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PREVIEW

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Effect of plane of nutrition on visceral organ growth and metabolism

Burrin, Douglas Guy, Ph.D.

The University of Nebraska - Lincoln, 1987

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PREVIEW

EFFECT OF PLANE OF NUTRITION ON
VISCERAL ORGAN GROWTH AND METABOLISM

by

Douglas G. Burrin

Presented to the Faculty of
The Graduate College in the University of Nebraska
In Partial Fulfillment of Requirements
For the Degree of Doctor of Philosophy

Major: Animal Science

Under the Supervision of Professor Robert A. Britton

Lincoln, Nebraska

October, 1987

TITLE

Effect of Plane of Nutrition on

Visceral Organ Growth and Metabolism

BY

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EFFECT OF PLANE OF NUTRITION ON
VISCERAL ORGAN GROWTH AND METABOLISM

Douglas G. Burrin, Ph.D.

University of Nebraska, 1987

Adviser: Robert A. Britton

Three studies were conducted to measure the effect of plane of nutrition on visceral organ growth and metabolism. In study 1, rats fasted for 72 h generally had lower visceral organ mass, RNA content and ratios of RNA/DNA and protein/DNA than fed rats. In vitro oxygen consumption and valine incorporation rates expressed per unit of tissue did not differ between treatments. However, nutrient restriction decreased total liver oxygen consumption by decreasing organ and cell size. In study 2, lambs were fed ad libitum (ADLIB) or a maintenance (MAINT) diet for 21 d. During the 21-d period at 7-d intervals, four lambs per treatment were slaughtered and measurements of visceral organ size and metabolic activity obtained. During the 21-d period, ADLIB lambs had higher visceral organ mass, protein and RNA contents but lower DNA concentrations than MAINT lambs. Ratios of visceral organ RNA/DNA and protein/DNA in ADLIB lambs were higher than MAINT lambs. Estimates of in vitro oxygen consumption, valine incorporation and substrate oxidation per unit of tissue were relatively similar between treatments, however, estimates of total liver metabolic activity were higher in ADLIB than MAINT lambs. Plane of nutrition affected visceral organ size and metabolic activity through changes in cellular hypertrophy. The design, feeding regimen and duration of study 3 was identical to study 2, and blood flow and

nutrient fluxes were measured in the liver and portal-drained viscera (PDV). During the 21-d period, ADLIB lambs had higher liver and PDV blood flow and oxygen consumption than MAINT lambs. In the control period, total splanchnic tissues (liver + PDV) represented an average of 52% of whole body oxygen consumption which increased to 69% in ADLIB lambs and decreased to 41% in MAINT lambs after 21 d. Net portal absorption of $\text{NH}_3\text{-N}$, α -amino-N (AAN) and essential amino acids and hepatic uptake of AAN, essential and nonessential amino acids in ADLIB lambs was higher than MAINT lambs. In ADLIB lambs, the estimated cost of urea synthesis and gluconeogenesis from nonessential amino acids was higher than MAINT lambs.

PREVIEW

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D.B.

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INTRODUCTION

In the study of whole body metabolism, the internal organs of the viscera play an integral role in absorption and interconversion of nutrients required for growth and development. It may seem logical therefore, that the visceral organs should comprise a significant proportion of mammalian basal metabolic energy requirements. Although technological developments have enabled quantitative empirical estimates of basal energy expenditures, the precise biochemical and physiological processes that are involved are considerably less well understood. The importance of visceral organs in basal metabolism and their key role in many biochemical and physiological processes make them a focal point of interest in attempting to understand the factors that influence basal energy expenditures in mammals.

Several researchers have attempted to define basal metabolic rate both quantitatively and biologically. Classical scientists observed basal metabolic rate to be a constant function of body size over a diverse range of mammalian species. A common distinction made between the biological components of basal energy expenditures is that of service functions (e.g., digestion, respiration and circulation) and processes of cellular maintenance (e.g., protein synthesis, ion transport and lipogenesis). The lack of understanding of these biological phenomena has not, however, prevented the application of empirical methods to quantify whole animal basal energy needs and further determine factors that influence these needs. Factors such as age, breed, stage of production and level of nutrition contribute to

the variation observed in basal metabolic rate. A common correlate to the observed variation in basal metabolic rate is that of visceral organ size and metabolic activity. Compared to the whole body, the visceral organs have a higher metabolic activity with regard to protein turnover and oxygen consumption.

