4-Alkynylphenylsilatranes: Insecticidal Activity, Mammalian Toxicity, and Mode of Action

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4-Ethynyl- and 4-(prop-1-ynyl)phenylsilatranes [$N(CH_2CH_2O)_3SiR$, R = C_6H_4-4-C≡CH or C_6H_4-4-C≡CCH_3] are highly toxic to houseflies (pretreated with piperonyl butoxide) and milkweed bugs (topical LD_{50}s 3–14 μg/g) and to mice (intraperitoneal LD_{50}s 0.4–0.9 mg/kg), and they are moderately potent inhibitors of the [^{35}S]-*tert*-butylbicyclophosphorothionate or TBPS binding site (GABA-gated chloride channel) of mouse brain membranes. Scatchard analysis indicates noncompetitive interaction of 4-ethynylphenylsilatrane with the TBPS binding site. Phenylsilatrane analogues with 4-substituents of H, CH_3, Cl, Br, and C≡$CSi(CH_3)_3$ are highly toxic to mice but have little or no activity in the insect and receptor assays. Radioligand binding studies with [4-^{3}H]phenylsilatrane failed to reveal a specific binding site in mouse brain. Silatranes with R = H, CH_3, CH_2Cl, CH=CH_2, OCH_2CH_3, and C_6H_4-4-CH_2CH_3 are of little or no activity in the insect and mouse toxicity and TBPS binding site assays as are the trithia and monocyclic analogues of phenylsilatrane. 4-Alkynylphenylsilatranes are new probes to examine the GABA receptor–ionophore complex of insects and mammals.

INTRODUCTION

The high convulsant potency and toxicity in mammals of 1-arylsilatranes [$N(CH_2CH_2O)_3Si$-aryl] was first reported in 1964 by Voronkov and his colleagues (Baltkajs et al., 1964), who subsequently prepared large numbers of silatranes and examined their activity in many biological systems (Voronkov and Lukevics, 1969; Voronkov, 1979; Voronkov et al., 1982). They noted that many arylsilatranes are highly toxic to mammals but failed to observe any insecticidal activity. The silatranes are one of a number of toxicants referred to on a trivial basis as "cage convulsants". Others are bicyclophosphorus esters [e.g., $(CH_3)_3CC(CH_2O)_3P$=S, also known as TBPS], their bicycloorthocarboxylate analogues [$RC(CH_2O)_3CR'$], picrotoxinin and related compounds, and polychlorocycloalkane insecticides, all of which inhibit TBPS binding in brain membrane preparations and are proposed to act in mammals as $GABA_A$ receptor antagonists, thereby blocking the GABA-gated chloride channel (Casida et al., 1976; Casida, 1987; Casida and Palmer, 1988; Obata et al., 1988). The potency of the cage convulsants other than the silatranes in blocking the TBPS binding site of brain membranes is generally more closely correlated with their toxicity to mice than with their insecticidal activity (Casida et al., 1988). In this correlation, 4-chlorophenylsilatrane is a poor inhibitor of TBPS binding (Squires et al., 1983; Cole et al., 1984) and GABA-induced chloride flux (Obata et al., 1988) relative to its mammalian toxicity.

The present study examined the structure–activity relationships, selective toxicity, and mode of action of the silatranes with particular respect to their potential insecticidal activity.

MATERIALS AND METHODS

Spectroscopy. Proton nuclear magnetic resonance (NMR) spectra were obtained at 300 MHz with a Bruker WM-300 spectrometer for samples dissolved in deuterochloroform unless otherwise stated. Mass spectra (MS) utilized the Hewlett-Packard 5985 system with chemical ionization (230 eV with methane at 0.8 Torr) or electron impact (70 eV).

