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Synthesis and preliminary *in vivo* evaluation of new 2-Aryl-6-methyl-1,2-dihydro-1*H*-pyridin-4-ones and 2-Aryl-6-methylpiperidin-4-ols, as potential anti-amnesiant agents

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Abstract

A revisited synthesis of 2-aryl-6-methyl-1,2-dihydro-1*H*-pyridin-4-ones and their saturated analogues 2-aryl-6-methylpiperidin-4-ols is described. A five steps procedure, using the sulfinimine chemistry, to prepare a key intermediate β-(6-chloronicotinic)-β-amino ester is also reported. *In vivo* spontaneous working memory activity of these compounds has been investigated in the mouse. Results obtained with 2-(3-chlorophenyl)-6-methyl-1,2-dihydro-1*H*-pyridin-4-one **9b**, the most effective derivative in this model, have been reported.

Keywords: β-*Amino esters, sulfinamides, pyridinones, working memory, spontaneous alternation, nAChRs ligands*

Introduction

The nicotinic acetylcholine receptors (nAChRs) belong to a superfamily of ligand-gated ion channels which includes muscle-type and neuronal-type nAChRs, 5-HT_3, $GABA_A$, $GABA_C$ and glycine receptors. Actually twelve neuronal nAChR subunits ($\alpha2-\alpha10$ and $\beta2-\beta4$) have been found. Several combinations of α and β subunits can be expressed in oocytes or other heterologous expression systems resulting in functional ion channels with distinct pharmacological properties. For a few decades neuronal nAChRs have hold considerable promise as therapeutic targets for the treatment of central nervous system disorders. Drugs aimed at nAChRs may have a potential for the treatment of neurodegenerative disorders such as Alzheimer's and Parkinson's diseases, Tourette's syndrome, schizophrenia, attention deficit, hyperactivity disorder, certain epilepsies, and nicotine addiction, as well [1–3].

Our laboratory has recently pointed out the interest of new 2-aryl-6-methyl-1,2-dihydro-1*H*-pyridin-4-one and 2-aryl-6-methylpiperidin-4-ol derivatives as memory enhancers in relation to their nicotinic acetylcholine receptor activity [4–6]. Herein, we present a preliminary communication devoted to pharmacomodulations on our series and the first *in vivo* experiments concerning our lead compound.

Materials and methods

Chemistry

Instrumentation. Commercial reagents were used as received without additional purification. Melting points were determined on a Kofler melting point apparatus and are uncorrected. Infrared (IR) spectra were obtained in KBr pellets with a Genesis Series FTIR spectrometer. ^1H NMR (400 MHz) and ^{13}C NMR (100 MHz) were recorded on a JEOL Lambda

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400 spectrometer. Chemical shifts are expressed in ppm downfield from the residual deuterated solvent or the internal standard tetramethylsilane. Thin layer chromatographies (TLC) were performed on 0.2 mm precoated plates of silica gel 60F-254 (Merck). Visualization was made with ultraviolet light (254 nm). Column chromatographies were carried out using silica gel 60 (0.063–0.2 mm) (Merck). Elemental analyses for new compounds were performed at the "Institut de Recherche en Chimie Organique Fine" (Rouen).

*Synthesis method **a**: 6-chloropyridin-3-ylmethanol **1c**.* To a solution of 6-chloronicotinic acid (3.15 g, 20.0 mmol) in dry THF (60 mL), cooled to −10°C, was added triethylamine (2.93 mL, 20.0 mmol). The reaction mixture was stirred for 30 min and ethyl chloroformate (2.10 mL, 22.0 mmol) was added dropwise. After stirring for 30 min the solution was filtered, H_2O (30 mL) was added and the mixture was cooled to 0°C. $NaBH_4$ (1.89 g, 50.0 mmol) was added and the reaction mixture was allowed to warm to RT under stirring for 2 h. The mixture was extracted with ethyl acetate. The organic phase was washed with a saturated aqueous $NaHCO_3$ solution, dried ($MgSO_4$) and concentrated. The residue was purified by chromatography on silica gel eluting with ethyl acetate/cyclohexane (1:1) to afford **1c** (2.27 g) as a white solid. Yield: 79%. mp 44–45°C. IR (KBr) ν 3303, 2890, 1590, 1570, 1459, 1298, 1104, 1065, 1022, 821, 638 cm^{-1}. ^1H NMR ($CDCl_3$) δ 8.31 (d, $^4J_{HH} = 2.0$ Hz, 1H, H-2), 7.69 (dd, $^3J_{HH} = 8.2$ Hz, $^4J_{HH} = 2.0$ Hz, 1H, H-4), 7.31 (d, $^3J_{HH} = 8.2$ Hz, 1H, H-5), 4.71 (s, 2H, CH_2OH), 3.29 (bs, 1H, OH). ^{13}C NMR ($CDCl_3$) δ 150.4 (C-6), 148.1 (C-2), 137.8 (C-4), 135.5 (C-3), 124.2 (C-5), 61.6 (CH_2OH).

