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No efficacy of processed Fabuless[™] (Olibra[™]) in suppressing appetite or food intake

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Running Title: Loss of efficacy in processed Fabuless™

Abstract

Background/Objectives: To investigate the feasibility of Fabuless[™] (previously called Olibra[™] and Reducal[™]) as a food ingredient for food intake and appetite reduction, by assessing the effects of food processing on efficacy.

Subjects/Methods: Twenty-four healthy volunteers (16 female, 8 male; age: 18-43 years; BMI: 18-37 kg/m²) took part in a randomised, placebo controlled, double-blind, cross-over trial. Yoghurtbased meal replacement drinks (containing processed or unprocessed Fabuless[™], or a control fat) were followed by an *ad libitum* lunch and evening meal (dinner). Key outcome measures were energy intake and self-reported appetite ratings.

Results: Compared with control, only unprocessed FabulessTM reduced subsequent energy intake, though only during the evening meal (P<0.01; control, processed and unprocessed: 4.3, 3.9 and 4.2MJ respectively) and not during lunch (3.6, 3.7 and 3.6MJ). Self-reported appetite scores did not differ between treatments.

Conclusions: While modest effects of unprocessed Fabuless[™] were seen on food intake, but not on appetite, the ingredient was not robust to common food manufacturing processes (thermal and shear processing). Claims on reduced food intake and appetite relating to this ingredient in food products are therefore only valid where functionality has been demonstrated after all relevant processing and storage steps.

Keywords (MeSH): Fats, emulsions, energy intake, appetite, satiety response, food manufacture.

Introduction

Fabuless[™] (previously called Olibra[™] and Reducal[™]) is a 42% fat emulsion formulated from palm oil and oat oil fractions, used as a food ingredient and claimed to reduce appetite and food intake. It is produced by Lipid Technology Provider (LTP, Sweden) and marketed for satiety benefits in food applications by DSM (The Netherlands). Initial published research, performed in one laboratory by the same research group, reported significant decreases in energy and macronutrient intakes 4-8h post-treatment when Fabuless[™], compared to a control fat, was added to yoghurt (Burns *et al.*, 2000; 2001), and that these effects were dose-dependent (Burns *et al.*, 2002). However, no subsequent studies have replicated these initial results. Indeed, a later study by the same group failed to replicate these positive findings (Logan *et al.*, 2006) after either acute or chronic (3-week) intakes. Similarly, in a different laboratory, Diepvens *et al.* (2008) did not find any overall effects of Fabuless[™] on food intake. Because Fabuless[™] is a complex structured emulsion, rather than a single chemical entity, it is possible that unspecified differences in the material or its handling could contribute toward inconsistencies in its reported efficacy.

In the studies that showed a significant inhibitory effect of Fabuless[™] on food consumption (Burns *et al.*, 2000; 2001; 2002), it is not clear exactly how the active ingredient was added to the test yoghurts, and the amount of processing (i.e., significant mixing or heating) involved. The physical structure of the Fabuless[™] emulsion is thought to be critical for its putative functionality within the gastrointestinal tract and it is not clear to what extent normal food processing may affect efficacy. It is therefore also not clear if research carried out under one set of processing conditions can support claims where this ingredient may undergo a different set of conditions (or for that matter, potential changes during product shelf life). Commercial food manufacturing processes, which guarantee food safety, expose ingredients to shear stress (through mixing and homogenisation) and raised temperatures (through pasteurisation etc). The goal of the study presented here was to assess the feasibility of Fabuless[™] as a hunger control ingredient in yoghurt-based meal replacement beverages, and to establish the impact of specific production processes on its ability to reduce appetite and food intake.

Materials/Methods

Study Design

Twenty-four healthy volunteers were recruited for the study from the Bristol University campus and its surrounding area through an existing participant database, and through posters displayed in

various Departments of Bristol University. Exclusion criteria were: currently dieting to lose weight; getting up and/or having breakfast after 9 A.M. regularly; skipping breakfast, lunch or dinner regularly; self-defined 'picky' eater; simultaneous involvement in other intervention studies, smoker, taking any prescription medication; pregnancy or breastfeeding. In order to minimise expectancy effects, the exact objectives of the study were concealed and the study was therefore presented as investigating 'Effects of a new yoghurt product on mood and hunger'. In addition, subjects were excluded if they scored high for cognitive restraint according to the Dutch Eating Behaviour Questionnaire (DEBQ; van Strien *et al.*, 1986a). The exact exclusion criteria used were (van Strien, personal communication, updated from: van Strien *et al.*, 1986b):

- men with BMI < 27 and DEBQ-restraint score > 2.37;
- men with BMI \geq 27 and DEBQ-restraint score > 3.04;
- women with BMI < 26 and DEBQ-restraint score > 3.24;
- women with BMI \geq 26 and DEBQ-restraint score > 3.41.

