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Cyclophosphamide Therapy of Idiopathic Nephrosis

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• *Thirty-three children with steroid-dependent or steroid-resistant idiopathic nephrosis were treated with a combination of cyclophosphamide and prednisone. Remission occurred in all 23 steroid-dependent patients and, at last report, 12 had gone without any medication for periods of from two months to 21 months without relapse. Remission occurred in five of ten steroid-resistant patients.*

The renal biopsy findings in these patients demonstrate a correlation between the "minimal change" lesion and a positive response to cyclophosphamide.

FOR THE PAST 15 years, ACTH or corticosteroids have been the primary therapy for idiopathic nephrosis of childhood. Corticosteroids have resulted in a significant improvement in the morbidity and short-term mortality from this disease.[1,2,3] There has emerged, however, a significant number of patients, who are controlled by corticosteroid therapy but have relapse upon termination of drug therapy. Continuous or intermittent therapy must be used to keep these children free of symptoms and their urine free of protein. The term "steroid-dependent" has been used to categorize these patients. These patients are distinct from a smaller group of children whose disease is completely resistant to corticosteroids. In the early 1960's several investigators began exploring the use of immunosuppressive therapy as an alternative therapy for these two groups of patients.[4-8] The results of the past few years have shown that this form of therapy seems to be effective in both the steroid-dependent and, to a lesser degree, the steroid-resistant patients.[7] In August, 1967, we began using cyclophosphamide in the treatment of steroid-dependent and steroid-resistant idiopathic nephrosis. This paper presents our experience in 33 patients.

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TABLE 1.—*Growth retardation caused by corticosteroid therapy in 12 children with idiopathic nephrosis, and subsequent catch-up growth following cyclophosphamide therapy*

Patient No.	Age at Time Cyclophosphamide Begun (Years)	Total Duration of Steroid Therapy (Years)	Height (Percentile)	Total Length of Remission (Months)	Expected* Growth (Inches)	Actual Growth (Inches)
1	14	> 10	< 3rd	17+	1.2	3.5
2	12	10	< 3rd	11+	1.6	3.0
3	17	10	< 3rd	16+	.2	0.5
4	12	10	25th	11+	1.7	3.0
5	12.5	> 10	10th	3+	.4	1.0
6	11	6	< 3rd	12+	2.2	3.5
7	10	6	< 3rd	12+	1.8	4.5
8	14.5	9	< 3rd	11+	2.1	4.5
9	10.5	9	< 3rd	12+	2.1	5.5
10	9	7	50th	12+	1.9	3.75
11	5.75	5	< 3rd	7+	1.2	2.0
12	7	7	< 3rd	7+	1.2	2.0

*Reference Standard: Howard V. Meredith in Textbook of Pediatrics, Editor, W. E. Nelson, 8th Edition, W. B. Saunders, Philadelphia, 1964, pp 50-53.

Materials and Methods

Between August 1967 and May 1969, 33 children aged 18 months to 17 years with idiopathic nephrosis characterized by proteinuria, hypoalbuminemia, hyperlipemia and edema were treated with cyclophosphamide and prednisone.

None had hematuria nor hypertension. All had normal blood urea nitrogen levels. Twenty-three patients exhibited steroid dependency, which we define as five or more relapses or the necessity for continuous steroid medication to prevent recurrence of proteinuria over a period of 12 to 18 months. The remaining ten patients were steroid-resistant, with persistent proteinuria (more than 1 gram in 24 hours) after 28 days of daily prednisone therapy at a dose of 75 mg per square meter of body surface per day. No patient was hypocomplementemic.

