D-PENICILLAMINE IN RHEUMATOID ARTHRITIS

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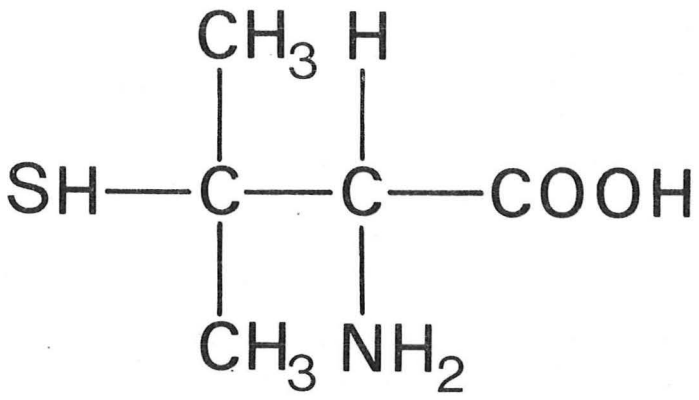


Penicillamine

"The treatment of rheumatoid disease is, for most patients, palliatory, and for all it is empirical. Those few drugs which seem to suppress the disease have limitations, so that any new preparation which promises to induce a remission at less hazard is worthy of close study."

Multicentre Trial Group, 1973

INTRODUCTION

In the fall of 1978 the FDA released D-penicillamine for use in treating severe rheumatoid arthritis (RA). This occurred thirty-five years after the compound was first discovered (1) and eighteen years after the first preliminary reports of its potential usefulness in RA appeared (2,3). It would seem reasonable at this time, therefore, to examine some of the characteristics of this drug and to review the clinical experience with the use of D-penicillamine in RA. At the outset, it should be understood that the basis for the effectiveness of D-penicillamine therapy in RA is unknown. However, since therapy with the drug appears to be able to induce remissions in disease activity in some patients, an attempt to understand its mechanism of action in RA may well provide some insight into the pathogenesis of this disease.

D-penicillamine is one of a group of drugs that share the characteristic of being effective in the treatment of RA (4). These drugs have been variously referred to as "specific anti-rheumatoid agents", "remission-inducing drugs", or "slow-acting anti-inflammatory drugs". Each of these terms connotes a feature of the action of these drugs in RA but none is entirely accurate. The concept that these drugs are specific anti-rheumatoid agents derives from the observation that they are active in treating RA but have little non-specific anti-inflammatory or analgesic action. It is unclear, however, that these drugs exert a unique action on rheumatoid inflammation. Although their usefulness in treating RA has been demonstrated, most of them have also been shown to be somewhat effective in other rheumatologic conditions. Thus, for example, gold and immunosuppressive agents have been shown to be effective in psoriatic arthritis (5-8). In addition, the activity of many of these agents in other conditions has not been adequately examined to conclude that their activity is specifically directed against the rheumatoid process.

The onset of clinically apparent suppression of disease activity is usually not seen until weeks or even months after the initiation of therapy and, hence, they are referred to as "slow-acting" agents. The question of whether these drugs induce actual remissions in disease activity is unclear. While therapy with these drugs can lead to significant suppression of the signs and symptoms of activity and can probably retard the progress of the disease, withdrawal of the drug is often associated with recrudescence of disease activity (9).

Despite these caveats, it is useful to think of these drugs as a group because of the similarity of their effects in patients with RA. The first drugs of this group that were recognized

were the gold compounds which have been used clinically for the past fifty years (10-14) and are the standards by which the other drugs are judged. Newer drugs include the immunosuppressive agents (15-20), D-penicillamine and perhaps levamisole (21,22).

Characteristics of therapy with these drugs include a delayed onset of clinical effect. The gradual suppression of the signs and symptoms of RA may not be apparent until weeks or months after the initiation of therapy and can take 4 to 6 months or longer to reach a maximum. During this time, there is often a slow reduction in clinical signs of disease activity with decreased pain, swelling and stiffness. In addition, there may be increases in hemoglobin concentrations along with decreases in the erythrocyte sedimentation rate and titers of rheumatoid factor. This differs from the results of treatment with non-steroidal anti-inflammatory agents such as aspirin or indomethacin. Although these agents may suppress symptoms of RA more rapidly, there is usually no significant change in these serological correlates of disease activity, as indicated below (23).

Effect of Therapy with Aspirin and Indomethacin on Acute Phase Reactants in Rheumatoid Arthritis (Aylward et al, Rheum. Rehab. 14:101, 1975)

	Aspirin*		Indomethacin*	
	Pre-treatment	Post-treatment	Pre-treatment	Post-treatment
ESR (mm/hour)	49.4	48.4	51.1	47.0
CRP (μg/ml)	156.4	169.3	145.4	143.6
Haptoglobin (mg/100ml)	201.4	198.7	219.2	231.0

*Groups of 10 patients with classical or definite rheumatoid arthritis were treated with aspirin (4-5gm daily) or indomethacin (150mg/day) for 6-8 weeks.

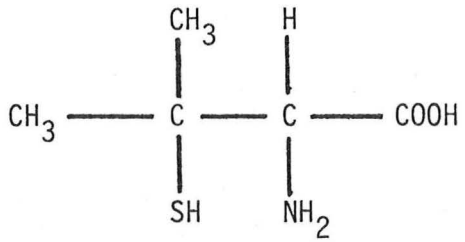
The second characterisitic of these drugs is the capacity to alter the course of the disease and retard its progress. Thus, therapy with these agents has been shown to have the potential to suppress the development of new erosive changes in peri-articular bones (14,17,19,24). Such bone erosions are accepted as an objective radiographic measure of the destructiveness of the underlying progressive inflammatory process. This is to be contrasted with the results of therapy with non-steroidal anti-inflammatory agents. While therapy with such non-specific agents may relieve pain and clinical signs of inflammation, it has not been shown to alter the erythrocyte sedimentation rate, titers of rheumatoid factor, the development of bone erosions, or the outcome of the disease.

The third characteristic of these drugs that limits their usefulness is the marked degree of toxicity associated with the use of each of them (25). The side-effects probably have a greater influence on the use of these drugs than their therapeutic efficacy. Because of the potential toxicity of these agents, they are not used for all patients. Many patients with RA receive adequate symptomatic relief with conventional non-steroidal anti-inflammatory drugs. A trial of these agents is usually warranted as first-line therapy, especially during the first months of the disease when spontaneous remissions may occur and the outcome of the disease is uncertain. After six months or so of such therapy, more definitive treatment is indicated if there is persistent disease activity. This is especially the case in patients whose disease is clearly progressive with developing deformities or destructive changes on radiographic evaluation.

It should be borne in mind that while these drugs have been shown to be effective in treating patients with RA, their use remains empirical. At the present time, there is no way to match patients with the appropriate therapeutic agent. Thus, the characteristics of individual patients that determine whether they will have a therapeutic response or develop toxicity to one of these drugs remain unknown. In addition, the specific indications for each of these agents are unclear and their use, therefore, remains arbitrary.

D-penicillamine: Historical Perspective

D-penicillamine is a structural analogue of the naturally occurring amino acid cysteine in which methyl groups replace the two hydrogen atoms in the beta carbon position. As an amino acid, it may occur as either the D or L isomer. Early studies utilized the racemic mixture D,L penicillamine, but more recently only the D isomer is used in clinical medicine.



β, β-dimethylcysteine
(β-mercaptovaline)

Penicillamine was first described by Abraham et al in 1943 as an acid hydrolysis product of penicillin (1). In 1953, Walshe was the first to identify penicillamine *in vivo* in the urine of individuals treated with penicillin and, because of its capacity to chelate copper, suggested that this compound might be used to promote copper

excretion in patients with Wilson's disease (26,27). In 1954, Tabachnick et al showed that D-penicillamine could take part in thiol-disulfide interchange reactions with cystine (28). The subsequent observation that the resultant mixed disulfide consisting of penicillamine and cysteine was much more soluble than cystine (cysteine disulfide) served as the basis for the use of this drug in cystinuria.

The observation that eventually led to the use of D-penicillamine in rheumatoid arthritis was made by Deutsch and Morton in 1957. They observed that sulfhydyl reducing agents could reduce the disulfide bonds of the IgM molecule and cause the dissociation of pentameric IgM into monomeric subunits (29). Rheumatoid factor (RF) found in a high percentage of patients with severe rheumatoid arthritis (30,31) is an IgM antibody directed against human immunoglobulin G. It had been rediscovered in 1948 by Rose et al (32) and was thought at that time to be involved in the pathogenesis of rheumatoid arthritis. In 1958, Heimer and Federico (33) showed that IgM RF could be dissociated with sulfhydryl reducing compounds to 7S subunits with a consequent loss of its capacity to agglutinate IgG coated particles. In 1960, Ritzmann et al (34) demonstrated that penicillamine, like other thiols, could dissociate macroglobulins and, based upon this observation, used penicillamine to treat a patient with IgM cold agglutinin-mediated hemolytic anemia (35). The resultant decrease in antibody titer was attributed to the capacity of penicillamine to dissociate IgM antibody *in vivo*. In the same year, Griffin et al (2) and Dresner and Trombly (3) showed that penicillamine could dissociate IgM RF *in vitro* and presented preliminary data that administration of this drug to patients could lead to decreases in the titer of circulating rheumatoid factor and improvement in symptoms.

Based upon this work Jaffe, in 1962, attempted to confirm that penicillamine could dissociate IgM RF *in vivo*. Intra-articular administration of penicillamine was found to decrease synovial fluid RF titers (36). Subsequently, Jaffe was able to confirm that systemic administration of penicillamine led to a fall in serum RF titers (37). In 1964, Jaffe reported that penicillamine therapy not only led to a fall in RF titers, but also to amelioration of symptoms in a patient with RA and vasculitis (38). It was not until 1973, nine years later, however, that a controlled trial confirmed that penicillamine was effective in the treatment of rheumatoid arthritis (39).

Clinical Uses of D-Penicillamine

I. FDA Approved Uses:

a) Wilson's disease
b) Cystinuria
c) Rheumatoid arthritis

II. Other Uses:

a) Heavy metal intoxication - lead, arsenic, mercury
b) Scleroderma (54-62)
c) Avian muscular dystrophy (63)
d) Primary biliary cirrhosis (64-66)
e) Juvenile rheumatoid arthritis (67,68)
f) Chronic active hepatitis (69,70)

III. Reports of Successful Use:

a) Polymyositis (71)
b) Palindromic rheumatism (72)
c) Macroglobulinemia (73)
d) IgM cold agglutinin-mediated hemolytic anemia (35)
e) Neonatal ABO incompatibility (74)
f) Neonatal hyperbilirubinemia (75)
g) Schistosomiasis (adjuvant to antimonial therapy) (76,77)
h) Paracetamol overdose (78)
i) Porphyria cutanea tarda (79)
j) Tracheal stenosis (80)
k) Aldehyde intoxication (81)

IV. Questionable Usefulness:

a) Essential cryoglobulinemia (82)
b) Ankylosing spondylitis (71,83-85)
c) Schizophrenia (86-88)
d) Amyloidosis (89,90)
e) Alcoholic liver disease (91)
f) Gold toxicity (92-95)

V. Shown to be of No Value:

a) Multiple sclerosis (96)

D-Penicillamine: Current Clinical Usage

Currently, D-penicillamine has been approved for use in three clinical conditions. It is the mainstay of the treatment of Wilson's disease (27,40-43) and cystinuria (44-47) and appears to have a role in the treatment of rheumatoid arthritis. It has also been widely and effectively used to treat intoxication with a number of heavy metals including lead, mercury and arsenic (48-53), but has not been approved by the FDA for this purpose.

In addition, preliminary clinical trials have indicated that it may be effective in a number of other conditions such as scleroderma, primary biliary cirrhosis, chronic active hepatitis and juvenile rheumatoid arthritis and controlled trials are currently in progress to evaluate its usefulness in these conditions.

As indicated in the above Table, D-penicillamine has also been employed in a number of other clinical situations with varying degrees of success. The suspected utility of D-penicillamine in most of these clinical situations is based upon its known biochemical properties.

D-penicillamine: Biochemical Properties

Penicillamine is a trifunctional amino acid in which a carboxyl group and an amino group are attached to one carbon atom and a sulfhydryl and two methyl groups to a second carbon atom. As an amino acid, penicillamine can exist as either the D or L optical isomer. Early studies utilized the racemic mixture, D,L penicillamine, but more recently only the D isomer has been used because of its decreased toxicity (97). The three functional groups in penicillamine undergo characteristic chemical reactions including acid-base equilibria, nucleophilic addition and displacement, combination with various metals, oxidation and free radical transformations (98). The biochemical properties of D-penicillamine that are most important toward understanding its action in these various clinical situations are: 1) the capacity to form stable chelates with various metals; 2) the ability to participate in thiol-disulfide interchange reactions, and 3) the capacity to react with various carbonyl compounds to form stable thiazolidines.

