Intermittent fasting modulation of the diabetic syndrome in sand rats. II. *In vivo* investigations

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Abstract. This study deals with the effects of daily intermittent fasting for 15 h upon the development of diabetes in sand rats exposed to a hypercaloric diet. The same pattern of daily intermittent fasting was imposed on sand rats maintained on a purely vegetal diet (control animals). Over the last 30 days of the present experiments, non-fasting animals gained weight, whilst intermittently fasting sand rats lost weight. In this respect, there was no significant difference between control animals and either diabetic or non-diabetic sand rats exposed to the hypercaloric diet. The postprandial glycemia remained fairly stable in the control animals. During a 3-week transition period from a purely vegetal to a hypercaloric diet, the postprandial glycemia increased by 5.95±1.26 mM (n=6) in diabetic sand rats, as distinct from an increase of only 0.45±0.56 mM (n=6) in the non-diabetic animals. During the intermittent fasting period, the postprandial glycemia decreased significantly in the diabetic animals, but not so in the non-diabetic sand rats. Before the switch in food intake, the peak glycemia at the 30th min of an intraperitoneal glucose tolerance test was already higher in the diabetic than non-diabetic rats. In both the non-diabetic and diabetic sand rats, intermittent fasting prevented the progressive deterioration of glucose tolerance otherwise observed in non-fasting animals. These findings reveal that, at least in sand rats, intermittent daily fasting prevents the progressive deterioration of glucose tolerance otherwise taking place when these animals are exposed to a hypercaloric diet.

Introduction

Overabundant food intake with chronic positive energy balance leads to metabolic disorders such as obesity and type 2 diabetes. Chronic moderate reduction in energy intake results in the opposite effects, namely increased insulin sensitivity and improved glucose homeostasis (1-3). In fact, caloric restriction is considered as a common treatment for obesity, insulin resistance and type 2 diabetes (4).

The caloric restriction may occur in different manner, either a relative decrease of food intake, e.g. by regimens restricted in their carbohydrate (5-7) or lipid content (8), or otherwise a total short (9,10) or prolonged (8) fasting.

In this study, we explored another modality of caloric restriction consisting in a daily intermittent fasting for 15 h, i.e. from 5 p.m. to 8 a.m., over a period of 30 days. To study this, we exploited a desert gerbil, *Psammomys obesus* in which the diabetes syndrome is similar to that found in type 2 diabetic human patients (11). Sand rats were indeed selected for the present investigations because they represent the most appropriate animal model for induction of diabetes as a result of exposure to a diet of increased caloric value.

In the first report in this series, we dealt mainly with such items as the biotope and type of vegetal diet of sand rats *(Psammomys obesus)* in their desertic habitat, as well as capture modalities, emphasis being placed on the daily pattern of food intake by these animals in their natural environment (12). Taking such a pattern into consideration, the major aim of the present study was to explore the possible effects of daily intermittent fasting for 15 h, i.e. from 5 p.m. to 8 a.m., upon the time-related changes in body weight and glucose tolerance provoked in sand rats by the transition from a purely vegetal diet to a hypercaloric one.

Materials and methods

The design of the present experiments was already described in details in a prior report (12). Briefly, 52 sand rats were captured in April in the region of Abadla (Béchar Wilaya, Algeria) and transported to the Laboratory (Mostaganem University, Algeria). After 15 days of acclimatization, during which the sand rats had only access to a vegetal diet of Chenopodiaceae, and during a subsequent 20-day transition period, the 14 heavier (and presumably oldest) sand rats were maintained on the same vegetal diet (control animals) whilst the other 38 rats were given access to salty water and a mixed diet composed to 20 g per animal and per day of a standard laboratory chow, with an energetic value of 2.90 kcal/g, and

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Table I. Flow chart of study design.