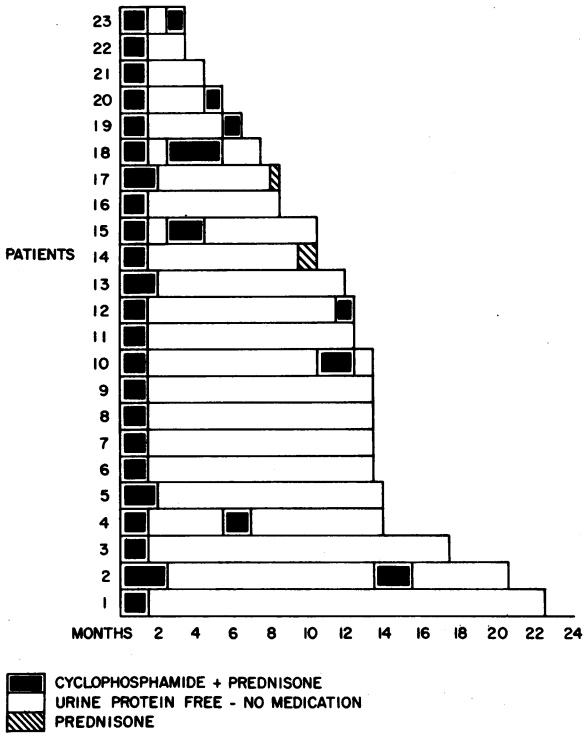

Chart 1.—Response to Cyclophosphamide in Steroid-Dependent Patients.

The dose of cyclophosphamide was 5 mg per Kg of body weight per day to a maximum of 150 mg per day continued for a period of six weeks after proteinuria had cleared. During this time prednisone was continued at a dose of 50 to 75 mg per square meter per day given on alternate days. The white blood cell count was measured two to three times a week. The dose of cyclophosphamide was halved if the white blood cell count fell below 5000 per cu mm and the drug was discontinued if the white blood cell count was less than 4000 per cu mm. Cyclophosphamide therapy was reinstituted when the white blood cell count rose to 4000 per cu mm. Nine patients received two courses of cyclophosphamide. A patient was considered resistant to cyclophosphamide if significant proteinuria (more than 1 gram in 24 hours) continued after two months of daily therapy. Renal biopsy specimens were obtained before cyclophosphamide therapy was begun in 21 of 23 steroid-dependent children and in all ten steroid-resistant patients. Kidney sections were stained, using hematoxylin and eosin, periodic acid/Schiff, trichrome, and Jones–methenamine–silver, and examined by light microscopy. Immunofluores-

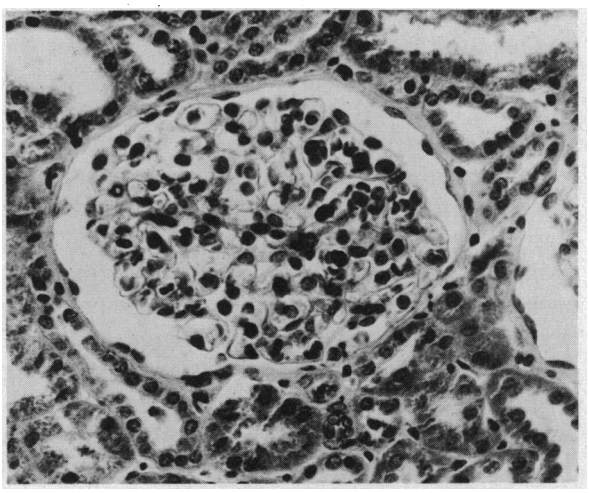

Figure 1.—Representative photomicrograph showing biopsy with "minimal change" lesion.

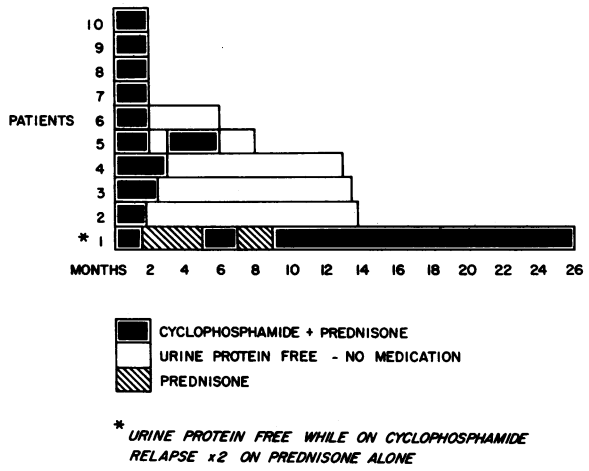

Chart 2.—Response to Cyclophosphamide in Steroid-Resistant Patients.

cent biopsy studies were carried out on 19 of the 21 specimens from steroid-dependent children and on seven of the ten from steroid-resistant children.