Synthesis of Silatranes and Related Compounds (Tables I and II). Arylsilatranes 1 and 7 were obtained from Petrarch Systems Inc. (also known as Hulls America Inc.) (Bristol, PA) and M & T Chemicals Inc. (Rahway, NJ), respectively. Compounds 2–6 and 8 were synthesized for this study, and each compound was pure by TLC and NMR and gave appropriate quasimolecular ions (CI-MS) or molecular ions (EI-MS) and a base peak of m/z 174 corresponding to $[N(CH_2CH_2O)_3Si]^+$ (Voronkov et al., 1982).

[4-^{3}H]Phenylsilatrane. [4-^{3}H]Phenylsilatrane (5.5 Ci/mmol) was prepared by reductive dechlorination of 4-chlorophenylsilatrane with tritium gas, the final radiolabeling being carried out by New England Nuclear Corp. In developing the method, a solution of 4-chlorophenylsilatrane in dry, oxygen-free ethyl acetate containing 1 equiv of triethylamine and a catalytic amount of 10% palladium on carbon catalyst was stirred vigorously under a hydrogen atmosphere overnight. The resulting mixture was filtered through Celite and evaporated. Purification on silica eluting with chloroform–methanol (19:1) afforded an almost quantitative yield of phenylsilatrane. An analogous procedure was used to prepare [4-^{3}H]phenylsilatrane with final purification by HPLC on Zorbax silica eluting with methylene chloride–methanol (199:1).

4-Substituted Phenyltriethoxysilane Intermediates. 4-Methylphenyltriethoxysilane was prepared by the procedure of Selin and West (1962) [bp 85–88 °C (0.2 mmHg); reported 104–106 °C (1.8 mmHg)], and an analogous procedure was used for 4-bromophenyltriethoxysilane [bp 118–122 °C (1.6 mmHg); reported 118 °C (1.5 mmHg) by Pepe (1961)]. 1-Bromo-4-ethynylbenzene (from 4-bromoacetophenone; Dufraisse and Dequesnes, 1931) was converted to the trimethylsilyl derivative [bp 93 °C (1 mmHg); reported 96–97 °C (2 mmHg) by Eaborn et al. (1967)]. The Grignard reagent, 4-[(trimethylsilyl)ethynyl]-phenylmagnesium bromide, prepared by refluxing 1-bromo-4-[(trimethylsilyl)ethynyl]benzene (40 mmol) with magnesium (41 mmol) in dry tetrahydrofuran (THF), was added to tetraethoxysilane (15 mL) in dry THF (10 mL). After the mixture was stirred for an additional 2 h at room temperature, the THF was removed by distillation under N_2. On cooling, an oil separated and solidified. Following filtration, the residue was washed with

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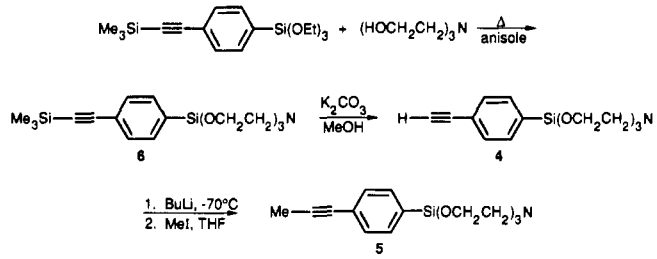

Figure 1. Synthesis of 4-ethynylphenylsilatrane and related compounds.

dry ether and the solvent was removed in vacuo. Distillation yielded 4-[(trimethylsilyl)ethynyl]phenyltriethoxysilane (30%): bp 125–130 °C (0.6 mmHg); [M + 1]$^+$ 337; NMR δ 0.23 [9 H, s, $(CH_3)_3Si$], 1.22 [9 H, t, J = 7.0 Hz, $(CH_3CH_2O)_3$], 3.83 (6 H, q, J = 7.0 Hz, $(OCH_2CH_3)_3$], 7.44 (2 H, d, J = 8.5 Hz, Ar H), 7.59 (2 H, d, J = 8.5 Hz, Ar H).