The study protocol was approved by the relevant Human Research Ethics Committee at the University of Bristol, and participants provided written informed consent at the end of the practice/familiarisation visit. At the very end of the study, they received a full written debriefing including an explanation of the purpose of the study.

Participants were assigned to the three conditions according to a balanced randomised placebocontrolled, double-blind, cross-over design. The study was conducted over two periods of two weeks; however, due to the limited shelf life of the test product, participants 1-12 were tested in week 1-2 and participants 13-24 were tested in week 3-4. The design of the study balanced treatments within these cohorts.

Before testing commenced, subjects attended an introduction/familiarisation session in the laboratory, where they practiced the main visual-analogue scale (VAS) questionnaire of the study (see below) several times, and completed the DEBQ. Height and weight were measured and they were given a 'Drink, Activity & Evening Snack Diary' for additional data collection, with extensive instructions.

Subsequently, participants returned for their three testing days after fasting from 20:00h the previous evening. On test days, participants consumed their treatment (breakfast as meal replacement drink) at 9:00h, their *ad lib* lunch at 13:00h and their *ad lib* dinner at 17:00h. Before each *ad lib* meal, they were reminded to "eat until you feel comfortably full – no more and no less". The meals selected were intended to be of average liking (not disliked so participants would eat

much less than normal and possibly compensate with larger snacks in the evenings; and not overly liked so participants would eat much more than normal). This was explained, as was the food they would receive and they were asked to consider not taking part if they felt the meals were not moderately liked. Additionally, they were asked to keep an activity and drinks diary on the first testing day, which they had to follow as closely as possible on each of the subsequent testing days with the explicitly explained aim of keeping all testing days as identical as possible and thereby keep inter-day variability to an absolute minimum. Exercise was not allowed on a testing day until later that evening, nor were additional evening meals. Participants were asked not to eat anything else during the testing day until the evenings of the testing days, when snacks and alcoholic drinks were allowed, although these were recorded using the same diary. Also, participants were instructed to bring some light reading material to entertain themselves during in-laboratory waiting times, and magazines were also available. Each participant occupied an individual testing booth where distraction was kept to a minimum: they were visually separated; talking amongst participants was prohibited; and mobile phones were handed to the experimenter to avoid any further distractions. Participants were allowed to leave when all present had finished eating their test meal, although for lunch and dinner, subjects could leave only after the first post-meal questionnaires (30 minutes after the start of the meal; see below) were completed.

At baseline and every 30 minutes post-treatment until after dinner, VAS questionnaires were filled out using a pre-programmed Palm Zire[™] 21 handheld Personal Digital Assistant (PDA). VAS questionnaires recorded appetite and mood related feelings (see below). Five of these questionnaires were followed by a 4-point 'physical symptoms' questionnaire, including one immediately premeal, one at 11:00h and one at 15:00h. Additionally, a palatability questionnaire was completed for each of the treatments.

A schematic representation of the various activities can be found below in Figure 1.

INSERT FIGURE 1 HERE

Test Foods

Breakfast (Treatment) The three treatment yogurt-based meal replacement drinks or 'test drinks' (325ml each) contained either 12.5g FabulessTM (5g fat; added during manufacture, thus 'processed'); 12.5g FabulessTM (5g fat; added at the end of manufacture, thus 'unprocessed') or 5g

control fat (milk fat +corn oil). Additions of Fabuless™ at or below 12.5g has been reported to substantially reduce food intake in the original efficacy studies (Burns et al., 2000; 2001; 2002). In the "processed" condition Fabuless was mixed with all other ingredients at the start of the production process. Subsequent processing steps involved pre-heating to 72°C, homogenisation at 65°C, pasteurisation at 92°C for 5 minutes, cooling to 42°C for 5 hours, pH adjustment to 4.6 and final homogenisation with fruit-based ingredients. In the "unprocessed" condition, Fabuless was added after this final step with minimal mixing, then manually filled into aseptic containers for storage at $< 8^{\circ}$ C. The product description of the test drinks was "A fresh raspberry voghurt beverage containing vitamins and minerals, with sugars and sweeteners", and contained the following ingredients: Live yoghurt (milk, whey, milk protein); raspberry puree; sucrose; glucose; oligofructose (fibre); vegetable oils; minerals; vitamins; stabiliser (pectin); sweeteners (aspartame, acesulfame K); flavouring; colour (carmine). They were stored at max 8°C to ensure a shelf-life of 14 days. Serving size for all three treatments were 325ml (336g). Energy content was identical, and macronutrient composition was virtually identical between treatments. FabulessTM containing test drinks (both processed and unprocessed): 266kcal, 15.2g protein (23% of delivered energy), 5.7g fat (19% of delivered energy) and 38g carbohydrate per serving; Control test drinks: 268kcal, 15.4g protein (23% of delivered energy), 5.8g fat (19% of delivered energy) and 39g carbohydrate per serving. The test drinks were manufactured by International Food Network Ltd, Reading, UK.

