Opposite effects of variations in food intake on carbohydrate and fat oxidation in ad libitum fed mice

J.P. Flatt

Department of Biochemistry, University of Massachusetts Medical School, Worcester, MA, USA

Daily carbohydrate and fat oxidation rates were determined during 160 consecutive days in 10 female CD1 mice. The animals had free access to a running wheel and to one of two synthetic diets whose composition resembled either lab chow or a mixed western diet. Carbohydrate oxidation was found to be correlated positively with daily variations in food intake, matching carbohydrate intake closely, regardless of the diets' macronutrient contents (R = .83 and .72, respectively). By contrast, fat oxidation was correlated negatively with daily variations in food intake on each diet, being inhibited by carbohydrate intake (R = −.65). Such results reflect the priority given in metabolic regulation to the maintenance of carbohydrate balance over the maintenance of fat balance, as well as over energy balance, which was linked very tightly to the latter (R = .91 and .95). Fat oxidation appeared to be determined by the gap between overall energy expenditure and the sum of carbohydrate plus protein energy ingested (R = .9), rather than by the amounts of fat consumed. These findings thus illustrate fundamental differences in the control of carbohydrate and fat metabolism, which must be taken into account to understand body weight regulation and maintenance.

Keywords: carbohydrate oxidation; carbohydrate balance; fat oxidation; fat balance; food intake

Introduction

The regulation of the energy balance is a central issue in understanding the control of body weight and the causes of obesity.[1-5] Generally, it is accepted that daily nitrogen balances will oscillate around the mean corresponding to an even nitrogen balance when diets supplying more than enough protein and appropriate amounts of energy are consumed.[6] In studying energy balance regulation, only data on the sums of carbohydrate and fat intakes and oxidations are usually available and considered. This is sometimes justified by the belief that excess carbohydrate can be transformed readily into fat by de novo lipogenesis. However, fat synthesis in man can only be induced by ingestion of very high doses of carbohydrate for several consecutive days,[7] and lipogenesis does not provide a significant escape from the carbohydrate pool under usual dietary conditions.[8] In man, fat synthesis will not exceed the concomitant rate of fat oxidation, even after the ingestion of test meals providing 500 g of carbohydrate,[9] demonstrating that glycogen stores are maintained spontaneously in a range far below their maximal capacity. Since glucose cannot be made from fatty acids, carbohydrates and fats are in effect in separate compartments, and weight maintenance depends on the organism's ability to achieve balance between intake and oxidation in each.[10] To understand the metabolic phenomena which determine for which body composition weight maintenance tends to become established, it is necessary to be able to recognize separately how carbohydrate and fat balances are achieved.[1,11]

Maintenance of carbohydrate balance is possible by adjustment of carbohydrate oxidation to carbohydrate intake (plus the glucose yielded by gluconeogenesis), as evidenced by the ease with which man and animals adapt to imposed nutrient intakes, regardless of the amounts of carbohydrate which they supply. Under free-feeding conditions, the maintenance of carbohydrate balance may be further facilitated by corrective

Address reprint requests to J.P. Flatt, Department of Biochemistry, University of Massachusetts Medical School, Worcester, MA 01655, USA.
Received October 25, 1989; accepted September 24, 1990.

changes in food intake, which are correlated negatively with the previous day's carbohydrate and fat balances.[10,11] Deviations from the carbohydrate balance exert a more powerful impact on the next day's food intake than deviations from the fat balances.[10,11]

A new method which allows accurately determining 24 hr respiratory exchanges in mice[12] was used to establish daily and cumulative carbohydrate, fat, and energy balances in 10 ad libitum fed female CD1 mice during 160 consecutive days. The results show that carbohydrate balance is maintained more accurately than fat balance, a fact which previously could be inferred only over the long-term. Particularly noteworthy is the fact that daily variations in the amounts of food consumed were found to exert opposite effects on carbohydrate and fat oxidation. Thus, carbohydrate oxidation was correlated positively with daily food (and hence carbohydrate) intake, whereas fat oxidation was correlated negatively with daily food (and hence fat) intake. These fundamental differences in the manner in which carbohydrate and fat balances are achieved need to be taken into consideration in trying to understand the regulation of food intake and the mechanisms involved in the maintenance of stable body weights.

