Body Composition: Relative in vivo Determinations from Potassium-40 Measurements

Abstract. *Measurements of body composition from serial, in vivo determinations of potassium-40 were made on 51 obese human subjects who were placed on a low calorie diet. Weight losses in the group averaged from 12 to 36 pounds. There was no change in potassium content or in lean body weight. Weight loss resulted from the loss of excess body fat.*

Techniques for the in vivo estimation of total body potassium by counting of potassium-40 have been used to study the possible relationships between total body potassium and lean body mass (*1*). Recently Forbes *et al.* (*2, 3*) and Anderson and Langham (*1*) have suggested that the measurement of the K^{40} content of man in a whole-body counter can be used to determine the total body fat of man.

Attempts to assign absolute values for the body fat of man by this technique will result in unavoidable errors until the many factors influencing the accuracy of the technique can be assessed. Factors which may contribute substantial errors include body stature (which in turn affects reproducible geometry), bone size, total body water, distribution of muscle and fat within the body, and variation in concentration of potassium in various tissues. There are undoubtedly other factors. Even so, there is a reasonable correlation between values obtained by the various techniques (including chemical analyses), which indicate that a K^{40} determination by liquid- and crystal-scintillation counting techniques is a most useful approach to the in vivo estimation of lean body weight and body fat.

After consideration of the potential applications of scintillation-counting techniques, the fact is that absolute measurements of body composition are not essential for many research and practical applications. It is much more accurate and meaningful to measure differences in the same individual resulting from such factors as differences in nutrition, physical fitness, drug therapy, and disease. This can be accomplished with a high degree of accuracy by serial measurements on the same individual which give relative measurements in which errors are canceled. The technique under these circumstances is a reliably reproducible one, relatively free of errors.

We have studied the effect of nutritionally complete food-substitute therapy on the lean body weight and body fat of cosmetically obese human subjects by whole-body liquid-scintillation counting techniques for relative in vivo measurements of potassium content. The subjects were 35 males, aged 18 to 61 years, whose weights were from 172 to 280 pounds, and 16 females, aged 14 to 61 years, whose weights were from 137 to 263 pounds. All were judged to be in good physical health. Each was placed on a nutritionally complete food-substitute therapy (*4*). The food-substitute therapy consisted of 900 calories per day plus calorie-free liquids and low-calorie, bulk-containing foods as required on an individual basis. The therapy was continued for a period of 8 weeks.

The body weight, total body potassium, lean body weight, and body fat of each individual were determined before initiating the diet regimen, at approximately weekly intervals during the experiment, and after termination of the experiment. The K^{40} content was determined with a 4-pi large-volume liquid-scintillation counter (*5*), and from these data total body K, lean body weight, and body fat were calculated from factors reported by Forbes *et al.* (*2*). The reproducibility of the measurements was determined from values obtained from three individuals of constant body weight over a period of 8 months before the experiment and during the time of the experiment. Repeated measurements of these individuals at approximately weekly intervals over the period gave the same results for values of total K with observed standard deviations of 3.1, 3.5, and 3.3 percent. Accordingly, the K content, the lean body weight, and the body fat in adult humans who had no significant weight change over an extended period of time seem constant within the standard deviations reported.

Seven of the 51 subjects placed on the food-substitute therapy over the 8-week period lost 15 pounds or less with 12 pounds being the lowest, 28 subjects lost 16 to 25 pounds, and 16 lost over 25 pounds; 36 pounds was the greatest loss. In no instance with the weight reductions cited was there any indication of a change in the lean body weight. Observed standard deviations were of the same order of magnitude as, or less than, those of the three control subjects of constant weight except for three female subjects who showed deviations of 4.9, 5.7, and 8.0 percent. However, the individual values of these three subjects showed no trend toward a consistent change in lean body weight. Somewhat higher standard deviations were expected since these subjects had much lower lean body weights than other subjects in the group. This does not

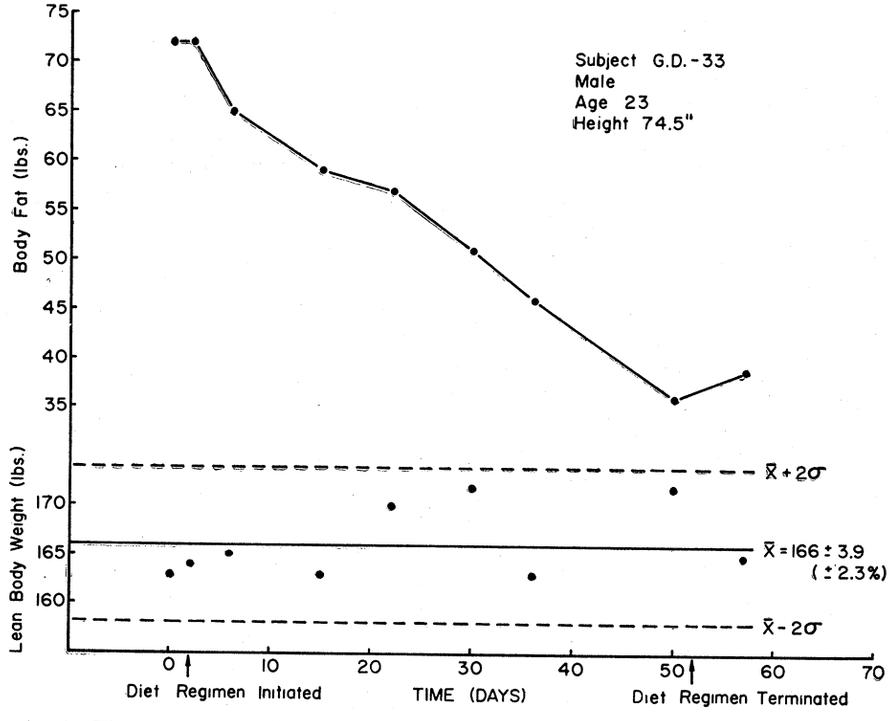

Fig. 1. The lean body weight and fat values of a male subject placed on a complete food-substitute therapy consisting of 900 calories per day for an 8-week period.

