BURGER'S MEDICINAL CHEMISTRY AND DRUG DISCOVERY

Fifth Edition

Volume 4: Therapeutic Agents

Edited by

Manfred E. Wolff

Technipharm Consultants
Laguna Beach, California

A WILEY-INTERSCIENCE PUBLICATION

JOHN WILEY & SONS, INC., New York • Chichester • Weinheim • Brisbane • Singapore • Toronto

BURGER'S MEDICINAL CHEMISTRY AND DRUG DISCOVERY

Fifth Edition

Volume 4: Therapeutic Agents

Edited by

Manfred E. Wolff

Technipharm Consultants
Laguna Beach, California

A WILEY-INTERSCIENCE PUBLICATION

JOHN WILEY & SONS, INC., New York, Chichester · Weinheim · Brisbane · Singapore · Toronto

Notice Concerning Trademark or Patent Rights.
The listing or discussion in this book of any drug in respect to which patent or trademark rights may exist shall not be deemed, and is not intended as a grant of, or authority to exercise, or an infringement of, any right or privilege protected by such patent or trademark.

This text is printed on acid-free paper

Library of Congress Cataloging in Publication Data:
Burger, Alfred, 1905–
[Medicinal chemistry]
Burger's medicinal chemistry and drug discovery.—5th ed. / edited by Manfred E. Wolff.
p. cm.
"A Wiley-Interscience publication."
Contents: v. 4. Therapeutic agents
Includes bibliographical references and index.
ISBN 0-471-57559-3
1. Pharmaceutical chemistry. I. Wolff, Manfred E. II. Title. III. Title: Medicinal chemistry and drug discovery.
RS403.B8 1994
615'. 19—dc20 94-12687

Printed in the United States of America

10 9 8 7 6 5 4 3 2 1

Preface

Volume 4 of this series is concerned primarily with the discussions of cardiovascular drugs and chemotherapeutic agents that were not considered in Volume 2. As was indicated there, in the interests of publishing the chapters in a timely manner, a decision was taken to divide each subject area into two parts, which could be published in an early and a later volume.

In an era of medical cost containment, it is important to note the large economic advantage provided to society by the new agents whose discovery and development is chronicled in these volumes. Today, heart disease is the most important killer in western-style countries. The costs of healthcare and hospitalization for such diseases is reduced tremendously by the availability of safe and effective medicines. Another notable point regarding drug development of particular importance to these agents is the difficulty of designing a definitive clinical trial in a multifactorial disease.

Industrial support for the research groups in the field of chemotherapy has declined substantially in past decades, owing to the availability of highly effective compounds. For example, the comparatively small changes in the status of antiparasitic drug discovery required only update discussions regarding the clinical development of existing agents. But drug resistance by pathogenic organisms has seriously eroded the usefulness of existing medicines, and this picture is changing drastically. Thus, the discovery and development of new medicines for chemotherapy, utilizing new mechanisms of action, is a high priority interest once more.

Again I thank my friends, the dedicated authors, who generously took time from their already overcrowded schedules to pass their expert knowledge on to others. I am grateful to Michalina Bickford, Managing Editor with John Wiley & Sons, Inc. for her work in connection with this series. As always I thank my wife, Gloria, for her steadfast support and encouragement in everything I do.

MANFRED E. WOLFF

Laguna Beach, California

Contents

Part I Radiological Agents, Pt. 1, 1

46. RADIOSENSITIZERS AND RADIOPROTECTIVE AGENTS, 3

William Foye
Massachusetts College of Pharmacy and Allied Science
Boston, Massachusetts, USA

Edward A. Bump
Harvard Medical School
Boston, Massachusetts, USA

Part II Cardiovascular Drugs, Pt. 2, 71

47. MYOCARDIAL INFARCTION AGENTS, 73

Ruth A. Altschuld and George E. Billman
Department of Medical Biochemistry
Ohio State University
Columbus, Ohio, USA

48. ENDOGENOUS VASOACTIVE PEPTIDES, 101

A. A. Houdi and K. F. Hauser
University of Kentucky
Lexington, Kentucky, USA

49. HEMATOPOIETIC AGENTS, 151

Maureen Harrington
Walther Oncology Center
University of Indiana Medical School
Indianapolis, Indiana, USA