Materials and methods

The study was conducted on female CD1 mice purchased from the Charles River Corporation. They were maintained on lab chow up to 90 days of age and then placed on one of two synthetic diets prepared by TEKLAD (Madison, WI) to contain 18% of their energy as casein. One of these was a "high carbohydrate diet" with a macronutrient distribution similar to lab chow (69% of total energy as carbohydrate, 13% as fat), whereas the other was a "mixed diet," formulated to contain 37% of its energy as carbohydrate and 45% as fat, which is in the range of macronutrient distribution typical for western diets. The carbohydrates in both diets were provided half in the form of sucrose and half as corn starch; both contained 10 grams of cellulose per 1000 kcal. When the mice were 130 days old, their weights had stabilized and indirect calorimetry and food intake measurements were initiated and continued for 160 days without interruption. The animals were housed individually in conventional plastic cages containing a running wheel, linked to a counter to determine the number of revolutions which the animals chose to perform each day. They had free access to water and to the diet to which they had adapted. Each cage was placed into a 55-gallon plastic drum, which can be sealed hermetically with a plexiglass cover through which they were exposed to a 12 hr light-dark cycle. This set-up allows measurement of the respiratory exchanges of individual mice during an entire day (during which the P_{CO_2} increases to 1.2-2%, while the P_{O_2} declines from 21% to 19-20%), except for a 45 to 60 minute period every day in the early afternoon (when food intake is low), to permit venting (with a fan), cleaning, and measuring of body weights and food consumption. The experimental procedures, the exact composition of the diets, and the calculations involved in determining daily carbohydrate and fat oxidations and balances are described in detail in an acompanying paper.[12] After one month, the diets were switched; two months later, the animals were returned to their initial diet for the last two months of the experiment. When the diets were switched, the mice transferred from the high carbohydrate to the mixed diet gained weight; this was associated with deposition of body fat, as shown by cumulating the daily fat balances determined by indirect calorimetry. Conversely, the animals which where changed from the mixed diet to the high carbohydrate diet lost weight, mostly as fat. These changes were reversed when the mice were returned subsequently to their initial diets. (The time course of these changes is shown in *Figure 2.*)[12]

Results

Data obtained by indirect calorimetry allow establishing of changes in the body's carbohydrate, fat, and protein contents over a given period of observation, but it is common practice to refer to these as rates of substrate oxidation. When this is done, one may encounter negative values for fat oxidation, which indicate that more fat was synthesized from carbohydrate than oxidized. This is the case whenever non-protein Respiratory Quotients (RQs) are greater than 1.0 (i.e., whenever overall RQs excede .96 on diets containing 18% protein) on a given day. The relationships between daily variations in food intake and rates of carbohydrate and fat oxidation on the two diets are shown in *Figure 1.* The regression lines obtained for each of the 10 mice on the two diets form rather tight bundles and they are all statistically significant ($P \leq 0.0001$, except for two cases where $P < 0.001$ and $P = 0.01$). Essentially identical conclusions are obtained when the 800 daily measurements obtained on each diet with the ten different mice are pooled (cf. equations shown in inserts in the various panels of *Figure 1*). Subsequent data analysis, therefore, were performed on these data pools. In this instance, they show positive correlations between carbohydrate oxidation and food intake with R^2 values of .69 and.52 ($P \leq 0.0001$), whereas fat oxidation is correlated negatively with daily variations in energy intake ($R^2 = .42$ and .08; $P \leq 0.0001$).

It is of interest to note and to understand the reason for the difference in the slopes observed for the two diets. This becomes evident by considering *Figure 2,* where carbohydrate and fat oxidation are shown in relation to carbohydrate and fat intake, rather than total energy intake. (In *Figure 2* and in subsequent figures, parameters in the regression equations pertaining to the high carbohydrate or to the mixed diet are identified by 'prime' or 'double-prime' symbols, respectively.) The slopes relating carbohydrate oxidation to carbohydrate intake are now nearly the same for the two diets (i.e., .76 and .73, respectively; upper panel of *Figure 2*). This indicates that the effect of

