Effect of Intermittent Feeding on Glucose-Insulin Relationship in the Chicken

JEAN SIMON AND GABRIEL ROSSELIN *
*Station de Recherches Avicoles (Institut National de la Recherche Agronomique), Centre de Recherches de Tours, 37380 Nouzilly, France; and * Unité de Recherches de Diabétologie et d'Etudes Radioimmunologiques des Hormones Protéiques, U. 55 (Institut National de la Santé et de la Recherche Médicale), Hôpital Saint Antoine, 75012 Paris, France*

ABSTRACT The effects of training to various rhythms of intermittent total starvation (ITS) or intermittent protein starvation (IPS) on the plasma glucose and the plasma insulin levels were studied in the growing chicken. Both types of feeding improved the glucose tolerance in spite of a decrease in the insulin response. After an oral glucose load, plasma free fatty acids showed opposite variations to plasma insulin and plasma glucose. The insulin released in response to a test meal was unchanged. In the ITS 1-1 group (1 day fasting–1 day feeding cycles), low glycemia-low insulinemia were observed during the fasting period of the cycle and high glycemia-hyperinsulinemia during the repletion period in response to the "adaptive hyperphagia." In the IPS 1-1 group (1 day feeding with the protein free diet–1 day feeding with the balanced diet cycles), glycemia was sustained at a high level during both periods of the cycle and insulinemia was depressed by feeding with the protein-free diet and highly stimulated by refeeding with the balanced diet. Therefore, in the chicken, intermittent feeding increases the insulin sensitivity of target tissues and modifies the B-cell sensitivity to glucose. The highest decrease in B-cell sensitivity to glucose was obtained with the protein free diet which further emphasizes the glucose-amino acid synergism previously observed for insulin release. J. Nutr. *109:* 631–641, 1979.

INDEXING KEY WORDS chicken · periodicity of eating · protein glucose tolerance · insulin · insulin sensitivity · insulin release

The effect of irregular and spaced food intake has been studied by numerous authors in different species (1–5). Such an interest was warranted in view of the clinical implications (4) and practical applications for animal production where feed restrictions and growth retardation are of interest, i.e., for growing pullets and aged broilers (6, 7). Thorough metabolic studies in the rat (8–16), the mouse (17), the hamster (18), and the chicken (19–23) have shown that the long term effects of spaced food intake result in hyperphagia during the repletion periods and a stimulation of lipogenesis and enzyme activities in liver and adipose tissue. These modifications allow the organism to store nutrients rapidly during refeeding after a period of restriction. Whereas in the rat, insulin is thought to be one, if not the main hormone, responsible for this metabolic adaptation (15, 24–30), this role has not been recognized in the chicken, mainly because it is generally thought that the chicken is re-

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TABLE 1

Composition of the diets

Balanced diet (experiments 1, 2, and 3)		Protein-free diet (experiment 3)	
Ingredients	Percent	Ingredients	Percent
Ground yellow corn	64	Glucose	53
Norway fish meal (72% protein)	8	Corn starch	32
Soybean meal (50% protein)	19	Corn oil	3
Dehydrated alfalfa meal	2	Cellulose	6
Corn oil	3	Mineral[3] and vitamin[4] premix	6
Dicalcium phosphate	1.5		
Shell limestone	1.3		
Mineral[1] and vitamin[2] premix	1.2		
Calculated metabol·energy content (kcal/kg)	3,180		3,200
Calculated protein content (%)	21		—

[1] Mineral mixture (%): experiment 1: iodinated sodium chloride 0.41; trace elements 0.2; experiment 2 and 3: iodinated sodium chloride 0.04; sodium carbonate 0.2, and trace elements 0.2. The trace element mixture contained (g/kg): Cu 1.4; Fe 14; I 0.7; Mn 43.3; Zn 38.6 and Co 0.135. [2] Vitamin mixture (g/100 kg diet): retinyl propionate (50,000 IU/g) 16; cholecalciferol (100,000 IU/g) 1; calcium pantothenate 0.7; nicotinic acid 1.2; vit B_{12} (1/10,000) 1; menadione 1; tocopherol (25%) 21; BHT 12.5; choline (25%) 400; riboflavin 0.4; $MnSO_4 \cdot H_2O$ 35.2; DL-methionine was added to vitamin mixture to the following extent: 100 g/100 kg diet (experiment 1) and 120 g/100 kg diet (experiments 2 and 3). In experiments 2 and 3 these ingredients were premixed with ground yellow corn to make 760 g. [3] Mineral mixture (g/100 kg diet): $CaCO_3$ 1,480; K_2HPO_4 758; iodinated sodium chloride 230; Na_2CO_3 242; $MgSO_4 \cdot 7\ H_2O$ 500; $CaHPO_4 \cdot 2H_2O$ 1,700; $MnSO_4 \cdot H_2O$ 33; $FeC_6H_5O_7 \cdot 5H_2O$ 33; $KAl(SO_4)_2 \cdot 12H_2O$ 1; $Na_2SeO_3 \cdot 5H_2O$ 0.055; NaBr 2.4; $ZnCl_2$ 8; $Na_2SiO_3 \cdot 9H_2O$ 5.5; $CoSO_4 \cdot 7H_2O$ 0.2; $NaMoO_4$ 0.9; $CuSO_4 \cdot 5H_2O$ 2; H_3BO_3 0.9; stabilized KI 4. [4] Vitamin mixture (g/100 kg diet): retinyl propionate (50,000 IU/g) 32; cholecalciferol (100,000 IU/g) 2; tocopherol (25%) 30; thiamin 2; Menadione 1; riboflavin 3; pyridoxine 3; calcium pantothenate 10; nicotinic acid 20; folic acid 0.6; biotin 0.1; choline (25%) 600; ascorbic acid 50; *p*-aminobenzoic acid 50; inositol 50; vit B_{12} (1/10,000) 100; BHT 12.5; mineral and vitamin mixtures were premixed with cellulose to make 6,000 g.