50. ANTICOAGULANTS, ANTHROMBOTICS, AND HEMOSTATICS, 179

William Bell and David F. Kong
Department of Medicine/Division of Hematology
Johns Hopkins University
Baltimore, Maryland, USA

Part III Chemotherapeutic Agents, Pt. 2, 247

51. SYNTHETIC ANTIBACTERIAL AGENTS, 249

William Remers and Soaring Bear
College of Pharmacy
University of Arizona
Tucson, Arizona, USA

52. LACTAM ANTIBIOTICS, 277

Robert Southgate, Neal F. Osborne, Michael J. Pearson, and George Burton
SmithKline Beecham Pharmaceuticals
Betchworth, Surrey, United Kingdom

53. ANTHELMINITIC AGENTS, 365

Peter J. Islip
The Wellcome Research Laboratories
Beckenham, Kent, United Kingdom

UPDATE ON ANTHELMINTIC AGENT DEVELOPMENT: CLINICAL RESULTS AND EXPERIENCE, 414

Adolfo Martínez-Palomo and Martha Espinosa Cantellano
Center for Research and Advanced Studies
Mexico City, Mexico

54. ANTIAMEBIC AGENTS, 429

William Ross
Lightwater, Surrey, United Kingdom

UPDATE ON ANTIAMEBIC AGENT DEVELOPMENT: CLINICAL RESULTS AND EXPERIENCE, 453

Adolfo Martínez-Palomo and Martha Espinosa Cantellano
Center for Research and Advanced Studies
Mexico City, Mexico

55. CHEMOTHERAPY OF AFRICAN TRYPANSOMIASIS, 459

Ching Cheng Wang
School of Pharmacy
University of California
San Francisco, California, USA

56. ANTIVIRAL AGENTS, DNA, 487

Kenneth F. Bastow and Pannarat Akanitapichat
The School of Pharmacy
The University of North Carolina
Chapel Hill, North Carolina, USA

Part IV Endocrine Drugs, Pt. 2, 551

57. FEMALE SEX HORMONES AND ANALOGS, 553

Peter Carmichael Ruentiz
University of Georgia
Athens, Georgia, USA

58. AGENTS AFFECTING THE IMMUNE RESPONSE, 589

Stewart Wong
Jing Xing Health and Safety Resources, Inc.
Annandale, Virginia, USA

INDEX, 633

PART I
RADIOLOGICAL AGENTS, Pt. 1

CHAPTER FORTY-SIX

Radiosensitizers and Radioprotective Agents

EDWARD A. BUMP

Joint Center for Radiation Therapy
Harvard Medical School
Boston, Massachusetts, USA

and

WILLIAM O. FOYE

Department of Chemistry
Massachusetts College of Pharmacy and Allied Health Sciences
Boston, Massachusetts, USA

CONTENTS

1 Protective Agents Against Ionizing Radiation, 5
1.1 Introduction, 5
1.2 Radiation damage, 5
1.3 Antiradiation testing, 6
1.4 Protective compounds, 7
1.4.1 Thiols and thiol derivatives, 8
1.4.2 Other sulfur containing compounds, 13
1.4.3 Metabolic inhibitors, 16
1.4.4 Agents involving metal ions, 16
1.4.5 Hydroxyl-containing compounds, 16
1.4.6 Heterocyclic compounds, 17
1.4.7 Physiologically active substances, 18
1.4.8 Metabolites of naturally occurring compounds, 19
1.4.9 Polymeric substances, 20
1.4.10 Miscellaneous substances, 20
1.5 Mechanisms of protective action, 21
1.5.1 Protection by anoxia or hypoxia, 21
1.5.2 Inhibition of free-radical processes, 21
1.5.3 Mixed disulfide hypothesis, 22
1.5.4 Biochemical shock, 23
1.5.5 Control of DNA breakdown, 23

Burger's Medicinal Chemistry and Drug Discovery,
Fifth Edition, Volume 4: Therapeutic Agents,
Edited by Manfred E. Wolff.
ISBN 0-471-57559-3

