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At 2 weeks of age, protein fractional breakdown rates were significantly higher in line LG compared to line HG, showing divergence in the rate of muscle protein degradation with selection for growth. By contrast, the usual major determinant of accretion, protein fractional synthesis rates remained unchanged, as previously suggested by Tomas *et al* [(1991) *Br Poult Sci* 32, 363-376] in chickens. In agreement with this, Seve *et al* [(1990). *In: Proc 4th Felasa Symposium* (10-15 June 1990, Lyon) 69-73] also recorded in 2 extreme genotypes of pigs an higher muscle protein turnover in the LG than in the HG line.

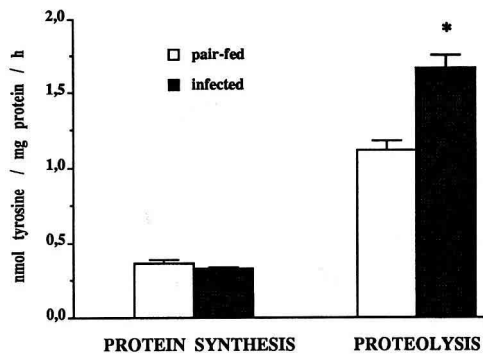
In 4-week-old chickens the difference between protein fractional breakdown rates between the lines was abolished, suggesting that changes in protein metabolism occur principally in the first days of life. However, when expressed in terms of absolute amount, protein deposition (g/d) was still higher in the fast growing line (2.5 or 1.8 times higher at 2 or 4 weeks of age).

### Involvement of Ca<sup>2+</sup>- and ATP-ubiquitin-dependent proteases in increased skeletal muscle proteolysis in septic rats.

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In bacterial infection, muscle protein wasting is partly due to anorexia. The aim of the present studies was to measure variations in both protein synthesis and breakdown in incubated muscles, and to identify the proteolytic pathways activated in infected rats.

Two groups of 8 rats (approximate initial body weight 300 g) were intravenously injected with 6 x 10<sup>8</sup> live *E coli* or saline. Control animals were pair-fed with infected rats. Animals were studied 2 d post-injection. Protein synthesis and breakdown measured *in vitro* using isolated epitrochlearis muscles, as described by Tischler *et al* [(1982) *J Biol Chem* 257, 1613-1621]. mRNA levels for proteases or co-factors involved in the 3 major proteolytic systems so far identified in skeletal muscle (*eg*, cathepsin D, a lysosomal protease; m-calpain, a Ca<sup>2+</sup>-dependent protease;



**Fig 1.** Rates of proteins turnover in septic (■) and pair-fed (□) rats. Values are means ± SD.  $p < 0.005$  vs pair-fed (L Voisin *et al*).

ubiquitin and the C8 proteasome subunit, both involved in the ATP-ubiquitin-dependent and proteolytic pathway) were measured in tibialis anterior muscles by using Northern blot procedures, as previously described by Taillandier [Taillandier (1993) Thèse de Doctorat de l'Université Blaise Pascal, Clermont-Ferrand II, France]. These mRNA levels appear strongly correlated with the rate of protein breakdown in many different muscle protein wasting conditions [Attaix *et al* (1994) *Reprod Nutr Dev* 34, 583-597].

The food intake of infected rats was depressed by 84% during the experimental period, and body weight loss was 20% within 2 d of infection. The mass of the tibialis anterior muscles was 19% lower in septic rats than in pair-fed controls. The rate of protein breakdown in the incubated epitrochlearis muscle of infected animals was 49% higher ( $P < 0.05$ ) than in the control group, but protein synthesis was not significantly depressed (fig 1). There was no variation in mRNA level for cathepsin D in the infected group compared to the control. By contrast, mRNA levels for the m-calpain, ubiquitin and the C8 proteasome subunit were systematically higher in septic animals than in pair-fed controls.

These data demonstrate that skeletal muscle protein wasting observed in septic animals mainly results from increased proteolysis in rigorous conditions of pair-feeding. They also suggest that the coordinate activation of both Ca<sup>2+</sup>- and ATP-ubiquitin-dependent proteolytic pathways is mainly responsible for skeletal muscle protein loss. Increased ATP-ubiquitin-dependent proteolysis has been reported in all muscle protein wasting

conditions studied so far [Attaix *et al* (1994) *op cit*], and could presumably be responsible for the breakdown of myofibrillar proteins.