sistant to insulin (31). However, we have shown that 1) chicken insulin is more potent than insulins from other species (32–33), 2) insulin receptors, although less numerous than in rat, are also present in chicken tissues (34), and 3) in chickens previously fed ad libitum, insulin release is controlled by the same stimuli as those operating in mammals, i.e., glucose, glucose-amino acid synergism, and the entero-insular axis (35). In this paper, we have investigated to what extent insulin is involved in the metabolic adaptation of the chicken accustomed to various rhythms of intermittent total starvation and intermittent protein starvation.

MATERIAL AND METHODS

Animals and diet. One-day old male chickens (heavy&"meat type" breeds) were obtained from a commercial hatchery. They were housed in individual wire cages provided with individual feeders and water bowls in a controlled environment (temperature and humidity) room. The room was lit continuously during the first 48 hours of life and thereafter, 14 hours per day from 0600 to 2000 hours. Chickens were fed a balanced or a protein-free diet whose compositions are indicated in table 1.

Experimental conditions. Two types of feed restriction were studied: (a) intermittent total starvation (ITS) where chickens were intermittently starved then fed the balanced diet ad libitum (experiments 1 and 2) and (b) intermittent protein starvation (IPS) where chickens were intermittently offered the protein-free diet then the balanced diet (experiment 3). The chickens submitted to ITS were trained to a 2-4 starvation-repletion cycle, i.e., 2 days of starvation-4 days of ad libitum feeding

(ITS 2-4 group) or to a 1-1 starvation-repletion cycle (ITS 1-1 group). The chickens submitted to IPS were trained to 1 day of feeding with the protein-free diet-1 day of feeding with the balanced diet cycles (IPS 1-1 group). These cycles result in similar growth rates (about 80% of controls) but different body composition (21–22, 36). As compared to controls body fat content is increased in the ITS 2-4 and IPS 1-1 groups (21–22, 36). Experimental groups contained from 2 to 8 chickens. Actual values are given in the figures.

In experiment 1 (ITS 2-4), 2-week old chickens (Hybro breed) were divided into two groups (control and ITS 2-4) selected for equal body weight (216 ± 2 g). The control group was fed the balanced diet ad libitum throughout the experiment. After 48 days of experiment, at the end of the repletion period and after one night fast (16 hours), chickens were submitted to an oral glucose tolerance test. Six days later, after one extra restriction-repletion cycle and one night fast (16 hours), chickens were fed a test meal of 20 g which represented about 1/6 to 1/8 of their daily food intake then sampled every 30 to 90 minutes.

In experiment 2 (ITS 1-1), 12-day old chickens (Hubbard breed) were divided into two groups (control and ITS 1-1) according to their body weight (188 ± 1 g) and body weight gain (106 ± 0.8 g) between 5 and 12 days. The control group was fed the balanced diet ad libitum throughout the experiment and ITS 1-1 every other day. On the 31st and 32nd days of the experiment, samples of blood were taken at the indicated times in order to measure the variations of plasma glucose and plasma insulin levels in response to fasting and refeeding. In order to characterize the long term effect of changes in the nutritional state, blood samples were taken starting at 8 hours after starvation and 2 hours after re-feeding. After 42 days of experiment and one night of fasting (16 hours), chickens were subjected to an oral glucose tolerance test.

In experiment 3, the two types of intermittent feeding were compared. At 12 days of age, chickens (Hubbard breed) were divided into two groups (IPS 1-1 and ITS 1-1) according to their body weight (192 ± 1.5 g) and submitted to the same measurements as those of experiment 2.

