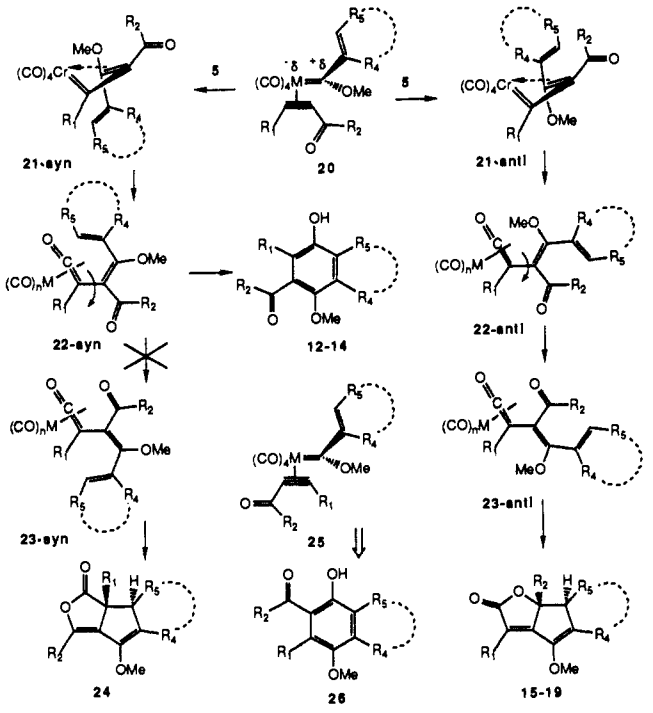
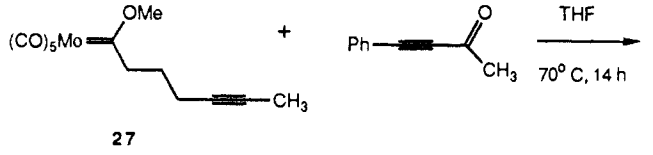
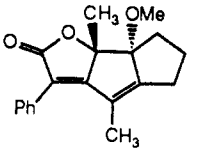
Scheme I



the acetylene substituent is controlling the regiochemistry of acetylene incorporation. Acetylenic ketones and esters have been examined previously, but in most cases they were terminal alkynes (R_1 = H) and in this situation steric factors may override electronic factors leading to phenols of the type **26** (R_1 = H). This may also explain why van Halban–White cyclization products have not been reported previously from the reactions of carbene complexes and alkynes.[8,9] Another unprecedented observation is that the regiochemistry of alkyne incorporation is affected by solvent.[10] The reaction of the molybdenum complex **7b** with 3-hexyn-2-one gives phenol **12a** and lactone **15a** in THF, whereas in acetonitrile, the phenol **26a** is the major product. The extent to which electronic factors and the nature of the solvent can control the regioselectivity in the reactions of carbene complexes with alkynes, and the extent to which the nature of the substituent R_1 can control the stereochemistry of the vinylcarbene intermediates in these reactions, are currently being investigated.



27



28 40 %

The reaction of the 4-hexynylcarbene complex **27**[11] with 4-phenyl-3-butyn-2-one illustrates the feasibility of a triple annulation for the preparation of tricyclic lactones via an in situ generated alkenylcarbene complex.[13] These observations associated with the reaction of alkenylcarbene complexes with ketoacetylenes suggest that further investigations are warranted with regard to the scope of the van Halban–White cyclizations of cross-conjugated ketenes in various configurations.

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Registry No. **7a**, 90150-06-4; **7b**, 124945-26-2; **8a**, 88426-08-8; **8c**, 111772-19-1; **9a**, 67746-50-3; **9b**, 124945-27-3; **10a**, 60920-65-2; **11a**, 109362-35-8; **12a**, 124945-09-1; **12b**, 124945-08-0; **12c**, 124945-18-2; **12d**, 124945-19-3; **13a**, 124945-10-4; **13b**, 124945-20-6; **14a**, 124945-11-5; **14b**, 124945-17-1; **15a**, 124945-12-6; **15b**, 124945-21-7; **15c**, 124991-96-4; **15d**, 124945-23-9; **16a**, 124945-13-7; **16b**, 124991-97-5; **17a**, 124945-14-8; **17b**, 124945-24-0; **18**, 124945-15-9; **19**, 124945-16-0; **27**, 124945-28-4; **28**, 124945-22-8; $H_3CC{\equiv}C(CH_2)_3I$, 28077-74-9; $Mo(CO)_6$, 13939-06-5; $CH_3COC{\equiv}CCH_2CH_3$, 1679-36-3; $PhC{\equiv}CCOCH_3$, 1817-57-8; $PhC{\equiv}CCHO$, 2579-22-8; $PhC{\equiv}CCO_2Et$, 2216-94-6; $TMSC{\equiv}CCOCH_3$, 5930-98-3; 1-cyclopentenyl-3-phenyl-2-propyn-1-one, 124945-25-1.