Results

Steroid-Dependent Patients

Remission, which we define as absence of proteinuria after cyclophosphamide and prednisone were discontinued, occurred in all 23 steroid-dependent patients. The response to cyclophosphamide in these patients is illustrated in Chart 1. At the time of this report, 12 of 23 patients had been in remission for from two to twenty-one months after therapy was ended. Of the 11 patients who had relapse, five had been in remission from six to 12 months and the remaining six for less than four months. In all the 21 cases in which renal biopsy was done, it showed the typical "minimal change" lesion of idiopathic nephrosis[9] (Figure 1).

Further analysis of this group (Table 1) revealed five patients had been treated with intermittent or continuous corticosteroid therapy for ten or more years and seven had been treated for periods of five to nine years before treatment with cyclophosphamide. These patients had many of the effects of long-term corticosteroid therapy, especially retarded linear growth. Following cessation of cyclophosphamide and corticosteroid therapy each child in this group had a growth spurt (defined as a height increase greater than that expected for a child of equivalent age and height percentile[10]) during the follow-up period.

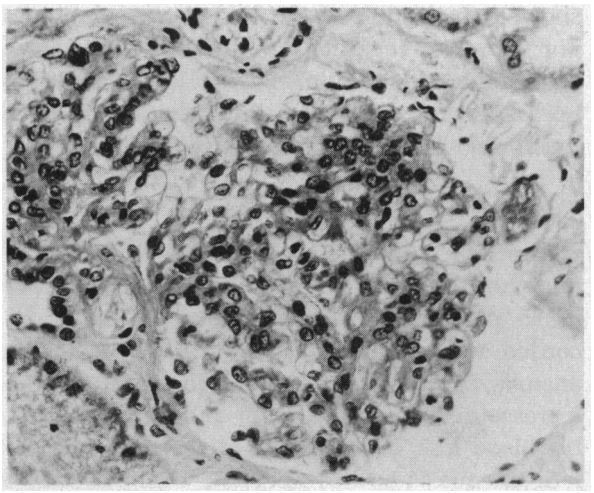

Figure 2.—Photomicrograph of renal biopsy specimen showing proliferative glomerular changes.

Steroid-Resistant Patients

Remission occurred in five of ten steroid-resistant patients (Chart 2). One subsequently had relapse; he received a second course of cyclophosphamide and, at the time of this report, had gone without all therapy for two months. Four patients were resistant to cyclophosphamide. The tenth patient responded to cyclophosphamide with clearance of proteinuria; however, proteinuria recurred upon discontinuance of the drug. A second course was given with the same result. He has been termed "cyclophosphamide-dependent" and is now on a protracted course of cyclophosphamide. In the five patients who had remittance on cyclophosphamide and on the one "cyclophosphamide dependent" patient renal biopsy showed

TABLE 2.—*Side effects of cyclophosphamide therapy in 33 patients*

	Number of Patients Affected
HEMATOLOGIC	
Leukopenia (< 4000 per mm³)	25
Anemia (<10 Gm%)	1
ALOPECIA	27
HEMORRHAGIC CYSTITIS	1
GASTROINTESTINAL	
Nausea or vomiting	11

no significant proliferation or inflammation and the changes were consistent with a "minimal change" lesion. Renal biopsy in the patients who did not respond to cyclophosphamide showed significant proliferative or exudative glomerular changes or both (Figure 2).

None of the biopsy material in either group showed thickening of the basement membranes nor the silver-negative "humps" characteristic of membranous glomerulonephritis.[11] Also there was no fraying of capillary walls characteristic of membrano-proliferative glomerulonephritis.[12]

Toxicity

The side effects attributed to cyclophosphamide are listed in Table 2. Leukopenia occurred in 25 patients; the lowest leukocyte count recorded was 1500 per cu mm. Mild hemolytic anemia, as manifested by a fall in hematocrit, an increase in reticulocyte count and no demonstrable blood loss, developed in one patient. Hemolytic anemia is not a recognized complication of cyclophosphamide therapy; the patient was receiving several other medications including spironolactone (Aldactone®) and chlorothiazide (Diuril®) at this time. In no case did thrombocytopenia, agranulocytosis or aplastic anemia develop. Alopecia, mild to almost complete, occurred in 27 cases; and in all instances the hair grew back. Mild hemorrhagic cystitis developed in one case, whereupon cyclophosphamide was discontinued for three days and then resumed without difficulty. No serious infections occurred in any patient. Approximately one-third of the group had occasional mild anorexia and occasional emesis while taking the drug. No patient showed signs of hepatotoxicity.