Capture of rats (n=52)		
Conditioning period: fed ad libitum a vegetal diet for 3 days (n=52)		
Acclimatization period: fed a vegetal diet (representing 100% of body wt.) for 15 days (n=52)		
Selection of control versus non-diabetic or diabetic sand rats		
Heaviest sand rats (n=14)	Lightest sand rats (n=38)	
(Control sand rats)	(Non-diabetic and diabetic sand rats)	
Transition period: 20 days starting at end of acclimatization period		
Maintained at vegetal diet	First week: 20 g chow + vegetal diet (75% body wt.)	
(100% body wt. for 3 weeks)	Second week: 20 g chow + vegetal diet (50% body wt.)	
	Third week: 20 g chow + vegetal diet (25% body wt.)	
Last day of transition period: postprandial glycemia		
Glycemia <0.7 mM (n=19)	Glycemia >8.3 mM (n=19)	
(Non-diabetic sand rats)	(Diabetic sand rats)	
Non-fasting versus intermittent fasting period: last 30 days of present experiments		
Non-fasting control rats (n=10)	Non-fasting non-diabetic rats (n=9)	Non-fasting diabetic rats (n=13)
Fasting control rats (n=4)	Fasting non-diabetic rats (n=10)	Fasting diabetic rats (n=6)
Fed vegetal diet	Fed chow diet	Fed chow diet

decreasing amounts of the vegetal diet, with an energetic value of 0.42 kcal/g, the latter amounts representing 75% (first week), 50% (second week) and 25% (third week) of the animal body weight, instead of 100% of the body weight during the acclimatization period (Table I). Based on the measurement of postprandial glycemia, these 38 sand rats were then divided in two groups of 19 animals each referred to as either non-diabetic (glycemia <7.0 mM) or diabetic (>8.3 mM). During the last 30 days of the present experiments, the animals were given access to either the vegetal diet (control animals) or hypercaloric diet (diabetic and non-diabetic rats), some of the sand rats in each group undergoing intermittent daily fasting from 5 p.m. to 8 a.m. the next day. In the intermittently fasting rats, the postprandial glycemia was measured 2 h after reintroduction of food and, if so required salty water, at 8 a.m. An intraperitoneal glucose tolerance test was performed in the fasting or non-fasting diabetic and non-diabetic sand rats at the 10th, 20th and 29th of the last 30 day period. For purpose of comparison with the fasting animals, the non-fasting sand rats were also deprived of food from 5 p.m. on the day before. After a 15 h of fasting, blood samples were collected from the saphaneous vein (0 min sample). Then, a 20% solution of glucose (2 g/kg body weight) was injected intraperitoneally, followed by blood sampling at 30 and 120 min after the injection. The body weight and postprandial glycemia, measured with a glucometer in a blood sample obtained from the saphenous vein, were recorded at times indicated in the text and figure. During the last 4 weeks of the present experiments, the food intake was measured every day.

All results are presented as mean values (± SEM) together with either the number of individual observations (n) or degree of freedom (df). The statistical significance of

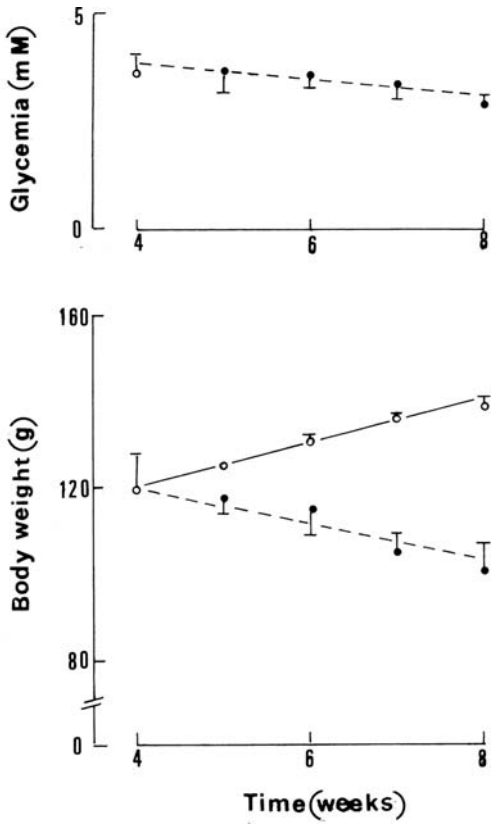

Figure 1. Upper panel: time course for the changes in postprandial glycemia in intermittently fasting control sand rats. Mean values (± SEM) refer to 4 animals at each time point. Lower panel: time course for the changes in body weight during the fasting or non-fasting period in control sand rats. At week 4, the mean absolute value (± SEM) refer to 8 animals. At weeks 5 to 8, the results (mean ± SEM) refer to 4 animals and correspond to the paired changes above (non-fasting animals; open circles) or below (fasting animals; closed circles) the measurement made at week 4.

