Effect of Exercise and Protein Intake on Energy Expenditure in Adolescents*

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In order to evaluate the influence of physical exercise and protein intake on Resting Metabolic Rate (RMR) and Postprandial Energy Expenditure (PEE), 16 healthy, normal-weight, 15 year-old, adolescent males at the same stage of pubertal development were studied. They were assigned to two dietary groups receiving the same energy intake (1.3 x by measured RMR) and different proportions of macronutrients (13 % protein, 39 % fat, 48 % CHO in Group A; 30 % protein, 32 % fat, 38 % CHO in Group B). An increase in postprandial energy expenditure, relative to basal, was observed in all individuals. The postprandial energy expenditure was higher in group B than in group A. Postprandial Post-exercise Thermogenesis (expressed as Kcal/3 h) was significantly higher in group B than group A (p<0.05). Although the RMR on the test day was not different between the groups, the RMR on day 2 was significantly higher than on day 1 in group B (p<0.01). In group B, the post-exercise RQ was significantly lower than the preexercise RQ (p<0.01). It is concluded that in normal-weight-adolescents, a hyperproteic diet followed by moderately-intensive exercise induces increases in EE and decreases in RQ in the postprandial post-exercise period and is accompanied by increase in the RMR the following day.

Key words: Hyperproteic diet, Postprandial energy expenditure, Indirect calorimetry, Adolescents, Resting metabolic rate.

Total Energy Expenditure can be divided into three components: 1) Basal Energy Expenditure (BEE), 2) physical activity and 3) thermogenesis which includes the increased rate of resting metabolism in response to such stimuli as a meal, cold exposure, certain drugs or stress.

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Exercise may induce negative energy balance through one or more of the following mechanisms: by directly enhancing EE (cost of the exercise per se); by affecting the other two components of EE (BEE and postprandial thermogenesis) and by affecting energy intake. Exercise might change the Resting Metabolic Rate (RMR) as an acute phenomenon and/or as a part of the long-term adaptation to training. Although the acute effect to exercise on RMR is generally accepted (7), discrepancies exist in relation to magnitude and duration of this post-exercise increase in VO2. Potentially, any effect of exercise on RMR could be very important in total energy expenditure represented by the RMR.

Exercise may affect the thermic effect of food either acutely or as a chronic phenomenon associated with the training effect. Several investigators have studied whether physical exercise influences Postprandial Thermogenesis (PT). Some studies were unable to show this additional effect of exercise on PT (23, 26) whereas others suggest that exercise raises PT when closely followed by a meal (28, 29). Some of the differences might be explained by the type of exercise, exercise intensity and type of meal ingested. Many studies suggest that PT, or the increase in Postprandial Energy Expenditure (PEE) relative to RMR, depends on the energy content of

Table I. Anthropometric characteristics (means ± S.D.) of the subjects at the beginning of the study. All comparisons between groups are not significantly different. Group A and group B received a "balanced" and a hyperproteic diet respectively (n = 8).

Group A	Group B
56.1±7.6	59.2±8.3
163±3.0	166±8.0
21.2±2.8	21.5±2.9
25.2±2.9	25.6±1.8
17.3±8.4	17.3±9.5
11.5±4.8	12.7±5.9
19.9±6.6	20.7±7.4
44.6±4.1	46.6±4.7
	56.1±7.6 163±3.0 21.2±2.8 25.2±2.9 17.3±8.4 11.5±4.8 19.9±6.6

the meal (10, 17). The influence of the macronutrient composition of the meal on PT remains controversial (1, 10). However, it could seem logical that a larger proportion of protein in the diet may induce a higher PT since, from the stoichiometric aspect, the expenditure that protein absorption represents, its metabolism to urea and glucose or towards new protein synthesis is higher than in other metabolic pathways (18). If either the physical exercise or a larger proportion of protein in a meal raises the PT, then the conjunction of the two should have and additive effect on PT.

The objective of this study was to evaluated the influence of physical exercise and protein intake on RMR and PEE to determine new modes of treatment for obesity.