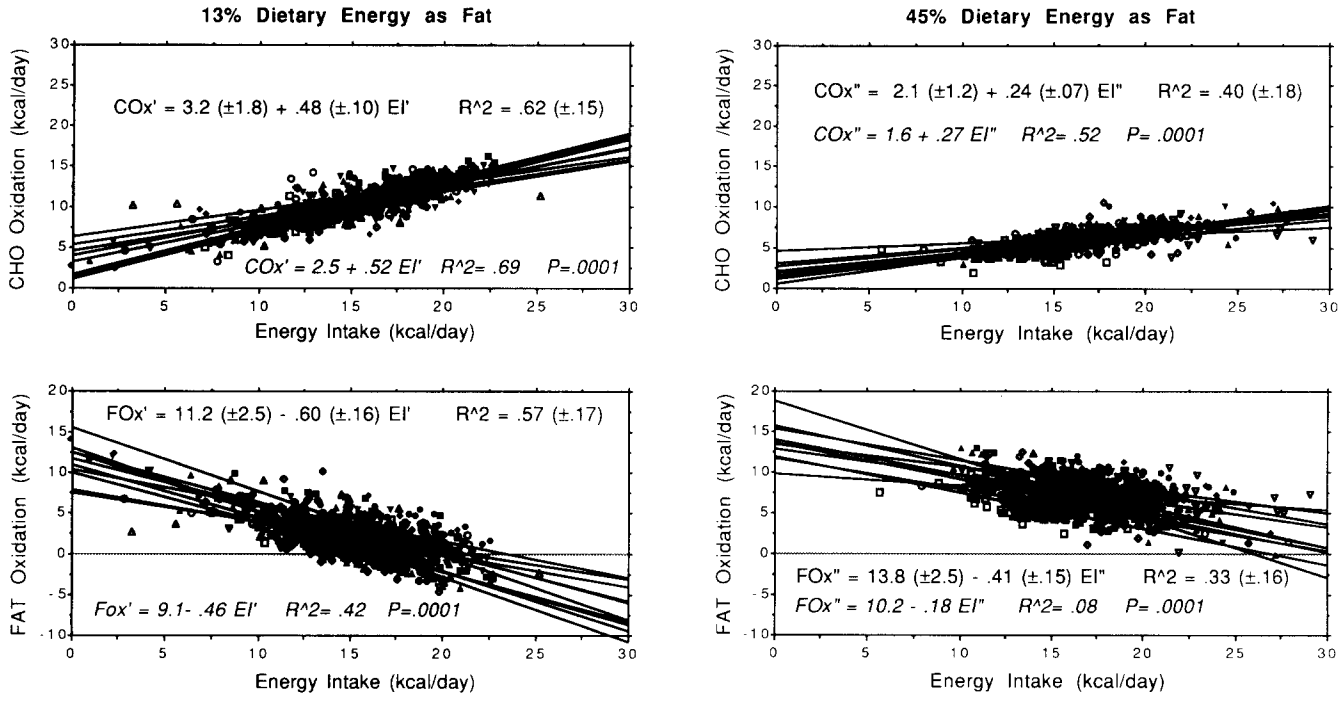

Figure 1 Relationships between daily rates of carbohydrate (upper) and fat oxidation (lower panels) and energy intake in ad libitum fed mice, when maintained on a high carbohydrate diet containing 13% (left) or on a mixed diet with 45% (right) of dietary energy as fat. The numerical coefficients are the means (± SD) of those obtained by separate regression analysis of the data obtained for each of 10 mice or those obtained for the pooled data from all 10 mice.

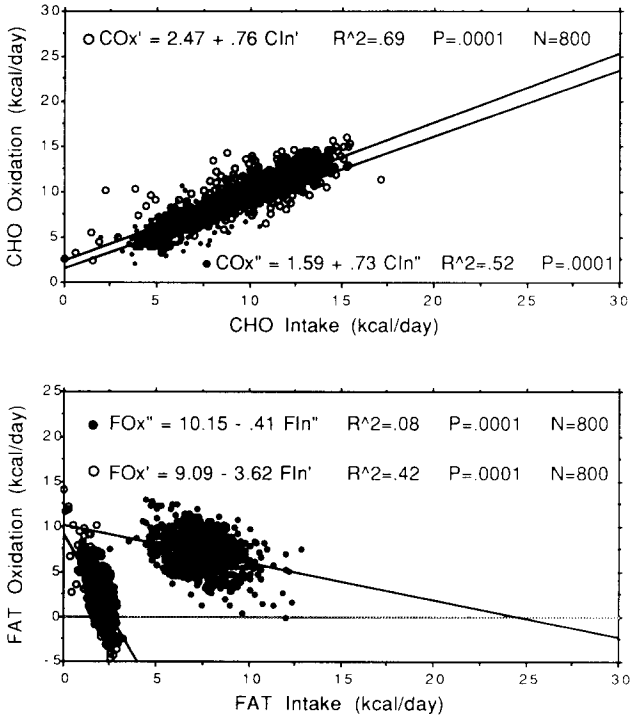

Figure 2 Relationships between daily carbohydrate (upper) and fat oxidation rates (lower panel) and carbohydrate or fat intake in 10 ad libitum fed female CD1 mice, when maintained on a high carbohydrate diet containing 13% (○), or on a mixed diet with 45% (●) of dietary energy as fat. It is possible to pool the 2 sets of 800 data pairs only for the carbohydrate oxidation data; the correlation equation obtained in this case is: COx = 0.77 + 0.90 CIn; R^2 = 0.85; $P \leq 0.0001$.

carbohydrate intake on carbohydrate oxidation is independent of diet composition, and that one can expect this phenomenon to manifest itself regardless of a diet's carbohydrate content. By contrast, the slopes of the negative correlations relating fat oxidation to fat intake on the two diets remain very different (lower panel of *Figure 2*). This reflects the fact that fat oxidation is not determined by fat intake, but rather that fat oxidation is inhibited by carbohydrate intake, as shown in the upper panel of *Figure 3*. Accordingly, average rates of fat oxidation are "allowed" to be greater on a mixed diet than on a high carbohydrate diet, and average rates of fat oxidation thereby can be commensurate with the fat content of various diets. This is illustrated by the position of the two clusters of fat oxidation rates in the lower panel of *Figure 2*, and is consistent with the fact that weight maintenance can occur on diets with widely differing fat contents.

