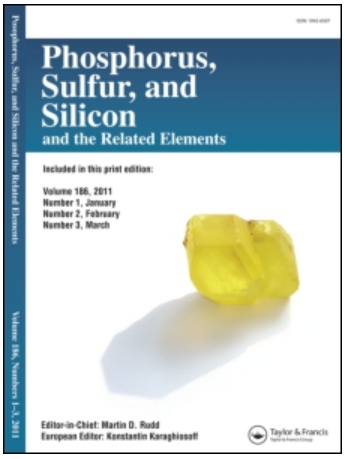

Phosphorus, Sulfur, and Silicon and the Related Elements
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713618290

Cytotoxic Activity of Ammonium Tellurates

Pavel Arsenyan[a]; Irina Shestakova[a]
[a] Latvian Institute of Organic Synthesis, Riga, Latvia

To cite this Article Arsenyan, Pavel and Shestakova, Irina(2009) 'Cytotoxic Activity of Ammonium Tellurates', Phosphorus, Sulfur, and Silicon and the Related Elements, 184: 9, 2427 — 2430
To link to this Article: DOI: 10.1080/10426500802487656
URL: http://dx.doi.org/10.1080/10426500802487656

Phosphorus, Sulfur, and Silicon, 184:2427–2430, 2009

ISSN: 1042-6507 print / 1563-5325 online
DOI: 10.1080/10426500802487656

Taylor & Francis
Taylor & Francis Group

Cytotoxic Activity of Ammonium Tellurates

Pavel Arsenyan and Irina Shestakova
Latvian Institute of Organic Synthesis, Riga, Latvia

The cytotoxic activity of a series of methylammonium tellurates on human fibrosarcoma HT-1080, mouse hepatoma MG-22A, and mouse fibroblasts 3T3 cell lines is described. The role of tellurates as free radical regulators is discussed.

Keywords Acute toxicity; cytotoxicity; tellurium

INTRODUCTION

In last decade the investigation of tellurium compounds as antitumor agents has attracted much attention.[1] Selenium and tellurium share unique chemical characteristics; however, there are some differences that make tellurium compounds of interest. Inorganic tellurium derivatives exhibit high toxicity to the central nervous system of rodents.[2] Inorganic tellurium (IV) compounds are metabolized by a route similar to selenium (IV), but in contrast to selenium, the methylated products of tellurium are considered more toxic to mammals.[3] It has been shown that sodium tellurite exhibits a cytotoxic effect on cultured HeLa cells.[4] The immunomodulator ammonium trichloro(dioxyethylene-O-O′) tellurate (AS101) has been shown to possess antitumor properties in the early stage of Madison 109 lung adenocarcinoma. Treatment with optimal doses of Taxol (25 and 17 mg/kg) and AS101 (0.5 mg/kg) resulted in 66.6 and 43.3% cures.[5] Previously, we reported that hydroselenites are able to activate nitric oxide generation in various tumor cell cultures.[6–8] The activation of nitric oxide production is especially prolonged in HT-1080 and MG-22A cell lines. The amount of nitric oxide produced depends on the type of tumor and the cation structure. According to the literature and our results, we propose that the antitumor

Received 8 March 2008; accepted 18 September 2008.
The financial support for this work provided by the Latvian Council of Science (grant No. 05.1757) is gratefully acknowledged.
Address correspondence to Pavel Arsenyan, Latvian Institute of Organic Synthesis, Aizkraukles 21, LV-1006, Riga, Latvia. E-mail: pavel@osi.lv

effect of hydroselenites stems from the fact that they are able to regulate the free radical balance *in vitro* and *in vivo*. In the current study, we report the cytotoxic activity of selected organoammonium tellurates *in vitro* against HT-1080, MG-22A, and NIH 3T3 cell lines. The role of tellurates as free radical regulators is discussed.

RESULTS AND DISCUSSION

It has been shown that telluric acid (**1**) readily reacts with ammonia, methyl amine, and dimethyl amine in water to give the corresponding ammonium tellurates **2**–**4**.

The reaction proceeds easily at room temperature with quantitative yields.

The results of *in vitro* experiments are summarized in Table I. The majority of tested tellurates **1**–**4** exhibited slight activity *in vitro* on human fibrosarcoma HT-1080 (MTT test) and good activity on mouse hepatoma MG-22A tumor cell lines. Telluric acid (**1**) shows high cytotoxic effect on the MG-22A cell line ($TD_{50} = 0.2$ μg/mL, MTT test). The conversion of **1** to ammonium salts (**2–4**) leads to a decrease of the cytotoxic effect on both studied tumor cell lines ($TD_{50} = 14 \div 61$ μg/mL, MTT test). It should be noted that in the same concentrations telluric acid (**1**) and their ammonium tellurates **2**–**4** very selectively act against tumor and normal mouse fibroblast (NIH 3T3) cells. Moreover, in spite of the literature data, this series of tellurium containing compounds exhibits very low LD_{50} toxicity (1236–2080 mg/kg).