Materials and Methods

 Sixteen healthy, adolescent, Subjects. -15 year-old, male volunteers were studied. They were selected from a previous study (8), were of normal body weight and at stage 3 of puberty as described by TANNER (32). All subjects were healthy with no personal or family history of diabetes mellitus or other metabolic diseases (table I). Highly aerobically-trained individuals were not accepted into the study so as to eliminate possible gross variations in metabolic response resulting from differences in levels of cardiorespiratory fitness. The individuals were randomly assigned into two groups on the basis of the diet administered. Fully informed written consent to participation was obtained from the parents and the study was approved by the Hospital's Ethical Committee.

Test-Meals. — Figure 1 shows the study design. For each subject, the evening Test Meal (TM) on the day prior to the study consisted of 600 Kcal in the form of a liquid formula (Pentadrink®, Nutrition Laboratories, Zoetermeer, Holland). The protein source of the TM was casein derivates. On the day of the test, each individ-

ual was given 1.3-fold the previously-determined RMR calculated by indirect calorimetry for half an hour and using WEIR's formula (35). This excess-to-requirement was to ensure that maintenance body weight during the test period (37). The

Aerobic fitness test. — Submaximal aerobic fitness was determined 3 days before the study by continuous, multi-stage exercise test on a cycle ergometer (ERG 550 Bosch, Germany). Prior to the submaximal test, time was allotted for the subjects to famil-

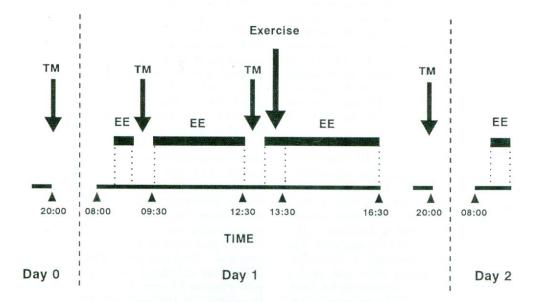


Fig. 1. Study design.

Time of the study protocol is represented at the bottom of the figure. Energy Expenditure (EE) at basal (day 1 and day 2), postprandial and postprandial post-exercise conditions were measured. The little arrows indicate the Test Meal (TM) administration and the big arrow indicates 30 min exercise test period.

energy requirement was calculated and administered in three isocaloric aliquots; two of which were administered in the metabolic lab during the day and the third one the same night at home. In group A the energy intake was supplied in the form of a liquid formula. Group B was given the same liquid formula but with 10 g/100 ml of a dietary supplement composed of a protein solution (Clinical Nutrition, Mataró, Spain). The energy content of the test meal was 792 ± 83 Kcal (13 % protein, 39 % fat, 48 % carbohydrate) and 795 ± 113 Kcal (30 % protein, 32 % fat, 38 % carbohydrate) in groups A and B respectively.

iarize themselves with cycling on the ergometer at a constant pedalling rate while breathing through the apparatus used for metabolic measurements. The subjects began pedalling at 50 rpm with zero external resistance. The work-rate was increased in 25 W stages every two minutes until heart rate was ≥170 beats/min (defined as Physical Working Capacity «PWC170») (34). This test measures the work required by a subject when his cardiac frequency reaches 170 beats/min. This system is considered better for measuring the aerobic resistance capacity than VO₂max in adolescents (33). A linear relationship exists between physical effort and cardiac frequency at the fre-

quencies between 100 to 170 beats/minute level. A relationship between pulse and oxygen uptake during physical activity has also been reported (19). Hence the cardiac frequency is a useful parameter for pre-

dicting oxygen uptake.

From the results of the multi-stage exercise test and by regression analysis, the intensity of physical activity required to reach a cardiac frequency of 170 beats/min (PWC170) was calculated. On the day of the test, the exercise intensity was adjusted according to each subject's level of aerobic fitness and the level of physical exercise was maintained at 70 % of the PWC170 for a period of 30 min.