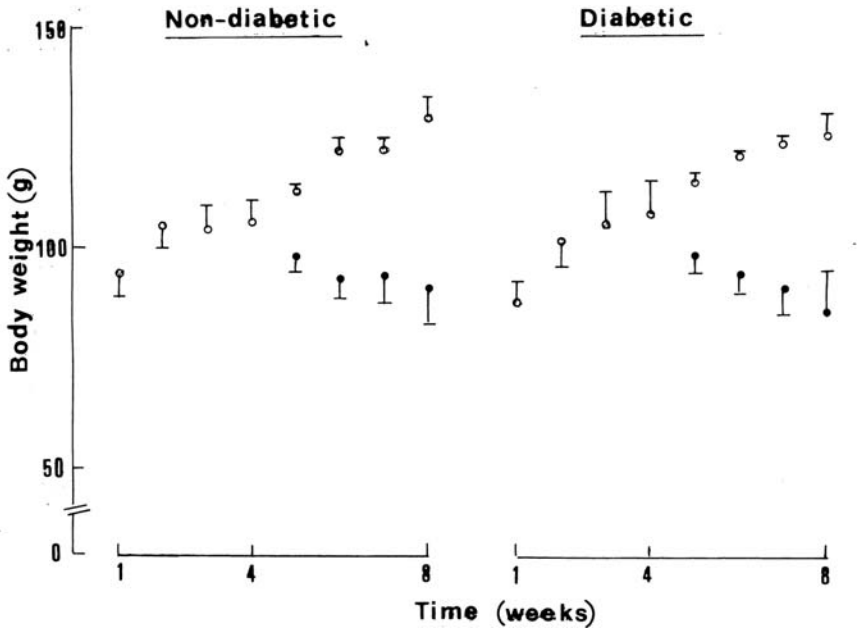

Figure 2. Time course for the changes in body weight during the transition period (week 1 to week 4) and fasting (or non-fasting) period (week 4 to week 8) in non-diabetic and diabetic sand rats. Mean absolute values (± SEM) refer to 19 rats from week 1 to week 4. The results from week 5 to week 8 refer to the mean values (± SEM) for 6-13 animals and correspond to the paired changes above (non-fasting animals; open circles) or below (fasting animals; closed circles) the measurement made at week 4.

differences between mean values was assessed by use of Student's t-test.

Results

Time course of changes in body weight. During the 3-week transition period, the change in body weight failed to differ significantly (df=34; $p>0.1$) in diabetic and non-diabetic rats. It consisted in a gain of 15.5±2.6 g (n=38), representing a 17.9±2.7% ($p<0.001$) increase in body weight relative to the value recorded at the onset of the transition period.

At the end of this transition period, the mean body weight also failed to differ significantly ($p>0.8$) in diabetic rats (107.4±7.9 g; n=19) and non-diabetic rats (105.5±5.3 g; n=19). These values remained somewhat lower, albeit no more significantly so, from that recorded at the same time in the control rats (120.0±8.6 g; n=8). When these control rats were further examined over the ensuing 4 weeks, their body weight increased by 18.8±2.4 g (n=4; $p<0.005$), representing a 17.2±1.6% increase relative to the value measured at the onset of this 4-week period. The intermittently fasting control rats, however, lost 20.0±7.4 g (n=4) over the same period, this representing a 15.3±4.6% ($p<0.05$) relative decrease. Neither the gain in body weight recorded in the non-fasting control rats (+18.8±2.4 g), nor the fall in body weight observed in the fasting control rats (-20.0±7.4 g) were significantly different ($p>0.7$ or more) from the corresponding mean values found in the diabetic and non-diabetic rats, i.e. +21.1±2.8 g (n=22) and -18.4±6.4 g (n=16) in non-fasting and fasting animals, respectively. The pattern of changes in body weight observed in the control animals over the four weeks preceding sacrifice is illustrated in Fig. 1.

Over the last 30 days of the present experiments, the non-fasting diabetic rats gained 19.2±3.5 g body wt. (n=13; $p<0.001$); this representing a 21.7±3.7% (n=13) relative increase over the value recorded at the onset of the same period. In the fasting diabetic rats, the situation was quite different ($p<0.001$) with a mean fall in body wt. of 24.2±11.4 g (n=6; $p<0.09$), corresponding to a 15.0±7.2% relative decrease ($p<0.1$).