Supplementary Material Available: Spectral data for all new compounds (**12a–d**, **13a,b**, **14a,b**, **15a–d**, **16a,b**, **17a,b**, **18**, **19**, **26**, and **28**) and X-ray crystallographic data for compound **15b** including tables of fractional coordinates, isotropic and anisotropic thermal parameters, bond distances, and bond angles (11 pages); a listing of F_o and F_c for compound **15** (5 pages). Ordering information is given on any current masthead page.

Solid-State ^{13}C Nuclear Magnetic Resonance Studies of Lithium Fluorenide Complexes

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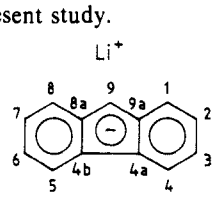
Exceptional interest in the structures of organolithium compounds has mainly been focused on the solution structure as probed by various experimental spectroscopic techniques. The solid-state structures have so far been determined by X-ray crystallography, mostly in the presence of a strong complexing agent.[1] Reports using solid-state cross-polarization/magic angle spinning (CP/MAS) NMR techniques[2] are scarce and limited to a few alkyllithium systems.[3]

In this communication we report for the first time a ^{13}C CP/MAS NMR study of a delocalized carbanion system, lithium fluorenide (**1**),[4] as a function of the complexation agent. Conflicting X-ray and solution structures of this system,[5,6] as suggested earlier, in relation to various calculations[5,7] made **1** an attractive candidate for the present study.



1

(1) Setzer, W. N.; Schleyer, P. v. R. *Adv. Organomet. Chem.* **1985**, *24*, 353.
(2) Yannoni, C. S. *Acc. Chem. Res.* **1982**, *15*, 201.
(3) (a) Gurak, J. A.; Chinn, J. W., Jr.; Lagow, R. J.; Steifink, H.; Yannoni, C. S. *Inorg. Chem.* **1984**, *23*, 3717. (b) Gurak, J. A.; Chinn, J. W., Jr.; Lagow, R. J.; Kendrick, R. D.; Yannoni, C. S. *Inorg. Chim. Acta* **1985**, *96*, L75.
(4) Complexed **1** was prepared in benzene, using *n*-butyllithium, according to a modified procedure to that given in ref 5, in the case of **1**–bisquinuclidine. In the other preparations, hexane was used as solvent. Two equivalents of the ligands were added, except for the bidentate TMEDA ligand, where 1 equiv was added. The crystals were filtered and dried for 15 min under high vacuum, before being packed into the rotor. All handling of the material was conducted under an argon atmosphere. Solid-state ^{13}C CP/MAS spectra were obtained by using a Bruker MSL 100 NMR spectrometer. Samples were rotated at 3 kHz, the repetition time was 2.5 s, and the contact time was 1 ms. Specially designed zirconium dioxide/Kel-F rotors were used throughout the study.
(5) Brooks, J. J.; Rhine, W.; Stucky, G. D. *J. Am. Chem. Soc.* **1972**, *94*, 7339.
(6) (a) Edlund, U. *Org. Magn. Reson.* **1979**, *12*, 661. (b) O'Brien, D. H.; Russell, C. R.; Hart, A. J. *J. Am. Chem. Soc.* **1979**, *101*, 633.
(7) Bushby, R. J.; Tytko, M. P. *J. Organomet. Chem.* **1984**, *270*, 265.

