EFFECT OF GROWTH RATE OF LAMBS ON CHEMICAL AND HISTOLOGICAL CHARACTERISTICS OF MUSCLE

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Summary

Fourteen lambs of Dorset Horn-Poll Merino breeding (eight wethers and six ewes) were used. Half the number of each sex were kept on a high plane of nutrition throughout their growing period, while the other half were given only maintenance rations from approximately 10 weeks of age till slaughter of all animals at five to six months.

The *L. dorsi* muscle was excised and examined. Sarcoplasmic proteins, intramuscular lipid and myoglobin fractions were significantly lower, but water, stroma protein and collagen-elastin content were higher in the low than in the high plane group. Growth retardation appeared to have little effect on myofibril proteins and on the ash content of the muscle.

Mean muscle fibre diameter and mean sarcomere length tended to be greater in the high plane than in the low plane group. The microscopic evaluation of collagen, elastin and fat agreed with the chemical analyses.

I. INTRODUCTION

It has been suggested that well-fed growing animals produce more tender meat than under-nourished ones (Deatherage 1963; Yeates 1965). A number of workers have studied the effect of nutritional stress on carcass quality of various animals, but with the exception of important studies on non-ruminants by McCance and his colleagues (Dickerson and McCance 1960; Widdowson and McCance 1963), most investigations have been on growth and body composition in terms of bone : muscle : fat ratios and not on chemical changes in muscle. Extreme loss of weight due to severe underfeeding is associated in adult cattle with decreases in muscle size (cross-sectional area) and muscle fibre diameter which may increase when the animals return to a high plane of nutrition (Yeates 1964).

It is not known what changes prolonged underfeeding causes in the chemical and physical characteristics of the muscle nor in meat quality. This information is of particular importance in countries which experience recurrent droughts, when stock may lose much weight. The present investigations were made to help resolve these questions. Biochemical and histological results are described here; physical characteristics and meat quality will be reported elsewhere.

II. MATERIALS AND METHODS

(a) Animals and Experimental Design

Fourteen Dorset Horn Merino lambs (eight wethers and six ewes), all by one sire, were used. The ewes and four of the wethers were single lambs, while the other four wethers were each twin to a ewe lamb. All lambs were reared by their mothers from birth to 10 weeks of age, when they were divided into two groups, balanced for sex, age, weight and whether singles or twins. One group then

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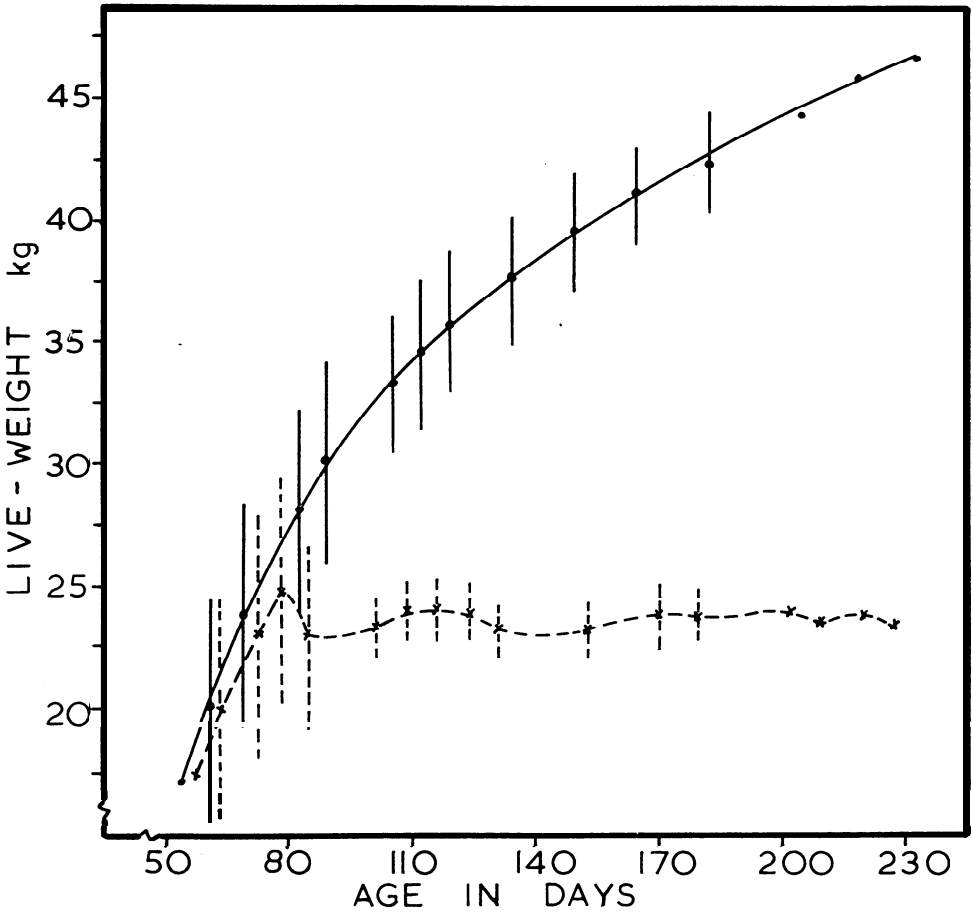