A comparable situation prevailed in the non-diabetic rats. Indeed, the non-fasting non-diabetic rats gained 23.9±4.5 g (n=9; $p<0.001$) over the same period of 30 days, this representing a 22.3±3.4% (n=9) relative increase. The fasting non-diabetic rats lost 15.0±7.9 g (n=10; $p<0.1$), this corresponding to an 11.6±7.1% relative decrease ($p>0.1$). None of these values differed significantly from those recorded in diabetic rats. Fig. 2 illustrates the results collected in both the non-diabetic and diabetic rats.

Food intake. The mean values for food intake, as derived from daily measurements during each successive weeks of the non-fasting or intermittent fasting period, are listed in Table II. In the non-fasting control animals, it averaged 82.3±9.5 g/day per rat, corresponding to a caloric intake of 34.6±4.0 kcal/day per rat. In the non-fasting non-diabetic and diabetic animals, respectively, it averaged 16.6±0.6 and 16.3±0.6 g/day per rat, corresponding to a caloric intake close to 47.7±1.7 kcal/day per rat. Such a caloric intake thus appeared somewhat higher ($p<0.02$) in the animals fed the chow diet as distinct from vegetal diet.

When the latter animals underwent intermittent fasting, the decrease in food intake ($p<0.004$) from 82.3±9.5 to 33.5±6.3 g/day was commensurate with the length of the fasting period (15 h/24 h), i.e. an expected decrease in food intake from 82.3±9.5 to 30.9±0.6 g/day. However, when the non-diabetic or diabetic animals underwent intermittent fasting the decrease in food intake ($p<0.001$) from 16.4±0.4 g/day to

Table II. Food intake.

Rats	Week 5	Week 6	Week 7	Week 8
Non-fasting control (n=6)	87.3±9.5[a]	80.4±9.2	79.5±8.0	81.9±11.3
Fasting control (n=4)	34.6±6.7	33.8±7.9	32.7±5.2	33.0±5.2
Non-fasting non-diabetic (n=6)	15.9±0.7	16.6±0.6	16.9±0.5	16.8±0.6
Fasting non-diabetic (n=6)	11.7±0.5	10.0±0.6	9.8±0.5	9.4±0.7
Non-fasting diabetic (n=6)	15.8±0.5	16.3±0.5	16.6±0.6	16.6±0.7
Fasting diabetic (n=6)	10.3±1.3	9.5±0.8	9.7±0.5	9.4±0.7

[a]All results are expressed as g/day per rat.

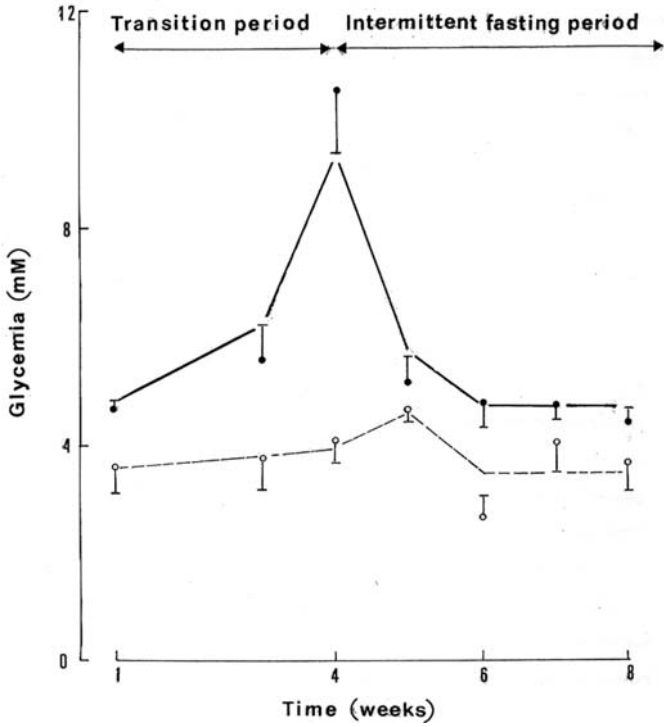

Figure 3. Time-related changes in the postprandial glycemia of fasting diabetic (closed circles and solid line) and non-diabetic (open circles and dashed line) sand rats. Mean values (± SEM) are derived from 6 individual observations at each time point. The horizontal lines (weeks 6 to 8) refer to the mean values during the last 2 weeks of the intermittent fasting period.