In all experiments, in order to prevent the "stress" effect hyperglycemia induced by handling (37) or by multiple blood sampling (38), chickens were handled and weighed 5 to 7 times without blood sampling during the rearing period and sampled only once on the day of experiment. At each sampling time, the different groups of chickens were selected for equal body weight. The glucose tolerance was measured using an oral glucose load of 2 g/kg body weight (with a 50% glucose solution, w/v) i.e., about 1/20 of a normal daily carbohydrate intake. Glucose was administered into the crop by oral intubation.

Chemical and statistical analyses. Single blood samples of 5 to 10 ml/chicken were collected into syringes containing 0.05 to 0.1 ml of a 1% sodium heparinate solution (w/v), cooled at 0° and centrifuged at 4°. The plasmas were separated, divided into aliquot samples, and stored at −20° until assay. Plasma glucose was determined with the ferricyanide method (39) adapted to an auto analyzer [1] with modifications described recently (40). In the chicken, the ferricyanide method overestimates the plasma glucose level by about 10% when compared with the glucose oxidase method. Plasma free fatty acids were extracted according to the method of Dole and Meinertz (41) using caprylic acid as carrier and margaric acid as internal standard. After extraction, free fatty acids were separated by thin layer chromatography, methylated, and analyzed by gas chromatography. The area of each fatty acid peak was measured and the sum of all peaks was calculated. The level of free fatty acid was quantitatively deduced from the area of the peak of the internal standard and expressed as μg margaric acid equivalents/ml. Chicken plasma insulin was determined by a specific and sensitive radioimmunoassay as previously described (32) with a guinea-pig anti-porcine insulin serum (Ab 27-6), pure chicken insulin as standard and (^{125}I)chicken insulin as tracer at a specific activity of 140 to 160 μCi/μg. Statistical analyses were performed using Student's *t*-test or analyses of variance (42).

[1] Technicon Instruments Corp., Ardsley, New York.

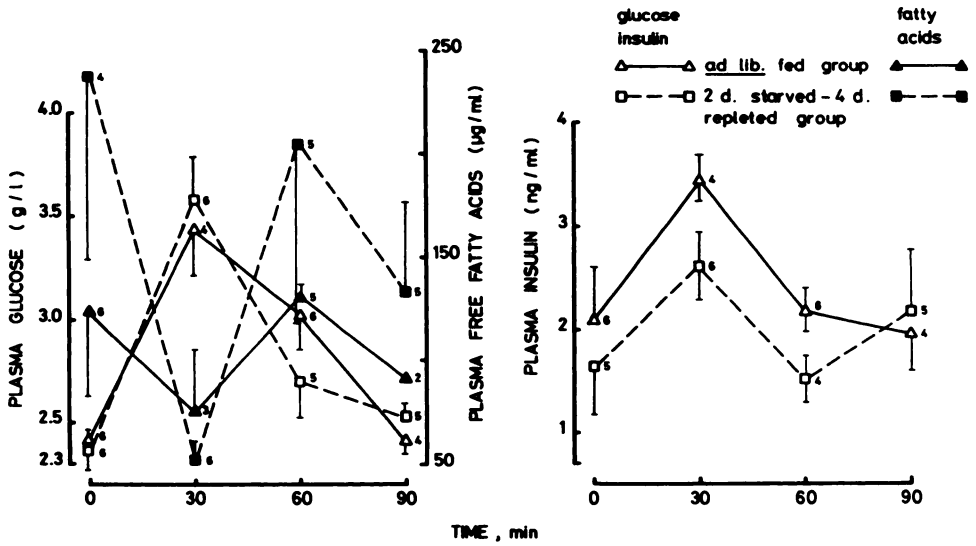

Fig. 1 Effect of intermittent total starvation on glucose tolerance (experiment 1). At 2-week old, male chickens were either fed ad libitum or submitted to intermittent total starvation with a cycle of: 2 days of starvation–4 days of ad libitum feeding (ITS 2-4 group). After 48 days of experiment, chickens being 9-week old, at the end of the repletion period of ITS 2-4 group and after an overnight fast (16 hours) chickens were loaded per os with glucose (2 g/kg body weight). Live body weight and total food intake (mean ± SEM) were: 2,307 ± 30 and 4,659 ± 61 g in the control group and 1,665 ± 21 and 3,378 ± 44 g in the ITS 2-4 group, respectively. Single blood samples per chicken were obtained by frontal cardiac puncture at the indicated times. Mean values of plasma glucose (left panel), free-fatty acids (left panel) and insulin (right panel) ± SEM for the indicated number of chickens are reported.