The role of NO in biosystems has attracted considerable attention in the last decade. NO is formed by enzymatic and nonenzymatic mechanisms. Because of its molecular weight and high lipophilicity, NO has

TABLE I *In Vitro* Cytotoxicity in Monolayer Tumor Cell Lines HT-1080 (Human Fibrosarcoma), MG-22A (Mice Hepatoma), NIH 3T3 (Normal Mouse Fibroblasts) and Caused by Tellurates 1–4[a]

	HT-1080			MG-22A			3T3	
Compound	TD_{50}, CV	TD_{50}, MTT	NO 100%	TD_{50}, CV	TD_{50}, MTT	NO 100%	TD_{50}	LD_{50}, mg/kg
1 H_6TeO_6	>100	26	13	34	0.2	72	427	1236
2 $(NH_4)_6$ TeO_6	>100	52	13	71	15	25	545	1726
3 $(NMeH_3)_6$ TeO_6	>100	61	11	74	16	18	614	2080
4 $(NMe_2H_2)_6$ TeO_6	>100	52	9	30	14	88	330	1870

[a]TD_{50}: Concentration (μg/mL) providing 50% cell killing effect; NO concentration (%) (CV: coloration).

good diffusion properties. It may act not only in the cell where it is produced but also in nearby tissues. Biologically produced NO originates from oxygen and L-arginine in the reaction catalyzed by NO synthase. NO, a long-lived radical with a wide range of actions, is known as a regulator of a variety of biological processes.[9–11] It was shown (Table I) that telluric acid (**1**) is NO radical protector on MG-22A cell line (TD_{100} = 72%). An inspection of the ability of ammonium tellurate (**2**) to protect cells from free radical influence shows that compound **2** is a strong NO-inhibitor (TD_{100} = 25%). The introduction of one methyl group into the ammonium cation **3** leads to extended NO-protecting properties (TD_{100} = 18%). Besides, the experimental data of the dimethyl ammonium analogue **4** show only a slight free radical protecting effect (TD_{100} = 88%). According to our investigations, the studied ammonium tellurates have no influence on tumor and normal cell phenotype.

EXPERIMENTAL

In Vitro Cytotoxicity Assay

Monolayer tumor cell lines: MG-22A (mouse hepatoma), HT-1080 (human fibrosarcoma), and normal mouse fibroblasts (NIH 3T3) were cultured in a standard medium DMEM (Dulbecco's modified Eagle's medium) without an indicator (Sigma) supplemented with 10% heat-inactivated fetal bovine serum (Sigma). After the ampoule was thawed, the cells from 1 to 4 passages were used. About 2–5 $\times$ 10^4 cells/mL (depending on line nature) were placed in 96-well plates immediately after the compounds were added to the wells. The control cells without test compounds were cultured on a separate plate. The plates were incubated for 72 h, 37°C, 5% CO_2. The number of surviving cells was determined using tri(4-dimethylaminophenyl)methyl chloride (Crystal Violet) or 3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolinium bromide (MTT). The quantity of live cells on the control plate was taken in calculations as 100%.[12,13] The concentration of NO was determined according to Gryess method (by NO_2 level in cultural medium). Sodium nitrite standard solution was used for the calibration curve.[12]

Morphology Assay

The change in cell morphology caused by tellurates was investigated on Nikon ECLIPSE TE 300 microscope slides. Crystal violet and acridine orange stains were used. The adherent cells were stained in the plate wells following culture of the cells with tellurates. Chromatin

condensation in apoptotic cells was visualized by staining the cellular DNA with the dye acridine orange.[14]

REFERENCES

[1] C. W. Nogueira, G. Zeni, and J. B. T. Rocha, *Chem. Rev.*, **104,** 6255 (2004).
[2] R. E. Perez-D'Gregorio and R. K. Miller, *Teratology*, **37,** 307 (1988).
[3] B. P. Laden and T. D. Porter, *J. Lipid Res.*, **42,** 235 (2001).
[4] W. J. Ding, T. Hasegawa, D. Peng, H. Hosaka, and Y. Seko, *J. Trace Elem. Med. Biol.*, **16,** 99 (2002).
[5] Y. Kalechman, A. Shani, S. Dovrat, J. K. Whisnant, K. Mettinger, M. Albeck, and B. Sredni, *J. Immunol.*, **156,** 1101 (1996).
[6] E. Lukevics, P. Arsenyan, I. Shestakova, I. Domracheva, I. Kanepe, S. Belyakov, J. Popelis, and O. Pudova, *Appl. Organometal. Chem.*, **16,** 228 (2002).
[7] E. Lukevics, P. Arsenyan, K. Rubina, I. Shestakova, I. Domracheva, I. Kanepe, J. Popelis, and O. Pudova, *Appl. Organometal. Chem.*, **16,** 235 (2002).
[8] P. Arsenyan, I. Shestakova, K. Rubina, I. Domracheva, A. Nesterova, K. Vosele, O. Pudova, and E. Lukevics, *Eur. J. Pharm.*, **465,** 229 (2003).
[9] F. A. Lopez, L. S. De Miguel, C. Caramelo, M. J. Gomez, R. Garcia, J. R. Mosquera, T. De Frutos, I. Millas, F. Rivas, G. Echezarreta, and S. Casado, *Am. J. Physiol.*, **272,** H760 (1997).
[10] F. Brockhaus and B. Bruene, *Exp. Cell Res.*, **238,** 33 (1998).
[11] K. H. Edmiston, Y. Shoji, T. Mizoi, R. Ford, A. Nachman, and J. M. Jessup, *Cancer Res.*, **58,** 1524 (1998).
[12] D. J. Fast, R. C. Lynch, and R. W. Leu, *J. Leucocyt. Biol.*, **52,** 255 (1992).
[13] P. J. Freshney, *Culture of Animal Cells (A Manual of Basic Technique)* (Wiley-Liss, New York, 1994), pp. 296–297.
[14] A. C. Loweth and N. G. Morgan, Methods for the study of NO-induced apoptosis in cultured cells, In: *Methods in Molecular Biology: Nitric Oxide Protocols*, M. A. Titheradge, Ed. (Humana Press, Totowa, NJ, 1998), pp. 311–320.