Basal energy expenditures account for a significant proportion of feed energy requirements in all phases of animal production. This necessitates a better understanding of the factors that influence energy needs. The manipulation of basal energy expenditures could, potentially, result in improved efficiency of production in domestic animals. The ability to modulate energetic efficiency of growth by plane of nutrition is related to changes in relative growth rates of visceral organs. However, the underlying components of visceral organ metabolism that may contribute to improved efficiency of whole animal growth rate have not yet been adequately characterized.

PREVIEW

BASAL ENERGY EXPENDITURES

Metabolic body size and the surface area law

In discussing basal metabolic energy expenditures, it is appropriate to begin with a chronology of events that have become the foundation of mammalian bioenergetics. Most of the discussion that follows is a synthesis of information reviewed in the classical treatise of animal energetics by Brody (1945) and Kleiber (1947). In 1780, Lavoisier and Laplace suggested that animal heat originates from the combustion of atmospheric oxygen and organic substances in the animal body. This concept was later confirmed by Rubner (1894) in experiments with fasting dogs and further by Atwater and Benedict (1903) with measurements of heat production in working men. Rubner (1894) also observed a systematic decline in metabolic rate per unit of body weight as body weight increased in fasting dogs. Sarrus and Rameaux (1839) first suggested the relationship between animal heat production and surface area in their thesis presented to the Royal Academy of France during its meeting on July 23, 1839. Based on the theoretical considerations of Rameaux and Sarrus and the practical observations of Rubner and others, the surface area law was developed, which states that in animals of different body size the rate of metabolism is proportional to the animal's body surface area. Bodies of a shape similar to that of a cylinder have a surface area proportional to the $2/3$ power of the volumes. If one assumes a density of 1 kg/liter, Richet (1889) estimated the surface area of a rabbit to be $4.84 W^{2/3}$ square decimeters when W is body weight in

kilograms. Meeh (1879) substituted the 4.84 in Richet's formula with a constant, k , which accounts for variation in shape of the animal, to yield an equation $S = kW^{2/3}$ where S is surface area in square decimeters and W is body weight in kilograms.

Realizing the inherent difficulty in measuring surface area of animals, Kleiber (1947) discussed the validity of applying the surface area law to animal metabolism and proposed some theories to be considered.

The metabolic rate of animals must be in proportion to their body surface because:

1. the rate of heat transfer between animal and environment is proportional to the body surface area.
2. the intensity of flow of nutrients, in particular of oxidizable material and oxygen, is a function of the sum of the "internal surfaces", which in turn is proportional to the body surface.
3. the rate of supply of oxidizable material and oxygen to the tissues is a function of the mean "intensity of blood flow", which is proportional to the cross-sectional area of the blood vessels, which in turn is proportional to the area of body surface.
4. the chemical and anatomical composition of the animal is a function of body size.
5. an inherited metabolic requirement of tissues is a function of body size.

Upon critical review of these theories, Kleiber rejected the

validity of the internal surfaces theory, as well as, chemical and anatomical composition. The rejection of anatomical considerations by Kleiber (1947) is interesting in light of the earlier observation by Brody et al. (1928) that weights of several glandular organs including liver, kidney and gastrointestinal tract increased directly with body weight at a rate similar to that of surface area. However, in his detailed discussion of animal bioenergetics, Brody (1945) neglected to emphasize the relative growth rates of the aforementioned visceral organs and instead focused on organs such as brain, heart and pituitary. The idea that variation in metabolic rate could be explained by relative changes in anatomical composition was suggested by Kestner (1936), although much of his evidence was based on brain weights.

