tion studied. The significance of this apparent impermeability of the rat placenta to these azoproteins is discussed.

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Proposed Method for Determining Biological Value of Protein in Ruminants. (24661)

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Little attention has been directed to the biological value of protein for ruminant animals because of the ability of the rumen microflora to synthesize amino acids. With the advent of nutritional interest in non-protein nitrogen, and more recently the interest in chemical stimulation of growth in ruminants, the need for an adequate simplified measure of biological value of protein has become important. Protein digestion and nitrogen retention have been used as methods for determining protein utilization by ruminants because no other satisfactory methods have been available. The Thomas-Mitchell method(1) for biological value determination, which has been generally used as the criterion in monogastric animals, has not proven satisfactory for ruminants. Because of the unique physiological functions of the ruminant digestive tract, accurate measurements of metabolic and endogenous nitrogen, required by the Thomas-Mitchell method, are not obtainable. However, estimates of endogenous and metabolic nitrogen calculated by Harris and Mitchell(2) from body weight and feed consumption, respectively, have been used in biological value determinations in ruminants.

In this investigation, alteration of blood and urinary nitrogen constituents from lambs was studied to develop a simplified method of: (a) predicting the biological value of protein, and (b) determining magnitude of stimulation from anabolic drugs.

TABLE I. Constituents of Experimental Rations, %.

Constituents	CSM	CSM-urea	Urea
Molasses beat pulp	36.5	36.5	36.5
Cotton-gin waste	35	35	35
Barley		12.05	24.1
Cottonseed meal	27.5	13.75	
Urea		1.7	3.4
Salt	1	1	1

Method. Twenty-six lambs were fed 3 rations (Table I) which varied only in source of nitrogen: (a) cottonseed meal, (b) 50% cottonseed meal : 50% urea, and (c) urea.

One-half of the lambs fed each ration was subcutaneously implanted at the base of the ear with 12 mg diethylstilbestrol. Rations were fed *ad libitum* for 10 days, followed by a 10-day period in which rations (17% crude protein) were fed in accord to 2.3% body weight. Blood samples, plus 24-hour urine and fecal samples were obtained from all lambs for analyses of nitrogen constituents. Apparent nitrogen (N) digestion was determined by means of the chromic oxide ratio technic. Thereafter, lambs were fed their respective rations *ad libitum,* and 45-day weight gains were calculated.

Observations. Illinois workers(3) reported that rations containing 11% protein equivalent satisfied the N requirements for maintenance and growth of lambs. Diets that contained 12% protein equivalent derived from urea N resulted in similar biological values

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TABLE II. Summary of Results.

	Diethylstilbestrol			None		
	CSM*	CSM-urea	Urea	CSM	CSM-urea	Urea
Avg body wt, lb	94.7	94.8	90.7	96.3	98.1	83.8
Avg day gain, lb†	.506	.445	.402	.337	.218	.284
Urine constituents:						
Urine N, mg/ml	7.56	5.90	8.26	7.01	9.06	6.06
NH_3N, "	.34	1.26	.52	.26	.47	1.22
Creatinine, "	.648	.605	.753	.458	.543	.479
Creatinine N, "	.248	.226	.281	.171	.203	.179
Total N, "	11.37	11.10	13.40	8.99	11.28	11.06
Creatinine N, %	2.13	2.04	2.09	1.90	1.79	1.61
Apparent N digestibility, %	65.34	69.88	73.40	66.76	66.85	69.35
Creatinine N, % / Apparent N digested	3.26 ±.51	2.92 ±.92	2.84 ±.24	2.84 ±.36	2.67 ±.20	2.32 ±.53

* CSM refers to cottonseed meal. † Calculated for 45 days.

and N retention values in lambs as soybean meal N(3). However, when protein equivalent was increased to 17%, the N from protein sources proved superior to urea N(2,3). Johnson *et al.*(4) emphasized that the biological value of protein from various sources for lambs has been reported by many stations to approximate 60; however, these studies were conducted with lambs fed 10% to 12% crude protein. In the study reported here, 17% protein equivalent rations were fed in accord to body weight, with the attempt to emphasize the biological value difference between sources of dietary N and diethylstilbestrol.