Fig. 1.—Mean growth rates of high plane (•—•—•) and low plane (x—x—x) lambs. The vertical lines represent the standard deviation from means.

remained on good pasture with their mothers (high plane) while the other group was weaned and given restricted grazing in a small paddock (low plane), the aim being to prevent any further weight gain (See Figure 1). Water was freely available to both groups. The lambs were weighed at intervals of 7 to 14 days. They were slaughtered in pairs, comprising one animal from each group matched for sex, type of birth and age. Six pairs were killed at ages of five to six months and the last pair at eight months.

(b) Carcass Appraisal

After slaughter and evisceration, each carcass was appraised by the method of Thwaites, Yeates and Pogue (1964). The outline of the transverse section of the *L. dorsi* muscle at the level of the 12th-13th rib (right side) was traced and the area was measured with a planimeter.

(c) Sampling for Chemical Analysis

The 3rd-6th lumbar portion of the *L. dorsi* muscle (left side) was excised, wrapped in polythene and kept in cold storage at 2.0 ±1°C until time of analysis

48 h post-mortem. Before analysis of the muscle, a thin layer of the exposed surface was removed to minimise microbial contamination, and it was then thoroughly minced and mixed to give homogeneity. Water content was determined by drying at 105°C (Hamm and Deatherage 1960) and ash content was obtained by A.O.A.C. methods (1960). Intra-muscular lipids were extracted by the method of Wierbicki et ***al.*** (1965).

(d) Protein Fractionation

The proteins of the muscle were studied by Helander's (1957) procedure, except that the meat was minced instead of frozen and sectioned into 10 μ slices, and an extraction apparatus similar to that of Ahmad and Cook (personal communication) was used. Fractionations were made in triplicate at 2.0 $\pm$ 1 °C to guard against denaturation of proteins. Sarcoplasmic proteins were extracted with 0.03 M K-phosphate buffer at pH 7.4 (low ionic extraction(A)). High ionic extraction (B) was effected with 1.1 M KI in 0.1 M K-phosphate buffer at pH 7.4. The difference between fraction (B) and fraction (A) represented the soluble myofibril proteins. The residue from low ionic extraction was treated with 0.1 M NaOH and the extract represented the total myofibril proteins (Hegarty, Bratzler and Pearson 1963). The residue was further treated according to the method of Lowry, Gilligan and Katersky (194 1) for combined collagen-elastin determination. The residue from high ionic extraction was neutralized, washed, treated with 3 : 1 (v/v) alcohol-ether mixture to remove adipose tissue, dried and recorded as stroma protein fraction. The extracted proteins were centrifuged for 15 min at 3,000 r.p.m. and filtered through Whatman No. 1 filter paper before determination.

(e) Protein Determination

Protein estimations were made by the biuret method of Gornall, Bardawill and David (1949) on duplicate 1 ml fractions from each extract, containing 3-6 mg protein. Samples were treated with petroleum-ether to remove any lipid contamination before developing the colour which was measured at wavelength of 540 mμ. Bovine albumin (Koch-Light Laboratories) was used to prepare the standard curves for low ionic, high ionic and 0.1 M NaOH extracts. The purity of the albumin was checked by determination of its nitrogen content. Myoglobin was determined according to the method of Wierbicki ***et al*** (1955).