The inhibition of fat oxidation by increases in food intake appears to be less pronounced on the mixed diet (R^2 = .08, cf. lower panel of *Figure 2*). This is due to the fact that variations in spontaneous running activity have a substantial effect on fat oxidation when the mixed diet is consumed. The impact of these parameters on carbohydrate and on fat oxidation when this diet is consumed, therefore, was assessed by multiple regression analysis (*Table 1*). As can be seen from the first equation, exercise does not alter the fact that carbohydrate oxidation is determined primarily by carbohydrate intake. Beyond the influence which running activity and body weight may have on the

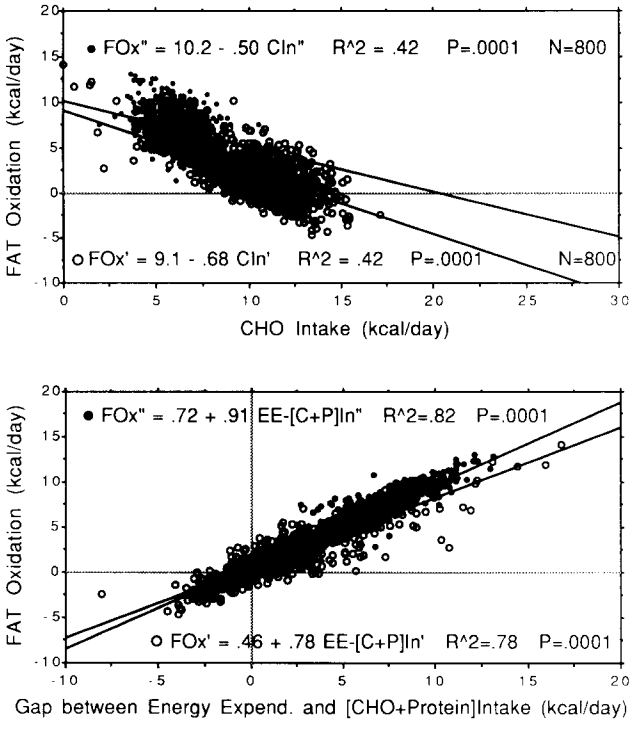

Figure 3 Fat oxidation rates as a function of carbohydrate intake (upper), or of overall energy expenditure minus protein plus carbohydrate energy intake (lower panel) in 10 ad libitum fed female CD1 mice, when maintained on a high carbohydrate diet providing 13% (○) or on a mixed diet with 45% (●) of dietary energy as fat. The data plotted in the lower panel for the two diets can be pooled, which yields the following correlation: Fox = 0.41 + 0.92 EE − [C + P]In' R^2 = .95; $P \leq 0.0001$.

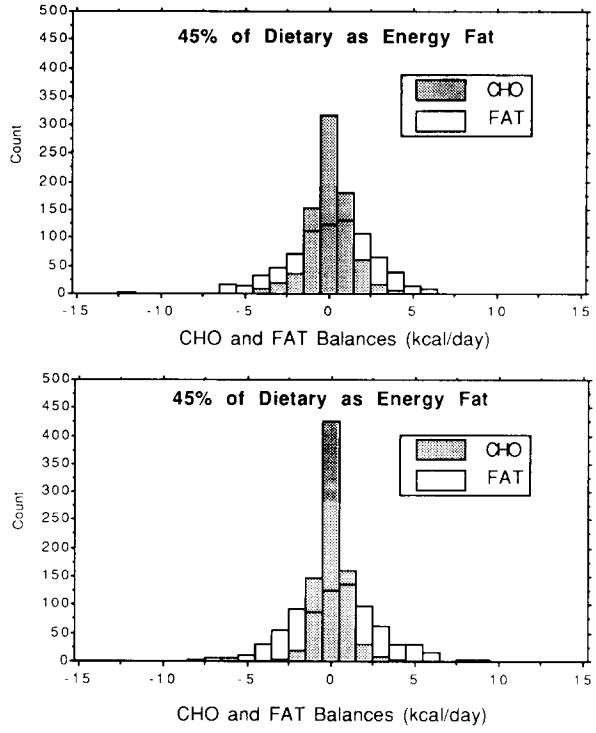

Figure 4 Comparison of daily carbohydrate and fat balances by frequency distribution in 10 ad libitum fed female CD1 mice, when maintained on a high carbohydrate diet providing 13% (upper), or on a mixed diet with 45% (lower panel) of dietary energy as fat (N = 800).