10.0±0.5 g/day (n=12 in both cases) was less pronounced ($p<0.001$) than that theoretically calculated from the length of the fasting period, i.e. an expected decrease in food intake down to 6.3±0.1 g/day. Nevertheless, when the intermittent fasting-induced decrease in food intake was converted to a decrease in caloric intake, no significant difference was anymore observed between control animals (48.8±12.9 g x 0.42 kcal/g = 20.5±5.4 kcal), and non-diabetic or diabetic animals (6.4±0.6 g x 2.90 kcal/g = 18.6±1.7 kcal). This coincides with the fact that the changes in body weight in non-fasting versus fasting animals are also comparable in control animals and non-diabetic or diabetic animals.

The comparison between food intake and changes in body weight allows to rule out, in the fasting rats, any increase in caloric expenditure, as could otherwise conceivably be attributed to the stress and anxiety caused by intermittent fasting. For instance, in the fasting control rats, the sum of caloric intake (14.1 kcal/day) and estimated caloric generation attributable to the daily decrease (0.71 g) in body weight (assuming mainly a loss of fat, i.e. 0.71 g times 7 kcal/g or 5.0 kcal/day) would imply a caloric expenditure close to 19.1 kcal/day. In the non-fasting control rats, the difference between caloric intake (34.6 kcal/day) and postulated caloric expenditure (if indeed comparable to that of fasting animals) would corresponding to a daily gain of 15.5 kcal/day or, relative to the daily gain in body weight (0.67 g/day), to an unrealistic caloric investment of 23.1 kcal/g to cover the gain in body weight. In turn, this implies that the caloric expenditure was higher in non-fasting sand rats than the intermittently fasting animal. For instance, if it were postulated that the gain in body weight in non-fasting animals correspond mainly to an increase in adipose tissue mass, the caloric expenditure would correspond to the difference between caloric intake (34.6 kcal/day) and postulated caloric investment (0.67 g/day times 7 kcal/g or 4.7 kcal/day), i.e. 29.9 kcal/g. The caloric expenditure would then appear about one third lower in fasting than in non-fasting control sand rats.

Postprandial glycemia. The postprandial glycemia was measured in the intermittently fasting control animals 2 h after allowing them again access to food at 8.00 a.m. Before the intermittent fasting period, it averaged 3.63±0.45 mM (n=4). As illustrated in Fig. 1 (upper panel), it then remained fairly stable. Even after 4 weeks of intermittent fasting, it was only 0.74±0.30 mM lower (n=4; $p<0.1$) than that measured at the onset of this fasting period.

A different situation prevailed in the non-diabetic and diabetic fasting animals. Just before the switch in food intake, the postprandial glycemia was already somewhat lower, albeit not significantly so ($p<0.1$) in non-diabetic rats (3.61±0.49 mM; n=6) than in diabetic ones (4.68±0.15 mM; n=6). Such a difference became more pronounced ($p<0.06$) 2 weeks after the switch in food intake with mean values of 3.75±0.58 mM (n=6) in non-diabetic rats and 5.59±0.60 mM (n=6) in diabetic rats (Fig. 3). It became highly significant ($p<0.001$) 3 weeks after the switch, at which time all measurements in the diabetic rats exceeded 8.3 mM (10.63±1.22 mM; n=6), whilst averaging no more than 4.06±0.41 mM (n=6) in the non-diabetic rats. As a matter of fact, over this 3-week

