It is clear, then, that a satisfactory extraction technique must somewhere provide for the separation of the *two* groups of growth substances which are active in the *Avena* test. Unless this is done, the *Avena* assay can only approach reasonable accuracy when either group of substances is extremely low in relation to the other.

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Comparative Nutritive Value of Casein and Lactalbumin for Man

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The classic work of Osborne and Mendel (*5*) showed that casein is deficient in cystine and that its companion protein, lactalbumin, is superior in promoting the growth of rats. This finding was promptly (and at that time, perhaps, properly) used in partial explanation of the superiority of breast milk over cow's milk in the feeding of infants (*2*). In confirmation of these early observations, we have recently reported (*1*) that the addition of methionine to an enzymic hydrolysate of casein (amigen) resulted in an accelerated rate of growth in rats, and when the supplemented hydrolysate was given intravenously, it improved nitrogen retention in dogs. When the same type of supplementation studies were made in humans, there was no improvement in nitrogen retention. Four groups of humans were employed: (1) surgical patients fed intravenously, (2) normal infants, (3) normal adults on a maintenance nitrogen level, and (4) normal adults who were protein depleted and fed just enough nitrogen to supply the approximate endogenous needs. Methionine supplementation did not increase the efficiency of nitrogen utilization from the casein hydrolysate.

These findings naturally led us to determine the comparative effectiveness of intact casein and of lactalbumin in promoting nitrogen retention *in man*. Four normal adults were placed on a protein-low diet (*4*) for 12 days, until a constant nitrogen excretion (assumed to be approximately endogenous) had been reached. The nitrogen intake during all depletion periods was 0.008 gram/kg. body weight/day, and for the last 4 of the initial 12 days the average nitrogen loss was 3.22 grams daily. The caloric intake during the entire 40-day study was maintained constantly at 40 calories/kg. body weight.

Four-day periods of supplementation with either casein or lactalbumin were then alternated with 4-day depletion periods. The two proteins were fed alternately, and each was administered for two 4-day experimental periods in order to minimize the effect of antecedent conditions on nitrogen balance. A level of supplementation insufficient to produce positive nitrogen balance was intentionally chosen. This level was 0.033 gram of nitrogen/kg. body weight as the test protein, equal to 2 per cent of the total calories. The average nitrogen balances for each subject for each period are given in Table 1. Subject C contracted a mild type of prevalent "influenza" and could not complete the test.

TABLE 1

NITROGEN BALANCES FOR EACH PERIOD

(Data expressed in grams nitrogen/day)

Periods (4 days each)	Subject				
	A	B	C	D	Avg.
Depletion	−3.92	−2.26	−3.81	−2.90	−3.22
Casein	−.76	−.64	−.83	−.93	−.79
Depletion	−4.13	−2.88	−3.52	−3.00	−3.38
Lactalbumin	−.65	−.47	−.69	−1.40	−.80
Depletion	−3.48	−2.29	−2.81	−2.75	−2.83
Casein	−.74	−.32	−.87	−.79	−.68
Depletion	−4.09	−2.10	—	−2.34	−2.84
Lactalbumin	−1.44	−.05	—	−.66	−.72

These experiments reveal no significant difference between the two proteins in maintaining nitrogen balance in man. The average endogenous value for the four depletion periods was −3.07 grams. The addition of casein spared 2.33 grams of body nitrogen, and the addition of lactalbumin, 2.31 grams. These average values are closer than might again be encountered, but the individual balances were within the range of variation that we have observed in this type of study. All pertinent details of the study will be published elsewhere.

We have approached the calculation of "biological value" with temerity, since, in our experience, such comparative values more often mask than disclose information. Using an average for the four depletion periods as the endogenous level of urinary and of fecal nitrogen excretion, average "biological values" are: for casein, 89.0 and 94.7, respectively; for lactalbumin, 90.2 and 93.3. In spite of the similarity in these final average figures, it should be noted that the variation in the individual values is as great as that observed in experimental animals (*3*).

We have attributed the failure of methionine to improve the nitrogen retention of a casein hydrolysate in man to the fact that man is not covered with hair. Hair contains a high percentage of cystine, and it is logical that the requirement of the rat and the dog for sulfur-containing amino acids should be greater than that of man (*1*). Whether or not this explanation is valid, these data demonstrate that lactalbumin is not superior to casein in promoting nitrogen retention in man.

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