amounts of food consumed, they have only a barely recognizable effect on carbohydrate oxidation. By contrast, fat oxidation is substantially enhanced by high body weight and running activity, but not by fat intake, as shown by the second equation. When relevant parameters other than fat intake are taken into consideration, variations in fat oxidation now appear to be predictable to an extent (R^2 = .41), comparable to that for carbohydrate oxidation (R^2 = .52). Since body weights are relatively stable and the intensity of running rather characteristic for a given animal, the R^2 values obtained for individual mice are in fact substantially higher (means ± SD = 0.33 ± 0.17) than the R^2 value of .08 obtained on the pool of data from all ten mice. In effect, the oxidation of fat corresponds to the gap between total energy expenditure minus the sum of carbohydrate plus protein intake (lower panel of *Figure 3*). This occurs with a rather remarkable consistency (R^2 = .8), regardless of the proportions of carbohydrate and fat in the diets; it is indeed an appreciably more precise relationship than that offered by considering only inhibition of fat oxidation by carbohydrate intake (compare the two panels of *Figure 3*).

Thanks to the metabolic responses which operate to adjust carbohydrate oxidation to carbohydrate intake, the organism can maintain carbohydrate balance with relative accuracy. On about half of the days, the deviations from an even carbohydrate balance are less than 0.5 kcal (*Figure 4*). Given the lack of impact of fat intake on fat oxidation, deviations from even fat balances can be quite considerable, particularly when fat is a substantial component of the diet (lower panel of *Figure 4*). Errors in the fat balance are smaller on the high carbohydrate diet (compare the 2 panels of *Figure 4*), on account of its rather low content. But even on this diet, where fat provides only 1/5 as much

Table 1 Effects of running, body weight, and food intake on carbohydrate and fat oxidation[a]

(1) CHO oxidation = 1.24	+ 0.01 x 10-4 Revolutions/day	+ 0.011 BWt	+ 0.71 CHOintake
R^2 = .52	± 0.03 x 10-4 [P = .6]	± 0.006 [P ± .05]	± 0.03 [P = 0.0001]
(2) FAT oxidation = 4.0	+ 1.09 x 10-4 Revolutions/day	+ 0.16 BWt	− 0.63 FAT(or- 0.77 CHO)intake
R^2 = .41	± 0.06 x 10-4 [P = .0001]	± 0.01 [P = .0001]	± 0.04 [P = 0.0001]

[a] Mice had free access to a running wheel and to a synthetic diet providing 45% of its energy as fat. Intakes and oxidations are in terms of kcal/day; body weight is in grams (N = 800).

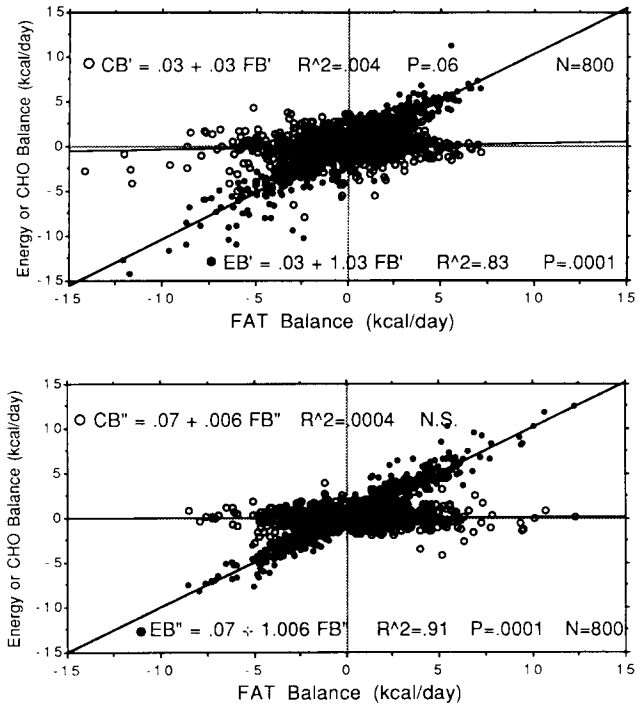

Figure 5 Relationships between daily energy (•) or carbohydrate (○) balances and fat balances in 10 ad libitum fed female CD1 mice, when maintained on a high carbohydrate diet providing 13% (upper panel), or on a mixed diet with 45% (lower panel) of dietary energy as fat.

energy as carbohydrate, deviations from an even fat balance remain larger than the deviations from carbohydrate balance. Furthermore, it is of interest to note that carbohydrate and fat balances are essentially uncorrelated (open circles in the 2 panels of *Figure 5*). Thus, fat accumulation does not require, nor is it dictated by, positive carbohydrate balances. Finally, it is apparent that deviations from energy balance (black dots in the 2 panels of *Figure 5*) are explained almost entirely by gains or losses of fat, the slope of the two regression lines being almost exactly equal to one (1.03 and 1.006), with R^2 values of .83 and .91 on the high carbohydrate and the mixed diet, respectively.

