

Spermic Acid Diester Derivatives as Pharmacological Carriers for Long-Term Controlled Nitric Oxide Delivery

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Received July 31, 1998; accepted September 25, 1998

Water-insoluble diazeniumdiolates with extended nitric oxide (NO) release half-lives were synthesized based on diesters of spermic acid. These diesters were prepared *via* Michael addition reactions from 1,4-diaminobutane and various acrylate esters. Synthesis and NO release profiles of dimethyl, diethyl, dibutyl, dihexyl, and dilauryl spermate diazeniumdiolates are reported.

Key words nitric oxide; 1-substituted diazen-1-ium-1,2-diulates; spermic acid; alcohol; esters

1-Substituted diazen-1-ium-1,2-diulates (NO/nucleophile adducts, abbreviated as diazeniumdiolates) have been a reliable and important type of nitric oxide (NO) donor for the treatment of various diseases that are associated with NO deficiencies in humans. Research in the development of water-soluble diazeniumdiolates has been successful over the past few years with the evaluation of diazeniumdiolates in many clinical trials. Although pharmacological effects have been observed in these trials due to the NO release, systemic side effects were observed because of their water solubility, which lead to the permeation of these compounds into the bloodstream. Furthermore, these water-soluble diazeniumdiolates demonstrated short half-lives ranging from seconds to several hours. Certain clinical situations require sustained and continuous exogenous NO release without frequent doses, such as anti-aggregatory coatings of implantable biomedical sensors and lung inhalation therapy. Subsequently, new water-insoluble diazeniumdiolates with long NO release half-lives are desired. Lipophilic diazeniumdiolates of dihexadecanyl spermate and dicholesteryl spermate were previously prepared and studied.¹⁾ These compounds had long NO release half-lives of 168 h and 552 h, respectively. Based on these, we developed and studied a new family of lipophilic diazeniumdiolates from spermate diesters.

In general, spermic acid diester hydrochloride derivatives were synthesized *via* Michael addition reaction of 1.0 eq of 1,4-diaminobutane with 2.0 eq of various acrylate esters in an appropriate solvent at room temperature for 36 h, followed by the addition of hydrochloric acid.²⁾ Dilauryl spermate (DLS), however, was directly obtained in 58% yield from 1,4-diaminobutane and lauryl acrylate in THF without the addition of hydrochloride. Dimethyl spermate (DMS) dihydrochloride was synthesized in methanol by reacting 1,4-diaminobutane with methyl acrylate, followed by the addition of concentrated HCl to the solution after it was cooled with ice. White crystal DMS/2HCl was obtained in 91% yield after recrystallization from 95% methanol. Similarly, diethyl spermate (DES) dihydrochloride was prepared in ethanol, dibutyl spermate (DBS) dihydrochloride was prepared in 1-butanol, and dihexyl spermate (DHS) dihydrochloride was prepared in THF in 86%, 71%, and 56% yields, respectively. Spermic acid diesters were obtained after neutralization with excessive triethylamine (TEA) and extraction with ethyl ether in the yield of approximately 30%. These diesters subsequently reacted with NO in acetonitrile at 100 psi for 3 d, forming the corresponding water-insoluble diazeniumdiolates.

UV-Vis spectroscopy was used to identify the presence of the NONO[−] group in the diazeniumdiolates. In chloroform all of these compounds showed a maximal absorbance peak at 240–250 nm, which is characteristic of diazeniumdiolates.³⁾ These water-insoluble diazeniumdiolates were mixed with pH 7.4 phosphate-buffered saline at 37 °C and were able to release NO spontaneously. Their NO release profiles were obtained by the chemiluminescence method,⁴⁾ and showed extra-long NO release half-lives ranging from 91 to 517 h. The increase of the end-chain length resulted in an aggressive decrease in NO release half-life and a significant increase in UV extinction coefficient because the longer chain in the ester group increases the amphiphilicity, thus making the polar diazen-1-ium-1,2-diolate group more accessible to solvent protons. From the UV data, we conclude that intermolecular diazeniumdiolate was formed in DMS-D, DES-D, and partially in DBS-D, while intramolecular diaze-

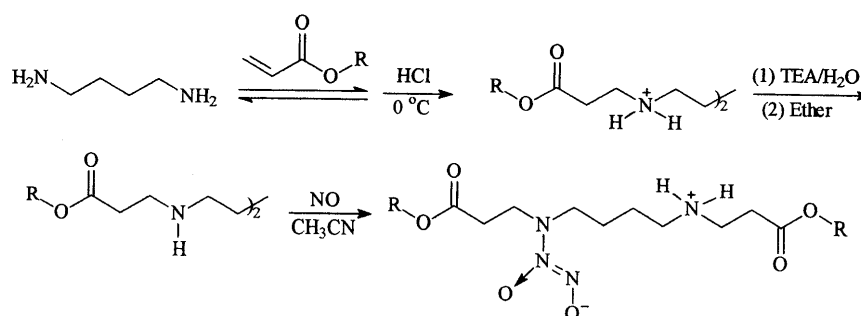


Fig. 1. Synthesis Scheme for the Diazeniumdiolates of Spermic Acid Derivatives

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Table 1. Physiological Data of the Diazeniumdiolates

