2-(Alkylamino)tetralin Derivatives: Interaction with 5-HT_{1A} Serotonin Binding Sites

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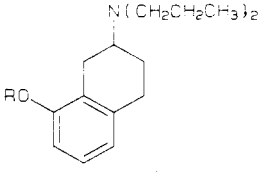
8-Hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT) is a selective 5-HT_{1A} serotonin agonist. Derivatives of 8-OH-DPAT with amine substituents larger or more bulky than *n*-propyl appear to be inactive in a presynaptic biochemical assay measuring agonist-induced feedback inhibition of 5-HT synthesis but have never been examined in brain binding assays. A series of *N*-phenylalkyl derivatives of 8-methoxy-2-aminotetralin was evaluated at [^{3}H]-8-OH-DPAT-labeled 5-HT_{1A} sites in rat brain hippocampal membranes. All of the phenylalkyl derivatives displayed significant affinity for these sites and, of the agents examined, the 3-phenylpropyl 8-hydroxy analogue appears to be optimal and had an affinity (K_i = 1.9 nM) comparable to that of 8-OH-DPAT (K_i = 1.2 nM). In addition, the presence of an oxygen-containing substituent at the 8-position of the tetralin ring is not necessary for good affinity, and secondary amines and tertiary amines displayed equal affinity at central 5-HT_{1A} binding sites. 5-HT_{1A} sites are found both pre- and postsynaptically; thus, differences observed in the biochemical assay as compared to the results of the present binding study could be due to different structural requirements of these two receptors. This seems unlikely, however, because there was little difference in the affinities of several selected analogues for striatal versus hippocampal binding sites. Because we have now demonstrated that amine substituents larger than propyl, and an unsubstituted 8-position, are well tolerated by central 5-HT_{1A} sites, future studies aimed at the development of new serotonergic tetralin analogues need not be limited to *N*-propyl or 8-hydroxy derivatives of 2-aminotetralin.

8-Hydroxy-2-(di-*n*-propylamino)tetralin (8-OH-DPAT; **1a**) was identified as a serotonin (5-HT) agonist by Arvidsson and co-workers[1-3] and was subsequently shown by others to bind with high affinity and selectivity to central 5-HT_{1A} sites.[4-7] In the biochemical assay used by Arvidsson et al.[1,3,8] (agonist-mediated feedback inhibition of 5-HT synthesis, presumably through presynaptic autoregulatory receptors), di-*n*-propyl substitution on the terminal amine seemed optimal for agonist activity.[3] Monoalkyl derivatives, as well as compounds with substituents smaller than propyl (e.g., dimethyl, diethyl), were less potent. Derivatives with amine substituents larger than propyl (e.g., *n*-butyl, *n*-octyl, benzyl) resulted in decreased activity or were inactive; similar results were seen with branched alkyl derivatives.[3] On the basis of their findings, Arvidsson and co-workers[3] proposed that the 5-HT receptor (now thought to be a 5-HT_{1A} receptor) has a cavity near the position where the terminal amine group interacts that can accommodate ethyl or *n*-propyl groups, but that is too small to accommodate larger or more bulky substituents.



1a R = H
1b R = CH_3

[^{3}H]-8-OH-DPAT binds with nanomolar affinity both to presynaptic and postsynaptic 5-HT_{1A} binding sites in mammalian brain.[9,10] Gozlan et al.,[9,11] for example, have reported that those sites labeled by [^{3}H]-8-OH-DPAT in the hippocampus constitute (predominantly) 5-HT_{1A} sites (corresponding to postsynaptic 5-HT_{1A} sites) and have termed the (presynaptic) sites labeled by [^{3}H]-8-OH-DPAT in the striatum as 5-HT_3 sites. (*Note*: These 5-HT_3 sites should not be confused with the 5-HT_3 sites defined by currently accepted 5-HT receptor nomenclature.[12]) Because these two sites appear to be different, there is a possibility of ultimately exploiting potential differences in binding to develop selective presynaptic or postsynaptic 5-HT_{1A} agents. Though Arvidsson et al.[3] have demonstrated the ineffectiveness of aminotetralin derivatives bearing bulky amine substituents in a presynaptic biochemical model, aminotetralin derivatives with substituents larger than propyl have never been examined by radioligand binding techniques. The purpose of the present investigation, then, was to examine several such derivatives in order to determine if they would bind at hippocampal 5-HT_{1A} sites.