D-penicillamine can form stable complexes with various metals and thus promote their excretion from the body. This is the basis of its use in Wilson's disease (27,40-43), heavy metal intoxication (48-53), and the suggestion that it might be useful in treating the manifestations of gold toxicity in patients with rheumatoid arthritis undergoing chrysotherapy (92,93,95). The use of D-penicillamine in primary biliary cirrhosis was stimulated by the finding that patients with this disease had increased concentrations of hepatic copper and the belief that deposition of free copper in liver tissue might incite an inflammatory response (99,100). The capacity of penicillamine to chelate antimonials has been suggested as a means to decrease the toxicity of these compounds without inhibiting their

therapeutic effect in Schistosomiasis (76,77). The capacity of D-penicillamine to inhibit copper-containing enzymes involved in catecholamine synthesis has been given as a rationale for its use in treating schizophrenia (86).

Biochemical Rationale for the Use of Penicillamine

I. Chelation:

1) Wilson's disease
2) Heavy metal intoxication
3) Primary biliary cirrhosis
4) Gold intoxication
5) Schistosomiasis
6) Schizophrenia

II. Thiol-disulfide interchange reactions:

1) Cystinuria
2) IgM cold agglutinin-mediated hemolytic anemia

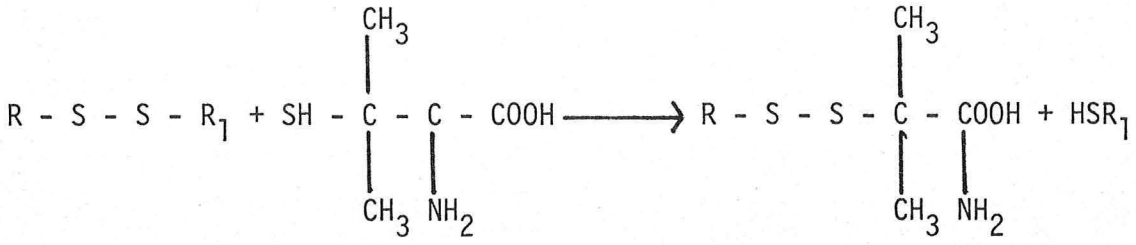
III. Thiazolidine formation:

1) Scleroderma
2) Cirrhosis
3) Aldehyde intoxication

IV. Unknown:

1) Rheumatoid arthritis

D-penicillamine can also take part in thiol-disulfide interchange reactions, as shown here:

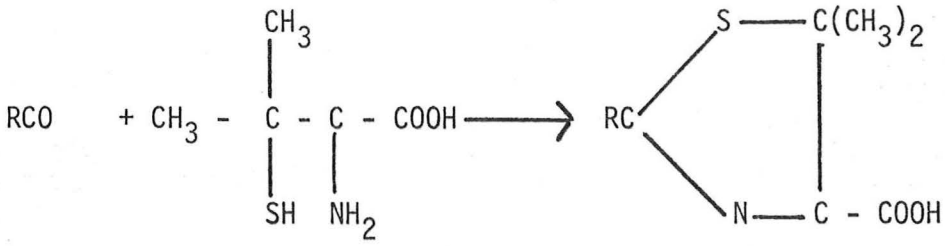


Disulfide + Penicillamine ⟶ Penicillamine mixed disulfide + free thiol

This capacity is the basis of the use of D-penicillamine in cystinuria since the mixed disulfide, penicillamine-cysteine is considerably more

soluble than cysteine disulfide (cystine). Thus, the administration of D-penicillamine-cystine to patients with cystinuria has been shown to prevent the formation of renal stones and also to dissolve pre-existing calculi (44-47). The capacity of penicillamine to take part in thiol-disulfide interchange reactions with IgM molecules leading to dissociation of pentameric IgM into its monomeric subunits was the rationale for its usage in macroglobulinemia (73) and IgM cold agglutinin-mediated hemolytic anemia (35). In addition, the impetus for its original use in rheumatoid arthritis came from the belief that penicillamine could inhibit rheumatoid factor activity by dissociating IgM molecules *in vivo* (2,3,36-38). It should be pointed out that this is an unlikely explanation for the effectiveness of penicillamine in rheumatoid arthritis since the concentrations of penicillamine achieved *in vivo* are more than 200 fold less than needed to achieve this biochemical effect on macroglobulins (2,101).

Finally, penicillamine can react with various carbonyl groups to form stable thiazolidines as shown here.



aldehyde + penicillamine ⟶ thiazolidine

D-penicillamine is thought to inhibit collagen cross-linking by forming thiazolidine rings with the lysyl-derived aldehydes that are intermediates in cross-link biosynthesis. By trapping allysyl residues in thiazolidine rings, D-penicillamine makes them unavailable for cross-link formation (102). Thus, the chronic administration of penicillamine to experimental animals or humans leads to the development of abnormalities of collagen similar to those found in lathyrism (103-105). This action of D-penicillamine on collagen cross-linking is the rationale for its use in scleroderma, tracheal stenosis, chronic active hepatitis, primary biliary cirrhosis and alcoholic liver disease.

Penicillamine can also form a thiazolidine with pyridoxal phosphate. This may explain the anti-vitamin B_6 effect associated with the use of this drug (106). The effect of penicillamine on pyridoxine metabolism *in vivo* is more pronounced with the L enantiomorph but may also be seen with D-penicillamine although it is rarely of clinical importance.

Finally, the capacity of penicillamine to form thiazolidines may have additional practical significance. Penicillamine can interact with acetaldehyde and thus, counter some of the manifestations of ethanol intoxication (81). In addition, it may act as a protective agent against overdoses of other more toxic aldehydes such as chloral hydrate, paraldehyde and formaldehyde derived from methanol intoxication (98).

D-penicillamine: Pharmacology:

After oral administration of D-penicillamine, about 40% of the total dose is absorbed (107). It is rapidly absorbed from the gastrointestinal tract reaching a concentration of 2-5% of the administered dose per liter of plasma within two hours of ingestion (108). Of this, about 25% can be detected in the reduced form as the free thiol, while the remainder is found as either the autologous or a mixed disulfide (101). Penicillamine appears to be distributed throughout the total body water (108). For several hours after administration of the drug, penicillamine is cleared from the plasma at a rate approximating the creatinine clearance (108). The initial plasma half life of the drug is about six hours (101). As protein binding occurs, the rate of excretion declines and little penicillamine appears in the urine after 24 hours. Approximately 80% of the plasma penicillamine is protein bound at this time (108). The plasma half life of the drug at this period is about eight days (101). Of the D-penicillamine appearing in the urine, about 90% is found as either penicillamine disulfide or cysteine-penicillamine disulfide, 8% as a methylated metabolite, S-methyl-D-penicillamine, and the remainder as free D-penicillamine thiol (107). Patients with toxic reactions to penicillamine do not appear to handle the drug differently than individuals without side effects (108).

D-penicillamine has a number of unique pharmacological properties resulting from its molecular structure. First,autooxidation to the internal disulfide is considerably slower than for other thiols (109). While the majority of penicillamine has been reported to be excreted as either a mixed or internal disulfide (107), it has been suggested that because of the available mechanisms for reduction of disulfides *in vivo*, it is likely that it is maintained in the body either as the free thiol or as a mixed disulfide bound to various proteins (109).

Secondly, D-penicillamine is resistant to enzymatic degradation by either amino acid oxidases or cysteine desulfhydrase (110). Less that 10% of an administered dose is metabolized (107), and as a result, D-penicillamine has an extremely long biological half life. The conclusion that D-penicillamine can be maintained in the body in an active form for prolonged periods of time is supported by the observation that it can be found in the urine for as long as three months after administration of the drug has been discontinued (111).

D-penicillamine: Treatment of Rheumatoid Arthritis:

After Jaffe's initial report of success in treating a patient with rheumatoid arthritis and vasculitis (38), reports of three uncontrolled trials appeared in the literature. Zuckner et al (112) treated 15 patients for 12-18 months with 1-2gm of D-penicillamine per day. Of the 15 patients, 10 improved clinically; eight were thought to have achieved more than a 50% improvement in symptoms and two others achieved some improvement, such that their maintenance dosages of non-specific anti-inflammatory drugs could be decreased. Onset of clinical improvement was first seen after six weeks of therapy and in one patient was not seen for six months after the initiation of therapy. A gradual decrease in the erythrocyte sedimentation rate was seen in nine patients. The titer of rheumatoid factor decreased in all treated patients, including those who showed no clinical improvement. Agranulocytosis developed in one patient and more minor side effects were seen in a number of others. Jaffe (113) also reported on his experience treating 49 patients with penicillamine. Over an eight year period, these patients received 60 twelve month courses of penicillamine. He reported that 80% of these courses of treatment resulted in a 50% or greater degree of improvement as evidenced by a reduction in subjective and objective signs of active disease, a sustained fall of 50% or more in ESR and a significant drop in rheumatoid factor titers. He also noted that subjective improvement usually began after 3-4 months of therapy and that objective improvement, such as decreased synovial swelling, joint tenderness or size of nodules, was usually not seen for six months. Toxic reactions to penicillamine were seen in about 25% of the patients. Finally, Huskisson and Hart (9) reported the results of penicillamine therapy (1,800mg/day) in 10 patients with rheumatoid arthritis who had failed to respond to conventional therapy including "rest in hospital" and gold. Seven of these patients responded favorably with decreases in joint inflammation, erythrocyte sedimentation rate and rheumatoid factor titers. Nine of ten developed side effects, and four developed proteinuria necessitating cessation of therapy.

The first controlled trial of D-penicillamine in rheumatoid arthritis was carried out in England by the Multicentre Trial Group and reported in 1973 (39). This attempted to determine in a controlled study whether D-penicillamine therapy would have a favorable influence on the course of rheumatoid arthritis. The study involved 105 patients with severe disease that had not responded to orthodox treatment. The study was double blind in design with controls and lasted for 12 months. Assignment of patients to therapy or placebo groups was done after stratification of the patients by age, sex and use of corticosteroids. The criteria for admission to the study were quite strict, as shown here, and ensured that only patients with severe RA would be included.

Criteria for Admission to
Multicentre Trial

1) ARA criteria for definite or classical RA.

2) Disease duration > 2 years.

3) Failure to respond to current therapy.

4) At least 4 of the following signs of active disease:

   a. ESR > 50mm/hr (Westergren).
   b. Hemoglobin < 11.7gm%.
   c. Weight loss > 2.7Kg in 6 months.
   d. > 2 large weight bearing joints involved.
   e. Neuropathy or vasculitis.
   f. Morning stiffness > 1 hour.
   g. Articular index > 50.
   h. Early fatiguing.

Excluded from the trial were patients who had had gold therapy within six months, immunosuppressive or anti-malarial therapy within two months, or any major change in therapy within one month. In addition, patients with renal dysfunction or a record of poor clinic attendance were excluded.

The patients received a maintenance dose of 1.5 grams of D-penicillamine per day and continued their ongoing regimen of anti-inflammatory medication. A number of subjective and objective clinical signs of disease activity, as well as laboratory correlates, were evaluated at 3, 6 and 12 months after the start of therapy.

After six months of therapy 41 of 52 patients were left in the penicillamine group and 45 of 53 in the control group. Each patient assessed his subjective judgment of the value of therapy at this time. A significantly greater number of individuals in the penicillamine group felt they had received successful therapy.

Participants' Assessment of Treatment
-Multicentre Trial Group-

Group	Opinion of Treatment Successful	Doubtful	No Value	
Penicillamine (N=41)	26	8	5	P<0.01
Control (N=45)	3	14	26	

After 12 months of therapy, 30 out of 52 patients remained in the penicillamine group and 38 of 53 in the control group. The reasons for failure to complete the year trial are tabulated below. It is worth noting that failure to complete the 12 month trial was usually related to adverse reactions in the penicillamine group, while failures in the control group usually related to worsening symptoms.

Reasons for Failure to Complete 12 Month
D-penicillamine Trial
-Multicentre Trial Group-

Cause	Penicillamine N=52	Control N=53
Adverse reactions	16	1
Therapeutic Failure	0	9
Extraneous	6	5
Total	22	15

The adverse side effects that caused withdrawal from the study included rash, thrombocytopenia, proteinuria, loss of taste, anorexia, nausea and vomiting. Gastrointestinal disturbance in one patient of the control group also led to withdrawal.