RESULTS

Experiment 1. Intermittent total starvation (ITS 2-4) versus ad libitum feeding

Effect on glucose tolerance (fig. 1). After overnight (16 hour) fasting, basal plasma glucose levels were similar in the ITS 2-4 group and the control group (fig. 1, left panel). After the oral glucose load, the plasma glucose and insulin (fig. 1, right panel) values of ITS 2-4 and control groups exhibited a similar pattern. However, the glucose disposal was slightly delayed in the control as compared to the ITS 2-4 group, the 60 minute level being significantly different from the initial level ($P < 0.05$). Plasma insulin levels were consistently lower in the ITS 2-4 group than in the control group from 0 to 60 minutes after the glucose load ($F_{1,26} = 4.64$, $P < 0.05$) which suggests a decrease in the B-cell sensitivity to glucose after intermittent feeding.

The plasma levels of free fatty acids (fig. 1, left panel) showed opposite variations to plasma glucose and were decreased at 30 minutes after the oral glucose load. However, no significant differences were observed between the two groups.

Effect on the utilization of a test meal (fig. 2). After overnight fasting (16 hours) chickens were offered a test meal of 20 g. In both groups, food intake (fig. 2, lower part left panel) was similar at 30 minutes. Thereafter food intake plateaued at 16 g in the control group. This was significantly lower than in the ITS 2-4 group from 30 to 90 minutes ($P < 0.01$). Plasma glucose levels significantly increased above initial levels at 30 minutes ($P < 0.01$) in the control group and at 30 ($P < 0.01$) and 60 minutes ($P < 0.05$) in the ITS 2-4 group. At 60 minutes, plasma glucose levels were significantly higher ($P < 0.05$) in the ITS 2-4 than in the control group. The test meal evoked a rapid rise in the plasma insulin levels at 30 ($P < 0.02$) and 60 minutes ($P < 0.05$) in both groups. No differences were observed in insulin release whether or not the chickens were trained to intermit-

tent feeding. This result, together with the plasma glucose level at 60 minutes, further indicate a decrease in the B-cell sensitivity to glucose in the starved-repleted chickens.

Experiment 2. Intermittent total starvation (ITS 1-1) versus ad libitum feeding

Variations in food intake, plasma glucose, and plasma insulin levels during a starvation-repletion cycle (fig. 3). On the repletion day, food intake amounted to 50.5 g between 0900 and 1100 hours in the ITS 1-1 group when it did not exceed 18.8 g in the controls in the same interval of time. This hyperphagia led to a significant increase of food intake during the repletion of ITS 2-4 ($P < 0.01$ at 1700 hours), although the total food intake remained lower than that of the controls during the 2-day measurement period.

The plasma glucose levels showed only minor and non-significant variations throughout the 2-day measurement period in the control group (fig. 3, upper part, left panel). In the ITS 1-1 group, the plasma glucose levels were constant from 0900 to 1700 hours on the first day which suggests that nutrients stored the day before were sufficient to maintain the plasma glucose constant for at least 8 hours. However at 2000, i.e., after 11 hours of fasting, plasma glucose was significantly decreased ($P < 0.01$). A further decrease was observed after the 24 hour fasting period at 0900 just before feeding ($P < 0.01$). Refeeding the ITS 1-1 group greatly increased the plasma glucose and led to high glycemia between 1100 and 1700 hours as compared to the control group ($P < 0.01$).

The plasma insulin levels (fig. 3, right panel) in the control group did not show any significant variations during the 2-day measurement period, as was observed with plasma glucose. In the ITS 1-1 group, the plasma insulin level, at 0900 just before refeeding, was not significantly different from the initial level. Such insulin levels together with the low glucose levels may be of consequence to the subsequent hyperphagia observed in this group. After refeed-