0002-7863/90/1512-1647$02.50/0

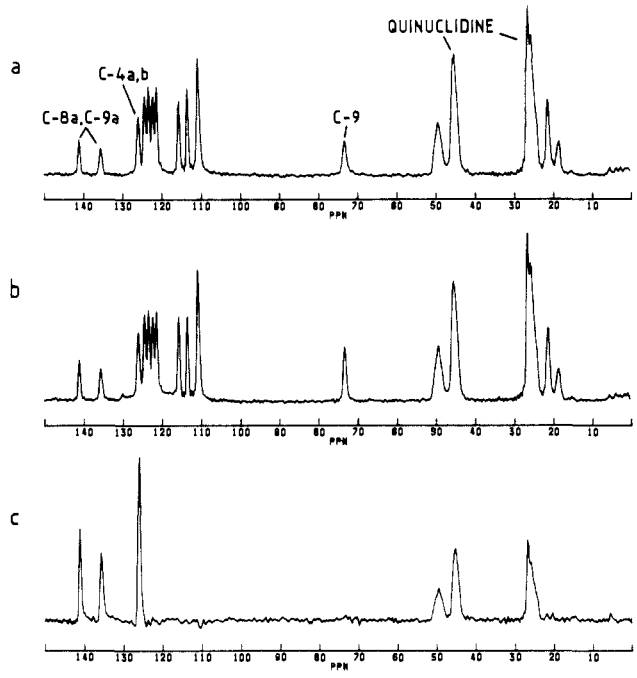


Figure 1. ^{13}C CP/MAS NMR spectrum of (a) lithium-7 fluorenide bisquinuclidine, (b) ^{13}C CP/MAS NMR spectrum of lithium-6 fluorenide bisquinuclidine; and (c) ^{13}C selective polarization inversion CP/MAS NMR spectrum of lithium-7 fluorenide bisquinuclidine with the phase inversion interval set to suppress the protonated carbons of **1**.

According to an X-ray study of **1**–bisquinuclidine, the lithium cation was asymmetrically positioned above the fluorenyl unit. The lithium cation was mainly interacting with the C-1 and C-9 positions.[5] This contrasts with the results from electrostatic calculations[7] and from solution ^{13}C NMR studies[6] where, in ethereal solvents, a symmetrical structure was suggested. The cation is then symmetrically located over the five-membered ring. However, it can be argued that solution NMR studies can not differentiate between such structures due to fast lithium exchange.

The ^{13}C CP/MAS NMR spectrum of ^{7}Li **1**–bisquinuclidine (Figure 1a) gave rise to 11 resolved lines, indicating an asymmetrical structure. This is in accordance with X-ray results.[5] In contradiction to the earlier studies of organolithium compounds,[3] lithium decoupling was not a prerequisite even when ^{7}Li was used. The carbon signals were significantly shifted relative to the shift values obtained of **1** in solution.[6] The increased shielding observed for C-9 and the splitting of the C-8a,9a resonance reflect a change in charge distribution according to the proposed X-ray structure.

Additional information of the carbon–lithium bonding could be obtained by preparing **1**–bisquinuclidine using ^{6}Li. Substitution of ^{7}Li by ^{6}Li is expected to cause a significant decrease in carbon signal line widths since ^{6}Li has a quadrupole moment only 0.5% of that of ^{7}Li. In fact, earlier studies of alkyllithium compounds showed that, even when ^{6}Li was used, a triple resonance technique, i.e. irradiation of ^{6}Li as well as ^{1}H and ^{13}C, was necessary in order to obtain reasonably narrow lines at room temperature.[3] As seen in Figure 1b, the spectrum is affected only to a minor extent by the change of lithium isotope. The minor broadening of C-9 observed for the ^{7}Li system suggests that the carbon–lithium bond is more ionic in this delocalized carbanion system compared with the bonding in alkyllithiums.

The assignment of the carbon signals is an inherent problem in ^{13}C CP/MAS NMR, especially if the solid-state and solution spectra are vastly different. Due to the poor ^{1}H resolution, only very few techniques are suitable. There is, however, one easily applicable technique to distinguish carbons with different polarization transfer rates,[8] i.e., protonated and nonprotonated carbons in delocalized systems. This technique is illustrated in Figure 1c, where the phase inversion interval was set to suppress the protonated carbons. It is seen from this figure that the C-8a and C-9a signals are distinct from each other whereas the C-4a and C-4b signals still are overlapping. The slow polarization transfer rate indicated for the methylene carbons in quinuclidine is due to rapid rotation along the quinuclidine molecular symmetry axis.