At the termination of the 12 month trial, the patients in each group were analyzed. The characteristics of the patients in each group at the outset of the study were comparable, as shown below:

Characteristics at the Outset of the Trial
of Patients Completing 12 Months
-Multicentre Trial Group-

	Penicillamine n=30	Control n=38
Age	55	57.9
Sex	8M, 22F	6M, 32F
Duration of Disease (yr)	11.1	11.2
Steroids	20	24
Weight (Kg)	64.5	64.1
ESR (mm/hr)	59	56
Hb (g/100ml)	12.0	12.4
Articular Index	57.1	56.8

The results of the trial indicated that by most methods of assessing the patients, the group receiving D-penicillamine significantly improved compared to the group receiving placebo.

Results of 12 Month Trial of D-penicillamine
in Rheumatoid Arthritis
-Multicentre Trial Group-

Measurement	Group	Time of Assessment 0 Months	12 Months	p
Pain (0-3)	Pen	2.11	1.14 (54%)	<0.005
	Control	2.24	1.76 (79%)	
Morning Stiffness (hr)	Pen	1.70	0.30 (18%)	<0.02
	Control	2.28	1.59 (70%)	
Grip Strength, R (mm Hg)	Pen	1.07	135 (126%)	<0.01
	Control	1.14	116 (102%)	
Articular Index (0-182)	Pen	57.1	26.3 (40%)	<0.01
	Control	56.8	40.0 (70%)	
Functional Assessment (0-192)	Pen	76.0	53.7 (71%)	<0.01
	Control	76.3	73.1 (96%)	
ESR (mm/hour)	Pen	59	39 (66%)	<0.01
	Control	56	53 (94%)	
Hemoglobin (g%)	Pen	12.0	13.4 (112%)	<0.01
	Control	12.4	12.7 (102%)	
Well-being (0-3)	Pen	1.04	0.61 (59%)	N.S.
	Control	1.26	0.89 (71%)	

Terms in parentheses indicate the percent of the initial assessment.

The differences between the control group and the drug group were probably greater than is apparent from these data because of the loss from the control group of patients whose arthritis became worse. None of the penicillamine group dropped out because of worsening symptoms whereas 17% of the control group withdrew for this reason.

Penicillamine therapy also had a significant effect on rheumatoid factor titers. After six months of therapy 48% of the penicillamine group showed decreased RF titers while this was only seen in 11% of the control group. No significant differences in RF titers were seen after three months of the trial.

Effect of D-penicillamine Therapy
on Rheumatoid Factor Titers

Length of Treatment	Patient Group	Percent of Patients* with Improvement in RF titer	p
3 months	Pen	8	N.S.
	Control	6	
6 months	Pen	48	<0.01
	Control	11	

*Reduction of greater than three twofold dilutions in sensitized sheep cell agglutination titer.

Radiographic evaluation of the joints of the hands and feet did not show any change in either group during the twelve months of the trial. However, all of the patients had severe erosive changes and twelve months of treatment may not have been long enough to reveal statistically significant changes.

Penicillamine therapy appeared to have an effect on extra-articular manifestations of rheumatoid arthritis as indicated below, but when compared to the control group this did not achieve statistical significance. Of note, however, was the observation that no patient taking D-penicillamine developed new vasculitic or neuropathic lesions during the trial.

Effect of D-penicillamine on Extra-articular Manifestations of RA
-Multicentre Trial Group-

Extra-articular Manifestation	D-penicillamine 0 Months	D-penicillamine 12 Months	Control 0 Months	Control 12 Months
	(Number of Patients with Each Finding)			
Nodules	11	9	17	15
Tenosynovitis	10	3	12	11
Ganglion	14	7	22	18
Vasculitis	5	2	0	1
Neuropathy	10	5	2	3
Episcleritis	4	3	2	1
Skin ulcer	5	2	1	2
Lymphadenopathy	4	1	7	3

During the initial 3 months of therapy, 63% of the D-penicillamine group and 34% of the control group experienced adverse reactions. After 12 months of therapy, 29% of the control group continued to experience side effects, whereas 35% of the D-penicillamine group experienced side effects even though 16 had dropped out because of adverse reactions. The side effects encountered during the trial are listed below. The addition of copper sulfate to the therapeutic program had no effect on the incidence of side effects.

Frequency of Side Effects
-Multicentre Trial Group-

	Penicillamine n=52	Control n=53
	(Number)	
I. Penicillamine only:		
Proteinuria	4	0
Thrombocytopenia	11	0
Miscellaneous	3	0
II. More Common with Penicillamine:		
Impaired taste	17	9
Bruising	4	2
Nausea	6	2
III. Both Groups:		
Rash	7	6
Oral ulcers	5	5
Gastrointestinal upset	10	10
Miscellaneous	5	6
IV. Controls only:		
Miscellaneous complaints	0	6

This study clearly indicated that D-penicillamine was effective therapy in severe rheumatoid arthritis. Using standard and widely accepted criteria of disease activity, improvement was noted in articular index, grip strength, functional assessment, ESR, hemoglobin and rheumatoid factor titer. Signs of improvement were usually evident after three months of D-penicillamine therapy. Of note was the observation that none of the patients who had taken D-penicillamine for more than six months left the trial with arthritis worse than it had been at the start. These findings were even more striking since the trial included only patients with far advanced disease who had not responded to orthodox therapy and thus was weighted against seeing a favorable response to D-penicillamine. There was no evidence, however, that D-penicillamine could prevent the development of bone erosions or facilitate their healing once present. In addition to the demonstration of therapeutic efficacy, this study confirmed that administration of D-penicillamine was associated with a high incidence of toxic reactions, some of which were life-threatening.

The conclusions of the Multicentre Trial Group have been confirmed by a large number of subsequent clinical trials. As indicated below, 22 open trials have been published through 1977 (24,112-132). In all, the trials involved 1,343 patients and lasted anywhere from 1-50 months, with a median of about 6-12 months. Although the information is incomplete in some series and difficult to interpret in others, improvement was seen in about 2/3 of the patients. Side effects were reported in about 1/3.

Results from a total of six controlled double-blind trials of D-penicillamine in rheumatoid arthritis have also been published (39,133-137). These were standard double blind trials lasting from 4 to 12 months with a median duration of six months. In the three trials where information can be extracted, 98 out of 153 patients, or 64%, showed improvement. Serious side effects were seen in a mean of 24%.

Finally, a number of ongoing trials of D-penicillamine therapy in rheumatoid arthritis were reported at the International Congress of Rheumatology in June 1977. Nineteen studies involving 1,600 patients were reported. The studies used a number of different dosage schedules, lasted for various periods of time, and included both controlled and uncontrolled trials. The results indicated that therapy with D-penicillamine led to improvement in a mean of 75% of treated patients with initial improvement seen after 2-4 months of treatment. Side effects were seen in 10-65% of treated patients (mean 43%). Side effects necessitated withdrawal from the study in 27% and there was one death from thrombocytopenia.

Clinical Trials of D-Penicillamine in Rheumatoid Arthritis

a) 22 uncontrolled trials (24,112-132)
number of patients - 1,343
duration of therapy - 1-50 months
improvement - 739/1190 (62%)
side effects - 360/981 (37%)

b) 6 controlled trials (39,133-137)
number of patients - 317
duration of therapy - 4-12 months
improvement - 64%
side effects - 24%

c) Ongoing trials (Internat'l. Congress of Rheumatology, San Francisco, Ca., June, 1977)

number of studies - 19
number of patients - 1,600
duration of therapy - 4-24 months
penicillamine dose - 300-1,500 mg/day
improvement - 75% (68-85%)
initial improvement - 2-4 months
side effects - 43% (10-65%)
withdrawals - 27% (5-39%)
deaths - 1

The Multicentre Trial Group found that therapy with D-penicillamine had no effect on the radiographic appearance of the joints of the hands and the feet. However, the patients studied had such advanced destructive changes that demonstration of improvement may not have been possible. In another blind trial, penicillamine therapy was also found to have little effect on the radiographic appearance of the joints (133). However, this trial was of short duration (24 weeks) and involved patients with advanced disease. One trial has suggested that prolonged therapy with D-penicillamine may suppress the development of new bone erosions and in a few instances may facilitate healing of such bone defects (24). As indicated in the table below, the data from this study also suggested that therapy with D-penicillamine may be better than gold therapy in preventing new erosions, although another trial from Scandinavia has found no difference between gold and D-penicillamine in this regard (138).

Effect of Treatment with Gold and D-Penicillamine on Bone Erosions in Rheumatoid Arthritis (Gibson et al, Rheum. Rehab. 15:211, 1976)

Treatment	Number of Patients	Bone Erosions* 0 Years	2 Years	Change	p
		(mean±SD)			
D-penicillamine	24	17.7±18.1	18.2±18.1	0.5±5.0	N.S.
Gold	20	34.5±30.5	42.4±31	7.7±12.4	<0.02

*The total number of bone erosions on each articular surface of both hands and both feet detected radiographically.

Finally, data from one of the double-blind trials has provided some support for the point of view that therapy with D-penicillamine may suppress the development of new bone erosions (130). In this study, x-rays of the hands and fingers were evaluated before and after a 24 week trial of D-penicillamine at therapeutic or homeopathic doses. The x-rays were examined in a blinded manner by two groups of observers; one group consisted of three orthopedists and the second of two internists. The orthopedists concluded that drug therapy had not affected osteoporosis, narrowing of the joint space or erosive changes. By contrast, the group of internists found that the number of new erosions developed per joint by the patients treated with D-penicillamine was significantly less ($p<0.05$) than that developed by the control group.

A number of trials have compared the therapeutic efficacy of D-penicillamine with that of other "specific" anti-rheumatoid agents. D-penicillamine and gold were found to have comparable therapeutic effects in three trials (138-150). In one trial involving 89 patients, the incidence of side effects in the D-penicillamine treated group was higher but the incidence of adverse reactions necessitating cessation of therapy was greater in the gold treated group (139). A second study involved 100 patients and also found the two drugs to be of comparable therapeutic efficacy (138). It was noted, however, that the incidence of side effects requiring discontinuation of therapy was also similar for the two drugs (26% for D-penicillamine and 30% for chrysotherapy). The effectiveness of D-penicillamine has also been compared to that of azathioprine in one trial involving 65 patients (134). After six months of therapy, D-penicillamine appeared to be more effective and less toxic, but by one year of therapy both drugs were comparable in efficacy and toxicity.

The optimal dosage regimen for the administration of D-penicillamine is not known. A number of clinical trials have addressed the question and the feeling has developed that a smaller daily dose may be effective and perhaps less toxic. In a double-blind controlled trial involving 121 patients, Dixon et al (133) compared D-penicillamine at daily doses of 600 or 1200 mg. After six months of therapy, the higher dose had not produced a greater therapeutic effect in the group of patients, although individual patients at times appeared to derive greater benefit from the higher dose. The frequency of rashes, blood dyscrasias and withdrawals from the trial were greater with the higher dose regimen. The overall impression that the lower dose regimen consisting of 500-600 mg of D-penicillamine per day is comparable in effectiveness to higher dose programs (1-2gm per day) is supported by four additional studies (114,126,135,141). At present, two ongoing double-blind studies are looking at even lower daily doses of D-penicillamine (150-250 mg/day) to determine whether these regimens might be equally effective but less toxic than presently used dosage schedules.

D-Penicillamine: Toxicity

Considerable attention has been given to the adverse side effects of D-penicillamine therapy. Most studies agree that somewhere between 30-65% of patients treated with D-penicillamine will experience adverse reactions. As many as 25 to 40% of treated patients may have to stop therapy because of the severity of the side effects. A compilation of the most frequently encountered side effects is given below. The most common adverse reactions are gastrointestinal. Aberrations of taste are found in about 12% of patients and usually occur in the first few months of therapy. Initially, it was felt that dysgeusia was secondary to depletion of zinc and/or copper associated with penicillamine therapy. However, administration of copper and/or zinc has not been found consistently to correct these abnormalities (39,135) and taste sensation usually returns to normal at the same rate whether or not penicillamine is discontinued. Dysgeusia may last for 3-4 months and, although annoying, rarely is disconcerting to the point where it necessitates cessation of therapy (126).

FREQUENCY OF SIDE EFFECTS IN PATIENTS TREATED WITH D-PENICILLAMINE

Side Effect	Frequency (%) A	B
Dysgeusia	13	12
G.I. distress, anorexia, nausea, vomiting,diarrhea	12	17
Rash	12	5
Proteinuria	9	6
Thrombocytopenia	7	4
Neutropenia	2	2
Oral ulceration	1	-

A - J. Baum, 1979, Scan. J. Rheum.; 23 clinical trials, 1,100 patients (142).

B - Merck Sharp & Dohme; 17 clinical trials, 1,270 patients.

A variety of other gastrointestinal side effects have been frequently reported, but these can usually be dealt with effectively without cessation of therapy.