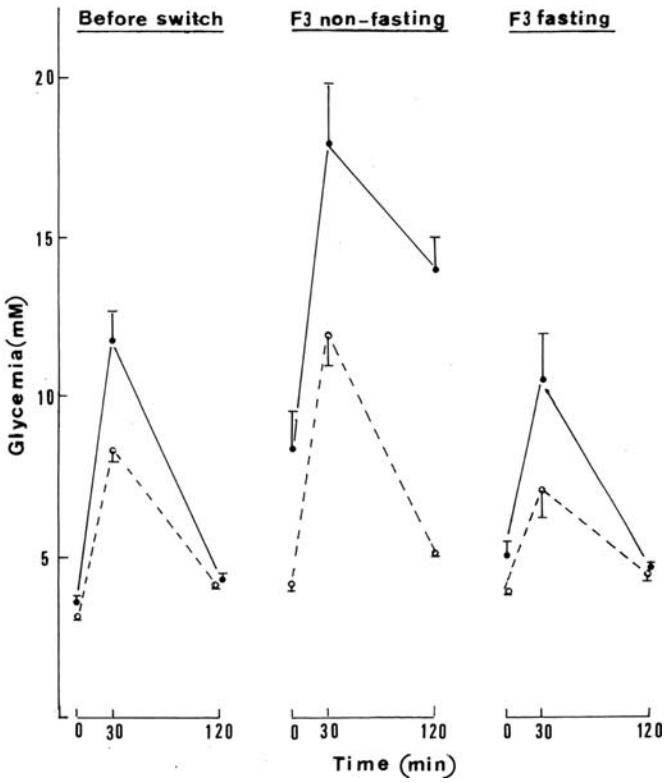

Figure 4. Pattern of changed in glycemia during intraperitoneal glucose tolerance tests conducted in either non-diabetic (open circles and dashed line) or diabetic (closed circles and solid line) sand rats examined before the switch in food intake (left) and at the end of the non-fasting (middle) or intermittent fasting (right) period. Mean values (± SEM) refer to 12 rats before the switch and 5 animals thereafter.

transition period, the postprandial glycemia increased by 5.95±1.26 mM (n=6) in the diabetic rats, as compared ($p<0.005$) to an increase of only 0.45±0.56 mM (n=6; $p>0.4$) in the non-diabetic rats (Fig. 3).

A comparable situation was observed over the same 3-week transition period in the non-fasting rats, in which the postprandial glycemia averaged before the switch in food intake and at the end of the transition period, respectively, 3.51±0.26 and 4.96±0.39 mM in the non-diabetic rats (n=9) and 3.97±0.19 mM and 11.70±0.92 mM in the diabetic rats (n=13). Pooling all available data, the postprandial glycemia was thus significantly higher ($p<0.03$) in diabetic rats (4.19±0.16 mM; n=19) than in non-diabetic rats (3.55±0.24 mM; n=15) already before the switch in food intake.

During the intermittent fasting period, the postprandial glycemia failed to decrease significantly in the non-diabetic rats, with a mean weekly fall averaging 0.09±0.39 mM (n=24; $p>0.8$). In the fasting diabetic rats, however, such a weekly fall averaged 1.53±0.58 mM (n=24; $p<0.02$). As documented in Fig. 3, such a decrease of postprandial glycemia in the diabetic rats was most pronounced during the first week of intermittent fasting when it averaged 5.45±1.11 mM (n=6). The data illustrated in Fig. 3 also indicate that over the last two weeks of the intermittent fasting period the mean postprandial glycemia remained significantly higher ($p<0.005$) in diabetic rats (4.67±0.21 mM; n=18) than in non-diabetic rats (3.44±0.30 mM; n=18).

Intraperitoneal glucose tolerance test (IPGTT). As expected, before the switch in food intake, the IPGTT yielded virtually identical results in rats later assigned to a non-fasting or intermittently fasting schedule. Already at this early stage, however, the results of the IPGTT differed in rats later allocated to the non-diabetic and diabetic group (Fig. 4). Indeed, whilst the zero and 120th min of the test yielded in the diabetic rats mean values slightly but not significantly higher than those recorded in the non-diabetic rats, the peak glycemia at the 30th min was much higher ($p<0.003$) in diabetic rats (11.80±0.89 mM; n=12) than in non-diabetic rats (8.38±0.39 mM; n=12).

During the non-fasting or intermittently fasting period, the situation also differed in non-diabetic and diabetic animals. In the non-fasting non-diabetic rats, a progressive deterioration of glucose tolerance was observed (n=6). The area under the curve indeed increase ($p<0.005$) from 732.9±42.5 mM.min before the switch in food intake to 1008.5±59.3 mM.min (n=6) at the end of the non-fasting period. Moreover, there was a highly significant positive correlation (r= +0.5172; n=24; $p<0.01$) between the individual values for such an area and the ranking (1 to 4) of the test day (before the switch in food intake and 10, 20 or 29 days after the onset of the non-fasting period). Such a deterioration failed to occur in the intermittently fasting non-diabetic animals, in which the area under the curve was no more higher at the end of the intermittent fasting period (689.3±54.1 mM.min; n=6) than before the switch in food intake (745.1±39.6 mM.min; n=6). As a matter of fact, in these fasting non-diabetic rats, the correlation coefficient between the individual values for the area under the curve and the ranking of the test day now yielded a negative value (r= -0.2882; n=24), which failed however to achieve statistical significance ($p>0.1$). In other words, after 20 to 29 days of intermittent fasting, the area under the curve during the IPGTT represented no more than 57.6±4.4% (n=12; $p<0.001$) of the mean corresponding values found at the same time in the non-fasting non-diabetic rats (100.0±6.5%; n=12).