Chemistry

2-Tetralone or 8-methoxy-2-tetralone[13] was reductively

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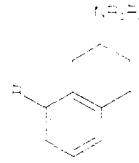
Table I. 2-(Alkylamino)tetralin Derivatives: Physical Properties and Binding Data



compound	R	R_1	R_2	method[a]	% yield	mp,[b] °C	formula[c]	5-HT_{1A} binding[d] K_i,[a] nM	5-HT_{1A} binding[d] Hill slope[e]
2	H	H	*n*-Pr	A	68	244–245[f]		19 ± 2	0.74 ± 0.02
3	OMe	H	H	A	39	275–278[g]		53 ± 5	0.77 ± 0.05
4	OMe	H	*n*-Pr	A	25	191–193[h]		2.3 ± 0.3	0.80 ± 0.03
5	OMe	Me	*n*-Pr	A	37	181–182[i]		2.1 ± 0.2	0.91 ± 0.01
6	OMe	Me	CH_2Ph[j]	A	37	159–161	$C_{19}H_{23}NO{\cdot}HCl{\cdot}^1/_2H_2O$	78 ± 3	0.91 ± 0.02
7	OMe	H	$(CH_2)_2Ph$	A	13	208–209	$C_{19}H_{23}NO{\cdot}HCl$	44 ± 3	0.85 ± 0.04
8	OMe	Me	$(CH_2)_2Ph$	A	23	184–186	$C_{20}H_{25}NO{\cdot}HCl$	7.9 ± 0.6	0.90 ± 0.03
9	OMe	H	$(CH_2)_3Ph$	A	63	221–223	$C_{20}H_{25}NO{\cdot}HCl$	2.5 ± 0.1	0.83 ± 0.05
10	OMe	H	$(CH_2)_4Ph$	A	32	191–193	$C_{21}H_{27}NO{\cdot}HCl$	5.6 ± 0.2	0.92 ± 0.05
11	H	H	$(CH_2)_3Ph$	A	79	225 dec	$C_{19}N_{23}N{\cdot}HCl$	13 ± 2	1.06 ± 0.06
12	OH	H	$(CH_2)_3Ph$	B	31	254–256	$C_{19}H_{23}NO{\cdot}HCl$	1.9 ± 0.2	0.87 ± 0.01
13	OMe	Me	$(CH_2)_3Ph$	C	40	148–150	$C_{21}H_{27}NO{\cdot}HCl$	2.0 ± 0.3	0.94 ± 0.05
14	OBz[j]	H	$(CH_2)_3Ph$	D	52	176–178	$C_{26}H_{29}NO{\cdot}HCl$	10 ± 5	0.91 ± 0.06
8-OH-DPAT (**1a**)	OH	*n*-Pr	*n*-Pr					1.2 ± 0.1	1.01 ± 0.01
8-OMe-DPAT (**1b**)	OMe	*n*-Pr	*n*-Pr					1.3 ± 0.3	0.78 ± 0.07

[a] Method A: Reductive amination (catalytic, except for **3**) of the appropriate tetralone; see **9** in the Experimental Section. B: O-Demethylation of **9** using HBr. C: Reductive methylation of **9**. D: Alkylation of **12** with benzyl bromide. [b] All compounds were recrystallized from absolute EtOH/Et_2O except for **9**, **10**, **11**, and **12**, which were recrystallized from absolute EtOH. [c] All compounds analyzed for C, H, and N are within ±0.4% of theory. [d] [^{3}H]-8-OH-DPAT-labeled 5-HT_{1A} sites. [e] K_i values and Hill slopes are followed by ±SEM. [f] Reference 21, mp 249–250 °C. [g] Reference 18, mp 273–275 °C. [h] Reference 18, mp 189–190 °C. [i] Reference 18, mp 180–182 °C. [j] Ph = phenyl; Bz = benzyl.