In Kleiber's discussion of applicability of the surface law to animals, he continued by suggesting that an integration of the rate of heat transfer and intensity of blood flow to the tissues were the most plausible theories on which to base the surface law. Brody (1945) discussed various indices of work capacity and discovered that basal metabolism of mature animals of different species is directly proportional to the product of body weight and oxygen consumption per heart beat. The final theory related to the surface law was that of inherited tissue metabolic requirement. Experiments reported by Grafe et al. (1925) and Terroine and Roche (1925) derived from in vitro oxygen consumption of excised tissues concluded essentially that the respiration rate of similar tissues in vitro is unchanged in different

size animals.

Later studies by Kleiber (1941) and Krebs (1950) demonstrated that in vitro oxygen consumption per unit tissue weight decreased systematically with increasing body size. Krebs (1950) also noted that liver tissue exhibited the greatest change in oxygen consumption over the range in animal body sizes. Thus, the theory relating the surface law to a constant cellular metabolic rate was rejected. Due to the failure to accurately define and measure body surface area, its use as a basis to adjust body size for metabolic rate was abandoned. The use of a power function of body weight as a reference base for basal energy needs was proposed by Krogh (1916). Kleiber (1932) plotted the logarithms of fasting metabolic rate against the logarithms of body weight and concluded that metabolic rate was more nearly proportional to the $3/4$ power of body weight than surface area. Similarly, using different data, Brody and Proctor (1932) suggested that basal metabolic rate and endogenous nitrogen excretion were proportional to the $.73$ power of body weight. The results of both Kleiber and Brody were based on measurements collected from mature animals and were intended for comparison of metabolic rate between species of mature animals. Data published by Benedict (1938) over a wide range of body sizes (20 g mouse to 4 ton elephant) under standardized conditions demonstrated a similar relationship between body size and metabolic rate, although some species did appear to deviate from this trend. Benedict (1938) went on to criticize the $.73$ power of body weight as a reference and also stated, "However satisfactory this relationship may be mathematically, this method of

presenting the data completely masks metabolic differences within species."

The relevance of the .73 or $3/4$ power of body weight as a function for predicting metabolic rate of animals within a species has been questioned by Van Es (1972) and Thonney et al. (1976). The variability in the exponent used to estimate basal metabolism from body weight of a given species is apparent, however, much of this variation may likely be a result of factors such as age, diet, breed and prior nutritional history. Nevertheless, many researchers in the nutritional sciences have adopted Kleiber's expression of $W^{.75}$ as a reference upon which many physiological and biochemical processes are based.

Empirical Estimates of Energy Partitioning

Several terms have been used to describe basal energy expenditures. Brody (1945) defined basal metabolic rate as the heat production during rest in a thermoneutral environment under post-absorptive or fasting conditions. The term maintenance energy expenditures or requirement can be derived from analyzing the process of energy partitioning in animals. The partitioning of energy originates from gross energy or the heat of combustion of a particular feedstuff. Gross energy minus fecal energy equals the apparent digestible energy. Fecal energy represents energy contained in undigested feed residues, bacterial cells and tissue sloughed from the intestinal lining. Digestible energy can be partitioned into energy lost as gas and urine to yield metabolizable energy. In ruminants, a

significant loss of energy can be attributed to methane gas lost during microbial fermentation in the foregut. Metabolizable energy (ME) represents that energy available to the body tissues for metabolic purposes (N.R.C., 1981).

A final energy loss is that of heat increment. Brody (1945) defined heat increment as the waste heat from oxidation and intermediary reactions involved in the nutritive process. Dock (1931, 1933) estimated that 80% of heat increment originates in the viscera, probably associated with processes of digestion and absorption of nutrients.

After accounting for heat increment, the remainder of ME can be categorized as net energy (NE), which is energy available for maintenance of body tissues (NE_m) and conversion into some measurable production output (NE_p). Blaxter (1961) determined that of the total gross energy fed to sheep at maintenance, 35% was lost in the feces, 8% in methane, 4% in urine and 6% as heat. The net energy available for maintenance was approximately 47% of gross energy intake. Several approaches to measuring maintenance energy have been derived from the equation expressing the relationship between ME intake, retained energy (RE) and heat loss (H) as follows:

$$ME_I = RE + H$$

By inspection of this equation, if RE is equal to zero then ME_I is equal to the amount of heat produced.