The growth stimulation due to diethylstilbestrol administration (Table II) was similar to the results found elsewhere(5). Furthermore, growth response was similar to the results reported by Light, *et al.*(6), who found that the value of urea was increased when fed to diethylstilbestrol-treated lambs.

Neither creatinine nor non-protein N content of the blood (Table II) were indicative of chemical growth stimulation or source of dietary N. Also, urinary ammonia and urea were not consistently influenced by treatment (Table II). However, total N per ml of urine was increased progressively with increasing amounts of urea in the ration.

Whereas, creatinine concentration/ml of urine was variable, the percentage of total urinary N from creatinine (creatinine N%) was directly related to the expected N utilization and to the actual growth rate of lambs in the respective groups (Table II).

Twenty-four hour urine collections may not be indicative of the average urine excretion in lambs; the short-time duration of urine collection adds extreme variability, particularly if the lambs are not accustomed to metabolism stalls. However, urinary creatinine N% determination does not require the recording of urine volume.

Urinary creatinine N has been considered as endogenous N independent of quantity or quality of dietary protein(7). For the most part, urinary N, other than that from creatinine, must originate primarily from metabolic degradation of dietary amino acids or from the non-protein N that was absorbed via the rumen wall. The percent of total urinary N that originates from creatinine or urinary creatinine N% should be related to the comparative amount of absorbed N that was retained in the body. Further, this measurement deducts body size differences between animals by correcting for endogenous N. For the most part, creatinine N% is indicative of relative amount of absorbed N that is retained by the tissue. Although this measurement does not possess the theoretical exactness of the numerator in the Thomas-Mitchell formula [*N* intake—(fecal N – metabolic N) – (urinary N – endogenous *N*)], it is more applicable for ruminant studies. With dogs, others(8) have obtained a 0.946 and a 0.988 correlation between urinary creatinine N% and the Thomas-Mitchell method of biological value determination.

When urine was obtained from the lambs, 24-hour fecal samples were also collected. Apparent N digestion (Table II), calculated by the chromic oxide ratio technic, was used as the denominator for correcting the respective creatinine N% values. This measurement was related to relative growth response of the lambs. Although metabolic fecal N was not obtained, this calculation stimulates the Thomas-Mitchell formula for determination of biological value. Both methods determine the comparative amount of absorbed N that is retained by the animal. While the Thomas-Mitchell method is most accurate for monogastric animals, the method proposed here provides a simple and practical procedure for determining comparative protein anabolism or biological value of protein in ruminant animals. Further, this method should be of particular value in determining relative duration of influences of relative magnitude of effects of growth-stimulating drugs.

Summary. A method for predicting comparative values of nitrogen sources and anabolic protein stimulation by drugs in ruminants has been proposed. The method requires determination of urinary creatinine N% divided by the apparent N digested (expressed as a decimal) in animals with similar N intake per unit body weight.

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Rapid Method for Determination of Lipid Bound Radioiodine in Blood. (24662)

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Studies of absorption of radioactive fats, and fatty acids, labeled with I^{131} have recently been shown(1,2,3) to provide useful clinical information in certain disorders of the gastrointestinal tract. Although much on radioactive fat absorption has described measurements on fecal collections(4), more recent studies have shown that measurements of radioactivity in blood could also be correlated with clinical status. For example, in idiopathic steatorrhea, pancreatitis, and pancreatic insufficiency, blood levels of radioactivity have been shown to be below values obtained with normal individuals. The ease with which blood radioactivity measurements could be made compared to data obtained from fecal collections, has also provided impetus for evaluation of blood radioactivity. In addition to measuring total I^{131} radioactivity in blood, some investigators(1,2,5) have determined amount of radioactivity in protein-bound form. One report(5) indicated that certain disease states, such as hypercholesteremia, lipemia and coronary artery disease are associated with alteration of ratio of lipid-bound radioiodine to inorganic radioiodine. Interest in protein-bound I^{131} measurements in fat absorption with I^{131} labeled fats, has led us to seek a more rapid procedure than the trichloroacetic acid precipitation of protein. Anion exchange resins, useful in removal of inorganic I^{131} in protein-bound "conversion ratio" studies(6,7,8) might also be suitable for separation of lipoprotein I^{131} radioactivity. If this separation could be performed on whole blood without prior re-