aminated with the appropriate amine with H_2 and PtO_2 to yield products **2** and **4–11** (Table I) according to the general procedure of Hacksell et al.[8] Compound **3** was synthesized by the reductive amination of 8-methoxy-2-tetralone with ammonium acetate and $NaCNBH_3$ according to the general procedure of Birch et al.[14] The 8-methoxy group of compound **9** was demethylated with 48% HBr to afford compound **12**, which was alkylated with benzyl bromide. Reductive methylation of **9** by the Eschweiler–Clarke procedure yielded compound **13**.

Results and Discussion

Methylation of the hydroxyl group of 8-OH-DPAT (**1a**; K_i = 1.2 nM) to give 8-OMe-DPAT (**1b**; K_i = 1.3 nM) has no effect on affinity at hippocampal 5-HT_{1A} sites (Table I). In fact, complete removal of the hydroxy/methoxy group has relatively little effect; a comparison of **4** with **2**, or **11** with **9** (or **12**), reveals that lack of this group decreases affinity by less than 1 order of magnitude (Table I). Furthermore, an *O*-benzyl group seems to be well tolerated (i.e., **14**; K_i = 10 nM). The primary amine analogue of 8-OMe-DPAT (i.e., 8-OMe AT, **3**) possesses an affinity for 5-HT_{1A} sites that is 40-fold less than that of 8-OMe-DPAT (**1b**) itself. This finding is consistent with the 60-fold decrease reported by Hoyer et al.[7] for 8-OH-DPAT as compared with its primary amine derivative. However, the necessity of the two *n*-propyl groups of **1b** does not seem all that important when the affinity of the monopropyl derivative **4** (K_i = 2.3 nM), or that of its *N*-methyl derivative **5** (K_i = 2.1 nM), is compared with that of the tertiary amine **1b** (K_i = 1.3 nM) (Table I). The affinity of the *N*-benzyl derivative **6** (K_i = 78 nM) is not significantly different from that of the unsubstituted derivative **3** (K_i = 53 nM); furthermore, as the chain length that separates the phenyl group from the amine is increased, affinity increases. Comparing affinities within the series **7**, **9**, **10**, or in the series **6**, **8**, **13**, it is seen that maximal affinity is obtained when the phenyl group is separated from the amine by a three-carbon chain. Indeed, the affinities of the two three-carbon chain (i.e., phenylpropyl) analogues, 8-OMe-PPAT (**9**; K_i = 2.5 nM) and its *N*-methyl derivative **13** (K_i = 2.0 nM), are not significantly different from those of **4** (K_i = 2.3 nM) or 8-OMe-DPAT (**1b**), or, for that matter, from that of 8-OH-DPAT itself (Table I). At this point, however, insufficient data are available to allow us to determine whether it is the length of the alkyl chain or the global lipophilicity of the amine substituent that is important for affinity. As anticipated from the results with **1a** and **1b**, O-demethylation of **13** (i.e. 8-OH-PPAT; **12**) had no effect on affinity (**12**, K_i = 1.9, and **13**, K_i = 2.0 nM; Table I).

The results obtained in the present study contrast sharply with what might have been expected on the basis of the results of Arvidsson and co-workers.[3] It is clear that hippocampal 5-HT_{1A} sites can accommodate amine substituents larger than *n*-propyl groups and that monoalkyl derivatives can bind with high affinity; 8-OH-PPAT (**12**), 8-OMe-PPAT (**9**), and its *N*-methyl analogue **13**, for example, bind at 5-HT_{1A} sites with affinities comparable to those of 8-OH-DPAT (**1a**) and 8-OMe-DPAT (**1b**). Whereas some of the differences in results of the two studies might be attributable to pharmacokinetic peculiarities, other explanations must be sought. 8-OH-DPAT (**1a**) binds with high affinity both to presynaptic and postsynaptic 5-HT_{1A} sites;[9,10,15] because the binding characteristics of these sites might be different, structure–activity relationships developed by using a presynaptic model might differ from those developed from radioligand binding studies with hippocampal homogenates. We explored this possibility by examining the binding of several of the aminotetralin analogues at striatal sites. The affinities of **9** (K_i = 2.9 ± 0.3 nM), **12** (K_i = 2.2 ± 0.2 nM), **13** (K_i = 4.0 ± 0.3 nM), and **14** (K_i = 16 ± 2 nM) for striatal