Discussion

Immunosuppressive therapy in the form of nitrogen mustard was first used for the treatment of the nephrotic syndrome as early as 1950.[13] Its use at that time was based upon the belief that the nephrotic syndrome was an immunologic disease. The drug appeared to be effective but was discontinued because of severe side effects. Corticosteroids then became the principal agent of therapy. Interest in the use of immunosuppressive therapy was again awakened with the recognition of a group of children who did not have adequate response to corticosteroids. In small groups of patients, Coldbeck,[4] West et al,[5] Grupe and Heymann[6] and Drummond et al[14] reported promising results with the use of cyclophosphamide. Etteldorf et al[7] treated 14 children successfully with cyclophosphamide and Travis and Dodge[8] reported their results in 35 patients. The largest and most recent series published to date is that of Moncrieff et al[15] in which 46 children with the nephrotic syndrome were treated with cyclophosphamide and corticosteroid therapy. Three of these children were steroid-resistant; the remainder were steroid-dependent. In all cases biopsy showed the "minimal change" lesion. Thirty-three of the 43 (77 percent) steroid-dependent children were in remission for from two to twenty-three months after one course of cyclophosphamide. Of the remaining patients, two died of pneumonia and adrenal failure and four remained in relapse. None of the three steroid-resistant patients responded to cyclophosphamide. Our series compares favorably with that of Moncrieff in that 12 of the 23 steroid-dependent patients had been in remission at the time of this report, for from two to twenty-one months after one course of cyclophosphamide. Of the 11 patients who had relapsed, three had been in remission for one year before relapse occurred and five again had remission following completion of a second course of therapy with cyclophosphamide. In the steroid-resistant group five of the ten patients treated with cyclophosphamide responded to therapy and were in remission at the time of this report. In four cases proteinuria continued. The tenth patient appears to be "cyclophosphamide dependent."

There are two major reasons why we strongly urge the use of cyclophosphamide in patients with steroid-dependent or steroid-resistant nephrosis. First, these patients have the highest morbidity and mortality of children with nephrosis. Recent large series such as those of Arneil and Lam[1] and Cornfeld and Schwartz[2] indicated that the ulti-

mate mortality rate of children with the nephrotic syndrome is in the range of 30 percent. This figure includes patients who are either steroid-resistant or steroid-dependent and must be maintained on corticosteroids for long periods. These patients are susceptible to all the severe side effects of corticosteroids, especially risk of infection, thrombosis and adrenal insufficiency.[16,17] While there is a risk in the use of cyclophosphamide and death has occurred in at least three patients who contracted varicella or rubeola,[7,18,19] we believe that this risk must be weighed against the ultimate prognosis in steroid-dependent and steroid-resistant nephrotic patients.

The second major objective in adopting immunosuppressive therapy for childhood nephrosis was to attempt to discontinue all medication for a long enough period to provide catch-up growth. Nine of the 12 steroid-dependent children in our series who had been receiving therapy for more than five years had severe growth retardation (were below the third percentile in height[10]). Initially these patients had been treated on a schedule of four days a week on and three days a week off corticosteroid therapy. In recent years this schedule has been changed to alternate days in all patients. We are impressed by the growth spurt ($P>0.01$) that occurred in all patients following cessation of corticosteroid therapy after cyclophosphamide treatment. In some patients the height increase was two to three times that expected. Of great significance is the fact that one patient (patient 3 in Table 1) grew only 0.5 inches in 16 months. This slowness suggests that epiphyseal closure already had occurred. We believe it is important to use cyclophosphamide in a steroid-dependent nephrotic child before epiphyseal closure prevents any chance for significant catch-up growth.