1.5.6 Modes of restoration, 24
1.5.7 Role of metal ions, 25
1.5.8 Metabolic effects, 27
1.5.9 Use of radioprotective agents in radiotherapy and chemotherapy of cancers, 29
2 Radiosensitizers, 30
2.1 Introduction, 30
2.2 Radiosensitization by alteration of energy absorption, 30
2.2.1 Boron neutron capture therapy (BNCT), 30
2.2.2 K-edge absorption and photoactivation of elements of high atomic number, 31
2.2.3 Photodynamic therapy, 33
2.3 Alteration of the primary radiolytic products, 34
2.4 Radiosensitization by reaction with DNA radicals, 35
2.5 Additional applications for electron-affinic drugs in cancer therapy, 38
2.5.1 Binding of nitroimidazoles to hypoxic cells: use in detection of hypoxia, 39
2.5.2 Additional sensitization by hypoxic metabolism of nitroimidazoles, 39
2.5.3 Bioreductive drugs, 39
2.6 Radiosensitization by alterations of oxygen delivery, 41
2.7 Radiosensitization by depletion of endogenous protectors, 45
2.8 Radiosensitization by inhibition of DNA repair, 46
2.8.1 Inhibition of PLD repair, 46
2.8.2 Radiosensitization by reaction with protein sulfhydryls, 47
2.9 Radiosensitization by perturbation of cellular metabolism, 49
2.9.1 Perturbation of energy metabolism, 49
2.9.2 Abrogation of G2 delay, 49
2.9.3 Radiosensitization by growth factors and cytokines, 50
2.9.4 Halogenated pyrimidines, 50
2.10 Radiosensitizers for which the mechanism of sensitization has not been established, 51
2.10.1 Metal–ion complexes, 51
2.10.2 Thiols and miscellaneous compounds, 52
2.10.3 Bacterial radiosensitizers, 52
3 Summary and Prospects for Future Development of Radiation Modifiers, 53

1 PROTECTIVE AGENTS AGAINST IONIZING RADIATION

1.1 Introduction

The protective action of certain substances against the damaging effects of ionizing radiation was first noted in 1942 (but not published until 1949) by Dale, Gray, and Meredith. A decrease was observed in the inactivation of two enzymes by X-rays on addition of several substances, including colloidal sulfur and thiourea, to aqueous preparations of the enzymes (1). Radioprotective effects for a bacteriophage were observed by Latarjet and Ephrati in 1948, using cysteine, cystine, glutathione, thioglycolic acid, and tryptophan (2). Radioprotection of mice against X-rays was achieved shortly after in three different laboratories, in Belgium, the USA, and Britain, using cyanide (3), cysteine (4), and thiourea (5), respectively. These protective effects were attributed at the time to inhibition of, or reaction with, cellular enzymes. The importance of sulfur-containing molecules for radioprotection was thus demonstrated from the very earliest experiments with living systems, although the reasons for selection of sulfur compounds were not clear.

The importance of the mercapto (or thiol) function was demonstrated in 1951 by Bacq (6), a Belgium physiologist, who removed the carboxyl group of cysteine and obtained 2-mercaptoethylamine (MEA, or cysteamine) (**1**), which proved to be a much

$$NH_2CH_2CH_2SH$$

(1)

stronger protective agent in mice than any previously tested. The presence of the amino group was also considered essential for good radioprotection, and most of the mercaptans and other sulfur-containing molecules, later synthesized, also contained an amino or other basic function. MEA and its derivatives, particularly those having greater lipophilic character, are still regarded as the most potent of the whole body radioprotective agents.

Since 1952, other types of structures have been found with radioprotective abilities, including a number of physiologically important agents, notably serotonin, but none has yet exceeded the amino alkyl mercaptans in effectiveness. Various explanations have been put forward for the protective ability of the thiols, but it was not until more knowledge was available regarding the radicals generated by ionizing radiation, and their effect on DNA, that present concepts became established.

Research attempts to explain the action of chemical radiation protectors have involved the use of not only mammals, but plants, bacteria, distinct types of cells, and even some synthetic plastics affected by ionizing radiation. This discussion attempts to list the various types of chemical structures that afford some protection against the deleterious effects of high energy ionizing radiation in mammals and to describe the more widely considered mechanisms of protection by which they may act.