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sites labeled by [^{3}H]-8-OH-DPAT (Hill slopes = 0.8–1.1) did not differ significantly from their affinities for hippocampal 5-HT_{1A} sites (Table I). Thus, it is unlikely that the observed differences can be attributed to different structural requirements for hippocampal versus striatal sites (if these are indeed good models of pre- versus postsynaptic binding). [On the other hand, while this work was in progress it was reported that [^{3}H]-8-OMe-2-[*N*-*n*-propyl-*N*-[3-[(2-nitro-4-azidophenyl)amino]propyl]-amino]tetralin, another aminotetralin derivative with a bulky terminal amine group, not only irreversibly labels hippocampal 5-HT_{1A} sites, but it displays only modest affinity for presynaptic (i.e., striatal) sites.[16-18]] A second explanation relates to the fact that Arvidsson and coworkers[1] examined (and based their hypotheses on the results of) agonist activity whereas we are examining binding affinity; thus, the possibility exists that some of the present high-affinity agents may lack intrinsic activity (i.e., they may constitute 5-HT_{1A} antagonists). Finally, we cannot overlook the possibility that the 5-HT sites mediating feedback control of 5-HT biosynthesis may constitute a different type of 5-HT (or 5-HT_{1A}) receptor than that present in rat hippocampus or striatum.

In summary then, hippocampal 5-HT_{1A} binding sites bind the *O*-methyl analogue of 8-OH DPAT (i.e., **1b**) and its mono *n*-propyl derivative (i.e., **4**) with an affinity comparable to that of 8-OH DPAT (**1a**) itself. Furthermore, there is associated with the terminal amine site a region of bulk tolerance that can easily accommodate bulky substituents. In three separate cases (i.e., **2**/**11**, **4**/**9**, **5**/**13**), it is seen that the phenylpropyl derivative binds as well as the corresponding *n*-propyl parent. Derivatives with large amine groups and/or that lack an 8-hydroxy group bind at hippocampal 5-HT_{1A} sites; the *N*-phenylpropyl derivatives also appear to bind well at striatal sites labeled by [^{3}H]-8-OH-DPAT. The results of present study are significant because they suggest that future studies with aminotetralin analogues need not be limited to derivatives with small terminal amine groups or to 8-hydroxy analogues; such agents might also provide entry to a novel class of 5-HT_{1A} antagonists. The affinity of the 8-benzyloxy analogue also reveals a new area for future exploitation.

Experimental Section

Synthesis. Proton magnetic resonance spectra were obtained with a JEOL FX90Q spectrometer with tetramethylsilane as an internal standard; infrared spectra were recorded with a Nicolet 5ZDX FT-IR spectrometer. Spectral data are consistent with the assigned structures. Melting points were determined with a Thomas-Hoover capillary melting point apparatus and are uncorrected. Elemental analysis was performed by Atlantic Microlab (Atlanta, GA), and values are within 0.4% of theory.