Energy Expenditure. — EE was measured by open circuit indirect calorimetry (SensorMedics Energy Expenditure Unit 2900, Holland) which includes a mass flowmeter transducer, a Bechman zirconium O2 analyzer and a Bechman LB-2 nondispersive infrared CO2 analyzer. The subjects breathed through a Hans-Rudolph non-rebreathing valve and used a face mask during the exercise period. During the postprandial and post-exercise period, exhaled air was collected using a hood. This system allows the continuous measurement of oxygen uptake (VO2), the production of carbon dioxide (VCO2) and the Respiratory Quotient (RQ = VCO₂/ VO2). Before and after each test, O2 and CO₂ sensors were calibrated using different gas mixtures of known N₂, O₂ and CO₂ concentrations. Periodically, the precision of the measurement was confirmed (24, 25). To calculate the energy expenditure, the following formula was used (6): EE(Kcal) = 3.581 VO₂(L) + 1.448 VCO₂(L) 1.773 urinary nitrogen (g/24h).

The urinary nitrogen was determined by the Kjendhal method (16) on the urine collected during 24 hours of the test day. On the night prior to the test, the subjects consumed the assigned 600 Kcal liquid formula diet (fig. 1). At 8 a.m. the following

morning, and after a period of at least 30 min of absolute rest, the VO2 and VCO2 at basal conditions were measured every minute for 30 minutes by indirect calorimetry and from which the RMR (day 1) was calculated. Following this, the first part of the test meal was consumed within 30 min and the VO₂ and VCO₂ were measured over the subsequent three hours. The PEE was calculated from the area under the curves of the EE measurements and expressed in Kcal/min. PT was calculated as [postprandial EE area (Kcal/3 h)] – [RMR Kcal/min) × 180 min] and expressed as Kcal/3 h or as a function of the energy contained in the test meal.

The second part of the test meal was then consumed over a 30 minute period and the VO₂ and VCO₂ were again measured over a 30 minute exercise period and for the following three hours of recuperation. From these data, the postprandial post-exercise energy expenditure was calculated and expressed as Kcal/min. Post-prandial Post-exercise Thermogenesis (PPT) was calculated from [postprandial post-exercise EE area (Kcal/3 h)] - [RMR Kcal/min) × 180 min] and expressed as Kcal/3 h or as a function of the energy contained in the test meal. Also the RQ in the basal, postprandial and the postprandial post-exercise states were calculated. At 20:00 h, the subjects consumed the third aliquot of the meal. At 8 a.m. the following morning the VO2 and VCO2 at basal conditions were repeated from which RMR (day 2) was calculated.

The calorimetric measurement was performed while the subject rested quietly watching television. Particular attention was given to prevent any extra movements of the subject since this contributes to in-

crease resting energy expenditure.

Anthropometric measurements. — Skin fold measurements were made at the triceps, biceps, suprailliac and subscapular regions. Chest size, height and weight were measured using the Holtain apparatus. The

measurements were made by a single investigator according to a standardized protocol. Body fat was estimated using the following equation of GARCÍA-LLOP et al. (15): -27.98 + (25.30 × log of the sum of the four skin-fold (mm) measurements + (0.2345 × BMI), where BMI (Body Mass Index) = body weight (Kg)/height² (m). The Fat-Free body Mass (FFM) was calculated by subtraction of the estimated fat mass from the total body weight.

Analysis of data. — Statistical analyses were conducted using the SPSS/PC package. For the comparison of means the Mann-Whitney U-test was used for unpaired data and the Willcoxon test for paired data. All values at the 0.05 % level were considered significant.

Results

There were no significant differences in weight, height, BMI or any other anthro-pometric measurement between the pometric measurement between the groups A and B (table I). The mean RMR on day 1 was not significantly different between the two groups (table II) when expressed either as Kcal/min or as Kcal/kg of FFM/h (1.69 \pm 0.07 Kcal/kg FFM/h and 1.64 ± 0.06 Kcal/kg FFM/h in groups A and B respectively).

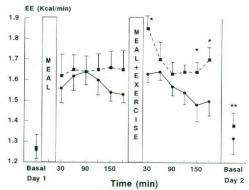


Fig. 2. Basal (day 1), Postprandial, Postprandial post-exercise and Basal (day 2) energy expenditure throughout the study period in relation to groups studied (13 % and 30 % of energy as protein in groups A (•) and B (■), respectively).