Discussion

It seems reasonable to consider that the marked asymmetry in the metabolic regulation of carbohydrate and fat metabolism, which manifests itself so strikingly in our experimental results, reflects the fact that glycogen reserves are much smaller than fat reserves, and that maintenance of glycogen stores within a physiologically desirable range has commanded much greater priority in the biological evolution of metabolic control mechanisms.[3,11] Increases in carbohydrate oxidation have the effect of limiting excessive expansion of the glycogen stores, whereas prompt declines in carbohydrate oxidation occur when carbohydrate intake and/or glycogen levels become low. They reach truly minimal rates during total starvation, when gluconeogenesis from amino acids and triglyceride-glycerol are the only sources of glucose.[13] Thus, respiratory quotients (RQs) tend to approach .7 when carbohydrate intake is curtailed (*Figure 6*). On the other hand, RQs will rise when food intake is high and more carbohydrate must be oxidized to limit glycogen accumulation. Thus, high RQs will then prevail (*Figure 6*), a fact from which one can learn to appreciate that fat oxidation on days with high food intakes is even less than average, further adding to the amounts of fat to be stored on such occasions.

If the difference in the regulation of carbohydrate and fat metabolism observed in mice is indeed a pattern dictated by the disparity in the size of the body's glycogen and fat reserves, a similar metabolic behavior should be expected to prevail in man. Indeed, recently it was reported that 24-hr carbohydrate balances are maintained more accurately than fat balances in human volunteers, and that only the latter correlate with the overall energy balance,[14,15] just as illustrated in *Figure 5*. The prompt rise of the RQ after food ingestion, even when the meal provides nutrients in the same proportions as the fuel mix oxidized before food intake,[16] reveals the power of the regulatory responses which control the composition of the fuel mix oxidized. The organism can achieve much more in modifying the composition of the fuel mix oxidized than in altering energy expenditure. The overall purpose of its endocrine and enzymatic regulatory responses evidently is directed primarily at adjusting carbohydrate oxidation to the supply of carbohydrate. This is evident also from the fact that the post-prandial RQ rise is primarily a function of the amount and nature of the carbohydrates consumed,[9,17] whereas it is not measurably affected by variations in the amounts of fat provided by the meal.[16]

As noted in considering the results shown in *Figures 1–3*, fat oxidation is determined by other factors than dietary fat intake. Overall substrate oxidation is coupled to ATP expenditure. Therefore, if the organism does indeed tend to maintain spontaneously carbohydrate and protein balances, one should expect the rate of fat oxidation to correspond approximately to

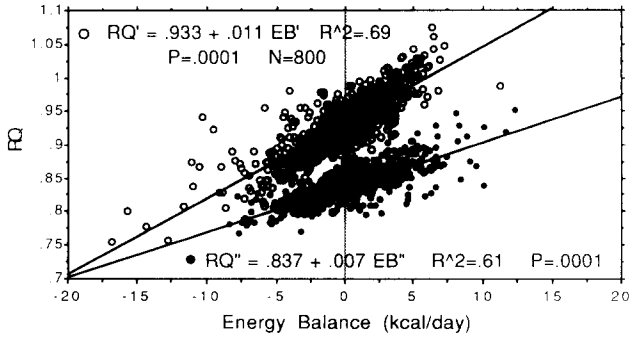

Figure 6 Relationships between 24-hr Respiratory Quotients (RQs) and energy balances in 10 ad libitum fed female CD1 mice, when maintained on a high carbohydrate diet providing 13% (○), or on a mixed diet with 45% (•) of dietary energy as fat.