*Method **b**: 6-chloropyridine-3-carbaldehyde **2c**.* To a mixture of pyridinium chlorochromate (4.85 g, 22.5 mmol) and Celite® (5.0 g) in dry CH_2Cl_2 (100 mL), cooled to 0°C, was added a solution of 6-chloropyridin-3-ylmethanol **1c** (2.15 g, 15.0 mmol) in CH_2Cl_2 (25 mL). After stirring for 2 h the mixture was filtered through Celite® and the filter cake was washed with diethyl ether. The organic layer was concentrated and the residue was purified by chromatography on silica gel eluting with CH_2Cl_2 to afford **2c** (1.74 g) as a white solid. Yield: 82%. mp 79-80°C. IR (KBr) ν 3095, 2875, 1702, 1588, 1560, 1458, 1353, 1110, 1020, 832, 734, 538, 489 cm^{-1}. ^1H NMR ($CDCl_3$) δ 10.07 (s, 1H, CHO), 8.84 (d, $^4J_{HH} = 1.9$ Hz, 1H, H-2), 8.12 (dd, $^3J_{HH} = 8.2$ Hz, $^4J_{HH} = 1.9$ Hz, 1H, H-4), 7.49 (d, $^3J_{HH} = 8.2$ Hz, 1H, H-5). ^{13}C NMR ($CDCl_3$) δ 189.2 (CHO), 156.9 (C-6), 152.4 (C-2), 138.0 (C-4), 130.4 (C-3), 125.2 (C-5).

*Method **e**: N-(6-Chloropyridin-3-ylmethylene)-p-toluenesulfinamide **5c**.* A mixture of 6-chloropyridine-3-carbaldehyde **2c** (991 mg, 7.0 mmol), *p*-toluene sulfinamide (1.14 g, 7.35 mmol) and titanium(IV) ethoxide (8.0 g, 35.0 mmol) in dry CH_2Cl_2 (100 mL) was refluxed for 12 h. The reaction mixture was quenched at 0°C by addition of H_2O. The turbid solution was filtered through Celite® and the filter cake was washed with CH_2Cl_2. The organic layer was separated, dried ($CaCl_2$) and concentrated. The residue was purified by chromatography on silica gel eluting with CH_2Cl_2 to afford **5c** (1.64 g) as a white solid. Yield: 84%. mp 142–143°C. IR (KBr) ν 3086, 3056, 2923, 1594, 1583, 1556, 1454, 1373, 1333, 1100, 805, 673, 552 cm^{-1}. ^1H NMR ($CDCl_3$) δ 8.77 (s, 1H, HC = N), 8.76 (d, $^4J_{HH} = 2.2$ Hz, 1H, H-2′), 8.13 (dd, $^3J_{HH} = 8.3$, $^4J_{HH} = 2.2$ Hz, 1H, H-4′), 7.6 (d, $^3J_{HH} = 8.2$ Hz, 2H, H-2 and H-6), 7.41 (d, $^3J_{HH} = 8.3$ Hz, 1H, H-5′), 7.32 (d, $^3J_{HH} = 8.2$ Hz, 2H, H-3 and H-5), 2.40 (s, 3H, CH_3). ^{13}C NMR ($CDCl_3$) δ 156.7 (HC = N), 154.9 (C-6′), 151.1 (C-2′), 142.0 (C-4), 140.8 (C-1), 138.0 (C-4′), 129.8 (C-3 and C-5), 128.4 (C-3′), 124.7 (C-5′), 124.5 (C-2 and C-6), 21.3 (CH_3). Anal. Calcd. For $C_{13}H_{11}N_2OClS$: C, 56.01; H, 3.98; N, 10.05. Found: C, 56.22; H, 4.24; N, 9.68%.