(f) *Histological Studies*

The ***L. dorsi*** muscle (left side) in the 13th thoracic-2nd lumbar region was excised and kept in an atmosphere of nitrogen at 24.0 $\pm$1°C. Core samples of 1.3 cm diameter were taken after the resolution of rigor. They were fixed in Susa, embedded, sectioned (8-12 μ thick) and stained in Orcein and van Gieson by a method modified from Carleton and Drury (1957). Observations were made on the quantity, type and distribution of muscle fibres, collagen, elastin, and fat; muscle fibre diameter and sarcomere length were measured.

III. RESULTS

(a) Carcass Characteristics

The data on carcass appraisal are summarised in Table 1. There were differences between high and low plane lambs in carcass weight, dressing percentage, and fleshing index. Photographs of the 12th-13th rib cut from representative

TABLE 1

Effect of Plane of Nutrition on Carcass Characteristics of Lambs

	Liveweight kg	Carcass Weight (cold) kg*	Dressing Percentage	Length of Carcass (cm)	Fleshing Index		T.S. of *L. dorsi*		Rib fat thickness (cm)
					Gross	Net	Area (cm^2)	Depth (cm)	
Low Plane (Mean of 7 Lambs)	23.08	9.76	42.2	53.00	—3.69	—3.69	9.83	2.47	<0.1
Standard Deviation	±1.13	±0.88	±1.81	±0.77	±2.05	±2.05	±0.85	±0.24	—
High Plane (Mean of 7 Lambs)	42.55	20.91	49.10	59.90	+7.5	+5.8	15.93	3.51	0.56
Standard Deviation	±3.77	±2.51	±1.96	±1.75	—	±3.47	±2.56	±0.45	±0.09

*Calculated by making 2% reduction in hot carcass weight.

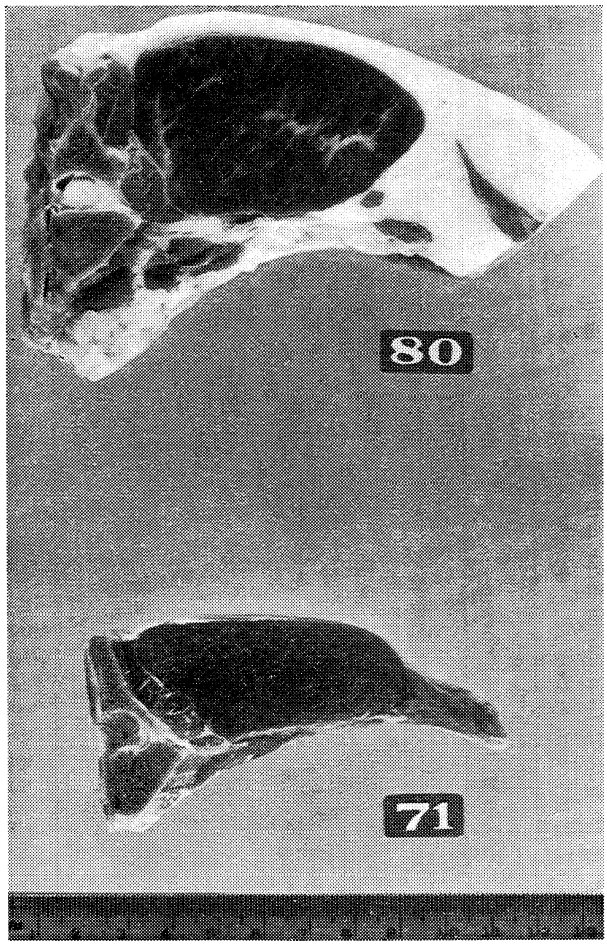

Fig. 2—Rib cuts of representative carcasses of high plane (80) and low plane (71) lambs.

animals of the high and low plane groups (Figure 2) show the differences in muscle development, subcutaneous fat thickness and intra-muscular fat (marbling). The low plane lambs were almost devoid of subcutaneous fat over the loin eye muscle. The width of the eye muscles was similar but the average muscle depth was only 2.47 cm for low plane lambs as compared to 3.5 1 cm for high-plane lambs.

