates of exchange were noted with the exception of the iodobenzoic acids. While good radiochemical yields were obtained for a variety of compounds by heating at 140–160 °C for 1–4 h, 12 h at 140 °C was required to produce ca. 30% exchange of both *m*- and *p*-iodobenzoic acids. In contrast, the radiochemical yield of *o*-iodobenzoic acid was 63% after only 15 min, implicating participation of the *o*-carboxyl group in the exchange process. Further heating at 140 °C for 24 h, however, reduced the yield to 35%.

Application of the technique to the production of radiolabeled aryl iodides via interhalogen exchange is rep-

Table V. Solid-Phase Exchange Radioiodination of Representative Compounds in the Presence of Ammonium Sulfate[a]

| no. | compound[b] | no. of trials | % yield (isolated)[c] | sp act., mCi/mg |
|---|---|---|---|---|
| 2 | (*m*-iodobenzyl)guanidine (mIBG)[d] | 30 | 92 (90-98) | 0.8-2.0[e] |
| 3 | (*p*-iodobenzyl)guanidine (pIBG)[d] | 4 | 85 (50-98) | 0.8-1.1 |
| 4 | (3,4-diiodobenzyl)guanidine[d] | 1 | 70 | 1.0 |
| 5 | (3,5-diiodobenzyl)guanidine | 1 | 50 | 1.0 |
| 6 | D,L-α-methyl-mIBG[d] | 1 | 72 | 1.0 |
| 7 | D-α-methyl-mIBG | 1 | 40 | 1.0 |
| 8 | L-α-methyl-mIBG | 1 | 32 | 1.0 |
| 9 | D,L-α-methyl-pIBG[d] | 3 | 74 (41-90) | 0.8-1.2 |
| 10 | (*m*-iodophenethyl)guanidine | 1 | 94 | 2.2 |
| 11 | (*p*-iodophenethyl)guanidine | 2 | 83 (75-90) | 1.0 |
| 12 | (*m*-iodophenyl)guanidine | 1 | 78 | 2.2 |
| 13 | (*m*-iodobenzyl)urea | 1 | 70 | 1.0 |
| 14 | *p*-iodophenethylamine | 1 | 90 | 1.2 |
| 15 | *p*-iodo-D,L-α-methylphenethylamine | 1 | 43 | 0.9 |
| 16 | *p*-iodo-D,L-phenylalanine[d,f] | 7 | 37 (22-58) | 0.5-3.0 |
| 17 | 5-iodo-D,L-tryptophan[d,f] | 2 | 59 (50-68) | 0.7-2.0 |
| 18 | 6-iodo-D,L-tryptophan[d,f] | 3 | 36 (14-53) | 0.2-2.7 |
| 19 | *o*-iodobenzoic acid[d,g] | 1 | 63 | 3.0 |
| 20 | *m*-iodobenzoic acid[d,h] | 1 | 40 | 1.0 |
| 21 | *p*-iodobenzoic acid[d,h] | 1 | 33 | 1.0 |
| 22 | (*p*-iodophenyl)acetic acid | 1 | 96 | 0.4 |
| 23 | (*p*-bromophenyl)acetic acid[i] | 1 | 34 | 0.1 |

[a] Conditions: compound (0.2-2.0 mg), $Na^{125}I$ (5-20 mCi), $(NH_4)_2SO_4$ (2-5 mg), initial volume ($H_2O$) 0.2-1.0 mL, solid phase, 120-160 °C, 1-4 h. [b] Compounds **2-12**, **14** were the sulfate salts; compound **15** was the HCl salt. [c] Unbound radioiodine removed on a Cellex-D (Bio-Rad) column. [d] Radiochemical purity confirmed by HPLC following exchange. [e] Specific activities for five 'high-specific-activity' syntheses of 100-300 mCi/mg in yields of 40-84% were obtained, using only 25 μg of mIBG sulfate, 5-10 mCi of $Na^{125}I$, and 4 mg of $(NH_4)_2SO_4$. [f] Significant activity was lost due to strong adherence to the reaction vessel. [g] After 15 min at 160 °C. [h] After 12 h at 160 °C. [i] Radiolabeled product was (*p*-[$^{125}$I]iodophenyl)-acetic acid.

resented by the last entry in Table V. A 34% radiochemical yield of (*p*-[$^{125}$I]iodophenyl)acetic acid was obtained by exchange of 2.5 mg of (*p*-bromophenyl)acetic acid with 1 mCi of Na$^{125}$I in the presence of 3 mg of ammonium sulfate at 160 °C for 2 h. (*p*-Iodophenyl)acetic acid treated similarly resulted in >95% yield of labeled product.

