Metabolic Abnormalities Associated With Skeletal Myopathy in Severe Anorexia Nervosa

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The aim of this study was to characterize the metabolic disturbance associated with the skeletal myopathy resulting from extreme weight loss in anorexia nervosa. Muscle function was examined in eight female patients with severe (40%) weight loss due to anorexia nervosa and histologically confirmed myopathy. A wide range of biochemical and hematologic investigations were carried out, including serum enzymes and the response of plasma lactate to ischemic exercise of forearm muscles. All patients showed proximal muscular weakness. A diminished lactate response to ischemic exercise was a consistent finding, and a reduction of serum carnosinase activity was also found. There were no other consistent biochemical or hematologic abnormalities apart from lymphopenia of no clinical consequence. These findings contribute to our understanding of severe protein-energy malnutrition on the musculoskeletal system. The resulting disorder is a metabolic myopathy from which the patients recover rapidly as their nutrition improves. Although the patients admitted to a variety of abnormal eating behaviors, no correlation was found between a specific type of abnormal eating behavior and subsequent biochemical abnormalities. Reinstating appropriate eating behavior will treat the myopathy. Nutrition 2000;16:192–196. ©Elsevier Science Inc. 2000

Key words: anorexia nervosa, myopathy, lactate response to exercise, carnosinase, type 2 fiber atrophy

INTRODUCTION

Anorexia nervosa (AN) is an important cause of a wide range of physical morbidity.1–3 It occurs most frequently in adolescent and young adult females in whom the prevalence may be as high as 1%,4 whereas males account for fewer than 10% of patients.5 The diagnostic criteria of AN are fully described by the American Psychiatric Association in the fourth edition of the Diagnostic and Statistical Manual of Mental Disorders (DSM-IV)6 but have also been outlined more briefly.7 Self-induced weight loss is a key feature of AN, and in the postpubertal female it is typically associated with amenorrhoea. The patients’ psychological preoccupations are best summarized as a morbid fear of fatness. Anorexic patients want to lose weight by avoiding nutritious foods. They tend to conceal this behavior by euphemisms: “being on a diet” or “being vegetarian.” Vegetarianism can be an excuse for adhering rigidly to a low-calorie diet consisting of vegetables and fruit. Patients who consume large amounts of carrots or tomatoes may develop carotinemia, with an orange coloration of the face and hands. The malnutrition may be further complicated by behaviors aimed at additional reduction of body weight: self-induced vomiting; abuse of laxatives, emetics, diuretics, appetite suppressants, or L-thyroxine; and excessive exercise to burn up calories. AN often runs a protracted course, and, in severe cases, the crude mortality rate is high—up to 18% in series of patients followed-up for 20 y.8,9

It may be surprising that anorexic patients seldom appear to suffer from vitamin deficiencies. In the female, during the early stages of weight loss, the negative energy balance leads to a depletion mainly of fatty tissues. An estimation of the composition of the tissue restored while patients are being refed during treatment showed that most of the weight gain was due to fat (77%), although there was a small degree of protein synthesis (7%) and some water retention (16%).10 In very severe cases there occurs protein-energy malnutrition.11 Refeeding studies in male patients with AN have demonstrated that protein synthesis is greater than in female patients.12 In both sexes not only is there loss of body fat but eventually skeletal muscle mass is reduced.13 In spite of their emaciation, anorexic patients often persist with overexercising.14 Usually only in the late stages do patients admit to symptoms of fatigue and muscular weakness. This weakness, usually proximal in distribution,15 is often attributed to hypokalemia secondary to vomiting or abuse of laxatives and diuretics. However, the most common cause is a skeletal muscle myopathy associated with type 2 muscle fiber atrophy.13,15,16

Despite awareness that myopathy may occur in AN, there have been relatively few studies of this complication. We recently reported a series of patients with AN and extreme weight loss who had skeletal myopathy characterized by severe type 2 fiber atrophy, in the absence of neurogenic change, plus the presence of abnormal accumulations of glycogen within muscle fibers and loss of contractile elements.17 The aims of the present study were to carry out a biochemical assessment of muscle function and to ascertain whether the skeletal myopathy in AN is associated with disturbances of serum enzymes or other biochemical changes.
PATIENTS AND METHODS

Patients

Eight female patients admitted consecutively to the Eating Disorders Unit of the Maudsley Hospital were enrolled in the study. Their mean (SD) age was 24.0 (3.0) y. The major criterion for admission to the unit was continuing life-threatening weight loss that had proved refractory to outpatient management. All patients met the DSM-IV diagnostic criteria for AN, and none had any intercurrent illness. The Hospital Ethics Committee approved the study, and all patients gave informed written consent. The following investigations were performed within a week of admission to establish baseline data.