4-Substituted Phenylsilatranes. Silatranes **2** and **8** were prepared by the procedures of Daneshrad et al. (1975) and Wu et al. (1982), respectively. A stirred mixture of the appropriate aryltriethoxysilane (10 mmol) and triethanolamine (10 mmol), in the presence of a catalytic amount of KOH in the case of **8**, was heated at 180 °C under N_2 in an oil bath until the theoretical amount of ethanol had distilled over, normally within 1 h. The solid residue was collected, washed with ether, and purified by recrystallization. Compound **2** (87%): mp 195.5–196 °C (chloroform/hexane), reported 195–195.5 °C; [M]$^+$ 265; NMR δ 2.27 (3 H, s, CH_3), 2.89 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 3.88 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 7.07 (2 H, d, J = 7.8 Hz, Ar H), 7.60 (2 H, d, J = 7.8 Hz, Ar H). Compound **8** (91%): mp 247 °C (hexane/acetone), reported 247–248 °C by Hencsei et al. (1983); [M + 1]$^+$ 330 (Br 79), 332 (Br 81); NMR δ 2.92 [6 H, t, J = 5.9 Hz, $N(CH_2CH_2O)_3Si$], 3.89 [6 H, t, J = 5.9 Hz, $N(CH_2CH_2O)_3Si$], 7.38 (2 H, d, J = 8.3 Hz, Ar H), 7.59 (2 H, d, J = 8.3 Hz, Ar H).

Silatrane **6** was prepared by coupling the corresponding triethoxysilane and triethanolamine (Figure 1) under conditions used earlier (Voronkov et al., 1979) for preparing unsaturated silatranes. A solution of triethanolamine (8.3 mmol) and 4-[(trimethylsilyl)ethynyl]phenyltriethoxysilane (8.6 mmol) in anisole (dried over $CaCl_2$) was refluxed for 20 h under N_2. Removal of the ethanol and anisole at atmospheric pressure yielded the product as a solid together with unreacted starting material as an oil. Addition of ether, filtration, and washing the residue with ether gave **6** as a white solid (60%): mp 248 °C; [M + 1]$^+$ 348; NMR δ 0.23 [9 H, s, $(CH_3)_3Si$], 2.92 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 3.89 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 7.35 (2 H, d, J = 8.1 Hz, Ar H), 7.66 (2 H, d, J = 8.1 Hz, Ar H). The trimethylsilyl group was removed by adding dry methanol (6 mL) and anhydrous K_2CO_3 (55 mg) to silatrane **6** (1.43 mmol) in dry chloroform (3 mL) and stirring the mixture at room temperature under N_2 overnight (Figure 1). Following solvent removal, the residue was taken up in chloroform which was washed successively with water and brine, dried ($MgSO_4$), and concentrated to give a white solid. Further washing with ether on a fritted disk gave **4** (76%): mp 233–236 °C (hexane/acetone); pure by GC–MS (EI); [M$^+$] 275; NMR δ 2.93 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 3.00 (1 H, s, $C{\equiv}CH$), 3.90 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 7.39 (2 H, d, J = 8.1 Hz, Ar H), 7.69 (2 H, d, J = 8.1 Hz, Ar H).

Silatrane **5** was prepared from **4** (Figure 1) (0.062 mmol) in dry THF (2 mL) at −70 °C under N_2 on addition of excess n-butyllithium (0.184 mmol) to give a permanent pink color (indicating complete formation of the anion) and, after 0.5 h at −70 °C, by addition of excess methyl iodide (2.41 mmol) (discharging the pink color). Following removal of excess methyl iodide after the mixture was warmed to room temperature, water was added, and the mixture was extracted with chloroform; workup as above including washing with ether and recrystallization gave **5** as a white solid (no starting material **4** was present by GC–MS) (82%): mp 263–264 °C (hexane/acetone); GC–MS(EI) [M$^+$] 289; NMR δ 2.02 (3 H, s, CH_3), 2.92 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 3.90 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 7.29 (2 H, d, J = 8.0 Hz, Ar H), 7.64 (2 H, d, J = 8.0 Hz, Ar H).

