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Fructo-oligosaccharides reduce energy intake but do not affect adiposity in rats fed a low-fat diet but increase energy intake and reduce fat mass in rats fed a high-fat diet☆

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ABSTRACT

The ingestion of low or high lipid diets enriched with fructo-oligosaccharide (FOS) affects energy homeostasis. Ingesting protein diets also induces a depression of energy intake and decreases body weight. The goal of this study was to investigate the ability of FOS, combined or not with a high level of protein (P), to affect energy intake and body composition when included in diets containing different levels of lipids (L). We performed two studies of similar design over a period of 5 weeks. During the first experiment (exp1), after a 3-week period of adaptation to a normal protein-low fat diet, the rats received one of the following four diets for 5 weeks (6 rats per group): (i) normal protein (14% P/E (Energy) low fat (10% L/E) diet, (ii) normal protein, low fat diet supplemented with 10% FOS, (iii) high protein (55%P/E) low fat diet, and (iv) high protein, low fat diet supplemented with 10% FOS. In a second experiment (exp2) after the 3-week period of adaptation to a normal protein-high fat diet, the rats received one of the following 4 diets for 5 weeks (6 rats per group): (i) normal protein, high fat diet (35% of fat), (ii) normal protein, high fat diet supplemented with 10% FOS, (iii) high protein high fat diet and (iv) high protein high fat diet supplemented with 10% FOS. In low-fat fed rats, FOS did not affect lean body mass (LBM) and fat mass but the protein level reduced fat mass and tended to reduce adiposity. In high-fat fed rats, FOS did not affect LBM but reduced fat mass and adiposity. No additive or antagonistic effects between FOS and the protein level were observed. FOS reduced energy intake in low-fat fed rats, did not affect energy intake in normal-protein high-fat fed rats but surprisingly, and significantly, increased energy intake in high-protein high-fat fed rats. The results thus showed that FOS added to a high-fat diet reduced body fat and body adiposity.

1. Introduction

Among the nutritional strategies adopted to control weight gain, diets enriched in dietary fiber or high-protein diets (HP) are both considered to acutely decrease appetite sensations [1–3], reducing energy intake [4–6], preventing fat accumulation and/or reducing body weight in the long term [7–13]. To our knowledge, only one recent study has investigated the effects of separate or concomitant supplementation with soluble fermentable dietary pectin or proteins in rats fed a high-fat diet on their long-term food intake and weight gain. As a result, pectin more than the high protein content decreased food intake and induced body fat loss without there being any effect of combining the two ingredients [14]. Nevertheless, the potential synergy of such a combination still needed to be studied because Adam's results could

Abbreviations: exp1, experiment 1; exp2, experiment 2; NP, normal protein normal fat; NP-FOS, normal protein normal fat with fructo-oligosaccharide; HP, high protein normal fat; HP-FOS, high protein normal fat with fructo-oligosaccharide; NP-HF, normal protein high fat; NP-HF-FOS, normal protein high fat with fructo-oligosaccharide; HP-HF, high protein high fat; HP-HF-FOS, high protein high fat with fructo-oligosaccharide; ACC, acetyl acyl carboxylase; AMM, active metabolic mass; AgRP, Agouti-related peptide; BW, body weight; CART, Cocaine- and amphetamine-related transcripts; CRH, Corticotropin releasing hormone; EI, energy intake; FAS, fatty acid synthase; FOS, fructo-oligosaccharide, LBM, lean body mass; MC_4R, Melanocortin-4 receptor; NEFA, non-esterified fatty acid; NPY, Neuropeptide Y; POMC, Pro-opiomelanocortin; RPL13A, ribosomal protein L 13a; SCFA, short chain fatty acids; TG, triglyceride

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also have been influenced by the type of fiber or protein included in the diet. Moreover, other factors may modulate these effects, including the high fat content of the diet [15]. Among the dietary fibers with interesting nutritional effects, the fermentable fiber fructo-oligosacharide (FOS) has been shown to reduce energy intake and limit body weight gain and fat storage [6,11]. FOS has a negligible viscosity when compared with other fermentable fibers such as pectin and beta-glucan which have high viscosity. Furthermore, proteins, and particularly total milk proteins, are widely acknowledged to reduce energy intake, fat storage and body weight gain [12].

The present study was therefore designed to investigate the ability of FOS (10%) combined or not with a high protein level (P) to affect energy intake and body composition when these ingredients were included in diets containing different levels of lipids (L). Two experiments were conducted in rats fed either a low-fat diet (10% oil/energy) (exp1), or a high-fat diet (35% oil/energy) (exp2), both associated with a normal (NP) (total milk protein) or high protein (HP) content, and with FOS (10%) or not. In both cases, we measured energy intake, body weight gain, body composition, plasma metabolites and the gene expression of hypothalamic peptides controlling food intake.