In the non-fasting animals, the deterioration of glucose tolerance was more rapid in diabetic than in non-diabetic rats. First and as alluded to above, the area under the curve during the IPGTT was already higher ($p<0.005$) in diabetic rats (954.8±60.6 mM.min; n=12) than in non-diabetic animals (739.0±27.8 mM.min; n=12) before the switch in food intake. Second, in the non-fasting diabetic rats it increased ($p<0.025$) from 967.2±113.4 mM.min (n=6) before the switch in food intake to 1494.3±155.2 mM.min (n=6) at the 10th day of the non-fasting period, whilst no significant change ($p>0.8$) was recorded over the same length of time in the non-fasting non-diabetic rats (732.9±42.5 mM.min versus 757.2±96.2 mM.min; n=6 in both cases). Last, whilst, in the latter animals, a later increase ($p<0.03$) to 1036.6±68.5 mM.min (n=12) was recorded 20 and 29 days after the onset of the non-fasting period, such was not the case in the non-fasting diabetic rats, in which the mean value reached at day 10 (1494.3±155.2 mM.min; n=6) failed to differ significantly ($p>0.4$) from that recorded at days 20 and 29 (1664.0±128.9 mM.min; n=12). In the intermittently fasting diabetic rats, the area under the curve of the IPGTT failed to differ significantly before the switch in food intake and at

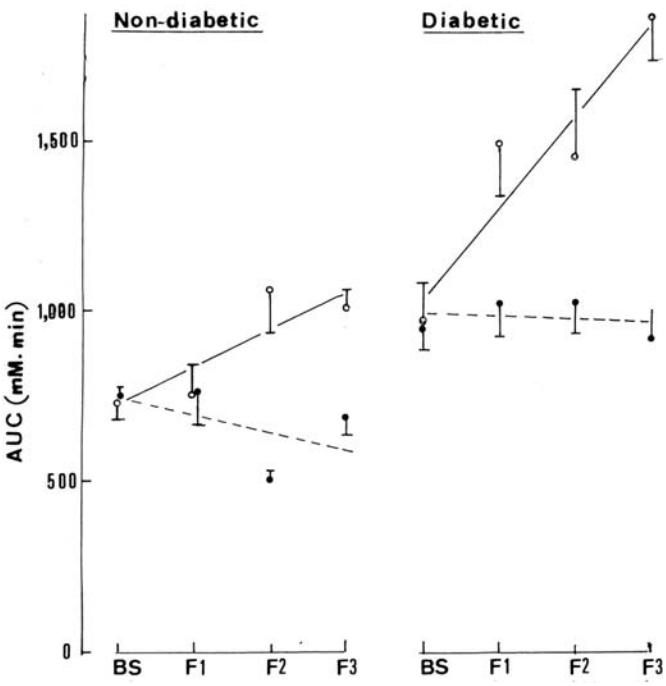

Figure 5. Area under the curve (AUC) during intraperitoneal glucose tolerance tests conducted in either non-fasting (open circles and solid line) or intermittently fasting (closed circle and dotted line) non-diabetic (left) and diabetic (right) sand rats examined before the switch in food intake (BS) at the 10th, 20th and 29th day (F1, F2 and F3) of the fasting (or non-fasting) period. Mean values (± SEM) refer to 6 individual observations at each time point. The oblique lines corresponding to the regression lines.

any time during the fasting period (Fig. 5). Such an area was lower in the fasting diabetic rats than in the non-fasting diabetic rats, whether during the first ($p<0.03$) or last ($p<0.001$) IPGTT performed during this period.

Discussion

An excess intake of calories favours the development of such diseases as obesity and type 2 diabetes. Inversely, an alimentary restriction may protect against these diseases. For instance, in rats and mice, a decrease by 30 to 40% of daily food intake or the alteration between 24 h of fasting followed by 24 h of refeeding were found efficient in such a perspective (9,13).

The present results clearly indicate that a third approach, i.e. a daily intermittent fasting for 15 h over a period of 30 days opposes the development of glucose intolerance or frank diabetes in sand rats otherwise exposed to a hypercaloric diet.