Clinical Assessment

Clinical assessment was carried out on all patients by the same examiner (D.M.M.) and included a detailed history of abnormal eating behaviors and symptoms of myopathy (weakness, fatiguability, and pain), a full physical examination, and anthropometric assessments of weight, height, and skinfold thicknesses at triceps, biceps, and subscapular sites.

Hematologic and Biochemical Investigations

Blood samples were collected for hematologic and biochemical assays. All subjects had assays for full blood count and white cell differential count performed on a Coulter JS-plus analyzer (Coulter Electronics Ltd., Beds, UK). Serum iron, ferritin, vitamin B12, and folic acid were analyzed by standard techniques.

Urea, electrolytes, and glucose were measured on an Astra 4 analyzer (Beckman Instruments Ltd, Bucks, UK). Calcium, magnesium, iron, phosphate, creatinine, total protein, albumin, and bilirubin were measured with the Cobas Fara analyzer. In addition, endocrine profile was assayed by standard methods and included growth hormone (GH), luteinizing hormone (LH), folliclestimulating hormone (FSH), free T4 (FT4), and thyroid-stimulating hormone (TSH). Laboratory performances met the criteria of external quality-assessment schemes throughout the period of the study.

Biochemical Assessment of Muscle Function

The response of the forearm muscles to ischemic exercise was measured.19,20 After the collection of a resting blood sample, a blood-pressure cuff was inflated around the upper arm to 20 mmHg above systolic pressure to occlude arterial blood flow. Subjects performed ischemic exercise for 1 min, during which time they were actively encouraged to persist in repeatedly squeezing their hand despite any discomfort incurred. The cuff was released after exercise, and venous blood draining from the forearm muscles was sampled at 1, 3, 6, and 10 min for the measurement of blood lactate. Lactate was assayed on the Yellow Spring Instrument Analyser using an immobilized enzyme (l-lactate) membrane and sensor probe system. In normal subjects, there is a three- to five-fold increase in plasma lactate concentration in the 1-min sample after ischemic forearm exercise and a gradual decline in lactate concentrations thereafter.19–21

Statistical Analysis

All data are expressed as mean (SD). Differences between the means of groups were analyzed by unpaired Student’s t tests using two-tailed tables. Differences were considered significant when \( P < 0.05 \). Comparisons were made between vegetarian and non-vegetarian patients; the presence or absence of subjective fatigue; and between overexercising and non-overexercising groups.

RESULTS

Clinical Assessment

The patients’ clinical details have been previously reported in detail17 and are summarized in Table I. The mean age of onset of

<table>
<thead>
<tr>
<th>Patient number</th>
<th>Age (y)</th>
<th>Duration of AN (y)</th>
<th>BMI</th>
<th>Methods of weight loss</th>
<th>Clinical features</th>
</tr>
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<tbody>
<tr>
<td>1</td>
<td>27</td>
<td>13.0</td>
<td>11.5</td>
<td>D, V, L, OE</td>
<td>W, F, P, R</td>
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<td>2</td>
<td>25</td>
<td>4.0</td>
<td>12.9</td>
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</tr>
<tr>
<td>3</td>
<td>26</td>
<td>13.0</td>
<td>12.6</td>
<td>D, OE</td>
<td>W, P</td>
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<tr>
<td>4</td>
<td>28</td>
<td>10.0</td>
<td>12.7</td>
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<td>13.3</td>
<td>D</td>
<td>W</td>
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<td>21</td>
<td>4.0</td>
<td>11.6</td>
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<td>W, F</td>
</tr>
<tr>
<td>7</td>
<td>21</td>
<td>6.0</td>
<td>12.8</td>
<td>D, V, OE</td>
<td>W, F, R</td>
</tr>
<tr>
<td>8</td>
<td>24</td>
<td>2.5</td>
<td>13.5</td>
<td>D, SIV</td>
<td>W, F, P, R</td>
</tr>
<tr>
<td>Mean (SD)</td>
<td>24 (3)</td>
<td>6.8 (4.6)</td>
<td>12.6 (0.7)</td>
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</tbody>
</table>