* p < 0.05 vs group A; ** p < 0.01 BEE day

1 vs BEE day 2 in group B.

Postprandial Pre-exercise Period. — In all individuals, a statistically significant increase in PEE relative to basal was observed (fig. 2). In group A, EE was maximal at 90 minutes after test-meal ingestion whereas in group B the augmentation was observed within the first 30 minutes and was maintained constant throughout the following three hours. In both groups, PEE at 180 minutes was significantly higher than basal values.

PEE, expressed as a percentage increase relative to basal values, was consistently

Table II. Comparisons of EE parameters (means \pm S.E.M.) during pre-exercise and post-exercise period between Group A ("balanced" diet) and group B (hyperproteic diet)

	Group A	Group B	"P"
RMR Day 1/(Kcal/min)	1.26 ± 0.05	1.27 ± 0.06	ns
Postprandial Thermogenesis		1121 2 0100	110
(Kcal/3 h)	54.2 ± 5.4	61.1 ± 8.9	ns
(% energy intake)	6.9 ± 0.8	7.9 ± 1.2	ns
Postprandial EE (Kcal/min)	1.56 ± 0.05	1.61 ± 0.07	ns
Postprandial Post-exercise Thermogenesis			710
(Kcal/3 h)	51.2 ± 5.7	69.4 ± 6.3	P < 0.05
(% energy expenditure)	6.6 ± 0.8	9.0 ± 1.0	ns
Postprandial Post-exercise EE (Kcal/min)	1.54 ± 0.05	1.66 ± 0.06	ns
RMR Day 2 (Kcal/min)	1.32 ± 0.06^{a}	1.38 ± 0.06^{b}	ns

^a(ns) and ^b(p < 0.01) vs RMR Day 1 in respective groups.

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higher in group B relative to group A, the differences being greater after 120 minutes. PEE increases relative to basal were 23.8 \pm 4.3 %, 28.6 \pm 2.9 %, 30.1 \pm 3.5 %, 27.0 \pm 2.9 %, 22.2 \pm 2.3 % and 21.4 \pm 2.6 % at 30, 60, 90, 120, 150 and 180 minutes in group A and 27.4 \pm 1.0 %, 30.4 \pm 4.4 %, 29.6 \pm 4.9 %, 30.4 \pm 5.7 %, 30.8 \pm 4.4 % and 29.9 \pm 4.2 % for group B. All postprandial and postprandial post-exercise measurements were significantly different (p<0.001) from Resting Metabolic Rate.

Postprandial Thermogenesis, expressed as Kcal/3 h or as a percentage of energy intake, was higher in group B than in group A. These differences failed to reach statis-

tical significance (table II).

Postprandial Post-exercise Period. — PPEE is consistently higher in group B than group A. Statistically significant differences were observed at 30, 150 and 180 minutes (fig. 2).

PPT (expressed as Kcal/3 h) was significantly higher in group B than group A (ta-

ble II) (P<0.05).

Pre-exercise vs Post-exercise Period. — No significant differences were found in PEE with respect to PPEE nor PPT with respect to PT for the two groups (table II).

In group A, when comparing the postprandial RQ pre and postexercise, a statistical difference was only observed at the 60 min period, whereas in group B, the

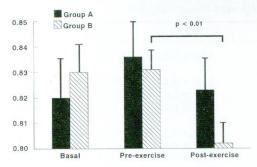


Fig. 3. Respiratory Quotient values during the study period.

postprandial post-exercise RQ was significantly lower than the postprandial post-exercise RQ at 60, 90, 120 at 180 min time points. These differences were consistent when comparing the RQ area under-the-curve (p<0.01) (fig. 3).

In group B, the RMR on day 2 was significantly higher than the corresponding

value on day 1 (table II).

Discussion

Total and basal energy expenditure is well documented in lean and obese adolescents (11) but a relative paucity of information exists regarding the other components of energy expenditure in humans particularly during the growth phase as in our study. An important part of the adolescent energy is expended in tissue synthesis and growth; the latter being the principal factor that distinguishes between adult and adolescent metabolism.