*Method **f**: ethyl 3-(6-chloropyridin-3-yl)-3-[(p-tolylsulfinyl)amino]propanoate **6c**.* To a solution of dry THF (20 mL), cooled to −78°C, was added NaHMDS 2M (4.40 mL, 8.8 mmol) followed by anhydrous ethyl acetate (806 μL, 8.25 mmol). The reaction mixture was stirred for 1 h and during this time a solution of *N*-(6-chloropyridin-3-ylmethylene)-*p*-toluenesulfinamide **5c** (1.53 g, 5.5 mmol) in THF (5 mL) was added dropwise at −78°C. After stirring for 6 h the reaction was quenched at −78°C with a saturated aqueous NH_4Cl solution. The organic phase was extracted with ethyl acetate, dried ($MgSO_4$) and concentrated. The residue was purified by chromatography on silica gel eluting with ethyl acetate/petroleum ether (1:1) to afford **6c** (1.57 g) as a yellow oil. Yield: 78%. Mixture of two diastereomers: ^1H NMR ($CDCl_3$) δ 8.42 and 8.12 (m, 1H, H-2″), 7.76 and 7.46 (m, 1H, H-4″), 7.57 and 7.45 (m, 2H, H-2′ and H-6′), 7.34 and 7.15 (m, 1H, H-5″), 7.32 and 7.18 (m, 2H, H-3′ and H-5′), 5.53 and 5.17 (m, 1H, NH), 4.85 and 4.75 (m, 1H, CH), 4.07 (q, $^3J_{HH} = 7.2$ Hz, 2H, CH_2O), 2.96 and 2.85 (m, 2H, $CH_2C=O$), 2.43 and 2.36 (s, 3H, CH_3), 1.18 (t, $^3J_{HH} = 7.2$ Hz, 3H, CH_3CH_2O).

*Method **g**: ethyl 3-amino-3-(6-chloropyridin-3-yl)propanoate **4c**.* To a stirred solution of ester **6c** (1.47 g, 4.0 mmol) in MeOH (15 mL), cooled to 0°C, was added TFA (1.23 mL, 16.0 mmol). The reaction

mixture was stirred at RT for 2 h and concentrated. The residue was dissolved in CH_2Cl_2 (30 mL) and the resulting solution was washed with a saturated aqueous $NaHCO_3$ solution. The organic layer was dried ($CaCl_2$) and concentrated. The residue was purified by chromatography on silica gel eluting with ethyl acetate/MeOH (9:1) to afford **4c** (796 mg) as a yellow oil. Yield: 87%. IR (KBr) ν 3365, 3292, 2983, 2941, 1725, 1429, 1372, 1185, 1022, 711 cm^{-1}. ^1H NMR ($CDCl_3$) δ 8.39 (d, $^4J_{HH} = 2.3$ Hz, 1H, H-2′), 7.73 (dd, $^3J_{HH} = 8.2$ Hz, $^4J_{HH} = 2.3$ Hz, 1H, H-4′), 7.31 (d, $^3J_{HH} = 8.2$ Hz, 1H, H-5′), 4.48 (dd, $^3J_{HH} = 5.5$ and 8.1 Hz, 1H, CH), 4.14 (q, $^3J_{HH} = 7.1$ Hz, 2H, CH_2O), 2.68 and 2.64 (ABX, $^2J_{AB} = 16.1$ Hz, $^3J_{AX} = 8.1$ Hz, $^3J_{BX} = 5.5$ Hz, 2H, $CH_2C=O$), 1.97 (bs, 2H, NH_2), 1.24 (t, $^3J_{HH} = 7.1$ Hz, 3H, CH_3CH_2O). ^{13}C NMR ($CDCl_3$) δ 171.1 (C = O), 150.5 (C-6′), 148.2 (C-2′), 138.5 (C-3′), 137.0 (C-4′), 124.2 (C-5′), 60.8 (CH_2O), 49.7 (CH), 43.5 ($CH_2C=O$), 14.1 (CH_3CH_2O). Anal. Calcd. For $C_{10}H_{13}N_2O_2Cl$: C, 52.52; H, 5.73; N, 12.25. Found: C, 52.79; H, 5.97; N, 11.91%.