1.2 Radiation Damage

Biologic effects of radiation have been reviewed in detail by a number of authors, including von Sonntag (7), Pizzarello and Witcofski (8), Casarett (9), Okada (10), and Dertinger and Jung (11). In general, ionizing radiation results in damage to the blood-forming organs, gastrointestinal system, or central nervous system, depending on radiation dose. Hematopoietic death from bleeding, infection, or anemia is the type usually observed from antiradiation screening of potential protectants, and follows 7–30 days after exposure to a lethal dose (~1000 rads) in mice.

Absorption of radiation energy by biological molecules has been considered to be either direct or indirect, although the distinction between them is not clear. Direct action

involves absorption of radiation energy by target molecules, such as DNA or RNA, resulting in molecular damage. Indirect action involves the release of radiation energy in the environment of a target molecule with transfer of the energy to the target molecules by free radicals. These are generally considered to be the radiolytic products of water, which deposit their energy in biological systems and create excited molecules and ions, or ultimately semistable bioradicals, which lead to molecular alterations and resulting loss of normal biological activity. The following model reactions have been suggested (12):

$$RH \leftrightarrow R\cdot + H\cdot \qquad (46.1)$$

$$RH + H\cdot \rightarrow R\cdot + H_2 \qquad (46.2)$$

In the presence of oxygen:

$$R\cdot + O_2 \rightarrow RO_2\cdot \qquad (46.3)$$

And in the presence of a sulfhydryl compound:

$$R\cdot + RSH \rightarrow RH + RS\cdot \qquad (46.4)$$

These model reactions show how molecular oxygen sensitizes cellular molecules to radiation damage by preventing the reversal of reaction (46.1) and producing a damaging peroxy radical. A rapid hydrogen atom donor, such as a thiol, is considered responsible for instantaneous repair or restoration (13). Repair usually refers to an enzymatic process, whereas the term "restoration" includes introduced agents as well.

The radiolytic products of water are hydrogen atoms, hydroxyl radicals, hydrated electrons, and several nonradical species, hydrogen molecules, hydrogen peroxide, and hydronium ions:

$$H_2O \rightarrow H\cdot + OH\cdot + e^-_{aq} + H_2O_2 + H_3O^+ + H_2 \qquad (46.5)$$

Regardless of the direct or indirect transfer of energy to a molecule, radiation inactivation can be transferred to other molecules through coupled reactions (14). Thus a marked amplification of radiation damage can result. It can be seen, however, that the presence of a sulfhydryl group, or molecule capable of scavenging the radiolytic radicals from irradiation of water molecules, can bring about radiation protection. Hydroxyl radical scavenging may be difficult in cellular conditions because of the short half life of the radical and the concentration required of the protection agent.

1.3 Antiradiation Testing

Much of the early testing of antiradiation agents employed X- or γ-rays from an external source. These high energy radiations cause the ejection of electrons from the atoms through which they pass with resulting ionization. Neutrons have been used infrequently. Test animals most often have been mice or rats, with guinea pigs used less frequently. Antiradiation testing with dogs or monkeys has been limited to the more effective compounds as determined from screening with mice or rats. Further information on this testing, using 30-day survival as criterion for protection, may be found in texts devoted to radiobiology (15, 16) from that era.

Various physiological effects may be observed, depending on the dose and type of radiation, as well as on the type of animal used. In theory, the appearance of any observable symptom of radiation damage may be used as the basis of a testing procedure, but historically lethality has generally been the criterion for protection. Sufficient numbers of animals must be employed for statistical significance, and in the case of mice irradiated with a lethal dose of X- or γ-rays, a 30-day survival period is generally observed. Testing results are expressed most com-

monly as the percentage survival for the observation period in comparison to the survival of control animals. Another method of expression of test data is in terms of the dose reduction factor (DRF) (the ratio of the radiation dose causing an effect such as LD_{50} in the treated animals or cells compared to the same effect from irradiation in the unprotected animals or cells). Recently, radioprotector studies have been most frequently carried out in cell culture (17). Particular mechanisms of protection may be more effective with regard to certain end-points (17). For example, protection against mutagenesis has been observed at lower concentrations of amino thiols than are required for protection against cell killing.