AN, anorexia nervosa; BMI, body mass index; D, dietary restriction; F, subjective fatiguability; L, laxative abuse; OE, overexercising; P, muscular pain; R, reduction/absence of tendon reflexes; SIV, self-induced vomiting; V, vegetarian; W, muscle weakness.
AN was 17.3 (3.1) y, and its mean duration was 6.8 (4.6) y. All patients had severely restricted their food intake right up to the day of admission. Five had become vegetarians. Patient numbers 1, 3, and 6 were taking oral multivitamin preparations on a regular basis. Oral calcium supplements and iron tablets were being ingested by patient numbers 3 and 6, respectively. Two patients admitted to laxative abuse, and only one patient admitted to self-induced vomiting before admission. None of the patients admitted to taking any emetic agents. Six of the patients admitted to overexercising; the most common form was walking or jogging and patient number 4 claimed that she performed 1500 sit-ups daily in the 7 y before admission. The symptoms of weakness, either on walking or climbing stairs, and fatiguability (exercise intolerance) were reported by all the patients as developing within the 6 wk before admission. Tendon reflexes were diminished or absent in three of the patients. None of the patients complained of any sensory loss, numbness, or paraesthesia and no signs of sensory loss were apparent.

When compared with published standard weight-for-height tables, all of the patients were severely below expected body weight. Mean weight loss was 41.5 (3.3)% of expected weight. Mean weight loss was 41.5 (3.3)% of expected weight.

Hematologic, Biochemical, and Endocrine Abnormalities

Hematologic, biochemical, and metabolic abnormalities are summarized in Table II. None of the patients was deficient in vitamin B12 or folate; indeed, three had elevated B12 levels: patient number 1 had nearly 2.5 times above the upper limit of the reference range = 130 000–450 000 nmol/L. Patient number 6 had a mild thrombocytosis (platelets were 509 000 mm^-3). None of these patients had any clinical evidence of a coagulopathy.

CK was elevated only in patient number 1 at 414 IU/L (reference range = 15–75 IU/L). Liver enzymes were profoundly disturbed in patient number 1, in whom AST was 522 IU/L (reference range = 10–50 IU/L) and AP was 142 IU/L (reference range = 30–120 IU/L). AST was also elevated in patient numbers 6 and 7, with values of 58 IU/L and 103 IU/L, respectively. y-GT was elevated only in patient number 2 at 84 IU/L (reference range = 5–55 IU/L). Otherwise CK, liver enzymes, and bilirubin results were within normal limits for the study sample.

Serum carnosinase activity was reduced in this series of patients with AN [91 (44) nmol · mL^-1 · min^-1] compared with an age and sex-matched control group [205 (31) nmol · mL^-1 · min^-1; P < 0.00005; Fig. 1]. Seven (88%) of eight patients had serum carnosinase activities less than 2 SD (143 nmol · mL^-1 · min^-1) below the control group mean.

Two patients (numbers 3 and 4) had hypokalemia, with values of 3.3 and 3.1 mmol/L, respectively. Serum urea was elevated at 12.2 mmol/L, only in patient number 1, probably reflecting dehydration on admission. Serum creatinine, sodium, and random glucose levels were within normal limits in all patients. Hypocalcemia occurred only in patient number 8 who had a calcium level corrected for serum albumin of 2.03 mmol/L (reference range =

1. Hematologic abnormalities (I)
   - Leucopenia
   - Lymphopenia
   - Microcytosis
   - Normocytic anemia

2. Hematologic abnormalities (II)
   - Iron
   - Ferritin
   - B12
   - Folate

3. Enzyme and metabolic abnormalities
   - AST
   - AP
   - CK
   - Carnosinase
   - Urea
   - PO4

<table>
<thead>
<tr>
<th>Patient number</th>
<th>Hematologic abnormalities (I)</th>
<th>Hematologic abnormalities (II)</th>
<th>Enzyme and metabolic abnormalities</th>
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</thead>
<tbody>
<tr>
<td>1</td>
<td>Leucopenia, thrombocytopenia</td>
<td>Iron↑, ferritin↑, B12↑, folate↑</td>
<td>AST↑, AP↑, CK↑, carnosinase↓, urea↑, PO4↓</td>
</tr>
<tr>
<td>2</td>
<td>Lymphopenia</td>
<td>Iron↓</td>
<td>γGT↑, carnosinase↓</td>
</tr>
<tr>
<td>3</td>
<td>Lymphopenia</td>
<td>Normal</td>
<td>K↓, carnosinase↓</td>
</tr>
<tr>
<td>4</td>
<td>Microcytosis</td>
<td>Iron↓, ferritin↓</td>
<td>K↓, Na↓</td>
</tr>
<tr>
<td>5</td>
<td>Lymphopenia</td>
<td>Iron↓, B12↑</td>
<td>Carnosinase↓</td>
</tr>
<tr>
<td>6</td>
<td>Normocytic anemia, lymphopenia, thrombocytosis</td>
<td>AST↑, carnosinase↓, HCO3↑, PO4↓</td>
<td>Carnosinase↓, Ca↓</td>
</tr>
<tr>
<td>7</td>
<td>Leucopenia, thrombocytopenia</td>
<td>Iron↑, B12↑</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>Normocytic anemia</td>
<td>Normal</td>
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</tr>
</tbody>
</table>

↑, elevated; ↓, reduced; AP, alkaline phosphatase; AST, aspartate aminotransferase; Ca, calcium; CK, creatine kinase; HCO3, bicarbonate; γGT, y-glutamyl transferase; K, potassium; Na, sodium; n/a, not available; PO4, phosphate.