## Discussion

Solid-phase exchange between no-carrier-added radioiodide and unactivated aryl iodides in the presence of ammonium sulfate is a mild, simple technique for the production of radiolabeled iodo aromatic compounds of high specific activity. The function of the ammonium sulfate is seemingly to cause a gradual increase in the acidity of the exchange medium via in situ thermal decomposition to ammonium bisulfate with resulting evolution of ammonia.[19] The conditions employed in this study are, however, insufficient to cause total conversion to bisulfate;[19] thus, the exchange is likely occurring in a medium containing both ammonium sulfate and bisulfate. The fact that a constant pH of 3 is obtained following exchange with varying amounts of ammonium sulfate suggests that an equilibrium mixture of ammonium sulfate and bisulfate and is established which provides a favorable acidic environment for the exchange process. Because of this, the method is particularly convenient as a general radioiodination technique—the amount of ammonium sulfate required for successful exchange is not critical and the acidity of the exchange medium is internally regulated.

The success of the exchange, which is diminished in the presence of reducing agents or the absence of oxygen, appears to be dependent on the in situ oxidation of the radioiodide in addition to the acidification of the reaction medium. The formation and loss of volatile (molecular) radioiodine which occurs to a small extent during the ammonium sulfate facilitated exchange is a reflection of the oxidizing conditions that exist. However, molecular radioiodine per se is probably not the reactive species responsible for the efficient exchange since no exchange was observed under conditions in which it is formed, i.e., in the presence of sodium iodate, sodium nitrite, or iodine monochloride at a slightly acidic pH. Whether the reactive species is derived from molecular radioiodine or formed via a separate oxidation route remains to be determined. Since sulfuric acid can be substituted for ammonium sulfate to promote the exchange, the presence of the ammonium group is not an absolute requirement. However, loss of volatile radioiodine is significantly decreased when the initial pH is neutral to slightly basic, as in the ammonium sulfate facilitated exchanges, rather than acidic.

Discussion of the mechanism of the exchange and of the nature of the reactive radioiodine species responsible is complicated by the nonclassical behavior of iodine in very small (1 mCi of Na$^{125}$I = 0.46 nmol) quantities. For example, several oxidation products of iodine which are quite unstable in macroquantities, such as HIO or $HIO_2$, are easily formed in no-carrier-added solutions of radioiodide and can exist for long periods of time in such solutions.[20,21] Nevertheless, results of this study, in which participation of an oxidation product of radioiodide is implicated and compounds containing electron-withdrawing substituents such as *m*- and *p*-iodobenzoic acids are less efficiently radiolabeled, are more consistent with an electrophilic rather than a nucleophilic process. Additional studies probing the mechanism of the exchange are in progress.

A popular technique in recent literature for the radioiodination of a variety of iodoaromatic compounds is the "melt" method introduced by Elias[14] for radioiodination of *o*-iodohippuric acid. In this technique, the compound to be labeled is heated to a molten state in the presence of radioiodide to effect exchange. The ammonium sulfate technique, carried out at temperatures below the substrate melting point, offers milder conditions which are particularly important, as previously mentioned, in the radioiodination of thermally unstable compounds. In spite of the fact that radioiodide exchange of mIBG was successful with the ammonium sulfate technique but failed under "melt" conditions, similarities exist between these techniques. For example, mildly acidic, oxidizing conditions appear to be important for the ammonium sulfate facilitated exchange of mIBG, as shown in this study. Similarly, radioiodide exchange of *o*-iodohippuric acid under melt conditions is always carried out under acidic conditions in which at least a portion of the iodihippurate is as the free acid.[22] Indeed, the exchange fails dramatically when run under neutral or basic conditions,[22] suggesting a similar situation for the failure of the melt exchange of mIBG in the absence of ammonium sulfate. Evidence for the existence of mildly oxidizing conditions during the melt exchange of *o*-iodohippuric acid is provided by Sinn et al.[23] who noted that exchange is virtually quantitative even when 25% of the radioiodine used in the exchange is in the form of iodate. Disproportionation of the radioiodide and radioiodate to radioiodine under the acidic conditions of the exchange is certainly likely.