The dose of protective agent employed is usually the maximum tolerated dose, (MTD), i.e., the dose causing no deleterious effects. In a drug-screening program, candidate compounds are usually tested at their MTD level using a radiation dose that is lethal to all control animals in 30 days. The time interval between administration of the drug and irradiation of the animals is usually 15–30 min for intraperitoneal dosing and 30–60 min for oral dosing. Drugs believed to act by hypoxia or other metabolic changes usually must be administered several hours prior to irradiation. Rate of irradiation in screening programs has commonly been 50–250 rads/min. At lower rates, the time for maximum effectiveness of drug can be exceeded before the total dose is administered. In addition, repair processes could become significant before the irradiation is complete. Chronic radiation studies have been carried out with repeated administration of protectant, but results have been less decisive (18).

Several concepts should be kept in mind in regard to testing results of radiation protectors. (1) Since many compounds have been tested at the maximum tolerated dose, the best protectors are not necessarily the most active on a molar basis, but rather the most effective at a given level of toxicity. (2) Although many of these compounds are active as the parent compound, some are activated metabolically, or may act indirectly by inducing an endogenous protective mechanism.

Other testing procedures used to a lesser extent include the inhibition of bacterial or plant growth and the prevention of depolymerization of polymethacrylate or polystyrene (19) or of DNA (20). Plaque-forming ability of coliphage T (21), effect on Eh potential (22), inhibition of peroxide formation of unsaturated lipids or β-carotene (22), and inhibition of chemiluminescence of γ-irradiated mouse tissue homogenates (23), as well as use of spleen colony counts (24) have also been employed as test procedures.

1.4 Protective Compounds

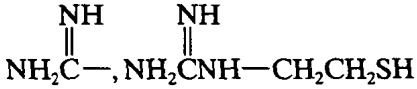
The more extensively investigated compounds have been discussed in books by Thomson (16), Bacq (25), Balubukha (26), Nygaard and Simic (27), Livesey, Reed and Adamson (28), and Bump and Malaker (17). A catalog of compounds tested for radiation protection up to 1963 was compiled by Huber and Spode (29). Extensive reviews on protective agents since 1963 have been written by Melching and Streffer (30), Overman and Jackson (31), Romantsev (32), Foye (33), Klayman and Copeland (34), Yashunskii and Kovtun (35), and Bump and Brown (36). Reports of two international symposia on radioprotective and radiosensitizing agents have been published by Paoletti and Vertua (37) and by Moroson and Quintiliani (38). A series of symposia on radiation modifiers has also been held (39). Chapters on radioprotective agents have appeared in *Annual Reports in Medicinal Chemistry* in 1966 and 1967 (40) and in 1968 and 1970 (41), and in *Military Radiobiology in 1987* (42).

In the following discussion of structure–activity relationships, results on radioprotec-

tion of mice are compared unless otherwise stated. Relevant details concerning radiation dose, compound dose, route of administration, or strain of test animal, variations of which can alter results significantly, may be found in the original references.

1.4.1 THIOLS AND THIOL DERIVATIVES. 2-Mercaptoethylamine (MEA, cysteamine) and its derivatives have constituted the most effective class of radioprotective compounds. Since the initial discoveries of the protective action in mice of cysteine by Patt et al. (4) and its decarboxylated derivative, MEA, by Bacq et al. (6), hundreds of derivatives and analogs of the mercaptoethylamine structure have been synthesized and tested for radioprotective activity. In the USA, the Walter Reed Army Institute of Research funded a large synthetic program and developed a screening procedure for compounds mainly of this type, during the period of 1959–1986. A compilation of the compounds tested in this program was made by T. R. Sweeney of the WRAIR in 1979. Many European countries also supported research programs on the development of radioprotective compounds joined more recently by China and India. Other types of agents have been found with protective activities, but sulfur-containing molecules have been by far the most numerous.

Several structural requirements for activity in this series have become established. The presence of a free thiol group, or a thiol derivative that can be converted to a free thiol *in vivo*, is essential for activity. The presence of a basic function (amino, amidino, or guanidino (43)) (**2**) located two or three



(2)

carbon atoms distant from the thiol group is favorable for the best activity. Activity for these basic thiols drops off drastically with more than a three-carbon distance (44). The benefit due to the basic group has not yet been explained, however. Several acyl thiol derivatives (**3**), such as the thiosulfuric acid

$$^{+}NH_3CH_2CH_2SX \quad X{=}SO_3^-, PO_3H^-, CS_2^-$$

(3)

(45), phosphorothioic acid (46), and trithiocarbonic acid (47), most likely liberate free thiol in the animal.