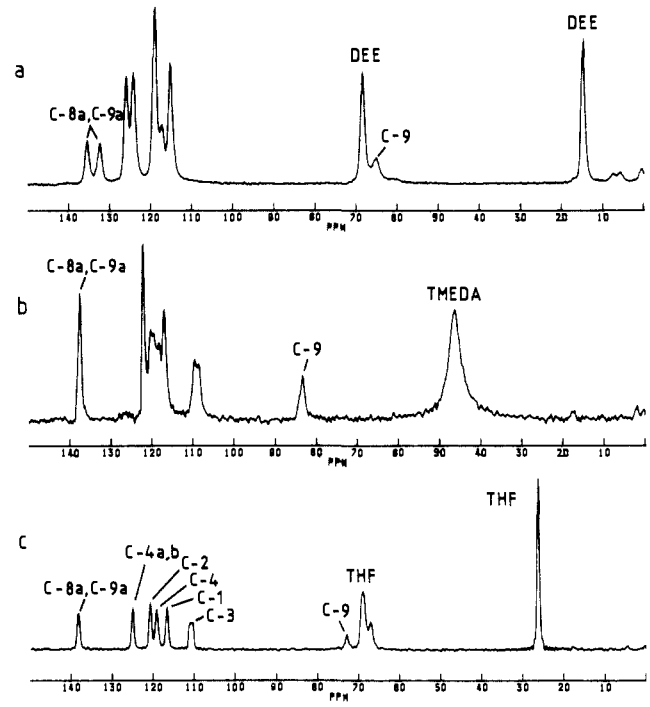


Figure 2. ^{13}C CP/MAS NMR spectra of lithium-7 fluorenide as (a) DEE, (b) TMEDA, and (c) THF complexes.

If the cation coordination is changed, one notices in diethyl ether (DEE) that the asymmetrical structure is retained (Figure 2a). C-9 is shielded relative to the value obtained when **1**–bisquinuclidine is used. This is expected, since cation-induced polarization of charge is more efficient having the weaker coordinating DEE ligand.

Using an agent that is common in X-ray studies, *N,N,N',N'*-tetramethylethylenediamine (TMEDA), we observe only a single C-8a,9a line, which suggests a symmetrical structure (Figure 2b). A similar result, with seven resolved lines, was obtained by using tetrahydrofuran (THF) as complexation agent (Figure 2c). The shift values for the aromatic carbons are all within 0.5 ppm from the values obtained in solution under contact ion pair conditions (2-methyltetrahydrofuran; room temperature).[6a] The assignments given in Figure 2c are in part based on the similarity to the solution values.

In order to clarify whether a dynamic lithium exchange process could account for the proposed symmetry, we undertook a variable-temperature study of **1**–TMEDA. The NMR signals of the fluorenyl system were practically unaffected by lowering the temperature to −60 °C. The only noticeable change was that the broad TMEDA signal at $\delta = 43$ ppm was resolved in to three peaks ($\delta = 54$, 45, and 39 ppm) due to a restriction of a dynamic process of the TMEDA molecule at low temperature.[9] The fact that the fluorenyl but not the TMEDA peaks were unaffected by lowering the temperature strongly support a symmetric arrangement of the counterion. From comparative UV studies it has been argued that **1** prefers an asymmetric structure both as a THF complex and as a TMEDA complex.[10] A fast lithium exchange is highly unlikely in the solid phase, and if it were possible, it is hard to

(8) Xiaoling, W.; Shanmin, Z.; Xuewen, W. *J. Magn. Reson.* **1988**, *77*, 343.

(9) (a) Jutzi, P.; Schlüter, E.; Siegfried, P.; Saak, W. *Chem. Ber.* **1985**, *118*, 1959. (b) Schade, C.; Schleyer, P. v. R.; Gregory, P.; Dietrich, H.; Mahdi, W. *J. Organomet. Chem.* **1988**, *341*, 19.