![FIG. 1. Serum carnosinase activity in patients with anorexia nervosa compared with age- and sex-matched controls.](image)
all subjects demonstrated an increase in the blood lactate level to above the reference range for follicular phase. Six (75%) patients had raised basal lactate concentrations (refer to Figure 2). A similarly impaired response is frequently seen in AN and is usually without clinical consequence. However, the abnormal findings from standard biochemical and hematologic investigations are broadly in line with results from previous studies. The lymphopenia found in most of the patients is frequently seen in AN and is usually without clinical consequence. Therefore, serum carnosinase activities may be reduced in patients with AN due in part to the associated skeletal myopathy and in part by a central mechanism due to damage of carnosinase-producing brain cells.

Biochemical Assessment of Muscle Function

Figure 2 shows the changes in blood lactate after ischemic exercise. The mean resting blood lactate level was 2.24 (0.82) mmol/L. Six (75%) patients had raised basal lactate concentrations (reference range = 0.7–2.0 mmol/L). After 1 min of ischemic exercise, all subjects demonstrated an increase in the blood lactate level to between 1.0 and 2.5 times the resting level [mean = 1.81 (0.52)] compared with values in normal subjects of three- to five-fold.

DISCUSSION

Extreme weight loss in AN is obviously the cause of a profound whole-body metabolic disturbance. The present study has shown that there is also a specific myopathy causing proximal muscle weakness in AN that is associated with a selective disturbance of skeletal muscle metabolism. Indeed, serum carnosinase activities are reduced in patients with anorexia nervosa (AN). Interestingly, patient number 6 had an albumin level of 29 g/L (reference range = 35–50 g/L). Patient numbers 1 and 7 had markedly low phosphate levels of 0.48 and 0.29 mmol/L, respectively (reference range = 0.8–1.4 mmol/L). In no patient was the cholesterol level higher than desirable (<5.2 mmol/L).

Free T4 and TSH levels were normal in all the patients. Elevated GH concentration of 50 μg/L was noted in patient number 1, and low GH levels (<2.5 μg/L) were observed in four other patients. LH was reduced in all patients (reference range for follicular phase = 2–10 U/L). Similarly, FSH was reduced in seven patients; patient number 4 had a level of 5.3 U/L (reference range for follicular phase = 2–10 U/L).

FIG. 2. Blood lactate response to ischemic exercise. Data shown are mean (SD). Normal subjects show three- to five-fold peak rise in lactate, i.e., >6 mmol/L.

2.30–2.65 mmol/L). Magnesium levels were within the reference range in all patients. Hypoalbuminemia was found only in patient number 6 who had an albumin level of 29 g/L (reference range = 35–50 g/L). Patient numbers 1 and 7 had markedly low phosphate levels of 0.48 and 0.29 mmol/L, respectively (reference range = 0.8–1.4 mmol/L). In no patient was the cholesterol level higher than desirable (<5.2 mmol/L).

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DISCUSSION

Extreme weight loss in AN is obviously the cause of a profound whole-body metabolic disturbance. The present study has shown that there is also a specific myopathy causing proximal muscle weakness in AN that is associated with a selective disturbance of skeletal muscle metabolism, most notably the diminished lactate response to ischemic exercise. A similarly impaired response is seen in the partial expression of myophosphorylase deficiency in McArdle’s disease.21 A glycogen-storage disease in which young patients complain of fatigueability and muscle pain after exercise and, as they grow older into adulthood, develop proximal muscle wasting and weakness. As a result of the myophosphorylase deficiency, glycogen accumulates within skeletal muscle in McArdle’s disease. Interestingly, ultrastructural studies using electron microscopy have demonstrated abnormal accumulations of glycogen within muscle fibers in biopsy samples from several of the anorexic patients in this study. It is possible that, in the AN patients reported here, this is due to a defect in anaerobic glycolysis, which also becomes manifest in the impaired lactate response to ischemic exercise.