Hydrogenation of silatrane **4** (0.15 mmol) in dry ethyl acetate (25 mL) with 5% Pt/C (~10 mg) at atmospheric pressure for 1 h and filtration through Celite eluting with ethyl acetate gave **3** after solvent removal (40%): mp 148–149 °C; [M + 1]$^+$ 280; NMR δ 1.17 (3 H, t, J = 7.5 Hz, CH_3), 2.57 (2 H, q, J = 7.5 Hz, CH_2), 2.91 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 3.89 [6 H, t, J = 5.85 Hz, $N(CH_2CH_2O)_3Si$], 7.10 (2 H, d, J = 7.9 Hz, Ar H), 7.63 (2 H, d, J = 7.9 Hz, Ar H).

1-Phenyl-2,8,9-trithia-5-aza-1-silatricyclo[3.3.3.0^{1,5}]undecane (Referred to as Phenyltrithiasilatrane). 2,2′,2″-Trimercaptotriethylamine [prepared according to the method of Harley-Mason (1947)] was converted to phenyltrithiasilatrane according to a literature procedure (Lukevics et al., 1977). A stirred mixture of trimercaptotriethylamine (4.0 mmol) and tris(dimethylamino)phenylsilane (Petrarch Systems Inc.) (5.9 mmol) was heated by means of an oil bath maintained at 110 °C for 2 h. Dimethylamine liberated was removed by a constant flow of N_2. The white solid which separated was collected and washed on a fritted disk with ether (~50%, poorly soluble in organic solvents and possibly containing a small amount of insoluble residue): mp 304–305 °C (reported 300 °C); [M + 1]$^+$ 300, [M − C_6H_5]$^+$ 222; NMR (CD_2Cl_2) δ 2.86–2.91 [12 H, m, $N(CH_2CH_2S)_3Si$], 7.20–7.23 (3 H, m, Ar H), 7.69–7.72 (2 H, m, Ar H).

1,3-Dioxa-6-aza-2-silacyclooctanes (Also Known as Pseudosilatranes or Silocanes; Hegyes et al., 1983). Two pseudosilatranes (Table II) were prepared according to the procedure of Urtane et al. (1985). A stirred mixture of methylphenyldiethoxysilane (Petrarch Systems) and N-methyl- or N-$tert$-butyldiethanolamine (Aldrich Chemical Co., Milwaukee, WI), with a catalytic amount of sodium methoxide in the case of the N-methyl analogue, was heated at 160 °C under N_2 in an oil bath until the theoretical amount of ethanol was distilled over, normally within 1 h. Distillation gave the desired compounds. N-Methyl derivative (76%): bp 104–110 °C (0.5 mmHg), redistilled 112 °C (0.5 mmHg), reported 123–124 °C (3 mmHg) by Urtane et al. (1985); [M + 1]$^+$ 238; NMR δ 0.27 (3 H, s, $SiCH_3$), 2.05 (3 H, s, NCH_3), 2.55–2.61 [4 H, m, $N(CH_2CH_2O)_2Si$], 3.84 [4 H, t, J = 5.0 Hz, $N(CH_2CH_2O)_2Si$], 7.30–7.32 (3 H, m, Ar H), 7.56–7.59 (2 H, m, Ar H). N-$tert$-Butyl derivative (30%): bp 110–123 °C (0.2 mmHg), reported 118 °C (2 mmHg) by Urtane et al. (1985); [M + 1]$^+$ 280; NMR δ 0.44 (3 H, s, $SiCH_3$), 1.04 [9 H, s, $C(CH_3)_3$], 2.52–2.60 (2 H, m), 2.82–2.89 (2 H, m) [$N(CH_2CH_2O)_2Si$], 3.85–3.91 [4 H, m, $N(CH_2CH_2O)_2Si$], 7.33–7.36 (3 H, m, Ar H), 7.64–7.66 (2 H, m, Ar H).

