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A protein in urine associated with muscle disease and muscle damage

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Abstract

Analysis of the protein composition of human urine by high-resolution two-dimensional electrophoresis showed that several features are associated with neuromuscular diseases, the best defined being the appearance in the urine of a small amount of a protein that migrates on the electropherogram as a characteristic spot (spot C). This spot consists of a protein of apparent molecular weight 26 000 and isoelectric point 5.3. The spot was usually present in the urine of patients suffering from diseases in which the musculature was directly affected but was rarely found in other patients and normal subjects.

The protein responsible for spot C appears to be an index of muscle damage caused by a number of conditions. Attempts are being made to isolate enough of the protein to permit its identification.

Introduction

The analysis of the protein composition of human urine by high-resolution two-dimensional electrophoresis provides a highly sensitive index of the functional state of an individual.^{1,2} By applying this technique to patients with neuromuscular diseases we have observed several features in the urinary protein pattern that are associated with these diseases.¹ The best defined

and most consistent feature is the appearance in the urine of a small amount of a protein that migrates on the electropherogram as a characteristic spot; we have designated this spot C.

The protein and its prevalence

From its electrophoretic behaviour spot C consists of a protein of apparent molecular weight 26 000 and isoelectric point 5.3.

We studied the prevalence of spot C in electropherograms of urine from 107 normal subjects and 130 patients with various neuromuscular diseases. Spot C was most pronounced and was invariably observed on electropherograms of the urine of boys with Duchenne muscular dystrophy, who were estimated to excrete about 100 µg a day (table). It was also usually present in the urine of patients suffering from limb-girdle dystrophy, spinal muscular atrophies, and dystrophias myotonica. Spot C was not observed in the urine of patients partially immobilised by other disabilities such as spina bifida, cerebral palsy, and congenital deformity, in which the musculature is not directly affected. In preliminary studies we did not detect the protein responsible for spot C in the urine of patients with myasthenia gravis and multiple sclerosis.

Discussion

We believe that the presence of the protein responsible for spot C in urine is an index of muscle damage produced by a number of conditions and not the direct product of a modified gene associated with a particular neuromuscular disease. The protein is probably present in normal muscle, although the evidence for this is so far circumstantial for we have not been able to show its presence by electrophoresis of extracts of this tissue and have not detected it in the serum of normal people or patients with Duchenne muscular dystrophy. It was detected in the urine of normal men for a day or two after a bout of vigorous exercise. It was also observed in the urine of patients with a history of circu-

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latory insufficiency—for example, after myocardial infarction or in cases of arterial insufficiency of a leg.

Spot C was detected in about 60% of the urine samples from carriers of Duchenne muscular dystrophy. Only rarely (4% of cases) were traces of it seen in the electropherograms of urine from normal subjects (table). In these cases the presence of the protein in the urine may possibly have been due to mild muscle damage from exercise, sprains, or falls, etc.

The protein responsible for spot C does not appear to be identical with any of the muscle enzymes known to be present in

Prevalence of spot C in electropherograms of urine from normal subjects and patients with neuromuscular diseases

	Age (years)		Total No studied	No with spot C
	Range	Mean		
<i>Normal subjects</i>				
Adult women	18-55	27.5	34	2
Pregnant women	18-33		24	0
Adult men	20-35	28.2	25	0
Boys	6-18	10.0	26	2
<i>Disabled people</i>				
Male patients	6-39	18.6	14	1
<i>Patients with Duchenne muscular dystrophy</i>				
Boys	6-22	17.2	31	31
Mothers:				
Obligatory carriers	39-57		21	12
Isolated cases	23-45	36.0	11	3
Sisters	11-25	17.1	10	5
<i>Patients with other neuromuscular diseases</i>				
Limb-girdle muscular dystrophy			2	2
Werdnig Hoffman paralysis	14-21	18	3	2
Spinal muscular atrophy	11-19		2	2
Peroneal muscular atrophy*			5	3-4
Dystrophia myotonica	15-62	39.0	12	9
Myasthenia gravis	16-61	43.8	10	0
Multiple sclerosis	29-64	46.7	9	0

*Including Charcot-Marie-Tooth atrophy.

the serum of patients with muscular dystrophy. In this respect it is interesting that no spot corresponding to creatine kinase could be detected on electropherograms of the urine of these patients, even though spot C was clearly identifiable. On the other hand, unlike spot C, creatine kinase was clearly visible as a protein-staining spot on electropherograms of serum from these patients.

Our studies thus indicate that the presence in the urine of protein migrating as spot C is characteristic of diseases in which the muscle is clearly affected. The detection of this protein in urine, particularly when methods for its radioimmune assay have been developed, might be a useful sensitive aid in confirming the diagnosis of muscle disease and under certain conditions in assessing muscle damage. It might also have a role in the detection of the carrier state in Duchenne muscular dystrophy. We are now attempting to isolate the protein from the urine of boys with Duchenne muscular dystrophy in sufficient amounts to permit its identification.

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Haemolytic anaemia after cisplatin treatment

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Abstract

Normochromic or normocytic anaemia is a common side effect of treatment with cisplatin. Two patients treated with cisplatin 100 mg/m² in combination with vinblastine, bleomycin, and actinomycin D developed haemolytic anaemia. Neither patient had evidence of haemolysis before treatment, and in both cases severe haemolytic anaemia developed after several courses of cisplatin and when the cancer had regressed almost completely.

The importance of haemolysis in the development of anaemia after cisplatin treatment has not been investigated fully and further studies are needed.

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Introduction

cis-Dichlorodiammineplatinum (II) (cisplatin) is a cancer chemotherapeutic agent of value in several types of tumour.¹ Major side effects reported with cisplatin include gastrointestinal and renal toxicities, and ototoxicity²; anaemia has been reported in 9-40% of patients treated with cisplatin.² The anaemia is usually normochromic and normocytic in type and considered to be secondary to depression of erythropoiesis, though adequate studies to clarify the mechanisms responsible have not been reported. We report two cases of haemolytic anaemia occurring after cisplatin treatment.

Case reports

Case 1—A 37-year-old man with metastatic mixed embryonal cell carcinoma and teratocarcinoma of the testis affecting lungs and retroperitoneal lymph nodes started chemotherapy with cisplatin 100 mg/m² every three weeks in combination with vinblastine, bleomycin, and actinomycin D on 7 September 1978. His initial haemoglobin concentration was 16.3 g/dl. There was virtually complete regression of all metastatic disease over the next nine weeks. After the third course of cisplatin (total dose 503 mg) his haemoglobin