Skin rashes occur in 5-12% of patients. The rashes that occur early during the course of therapy are usually morbilliform, maculopapular and pruritic, urticarial or eczematous. These usually can be adequately managed with local steroids and antihistamines and do not necessitate cessation of treatment. Skin reactions occurring later in therapy are usually more serious. These include pemphigus foliaceus, pemphigus vulgaris (143-150) and elastosis perforans serpiginosa (151) and require discontinuance of therapy.

Proteinuria is a more serious adverse side effect and may occur in 6 to 9% of treated patients. It usually develops late in the course of therapy (after 7 to 12 months) but may develop anytime as depicted below (126). Of interest, it appears that penicillamine induced nephropathy is seen more commonly in RA and cystinuria than in Wilson's disease (152). The reason for this observation is not clear.

Incidence of the Development of Proteinuria
H.F.H. Hill, Sem. Arth. Rheum. 6:361, 1977

Treatment Period (months)	Number of Patients	Proteinuria	Proteinuria >2gm/24hr
0-6	93	2 (2%)	1 (1%)
6-12	68	20 (30%)	17 (25%)
12-18	32	2 (6%)	1 (3%)
18-24	21	1 (5%)	1 (5%)

Proteinuria may become marked resulting in the development of the nephrotic syndrome. In one series, 4 out of 25 patients who developed D-penicillamine-induced proteinuria developed massive proteinuria exceeding 10gm/24hr and other evidence of the nephrotic syndrome (126). Clinical and laboratory abnormalities persisted for 12-24 months but recovery of renal function was eventually complete.

A number of studies have examined the histopathology of penicillamine-induced nephropathy. Renal biopsies usually have demonstrated subepithelial deposits in the glomerular basement membrane containing both IgG and C3, as indicated below:

RENAL BIOPSY FINDINGS IN PENICILLAMINE NEPHROPATHY

Patients (Number,Diagnosis)	Light Microscopy	Electron Microscopy	Immuno-fluorescence	Reference
3, cystinuria	Focal proliferative glomerulitis	Subepithelial deposits	N.D.	153
2,RA, scleroderma	Basement membrane deposits	Subepithelial deposits	IgG, C3	154
1,Wilson's disease	Minimal	N.D.	IgG, IgM, C3	155
4, RA	Minimal changes	Subepithelial deposits	IgG, C3	156
14, RA	Minimal changes	Subepithelial deposits	IgG, C3	152

Although findings consistent with immune complex deposition have been shown, there is no evidence of either circulating immune complexes in these patients or of peripheral complement consumption (152). Neither the identity of the putative antigen, nor the antigenic specificity of the immunoglobulin in the deposits has been defined.

This renal lesion tends to be reversible but proteinuria may persist for months, or even years, before it completely resolves. There is usually no residual renal dysfunction. Gärtner et al (157) described 31 patients who developed membranous nephritis a mean of seven months after beginning penicillamine therapy. In five of these patients, repeat biopsies were performed one year later and in each the extent of the changes seen by electron microscopy was reduced. In another study involving 14 patients with underlying rheumatoid arthritis who developed D-penicillamine nephropathy, striking persistence in the renal lesions was seen. In two patients, repeat renal biopsies one year after the cessation of D-penicillamine therapy showed no resolution of the histopathological findings (152). It is generally accepted, however, that this renal lesion is reversible but that proteinuria may persist for months, or even years, before it completely resolves.

It is currently felt that treatment with penicillamine can be continued until the degree of proteinuria reaches 2 grams per 24 hours (158). At this point, the drug can be discontinued or the dose can be reduced. If the drug is discontinued, it can usually be restarted at a lower dose when the proteinuria has diminished. The rate of clearance of proteinuria appears to be independent of whether the drug is discontinued, but the renal lesion is almost always reversible.

Rapidly progressive glomerulonephritis (159) and renal vasculitis (160) have also been reported to develop in patients treated with penicillamine. Each can lead to rapid renal failure and necessitates discontinuance of the drug.

Thrombocytopenia occurs in about 4-7% of treated patients. It is usually seen between the tenth and twentieth week of therapy (126), but has been seen during the first month of therapy. The platelet count most often falls progressively and not abruptly. Therapy must be discontinued if the platelet count drops below $100,000/mm^3$. After penicillamine is stopped and the platelet count returns to normal, penicillamine may be cautiously reinstituted at a lower dose (158). Neutropenia, which may occur in as many as 2% of the patients, requires discontinuance of therapy.

Administration of D-penicillamine has been associated with a wide variety of other side effects that fortunately occur much less frequently.

SIDE EFFECTS IN PATIENTS
TREATED WITH D-PENICILLAMINE

a) Gastrointestinal:

hematemesis
diarrhea
peptic ulceration
cholestatic jaundice
pancreatitis

b) Neurologic:

diplopia
tinnitus
optic neuritis
neuropathy

c) Dermatologic:

skin odor
toxic epidermal necrolysis
increased skin friability
elastosis perforans serpiginosa
benign mucous membrane pemphigoid
lichenoid eruption

d) Miscellaneous:

fever
bone marrow aplasia
obliterative bronchiolitis
mammary hyperplasia
proliferative glomerulonephritis
thrombophlebitis
facial edema
cystitis
chronic lymphatic leukemia
IgA deficiency
miliary pulmonary infiltrates
red cell aplasia

The administration of D-penicillamine has also been associated with the development of a number of side effects that appear to involve the development of autoantibodies or other auto-immune phenomena. It should be noted that these side effects do not appear to result from some unique imbalance of immunoregulatory mechanisms found in rheumatoid arthritis patients as most have also been reported in patients with Wilson's disease or cystinuria treated with penicillamine.

"AUTO-IMMUNE" SIDE EFFECTS IN
PATIENTS TREATED WITH D-PENICILLAMINE

Myasthenia gravis
Pemphigus foliaceus
Pemphigus vulgaris
Goodpasture's syndrome
Drug-induced SLE
Immune complex nephropathy
Auto-immune hemolytic anemia
Thyroiditis
Polymyositis, dermatomyositis
Thrombotic thrombocytopenic purpura

Myasthenia gravis has been reported to develop in a small number of individuals with either Wilson's disease or rheumatoid arthritis treated with D-penicillamine (161-168). In general, this is a reversible clinical syndrome that closely resembles idiopathic

myasthenia gravis and remits with discontinuance of the drug. The presence of anti-acetylcholine receptor antibodies has been found in most of these patients (165-168). This antibody is not found in D-penicillamine treated patients who do not have the myasthenic syndrome. D-penicillamine treated patients do have an increased incidence of another anti-muscle antibody, however. Thus, 18% of them have been reported to have anti-striational antibodies although this does not appear to correlate with the development of myasthenia (162). Findings in 20 patients who developed myasthenia gravis are given below (168):

Findings in 20 Patients with Rheumatoid Arthritis
Who Developed Myasthenia Gravis Associated with
D-Penicillamine Therapy (Bucknall, Scan. J. Rheum. 1979)

D-penicillamine therapy
duration - 4-24 months
dose - 13 - >750mg/day
7 - <600mg/day

Associated autoimmune phenomena
1 - Nephropathy
1 - Drug-induced lupus syndrome
1 - Thyroiditis

Auto-antibodies
7/11 - ANA
8/9 - anti-acetylcholine receptor antibodies

Therapy
1 - nil
19 - anticholinesterase
4 - thymectomy

Outcome
10 - complete recovery
9 - partial recovery
1 - death

Therapy with D-penicillamine has also been associated with the development of pemphigus foliaceus and pemphigus vulgaris in a number of patients (142-150). These syndromes have been reported in Wilson's disease patients and individuals with rheumatoid arthritis and scleroderma. IgG antibodies to epidermal intercellular material has been demonstrated in many of the cases.

A clinical syndrome compatible with Goodpasture's syndrome has been reported in three patients with Wilson's disease (169) and three patients with rheumatoid arthritis (170,171). Although the clinical syndromes were consistent with the diagnosis, there has been no documentation of the presence of anti-glomerular basement membrane antibodies in these patients.

In addition, the drug-induced lupus syndrome (172-174), hemolytic anemia (175), polymyositis (176-178) and thrombotic thrombocytopenic purpura (179) have all been reported to develop in patients treated with D-penicillamine.

A number of studies have been undertaken to develop treatment protocols that might decrease the incidence of adverse reactions in D-penicillamine treated patients. Most of these have attempted to use lower daily doses of D-penicillamine. In a double blind trial, Dixon et al (133) found that a daily dose of 600mg of D-penicillamine was as effective as higher doses but was less toxic. The results of these findings are shown here.

Incidence of Side Effects with
D-penicillamine Therapy
Dixon et al, Ann. Rheum. Dis. 34:416, 1975

	Patient Group		
		D-penicillamine(mg/day)	
	Control (43)	600 (34)	1200 (44)
Number of adverse reactions	21	25	48
Number complaining	15 (34%)	17 (50%)	30 (67%)
Number of withdrawals	4 (9%)	9 (26%)	18 (40%)

These results were supported by a number of other trials (114,126,135) and have been enthusiastically embraced by the rheumatologic community. However, a recent trial suggests that the use of low dose D-penicillamine therapy may not be associated with a diminished incidence of toxicity. Thus, in a retrospective study of 114 patients, Webley and Coomes (141) found that both the therapeutic effect and the incidence of toxicity were comparable in patients treated with low and high dose penicillamine.

Comparison of High and Low D-Penicillamine
Dose Schedules
(Webley and Coombs, J. Rheum. 6:20, 1979)

	D-Penicillamine Dose	
	>600mg/day n=43	<600mg/day n=73
	(percent)	
Improvement	54	47
Side Effects	52	42
Proteinuria	10	8
Marrow Depression	10	6
Withdrawal	41	43

Corke and Huskisson (180) similarly found no relationship between the dose of D-penicillamine and the occurrence of side effects. A number of ongoing trials are currently attempting to determine whether lower dose regimens will alter toxicity. A preliminary report from a prospective cooperative trial sponsored by the European League Against Rheumatism indicates that the incidence of adverse reactions is similar in patients treated with 250 or 750mg per day (181).

Corke and Huskisson (180) found that the development of toxic reactions to D-penicillamine could not be correlated with any of a variety of characteristics of the patients treated including sex, age, duration of disease, latex fixation titer, ANA titer, complement level, serum immunoglobulin concentrations or presence of Sjögren's syndrome. A recent report suggests that the development of toxicity to both gold and D-penicillamine may be related to the HLA type of the individual. Panayi et al (182) studied 95 patients with rheumatoid arthritis and 200 health controls. They found that the incidence of HLA-DRW4 was significantly more prevalent among the RA patients, confirming the findings of Peter Stastny (183).

Prevalence of HLA-DR Antigens in
Patients with Rheumatoid Arthritis
Panayi et al, Br. Med. J. 2:1326, 1978

Antigen	Rheumatoid Arthritis n=95	Control n=200	p
	(percent)		
DRW2	13.7	30.0	<0.002
DRW3	29.5	27.0	NS
DRW4	55.8	33.5	<0.0005

In addition, they found that there was a significant association between the presence of DRW2 and DRW3 and toxic reactions to either gold thiomalate or D-penicillamine. These data suggest that the HLA-DR phenotype of the individual is associated not only with the likelihood of developing RA, but also with the potential for adverse reactions to gold or D-penicillamine.

Association between HLA-DRW2 and/or DRW3 and
Toxic Reactions to D-penicillamine and Gold

Antigen	Number of Patients Toxic Reactions	No Toxic Reactions	p
DRW2+DRW3	21	5	<0.025
Other	16	17	

The relationship between antecedent gold therapy and the incidence of toxic reactions to D-penicillamine has also been examined. The Multicentre Trial Group found that previous chrysotherapy did not influence the likelihood of developing an adverse reaction to D-penicillamine (184). However, more recent studies have suggested that patients who have received gold therapy in the past may have a greater incidence of D-penicillamine induced rashes (141) and those who have had previous toxic reactions to gold may have an increased incidence of D-penicillamine induced thrombocytopenia, pemphigus or proteinuria (185).

One additional adverse effect of D-penicillamine therapy results from its capacity to inhibit collagen cross-linking (102-105). Thus, it has been shown in both experimental animals (186,187) and in man (188,189) that administration of D-penicillamine can inhibit wound healing. The mean time to wound healing after 42 orthopedic surgical procedures in 21 patients with rheumatoid arthritis treated with penicillamine was 19.8±13.1 days (189). Patients with rheumatoid arthritis healed similar wounds in 16.6±7.5 days and normals healed in 15.2±7.9 days. There was no greater incidence of complications in the penicillamine treated patients, however. Teigland (190) also found no increased rate of complications in 22 orthopedic procedures carried out on 20 patients with rheumatoid arthritis treated with penicillamine. Despite the report of one patient with rheumatoid arthritis treated with penicillamine who had a complete failure of wound healing following thoracotomy (188), it is currently felt that the modest increase in the time of wound healing does not pose a significant problem and that it is not necessary to discontinue penicillamine for surgical procedures.