The two groups investigated in the present study were homogeneous with respect to age, pubertal stage and other anthropometric parameters. The RMR (day 1) observed in adolescents in this study were similar to that described by DIETZ et al. (11). The RMR on the first day of the study was not significantly different between the groups expressed as Kcal/day or as

Kcal/Kg FFM/h.

In the present study, to compare the effect of a «balanced» diet as opposed to a hiperproteic diet the relative proportions of protein-derived calories were 13 % and 30 % in groups A and B respectively as compared to BELKO et al. (1) who, varying the protein proportion of the diet, showed that the maximum PT was achieved in adults with a test meal containing 30-45 % of calories derived from protein. The exercise intensity was adjusted according to each individual's level of aerobic fitness to preclude contribution of cardio-respiratory fitness. All the thermogenesis measurements were performed on the same day to

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minimise the 4-7 % day-to-day variability in basal energy expenditure (26) on the measured parameters.

Postprandial Pre-exercise Period. — In each individual a significant increase in EE was observed at 30 minutes postprandially, corresponding to PT, and which remained high for the three hours of the test period. These results are similar to those obtained by SEGAL et al. (27) in adults indicating that adolescents' PT could extend well beyond a three-hours period. Subjects with an intake of high protein and low carbohydrate derived energy showed an earlier, sustained increase in PEE than did individuals who had received a more «balanced» diet. The same results were obtained when PT was evaluated as Kcal/3 h or as a function of the energy contained in the test meal. These findings were not statistically significant and may have resulted from limitations in the study design. Increasing the numbers of individuals studied in each group or by extending the study period to six hours or more would have made the differences clearer but such protocol was not acceptable to our subjects' parents. A physiological explanation could be that the diet of group B contained more energy derived from protein, whereas the energy derived from carbohydrates was less than in group A. The highest effect on PT is protein, followed by carbohydrates, with fats having a minor effect, accounting for 28-30 %, 9-26 % and 4 %, respectively (18). Hence the increased protein intake in group B may augment the difference in proportional contribution of protein and carbohydrate to the PT.

Of the macronutrients, carbohydrate utilization produces the highest RQ and this could explain the observation of a lower RQ in group B (high protein diet) than in group A (high carbohydrate diet) in this period (14).

Postprandial Post-exercise Period. — Several conflicting assessments of the effect of exercise on PT in obese and non-obese subjects under various experimental conditions have been reported (1, 4, 5, 9, 23, 26, 29-31, 36). While a number of investigators have demonstrated that exercise facilitated PT in lean (4, 20, 28, 29, 31) and obese subjects (3), other investigators (9, 23, 36) have found no changes. SEGAL et al. (30) found that postprandial thermogenesis was facilitated by exercise in lean women and men while BRADFIELD et al. (3) showed that this effect occurred in obese subjects, but only after the completion of exercise.

In the present study, hyperproteic diet administered prior to exercise raises the Postprandial post-exercise energy expenditure. Different mechanisms can explain the PEE increase during the post-exercise recovery period. Of primary importance is the high energy cost of storage of carbohydrate as glycogen in the liver and muscles (13). Stoichiometric calculations indicate that 2 mol of ATP are required per mol of glucose transformed into glycogen, i.e. 5.3 % of the energy content of glucose (13). Cessation of exercise is characterised by a rapid increase, within 2-10 minutes, in insulin levels and an elevation of muscle sensitivity to insulin (12). Consequently, mobilisation of glucose stores is observable as an increase in EE. On the other hand, MILLWARD et al. (21) observed that during the postprandial post-exercise period there is a higher contribution of protein oxidation than in basal conditions. While exercising, protein synthesis is inhibited and subsequently compensated in the post-exercise period (21). Hence, the administration of a hyperproteic meal prior to exercise would facilitate a higher protein turnover due to the greater availability of the substrate. This higher turnover would be observed as an EE rise. In addition exercise increases circulating levels of thermogenic hormones (adrenalin and nor-adrenalin) causing an increase in body temperature. Further more, as a mechanism for enhancing metabolic control, substrate cycles may be activated to process the various metabolites which increase during and after exercise (22). This could account for the 12 to 24 hour postexercise rise in energy expenditure observed (22).