Properties	DMS-D	DES-D	DBS-D	DHS-D	DLS-D
λ_{\max} (nm)	240	242	242	242	244
ϵ_{\max} (mm ⁻¹ ·cm ⁻¹)	2.84	1.65	4.13	5.67	6.45
$t_{1/2}$ (hours)	488	517	316	93	91

Table 2. ¹H-NMR Spectroscopic Data of DMS, DES, DBS, DHS, DLS, and Their Diazeniumdiolates DMS-D, DES-D, DBS-D, DHS-D, and DLS-D

DMS: 4.18 (t), 3.36 (t), 3.14 (t), 2.86 (t), 1.80 (m), 1.65 (m), 1.37 (m), and 0.91 (t)

DMS-D: 5.35 (s), 4.65 (s), 4.37 (m), 4.16 (m), 3.80 (m), 3.65 (s), 3.02 (m), 2.69 (t), 2.56 (m), and 1.26 (m)

DES: 4.12 (q), 2.85(t), 2.60 (s), 2.45 (t), 1.50 (m), and 1.22 (t)

DES-D: 4.12 (q), 3.05 (t), 2.82 (s), 2.67 (t), 1.75 (m), and 1.25 (t)

DBS: 4.08 (t), 2.87 (t), 2.62 (t), 2.51 (t), 1.61 (m), 1.52 (m), 1.37 (m), 1.05 (m), and 0.93 (t)

DBS-D: 4.08 (t), 2.94 (t), 2.70 (m), 2.58 (m), 1.62 (m), 1.37 (m), and 0.92 (t)

DHS: 4.13 (t), 3.52 (t), 3.09 (m), 2.80 (t), 1.79 (m), 1.65 (m), 1.28 (m), and 0.85 (t)

DHS-D: 4.08 (m), 3.64 (m), 3.18 (m), 2.97 (m), 2.88 (m), 1.94 (s), 1.59 (m), 1.25 (s), and 1.88 (m)

DLS: 4.07 (t), 3.45 (q), 2.87 (30, 2.62 (t), 2.58 (s), 2.53 (m), 1.60 (q), 1.51 (m), 1.30 (s), 1.21 (t), 1.03 (t), and 0.89 (t)

DLS-D: 4.34 (t), 4.19 (m), 4.07 (m), 3.23 (t), 3.10 (q), 2.97 (m), 2.81 (m), 2.54 (m), 1.61 (m), 1.30 (m), and 0.88 (m)

300 MHz, in CDCl₃.

numdiolate was formed in DHS-D and DLS-D.⁵⁾ The structures of these molecular species were partially identified by NMR spectroscopy, although it is not certain if NMR spectroscopy could differentiate the intermolecular and the in-

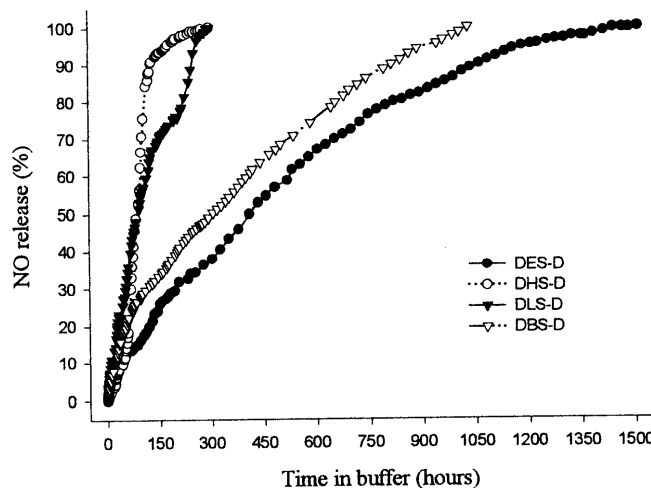


Fig. 2. NO Release Profiles of Diazeniumdiolates under Physiological Conditions

tramolecular salt species. Their water insolubility will lead to site-specific NO release for pharmacological applications, thus reducing adverse systemic side effects. The spermate ester carrier should be able to decompose into nontoxic materials since poly(butanediol spermate) rapidly decomposes after it delivers NO under physiological conditions.⁶⁾

References

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