Some radioprotectors may act by releasing endogenous nonprotein thiols normally bound by disulfide linkages with serum or interstitial proteins. An increase in tissue nonprotein thiol levels results after administration of the thiosulfate and phosphorothioate of MEA (48).

Alkylation of the nitrogen of MEA causes loss of activity in some cases, but has also resulted in some of the most potent of the MEA derivatives, of which WR2721 (**4**) is

$$^{+}NH_3CH_2CH_2CH_2NHCH_2CH_2SPO_3H-$$

(4)

the best known. The *N*-β-phenethyl and *N*-β-thienylethyl derivatives have good activity (49). Dialkylation of the nitrogen of MEA usually results in some loss of activity. Whereas the *N*,*N*-dimethyl and *N*,*N*-diethyl derivatives retain much of the activity of the MEA, the *N*,*N*-dipropyl and *N*,*N*-diisobutyl derivatives retain a little activity, the di-*n*-butyl derivative is inactive (44). *N*,*N*′-Polymethylene bridging of the MEA structure provides compounds (**5**) that are active

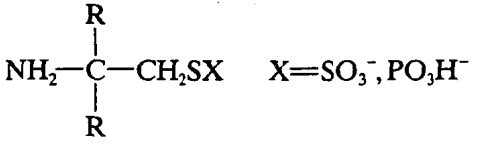
$$XS(CH_2)_2NH(CH_2)_nNH(CH_2)_2SX$$

(5)

where X is PO_3H_2 and n is 3 or 4, but inactive where X is SO_3H (50).

Alkylation of the carbon atoms of the MEA structure has given varied results. Active compounds have been found among C-monoalkyl derivatives, 2-aminopropan-1-

thiol having moderate activity and 1-aminopropan-2-thiol having good activity (51, 52). Whereas α, α-dialkyl-β-aminoethanethiols are inactive (53, 54), some β,β-dialkyl-β-aminoethane thiosulfates and phosphorothioates (6) have substantial activity



(6)

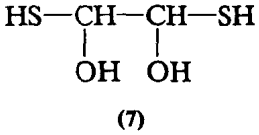
(55). 2-Amino-1-pentanethiol and 2-amino-3-methyl-1-butanethiol also have good activity (55). C-Trialkyl derivatives of MEA (54), *sec*–mercaptoalkylamines (56), and 2-mercapto-2-phenethylamine (57) are inactive. Generally, the presence of a phenyl group in the MEA structure blocks activity (58). α,α-Dimethyl-2-aminoethanethiol, derived from penicillamine, is not protective but has radiosensitizing activity (59).

Alkylation or arylation of the mercapto group generally results in loss of activity. The *S*-benzyl derivative of MEA has some activity, probably resulting from *in vivo* debenzylation (60).

Attempts to determine whether the stereochemical structure of the aminoalkanethiols is important have revealed that a given stereoisomer may provide greater radioprotection than others. A small difference in activity was found for the cis and trans isomers of 2-aminocyclohexane-1-thiol (61). The cis forms of 2-mercaptocyclobutylamine and 2-mercaptocyclobutyl-*N*-methylamine have higher radioprotective activities in mice than the trans forms. No correlation could be found between protective activity and ability to protect against either induction of DNA single-strand breaks or inactivation of proliferative capacity of hamster cells in vitro (62), however. On the other hand, the trans forms were less toxic and somewhat more effective in competing for free radicals in DNA. The D and L isomers of 2-aminobutylisothiuronium bromide have been separated, and the D isomer was twice as active in mice as the L isomer (63). The optical isomers of dithiothreitol show a greater difference in protective ability, the Dg isomer protecting 50% of mice exposed to 650 rads, whereas the Lg isomer afforded no protection (64). The Dg isomer was also less toxic.

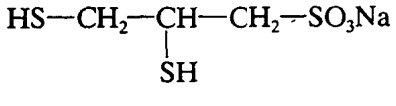
Other functional groups in the MEA structure have generally caused diminution or loss of protective ability. The presence of a carboxyl group frequently causes lower activity; cysteine, for instance, has the same dose reduction factor (1.7) in mice as MEA or MEG, but a much larger dose is required (65). This may be explained by the charge, or Z value, of the RSH molecule, which determines the concentration of thiol in the immediate vicinity of DNA, which has been shown to be in agreement with scavenging and chemical repair reactions (66). The negative charge of a carboxyl group would thus be repelled by negative charges on DNA and prevent close accumulation of the thiol, necessary for DNA protection and repair.