In the two groups of adolescents, postprandial post-exercise RQ was lower than in the postprandial pre-exercise period. This finding was similar to that observed in adults by others (2, 5) as to the fact that exercise slightly decreased the RQ. In group B, the postprandial post-exercise RQ was significantly lower thant the preexercise period. This suggests that the hyperproteic and low carbohydrate diet intake, before physical activity, increases fat utilisation.

In our study, in group B a consistent increase of RMR on day 2 in relation to day 1 was observed, suggesting that there was a prolonged effect of the meal with exercise on energy expenditure. The influence of a hyperproteic diet-with-exercise on PPT represents a smaller effect on the total energy balance, while its relative influence on RMR is more important since the RMR represents a larger percentage of the total

In conclusion, in the present study of normal-weight-adolescents, the administration of a hyperproteic diet before moderately-intensive exercise induces increases in EE and decreases in RQ in the postprandial post-exercise period together with an increase in the RMR the following day. These results would suggest that the inclusion of physical activity together with a hyperproteic diet in an obesity treatment program would not only favour the conservation or increase of FFM in the subject but would also increase EE and fat utilization during the post-exercise recovery period.

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Resumen

Se evalúa la influencia del ejercicio físico y de la ingesta proteica sobre el Gasto Energético Basal (GEB) y el Gasto Energético Postprandial (GEP), en 16 varones sanos, adolescentes de 15 años de edad en el mismo estadio de desarrollo puberal, en normopeso. Se reparten al azar en dos grupos que reciben la misma cantidad de energía (1,3 × GEB medido) y diferente proporción de macronutrientes (grupo A: 13 % proteínas, 39 % lípidos, 48 % hidratos de carbono, grupo B: 30 % proteínas, 32 % lípidos, 38 % hidratos de carbono). En todos los individuos se observa un aumento en el GEP respecto al GEB, el cual es superior en el grupo B. La termogénesis postprandial post-ejercicio (Kcal/3 h) es significativamente superior en el grupo B (p<0,05). Aunque el GEB en el día 1 no se diferencia entre los dos grupos, el del día 2 es superior en el grupo B (p<0,01), siendo en este grupo, el cociente respiratorio post-ejercicio, significativamente menor respecto al cociente respiratorio pre-ejercicio (p<0,01). Se concluye que, en adolescentes en normopeso, una dieta hiperproteica seguida de un ejercicio físico de intensidad moderada, induce un aumento del GE y una disminución del cociente respiratorio durante el período postprandial post-ejercicio, acompañándose de un aumento del GÉB del día siguiente.

Palabras clave: Dieta hiperproteica, Gasto energético postprandial, Calorimetría indirecta, Adolescentes, Gasto energético basal.

References

- Belko, A. Z., Barbieri, T. F. and Wong, E.: Am.
- J. Clin. Nutr., 43, 863-869, 1986. Bielinski, R., Schutz, Y. and Jéquier, E.: Am. J. Clin. Nutr., 42, 69-82, 1985. Bradfield, R. B., Curtis, D. E. and Margen, S.:
- Am. J. Clin. Nutr., 21, 1208-1210, 1968.
- Bray, G. A., Whipp, B. J. and Koyal, S. N.: Am. I. Clin. Nutr., 27, 254-259, 1974.
- Broeder, C. E., Brener, M., Hofman, Z., Paijmans, I. J. M., Thomas, E. L. and Wilmore, J. H.: *Int. J. Obesity*, 15, 95-104, 1991.