D-penicillamine: Current Recommendations (158)

Penicillamine can be recommended for the treatment of severe rheumatoid arthritis. The indication for penicillamine therapy is sustained progressive disease unresponsive to anti-inflammatory agents. Other indications are the presence of extra-articular manifestations, such as vasculitis or Felty's syndrome, although use of D-penicillamine in these syndromes is based largely on anecdotal information (9,38,126,191-197). Preliminary reports have indicated that juvenile rheumatoid arthritis may also respond to therapy with penicillamine and that the incidence of toxicity in children may be less than in adults (67,68). However, the drug has not been approved for general use in children.

The drug should be begun at an initial dose of 250 mg per day, given on an empty stomach. This is to ensure uniformity of absorption. If needed, the dose may be increased by 250 mg daily every three months until a response is seen or a total dose of one gram per day is reached. In rare patients, a dose of greater than one gram per day may be needed. The maximum duration of therapy is unknown, but penicillamine therapy appears to be a life-long proposition. Some patients have been treated for as long as 10 years and discontinuance of therapy has been associated with the development of increased disease activity (9).

What result is expected when a patient with rheumatoid arthritis is begun on penicillamine? First, the response to therapy will be delayed. Three months of therapy may be needed before the earliest signs of improvement are seen. If a patient is going to respond to penicillamine, some evidence will usually be seen by six months. If there is no evidence of toxicity, it is probably worthwhile

continuing for one year before abandoning therapy. Until a response is observed, non-specific anti-inflammatory drugs must be continued as penicillamine itself has little anti-inflammatory action. As mentioned before, one can expect improvement in 2/3 to 3/4 of treated patients. Even in patients who have failed on gold, one can expect improvement with penicillamine in a similar number (184). Toxic reactions may be observed in 1/4 to 2/3 of treated patients and may require discontinuance of therapy in anywhere from 10-40%.

Because of the incidence of side effects, patients undergoing penicillamine therapy must be followed carefully. It is recommended that they receive complete blood counts and urinalyses every two weeks for the first six months of therapy and monthly thereafter. Depressed white counts or platelet counts below $100{,}000/mm^3$ are indications to stop therapy. In addition, a progressive fall in the WBC or platelet count on three successive occasions, although both may remain within the normal range, may be a sign of impending marrow depression and probably indicates that the drug should be discontinued. Proteinuria of greater than 2 gm per 24 hours (the F.D.A. suggests one gm per day) is also an indication to stop therapy.

Pregnancy is a contra-indication to the use of D-penicillamine. In 1971, Mjølnerød et al (198) reported that a woman with cystinuria treated with two grams of D-penicillamine daily during pregnancy gave birth to a child with a generalized connective tissue defect including lax skin, hyperflexibility of the joints, vascular fragility and impaired wound healing. Similarly, a woman with rheumatoid arthritis who received 900 mg of D-penicillamine per day during pregnancy gave birth to a child with multiple congenital anomalies including lax skin (199). Although it has been reported that a number of women with Wilson's disease or cystinuria treated with D-penicillamine during pregnancy have produced normal offspring (22-202), it is recommended that D-penicillamine therapy for rheumatoid arthritis be discontinued during pregnancy.

Penicillin allergy is not a contra-indication to continued penicillamine therapy as no increased incidence of reactions to penicillamine in penicillin-allergic individuals has been reported.

D-Penicillamine: Mechanism of Action

The mechanism of action of D-penicillamine in rheumatoid arthritis is unclear at the present time. Penicillamine was first used to treat RA because of the belief that it might directly dissociate IgM rheumatoid factor *in vivo* and thus act to ameliorate the disease. Treatment of RA with D-penicillamine has been demonstrated to lead to decreased RF titers. However, it is unlikely that this effect results from D-penicillamine-induced dissociation of IgM RF since the concentration of D-penicillamine attained *in vivo* is about 200 fold less than that needed to dissociate

macroglobulins (2,101). In addition, RF titers remain depressed for prolonged periods after the cessation of drug therapy (37,203) and titers of IgG RF, which cannot be dissociated directly by D-penicillamine are also decreased (113,197). It would thus appear that the reduction in RF titers is a secondary phenomenon which reflects an amelioration of the underlying rheumatoid process. This suggests that the primary action of penicillamine in RA may result from a direct anti-inflammatory effect of the drug, although such an action is not apparent clinically. In most animal models of inflammation, such as adjuvant arthritis, D-penicillamine exerts little inhibitory action (204-207) and in some models may actually enhance inflammation (208). However, complexes formed between penicillamine and copper have been shown to have potent anti-inflammatory activity in a number of experimental models of inflammation (207,209). In addition, it has been shown that these penicillamine-copper complexes have potent superoxide dismutase activity (210,211). This might be important in mitigating the toxic effects of superoxide anions produced in inflammatory sites. Finally, it has been shown that penicillamine may depress neutrophil chemotaxis, although this effect is usually seen only with concentrations of D-penicillamine far in excess of that attained *in vivo* (212,213).

It has been suggested that D-penicillamine might exert an immunosuppressive action and, thus, act by suppressing the ongoing immune response that underlies the chronic inflammation (214). A number of observations support this idea. First, successful therapy of RA with penicillamine results in decreased rheumatoid factor titers and often immunoglobulin levels (82,215,216), as well as decreased levels of circulating immune complexes (66,197). Evidence as to whether penicillamine acts to suppress the immune response in laboratory animals is conflicting. Thus, a number of studies have indicated that it may either inhibit (217,218), enhance (219), or have no effect (205,220) on the immune response of intact animals. However, more recently Hunneyball et al (221,222) have shown that rabbits treated with 15mg per kg of D-penicillamine exhibited a depressed *in vivo* antibody response to immunization with egg albumin. This was especially marked late in the immune response and seemed preferentially to effect high avidity IgG antibody production. Concomitant with this, there was a more striking decline in cell mediated immunity. After 230 days of penicillamine therapy (15mg/kg), skin test reactivity to the immunizing antigens was markedly depressed. This suggested that penicillamine may well exert its action by depressing the function of T lymphocytes *in vivo*.

In vitro experiments using human lymphocytes have further defined an action of D-penicillamine. Experiments carried out in our laboratory have shown that D-penicillamine may selectively inhibit T lymphocyte function (223). Helper T cells, or those involved in facilitating the generation of antibody forming cells from B cell per-cursors, appear to be preferentially inhibited in their activity (224).

It has previously been shown that the signs and symptoms of rheumatoid arthritis can be ameliorated by thoracic duct drainage with removal of large numbers of T lymphocytes (225-228). Therefore, the capacity of D-penicillamine to pharmacologically alter T cell function could well explain its therapeutic efficacy in rheumatoid arthritis.

REFERENCES

1) Abraham, E.P., E. Chain, W. Baker and R. Robinson. 1943. Penicillamine, a characteristic degradation product of penicillin. Nature (Lond.) 151:107.

2) Dresner, E. and P. Trombly. 1960. Chemical dissociation of rheumatoid factor in vitro and in vivo. Clin. Res. 8:16.

3) Griffin, S.W., A. Ulloa, M. Henry, M.L. Johnston, and H.L. Holley. 1960. In vivo effect of penicillamine on circulating rheumatoid factor. Clin. Res. 8:87.

4) Huskisson, E.C. 1976. Specific therapy for rheumatoid arthritis. Rheum. Rehab. 15:133.

5) Dorwart, B.B., E.P. Gall, H.R. Schumacher and R.E. Krauser. 1978. Chrysotherapy in psoriatic arthritis. Arth. Rheum. 21:513.

6) Black, R.L., W.M. O'Brien, E.J. Van Scott, R. Auerbach, A.Z. Eisen and J.J. Bunim. 1964. Methotrexate therapy in psoriatic arthritis. Double-blind study on 21 patients. JAMA 189:743.

7) Levy, J., H.E. Paulus, E.V. Barnett, M. Sokoloff, R. Bangert and C.M. Pearson. 1972. A double blind controlled evaluation of azathioprine treatment in rheumatoid arthritis and psoriatic arthritis. Arth. Rheum. 15:116.

8) Levine, S. and H.E. Paulus. 1976. Treatment of psoriatic arthritis with azaribine. Arth. Rheum. 19:21.

9) Huskisson, E.C. and F.D. Hart. 1972. Penicillamine in the treatment of rheumatoid arthritis. Ann. Rheum. Dis. 31:402.

10) Forestier, J. 1932. The treatment of rheumatoid arthritis with gold salts injections. Lancet. 1:441.

11) Research Subcommittee of the Empire Rheumatism Council: Gold therapy in rheumatoid arthritis. 1960. Report of a multicentre controlled trial. Ann. Rheum. Dis. 19:95.

12) Research Subcommittee of the Empire Rheumatism Council: Gold therapy in rheumatoid arthritis. 1961. Final report of a multicentre controlled trial. Ann. Rheum. Dis. 20:315.

13) The Cooperating Clinics Committee of the American Rheumatism Association: 1973. A controlled trial of gold salt therapy in rheumatoid arthritis. Arth. Rheum. 16:353.

14) Sigler, J.W., G.B. Bluhm, H. Duncan, J.T. Sharp, D.C. Ensign, and W.R. McCrum. 1974. Gold salts in the treatment of rheumatoid arthritis. A double-blind study. Ann. Int. Med. 80:21.

15) Fosdick, W.M., J.L. Parsons and D.F. Hill. 1969. Long-term cyclophosphamide therapy in rheumatoid arthritis: A progress report, six years experience. Arth. Rheum. 12:663.

16) Mason, M., H.L.F. Currey, C.G. Barnes, J.F. Dunne, B.L. Hazleman and I.D. Strickland. 1969. Azathioprine in rheumatoid arthritis. Brit. Med. J. 1:420.

17) Cooperating Clinics of the American Rheumatism Association. 1970. A controlled trial of cyclophosphamide in rheumatoid arthritis. New. Eng. J. Med. 283:883.

18) Urowitz, M.B., D.A. Gordon, H.A. Smythe, W. Pruzanski and M.A. Ogryzlo. 1973. Azathioprine in rheumatoid arthritis. A double blind cross-over study. Arth. Rheum. 16:411.

19) Currey, H.L.F., J.Harris, R.M. Mason, J. Woodland, T. Beveridge, C.J. Roberts, D.W. Vere, A.StJ. Dixon, J. Davies and B. Owen-Smith. 1974. Comparison of azathioprine, cyclophosphamide and gold in treatment of rheumatoid arthritis. Brit. Med. J. 3:763.

20) Townes, A.S., J.M. Sowa and L.E. Shulman. 1976. Controlled trial of cyclophosphamide in rheumatoid arthritis. Arth. Rheum. 19:563.

21) Multicentre Study Group. 1978. Levamisole in rheumatoid arthritis. A randomised double-blind study comparing two dosage regimens of levamisole with placebo. Lancet. 2:1007.

22) Multicentre Study Group. 1978. A multicentre randomized double-blind study comparing two dosages of levamisole in rheumatoid arthritis. J. Rheum. 5 (Suppl. 4):5.

23) Aylward, M., J. Maddock, R. Wheeldon and R.J. Parker. 1975. A study of the influence of various antirheumatic drug regimens on serum acute-phase proteins, plasma tryptophan, and erythrocyte sedimentation rate in rheumatoid arthritis. Rheum. Rehab. 14:101.

24) Gibson, T., E.C. Huskisson, J.A. Wojtulewski, P.J. Scott, H.W. Balme, H.C. Burry, R. Grahame and F.D. Hart. 1976. Evidence that D-penicillamine alters the course of rheumatoid arthritis. Rheum. Rehab. 15:211.

25) Franchimont, P., G. Heynen, and C.H. Hauwaert. 1978. Adverse reactions to the principal drugs used in rheumatoid arthritis.-A review. J. Rheum. 5(Suppl. 4):85.

26) Walshe, J.M. 1953. Disturbances of amino acid metabolism following liver injury. Quart. J. Med. 22:483.

27) Walshe, J.M. 1956. Penicillamine: new oral therapy for Wilson's disease. Amer. J. Med. 21:487.

28) Tabachnik, M., H.N. Eisen and B. Levine. 1954. New mixed disulfide; penicillamine-cysteine. Nature 174:701.

29) Deutsch, H.F. and J.I. Morton. 1957. Dissociation of human serum macroglobulins. Science 125:600.

30) Sharp, J.T., E. Calkins, A.S. Cohen, A.F. Schubart and J.J. Calabro. 1964. Observations on the clinical, chemical and serological manifestations of rheumatoid arthritis based on the course of 154 cases. Medicine (Baltimore) 43:41.