8-Methoxy-2-aminotetralin Hydrochloride (3). This compound was prepared according to the general procedure of Birch et al.[14] 8-Methoxy-2-tetralone[13] (1.1 g, 6.2 mmol) in MeOH (20 mL) was added to $NaCNBH_3$ (560 mg, 8.9 mmol) and ammonium acetate (4.8 g, 62.4 mmol). The reaction mixture was allowed to stir at room temperature for 89 h and was then acidified to pH <2 with concentrated HCl. The MeOH was removed in vacuo. A solution of the resulting white solid in H_2O (75 mL) was extracted with Et_2O (3 × 50 mL). The aqueous layer was basified with solid KOH to pH >10 and extracted with Et_2O (3 × 50 mL). The combined organic fractions were dried ($MgSO_4$), and the solvent was removed under reduced pressure. The resulting oil was chromatographed (column diameter 13 mm) on silica gel with 4:1 $CHCl_3$–MeOH as the eluent. The solvents were removed under reduced pressure, and the resulting oil was distilled (Kugelrohr, 48–52 °C, 0.13 mmHg, lit.[13] bp 110–112 °C, 0.8 mmHg) to yield 430 mg (39%) of the desired product, which solidified upon standing, mp 117–120 °C. Ethereal HCl was added to a solution of the compound in absolute EtOH until an acidic pH was achieved, and the solvents were removed in vacuo. Recrystallization of the resulting solid from absolute EtOH/Et_2O afforded **3**, mp 275–278 °C (lit.[13] mp 273–275 °C).

8-Methoxy-2-[*N*-(3-phenylpropyl)amino]tetralin Hydrochloride (9). Glacial acetic acid (1.62 g, 27 mmol) and 3-phenyl-1-aminopropane (3.65 g, 27 mmol) were added to a stirred solution of 8-methoxy-2-tetralone (1.58 g, 9. mmol) in absolute EtOH (25 mL) with 4A molecular sieves. The solution was allowed to stir at room temperature under an N_2 atmosphere for 14 h. The EtOH was heated to dissolve the precipitate, and the molecular sieves were removed by filtration. The filtrate was added to a Parr bottle containing PtO_2 (60 mg), washing with additional absolute EtOH (35 mL). The solution was hydrogenated until sufficient H_2 was taken up (3 h). The catalyst was removed by filtration, and the EtOH was removed under reduced pressure to yield a yellow-brown oil. The oil was added to a 10% HCl solution (25 mL), and the precipate was removed by filtration and washed well with anhydrous Et_2O. Recrystallization from absolute EtOH afforded 1.9 g (63%) of **9** as white crystals, mp 221–223 °C.

Compounds **2**, **4–8**, **10**, and **11** were prepared in the same manner as **9** by using the appropriate tetralone and amine (See Table I).

8-Hydroxy-2-[*N*-(3-phenylpropyl)amino]tetralin Hydrochloride (12). A suspension of **9** (0.35 g, 1.05 mmol) in freshly distilled 48% HBr (15 mL) was heated at 120 °C for 2 h under an N_2 atmosphere. The volatiles were removed under reduced pressure, and the resulting solid was suspended between a saturated aqueous solution of $NaHCO_3$ (30 mL) and Et_2O (30 mL) and was allowed to stir at room temperature for 20 h. The layers were separated, and the aqueous fraction was extracted with Et_2O (20 mL). The ether portions were combined and dried ($MgSO_4$). The inorganic matter was removed by filtration, and the solvent was removed under reduced pressure to yield a solid, crude mp 148 °C. Ethereal HCl was added to a solution of the solid in absolute EtOH to achieve a pH of 2, and the solvents were removed in vacuo to afford a white solid. Recrystallization from absolute EtOH yielded 140 mg (31%) of **12**, mp 254–256 °C dec.