Based on the above considerations, maintenance energy requirements can be estimated experimentally in adult, non-pregnant, non-lactating animals by feeding measured amounts of feed during

periods energy stasis, or if one assumes energy and weight stasis to be equivalent, weight stasis. Ledger and Sayers (1977), observed a progressive decline in feed energy required to maintain body weight in steers weighing 185, 275 and 450 kg. They concluded that the reduction in feed intake required to maintain live weight was a result of decreased basal metabolism and increased efficiency of utilization of the diet. The major limitations of using this method to estimate maintenance are the assumptions of a constant value for the efficiency of energy use for maintenance and that weight stasis equals energy stasis, which under some situations may be incorrect due to changes in body composition.

Another method involves trials in which animals are fed at different levels of intake and an estimate of retained energy is estimated from comparative slaughter as the difference between final and initial body composition (Lofgreen, 1965; Thomson, 1965). Regression analysis is performed to describe the relationship between energy intake and retained energy. Extrapolation of these data to an intercept which represents zero retained energy ($RE = 0$) yields an estimate of maintenance energy requirements. This method has been used in developing maintenance energy requirements in cattle and sheep (Garrett et al., 1959), as well as, providing a basis for a comprehensive system for expressing energy requirements in beef cattle (Lofgreen and Garrett, 1968). A modified version of this method has been used to estimate energy requirements in pregnant sheep (Rattray et al., 1973) and cattle (Ferrell et al., 1976).

A third method involves the measurement of heat exchange using direct or indirect (closed or open circuit) calorimetry. The latter method has been the most widely implemented means for measuring energy exchange in mammalian species and is based on analysis of expired gases (O_2 , CO_2 and CH_4) corrected for urinary N losses (Brouwer, 1965). The method enables estimation of maintenance or basal energy expenditures in animals at different stages of growth and levels of production. Net energy for maintenance is measured as the fasting heat production during the post-absorptive period in a thermoneutral environment to avoid obligatory corrections for heat increment and energy associated with tissue deposition or thermoregulation. In ruminants a large volume of digesta, relative to nonruminants, is present in the gastrointestinal tract and this may require prolonged periods for clearance of digesta. Blaxter (1962) identified that two criteria used to demonstrate a true fasting state in ruminants were disappearance of methane production and a decrease in the respiratory quotient to values that show no oxidation of carbohydrate. The use of respiration calorimetry is most appropriate for obtaining several acute estimates of maintenance while comparative slaughter is suited for measuring maintenance energy expenditures over an extended period of time.

As mentioned previously, the use of fasting heat production to estimate maintenance energy requirements has been widespread in the nutritional sciences, however, the method has not gone uncriticized. In the fasting state, the animal is maintained by mobilization of body tissue to provide energy. In this condition, heat produced

represents heat lost during mobilization and maintenance of existing tissue and work required to maintain physiological functions. The fact that this condition represents an energy deficit to the animal is indicative by the loss of body energy stores, namely tissue fat and protein. Webster et al. (1974) questioned the relevance of fasting metabolism for failure to define the changes in an animal's metabolism in response to energy deficit. They concluded that an imperfect description of the biological processes required to maintain an animal during fasting metabolism and conditions of growth make comparison of each irrelevant.

The purpose of the previous discussion is not to examine the quantitative estimates of basal metabolism but to briefly describe the various methodologies typically used to obtain such estimates. Even under standardized conditions of measurement, considerable variation exists between and within animals. Many of the factors responsible for such variation are sex, age, biological type and plane of nutrition.

Effect of Age, Sex, Biological Type and Plane of Nutrition

The classical observation by Brody (1945) and Kleiber (1947) is that the interspecies relationship between basal metabolism and body weight is a constant function. However, much of the following evidence suggests that maintenance energy expenditures are influenced by factors such as age, sex and productive potential. A general decline in maintenance energy requirements is observed with increasing age in several species. Maintenance tends to be higher in males