explain the high standard deviations completely, but we believe no significance can be attached to the somewhat wider scatter of values than is seen in the other measurements. The K values varied from 71.9 ± 4.1 g, corresponding to a 59-lb lean body weight for a 61-year-old female with an initial body weight of 137 lb, to 233.4 ± 5.3 g, corresponding to a 193-lb lean body weight for an 18-year-old male subject with an initial body weight of 227 lb. The average K value of all the 51 subjects prior to intiating the food-substitute therapy was 153.1 g, and at the end of the 8-week period it was 154.2 g. Since the final average K value is larger than the initial average value, there can be no statistical decrease in the average initial to final measurements. No statistical decrease means that the test statistic would accept the hypothesis of equality of the averages for the probability of rejecting a true hypothesis at least up to the 0.40 level. The lean body weight of each of the 51 subjects was thus considered to remain constant with the total weight loss resulting from loss of excess fat. A typical plot of the lean body weight and fat values for one of the male subjects over the period of the experiment is shown in Fig. 1.

These results confirm the findings reported by Berlin *et al.* (*6*), who studied the body composition of three obese subjects who showed weight losses of 65, 52, and 44 pounds when placed on a restricted calorie intake. Their method consisted of metabolic balance and body-water and body-density measurements. Nitrogen balance studies of obese subjects on weight-reducing regimens described by other authors (*7*) are also in agreement with these results. As cited by Berlin (*6*), a calculation of the density of the net tissue lost in his studies yields values which are not in good agreement with the density values previously reported (*8, 9*) for normal and overweight individuals on a weight-reduction regimen in which lean body mass was reported to be lost. Berlin indicates that data for the density of the tissue lost, when translated in terms of body composition, are in a large part misleading; the loss ought not be accredited to an anatomical entity for which there appears to be no evidence (*8*).

These results show the importance of relative measurements of body composition obtained by serial, in vivo determinations of potassium-40 (*10*).

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X-Ray Sensitivity and DNA Synthesis in Synchronous Populations of HeLa Cells

Abstract. *Inhibition with either 5-fluorodeoxyuridine or deoxyadenosine for specified periods during the division cycle of the HeLa S3 cell shows that the mid-interphase peak in sensitivity occurs just before DNA replication begins. Sensitivity subsequently decreases only after synthesis of DNA is resumed. One interpretation of the relation between fluctuations in sensitivity and in DNA synthesis is that the lethal radiation damage to these cells occurs in DNA.*

Sensitivity of HeLa S3 cells to 220 kev x-rays, as measured by loss of the capacity for sustained reproduction, fluctuates during the division cycle (*1*); synchronized populations display two peaks of sensitivity, one at mitosis and the second about midway through the cycle, 10 hours later (*2*). In this system about 60 percent of the cells have begun to synthesize DNA by the latter time (S phase cells), while the remainder are still in the postmitotic (G_1) phase of the DNA-synthetic cycle (*3*). It was not clear, therefore, whether maximal interphase sensitivity occurs before or after the inception of DNA synthesis. Furthermore, although we suggested (*2*) that the sensitivity fluctuations might be intimately related to the cyclic pattern of DNA synthesis, no evidence was adduced in support of any such relation. We have now measured the x-ray sensitivity of synchronous cell populations in which resolution was increased by inhibiting DNA synthesis for specified periods during the division cycle. Greatest sensitivity occurs just before the start of DNA duplication. In addition, the marked distortions which these treatments produce in the survival pattern correlate strongly with the changes induced in the DNA-synthetic cycle.

Replicate cultures, each containing roughly 10^3 selectively harvested mitotic HeLa S3 cells (*3*), were incubated in medium Nl6HHF (*4*) in plastic dishes, under the usual conditions. At appropriate times the growth medium was replaced with warmed medium containing either of two inhibitors of DNA synthesis: 5-fluorodeoxyuridine (FUdR) (*5*) at a concentration of $10^{-6}M$ (*6, 7*) and deoxyadenosine at $3.2 \times 10^{-4}M$ (or, in one experiment, $4.8 \times 10^{-4}M$) (*8*). All dishes in a given experiment were treated with inhibitor for the same period, irrespective of when they were irradiated. To reverse inhibition by deoxyadenosine, the medium was removed, the plates were rinsed once with Saline G (*4*), and fresh growth medium was added. Inhibition by FUdR was reversed with $10^{-5}M$ thymidine (*6*).

All operations were performed at 37°C, including irradiations which were carried out in single exposures of 300 rads (*9*). Cell survival after irradiation with this one dose is a satisfactory indicator of the changes in sensitivity that occur during the cell-division cycle (*2*). The shape of the dose-survival curves is not altered in the presence of FUdR (*10*) or deoxyadenosine. The criterion for cell survival was growth of a 50-cell colony within 10 days (*11*).

The rate of DNA synthesis was determined in normal synchronous populations and in those inhibited by deoxyadenosine by measuring the incorporation of C^{14}-labeled thymidine (*12*) during 30-minute periods (*3*). Inhibition