Insecticidal Activity. LD_{50} values were determined for adult female houseflies (*Musca domestica* L., SCR strain, ~20 mg each) and adult milkweed bugs (*Oncopeltus fasciatus* Dallas, ~40 mg each) held 24 h at 25 °C after application of the test compound to the ventrum of the abdomen. Synergized toxicity was evaluated for flies pretreated topically with piperonyl butoxide (PB) at 250 μg/g 2 h before the toxicant was administered. The carrier vehicle for each application was 0.5 μL of acetone for houseflies and 2.0 μL of acetone for milkweed bugs. The experiments were repeated as appropriate to establish the reproducibility of the results.

Mouse Toxicity Tests. Mortality was determined 24 h after intraperitoneal (IP) administration of the test compounds to male abino Swiss-Webster mice (18–22 g) with methoxytriglycol (50 μL) as the carrier vehicle. To establish LD_{50} values, five to nine mice were used at each dosing level.

Intracerebroventricular (ICV) toxicity was determined according to the procedure of Lawrence and Casida (1982).

Receptor Potency. The P_2 membrane fraction was prepared from the brains of mice by homogenizing in 0.32 M sucrose, centrifuging at 1500g for 10 min, decanting, and recentrifuging at 15000g for 20 min to obtain the P_2 pellet. This was then resuspended in 1 mM EDTA and dialyzed three times against water to remove endogenous GABA (Cole and Casida, 1986). The final pellet was resuspended in 200 mM NaCl–50 mM sodium phosphate, pH 7.4, assay buffer. Receptor assays involved 200 μg of protein in 1 mL of assay buffer containing [^{35}S]TBPS (2 nM) alone or with unlabeled TBPS (2 μM) to correct for nonspecific binding (12% relative to total binding). Following incubation for 30 min at 37 °C to achieve equilibrium between [^{35}S]TBPS and its binding site, the suspensions were

Table I. Structure–Activity Relationships for 4-Substituted Phenylsilatranes, $N(CH_2CH_2O)_3SiPh$-4-R

no.	R	LD_{50}, mg/kg: milkweed bug	LD_{50}, mg/kg: housefly (with PB)[a]	LD_{50}, mg/kg: mouse	TBPS receptor IC_{50}, μM
1	H	>500	>500	0.55 (0.33)[b]	>10[c]
2	CH_3	>500	>500	0.30 (0.15)	>10
3	CH_2CH_3	>500	>500	25–250	>10
4	C≡CH	3.0	3.4	0.36	1.0
5	$C{\equiv}CCH_3$	14	3.3	0.85	0.50
6	$C{\equiv}CSi(CH_3)_3$	>500	>500	2.5	10
7	Cl	>500	>500	0.22 (1.7)	>10
8	Br	>500	>500	0.88	10

[a] None of the compounds are active at 500 μg/g without PB. [b] Numbers in parentheses are data of Voronkov (1979) for mouse IP LD_{50} values. [c] No significant inhibition at 10 μM.

Table II. Silatranes and Related Compounds of Low Potency as Toxicants for Insects and Mammals and as Inhibitors of the [^{35}S]-*tert*-Butylbicyclophosphorothionate Binding Site of Mouse Brain Membranes[a]

silatranes, $N(CH_2CH_2O)_3SiR$[b]
- R = H (100)[c]
- R = CH_3 (3000)[c]
- R = CH_2Cl (2800)[c]
- R = $CH{=}CH_2$ (3000)[c]
- R = OCH_2CH_3 (3000)[c]

trithiasilatrane[d]
- $N(CH_2CH_2S)_3SiPh$ (>500)[e]

pseudosilatranes or silocanes[d]
- $CH_3N(CH_2CH_2O)_2Si(CH_3)Ph$ (>1000)[f]
- $(CH_3)_3CN(CH_2CH_2O)_2Si(CH_3)Ph$ (>1000)[f]