**Coordinate activation of lysosomal, Ca<sup>2+</sup>- and ATP-ubiquitin-dependent proteolytic pathways in the soleus muscle in hind-limb suspended rats.** D Taillandier<sup>1</sup>, E Aourousseau<sup>1</sup>, A Obled<sup>1</sup>, D Béchet<sup>1</sup>, M Ferrara<sup>1</sup>, P Cottin<sup>2</sup>, A Ducastaing<sup>2</sup>, X Bigard<sup>3</sup>, CY Guézennec<sup>3</sup>, D Attaix<sup>1</sup> (<sup>1</sup>INRA-Theix, Centre de Recherche en Nutrition Humaine et Unité d'Étude du Métabolisme Azoté, 63122 Saint-Genès-Champagnelle; <sup>2</sup>ISTAB et Université de Bordeaux I, 33405 Talence; <sup>3</sup>Centre d'Études et de Recherches de Médecine Aérospatiale, 91228 Brétigny-sur Orge, France)

During simulated weightlessness, increased protein breakdown is the major determinant of muscle atrophy and protein wasting in antigravity muscles, *ie* the soleus [Thomason and Booth (1990) *J Appl Physiol* 68, 1-12]. The aim of this study was to identify the proteolytic pathways responsible for these alterations in skeletal muscle.

Twenty male Wistar rats with an initial body weight of 120 ± 5 g were randomly assigned to a control and a non-weight-bearing group. The experimental group was suspended by the tail according to the protocol described by Taillandier [(1993) Thèse de Doctorat de l'Université Blaise-Pascal, Clermont-Ferrand II, France]. Animals were studied after 9 d of treatment. Isolated soleus muscles were incubated to measure protein synthesis and breakdown simultaneously, as described by Tischler *et al* [(1982) *J Biol Chem* 257, 1613-1621]. Two other groups of rats raised in identical conditions were used to measure either the rate of proteolysis after inhibiting both lysosomal and Ca<sup>2+</sup>-dependent proteases [Wing and Goldberg (1993) *Am J Physiol* 264, E668-E676], or mRNA levels for multiple components of proteolytic systems, in the soleus muscle.

The soleus muscle atrophy was higher than 50% ( $P < 0.001$ ) after 9 d treatment. Total protein breakdown of suspended rats was enhanced by 66% ( $P < 0.001$ ), compared to control animals. By contrast, the rate of protein synthesis was not significantly modified by hindlimb suspension. The

lysosomal and Ca<sup>2+</sup>-dependent proteolysis was greatly enhanced (254%,  $P < 0.001$ ) in the soleus muscle of non-weight-bearing rats, but did not exceed 18% of total proteolysis. In addition, the inhibition of both lysosomal and Ca<sup>2+</sup>-dependent proteases did not suppress increased proteolysis in the soleus muscle of suspended rats. Northern blot procedures revealed large increases in mRNA levels for lysosomal (cathepsins B, L and D), and Ca<sup>2+</sup>-dependent (m-calpain) proteases, and for components of the ATP-ubiquitin-dependent proteolytic pathway (ubiquitin, 14-kDa ubiquitin carrier protein E2, C2 and C9 proteasome subunits) in the soleus muscle of non-weight-bearing rats. Similar data were observed in a phasic muscle, the extensor digitorum longus.

These data clearly demonstrate that the major proteolytic systems are coordinately activated in skeletal muscle during simulated weightlessness. Cathepsins, calpains and the proteasome could degrade different classes of proteins, the ATP-ubiquitin-dependent proteolytic pathway being presumably responsible for the breakdown of the major contractile proteins.

## VII. Nutrition and pathology

**Effects of moderate amounts of dietary fat on post-prandial lipemia in healthy human subjects.** C Dubois<sup>1</sup>, M Armand<sup>1</sup>, P Borel<sup>1</sup>, M Senft<sup>1</sup>, H Portugal<sup>2</sup>, AM Pauli<sup>2</sup>, PM Bernard<sup>2</sup>, V Azais-Braesco<sup>3</sup>, C Latge<sup>4</sup>, D Lairon<sup>1</sup> (<sup>1</sup>Unité 130-INSERM (National Institute of Health and Medical Research), 18, avenue Mozart, 13009 Marseille; <sup>2</sup>Hôpital Sainte-Marguerite, 13000 Marseille; <sup>3</sup>Unité Vitamines, LNSA-INRA, 78000 Jouy-en Josas; <sup>4</sup>Crealis, Brive, France)

There have been considerable developments in postprandial studies on human subjects during recent years. These experiments have generally studied blood postprandial responses to amounts of fat (70–140 g) greatly exceeding that usually ingested during a meal, even for a western high-fat diet (40–50 g per meal).

The present study therefore compares the post-prandial responses to 2 different and moderate amounts of dietary fat (30 and 40 g) in the form of well-defined dietary emulsions of sunflower oil (droplet size: 30–44 µm). The 30 g fat