the difference between overall energy expenditure and the sum of carbohydrate and protein energy consumed. This quite clearly is the case (cf. lower panel of *Figure 3*). Furthermore, this phenomenon appears to be valid for diets of different compositions, just like the finding that carbohydrate oxidation is determined by carbohydrate intake, regardless of diet composition (*Figure 2*). The high power of the correlations shown in *Figure 3* (R^2 = .8) suggests that the reactions involved in the storage, mobilization, and oxidation of fat are not in a position to exert much influence on short-term adjustments of substrate oxidation. White adipose tissue appears to have enough enzymatic capacity and flexibility to handle variations in fat influx and efflux as needed and dictated by demands created elsewhere. Such a view would be consistent with the finding that adipose cells transplanted from lean to genetically obese mice, or vice versa, tend to become small or large depending on the environment encountered in the host (i.e., lean or obese), regardless of their genetic origin.[18] This does not preclude that the size of the adipose tissue mass, and the degree of insulin resistance which it may elicit, may play a role in influencing the average RQ and hence the body composition for which weight stability tends to become established.[10]

RQs are measured frequently as they provide an easily obtained measure of the relative proportions of carbohydrate and of fat in the fuel mix oxidized. As noted in considering *Figure 6*, the spontaneous adjustment of carbohydrate oxidation to carbohydrate intake leads to high RQs during periods of positive energy balance, while relatively low RQs prevail during periods of energy deficits. But RQ values are obviously also markedly influenced by the carbohydrate content of the diet. In order to use RQ values in assessing how the proportions of carbohydrate and fat in the fuel mix oxidized compare to the relative proportions of carbohydrates and fat in the diet, it is convenient to compare the RQ to the "Food Quotient" (FQ). The FQ is defined as the ratio of CO_2 produced to oxygen consumed during the oxidation of a representative sample of the diet consumed,[19] it being assumed that the relative distribution of macronutrients absorbed is about the same as in the diet. Knowing whether the average RQ is greater, equal, or smaller than the FQ allows predicting of whether the energy balance is positive, even, or negative (*Figure 6*). This rule can be generalized and made independent of diet composition, by expressing the energy balance in terms of the ratio of energy intake divided by energy expenditure, and relating this to the RQ/FQ ratio.[11] This relationship (R^2 = .63, *Figure 7*) provides a useful concept to consider the problems of weight maintenance and weight control, in another manner[10,19] than by the usual arguments based on energy balance. It reveals that average energy balance is only sustained when the RQ is equal to the FQ (i.e., when the composition of the fuel mix oxidized is equivalent in the average to the macronutrient distribution in the diet). This helps to recognize measures which may be conducive

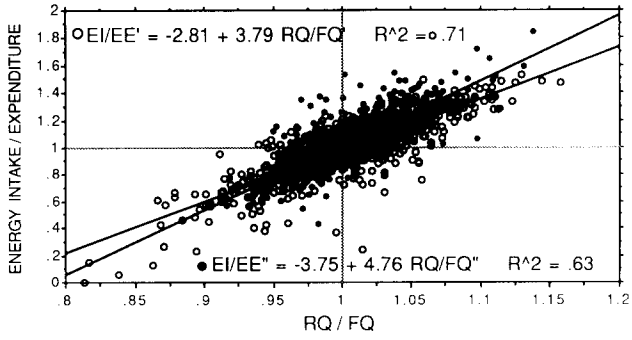

Figure 7 Relationships between Energy Balance (expressed as the ratio of Energy Intake/Energy Expenditure) and the RQ/FQ ratio in 10 ad libitum fed female CD1 mice, when maintained on a high carbohydrate diet containing 13% (○), or on a mixed diet with 45% (●) of dietary energy as fat. When the data obtained on the two diets are pooled, the correlation is described by: EI/EE = −2.80 + 3.76 RQ/FQ; R^2 = .63; P = 0.0001.

to weight loss, since measures that cause the RQ to become lower than the FQ will also be effective in causing a negative energy balance. As one must expect it to be more difficult to maintain an RQ equal to (or lower than) the FQ when the FQ is low, it becomes understandable why diets with a rather low FQ (i.e., diets with a substantial fat content) are more conducive to the development of obesity than high carbohydrate diets with a high FQ.[11,15,20-23] One can also understand why sustained exercise of low to moderate intensity is most likely to help in limiting[21] or reversing excessive accumulation of body fat,[11] since this type of exertion is most effective in lowering the RQ, and thereby in increasing the fat content of the fuel mix oxidized.[10]

In conclusion, data providing information on the carbohydrate and fat sectors of metabolism, rather than merely on overall energy expenditure and energy intake, show that carbohydrate oxidation is correlated positively, whereas fat oxidation is correlated negatively with daily variations in food intake. The fundamental difference which this implies in the manner by which carbohydrate and fat balances have to be achieved must be taken into consideration in trying to formulate concepts about the regulation of energy balance.

Acknowledgments

The collaboration of K.E.G. Sargent and B.R. Krauss is gratefully acknowledged. Support for these studies was provided by NIH Grant DK 33214.