31) Sievers, K. 1965. The rheumatoid factor in definite rheumatoid arthritis. Acta Rheum. Scand. (Suppl) 9:1.

32) Rose, H.M., C. Ragan, E. Pearce and M.O. Lipman. 1948. Differential agglutination of normal and sensitized sheep erythrocytes by sera of patients with rheumatoid arthritis. Proc. Soc. Exp. Biol. Med. 68:1.

33) Heimer, R. and O.M. Federico. 1958. Depolymerization of the 19S antibodies and the 22S rheumatoid factor. Clin. Chim. Acta. 3:496.

34) Ritzmann, S.E., S.I. Coleman and W.C. Levin. 1960. The effect of some mercaptanes upon a macrocry-gelglobulin; modifications induced by cysteamine, penicillamine and penicillin. J. Clin. Invest. 39:1320.

35) Ritzmann, S.E. and W.C. Levin. 1961. Effect of mercaptanes in gold agglutinin disease. J. Lab. Clin. Med. 57:718.

36) Jaffe, I.A. 1962. Intra-articular dissociation of the rheumatoid factor. J. Lab. Clin. Med. 60:409.

37) Jaffe, I.A. 1963. Comparison of the effect of plasmapheresis and penicillamine on the level of circulating rheumatoid factor. Ann. Rheum. Dis. 22:71.

38) Jaffe, I.A. 1964. Rheumatoid arthritis with arteritis. Report of a case treated with penicillamine. Ann. Int. Med. 61:556.

39) Multicentre Trial Group. 1973. Controlled trial of D(-)Penicillamine in severe rheumatoid arthritis. Lancet 1:275.

40) Scheinberg, I.H. and I. Sternlieb. 1960. The long-term management of hepatolenticular degeneration (Wilson's disease). Amer. J. Med. 29:316.

41) Sternlieb, I. and I.H. Scheinberg. 1968. Prevention of Wilson's disease in asymptomatic patients. New Eng. J. Med. 278:352.

42) Strickland, G.T., D. Frommer, M.L. Leu,, R. Pollard, S. Sherlock, and J.N. Cumings. 1973. Wilson's disease in the United Kingdom and Taiwan. Quart. J. Medicine. 42:619.

43) Grand, R.J. and G.F. Vawter. 1975. Juvenile Wilson's disease: Histologic and functional studies during penicillamine therapy. J. Pediat. 87:1161.

44) Crawhall, J.C., E.F. Scowen and R.W.E. Watts. 1963. Effects of penicillamine on cystinuria. Brit. Med. J. 1:588.

45) Lotz, M. and F.C. Bartter. 1965. Stone dissolution with D-penicillamine in cystinuria. Brit. Med. J. 2:1408.

46) Seegmiller, J.E., T. Friedmann, H.E. Harrison, V. Wong, and J.A. Schneider. 1968. Cystinosis. Ann. Int. Med. 68:883.

47) Dahlberg, P.J., C.J. VanDenBerg, S.B. Kurtz, D.M. Wilson, L.H. Smith. 1977. Clinical features and management of cystinuria. Mayo Clin. Proc. 52:533.

48) Chisholm, J.J. 1968. The use of chelating agents in the treatment of acute and chronic lead intoxication in childhood. J. Pediat. 73:1.

49) Sachs, H.K., L.A. Blanksma, E.F. Murray and M.J. O'Connel. 1970. Ambulatory treatment of lead poisoning. Report of 1,155 cases. Pediat. 46:389.

50) Vitale, L.F., A. Rosalinas-Bailon, D. Folland, J.F. Brennan, and B. McCormick. 1973. Oral penicillamine therapy for chronic lead poisoning in children. Ped. Pharm. Ther. 83:1041.

51) Kuruvilla, A., P.S. Bergeson and A.K. Done. 1975. Arsenic poisoning in childhood. An unusual case report with special notes on therapy with penicillamine. Clin. Toxicol 8:535.

52) Peterson, R.G., and B.H. Rumack. 1977. D-penicillamine therapy of acute arsenic poisoning. J. Pediat. 91:661.

53) Kark, R.A.P., D.C. Poskanzer, J.D. Bullock, and G. Boylen. 1971. Mercury poisoning and its treatment with N-acetyl-D, L-penicillamine. New Eng. J. Med. 285:10.

54) Fulghum, D.D. and R. Katz. 1968. Penicillamine for Scleroderma. Arch. Derm. 98:51.

55) Bluestone, R., R. Grahame, V. Holloway, and P.J.L. Holt. 1970. Treatment of systemic sclerosis with D-penicillamine. A new method of observing the effects of treatment. Ann. Rheum. Dis. 29:153.

56) Moynahan, E.J. 1973. Morphea (Localized cutaneous scleroderma) treated with low-dosage penicillamine (4 cases, including Coup de Sabre). Proc. Roy. Soc. Med. 66:1083.

57) Tio, H., L. van Wijk and E. deHaan. 1973. Treatment of progressive systemic sclerosis (PSS) with penicillamine. Acta Med. Scand. 193:477.

58) Thomson, J., J.A. Milne. 1974. Two years of penicillamine for progressive systemic sclerosis: a case report. Post Grad. Med. J. 50(suppl 2):36.

59) Moynahan, E.J. 1974. Penicillamine in the treatment of morphoea and keloid in children. Post. Grad. Med. J. 50(suppl 2):39.

60) Asboe-Hansen. 1975. Treatment of generalized scleroderma with inhibitors of connective tissue formation. Acta Dermatovener 55:461.

61) Bröll, V.H., G. Tausch and R. Eberl. 1976. Zur Behandlung der interstitiellen Lungenfibrose mit D-penicillamin bei progressiver Sklerodermie (Langzeitstudie). Wiener Klinische Wochenschrift 88:292.

62) Mellstedt, H., B. Fagrell, G. Holm and M. Bjorkholm. 1977. D-penicillamine treatment in systemic sclerosis. Scan. J. Rheum. 6:92.

63) Chou, T-H., E.J. Hill, E. Bartle, K. Woolley, V. LeQuire, W. Olson, R. Roelofs and J.H. Park. 1975. Beneficial effects of penicillamine treatment on hereditary avian muscular dystrophy. J. Clin. Invest. 56:842.

64) Jain, S., P.J. Scheuer, S. Samourian, J.O. McGee and S. Sherlock. 1977. A controlled trial of D-penicillamine therapy in primary biliary cirrhosis. Lancet 1:831.

65) Fleming, C.R., J. Ludwig and E.R. Dickson. 1978. Asymptomatic primary biliary cirrhosis. Presentation, histology and results with D-penicillamine. Mayo Clin. Proc. 53:587.

66) Epstein, O., D. DeVilliers, S. Jain, B.J. Potter, H.C. Thomas and S. Sherlock. 1979. Reduction of immune complexes and immunoglobulins induced by D-penicillamine in primary biliary cirrhosis. New Eng. J. Med. 300:274.

67) Schairer, H. and E. Stoeber. 1976. Long term follow-up of 235 cases of juvenile rheumatoid arthritis treated with D-penicillamine . in Penicillamine Research in Rheumatoid Disease. edited by E. Munthe. Fabritius and Sønner, Oslo, p. 279.

68) Ansell, B. and M.A. Hall. 1977. Penicillamine. Arth. Rheum. 20(suppl):536.

69) Wildhirt, E. 1974. The treatment of chronic liver disease with D-penicillamine. Postgrad Med. J. 50(suppl 2):42.

70) Stern, R.B., S.P. Wilkinson, P.J.N. Howorth and R. Williams. 1977. Controlled trial of synthetic D-penicillamine and prednisone in maintenance therapy for active chronic hepatitis. Gut. 18:19.

71) Golding, D.N. 1974. D-penicillamine in ankylosing spondylitis and polymyositis. Postgrad Med. J. 50(suppl 2):62.

72) Huskisson, E.C. 1976. Treatment of palindromic rheumatism with D-penicillamine. Br. Med. J. 2:979.

73) Bloch, H.S., A. Prasad, A. Anastasi and D.R. Briggs. Serum protein changes in Waldenströms macroglobulinemia during administration of a low molecular weight thiol (penicillamine) J. Lab. Clin. Med. 56:212, 1960.

74) Lakatos, L., B. Köver, Gy. Oroszlán and Z. Vekerdy. 1976. D-penicillamine therapy in ABO hemolytic disease of the newborn infant. Eur. J. Pediat. 123:133.

75) Korányi, G., J. Kovacs and I. Vörös. 1978. D-penicillamine treatment of hyperbilirubinaemia in preterm infants. Acta Pediat. Acad. Sci. Hung. 19:9.

76) Tarrant, M.E., S. Wedley and T.J. Woodage. 1971. The effect of penicillamine in the treatment of experimental schistosomiasis with tartar emetic. Ann. Trop. Med. Parasitol. 65:233.

77) Henry, W., N.I. Girgis and N.S. Mansour. 1976. In vitro studies on the use of penicillamine with tartar emetic against Schistosoma mansoni worms. Ann. Trop. Med. Parasitol. 70:425.

78) Prescott, L.F., G.R. Sutherland, J. Park, I.J. Smith and A.T. Proudfoot. 1976. Cysteamine, methionine and penicillamine in the treatment of paracetamol poisoning. Lancet 2:109.

79) Hunter, G.A. and G.F. Donald. 1970. The treatment of cutaneous porphyria with chloroquine or D-penicillamine. Br. J. Derm. 83:702.

80) Fluur, E. and B. Olhagen. 1975. Pharmacotherapy in tracheal stenosis. Acta Otolaryngol. 79:442.

81) Nagasawa, H.T., D.J.W. Goon, E.G. DeMaster and C.S. Alexander. 1977. Lowering of ethanol-derived circulating blood acetaldehyde in rats by D-penicillamine. Life Sciences. 20:187.

82) Goldberg, L.S. and E.V. Barnett. 1970. Essential cryoglobulinemia. Immunologic studies before and after penicillamine therapy. Arch. Int. Med. 125:145.

83) Leca, A.P. and J.P. Camus. 1975. Ankylosing spondylitis: treatment failures with D-penicillamine. Nouv. Presse Med. 4:112.

84) Bird, H.A. and A. StJ. Dixon. 1977. Failure of D-penicillamine to affect peripheral joint involvement in ankylosing spondylitis or HLA B27 associated arthropathy. Ann. Rheum. Dis. 36:289.

85) Scharf, Y. and M. Nahir. 1976. Penicillamine in ankylosing spondylitis. Arth. Rheum. 19:122.

86) Nicolson, G.A., A.C. Greiner, W.J.G. McFarlane and R.A. Baker. 1966. Effects of penicillamine on schizophrenic patients. Lancet 1:344.

87) Affleck, J.W., A.J. Cooper, A.D. Forrest, J.R. Smythies and A.K. Zealley. 1969. Penicillamine and schizophrenia - a clinical trial. Brit. J. Psychiat. 115:173.

88) Mattke, D.J. and M. Adler. 1971. Mode of action of D-penicillamine in chronic schizophrenia. Dis. Nerv. Sys. 32:388.

89) Cohen, H.J., L.S. Lessin, J. Hallal and P. Burkholder. 1975. Resolution of primary amyloidosis during chemotherapy. Studies in a patient with nephrotic syndrome. Ann. Int. Med. 82:466.

90) Cohen, H.J. 1978. Combination chemotherapy for primary amyloidosis reconsidered. Ann. Int. Med. 89:572.

91) Resnick, R.H., J. Boitnott, F.L. Iber, H. Makopour and J.J. Cerda. 1974. Preliminary observations of d-penicillamine therapy in acute alcoholic liver disease. Digestion 11:257.

92) Eyring, E.J. and E.P. Engleman. 1963. Interaction of gold and penicillamine. Arth. Rheum. 6:216.

93) Davis, C.M. 1969. D-penicillamine for the treatment of gold dermatitis. Amer. J. Med. 46:472, 1969.

94) Davis, P. and D. Barraclough. 1977. Interaction of D-penicillamine with gold salts. Arth. Rheum. 20:1413, 1977.

95) Harth, M., J.P. Hickey, W.K. Coulter, J.M. Thompson and T.F. Disney. 1978. Gold-induced thrombocytopenia. J. Rheum. 5:165.

96) Cendrowski, W. and A. Czlonkowska. 1976. Penicillamine in multiple sclerosis. Acta. Neurol. Scand. 54:281.

97) Aposhian, H.V. 1971. Penicillamine and analogous chelating agents. Ann. N.Y. Acad. Sci. 179:481.

98) Friedman, M. 1976. Chemical basis for pharmacological and therapeutic actions of penicillamine. Adv. Exp. Med. Biol. 86B:649.