*Method **h**: tert-Butyl 3-[1-(6-chloropyridin-3-yl)-3-ethoxy-3-oxopropylamino]but-2-enoate **7c***. To a stirred solution of amino ester **4c** (572 mg, 2.5 mmol), in dry benzene (15 mL), was added AcOH (430 μL, 7.5 mmol), and *tert*-butylacetoacetate (497 μL, 3.0 mmol). The mixture was heated under reflux for 4 h and the water formed was removed azeotropically using a Dean-Stark apparatus. After cooling to RT, $CHCl_3$ (15 mL) was added and the solution was washed with a saturated aqueous $NaHCO_3$ solution. The organic layer was dried ($CaCl_2$), the solvents were evaporated and the residue was purified by chromatography on silica gel eluting with diethyl ether/petroleum ether (6:4) to afford **7c** (729 mg) as a colorless oil. Yield: 79%. IR (KBr) ν 3269, 3221, 2972, 2937, 1732, 1651, 1610, 1448, 1422, 1365, 1270, 1152, 1026, 788, 715 cm^{-1}. ^1H NMR ($CDCl_3$) δ 9.03 (d, $^3J_{HH} = 9.3$ Hz, 1H, NH), 8.34 (d, $^4J_{HH} = 2.7$ Hz, 1H, H-2′), 7.62 (dd, $^3J_{HH} = 8.3$ Hz, $^4J_{HH} = 2.7$ Hz, 1H, H-4′), 7.32 (d, $^3J_{HH} = 8.3$ Hz, 1H, H-5′), 5.03 (m, 1H, CH), 4.48 (s, 1H, C = CH), 4.14 and 4.10 (ABX_3, $^2J_{AB} = 10.7$ Hz, $^3J_{AX} = 7.2$ Hz, $^3J_{BX} = 7.2$ Hz, 2H, CH_2O), 2.85 and 2.78 (A′B′X′, $^2J_{A'B'} = 15.6$ Hz, $^3J_{A'X'} = 7.8$ Hz, $^3J_{B'X'} = 6.4$ Hz, 2H, $CH_2C=O$), 1.80 (s, 3H, CH_3), 1.47 (s, 9H, CH_3tBu), 1.22 (t, $^3J_{HH} = 7.2$ Hz, 3H, CH_3CH_2O). ^{13}C NMR ($CDCl_3$) δ 170.4 (C = O), 169.8 (C = O), 159.0 (C = CH), 150.7 (C-6′), 147.9 (C-2′), 136.7 (C-3′), 136.6 (C-4′), 124.5 (C-5′), 87.6 (C = CH), 78.4 (C*t*Bu), 61.2 (CH_2O), 51.1 (CH), 42.9 ($CH_2C=O$), 28.5 (CH_3tBu), 19.4 (CH_3), 14.0 (CH_3CH_2O).

*Method **i**: tert-Butyle 6-(6-chloropyridin-3-yl)-2-methyl-4-oxo-1,4,5,6-tetrahydropyridine-3-carboxylate **8c***. To a stirred solution of enamine **7c** (738 mg, 2.0 mmol), in dry *t*-BuOH (10 mL), was added *t*-BuOK (269 mg, 2.4 mmol), and the mixture was heated at 50°C. After completion (4 h monitored by TLC analysis), the reaction mixture was quenched with an aqueous HCl 0.5 M solution and extracted with $CHCl_3$. The organic layer was dried ($CaCl_2$) and concentrated. The crude product was purified by chromatography on silica gel eluting with ethyl acetate/MeOH (8:2) to give **8c** (542 mg) as a yellow solid. Yield: 84%. mp 180–181°C. IR (KBr) ν 3279, 3081, 2975, 2931, 1696, 1619, 1558, 1529, 1430, 1366, 1163, 1096, 715 cm^{-1}. ^1H NMR ($CDCl_3$) δ 8.35 (d, $^4J_{HH} = 2.3$ Hz, 1H, H-2′), 7.69 (dd, $^4J_{HH} = 2.3$ Hz, $^3J_{HH} = 8.3$ Hz, 1H, H-4′), 7.33 (d, $^3J_{HH} = 8.3$ Hz, 1H, H-5′), 6.52 (bs, 1H, NH), 4.77 (dd, $^3J_{HH} = 5.9$ and 11.4 Hz, 1H, CH), 2.61 and 2.49 (ABX, $^2J_{AB} = 15.8$ Hz, $^3J_{AX} = 11.4$ Hz, $^3J_{BX} = 5.9$ Hz, 2H, $CH_2C=O$), 2.31 (s, 3H, CH_3), 1.50 (s, 9H, CH_3tBu). ^{13}C NMR ($CDCl_3$) δ 186.7 (C = O), 166.4 (CO_2tBu), 165.5 (HN-C = C), 151.7 (C-6′), 148.3 (C-2′), 137.3 (C-3′), 134.0 (C-4′), 124.7 (C-5′), 100.4 (HN-C = C), 80.8 (C*t*Bu), 51.5 (CH), 42.8 ($CH_2C=O$), 28.4 (CH_3tBu), 22.6 (CH_3). Anal. Calcd. For $C_{16}H_{19}N_2O_3Cl$: C, 59.54; H, 5.93; N, 8.68. Found: C, 59.19; H, 5.98; N, 8.91%.