The ammonium sulfate method is particularly suited, but not limited, to the radioiodination of compounds which are at least moderately soluble in aqueous systems. This includes many pharmacologically active compounds of interest as potential radiopharmaceuticals. In such cases, the fact that no potentially toxic additives or organic solvents are introduced during or following the exchange greatly simplifies the workup and formulation of the radiolabeled product for in vivo administration. In most instances, removal of the unbound radioiodine on a simple ion-exchange column is the only workup required. In contrast, radioiodine exchange carried out in, for example, molten acetamide[17] or with copper salts in $Me_2SO$[24] necessitates the removal of the organic medium prior to formulation requiring additional and more involved procedures. The ammonium sulfate method requires no elaborate techniques, glassware, or apparatus to make it suitable for general, routine radiopharmaceutical preparation. The method has been used exclusively in the production of [$^{131}$I]mIBG and [$^{123}$I]mIBG for clinical studies at The University of Michigan.[25,26] The scope of the reaction with a variety of aryl iodides, in addition to those of Table V, is also being evaluated.

The greatest potential utility of the ammonium sulfate method is in the application to very high specific activity syntheses. Samples of [$^{125}$I]mIBG at 100 Ci/mmol, representing 5% of the theoretical maximum specific activity, have been easily prepared by use of the technique. For carrier-free aryl radioiodides, interhalogen exchange of the

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homologous bromo compound in the presence of ammonium sulfate followed by HPLC isolation promises to be a very viable approach. Efforts in this regard are currently underway.

## Experimental Section

The iodine-125 used in this study was a no-carrier-added solution of $Na^{125}I$ (ca. 500 mCi/mL) in reductant-free 0.1 N NaOH obtained from Union Carbide or New England Nuclear. The commercial solution was diluted immediately prior to use with deionized, distilled water to a radioiodide concentration of 10–20 mCi/mL. The pH of the diluted solution, as determined with a micro pH electrode, was 8.0–9.5. Radioactivity was quantified with a Capintec Model CRC-2 radioisotope calibrator or a Packard Model 5260 autogamma counter. Both instruments were calibrated against an NBS standard solution of $Na^{125}I$.

Synthesis of compounds **2–13** is reported elsewhere.[27] Compound **15** was synthesized according to Binovic et al.[28] The iodotryptophans **17** and **18** were synthesized by modifications of the procedure of Lambrecht et al.[29] All other compounds used in this study were obtained from commercial sources.

**Chromatographic Analyses.** TLC analyses were performed on 2.5 × 20 cm silica gel coated glass plates (Whatman K6F). The reaction solutions were spotted overlying a spot of 0.1 M NaI to minimize loss of volatile radioiodine by air oxidation. The plates were analyzed on a Packard Model 720 radiochromatogram scanner immediately after development and drying. Percent exchange was determined by integration of the chromatogram peaks (by the product of peak height × peak width at half-height) and calculated as the ratio of activity of exchanged product to the total activity chromatographed, averaged for duplicate chromatograms. The solvent systems and the respective $R_f$ values (with variability generally within ±5%) of free iodide and iodate for each system are as follows: A, EtOH/EtOAc (1/1), 0.75, 0.07; B, EtOH/EtOAc/concentrated $NH_4OH$ (20/20/1), 0.60, 0.06; C, EtOH/concentrated $NH_4OH$ (3/1), 0.90, 0.23; D, nBuOH/AcOH/$H_2O$ (5/2/1), 0.65, 0.06; E, *n*-BuOH/AcOH/$H_2O$ (6.5/1.5/2.5), 0.45, 0.06; F, EtOAc/EtOH (95/5), 0.05, 0.00. $R_f$ values for the compounds listed in Table V with the solvent systems used are as follows: **2–12**, 0.02 (A), 0.02 (B), 0.15 (C); **13**, 0.70 (C), 0.19 (F); **14**, **15**, 0.12 (A), 0.15 (B), 0.75 (C); **16–18**, 0.15 (B), 0.50 (D); **19–21**, 0.32 (B), 0.75 (E); **22**, **23**, 0.30 (B), 0.80 (E).