N-Monosubstituted derivatives of MEA containing thioureide or sulfone substituents are inactive, although sulfonic acid zwitterions, $HS(CH_2)_2NH_2^+(CH_2)_3SO_3^-$, are strongly protective (67). The presence of hydroxyl often favors activity; e.g., L(+)-3-amino-4-mercapto-1-butanol gives good protection to mice (68). An additional thiol group diminished activity in a series of 2-alkyl-2-amino-1,3-propanedithiols, which showed little activity in mice (67). Dithiothreitol (Cleland's reagent) (7) has protective ability (64), and Carmack et al. (64) found no protection from the oxidized dithiane form. Its protective activity *in vivo* may be due to release of other nonprotein thiols from mixed disulfides. Also, this may reflect



(7)

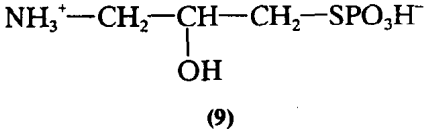
a requirement for a suitable redox potential. Dithiothreitol is so readily reduced that it quickly becomes oxidized in biological systems. It is a good protector in vitro, under conditions where it remains reduced. *N*-Carbamoyl ethyl derivatives of the phosphorothioate of MEA, however, had high protective activity (69). Sodium 2,3-dimercaptopropane sulfonate (Unithiol) (**8**),



(**8**)

which was studied in Russia, was claimed to be more protective and less toxic than MEA (70).

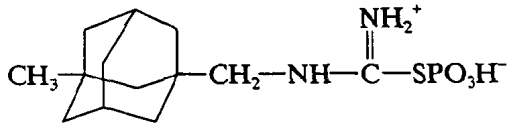
S-Acylation of the MEA structure has provided some very active compounds, particularly where zwitterions have resulted. The thiosulfate (Bunte salt) (45), phosphorothioate (46), and trithiocarbonate (47) of MEA, all of which form zwitterions, have protective activities comparable to that of MEA. Corresponding zwitterions of mercaptoethylguanidine (MEG) also give protection to mice corresponding to that of MEG (47, 71). Of these *S*-acyl derivatives, the phosphorothioates have been particularly effective; *S*-(3-amino-2-hydroxypropyl)phosphorothioate (**9**) and *S*-(2-aminopropyl)-



(**9**)

phosphorothioate have DRF values in mice of 2.16 and 1.86, respectively, in comparison to a DRF value of 1.84 for MEA (72). *S*-[2-(3-Aminopropylamino) ethyl]-phosphorothioate (**4**) (known as WR2721, from the screening program at the Walter Reed Army Institute of Research) (73) has high antiradiation activity and has been studied in numerous investigations. 3-Aminopropylphosphorothioate (71), however, and *N*-substituted derivatives of 2-aminoethylphosphorothioate are essentially inactive (71). Numerous publications concerning the synthesis and screening activities of the amino thiol derivatives submitted to the Walter Reed Army Institute of Research, leading to the selection of WR2721 as the most effective compound resulting from this screening program, have not been included here.

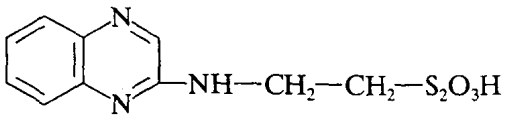
A comparison of the relative activities and toxicities of thiols with the corresponding thiosulfates showed the thiosulfates to be less toxic and comparable in activities (52, 74). In a series of 2-*N*-alkylaminoethanethiols, comprising 66 compounds, the thiosulfates were generally superior to either the corresponding thiols, disulfides, or thiazolidines (74), given intraperitoneally (ip) to mice. Another comparison of the relative effectiveness of thiols with the common mercapto-covering groups, the disulfide, thiosulfate, and phosphorothioate, was made with a series of 84 2-mercaptoacetamidine derivatives (75). Although generalities were not evident, by the ip route, the (3,5-dimethyl-1-adamantyl) methyl phosphorothioate (**10**) was