(b) Chemical Characteristics

The results given in Table 2 indicate that the ***L. dorsi*** muscle of high-plane lambs was about 4% lower in mean water content but contained about 2.8% more inter- and intra-cellular lipid. The percentage ash in both groups was similar. Sarcoplasmic proteins were markedly lowered as a result of nutritional stress, whereas the myofibril fraction which constitutes the contractile filaments of actin and myosin was unaffected. Both collagen-elastin and stroma fractions were rela-

TABLE 2

Chemical and Histological Characteristics of L. dorsi of Low and High Plane Lambs
Mean values expressed as percentages of fresh weight of muscle

			Low Plane (Mean of 7 Lambs)	High Plane (Mean of 7 Lambs)	Standard Error	Significance of Difference
Water content %			77.45	73.42	±0.41	$P<0.01$
Protein Fraction %	Sarcoplasmic	Total	5.65	7.57	±0.27	$P<0.01$
		Myoglobin*	0.32	0.44	±0.02	$P<0.01$
	Myofibril	Total	11.51	11.62	±0.29	N.S.
		Soluble	10.49	9.85	±0.37	N.S.
	Stroma	Total	3.21	2.24	±0.13	$P<0.01$
		Collagen-Elastin	1.12	0.75	±0.09	$P<0.05$
Lipid %			0.49	3.37	±0.29	$P<0.01$
Ash %			1.070	1.076	±0.154	N.S.
Fibre Diameter			36.1μ	39.3μ	±1.31μ	$P>0.05<(0.1)$†
Sarcomere Length			1.39μ	1.52μ	±0.47μ	$P>0.05<(0.1)$†

*Determined as cyanometmyoglobin.
†Derived by "t-test" analysis.
N.S. Not-significant.

tively high in low plane lambs but myoglobin content, which accounts for most of the colouring pigment of the muscle, was considerably lower.

(c) Histology

Muscle fibres varied in diameter within groups and the low plane group generally had smaller fibres than the high plane group ($0.1>P>0.05$). Sarcomere length also varied with treatment: mean values for the low and high plane groups respectively were 1.39 μ and 1.52 μ ($0.1>P>0.05$).

The microscopic evaluation of collagen, elastin and fat agreed with the chemical analyses. In all samples, collagen appeared to be of the same type, and occurred in roughly the same proportion to muscle fibres. Elastin was sparse in all samples, and-frequently could only be seen in walls of blood vessels; occasional single fibres were associated with the collagen of fat deposits. Fat was almost absent in low plane lambs except in one where moderate intra-muscular fat deposits were observed, and in another where fat deposits with very small fat cells were noted. In other low plane samples, the location of earlier fat deposits was indicated by the "collapsed" (i.e. depleted) nature of the fat cells. In all, high plane samples, intra-muscular fat deposits were obvious, and fat cells were well filled.

IV. DISCUSSION

This study indicates that significant biochemical and histological differences may exist between the muscle of quickly grown and retarded lambs. Concerning the differences in lipid, it is well known (Neptune, Sudduth and Foreman 1959; Issekutz, et *al*. 1964; Havel, et *al*. 1964) that intramuscular lipid serves as a source of energy during periods of caloric deficiency, but it has recently been shown (Masora, Rowe11 and McDonald 1964; Masora, et ***al.*** 1966) that some lipids of the muscle are structural-functional elements which are not depleted for the purpose of energy metabolism as a result of limited caloric supply. Thus about 0.5% lipid persisted in the muscle, even during the prolonged growth retardation involved in the present study.

The relative decrease in sarcoplasmic proteins in low plane lambs might be associated either with a decline in the synthesis of muscle sarcoplasm or with an increase in the catabolism of these proteins. The myofibril proteins and collagen-elastin or stroma fractions seem, on the other hand, to be relatively stable components of the muscle complex. The results show that these fractions of the muscle did not vary in absolute amount under the experimental conditions. However, with the decrease in sarcoplasmic proteins and intra-muscular fat, the proportion of collagen-elastin complex to total muscle increased in the low plane lambs.

Although there was some reduction in muscle fibre diameter in the low plane group, the small difference in the present experiment is in keeping with the finding of other workers (Joubert 1956; Yeates 1964), that very severe weight loss is necessary to cause pronounced decrease in the muscle fibre diameter.

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Note:—Since submission of the above manuscript estimation of myoglobin by the method of Fleming, H. P., Blumer, T. N., and Caraig, H. B., 1960 (J. Anim. Sci. **19:** 1164) in a subsequent experiment has shown myoglobin content to be unaffected by nutritional stress.