8-Methoxy-2-[*N*-methyl-*N*-(3-phenylpropyl)amino]tetralin Hydrochloride (13). Formic acid (97%, 0.15 g, 3.1 mmol) and formaldehyde (37%, 0.22 g, 2.7 mmol) were added to the free base of **9** (0.30 g, 1.02 mmol) at 0 °C. The solution was heated at 80 °C (oil bath temperature) for 14 h and then allowed to cool to room temperature, and the solvents were removed under reduced pressure. The resulting yellow oil was suspended in 10% HCl (10 mL), and the mixture was extracted with Et_2O (3 × 6 mL). The aqueous fraction was basified with 15% NaOH and extracted with Et_2O (3 × 8 mL), the combined ethereal fractions were dried ($MgSO_4$), and the ether was removed in vacuo to yield an oil, which was chromatographed on silica gel (10 g, column diameter 13 mm) by elution with $CHCl_3$–MeOH, 9:1. The first 30 mL of eluent were discarded and the next 45 mL were combined, and the solvent was removed under reduced pressure to yield a clear oil. An ethereal HCl solution was added to the resulting oil in absolute EtOH until an acidic pH was achieved. The solvents were removed in vacuo, and the resulting oil was triturated with anhydrous Et_2O to yield white crystals. Recrystallization from absolute EtOH/Et_2O yielded 140 mg (40%) of **13** as white crystals, mp 148–150 °C.

8-(Benzyloxy)-2-[*N*-(3-phenylpropyl)amino]tetralin Hydrochloride (14). Sodium hydroxide (80 mg, 2.0 mmol) in H_2O (1 mL) was added to the free base of **12** (60 mg, 0.21 mmol) in MeOH (2 mL), and the solution was allowed to stir at room temperature for 30 min; the solvents were removed under reduced pressure while the mixture was heated on a water bath (ca. 40 °C). The resulting solid was dissolved in dry THF, and the insoluble material was removed by filtration; the THF filtrate was evaporated in vacuo to yield a yellow gum. Benzyl bromide

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(0.025 mL, 0.21 mmol) in DMF (2 mL) was added to a solution of the gum in DMF (3 mL), and the solution was allowed to stir at room temperature for 1 h. The solution was poured into H_2O (5 mL) and acidified with 10% HCl. The aqueous solution was extracted with Et_2O (3 × 7 mL), basified with 15% NaOH, and extracted with Et_2O (4 × 8 mL). The combined Et_2O fractions were dried ($MgSO_4$), and the Et_2O was removed under reduced pressure to yield a yellow oil. An ethereal solution of HCl was added to a solution of the oil in absolute EtOH to achieve a pH of 2. The white precipate was collected by filtration, and recrystallization from absolute EtOH/Et_2O yielded 44 mg (52%) of the desired product, mp 174–176 °C.

Binding Studies. The radioligand binding assay was conducted in essentially the same manner as reported earlier.[19,20] Following decapitation, the brains of male Sprague–Dawley rats (ca. 220 g) were removed, placed in 0.9% ice-cold saline, and dissected over ice until the tissue was prepared. Tissues were stored in ice-cold saline for not longer than 1 h and, following blot drying and weighing, were prepared and frozen at −30 °C until used. Freshly dissected or frozen tissue was homogenized in 30 volumes of ice-cold buffer containing 50 mM Tris·HCl (pH 7.4 at 37 °C; pH 8.0 at 4 °C), 0.5 mM Na_2EDTA, and 10 mM $MgSO_4$ and centrifuged at 30000*g* for 15 min. The supernatant was discarded, and the pellet was resuspended and preincubated for 15 min at 37 °C. The pellet was washed twice by centrifugation and resuspension. The final assay buffer contained 50 mM Tris·HCl (pH 7.7), 10 μM pargyline, 0.1% ascorbate, 10 mM $MgSO_4$, and 0.5 mM Na_2EDTA. The 5-HT_{1A} receptor was labeled with 0.1 nM [^{3}H]-8-OH-DPAT (154 Ci/mmol; New England Nuclear) and 4-mg wet weight of rat hippocampal tissue. The presymaptic 5-HT_{1A} site was labeled with 1.0 nM [^{3}H]-8-OH-DPAT in 8 mg/mL wet weight of rat striatal homogenate in assay buffer. In both cases, 8-OH-DPAT (1 μM) was used to determine nonspecific binding. Eleven concentrations of nonradioactive competing drugs were made fresh daily in assay buffer. Following incubation with membranes and radioligand at 37 °C (20 min for hippocampal homogenates and 30 min for striatal homogenates), samples were rapidly filtered over glass fiber filters (Schleicher and Schuell) and were washed with 10 mL of ice-cold 50 mM Tris·HCl buffer. Individual filters were inserted into vials and equilibrated with 5 mL of scintillation fluid (ScintiVerse, Fisher) for 6 h before being counted at 45% efficiency in a Beckman 3801 counter. Results were analyzed by using the program EBDA in order to determine IC_{50}, K_i, and Hill values.

