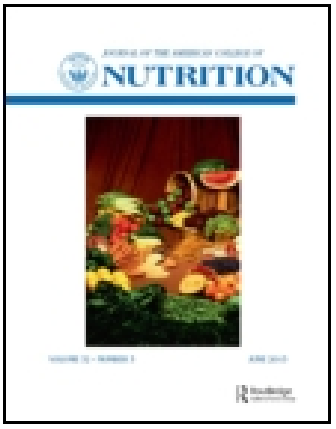

Journal of the American College of Nutrition

Publication details, including instructions for authors and subscription information:
http://www.tandfonline.com/loi/uacn20

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Published online: 02 Sep 2013.

To cite this article: M S Seelig (1982) Auto-immune complications of D-penicillamine—a possible result of zinc and magnesium depletion and of pyridoxine inactivation., Journal of the American College of Nutrition, 1:2, 207-214, DOI: 10.1080/07315724.1982.10718989

To link to this article: http://dx.doi.org/10.1080/07315724.1982.10718989

Journal of the American College of Nutrition 1:207-214 (1982)

Hypothesis

Auto-Immune Complications of D-Penicillamine – A Possible Result of Zinc and Magnesium Depletion and of Pyridoxine Inactivation

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Long-term high-dosage penicillamine treatment of patients with advanced stages of diseases with autoimmune components has resulted in very few adverse reactions in a series of over 50 such patients also given selected nutrients: pyridoxine, zinc and magnesium (which penicillamine inactivates or chelates), and vitamins B_1, B_{12}, and E (which have sulfhydryl-protective activity). The patients on this regimen have been essentially free of the side effects that occur in about a third of patients treated with penicillamine without such supplements. Reports of myasthenia gravis – a disease with abnormalities of the thymus and of T-cells, as a side effect of penicillamine – suggest that zinc, magnesium, and pyridoxine might be the agents most likely to be protective. Pyridoxine is necessary for cellular accumulation of zinc and magnesium, deficiencies of which have caused thymic and other immunologic abnormalities. Whether the other vitamins administered contribute to the favorable results requires further study.

Key words: autoimmune reactions, D-penicillamine, immunology, magnesium, zinc, pyridoxine, thymus, T cells, rheumatoid arthritis, Laennec cirrhosis

INTRODUCTION

D-penicillamine's effect on immunoregulation [1,2] is indicated by its activity against autoimmune diseases [3–5] and its paradoxical production of immunologically mediated side effects [1,2,5]. Depending on circumstances, it can either inhibit or augment lymphocyte transformation in vitro [2]. The development of myasthenia gravis during the course of administration of D-penicillamine, rare though it is [6–10], provides a clue to its immunologic toxicity. Thymic hyperplasia [6] and abnormal

Received December, 1981; accepted February, 1982.

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0731-5724/82/0102-0207$02.50

thymus-dependent lymphocytes (T cells) [2] have been reported in patients who developed this side effect. Similar changes have been described in spontaneous myasthenia gravis [11,12]. Thymic hyperplasia has also been seen in D-penicillamine toxicology studies [13]. The changes resemble those produced by experimental magnesium or zinc depletion [14–19] and those found in acrodermatitis enteropathica, a clinical zinc deficiency disorder [20]. Because penicillamine chelates zinc and magnesium [21] and because it inactivates pyridoxine [22], which is necessary for cellular uptake of zinc and magnesium [23–25], penicillamine's immunologic dysregulatory effects might be partially mediated by interference with utilization of these nutrients. In more than 50 patients with advanced chronic diseases (with autoimmune components) who were given selected nutrients before and during treatment with D-penicillamine in high dosages for prolonged periods, there was a very low incidence of adverse reactions [26–28]. We attribute this predominantly to protection against drug-induced deficiencies of magnesium, zinc, and pyridoxine.