The most commonly occurring form of muscle disease is characterized by type 2 fiber atrophy.22 Selective type 2 fiber atrophy is frequently associated with an underlying metabolic disturbance and has already been described in AN. It has been found in both iatrogenic28 and experimentally29 induced steroid myopathy and is a feature of chronic alcohol myopathy30–33 and various forms of muscular dystrophy34 and congenital myopathy.35 The significant reduction of serum carnosinase activity found in the present series of patients also supports an association between this finding and the myopathy due to AN. The mechanism of this reduction is uncertain. Serum carnosinase is a dipeptidase that catalyzes the hydrolysis of carnosine (β-alanyl-l-histidine) and anserine (β-alanyl-l-methylhistidine) and other histidine-containing dipeptides.36 Recent studies have indicated that the enzyme activity in serum is largely derived from the brain. Indeed, serum carnosinase activities are reduced in disorders of the central nervous system.38 In patients with AN, there may be an abnormality or damage of the cells producing carnosinase, resulting in the observed reduction in serum carnosinase activity seen in many of the patients in this study. In addition, serum carnosinase activity is reduced by approximately 50% in chronic alcohol misusers with histologic evidence of type 2 fiber atrophy.32 Reduced serum carnosinase activity has also been reported in various forms of muscular dystrophy34 and congenital myopathy.35 Therefore, serum carnosinase activities may be reduced in patients with AN due in part to the associated skeletal myopathy and in part by a central mechanism due to damage of carnosinase-producing brain cells.

The abnormal findings from standard biochemical and hematologic investigations are broadly in line with results from previous studies. The lymphopenia found in most of the patients is frequently seen in AN and is usually without clinical consequence.39 Deficits of potassium and phosphate, known to cause muscle weakness, were not consistently observed and did not correlate with the severity of the myopathy. However, both of these are found predominantly intracellularly, and serum levels do not necessarily reflect whole-body status.40 Interestingly, patient number 7 had a severe hypophosphatemia. Phosphate is necessary for the production of phosphorylated intermediates required by glycolysis, glycogen formation, and protein synthesis. The commencement of inpatient refeeding may have turned a chronic phosphate deficit into an acute one41 and serves as a reminder of one of the potential hazards entailed in refeeding extremely malnourished individuals. AST was elevated in three patients, one of whom (patient number 1) also had a raised CK level. The exact mechanism of enzyme release is uncertain but is thought to be due to a reduction in intracellular ATP, which requires phosphate, leading to increased muscle cell permeability.41

The endocrine results are consistent with previous well-documented findings in AN and essentially reflect the body’s appropriate adaptation to starvation rather than a primary disorder of hypothalamic–pituitary function. All the patients had secondary amenorrhea and hypogonadotrophic hypogonadism. GH secretion is frequently elevated in AN but is not associated with clinical signs of acromegaly because insulin-like growth factor 1 (somatomedin) production in the liver is restricted.42 Free-T4 and TSH levels were within normal limits in this patient group. Due to decreased peripheral conversion, free T3 levels are frequently reduced in AN. An association between a low T3 state and an increase in serum muscle indicators, such as CK, has been previously reported, although, as in the present study, these markers were elevated only in a minority of patients. The role of these endocrine changes in the etiology of skeletal myopathy in AN merits further investigation.
CONCLUSION

Severe weight loss due to AN provides a model for the effects of pure malnutrition, in the absence of any systemic disorder or infection, on the skeletal muscle system in previously well-nourished individuals. The resultant disorder is a metabolic myopathy associated with selective type 2 fiber atrophy, abnormal accumulation of glycogen within muscle fibers, diminished lactate response to exercise, and reduced serum carnosinase activity.

The mechanisms underlying these changes are unclear. In underdeveloped countries, myopathy in malnutrition is usually secondary to a neuropathy and is frequently associated with concomitant illnesses and infections. In this series, however, there was no clinical evidence of sensory loss and no evidence of neurogenic change in muscle biopsies, suggesting that myopathy in AN is not secondary to a neuropathy. Apart from AN-associated dietary restriction (the one behavior all the patients had in common) and severe weight loss, the patients suffered from no other illnesses. None of the sample had been abusing the emetic ipecac (emetine) syrup that has been reported as a rare cause of toxic myopathy in skeletal muscle. Treatment should be directed at reinstating appropriate eating behavior while attending to patients’ psychological health.

ACKNOWLEDGMENTS

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REFERENCES

23. Durbin JFG, Marsyers RC. Body fat assessed from total body density and its estimation from skinfold thickness; measurements on 481 men and women aged from 16 to 72 years. Br J Nutr 1974;32:77