Radiochemical purity for some of the $^{125}I$-labeled compounds listed in Table V was confirmed by HPLC analysis. A Waters Model 272 liquid chromatograph equipped with a Radiomatic Flo-one radioactive flow detector (200-μL solid scintillator cell) was used, employing simultaneous ultraviolet (254 or 280 nm) and radioactivity detection. A Waters μ-Bondapak C-18 column (4.6 × 250 mm) was used for all analyses with the following solvent systems: A, 0.1M $NaH_2PO_4$/THF (88/12); B, 0.05 M $KH_2PO_4$/$CH_3CN$ (85/15); C, 3% $Et_3N$ (pH 7.8 with $H_3PO_4$)/$CH_3CN$ (83/17). The $k'$ values for the compounds listed in Table V that were analyzed by HPLC are as follows: solvent A, 5.2 (**2**), 6.7 (**3**), 22.2 (**4**), 7.7 (**6**), 10.4 (**9**); solvent B, 2.5 (**16**), 4.8 (**17**), 5.4 (**18**); solvent C, 1.7 (**19**), 6.9 (**20**), 7.6 (**21**). In order to minimize the amount of radioactivity required for the HPLC analyses, we diluted the labeled product with unlabeled carrier compound and sodium iodide prior to injection.

**Exchange Radioiodinations. General Procedures.** Exchanges under reflux conditions were run in either 5- or 10-mL Pyrex flasks equipped with a reflux condenser connected to a sodium thiosulfate trap. Solid-phase reactions were performed either in 5-mL Pyrex flasks (with a septum-stoppered side arm) equipped with a specially designed Erlenmeyer-shaped condenser allowing for quick removal of the initial solvent by distillation or in 1- or 5-mL septum-closed multidose vials with a disposable glass syringe as the distillate condenser and receptacle. In some experiments, the reaction was carried out under a flow of nitrogen or air (ca. 5–10 $cm^3$/min) introduced via an 18 gauge needle through the septum of the reaction vessel. Heating was accomplished with an oil bath, and the temperature reported for nonrefluxing reactions are those of the equilibrated oil bath. At appropriate time intervals, TLC analysis and pH measurement (via combination microelectrode) were carried out either directly (refluxing solutions) or after redissolution of the reaction medium in a volume of solvent equal to the initial volume (solid-phase reactions).

For preparative radioiodide exchange of the compounds listed in Table V, 0.2–2.0 mg of substrate, 5–20 mCi or $Na^{125}I$, and 2–5 mg of ammonium sulfate were dissolved in 0.4–1.0 mL of water or aqueous alcohol, the mixture was heated to dryness, and the dry reaction mixture was maintained at 120–160 °C (below the melting point of the substrate) for 1–4 h. The reaction mixture was then dissolved in 1–4 mL of water or 5 mM pH 4.5 sodium acetate buffer. Passage of the resulting solution through a Cellex-D (Bio-Rad) anion-exchange column (1 × 4 cm or 1.5 × 5 cm), eluted with the aforementioned buffer, removed the unbound radioiodine from the final preparation. The product concentration of the eluant from the Cellex-D column was determined by ultraviolet absorption at its $\lambda_{max}$ and calculated from a standard curve of unlabeled compound generated on a Bausch and Lomb Spectronic 70 spectrophotometer against a blank of acetate buffer. Specific activity of the exchanged product was then determined from the specific concentration obtained by ultraviolet spectroscopy or by estimation based on starting quantities of substrate and final radiochemical yield.

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