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Registry No. **2**, 19485-87-1; **3**, 3880-77-1; **3**·HCl, 3880-76-0; **4**, 3902-22-5; **5**, 3902-23-6; **6**, 117145-81-0; **6**·HCl, 117145-90-1; **7**, 117145-82-1; **7**·HCl, 117145-91-2; **8**, 117145-83-2; **8**·HCl, 117145-92-3; **9**, 117145-84-3; **9**·HCl, 117145-93-4; **10**, 117145-85-4; **10**·HCl, 117160-93-7; **11**, 117145-86-5; **11**·HCl, 117145-94-5; **12**, 117145-87-6; **12**·HCl, 117145-95-6; **13**, 117145-88-7; **13**·HCl, 117145-96-7; **14**, 117145-89-8; **14**·HCl, 117145-97-8; $PrNH_2$, 107-10-8; PrNHMe, 627-35-0; $PhCH_2NHMe$, 103-67-3; $Ph(CH_2)_2NH_2$, 64-04-0; $Ph(CH_2)_2NHMe$, 589-08-2; $Ph(CH_2)_3NH_2$, 2038-57-5; $Ph(CH_2)_4NH_2$, 13214-66-9; 8-methoxy-2-tetralone, 5309-19-3; 8-hydroxy-2-tetralone, 53568-05-1.

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Prostaglandin Photoaffinity Probes: Synthesis and Biological Activity of Azide-Substituted 16-Phenoxy- and 17-Phenyl-$PGF_{2\alpha}$ Prostaglandins†

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The development of a prostaglandin $PGF_{2\alpha}$ photoaffinity probe led to the synthesis and biological evaluation of azide-substituted 17-phenyl-18,19,20-trinorprostaglandin $F_{2\alpha}$ and 16-phenoxy-17,18,19,20-tetranorprostaglandin $F_{2\alpha}$ derivatives. Two approaches for the preparation of iodinated versions of these prostaglandins were evaluated: (1) iodination of a phenyl azide bearing an activating hydroxyl group and (2) iodination of an aniline precursor to the phenyl azide group and subsequent conversion of the aniline to the phenyl azide. In the first approach, 17-(4-azido-2-hydroxyphenyl)-18,19,20-trinorprostaglandin $F_{2\alpha}$, 16-(5-azido-3-hydroxyphenoxy)-17,18,19,20-tetranorprostaglandin $F_{2\alpha}$, and 16-(4-azido-2-hydroxyphenoxy)-17,18,19,20-tetranorprostaglandin $F_{2\alpha}$ were prepared by using the Corey synthesis, but were biologically inactive presumably as a result of the hydrophilic phenolic hydroxyl group. In the second approach, the iodination of a 17-(4-aminophenyl)-18,19,20-trinorprostaglandin $F_{2\alpha}$ derivative delivered 17-(4-azido-3-iodophenyl)-18,19,20-trinorprostaglandin $F_{2\alpha}$, which exhibited competitive binding with natural [^{3}H]$PGF_{2\alpha}$ to ovine luteal cells and to plasma membranes of bovine corpora lutea. [^{125}I]-17-(4-Azido-3-iodophenyl)-18,19,20-trinorprostaglandin $F_{2\alpha}$ was utilized in a preliminary photoaffinity cross-linking experiment.

In the course of developing radiolabeled photoaffinity probes[1] for various products of the arachidonic acid cascade,[2] we required a synthesis of aryl azide substituted prostaglandin $F_{2\alpha}$ derivatives[3] **1** (Figure 1). It was anticipated that these probes (PG-ArN_3) would bind the

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