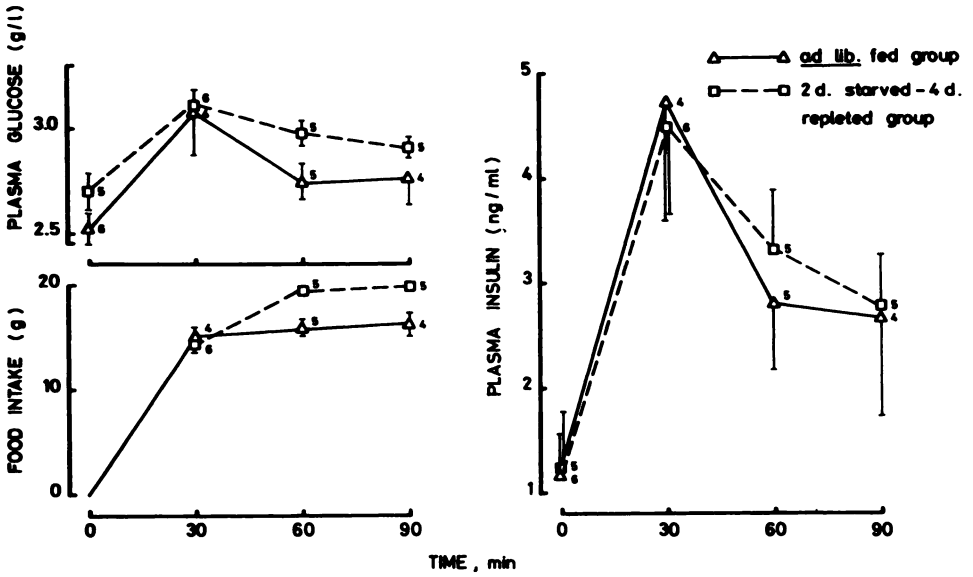

Fig. 2 Effect of intermittent total starvation on the utilization of a meal (experiment 1). After one extra starvation-repletion cycle of ITS 2-4 group (see legend to fig. 1) and after an overnight fast (16 hours) chickens (10-week old) were offered a meal of 20 g. Live body weight and total food intake (mean ± SEM) were 2,570 ± 23 and 5,527 ± 58 g, respectively, in the control group and 1,842 ± 22 and 3,998 ± 46 g in the ITS 2-4 group. Single blood samples per chicken were obtained by frontal cardiac puncture at the indicated times. Mean values of food intake (lower part, left panel), plasma glucose (upper part, left panel) and plasma insulin (right panel) ± SEM for the indicated number of chickens are reported.

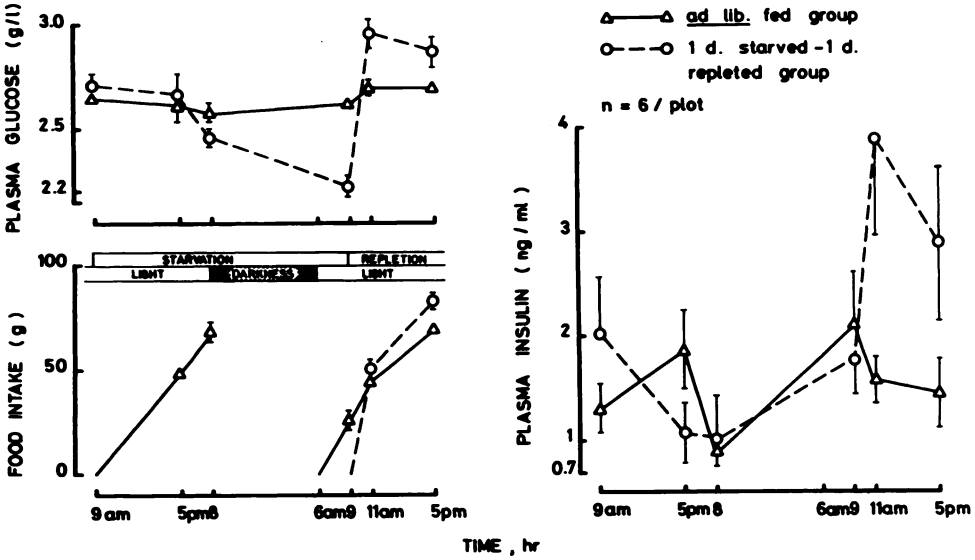

Fig. 3 Daily variations of the plasma glucose and insulin levels during a restriction-repletion cycle in chickens accustomed to intermittent total starvation (experiment 2). Twelve-day old male chickens were either fed ad libitum or submitted to intermittent total starvation with a cycle of: 1 day of starvation–1 day of ad libitum feeding (ITS 1-1 group). On the 31st and 32nd days of experiment, i.e., during the 16th restriction-repletion cycle of ITS 1-1 group, single blood samples per chicken were obtained by wing vein puncture at the indicated times. Mean values of cumulative food intake/day (lower part, left panel), plasma glucose (upper part left panel) and plasma insulin (right panel) ± SEM for the indicated number of chickens are reported. When not shown, the SEM was smaller than the plots.

ing, insulin release was highly stimulated and the plasma insulin levels were significantly higher from 1100 to 1700 hours ($P < 0.01$) in ITS 1-1 group than in the control group. Therefore, chickens accustomed to intermittent total starvation (ITS 1-1) exhibited large changes in plasma insulin levels related to large changes in plasma glucose levels concomitant with the hyperphagia which was observed on refeeding.