[a] LD_{50}s >500 μg/g insect topical (housefly with and without PB and milkweed bug). No significant inhibition at 10 μM for TBPS receptor. [b] Source: Petrarch Systems, Bristol, PA. [c] Numbers in parentheses are data of Voronkov (1979) for mouse IP LD_{50} values. [d] See Materials and Methods for synthesis. [e] Mouse IP LD_{50} as suspension in dimethyl sulfoxide. [f] Mouse IP LD_{50} for solutions in methoxytriglycol.

subjected to rapid filtration on Whatman GF/C filters, three rinses with 2 mL of cold assay buffer, and liquid scintillation counting.

Binding Studies with [4-^{3}H]Phenylsilatrane. The standard incubation mixture in 1 mL of Tris-HCl buffer (5 mM, pH 7.4) with 200 mM NaCl and 10 μM $CaCl_2$ contained 0.5 mg of P_2 membrane protein (prepared as above) and 4 nM [^{3}H]-phenylsilatrane. The mixture was incubated for 20 min at 25 °C followed by filtration. Nonspecific binding was determined as the difference between tritium bound from [^{3}H]phenylsilatrane alone and in the presence of 2 μM 4-chlorophenylsilatrane.

RESULTS

Insecticidal Activity (Tables I and II). Of the eight substituted phenylsilatranes examined, only the 4-ethynyl and 4-propynyl analogues (**4** and **5**) show significant insecticidal activity in topical assays with adult milkweed bugs and houseflies. Similar potency is observed for **4** and **5** to milkweed bugs without synergist and to houseflies with PB, and these compounds are not active on houseflies without synergist.

Eight other compounds including five silatranes, one trithiasilatrane, and two pseudosilatranes are not insecticidally active.

Toxicity to Mice. Six of the eight 4-substituted phenylsilatranes examined have mouse IP LD_{50}s in the range 0.22–0.88 mg/kg, whereas **6** and particularly **3** are less toxic (Table I). The remaining silatranes, lacking an aryl group, and the trithiasilatrane and pseudosilatranes are of little or no toxicity at the solubility limits of the test (Table II).

Four arylsilatranes have similar LD_{50}s, as micrograms per mouse, when administered ICV or IP, and this is also true for the trioxabicyclooctanes *tert*-butylbicycloorthobenzoate (TBOB) and TBPS (Table III).

Table III. Relative Toxicity of Arylsilatranes and Trioxabicyclooctanes Administered Intracerebroventricularly or Intraperitoneally to Mice

compound	LD_{50},[a] μg/mouse: ICV	LD_{50},[a] μg/mouse: IP
$N(CH_2CH_2O)_3SiPh$-4-R		
R = H	15	11
R = Cl	5	4
R = C≡CH	14	7
R = $C{\equiv}CCH_3$	7	17
trioxabicyclooctanes		
TBOB	23	26
TBPS	1.1	1.1

[a] Data reproducible in repeated tests within 2.0- and 1.2-fold for the ICV and IP treatments, respectively.