METHODS

A combination of magnesium, zinc, and vitamins B_6, B_1, B_{12}, C, and E was given to patients with Laennec cirrhosis (because of underlying deficiencies and anticipated drug-induced losses) while awaiting approval for use of D-penicillamine [28]. This new application was to lower the elevated copper levels of cirrhosis [29], to apply its effect on collagen turnover [30] to hepatic fibrosis, and to treat the autoimmune component of cirrhosis [4]. The supplements were given orally three times a day with meals: vitamins B_6 (50 mg), B_1 (50 mg), B_{12} (25 μg), C (500 mg), and vitamin E (400 IU), zinc (10–15 mg as the gluconate) and magnesium (100 mg as an amino acid chelate). Magnesium sulfate (100 mg of Mg) was given intramuscularly once a week to those with evidence of deficiency, as determined by percentage retention of a test dose. Vitamin B_{12} was also given intramuscularly, (2,000 μg) once a week because of its demonstrated malabsorption in cirrhotic patients with pancreatic insufficiency [28,31]. Partial improvement of iron-refractory anemia, strength, and sense of well-being led us to continue the supplements when D-penicillamine treatment was started (given at least 1 hour apart from meals and supplements) 1–3 months after the nutrient regimen had been started [28]. There was early response to 250-mg and 500-mg daily doses of penicillamine for as long as 3 months at each dosage level, but cessation of improvement led to increase of dosage according to the accepted gradual incremental regimen [5]. A few patients required increases to as much as 2.75 g/day to sustain improvement. The same regimen was utilized in treatment of patients with multiple sclerosis in an attempt to obtain benefit from its activity in autoimmune disease and from its activity against neurotropic viruses [26], and with rheumatoid arthritis with and without vasculitis (unpublished observations) in order to obtain data with identical treatment programs. Hospital staff shortages necessitated reduction in the scope of our studies (in terms of number of patients and number of daily doses given) after 5 years. Among those remaining in the program, daily dosage was increased (in those whose dosage had been reduced to 250–500 mg/day) when prior gains were not sustained on lower doses. Patients given up to 500 mg of penicillamine daily are usually given one-third the total daily dosage of the supplements (at one meal). Several patients in another study of rheumatoid arthritis, who were not on the nutritional supplement program and who

were intolerant of penicillamine, had treatment cautiously restarted in combination with only tocopherol (I.A. Jaffe, personal communication). In addition to frequent evaluation for clinical changes, all patients have been monitored at least monthly throughout the treatment program for hematologic, urinary, and other side effects.

RESULTS

The patients with Laennec cirrhoss, who received the full therapeutic regimen, exhibited early correction of refractory anemia and diminution of hepatic pain and enlargement in those with concomitant hepatitis, as well as correction of inverted albumin/globulin ratio and needle liver biopsy evidence of less fibrosis [28]. Several of those with multiple sclerosis showed decreased spasticity and improved vision, mobility, and electroencephalograms during the full treatment phase of the study [26], which subsequently plateaued and showed no further improvement or some regression. A few with rheumatoid arthritis, who have been more recently entered into the study, have shown striking improvement. The improvement of the cirrhotic patients was sustained only on doses of 1–2.0 g/day (or higher in a few), with the exception of cirrhotic patients with encephalopathy or peripheral neuropathy, whose neurologic signs improved markedly on only 250 mg/day plus the nutrients. A patient with advanced active rheumatoid arthritis, who had improved while receiving 1.75 g/day, suffered an exacerbation when her penicillamine dosage was reduced to 250 mg/day. She showed gradual relief of pain, lowering of sedimentation rate, and improvement in anemia as the penicillamine dosage was gradually increased to (750 mg/day). Two, whose reactivation of advanced rheumatoid arthritis was not controlled by corticoids, have similarly improved on 500–750 mg of D-penicillamine plus supplements. Another, with severe rheumatoid arteritis and extensive necrosis of both legs, improved remarkably, with re-epithelialzation and diminished pain, as dosage was gradually increased to 1,250 mg daily (M.S. Seelig and A.R. Berger, in preparation).