*Method **j**: 2-(6-Chloropyridin-3-yl)-6-methyl-2,3-dihydro-1H-pyridin-4-one **9c***. To a stirred solution of tetrahydropyridinone **8c** (484 mg, 1.5 mmol) in CH_2Cl_2 (5 mL) was added TFA (1.16 mL, 15.0 mmol) and the reaction mixture was heated at 50°C. After completion (4 h monitored by TLC analysis), the reaction mixture was neutralized with a saturated aqueous K_2CO_3 solution and extracted with CH_2Cl_2. The organic layer was dried ($CaCl_2$) and concentrated. The residue was purified by chromatography on silica gel eluting with ethyl acetate/MeOH (7:3) to afford **9c** (224 mg) as a yellow solid. Yield: 67%. mp 155-156°C. IR (KBr) ν 3235, 3059, 2963, 2936, 1616, 1581, 1536, 1435, 1358, 1269, 1105, 810 cm^{-1}. ^1H NMR ($CDCl_3$) δ 8.36 (d, $^4J_{HH} = 1.9$ Hz, 1H, H-2′), 7.68 (d, $^4J_{HH} = 1.9$ Hz, $^3J_{HH} = 8.1$ Hz, 1H, H-4′), 7.35 (d, $^3J_{HH} = 8.1$ Hz, 1H, H-5′), 6.23 (bs, 1H, NH), 5.02 (s, 1H, C = CH), 4.72 (dd, $^3J_{HH} = 5.9$ and 12.6 Hz, 1H, CH), 2.58 and 2.47 (ABX, $^2J_{AB} = 15.8$ Hz, $^3J_{AX} = 12.6$ Hz, $^3J_{BX} = 5.9$ Hz, 2H, $CH_2C=O$), 2.03 (s, 3H, CH_3). ^{13}C NMR ($CDCl_3$) δ 190.0 (C = O), 161.1 (C = CH), 149.2 (C-6′), 151.1 (C-2′), 136.2 (C-3′), 134.0 (C-4′), 124.7 (C-5′), 99.0 (C = CH), 54.3 (CH), 42.8 ($CH_2C=O$), 21.6 (CH_3). Anal. Calcd. For $C_{11}H_{11}N_2OCl$: C, 59.33; H, 4.98; N, 12.58. Found: C, 59.49; H, 5.17; N, 12.64%.

*Method **k**: 2-(3-Chlorophenyl)-6-methylpiperidin-4-ol **10b***. To a stirred solution of pyridinone **9b** (332 mg, 1.5 mmol) in EtOH (10 mL), cooled to 0°C, was added $NaBH_4$ (454 mg, 12.0 mmol) and the reaction mixture was stirred for 12 h at RT. The reaction