References

1 Bray, G.A. (1987). Obesity: A disease of nutrient or energy balance? *Nutrition Reviews* **45**, 33–43.
2 Sims, E.A.H. and Danforth, E. Jr. (1987). Expenditure and storage of energy in man. *J. Clin. Invest.* **79**, 1019–1925.
3 Mayer, J. and Thomas, D.W. (1967). Regulation of food intake and obesity. *Science* **156**, 328–337.
4 Stunkard, A.J. (1988). Some perspectives on human obesity: Its causes and treatment. *Bull. NY Acad. Sci.* **64**, 902–940.

5 Bouchard, C. and Pérusse (1988). Heredity and body fat. *Ann. Rev.* **8,** 259–277.

6 Sukhatme, P.V. and Margen, S. (1978). Models for protein deficiency. *Am. J. Clin. Nutr.* **31,** 1237–1256.

7 Acheson, K., Schutz, Y., Bessard, T., Anantharaman, K., Flatt, J.P., and Jéquier, E. (1988). Glycogen storage capacity and de novo lipogenesis during massive carbohydrate overfeeding in man. *Am. J. Clin. Nutr.* **48,** 240–247.

8 Passmore, R. and Swindells, Y.E. (1963). Observations on the respiratory quotients and weight gain of man after eating large quantities of carbohydrate. *Brit. J. Nutr.* **17,** 331–339.

9 Acheson, K.J., Schutz, Y., Bessard, T., Ravussin, E., Jéquier, E. and Flatt, J.P. (1984). Nutritional influences on lipogenesis and thermogenesis after a carbohydrate meal. *Am. J. Physiol.* **246,** E62–E70.

10 Flatt, J.P. (1988). Importance of nutrient balance in body weight regulation. *Diabetes/ Met. Rev.* **4,** 571–581.

11 Flatt, J.P. (1987). Dietary fat, carbohydrate balance, and weight maintenance; effects of exercise. *Am. J. Clin. Nutr.* **45,** 296–306.

12 Flatt, J.P. (1991). Assessment of daily and cumulative carbohydrate and fat balances in mice. *J. Nutr. Biochem.* **2,** 193–202.

13 Cahill, G.F., Jr. (1970). Starvation in man. *N. Engl. J. Med.* **282,** 668–675.

14 Abbott, W.G.H., Howard, B.V., Christin, L., Freymond, D., Lillioja, S., Boyce, V.L., Anderson, T.E., Bogardus, C., and Ravussin, E. (1988). Short-term energy balance: Relationship with protein, carbohydrate and fat balances. *Am. J. Physiol.* **255,** E332–E337.

15 Schutz, Y., Flatt, J.P., and Jéquier, E. (1989). Failure of dietary fat to promote fat oxidation: A factor favoring the development of obesity. *Am. J. Clin. Nutr.* **50,** 307–314.

16 Flatt, J.P., Ravussin, E., Acheson, K.J., and Jéquier, E. (1985). Effects of dietary fat on post-prandial substrate oxidation and on carbohydrate and fat balances. *J. Clin. Invest.* **76,** 1019–1024.

17 Jenkins, D.J.A., Wolever, T.M.S., Collier, G.R., Ocana, A., Rao, A.V., Buckley, G., Lam, Y., Mayer, A., and Thompson, L.U. (1987). Metabolic effects of a low-glycemic index diet. *Am. J. Clin. Nutr.* **46,** 968–975.

18 Ashwell, M. (1985). The use of the adipose tissue transplantation technique to demonstrate that abnormalities in the adipose tissue metabolism of genetically obese mice are due to extrinsic rather than intrinsic factors. *Int. J. Obes.* **9, Suppl. 1,** 77–82.

19 Flatt, J.P. (1978). The biochemistry of energy expenditure. In *Recent Advances in Obesity Research II* (G.A. Bray, ed.), pp. 211–228, Technomic Publishing Co. Inc., Westport, CT.

20 Schemmel, R., Michelson, O., and Tolgay, Z. (1969). Dietary obesity in rats: Influence of diet, weight, age, and sex on body composition. *J. Physiol.* **210,** 373–379.

21 Pitts, G.C. and Bull, L.S. (1977). Exercise, dietary obesity, and growth in the rat. *Am. J. Physiol.* **232,** R38–R44.

22 Lissner, L., Levitski, D.A., Strupp, B.J., Kalkwarf, H.J., and Roe, D.A. (1987). Dietary fat and the regulation of energy intake in human subjects. *Am. J. Clin. Nutr.* **46,** 886–892.

23 Tremblay, A., Plourde, G., Despres, J.P., and Bouchard, C. (1989). Impact of dietary fat content and fat oxidation on energy intake in humans. *Am. J. Clin. Nutr.* **49,** 799–805.