Effect on glucose tolerance (*fig. 4*). The initial plasma glucose level was higher ($P < 0.05$) in the ITS 1-1 group than in the control group (fig. 4, left panel). This difference was probably related to the fact that during the hours preceding fasting, chickens stored more nutrients in the ITS 1-1 than in the control group. After the oral glucose load, plasma glucose levels were significantly higher than initial levels from 30 to 60 minutes in the control group ($P < 0.001$) and from 30 to 45 minutes in the ITS 1-1 group ($P < 0.001$). In addition, plasma glucose levels were significantly lower ($P < 0.001$) from 30 to 60 minutes in the ITS 1-1 group than the corresponding values oberved in the control group. Therefore, one may conclude that intermittent total starvation with the ITS 1-1 rhythm improves the glucose tolerance in the chicken.

In response to the glucose load, plasma insulin levels were increased at 30 minutes in both groups ($P < 0.05$) and 45 minutes in the control group ($P < 0.05$). In addition, 30 to 60 minutes after the glucose load, plasma insulin levels were lower in the ITS 1-1 group than in the control group ($P < 0.05$). This lower insulin response, associated with the improved glucose tolerance, suggest that in ITS 1-1 group the insulin sensitivity of target tissues is increased.

Experiment 3. Intermittent total starvation (ITS 1-1) versus intermittent protein starvation (IPS 1-1)

Variations in food intake, plasma glucose, and plasma insulin levels during a restric-

tion-repletion cycle (*fig.* 5). Food intake (fig. 5, lower part left panel) in the IPS 1-1 group consisted of the protein-free diet between 0900 and 2000 and between 0600 and 0900, and of the balanced diet between 0900 and 1700 hours. In the ITS 1-1 group, food intake consisted of the balanced diet from 0900 to 1700 on the second day of the measurement period. On this day (the repletion day), food intake was significantly higher in the ITS 1-1 than in the IPS 1-1 group ($P < 0.01$ at 1700 hours) as previously observed (22).

Plasma glucose (fig. 5, upper part, left panel) and plasma insulin (fig. 5, right panel) levels in the ITS 1-1 group showed similar variations to those observed in experiment 2. In the IPS 1-1 group, the plasma glucose level was increased above the initial level at 1700 hours ($P < 0.01$) by feeding the protein-free diet and remained higher up to 1100 hours ($P < 0.05$) i.e., 2 hours after feeding the balanced diet. From 1700 (first day) to 0900 hours (just before refeeding the balanced diet) plasma glucose levels were significantly ($P < 0.05$ to $P < 0.01$) higher than the corresponding levels in the ITS 1-1 group. In the IPS 1-1 group, the plasma insulin level at 2000 hours was lower than initial level ($P < 0.01$) and the corresponding level in the ITS 1-1 group ($P < 0.05$). Feeding the balanced diet also increased insulin release ($P < 0.05$). Therefore, intermittent protein starvation evoked high glycemia and low insulinemia on the day of restriction. This finding further documents the previous observations (35) on the synergism between amino acid and glucose in stimulating insulin release.

Effect on glucose tolerance. After the oral glucose load, plasma glucose (fig. 6, left panel) and plasma insulin (fig. 6, right panel) increased to ($P < 0.01$ for glucose and $P < 0.02$ for insulin in the IPS 1-1 group) similar levels in both groups at 30 minutes and returned to the initial levels within 45 minutes. Therefore, glucose tolerance and insulin sensitivity would appear to be similar in ITS 1-1 and IPS 1-1 groups.