99) Fleming, C.R., E.R. Dickson, A.H. Baggenstoss and J.T. McCall. 1974. Copper and primary biliary cirrhosis. Gastroenterology. 67:1182.

100) Deering, T.B., E.R. Dickson, C.R. Fleming, M.G. Geall, J.T. McCall and A.H. Baggenstoss. 1977. Effect of D-penicillamine on copper retention in patients with primary biliary cirrhosis. Gastroenterology. 72:1208.

101) Van de Stadt, R.J. Personal communication.

102) Siegel, R.C. 1977. Collagen cross-linking. Effect of D-penicillamine on cross-linking in vitro. J. Biol. Chem. 252:254.

103) Nimni, M.E. and L.A. Bavetta. 1965. Collagen defect induced by penicillamine. Science. 150:905.

104) Nimni, M.E. 1968. A defect in the intramolecular and intermolecular cross-linking of collagen caused by penicillamine. J. Biol. Chem. 243:1457.

105) Harris, E.D. and A. Sjoerdsma. 1966. Effects of penicillamine on human collagen and its possible application to treatment of scleroderma. Lancet. 2:996.

106) Jaffe, I.A.. 1972. The antivitamin B_6 effect of penicillamine: Clinical and immunological implications. Adv. Biochem. Pharmacol. 4:217.

107) Perrett, D., W. Sneddon and A.D. Stephens. 1976. Studies on D-penicillamine metabolism in cystinuria and rheumatoid arthritis: Isolation of S-methyl-D-penicillamine. Biochem. Pharmacol. 25:259.

108) Gibbs, K. and J.M. Walshe. 1971. Studies with ^{35}S-labelled DL-penicillamine in patients with Wilson's disease. Quart. J. Med. 40:275.

109) Jellum, E. and S. Skrede. 1976. Biological aspects of thiol-disulfide reactions during treatment with penicillamine. in Penicillamine Research in Rheumatoid Disease. E. Munthe, editor. Fabritius and Sønner, Oslo. P. 68.

110) Aposhian, H.V. and L.S. Bradham. 1959. Metabolism in vitro of the sulphydryl amino acids, L and D-penicillamine. Biochem. Pharmacol. 3:38.

111) Wei, P. and A. Sass-Kortsak. 1970. Urinary excretion and renal clearances of D-penicillamine in humans and the dog. Gastroenterology. 58:288.

112) Zuckner, J. R.H. Ramsey, R.W. Dorner and G.E. Gantner, Jr. 1970. D-penicillamine in rheumatoid arthritis. Arth. Rheum. 13:131.

113) Jaffe, I.A. 1970. The treatment of rheumatoid arthritis and necrotizing vasculitis with penicillamine. Arth. Rheum. 13:436.

114) Day, A.T., J.R. Golding, P.N. Lee and A.D. Butterworth. 1974. Penicillamine in rheumatoid disease: A long-term study. Brit. Med. J. i:180.

115) Colombo, B., M. Carrabba, F. Fantini, G. Cherie Ligniere and S. Toxi. 1975. La D-penicillamina nel trattamento dell'artrite reumatoide: Nota Preliminare. Reumatismo 27:113.

116) Miehlke, K. 1975. Die Wirkung von D-penicillamine auf den Verlauf der chronischen Polyarthritis. Acta Med. Austriaca 2:127.

117) Tautz, V.I. 1975. Multicentrische ambulante Prufung von Trolovol (D-Penicillamin) bei chronischer Polyarthritis. Zeit Rheum. 34:87.

118) Camus, J.P., J. Grouzet, A.P. Leca and A. Preier. 1976. The treatment of rheumatoid arthritis with penicillamine. In Chronic Forms of Polyarthritis. (ed. F.J. Wagenhauser) Hans Huber, Bern Stuttgart Vienna, p. 245.

119) Gumpel, J.M. 1976. Cyclophosphamide, gold and penicillamine: Disease-modifying drugs in rheumatoid arthritis--Tailored dosage and ultimate success. Rheum. Rehab. 15:217.

120) Hadidi, T. 1976. D-penicillamin bei primar chronischer Poly-Arthritis. Med. Wett. 27:33.

121) Kogstad, O. 1976. D-penicillamine in rheumatoid arthritis: A study in 16 patients with special reference to a patient developing amyloidosis and kidney insufficiency. In Penicillamine Research in Rheumatoid Disease. E. Munthe, editor, Fabritius and Sønner, Oslo, p243.

122) Molony, J., A. McNamara, D. Doyle, H. Durkin, J. Murphy and E. Prenderville. 1976. Clinical experiences with penicillamine in rheumatoid disease. J. Irsh Med. Assoc. 69:41.

123) Munthe, E. 1976. Short term open study of penicillamine in classical rheumatoid arthritis. In Penicillamine Research in Rheumatoid disease. E. Munthe, editor, Fabritius and Sønner, Oslo, p247.

124) Nissila, M., P. Mäkisara, A. Kajander, J. Martio, R. von Essen and G.L. Mäkisara. 1976. Comparison of penicillamine and gold treatment in rheumatoid arthritis: A preliminary report. In Penicillamine Research in Rheumatoid Disease. E. Munthe, editor, Fabritius and Sønner, Oslo, p233.

125) Dippy, J.E. 1977. Penicillamine in rheumatoid arthritis - A 2-year retrospective study in 70 patients. Brit. J. Clin. Prac. 31:5.

126) Hill, H.F.H. 1977. Treatment of rheumatoid arthritis with penicillamine. Semin. Arthritis Rheum. 6:361.

127) Kalliomaki, J.L. 1977. D-penicillamine as an alternative to gold in the treatment of rheumatoid arthritis. Curr. Therap. Res. 21:815.

128) Lopez-Garcia, A. 1977. Revision de 36 casos de arthritis rheumatoide tratados con D-penicilamina. Rev. Espan. Reum. y Enferm Osteo. 20:9.

129) Santamaria, A., R. Ruiz de la Torre, P. Barcelo, Jr., M. Ripoll, J. Escarpenter and P. Barcelo, Sr. 1977. Tratamiento con D-penicilamine durante 3 anos de 100 enfermos affectos de poliartritis reumatoides. Rev. Espan. Rheum. y Enferm. Osteo. 20:335.

130) Saxer, M., A. Oeding, G. Kaganas and E. Vogt. 1977. Klinische Erfahrungen mit dem Basistherapeutikum D-penicillamin (Mercaptyl) bei chronischer Polyarthritis. Praxis 66:240.

131) Tausch, G., H. Bröll and R. Eberl. 1977. Langzeittherapie der chronischem Polyarthritis mit D-Penicillamin. Praxis 66:1427.

132) Tsang, I., C.A. Patterson, H.B. Stein, H.S. Robinson and D.K. Ford. 1977. D-penicillamine in the treatment of rheumatoid arthritis. Arth. Rheum. 20:666.

133) Dixon, A. St. J., J. Davies, T.L. Dormandy, E.B.D. Hamilton, P.J.L. Holt, R.M. Mason, M. Thompson, J.C.P. Weber and D.W. Zutshi. 1975. Synthetic D-penicillamine in rheumatoid arthritis. Double-blind controlled study of a high and low dosage regimen. Ann. Rheum. Dis. 34:416.

134) Berry, H., S.P. Liyanage, R.A. Durance, C.G. Barnes, L.A. Berger and S. Evans. 1976. Azathioprine and penicillamine in treatment of rheumatoid arthritis: A controlled trial. Brit. Med. J. i:1052.

135) Mery, C., F. Delrieu, R. Ghozlan, L. Saporta, F. Simon, B. Amor, C.J. Menkes and F. Delbarre. 1976. Controlled trial of D-penicillamine in rheumatoid arthritis. Scand. J. Rheumatol. 5:241.

136) Shiokawa, Y., Y. Horiuchi, M. Honma, T. Kageyama, T. Okada and T. Azuma. 1977. Clinical evaluation of D-penicillamine by multicentric double-blind comparative study in chronic rheumatoid arthritis. Arth. Rheum. 20:1464.

137) Berry, H., L. Fernandes, A.W. Ford-Hutchinson, S.J.W. Evans and E.B.D. Hamilton. 1978. Alclofenac and D-penicillamine. Comparative trial in rheumatoid arthritis. Ann. Rheum. Dis. 37:93.

138) Mäkisara, P., M. Nissilä, A. Kajander, J. Martio, R. von Essen, P. Anttila and G.L. Mäkisara. 1978. Comparison of penicillamine and gold treatment in early rheumatoid arthritis. Scand. J. Rheum. 7:166.

139) Huskisson, E.C., T.J. Gibson, H.W. Balme, H. Berry, H.C. Burry, R. Grahame, F.D. Hart, D.R.F. Henderson and J.A. Wojtulewski. 1974. Trial comparing D-penicillamine and gold in rheumatoid arthritis. Preliminary report. Ann. Rheum. Dis. 33:532.

140) Haataja, M., M. Nissilä and H.-M. Ruutsalo. 1978. Serum sulfhydryl levels in rheumatoid patients treated with gold thiomalate and penicillamine. Scand. J. Rheum. 7:212.

141) Webley, M. and E.N. Coomes. 1979. An assessment of penicillamine therapy in rheumatoid arthritis and the influence of previous gold therapy. J. Rheum. 6:20.

142) Baum, J. 1979. The use of penicillamine in the treatment of rheumatoid arthritis and scleroderma. Scand. J. Rheum. in press.

143) Marsden, R.A., T.J. Ryan, R.I. Vanhegan, M. Walshe, H. Hill, A.G. Mowat. 1976. Pemphigus foliaceus induced by penicillamine. Brit. Med. J. 2:1423.

144) Tan, S.G. and N.R. Rowell. 1976. Pemphigus-like syndrome induced by D-penicillamine. Brit. J. Dermatol. 95:99.

145) From, E. and P. Frederiksen. 1976. Pemphigus vulgaris following D-penicillamine. Dermatol. 152:358.

146) Davies, M.G. and P. Holt. 1976. Pemphigus in a patient treated with penicillamine for generalized morphea. Arch. Derm. 112:1308.

147) Marsden, R.A., R.P.R. Dawber, P.R. Millard and A.G. Mowat. 1977. Herpetiform pemphigus induced by penicillamine. Brit. J. Dermatol. 97:451.

148) Kristensen, J.K. and S. Wadskov. 1977. Penicillamine-induced pemphigus foliaceus. Acta Dermatovener. 57:69.

149) Sparrow, G.P. 1978. Penicillamine pemphigus and the nephrotic syndrome occurring simultaneously. Brit. J. Dermatol. 98:103.

150) Kennedy, C., L. Hodge and K. U. Sanderson. 1978. Skin changes caused by D-penicillamine treatment of arthritis. Report of three cases with immunological findings. Clin. Exp. Derm. 3:107.

151) Pass, F., S. Goldfischer, I. Sternlieb and I.H. Scheinberg. 1973. Elastosis perforans serpiginosa during penicillamine therapy for Wilson disease. Arch. Derm. 108:713

152) Bacon, P.A., C.R. Tribe, J.C. MacKenzie, J.V. Jones, R.H. Cumming and B. Amer. 1976. Penicillamine nephropathy in rheumatoid arthritis. A clinical, pathological and immunological study. Quart. J. Med. 45:661.

153) Hayslett, J.P., K.G. Bensch, M. Kashgarian and L.E. Rosenberg. 1968. Focal glomerulitis due to penicillamine. Lab. Invest. 19:376.

154) Jaffe, I.A., G. Treser, Y. Suzuki and T. Ehrenreich. 1968. Nephropathy induced by D-penicillamine. Ann. Int. Med. 69:549.

155) Lachmann, P.J. 1968. Nephotic syndrome from penicillamine. Postgrad Med. J. 44(Suppl.):23.

156) Dische, F.E., D.R. Swinson, E.B.D. Hamilton and V. Parsons. 1976. Immunopathology of penicillamine induced glomerular disease. J. Rheum. 3:145.

157) Gärtner, H.V., G.H Neild, A. Bohle, W. Hallauer, G. Hoppe-Seyler, F.M. Luttgen and F. Schollmeyer. 1975. Perimembranous Glomerulonephritis nach penicillaminetherapie. Bericht über 31 Fälle. Klin Wochenschr. 53:835.

158) Jaffe, I.A. 1978. D-penicillamine. Bull. Rheum. Dis. 28:948.

159) Jaffe, I.A. 1976. Penicillamine treatment of rheumatoid arthritis. In Penicillamine Research in Rheumatoid Disease. E. Munthe, editor, Fabritius and Sønner, Oslo, p11.

160) Falck, H.M. T. Tornroth, B. Kock and O. Wegelius. 1979. Fatal renal vasculitis and minimal change glomerulonephritis complicating treatment with penicillamine. Acta Med. Scand. 205:133.

