THE NUCLEIC ACID AND NITROGEN CONCENTRATION OF A BOVINE SKELETAL MUSCLE DURING EMACIATION AND REPLETION*

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A feature of animal production in the arid and semi-arid regions of the world is the seasonal emaciation of livestock due to chronic protein and energy depletion during the long, dry seasons and frequent droughts, followed by protein repletion during the shorter wet season (Payne and Hutchinson 1963; Robinson and Sagemen 1967). Bearing the brunt of the protein depletion and probably buffering the effect of protein loss in the essential organs of the body are the skeletal muscles which represent a major protein store in the body (Masters 1963). Although such protein depletion of body tissues occurs in animals exposed to similar climatic conditions throughout the world, little work has been carried out to investigate its severity with respect to nitrogen wastage from muscle cells.

One approach to the study of this problem arises from the observation that the DNA content of the diploid nucleus in interphase is constant (Boivin, Vendrely, and Vendrely 1948) and it has been proposed that the DNA content of organs or tissues can be used as a reference basis to demonstrate changes in other cellular constituents (Waterlow and Mendes 1957). This approach has now been used to measure changes in the nitrogen content of skeletal muscles of 11 adult Shorthorn cows severely depleted by drought conditions.

Methods

During mustering of cattle on natural pasture in north-western Australia in the course of another experiment in which the cattle required periodic weighing, 11 cows were found which were in a collapsed condition and unable to walk. They were brought by truck to the Kimberley Research Station, and samples weighing 0·7–1·5 g were taken from the M. triceps brachii (caput longum), right side, under local anaesthesia. This muscle was selected as a site for biopsy because its rate of growth is known to be similar to that of the musculature as a whole (Butterfield and Berg 1966). The cows regained body condition when given a high energy and high protein diet (whole cotton seed) *ad libitum* (see Fig. 1), and muscle samples were taken 3 months later from the corresponding site on the left side. The samples were weighed immediately on collection, placed in ethanol at —15°C, homogenized, and freeze-dried. Analysis for DNA and RNA was as recommended by Munro and Fleck (1966). Yeast core RNA (Mann Research Laboratory) and calf thymus DNA (Mann Research Laboratory), (Sigma Chemical Company) were used as standards, and their phosphorus content was determined (Allen 1940) as a check on purity. Nitrogen was determined by microKjeldahl digestion with potassium sulphate and selenium.

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Results

Table 1 compares the results of the above determinations on samples taken from skeletal muscles of depleted and recovering cattle with those taken from six "normal" animals raised on a high plane of nutrition throughout their lives.

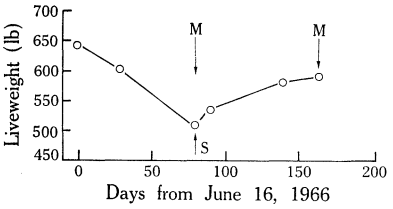

Fig. 1.—Liveweight changes of experimental cattle kept on natural pasture only, and then supplemented (S) during the late dry season (1966). M, times when muscle samples were taken.

The studies of Masters (1963) indicated a reduction of nearly 60% compared with normal values in the protein nitrogen : DNA phosphorus ratio in the muscles of thin sheep. In the present study the ratio of muscle nitrogen : DNA increased by 44% between samples taken when the animals were emaciated and when

TABLE 1

NUCLEIC ACID AND NITROGEN CONCENTRATION OF NORMAL, DEPLETED, AND RECOVERING BOVINE SKELETAL MUSCLE

Normal and depleted values were compared by the *t*-test, using pooled variance. Depleted and recovery values were analysed as repeated observations within animals. Values given are means ± S.E. Significance of differences between normal and depleted and between depleted and recovering values are indicated. n.s., not significant

Condition of Cattle	DNA (mg/100 g wet tissue)	RNA (mg/100 g wet tissue)	Nitrogen (mg/100 g wet tissue)	Ratio RNA/DNA	Ratio Nitrogen/DNA
Normal	25·8±3·9	85·8±6·6	2261±72	3·63±0·44	97·9±13·5
	*	n.s.	*	**	**
Depleted	39·1±2·8	86·8±6·8	1719±139	2·29±0·11	44·8±2·6
	n.s.	n.s.	**	*	**
Recovering	36·0±3·3	99·0±8·1	2280±106	2·85±0·23	67·7±5·6

* $0{\cdot}01 < P < 0{\cdot}05$. ** $P < 0{\cdot}01$.

they had regained about 16% in liveweight. This may be compared to the increase (Waterlow and Mendes 1957) of 45% in the ratio of non-collagen nitrogen : DNA phosphorus in samples taken from children between their entry to hospital in a depleted condition, and their recovery after 3 months on a good diet, when they had gained 29% in weight. In both cases the estimated increase in muscle nitrogen was much greater than would be expected from the liveweight gain. This is to be expected as other body tissues are not depleted by undernutrition to as great a degree as is the musculature (Keys *et al.* 1950).

In the present study the concentration of nitrogen per unit of wet tissue had apparently fallen markedly during depletion. Its increase during repletion was accompanied by a smaller increase in RNA concentration, in agreement with other work (Masters 1963; Robinson and Sagemen 1967).

Most of the evidence for the constancy of the DNA content of mammalian cells deals with specific tissues, particularly the liver. Masters (1963) reported changes in the nucleic acid concentration in skeletal muscle in sheep, but did not record total weight, nor therefore total DNA content, of muscle tissue. Robinson and Lambourne (unpublished data) found that the total DNA content of defined muscle groups in the mouse was virtually constant (175 ± 25 μg) over a twofold range in muscle weight caused by quantitative differences in nutrition.

Fat must be regarded as the major energy reserve in hibernatory animals of cold climates and in migratory species. The active wild animals of temperate and subtropical regions, like the free-grazing and "working" domestic species, have undergone little, if any, selection for fat deposition. The "adapted" cattle of northern Australia which graze over large areas of arid pasture land and experience frequent periods of undernutrition may be viewed as virtually "wild" animals. Muscle protein clearly represents an important labile reserve for such species and the present data suggest that muscle protein can be catabolized to a much greater degree than would be supposed from the observed loss in liveweight of such cattle.

If it is possible to confirm the constancy of the DNA content of easily accessible tissues (skin, muscle) then the ratio of nitrogen (lipid, mineral, etc.) to DNA may become a useful measure of the nutritional status of both feral and domesticated animals. It could be valuable in experimental ecology or as a guide to the necessity of feed supplementation or of adjustment in livestock numbers in pastoral regions.

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