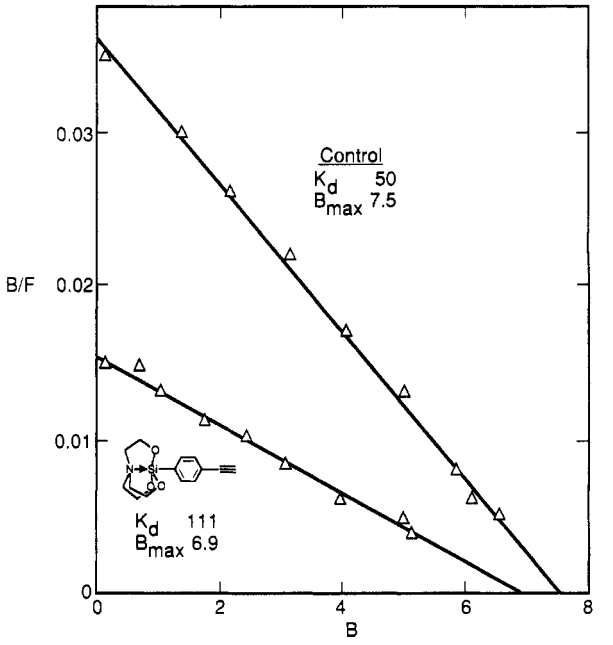

Figure 2. Scatchard plot of specific [^{35}S]TBPS binding to mouse brain membranes in the presence of 1.0 μM 4-ethynylphenylsilatrane (compound **4**). B and F are bound and free [^{35}S]-TBPS. K_d is given as nanomolar and B_{max} as picomoles per milligram of protein. Similar results were obtained in an independent experiment, i.e., control B_{max} = 7.4 and K_d = 50 and compound **4** B_{max} = 6.8 and K_d = 110. All Hill numbers were 0.99–1.01, and correlation coefficients were at least 0.998.

Potency at TBPS Receptor. Only two of the arylsilatranes are moderately potent inhibitors of TBPS binding (Table I). The potency order relative to the 4-substituent is propynyl (**5**) > ethynyl (**4**) > trimethylsilylethynyl (**6**) and bromo (**8**) > the other compounds. Test compounds in other series are inactive as inhibitors at the TBPS binding site (Table II).

Scatchard analysis indicates noncompetitive interaction of 4-ethynylphenylsilatrane (**4**) with the TBPS binding site (Figure 2).

Attempts To Recognize a [^{3}H]Phenylsilatrane Binding Site. [^{3}H]Phenylsilatrane yields little or no specific binding under the conditions used to assay the [^{35}S]TBPS receptor in mouse brain membranes. Mouse brain was therefore assayed by using the incubation mixture referred to as standard under Materials and Methods with the following variations: washed versus unwashed standard pellet; dialyzed versus undialyzed standard pellet; whole homogenate versus microsomes versus soluble fraction; protein levels of 0.1–1 mg; various

buffers of pH 5.5–9.5; temperatures of 0, 5, 25, and 37 °C; incubation times of 5 min–5 h; additions of EDTA, GABA, glycine, taurine, strychnine, $MgCl_2$, and various levels of $CaCl_2$. Studies were also made with P_2 fractions of rat and cow brain, various regions of cow brain, cow spinal cord, and housefly abdomen plus thorax preparations. Several agents were used in an attempt to increase recovery of the bound protein, i.e., poly(ethylene glycol), poly(ethylenimine), and γ-globulin. Preliminary attempts with a centrifugation assay at 5 °C in place of the filtration procedure also failed to give satisfactory results. In fact, none of these variations gave specific binding above 10–15%, and this amount was not considered appropriate for further investigation.

DISCUSSION

Voronkov (1979) cites earlier observations with silatranes of insect growth regulator activity but not of insecticidal activity. The insecticidal silatranes reported here are new compounds containing an alkynylphenyl substituent. They appear to undergo very facile oxidative detoxification in houseflies on the basis of the >150-fold synergism by PB, but oxidative detoxification is probably less important in milkweed bugs.

The toxicity to mice is greater for the 1-arylsilatranes than for the other silatranes examined, confirming the studies of Voronkov (1979). The new 4-alkynylphenylsilatranes fall in the general toxicity range of the 4-H, 4-CH_3, 4-Cl, and 4-Br analogues, indicating that the 4-substituent within those examined has very little effect on the mammalian toxicity. The mouse toxicity of the 4-ethynylphenyl compound is reduced 2-fold on methylation and 7-fold when the trimethylsilyl substituent is introduced. Comparison of ICV and IP toxicities of the cage compounds is of interest in three respects: the LD_{50} dose as micrograms of compound per mouse is almost the same by both routes of administration; the same relation is evident for the arylsilatranes and the trioxabicyclooctanes (TBPS and TBOB); these findings are in marked contrast to the much greater potency ICV than IP in similar assays with organophosphorus (Rainsford, 1978) and pyrethroid (Lawrence and Casida, 1982) insecticides. Action in the brain relative to other sites may be less important in poisoning by these inhibitors of the GABA-gated chloride channel than for inhibitors of the voltage-dependent sodium channel (pyrethroids) and of acetylcholinesterase (organophosphorus insecticides).