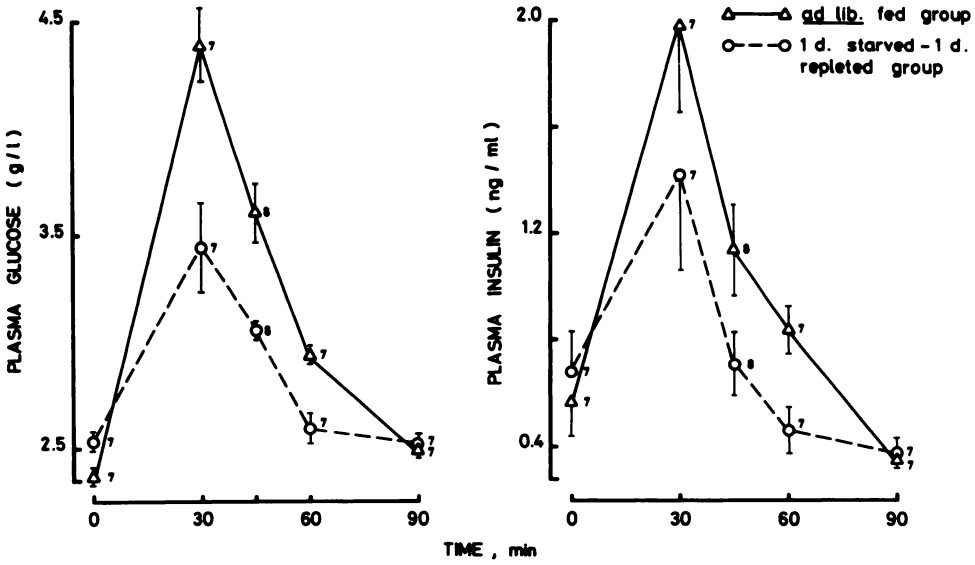

Fig. 4 Effect of intermittent total starvation on glucose tolerance (experiment 2). After 42 days of experiment, at the end of the repletion period of ITS 1-1 group (see legend to fig. 3) and after an overnight fast (16 hours), chickens were loaded per os with glucose (2 g/kg body weight). Live body weight and total food intake (mean ± SEM) were 1,770 ± 32 and 3,150 ± 54 g in the control group and 1,398 ± 19 and 2,454 ± 30 g in the ITS 1-1 group, respectively. Single blood samples per chicken were obtained by frontal cardiac puncture at the indicated times. Mean values of plasma glucose (left panel) and insulin (right panel) ± SEM for the indicated number of chickens are reported.

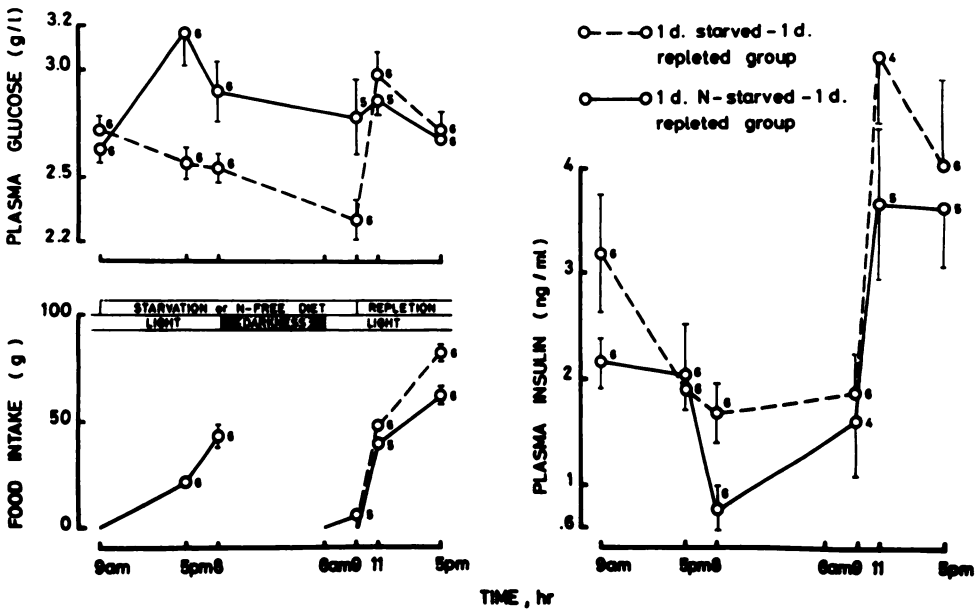

Fig. 5 Daily variations of the plasma glucose and insulin levels during a restriction-repletion cycle in chickens accustomed to either intermittent total starvation or intermittent protein starvation (experiment 3). Twelve-day old male chickens were submitted to intermittent total starvation with a cycle of: 1 day of starvation–1 day of ad libitum feeding (ITS 1-1 group) or to intermittent protein starvation with a cycle of: 1 day of feeding with the protein-free diet–1 day of feeding with the balanced diet (IPS 1-1 group). On the 31st and 32nd days of experiment, i.e., during the 16th restriction-repletion cycle of both groups, single blood samples per chicken were obtained by wing vein puncture at the indicated times. Mean values of cumulative food intake/day (lower part, left panel), plasma glucose (upper part, left panel) and plasma insulin (right panel) ± SEM for the indicated number of chickens are reported. When not shown, the SEM was smaller than the plots.