(10) Zerger, R.; Rhine, W.; Stucky, G. D. *J. Am. Chem. Soc.* **1974**, *96*, 5441.

rationalize why **1**–TMEDA and **1**-bisquinuclidine should have vastly different exchange rates. The condition that a change in solvation drastically could change the preferred lithium arrangement has been demonstrated recently in dilithium salts of benzocyclobutadienes.[11]

The results above are in accord with the proposal that the energy surface is rather shallow for systems of this kind and that other factors, such as crystal packing forces, may determine the actual crystal structure.[7,11,12]

It is also evident from this study that solid-phase ^{13}C CP/MAS NMR studies can provide a valuable source of structural information of organoalkali compounds. Variable-temperature studies are possible without changing the ion-pair structure as in solution. The method has a time scale that, in contrast to X-ray crystallography, can give more direct information about various dynamic processes.[13] Moreover, dipolar line broadening due to the coupling to the quadrupolar alkali nuclei can give detailed information about the nature of the carbon–alkali interaction.

Acknowledgment. We are indebted to the Swedish Natural Science Research Council for financial support.

(11) (a) Boche, G.; Etzrodt, H.; Massa, W.; Braum, G. *Angew. Chem.* **1985**, *97*, 858. (b) Bausch, J. W.; Gregory, P. S.; Olah, G. A.; Prakash, G. K. S.; Schleyer, P. v. R.; Segal, G. A. *J. Am. Chem. Soc.* In press.
(12) (a) Brooks, J. J.; Rhine, W.; Stucky, G. D. *J. Am. Chem. Soc.* **1972**, *94*, 7346. (b) Schleyer, P. v. R. *Pure Appl. Chem.* **1984**, *56*, 151.
(13) Lyerla, J.; Yannoni, C. S.; Fyfe, C. A. *Acc. Chem. Res.* **1982**, *15*, 208.

Enzyme-Facilitated Transport and Separation of Organic Acids through Liquid Membranes

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The selective separation of organic acids is important for the production of numerous biologially functional molecules, including amino acids, peptides, fats, and pharmaceuticals.[1] Enzymes, specifically lipases and proteases, have been used to selectively separate a variety of organic acids; however, multiple steps are usually required.[2] In addition, carbonic anhydrase has been used in a variety of liquid membranes for transport of CO_2.[3] In the present study, we have coupled the selectivity of enzymes with liquid membranes to provide a single-step method to selectively separate and purify organic acids.

Our experimental strategy was to use facilitative transport of a desired organic acid through a liquid membrane. This was carried out by the lipase-catalyzed esterification of the desired organic acid with a hydrophobic alcohol contained in an organic liquid membrane (see Figure 1A). The resulting ester partitions into the organic phase or is hydrolyzed to the parent acid. Once in the organic phase, the ester diffuses across the membrane where a second lipase catalyzes ester hydrolysis into the alcohol and the parent acid. If the enzyme-facilitated pathway is significantly faster than transport of the organic acids through the membrane, this will result in selective purification of the desired acid. The system employed for these studies is shown in Figure 1B.

(1) (a) Hansen, P. E.; Morgan, B. A. In *The Peptides: Analysis, Synthesis, Biology*; Udenfriend, S., Meinhofer, J., Eds.; Academic Press: Orlando, FL, 1984. (b) Pirkle, W. H.; Doherty, E. M. *J. Am. Chem. Soc.* **1989**, *111*, 4113–4114. (c) Kaiser, E. T.; Kezdy, F. J. *Science* **1984**, *226*, 249–255. (d) Wolfe, S.; Demain, A. L.; Jensen, S. E.; Westlake, D. W. S. *Science* **1984**, *226*, 1386–1392. (e) Macrae, A. R. *JAOCS* **1983**, *60*, 243A–246A.
(2) (a) Matson, S. L.; Quinn, J. A. *Annals N.Y. Acad. Sci.* **1986**, *469*, 152–165. (b) Chibata, I.; Tosa, T.; Takata, I. *Trends. Biotechnol.* **1983**, *1*, 9–11.
(3) (a) Donaldson, T. L.; Quinn, J. A. *Chem. Eng. Sci.* **1975**, *30*, 103–115. (b) Suchdeo, S. R.; Schultz, J. S. *Biochim. Biophys. Acta* **1974**, *352*, 412–440.