Considering the phenylsilatrane $N(CH_2CH_2O)_3SiPh$ as the parent compound, the trithia analogue $N(CH_2CH_2S)_3SiPh$ and pseudosilatranes or silocanes $R_1N(CH_2CH_2O)_2Si(R_2)Ph$ [$R_1 = CH_3$ or $(CH_3)_3C$ and $R_2 = CH_3$] are of very low mammalian toxicity. Although this might be attributable to low solubility for the trithiasilatrane, this is not the case with the pseudosilatranes. The pseudosilatranes have two conformational isomers in equilibrium of which the boat–boat conformer has a geometry similar to that of the phenylsilatrane but with a much weaker N → Si interaction (Hegyes et al., 1983). Insufficient information is available at present to evaluate the relative contribution of modified substituents, geometry, and N → Si bond strength to the inactivity of the trithiasilatrane and pseudosilatranes.

The substituted phenylsilatranes examined to date are generally more toxic to mammals than to insects; e.g., several compounds have mouse IP LD_{50} values of 0.2–0.8 mg/kg and insect topical LD_{50}s of >500 mg/kg, and even the insecticidal analogues are many fold more toxic to mice than to milkweed bugs or houseflies. The selective toxicity is not due entirely to differences in metabolic detoxification on the basis of PB synergism studies: the specificity of the target site may also be involved.

The poisoning signs of the arylsilatranes are very similar to those of several other types of cage convulsants. Phenobarbital pretreatment reduces the mouse toxicity of 4-chlorophenylsilatrane to the same extent as it does for selected TBPS and TBOB analogues (Casida et al., 1976). 1-(2-Thienyl)silatrane elevates cyclic GMP levels in rat cerebellum similar to the effect of GABA-receptor blockers and bicyclophosphorus esters which act as GABA antagonists, although the symptoms of poisoning are not identical; in each case the toxicity and elevation of cyclic GMP levels are reversed by phenobarbital and diazepam (Brandt et al., 1977; Mattsson et al., 1977). These observations provide suggestive evidence but not proof that the silatranes act at the GABA-gated chloride channel.

The binding site(s) of the 1-arylsilatranes is (are) not directly determined by the studies made to date with [^{3}H]-phenylsilatrane, indicating that this candidate radioligand either has insufficient specific activity or binding affinity or that the receptor preparations examined are not relevant. The specific activity would be adequate if the percentage specific binding, affinity, and titer of binding site in brain approximate those of TBPS. The target of phenylsilatrane may be in a different tissue or locus than that of TBPS.

The GABA-gated chloride channel of mammalian brain membranes is monitored directly by the TBPS binding assay (Squires et al., 1983; Obata et al., 1988). Only the alkynylphenyl derivatives of the silatranes examined to date are significantly active in inhibiting TBPS binding. However, the potency of the silatranes in the TBPS assay appears to be a poor predictor of their toxicity to mice. 4-Ethynylphenylsilatrane at 1.0 μM is a noncompetitive inhibitor of TBPS binding, suggesting that it acts at a closely coupled or allosteric site. It therefore appears that the toxicologically relevant binding site of the silatranes differs from that of the other cage convulsants. Interestingly, the two 4-alkynylphenylsilatranes that show the highest potency in the TBPS receptor assay are also those most toxic to insects.

The 4-alkynylphenylsilatranes are new probes for further definition of the comparative pharmacology of the $GABA_A$ receptor–ionophore complex of mammals and insects.

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