Several factors could conceivably participate in such a beneficial effect. The first and major factor consists in the lower caloric intake in intermittently fasting sand rats as compared to non-fasting animals. It could be speculated, however, that fasting and non-fasting animals also differ from one another in their caloric expenditure, e.g. as resulting from the stress and anxiety caused by intermittent fasting. Several findings argue against the latter hypothesis. First, the comparison between food intake and changes in body weight clearly indicates that the caloric expenditure is not higher, but actually lower, in fasting than in non-fasting sand rats. Second, while the sand rats, when first in captivity, exhibited stress by scraping and gnawing their cages, no obvious difference in behaviour was later recorded during the fasting versus non-fasting period. At the most, the fasting rats gave the impression of some tiredness in the morning, before being again given access to food. Third, in their natural biotope, the sand rats are considered as diurnal animals (12). Hence, the food deprivation in the intermittently fasting animals took place during the presumably resting part of their physiological activity cycle. It should not be ruled out, however, that, in their natural biotope, the sand rats remain also somewhat active at night in their burrows (12).

Our study also confirms that, in sand rats, the switch from a purely vegetal diet to a hypercaloric one may either result in overt diabetes or fail to increase significantly postprandial glycemia (14). This difference in the response to the hypercaloric diet is currently attributed to the genetic background, with the existence of two distinct lineages of sand rats, i.e. diabetes prone and diabetes resistant animals (15,16). The latter proposal is consistent with the present finding that, before the switch in food intake from the vegetal to hypercaloric diet, the postprandial glycemia was already higher ($p<0.03$) in sand rats later identified as diabetic (4.19±0.16 mM; n=19) than in the animals later considered as non-diabetic (3.55±0.24 mM; n=19). Likewise, before the switch in food intake, the area under the glycemic curve during an intraperitoneal glucose tolerance test was already higher ($p<0.005$) in the sand rats that eventually developed diabetes (954.8±60.6 mM.min; n=12) than in those who failed to do so (739.0±27.8 mM.min; n=12).

At variance with some other studies, in which the sand rats became diabetic after only 5 days exposure to a hypercaloric diet (16-18), the development of diabetes was more slow in the present study. Thus, as judged from either the difference between diabetic and non-diabetic sand rats at each time point or the changes recorded during the transition period in the diabetic animals, a significant increase of postprandial glycemia in the latter animals was only recorded 21 days after introduction of the hypercaloric diet, whilst still failing to achieve statistical significance 14 days after such an introduction. These two distinct time-related patterns in the development of diabetes may well be related to the fact that, in the present study but not so in the previous ones, the switch from the vegetal to hypercaloric diet was progressive over a 3-week transition period, instead of being immediate.

During such a transition period, no significant difference was found in terms of body weight between diabetic and non-diabetic sand rats. Such was also the case during the later 30-days non-fasting or intermittent fasting period. In both diabetic and non-diabetic sand rats, as well as in control animals, the intermittent fasting provoked a sizeable decrease in body weight, contrasting with the further gain in body weight recorded in the non-fasting sand rats. This decrease of body weight coincides, in both fasting diabetic and non-diabetic sand rats, with a loss of visceral fat mass observed at sacrifice (unpublished observation).

The reduction in body weight, especially in adiposity, is associated with improvement in glucose tolerance (19) and insulin action (3,5). In our study, this improvement of glucose tolerance was reflected both by a reduced area under the

glucose curve during the IPGTT in the fasting groups compared to the non-fasting ones and by a reduced postprandial glycemia which reached values comparable to those recorded before the switch period i.e., before the development of diabetes. The decrease in body weight observed in the intermittent fasting rats and resulting from a lesser food intake, may indeed contribute to the improvement of glucose tolerance, e.g. through a correction of insulin resistance (6,20-22). Such a change in insulin resistance was indeed observed in the present study at sacrifice (unpublished observation).

It cannot be ignored, however, that fasting does not always result in a body weight loss. For instance, in the investigations conducted by Ason *et al* (9) in mice submitted to an alternate day fasting, the food intake on the day following fasting was comparable to that consumed in two days by the non-fasting mice fed *ad libitum*. In human subjects, such an alternate day fasting schedule is uneasy to be respected over a long period because of the discouraging hunger feeling (23). Obese subjects even refuse to follow such a schedule. The results of this study, inspired by the daily fasting period during the Ramadan (12), indicate that, under the present experimental conditions, the food intake remained lower in intermittently fasting animals than in non-fasting rats, suggesting that the frustration during the fasting period did not result in a compensatory excessive food intake during the subsequent non-fasting period of the day.

To our knowledge, it remains to be investigated whether a comparable feeding pattern may prove to be acceptable and efficient in human subjects, e.g. in the perspective of restoring normal body weight in obese subjects.

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