DISCUSSION

Intermittent feeding (ITS 1-1 and IPS 1-1) induces considerable changes in the insulin pattern of the chicken which are characterized by high plasma insulin levels consequent upon the high level of nutrient intake during the repletion following a period of restriction (figs. 3 and 5). A similar reaction has been noticed in "meal-eater" rats (2 hours feeding/day) after their daily meal (43). These high levels of insulinemia are related to the rapid food intake due to the "adaptive hyperphagia" and are very likely of physiological importance permitting a rapid storage of the nutrients. In addition, the storage of nutrients would be further stimulated by the fact that either type of intermittent feeding (ITS 1-1 and IPS 1-1) markedly improved the glucose tolerance in spite of a constant decrease in the plasma insulin level. In the chicken, therefore, intermittent feeding may increase the insulin sensitivity of target tissues. In the rat, similar intermittent feeding schedules (2), meal eating (15–16), or force-feeding 80% of the voluntary and ad libitum food intake (14) also improved the glucose tolerance and in the chicken, meal-eating produced the same effect although less consistently (23). In "meal-eater" rats, the in vivo sensitivity of adipose tissue to insulin is enhanced (15, 24–25). Adipose tissue in the chicken is unlikely to be the main responsive tissue since in this tissue, lipogenesis is very low, unresponsive to insulin (44–46), and unadaptative to intermittent feeding (32). We can suppose that the enhanced glycogen and lipid synthesis (21–22) without changes in insulin release (fig. 2) demonstrated after a meal in the liver of chickens accustomed to intermittent feeding represent, at least in part, an example of this adaptation.

The effectiveness of glucose in inducing

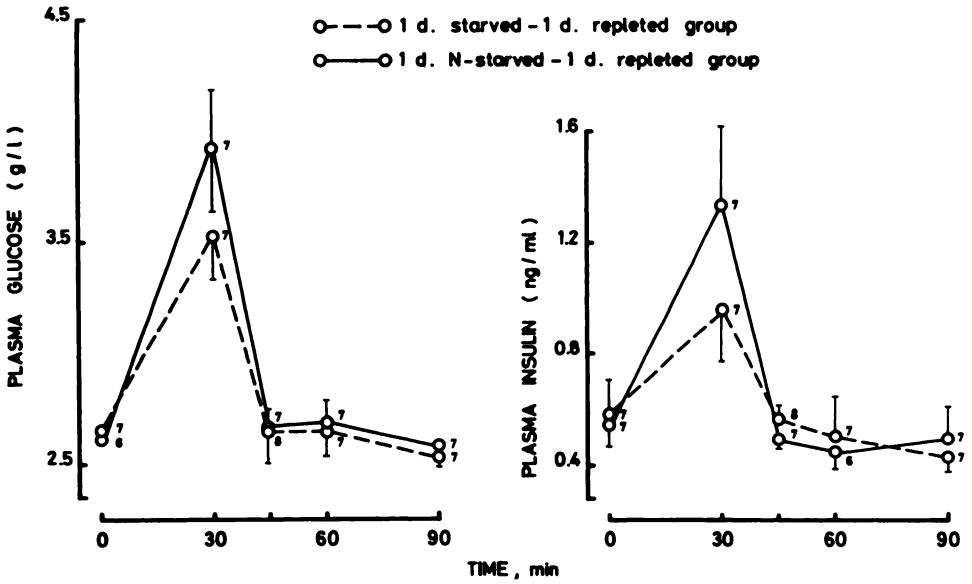

Fig. 6 Effect of intermittent total starvation and intermittent protein starvation on glucose tolerance (experiment 3). After 42 days of experiment, at the end of the repletion period of ITS 1-1 and IPS 1-1 groups (see legend to fig. 5) and after an overnight fast (16 hours), chickens were loaded per os with glucose (2 g/kg body weight). Live body weight and total food intake (mean ± SEM) were 1,365 ± 18 and 2,413 ± 31 g, in ITS 1-1 group and 1,376 ± 23 g and 692 ± 24 (protein diet) and 2,071 ± 35 g (balanced diet) in the IPS 1-1 group, respectively. Single blood samples per chicken were obtained by frontal cardiac puncture at the indicated times. Mean values of plasma glucose (left panel) and insulin (right panel) ± SEM for the indicated number of chickens are reported.

insulin release was dependent on both the nature and the rhythm of intermittent feeding. In this respect, plasma glucose was highly effective in inducing insulin release in ITS 1-1 group; this was particularly evident during the fasting period of the cycle (figs. 3 and 5). In the other experiments (ITS 2-4 and IPS 1-1 groups), the glucose induced insulin release appeared to be lower than in both the control and the ITS 1-1 group. In the ITS 2-4 group, this may be related to the longer period of fasting during the cycle since it has been observed in the chicken (35) as well as in mammals (47–50) that prolonged fasting results in a decrease in the B-cell sensitivity to glucose. In the IPS 1-1 group, the decrease in sensitivity is more apparent when carbohydrates are given in a normal solid diet (fig. 5) than when given in solution (fig. 6). This is likely to be due to the lack of amino acids which are known to exert a potent synergetic effect in stimulating the glucose-induced insulin release in the chicken (35) as well as in mammals (51–53). Finally, among the three feeding patterns which have been studied, ITS 1-1 results in the optimal glucose-insulin relationship characterized both by an increase of sensitivity of the B-cell to glucose and a rapid glucose disposal for a minimal insulin secretion. This rhythm is the only one that does not increase the body fat content of the chicken.

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