The renal biopsy findings in this series demonstrated an apparent correlation between the "minimal change" lesion and response to cyclophosphamide. All the steroid-dependent patients who had biopsy had "minimal change" lesions and in all of them remission was induced with cyclophosphamide. Furthermore the renal changes noted on biopsy in the five steroid-resistant patients who had remission when treated with cyclophosphamide were relatively mild and not inconsistent with a "minimal change" lesion. The patients who were resistant to both corticosteroids and cyclophosphamide showed significant proliferative or exudative glomerular changes although clinically there was no observable difference between them and the responsive groups.

As far as can be determined from the available light and fluorescent biopsy material examined, the cyclophosphamide-resistant patients did not exhibit early membranous glomerulonephritis.

The results of this report indicate that children with steroid-dependent and steroid-resistant nephrosis should be treated with cyclophosphamide. While cyclophosphamide appears to have at least partially altered the morbidity of idiopathic nephrosis, further follow-up is necessary to determine its long-term effects on this disease.

TRADE AND GENERIC NAMES OF DRUGS

Aldactone® ..... spironolactone
Diuril® ..... chlorothiazide

REFERENCES

1. Arneil GC, Lam CH: Long-term assessment of steroid therapy in childhood nephrosis. Lancet 2:189, 1966
2. Cornfeld D, Schwartz MW: Nephrosis, a long-term study of children treated with corticosteroids. J Pediat 68:507, 1966
3. Van Acker KJ, Hooft C: The influence of hormone treatment on the natural evolution of the idiopathic nephrotic syndrome in childhood. Acta Paediat Scand 57:479-486, 1968
4. Coldbeck JH: Experience with alkylating agents in the treatment of children with the nephrotic syndrome. Med J Aust 2:987, 1963
5. West CD, Hong R, Holland NH: Effect of cyclophosphamide on lipoid nephrosis in the human and on aminonucleoside nephrosis in the rat. J Pediat 68:516-524, 1966
6. Grupe WE, Heymann W: Cytotoxic drugs in steroid-resistant renal disease. Amer J Dis Child 112:448, 1966
7. Etteldorf JN, Roy S, Summitt RL, et al: Cyclophosphamide in the treatment of idiopathic lipoid nephrosis. J Pediat 70:758, 1967
8. Travis LB, Dodge WF: Evaluation of combined therapy (steroids and cyclophosphamide) in the steroid resistant (SR) and steroid dependent (SD) nephrotic syndrome. Abstract, Society for Pediatric Research, Atlantic City, 1967
9. Churg J, Habib R, White RHR: Pathology of the nephrotic syndrome in children. Lancet 1:1299, 1970
10. Meredith HV: Textbook of Pediatrics (8th Edition), *In* Nelson WE (Ed). Philadelphia, WB Saunders Co, 1964, pp 50-53
11. Ehrenreich T, Churg J: Pathology of membranous nephropathy, *In* Sommers SC (Ed): Pathology Annual. New York, Appleton-Century-Crofts Inc., 1968, pp 145-186
12. Northway JD, McAdams AJ, Forristal J, et al: A "silent" phase of hypocomplementemic persistent nephritis detectable by reduced serum B 1c- globulin levels. J Pediat 74:28-38, 1969
13. Chasis H, Goldring W, Baldwin DS: Effect of nitrogen mustard on renal manifestations of human glomerulonephritis. J Clin Invest 29:804, 1950
14. Drummond KN, Hillman DA, Marchessault JH, et al: Cyclophosphamide in the nephrotic syndrome of childhood. Canad Med Assoc J 98:524, 1968
15. Moncrieff MW, White RHR, Ogg CS, et al: Cyclophosphamide therapy in the nephrotic syndrome in childhood. Brit Med J 1:666, 1969
16. Lieberman E, Heuser E, Gilchrist GS, et al: Thrombosis, nephrosis and corticosteroid therapy. J Pediat 73:320, 1968
17. Good RA, Vernier RL, Smith RT: Serious untoward reactions to therapy with cortisone and adrenocorticotrophin in pediatric practice. Pediatrics (Part I) 19:95, 1957 and Part II 19:272, 1957
18. Scheinman JI, Stamler FW: Cyclophosphamide and fatal varicella. J Pediat 74:117, 1969
19. Meadow SR, Weller RO, Archibald RWR: Fatal systemic measles in a child receiving cyclophosphamide for the nephrotic syndrome. Lancet 2:876, 1969