mixture was made alkaline with a saturated aqueous K_2CO_3 solution and extracted with CH_2Cl_2. The organic layer was dried ($CaCl_2$) and concentrated. The residue was purified by chromatography on silica gel eluting with ethyl acetate/MeOH (8:2) to afford **10b** (291 mg) as a white solid. Yield: 86%. mp 94-95°C. IR (KBr) ν 3278, 3248, 3031, 2965, 2928, 2835, 1605, 1452, 1374, 1307, 1088, 1031, 847, 755, 699 cm^{-1}. ^1H NMR ($CDCl_3$) δ 7.39 (s, 1H, H-2′), 7.26-7.24 (m, 3H, H-4′, H-5′ and H-6′), 3.79 (m, 1H, H-4), 3.67 (dd, $^3J_{HH} = 2.1$ and 11.5 Hz, 1H, H-2), 2.84 (ddq, $^3J_{HH} = 2.2$, 6.2 and 11.0 Hz, 1H, H-6), 2.07 (m, 1H, H-3eq.), 1.99 (m, 1H, H-5eq.), 1.85 (bs, 2H, NH and OH), 1.37 (m, 1H, H-3ax.), 1.17 (d, $^3J_{HH} = 6.2$ Hz, 3H, CH_3), 1.16 (m, 1H, H-5ax.). ^{13}C NMR ($CDCl_3$) δ 145.9 (C-1′), 134.3 (C-3′), 129.7 (C-5′), 127.4 (C-4′), 126.9 (C-2′), 124.9 (C-6′), 69.5 (C-4), 59.2 (C-2), 50.9 (C-6), 43.3 (C-3), 43.2 (C-5), 22.4 (CH_3). Anal. Calcd. For $C_{12}H_{16}NOCl$: C, 63.86; H, 7.14; N, 6.21. Found: C, 63.99; H, 7.21; N, 6.06%.

Pharmacology

Animals. Experiments were performed on male NMRI mice (Centre d'Elevage René Janvier, Le Genest, France) weighing 25–30 g. Mice were housed in standard polycarbonate cages containing ten animals and maintained in a regulated environment (22 ± 1°C) under 12h–12 h light/dark cycle (light on between 20:00 and 8:00) with food and water freely available in the home cage. Behavioral tests were conducted during the dark phase of the cycle from 14:00 to 20:00.

Apparatus. Immediate spatial working memory performance was assessed by recording spontaneous alternation behavior in a single session in a Y-maze made of black painted wood. The maze consisted of three equally spaced arms (22 cm long and 6.5 cm wide with walls 10 cm high). An inclined mirror was suspended over the maze to allow recording of the behavior at distance (1.5 m). Temperature in the test room was kept at 22 ± 1°C.

Testing procedure [7,8]. Each mouse, naive to the maze, was placed at the end of one arm and allowed to freely explore the maze during a 5-min session. The number and the sequence of arms entries and the number of rears were recorded by the observer. An arm entry was scored when all four feet crossed into the arm. Alternation behavior, defined as entries into all three arms on consecutive occasions, was expressed in percent of total arm entries. Percentage alternation was calculated as the ratio of actual to possible alternation (defined as the total number of arm entries minus two), multiplied by 100 as shown in the following equation: % Alternation = {(Number of alternations)/(Total arm entries-2)} × 100. The sequence of arm entered (for example ACBCBACAB…) provides a measure of spontaneous alternation behavior and thus immediate working memory (spontaneous alternation required recalling of precedent visited arm). Each experience was completed for ten mice per group.

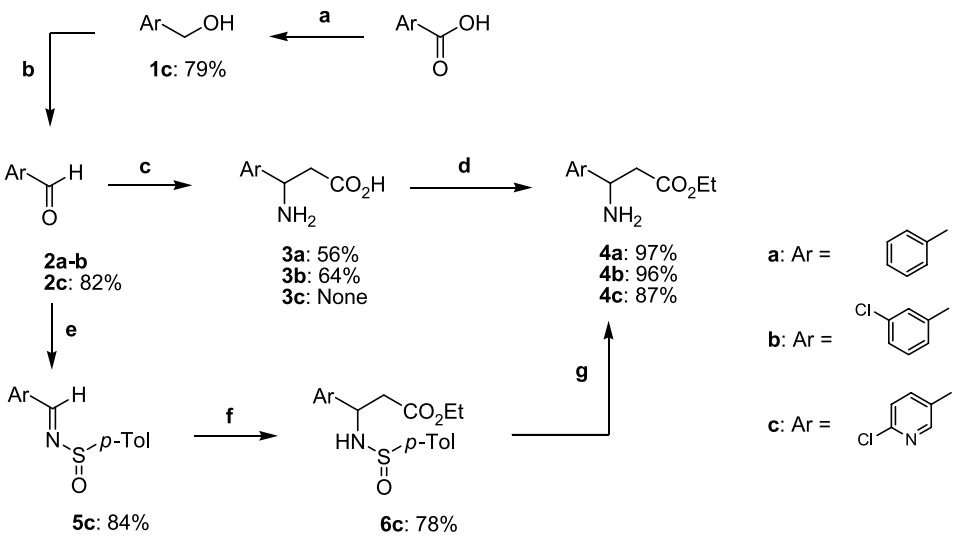
Drug administration. Compound **9b** was tested at 0.1, 0.3 and 1 mg/kg. For each dose, four groups were constituted: control (saline + saline), scopolamine (saline + scopolamine), tested compound (compound +saline) and association (compound+ scopolamine). Scopolamine (scopolamine hydrobromide, Sigma) was administered at 1 mg/kg. Arecoline (arecoline hydrobromide, Sigma) used as pharmacological anti-amnesic drug was tested at 1 mg/kg. All drugs, dissolved in saline as the vehicle, were administered intraperitoneally (i.p.) in a volume of 10 mL kg^{-1}, 30 min before testing.