Among the more than 50 patients in this series, most of whom were treated for 6 months to 7 years, only one or possibly two serious side effects developed, and there were few transitory discomforts [27]. Hematuria developed in one cirrhotic patient; cystoscopy disclosed hemorrhagic cystitis. Since a renal biopsy to rule out immunocomplex glomerulitis as a contributory factor was refused, penicillamine treatment (but not the nutritional supplements) was discontinued. The patient's signs of hepatic dysfunction recurred in the hospital. He left, returned to drink, and died in hepatic failure the following year. A multiple sclerosis patient, with a history of anaphylaxis to penicillin, developed frank albuminuria (1 g/day) while receiving 750 mg penicillamine/day as an outpatient. Treatment was stopped, and he was lost to further study. Several patients developed slight to moderate proteinuria (1 + to 3 +) at intervals, in association with urinary tract infections. Proteinuria subsided when the infection was controlled, without modification of penicillamine therapy. Hemorrhagic bullae over elbows and ecchymoses over legs and forearms developed in a cirrhotic patient while he was taking 2.75 g/day of penicillamine. The lesions cleared when the dosage was reduced to 500 mg/day and did not recur when clinical deterioration necessitated gradual increase of dosage to 1.75 g/day. Taste aberration or loss was encountered in two patients in their first month of treatment, before zinc supplementation had been

started. Transitory rashes were encountered in only two patients. No instance of thrombocytopenia or granulocytic leukopenia has been seen. Low pretreatment thrombocyte counts (in patients with Laennec cirrhosis or with multiple sclerosis), that necessitated very cautious institution of penicillamine therapy rose, during treatment. Low pretreatment hemoglobin and hematocrit levels also rose.

The rheumatoid arthritis patients who had been intolerant of D-penicillamine and had it restarted with tocopherol showed no improvement in tolerance of penicillamine (I.A. Jaffe, personal communication).

DISCUSSION

The unexpected occurrence of autoimmune disease complications in patients being treated with D-penicillamine [5–10], a drug with activity against autoimmune diseases [3–5,10,26,28], has led to many speculations and much study of its activity at the lymphocytic level [2,10] to elucidate the disparate findings. Thymic hyperplasia as a clinical [6] and experimental [13] toxicologic development, and the thymic and lymphocytic changes that are caused by depletion of two essential minerals namely, zinc and magnesium [14–20] that penicillamine chelates [21], suggest that the drug's first demonstrated clinical effect, as a chelator [32], might be responsible for the most serious problems associated with its use. Zinc is bound by penicillamine [21,33,34], which can cause retention [33,34] or loss of zinc [35,36], depending on whether it is given with or separately from the drug. Severe clinical manifestations of depletion developed in a child being treated for Wilson disease [35], and zinc levels were decreased in blood, brain, liver, and kidneys of mice given D-penicillamine [36]. Magnesium is low in the order of affinity of penicillamine for metals [21], but possibly when there is not an excess of trace or toxic minerals with greater affinity for the drug, even this macromineral can be removed. Preexistent magnesium inadequacy caused by cirrhosis or by drugs (such as diuretics or corticoids) might be intensified by penicillamine. The patients in this series have had marginally low serum magnesium levels while receiving penicillamine and have shown retention of most of parenterally administered test doses of magnesium (which is suggestive of deficiency), even though they all were being given magnesium supplements (100 mg three times daily), and several had received weekly intramuscular injections.

Inactivation of pyridoxine [22,36] has been demonstrated in rheumatoid arthritis patients being treated with penicillamine [22]; experimental vitamin B_6 deficiency causes cellular losses of both magnesium [23,24] and zinc [25], each of which is necessary for pyridoxine-dependent enzymes [37–39]. Cellular loss of magnesium and/or zinc might lead to aberrations of lymphoid tissue. Magnesium deficiency in rats has caused thymic hypertrophy [14–16] (or atrophy [16] under different conditions), lymphosarcoma, and leukemia [15,40] in certain strains. In mice, magnesium deficiency has reduced the number of antibody-synthesizing cells, thereby lowering immunoglobulin levels [39]. Optimal magnesium concentrations are required for normal in vitro lymphocyte transformation [42]. In rats, zinc deficiency causes inhibition of T-cell immunity [17], thymic atrophy and hypofunction [18], and impaired cell-mediated immunity [18]. Clinical zinc deficiency is associated with immunodeficiencies and T-cell dysfunction in infants [19,20] and thymic hypoplasia in calves [19,20] with acrodermatitis enteropathica, a disease that results from impaired intestinal zinc

absorption [20,43,44]. The effect of simultaneous deficiencies of magnesium, zinc, and pyridoxine, such as penicillamine can cause, has not been explored in animal models.