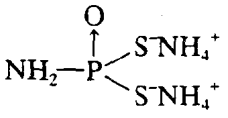
(**10**)

the most effective compound. Perorally, the disulfides appeared to be superior, the most effective compound being the 1-adamantylmethyl disulfide. In a series of *N*-heterocyclic aminoethyl disulfides and aminoethiosulfuric acids, the thiosulfates were generally more active and less toxic than the disulfides, administered either ip or perorally (76). The most effective compound of this series was 2-(2-quinoxalinylamino) ethanethiosulfate (**11**). It is believed that the phosphorothioate group aids in cellular transport (77).

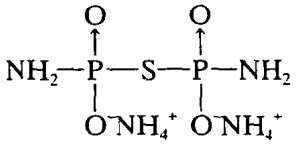
Two inorganic phosphorothioates, diammonium amidophosphorothioate (**12**) and diammonium thioamidodiphosphate (**13**)



(11)



(12)

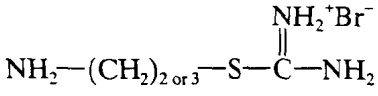


(13)

gave DRF values, respectively, of 2.30 and 2.16 at relatively low doses (72). Alkylation of the amidophosphorothioate lowered or eliminated activity, however (78).

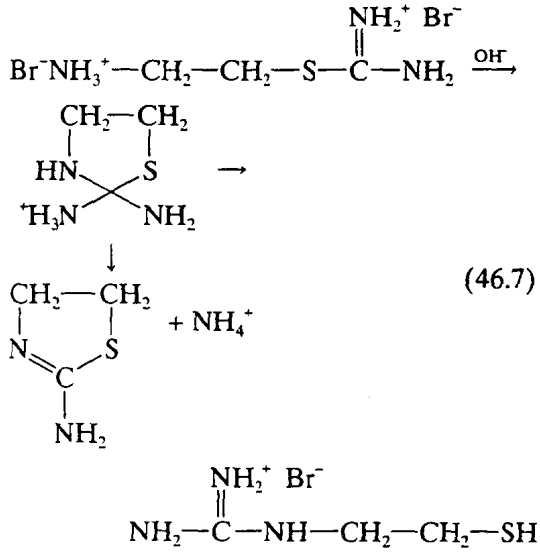
In a series of straight chain aliphatic thioesters of MEA, the best protection was found with the acetyl and octanoyl derivatives (79); the benzoyl ester was essentially inactive. *N*-Acetyl and *N*,*S*-diacetyl MEA showed minimal activity (80). In a series of hemimercaptals of MEA derived from glycolic acid, the most active protected mice at one-half the LD_{50} dose, with activity comparable to that of MEA (81).

Other basic functional groups can replace the amino group in the MEA structure to provide protective thiols. The inclusion of the guanidino group has provided very active compounds, notably 2-mercaptoethylguanidine (MEG) (**2**) and 2-mercaptopropylguanidine (MPG) (80). Solutions of these compounds were obtained by alkaline rearrangement of the aminoalkylisothiuronium (AET or APT) (**14**) salts. When these



(14)

compounds are employed for radiation protection tests, the hydrobromides of the aminoalkylisothiuronium bromides are rearranged in neutral or alkaline media. This rearrangement has been termed "intratransguanylation". Thus, AET or APT gives solutions of MEG or MPG (eq. 46.7). These compounds are usually not isolated by this procedure, but they may be isolated as the sulfates (82) or the trithiocarbonate esters (47).



(46.7)

Although AET is not subject to air oxidation, as are most thiols, it is affected by moisture, resulting in conversion to 2-amino-2-thiazoline. The disulfide, bis(2-guanidinoethyl) disulfide (GED), is readily prepared, however, and is relatively stable. With more than three carbon atoms between the amino and isothiuronium functions, rearrangement does not readily occur, and the isothiuronium salts give little protection. 2-Aminobutylthiopseudourea dihydrobromide, however, requires about one-fourth the molar quantity of AET for comparable protection in mice (63).

Replacement of the amino group by amidino has also resulted in compounds with good protective activity, particularly with Bunte salts of α-mercaptoacetamidines (**15**)