2. Materials and methods

2.1. Animals and diets

Forty-eight male rats (*Wistar* male Han) aged 8 weeks with an approximate body weight of 225 g, were studied. The animals were placed in a temperature (22 °C ± 1 °C) and humidity (53% ± 2%) controlled room under a 12:12 day: night cycle (lights off from 09:30 and lights on from 21:30). The adaptation period was seven days under a standard chow diet (16.1% protein, 3.1% fat, 59.9% carbohydrate, 5.1% minerals, 11.9% humidity and 3.9% fiber; Scientific Animal Energy Engineering). The animals had ad libitum access to water and food. All of the experiments were approved by the local Animal Care and Ethics Committee (COMETHEA 12/104, Jouy-en-Josas, France). The diets tested during this study varied in terms of their levels of protein and fat, the level of fat/E was low (10%) or high (38%) and that of protein/E was normal (14%) or high (55%) (Table 1).

2.2. Experimental procedure

Experiment 1 (exp1): Following adaptation to the housing conditions, 24 rats were fed with a normal-protein, low-fat diet (NP) for 3 weeks in order to adapt to the NP diet (baseline period). Thereafter, the rats continued with the NP diet or switched to a high-protein diet (HP), both supplemented or not with 10% FOS for 5 weeks. The rats therefore received one of the following four diets for 5 weeks (six rats per group): (i) NP diet, (ii) NP diet supplemented with 10% FOS (Supersmart) (NP-FOS), (iii) high-protein, low-fat diet (HP) and (iv) HP diet supplemented with 10% FOS (HPFOS) (Table 1; Fig. S1). Throughout the study period, the diets were available to the rats for 8 h each day (from 09:30 to 17:30). During the last week of the 5-week period, each group of rats received an 80 kJ preload of their respective diet from 09:30 to 10:00; the food was then removed between 10:00 and 10:30 before being offered again between 10:30 to 17:30. This pattern was chosen in order to train the animals to eat a standard meal within 1 h and thus to standardize both the amount of energy ingested and the physiological state of animals that were to be studied in a fed state on the day of the meal test and at the end of the experiment, the day of euthanasia. The rats ate the entire preload given to them each morning during the last week of the 5-week treatment period. The total daily energy intake encompassed the energy supplied by this preload.

Experiment 2 (exp2): We conducted the same experiment except that a high-fat diet was used instead of a low-fat diet (NP-HF). After the 3-week period of adaptation to NP-HF (baseline period), four groups were generated (six rats per group): (i) NP-HF diet, (ii) NP-HF diet supplemented with 10% FOS (NP-HF-FOS), (iii) HP-HF and (iv) HP-HF diet supplemented with 10% FOS (HP-HF-FOS) (Table 1; Fig. S1). Throughout the study period, the diets were supplied as in experiment 1 and the energy provided by the preload during the last week was 80 kJ. Again, the rats ate the entire preload given to them each morning during the last week of the 5-week treatment period.

2.3. Energy intake, body weight, adiposity and biochemical assays

Energy intake was measured daily during the 5-week treatment period. The rats were weighed every week at the same time (between 09:45 and 10:15) using Sartorius TE2101-L laboratory scales (maximum capacity: 2100 g, reproducibility/standard variation: 0.1 g). Adiposity was evaluated when the rats were euthanized at the end of the experiment. Following euthanasia, the rats were dissected and each organ and each specific tissue (including adipose tissue) was collected and weighed. Following euthanasia, the blood was collected and centrifuged at 3000 rpm for 10 min and the plasma was stored at − 80 °C for subsequent biochemical assays. Blood levels of glucose, non-esterified fatty acids, HDL and triglycerides were determined by the

Table 1
Composition of low-fat and high-fat diets enriched with FOS and protein. Fructo-oligosaccharide (10%) added to these diets did not supply any energy. The energy level (kJ/g) was not the same in the different diets.

	Experiment 1 (low-fat diets)				Experiment 2 (high-fat diets)			
(g/kg)	NP	HP	NP-FOS	HP-FOS	NP-HF	HP-HF	NP-HF-FOS	HP-HF-FOS
Milk protein	140	530	140	530	160	635	160	580
Starch	622.4	287	536.4	201	291.3	53.85	241.3	31.35
Sucrose	1003	45.7	86.3	31.7	291.4	53.85	241.3	31.35
Soybean oil	40	40	40	40	40	40	40	40
Lard	-	-	-	-	120	120	120	120
Minerals	35	35	35	35	35	35	35	35
Vitamins	10	10	10	10	10	10	10	10
Fiber FOS[a]	/	/	100	100	/	/	100	100
Cellulose	50	50	50	50	50	50	50	50
Choline	2.3	2.3	2.3	2.3	2.3	2.3	2.3	2.3
Total	1000	1000	1000	1000	1000	1000	999.9	1000
Energy %								
Protein	14	54,6	16	60,8	14	54.8	15	54.8
Carbohydrate	74	34	71	26	51	9	46	6
Fat	10	10	12	12	35	35	38	38
Energy (kJ/g)	14.57	14.63	13.08	13.14	17.55	17.45	16.02	15.94

[a] FOS fiber (Supersmart) did not supply energy.