161) Bucknall, R.C., A. St.J. Dixon, E.N. Glick, J. Woodland and D.W. Zutshi. 1975. Myasthenia gravis associated with penicillamine treatment for rheumatoid arthritis. Brit. Med. J. 1:600.

162) Dawkins, R.L., P.J. Zilko and E.T. Owen. 1975. Penicillamine therapy, antistriational antibody and myasthenia gravis. Brit. Med. J. 4:759.

163) Gordon, R.A. and J.W. Burnside. 1977. D-penicillamine-induced myasthenia gravis in rheumatoid arthritis. Ann. Int. Med. 87:578.

164) Bucknall, R.C. 1977. Myasthenia gravis associated with D-penicillamine therapy in rheumatoid arthritis. Proc. Roy. Soc. Med. 70(Suppl 3):114.

165) Masters, C.L., R.L. Dawkins, P.L. Zilko, J.A. Simpson, R.J. Leedman and J. Lindstrom. 1977. Penicillamine-associated myasthenia gravis, antiacetylcholine receptor and anti-striational antibodies. Amer. J. Med. 63:689.

166) Russell, A.S. and J.M. Lindstrom. 1978. Penicillamine-induced myasthenia gravis associated with antibodies to acetyl-choline receptor. Neurol. 28:847.

167) Vincent, A., J. Newsom-Davis and V. Martin. 1978. Anti-acetycholine receptor antibodies in D-penicillamine-associated myasthenia gravis. Lancet 1:1254.

168) Bucknall, R.C. 1979. Myasthenia gravis associated with D-penicillamine therapy in rheumatoid arthritis. Scand. J. Rheum., in press.

169) Sternlieb, I., B. Bennet and I.H. Scheinberg. 1975. D-penicillamine induced Goodpasture's syndrome in Wilson's disease. Ann. Int. Med. 82:673.

170) Gibson, T., H.C. Burry and C. Ogg. 1976. Goodpasture syndrome and D-penicillamine. Ann. Int. Med. 84:100.

171) McCormick, J.N., P. Wood and D. Bell. 1976. Penicillamine-induced Goodpasture's syndrome. In Penicillamine Research in Rheumatoid Disease. E. Munthe, editor, Fabritius and Sønner, Oslo. p268.

172) Harpey, J.P., B. Caille, R. Moulias and J.M. Goust. 1971. Lupus-like syndrome induced by D-penicillamine in Wilson's disease. Lancet 1:292.

173) Camus, J.P., J. Crouzet, J.F. Bach and J.C. Homberg. 1976. Autoantibodies in D-penicillamine treated rheumatoid arthritis. Agents and Actions. 6:351.

174) Harkcom, T.M., D.L. Conn and K.E. Holley. 1978. D-penicillamine and lupus erythematosus-like syndrome. Ann. Int. Med. 89:1012.

175) Harrison, E.E. and J.W. Hickman. 1975. Hemolytic anemia and thrombocytopenia associated with penicillamine ingestion. South. Med. J. 68:113.

176) Schraeder, P.L., H.A. Peters and D.S. Dahl. 1972. Polymyositis and penicillamine. Arch. Neurol. 27:456.

177) Cucher, B.G. and A.L. Goldman. 1976. D-penicillamine-induced polymyositis in rheumatoid arthritis. Ann. Int. Med. 85:615.

178) Petersen, J., P. Halberg. K. Højgaard, B.B. Lyon and S. Ullman. 1978. Penicllamine-induced polymyositis-dermatomyositis. Scand. J. Rheum. 7:113.

179) Ahmed, F., V. Sumalnop, D.M. Spain and M.S. Tobin. 1978. Thrombohemolytic thrombocytopenic purpura during penicillamine therapy. Arch. Int. Med. 138:1292.

180) Corke, C.F. and E.C. Huskisson. 1978. Factors affecting the development of penicillamine side-effects. Rheum. Rehab. 17:34.

181) Camp, A.V. 1979. EULAR prospective trial on adverse reactions to penicillamine. Scand. J. Rheum., in press.

182) Panayi, G.S., P. Wooley and J.R. Batchelor. 1978. Genetic basis of rheumatoid disease: HLA antigens, disease manifestations and toxic reactions to drugs. Brit. Med. J. 2:1326.

183) Stastny, P. 1978. Association of the B-cell alloantigen DRw4 with rheumatoid arthritis. New Eng. J. Med. 298:869.

184) Multi-centre trial group. 1974. Absence of toxic or therapeutic interaction between penicillamine and previously administered gold in a trial of penicillamine in rheumatoid disease. Postgrad. Med. J. 50(Suppl 2):77.

185) Hill, H. 1978. Penicillamine and previous treatment with gold. Brit. Med. J. 2:961.

186) Youssef, S.A., E.F. Geever and S.M. Levenson. 1966. Penicillamine effect on wound healing. Surg. Forum. 17:89.

187) Geever, E.F., S. Youssef, E. Seifter and S.M. Levenson. 1967. Penicillamine and wound healing in young guinea pigs. J. Surg. Res. 7:160.

188) Burry, H.C. 1974. Penicillamine and wound healing - a potential hazard? Postgrad. Med. J. 50(Suppl 2):75.

189) Schorn, D. and A.G. Mowat. 1977. Penicillamine in rheumatoid arthritis: Wound healing, skin thickness and osteoporosis. Rheum. Rehab. 16:223.

190) Teigland, J. 1976. Rheumatoid surgery in penicillamine-treated patients. In Penicillamine Research in Rheumatoid Disease E. Munthe, editor, Fabritius and Sønner, Oslo, p112.

191) Lorber, A. 1966. Penicillamine therapy for rheumatoid lung disease: effects on protein sulfhydryl groups. Nature 210:1235.

192) Jaffe, I.A. and R.W. Smith. 1968. Rheumatoid vasculitis-report of a second case treated with penicillamine. Arth. Rheum. 11:585.

193) Lake, B. and G. Andrews. 1968. Rheumatoid arthritis with secondary amyloidosis and malabsorption syndrome. Effect of D-penicillamine. Amer. J. Med. 44:105.

194) Golding, J.R., J.V. Wilson, and A.T. Day. 1970. Observations on the treatment of rheumatoid disease with penicillamine. Postgrad. Med. J. 46:599.

195) Blau, S. and L. Meiselas. 1971 Regression of splenomegaly with hematologic recovery in Felty's syndrome due to D-penicillamine. A case report. Meadowbrook Hosp. J. 5:16.

196) Zer, I., I. Machtey and O. Kurz. 1973. Combined treatment of scleromalacia perforans in rheumatoid arthritis with penicillamine and plastic surgery. Ophthalmol. 166:293.

197) Jaffe, I.A. 1975. Penicillamine treatment of rheumatoid arthritis: Effect on immune complexes. Ann. N.Y. Acad. Sci. 256:330.

198) Mjølnerød, O.K., S.A. Dommerud, K. Rasmussen and G.T. Gjeruldsen. 1971. Congenital connective-tissue defect probably due to D-penicillamine treatment in pregnancy. Lancet 1:673.

199) Solomon, L., G. Abrams, M. Dinner and L. Berman. 1977. Neonatal abnormalities associated with D-penicillamine treatment during pregnancy. New Eng. J. Med. 296:54.

200) Laver, M. and K.F. Fairley. 1971. D-penicillamine treatment in pregnancy. Lancet 1:1019.

201) Scheinberg, I.H. and I. Sternlieb. 1975. Pregnancy in penicillamine-treated patients with Wilson's disease. New Eng. J. Med. 293:1300.

202) Walshe, J.M. 1977. Pregnancy in Wilson's disease. Quart. J. Med. 46:73.

203) Jaffe, I.A. 1965. The effect of penicillamine on the laboratory parameters in rheumatoid arthritis. Arth. Rheum. 8:1064.

204) Klamer, B., E.T. Kimura and M. Makstenieks. 1968. Effects of oral cysteine, penicillamine and N-acetyl-penicillamine on adjuvant arthritis in rats. Pharmacol. 1:283.

205) Liyanage, S.P. and H.L.F. Currey. 1972. Failure of oral D-penicillamine to modify adjuvant arthritis or immune response in the rat. Ann. Rheum. Dis. 31:521.

206) Willoughby, D.A. and P.A. Dieppe. 1976. The effects of D-penicillamine in animal models. In Penicillamine Research in Rheumatoid Disease. E. Munthe, editor, Fabritius and Sønner, Oslo, p45.

207) Sorenson, J.R.J. 1976. Copper chelates as possible active forms of the antiarthritic agents. J. Med. Chem. 19:135.

208) Arrigoni-Martelli, E., E. Bramm, E.C. Huskisson, D.A. Willoughby, and P.A. Dieppe. 1976. Pertussis vaccine oedema: An experimental model for the action of penicillamine-like drugs. Agents Actions. 6:613.

209) Whitehouse, M.W., L. Field, C.W. Denko, and R. Ryall. 1975. Is penicillamine a precursor drug? Scan. J. Rheum. 4(Suppl. 8):183.

210) Younes, M. and U. Weser. 1977. Superoxide dismutase activity of copper-penicillamine: Possible involvement of Cu(11) stabilized sulphur radical. Biochem. Biophys. Res. Comm. 78:1247.

211) Lengfelder, E. and E.F. Elstner. 1978. Determination of the superoxide dismutating activity of D-penicillamine copper. Hoppe-Seyler's Z. Physiol. Chem. 359:751.

212) Chwalinska-Sadowska, H. and J. Baum. 1976. The effect of D-penicillamine on polymorphonuclear leukocyte function. J. Clin. Invest. 58:871.

213) Mowat, A.G. 1978. Neutrophil chemotaxis in rheumatoid arthritis. Effect of D-penicillamine, gold salts and levamisole. Ann. Rheum. Dis. 37:1.

214) Zvaifler, N.J. 1973. The immunopathology of joint inflammation in rheumatoid arthritis. Adv. Immunol. 16:265.

215) Bluestone, R. and L.S. Goldberg. 1973. Effect of D-Penicillamine on serum immunoglobulins and rheumatoid factor. Ann. Rheum. Dis. 32:50.

216) Huskisson, E.C. and H. Berry. 1974. Some immunological changes in rheumatoid arthritis among patients receiving penicillamine and gold. Postgrad. Med. J. 50 (Suppl. 2):59.

217) Altman, K. and M.S. Tobin. 1965 Suppression of the primary immune response induced by D-L-penicillamine. Proc. Soc. Exp. Biol. Med. 118:554.

218) Hübner, K.F. and N. Gengozian. 1965. Depression of the primary immune response by D-L-penicillamine. Proc. Soc. Exp. Biol. Med. 118:561.

219) Tobin, M.S. and K. Altman. 1964. Accelerated immune response induced by D-L penicillamine. Proc. Soc. Exp. Biol. Med. 115:225.

220) Schumacher, K., G. Maerker-Alzer, and W. Schaaf. 1975. Influence of D-penicillamine on the immune response of mice. Arzneim-Forsch. (Drug Res.) 25:600.

221) Hunneyball, I.M., G.A. Stewart and D.R. Stanworth. 1978. The effects of oral D-penicillamine treatment on experimental arthritis and associated immune response in rabbits. I. Effect on humoral parameters. Immunol. 34:1053.

222) Hunneyball, I.M., G.A. Stewart and D.R. Stanworth. 1978. The effects of oral D-penicillamine treatment on experimental arthritis and associated immune response in rabbits. II. The effects on cellular parameters. Immunol. 35:159.

223) Lipsky, P.E. and M. Ziff. 1978. The effect of D-penicillamine on mitogen-induced human lymphocyte proliferation: synergistic inhibition by D-penicillamine and copper salts. J. Immunol. 120:1006.

224) Lipsky, P.E. and M. Ziff. 1979. Inhibition of human helper T cell function by D-penicillamine and copper sulfate. Arth. Rheum. in press.

225) Dumont, A.E., D.J. Mayer and J.H. Mulholland. 1964. The suppression of immunologic activity by diversion of thoracic duct lymph. Ann. Surg. 160:373.

226) Wegelius, O., V. Laine, B. Lindström and M. Klockars. 1970. Fistula of the thoracic duct as immunosuppressive treatment in rheumatoid arthritis. Acta Med. Scand. 187:539.

227) Paulus, H.E., H. Machleder, R. Bangert, J.A. Stratton, L. Goldberg, M.W. Whitehouse, D. Yu and C.M. Pearson. 1973. A case report: Thoracic duct lymphocyte drainage in rheumatoid arthritis. Clin. Immunol. Immunopath. 1:173.

228) Paulus, H.E., H.I. Machleder, S. Levine, D.T.Y. Yu and N.S. MacDonald. 1977. Lymphocyte involvement in rheumatoid arthritis. Studies during thoracic duct drainage. Arth. Rheum. 20:1249.