Statistical analyses. Percentage alternation scores were statistically analyzed by a one-way analysis of variance (ANOVA) with "alternation" as interdependent factors and "pharmacological treatment" as an independent factor. In the case of significant differences in the variances, post-hoc multiple comparisons (PLSD of Fisher) were undertaken to locate a principal effect. p-Values less than 0.05 were considered to be significant. Results are expressed as means ± SEM.

Results and discussion

Chemistry

Key intermediates in the synthesis of the target compounds described here are the corresponding β-aryl-β-amino esters **4a–c**. To obtain these derivatives we applied the Rodionow-Johnson reaction [9–11]. This synthesis involves a single step in which corresponding arylcarboxaldehydes were refluxed with malonic acid and ammonium acetate. Contrary to previously described [12], in the case of the 6-chloro-3-pyridine carboxaldehyde reagent, prepared from 6-chloronicotinic acid *via* a reduction-oxidation sequence, this method failed. Alternatively, transformation of the aldehyde **2c** to *N*-sulfinyl aldimine **5c** was achieved by refluxing with racemic *para*-toluenesulfinamide in the presence of titanium(IV) ethoxide [13]. According to the Davis's methodology [14,15], treatment of derivative **5c** with the sodium enolate of ethyl acetate, generated from NaHMDS in THF at −78°C, afforded **6c**. The sulfinamide **6c** was then



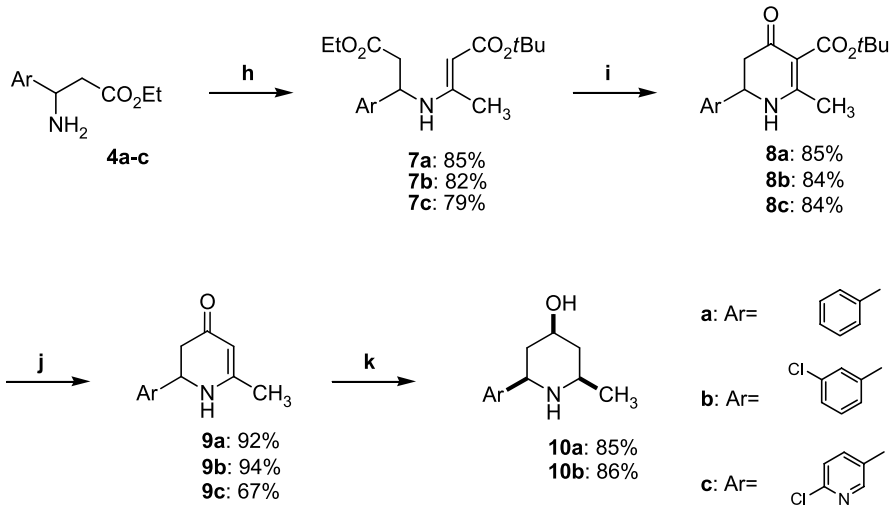
Scheme 1. Synthesis of β-aryl-β-amino esters **4**. (**a**) i) Et_3N, EtOCOCl, THF, 0°C, ii) $NaBH_4$, THF/H_2O, 0°C; (**b**) PCC, CH_2Cl_2, RT; (**c**) $CH_2(CO_2H)_2$, $AcONH_4$, EtOH, reflux; (**d**). i) $SOCl_2$, EtOH, reflux,. ii) NH_4OH, CH_2Cl_2, RT; (**e**) *p*-Toluenesulfinamide, $Ti(OEt)_4$, CH_2Cl_2, reflux; (**f**) NaHMDS, AcOEt, THF, −78°C; (**g**) i) TFA, MeOH, 0°C, ii) $NaHCO_3$, CH_2Cl_2, RT.

hydrolyzed by treatment with trifluoroacetic acid, affording after alkalinization, the aimed β-amino ester **4c** (Scheme 1).