- Bursztein, S., Elwyn, D., Askanazi, J. and Kinney, J. M.: In «Energy metabolism, indirect calorimetry, and nutrition». Williams and Wilkins. Baltimore, 1989. pp. 27-84.
- Calles-Escandón, J. and Horton, E. S.: Am. J.
- Clin. Nutr., 55, 533S-7S, 1992. Clivillé, R., Fernández-Ballart, J., Salas, J., Feliu, A., González-Hidalgo, R. and Martí-Henneberg, C.: Rev. Esp. Pediatr., 46, 495-501, 1990.
- Dalloso, H. M. and James, W. P. T.: Br. J. Nutr., **52**, 65-72, 1984.
- Dauncey, M. J. and Bingham, S. A.: Br. J. Nutr.,
- 50, 1-13, 1983.Dietz, W. H., Bandini, L. G. and Schoeller, D.
- A.: J. Pediatr., 118, 146-149,1991. Felig, P. and Wahren, J.: N. Engl. J. Med., 20, 1078-1084, 1975.
- Flatt, J. P.: In «Recent advances in obesity re search II». (Bray, G. A., ed.). Newman Publ.
- London, 1978. pp. 211-228.

 14. Flatt, J. P.: In «Rapport annuel 1991» (Jéquier, E., ed.). Fondation Nestlé pour l'étude de problèmes de l'alimentation dans le monde, Lausanne, 1991, pp. 53-76.
- García-Llop, L. A., Sarria-Chueca, A., Mollerón, M. P. and Bueno, M.: Rev. Esp. Pediatr., 46, 155-160, 1990.
- Hawk, P. B. and Bergeim, O.: Practical Physiological Chemistry (10th ed.). P. Blakiston's Son & Co, Philadelphia, 1931.
- 17. Hill, J. O., Heymsfield, S. B., McMannus, C. and DiGirolamo, M.: Metabolism, 33, 743-749, 1984.
- Jéquier E.: Encyclopédie Médico-Chirurgicale. Nutrition, Editions Techniques, Paris, 1980.
- Mellerowicz, H.: In «Ergometría» (Mellerowicz, H., ed.). Panamericana. Buenos Aires,
- 1984, pp. 53-70. Miller, D. S., Munford, P. and Stock, M. J.: *Am.* J. Clin. Nutr., 20, 1223-1229, 1967.

- Millward, D. J., Davies, C. T. M., Halliday, D., Wolman, S. L., Matthews, D. and Rennie, M.:
- Fed. Proc., 41, 2686-2691, 1982. Newsholme, E. A. and Leech, A. R.: Biochemistry for the Medical Sciences. John Wiley & Sons. Chichester, 1983.
- Pacy, P. J. Barton, N., Webster, J. D. and Garrow, J. S.: Am. J. Clin. Nutr., 42, 764-768, 1985.
- Salas-Salvadó, J., Barenys-Manent, M., Recasens, M. A. and Martí Henneberg, C.: Int. J. Obes., 1993 (in press).
- Salas-Salvadó, J., Molina, J., Figueres, J., Massó, J., Martí-Henneberg, C. and Jiménez, R.: Pediatr. Res., 33, 112-117, 1993. Schutz, Y., Bessard, T. and Jéquier, E.: Am. J.
- Clin. Nutr., 45, 1424-1432, 1987. Segal, K. R., Edaño, A. and Thomas, M. B.: Metabolism, **39**, 985-992, 1990.
- Segal, K. R. and Gutin, B.: Metabolism, 32, 581-589, 1983.
- Segal, K. R., Gutin, B., Nyman, A. M. and Pi-Sunyer, F. X.: *J. Clin. Invest.*, **76**, 1107-1112,
- Segal, K. R., Presta, E. and Gutin, B.: Am. J. Clin. Nutr., 40, 995-1000, 1984.
- 31. Stock, M. J.: Eur. J. Appl. Physiol., 43, 35-40, 1980.
- Tanner, J. M.: Growth at adolescence. Blackwell 32.
- Scientific Publications. Oxford, 1973.
- Tuxworth, W.: Med. Sport. Sci., 148, 13-21, 1988.
- 34 Wahlund, H.: Act. Med. Scand., 215, 1-78, 1948.
- Weir, J. B.: Physiol., 109, 1-9, 1949. 35.
- Welle, S.: Am. J. Clin. Nutr., 40, 990-994, 36. 1984.
- World Health Organization. Energy and protein requirements: report of a joint FAO/ WHO/UNU expert consultation. N.º 724. World Health Organization, Geneva, 1985.