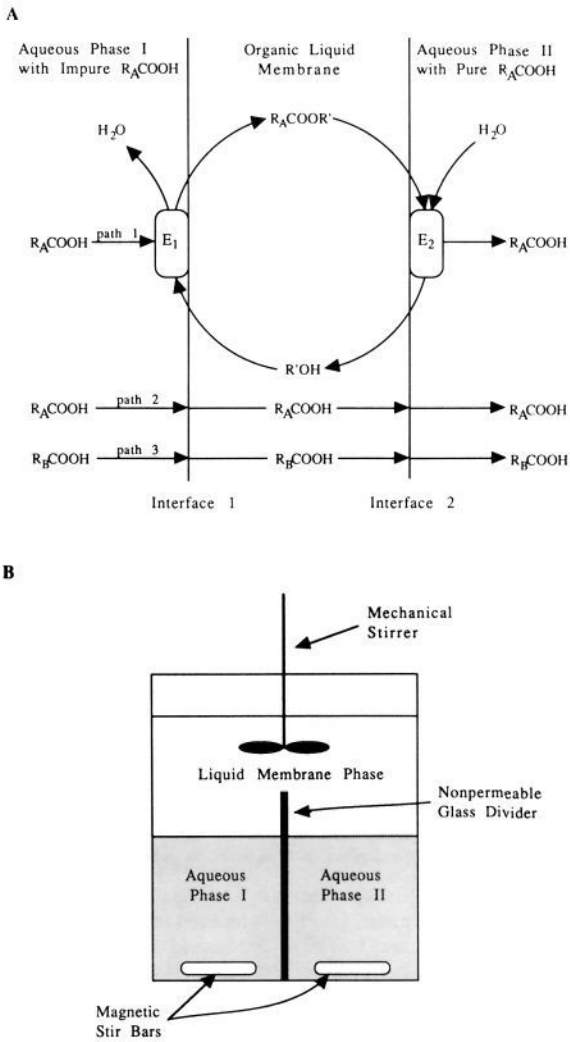

Figure 1. (A) Schematic of enzyme-facilitated liquid membrane transport for organic acid separations. R_ACOOH, R_BCOOH, R'OH, R_ACOOR', E_1, and E_2 represent the organic acids to be transported (where A is the desired acid), carrier alcohol, resulting ester, enzyme in aqueous phase I, and enzyme in aqueous phase II, respectively. Path 1 represents the enzyme-facilitated transport of the desired organic acid. Paths 2 and 3 represent nonfacilitated transport of both organic acids. (B) Enzyme-assisted membrane apparatus. The separation scheme was put into practice with a 1-L beaker with a diameter of 11 cm containing a 6-cm high impermeable glass septum separating the aqueous phases. Each aqueous phase contained 125 mL of solution and had a liquid height of 4 cm. The isooctane liquid membrane (450 mL) resided above each phase, yet the top of the isooctane phase was 3 cm above the top of the glass septum. Hence the organic solvent acted as a membrane between the two aqueous phases. Both aqueous phases were magnetically stirred at 150 rpm, and the isooctane phase was mechanically stirred at 75 rpm.

The results for the transport of 2-phenoxypropionic acid (PPA) are shown in Figure 2.[4] Lipase from *Candida cylindracea*[6] (CCL)

(4) 2-Phenoxypropionic acid was chosen as a model organic acid because it belongs to a known class of substrates of commercial lipase-catalyzed esterifications,[5] is easily analyzed by reversed-phase HPLC, and has the correct degree of hydrophobicity such that in its anionic form it resides almost solely in the aqueous phase while in its esterified form it partitions favorably into the organic liquid membrane.
(5) (a) Kirchner, G.; Scollar, M. P.; Klibanov, A. M. *J. Am. Chem. Soc.* **1985**, *107*, 7072–7076. (b) Chen, C.-S.; Wu, S.-H.; Girdaukas, G.; Sih, C. J. *J. Am. Chem. Soc.* **1987**, *109*, 2812–2817.
(6) Lipase from Candida cylindracea has been shown to be an effective esterification catalyst.[5] Lipase from porcine pancreas is a well-known hydrolytic catalyst[7] as well as transesterification catalyst yet is unable to catalyze esterification reactions.[5a] Hence reverse esterification and transport from aqueous phase II to phase I is eliminated.
(7) *Lipases*; Borgstrom, B., Brockman, H. L., Eds.; Elsevier: Amsterdam, 1984.

0002-7863/90/1512-1649$02.50/0