Treatment of β-aryl-β-amino esters **4a–c** with *tert*-butyl acetoacetate, in the presence of three equivalent amounts of acetic acid in refluxing benzene with azeotropic removal of water, generated the *tert*-butyl enaminoesters **7a–c**. The latter were then converted to the cyclic derivative **8a–c** using potassium *tert*-butoxide in *tert*-butanol. Removal of the *tert*-butyl group was finally performed by refluxing in dichloromethane in the presence of trifluoroacetic acid to give the target 2-aryl-6-methyl-2,3-dihydro-1*H*-pyridin-4-ones **9a–c** [16]. Access to the saturated 2-aryl-6-methylpiperidin-4-ols **10a** and **10b** was carried out using an excess of $NaBH_4$. Under these conditions only the diastereomers with substituents in the *cis* orientation were detected (^{1}H NMR and HPLC) and isolated (Scheme 2).

Pharmacology

Administered alone, or in association with scopolamine, the ability of compound **9b** to affect exploratory behavior in the Y-maze was investigated at doses of 0.1, 0.3 and 1 mg/kg. Statistical analysis revealed that, at the doses tested, compound **9b** administered alone had no effect on the alternation behavior when compared to saline controls (see for 0.3 mg/kg in Figure 1). Scopolamine significantly reduced the percentage of alternation when compared to saline



Scheme 2. Synthesis of 2-aryl-6-methyl-2,3-dihydro-1*H*-pyridin-4-ones **9**. (**h**) *t*-Butyl acetoacetate, AcOH, C_6H_6, reflux; (**i**) *t*-BuOK, *t*-BuOH, 50°C; (**j**) TFA, CH_2Cl_2, reflux, (**k**) $NaBH_4$, EtOH, RT.

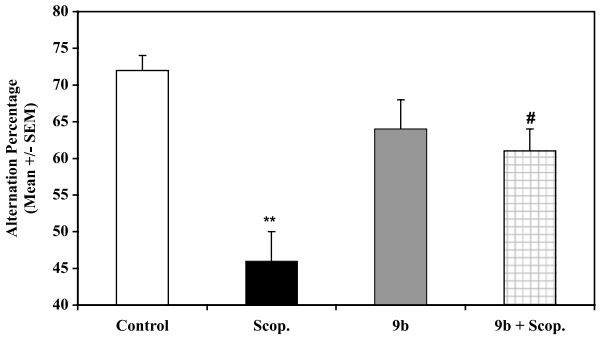


Figure 1. Effects of compound **9b** on spontaneous working memory in mouse. Mice (n = 10 in each group) received two injections 30 min before the test. Control = saline + saline; Scop. = saline + scopolamine (1 mg/kg); 9b = compound **9b** (0.3 mg/kg) + saline; 9b + Scop. = compound **9b** (0.3 mg/kg) + scopolamine (1 mg/kg). **p < 0.01 *versus* control (saline), #p < 0,05 *versus* scopolamine treated mice (ANOVA + PLSD of Fisher).

control ($p < 0.01$; PLSD of Fisher). However, in association with scopolamine experiments, **9b** being ineffective at 0.1 mg/kg dosage, reversed, at the doses of 0.3 mg/kg (Fig. 1) and 1 mg/kg (data not showns), the scopolamine-induced impairment of spontaneous alternation. This was illustrated for the 0.3 mg/kg dosage, both i) by a significant difference between the co-treatment and the scopolamine groups ($p < 0.05$; PLSD of Fischer), and ii) by the lack of significant difference between controls and co-treated animals. Under the same conditions, arecoline (1 mg/kg), used as a pharmacological reference, significantly reversed the scopolamine-induced deficit (49% for scopolamine group versus 67% for arecoline + scopolamine group, $p < 0.0001$ (PLSD of Fisher). The present findings demonstrate that compound **9b** at 0.3 mg/kg is able to reverse alternation deficits induced by scopolamine in the Y-maze test.

Conclusion

The *in vivo* pharmacological findings indicated that these new nicotinic ligands (especially compound **9b**) may provide a potential therapeutic approach to restore memory deficits in neurodegenerative diseases in which short-term working memory disorders were associated with cholinergic hypofunction. These promising results prompt us to continue the exploration of these new pyridinone and piperidinol series.

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