Few have investigated the effect of mineral supplements on the therapeutic or adverse effects of D-penicillamine. A mixture of minerals containing salts of magnesium, calcium, zinc, iron, and cobalt has been recommended to avoid depletion of essential minerals by the drug in Wilson disease patients [45]. The same supplement was given schizophrenic patients treated with penicillamine, with favorable results [46]. Improvement of schizophrenia with penicillamine was not confirmed by others who did not use the minerals [47]. It has been suggested that schizophrenics who improved on penicillamine plus minerals might have had pyroluria, a condition associated with zinc depletion, or kryptopyroluria (mauve factor), which is characterized by depletion of both zinc and pyridoxine and which improves somewhat on their administration [48]. Mineral supplements have been found to increase tolerance of high-dosage D-penicillamine in melanoma patients [49; and H.B. Demopoulos, personal communication].

The nutritional supplements given to the patients in this series [26–28] were selected to compensate for known penicillamine interference (with pyridoxine [22] and zinc [21,33–35]) and to substitute for the sulfhydryl-donor drug while we were awaiting clearance for administration of the drug, since increase in sulfhydryl levels might be a factor in therapeutic response to D-penicillamine [32,50,51]. Sulfydryl-protective vitamins—B_1 [52], B_{12} [53], E, and C [54,55]—were given at first with the intent of discontinuing them when penicillamine was started. A recent hypothesis, that autoimmune disorders might be mediated by free-radical-induced depression of T-cell suppressor function [56], suggests that the antioxidant activity of these vitamins, as well as that of D-penicillamine, might have played a role in the patients' improvement.

Subjective and hematologic improvement occurred while the patients were receiving the nutrients before penicillamine was started. Because vitamin assays disclosed persistently low levels, they were continued with penicillamine treatment. The same regimen was employed when new patients entered the study because of the absence of side effects, an incidence of 26–40% or more having been expected on the basis of reports of others [5,57,58]. Despite biochemical evidence of pyridoxine deficiency in rheumatoid arthritis patients on long-term penicillamine therapy [22], and of a disorder in pyridoxine metabolism in rheumatoid patients not receiving penicillamine [59], administration of only vitamin B_6 with penicillamine influenced neither the action of the drug nor its toxicity [5]. Zinc supplementation has corrected signs of severe zinc deficiency in a patient receiving penicillamine for Wilson disease [37], but its use has interfered with the therapeutic effects of low-dosage penicillamine in rheumatoid arthritis without preventing side effects [60]. In the present series, larger doses of zinc (30–45 mg/day) have not interfered with the therapeutic response to penicillamine in moderate to high doses (1–2.75 g/day) in patients with cirrhosis [28] or rheumatoid arthritis (unpublished observations).

Whether the vitamins that were provided in addition to pyridoxine,—which with magnesium and zinc are probably the most important components of the supplement mixture, contribute to the low incidence of side effects remains to be ascertained. Vitamin B_1 and magnesium activities are interdependent [61,62]. Magnesium deficiency intensifies some of the signs of vitamin E deficiency [63]. Vitamin E, magnesium, and zinc are each involved in maintaining membrane stability, and they and other vitamins and minerals play roles in immunosurveillance [64].

CONCLUSIONS

The most serious adverse reactions caused by D-penicillamine are those that develop late and that reflect its dysregulation of the immune system. The lack of such effects in a series of patients given high-dosage penicillamine might be the result of protecting against losses of cellular zinc and magnesium, depletions of which are associated with thymic and T-cell abnormalities.

ACKNOWLEDGMENTS

Appreciation is expressed to Herman Baker and Oscar Frank for vitamin assays and to Israeli A. Jaffe for unpublished data and for alerting me to the work demonstrating penicillamine's immunodysregulatory effects.

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