Lunch This *ad lib* meal consisted of: 1) A large plate of standardised cheese sandwiches made of 10 slices of bread with 5 slices of medium strength Cheddar cheese (24g each). Sandwiches were cut to the size of the cheese slice. Each slice of bread was buttered with 3.0g of margarine, and sandwiches were divided into four equal smaller triangles before serving; 2) A smaller plate containing one pack (200g) of 'custard creams' biscuits; 3) A clear jug containing 1 litre of non-carbonated water. Total energy content of the meal served was 10531 kJ.

Dinner This *ad lib* meal consisted of: 1) A large plate containing around 1000g bacon & leek pasta bake, prepared immediately before serving in a microwave oven according to food manufacturer's guidelines; 2) A bowl containing 450g strawberry yoghurt; 3) A clear jug containing 1 litre of non-carbonated water. Total energy content of the meal served was 8844 kJ.

Measures

The measures taken for statistical analysis were:

1) Energy intake (kJ) at lunch, dinner, and lunch and dinner combined;

- 2) Food intake (g) at lunch, dinner, and lunch and dinner combined;
- 3) Appetite and mood related feelings (100-point VAS), measuring 'Full', 'Hungry', 'Appetite for a snack', 'Thirsty', 'Satiated', 'How much you could eat', 'Energetic' and 'Drained';
- Physical symptoms experienced in the past hour (4-point scale, labelled 'not at all', 'mild', 'moderate' and 'severe');
- 5) Pleasantness ratings for various sensory aspects of the yoghurt-based meal replacement drink (100mm paper VAS).

Appetite, mood and physical symptom ratings were recorded electronically on a Palm Zire[™] PDA.

Statistical Analysis

Statistical analysis was carried out using SAS 8.0. As there were negligible differences between ANOVA and ANCOVA outcomes, estimated means adjusted for baseline differences were not calculated, and overall treatment effects for the VAS and food intake data were eventually analysed using ANOVA with subject, treatment and period as factors. P-values <0.05 were considered statistically significant. Next, 'active' treatments were compared against control using Fisher's Least Significant Difference (LSD) test as reported in the results section. The comparisons (1-tailed t-tests) were corrected for multiple comparisons by using a pooled error term. Frequencies of physical symptoms were compared between treatments using χ^2 .

RESULTS

Participant Characteristics

Twenty-four participants (16F, 8M) were recruited into the study, with an average age of 23.3 years (\pm 5.3; range 18-43), mean Body Mass Index (BMI) of 22.4 kg/m² (\pm 3.8; range 18.1- 37.0) and DEBQ-restraint scores of 2.1 (\pm 0.6; range 1.2-3.2). Age did not differ significantly between the two sexes, though females showed a predictable trend for higher scores on BMI and dietary restraint (DEBQ restraint scale) compared to males. Baseline characteristics are presented below in Table 1.

INSERT TABLE 1 HERE

Pleasantness and mood ratings, and physical symptoms

No differences in overall product pleasantness (taste, aroma, flavour), mood or adverse physical symptoms were observed between treatments (Ps>0.1).

Appetite and satiety ratings

No treatment effects were found for any appetite measures over various time periods using Area-Under-Curve (AUC) data (Ps>0.5). Additionally, sporadic effects at individual time points, usually immediately pre-meal, are not confirmed by other, related measures (see Figure 2).

INSERT FIGURE 2 HERE

Food and energy intake

Results for food intake at lunch and dinner are shown in Table 2

Lunch

Both energy intake (kJ) and food intake (g) at lunch were not significantly affected by either of the FabulessTM test drinks when compared to control (Ps>0.2).

Dinner

Energy intake at dinner was significantly lower (pasta alone: t(42)=2.79; P=0.004; dinner total: t(42)=2.46; P=0.009) following the unprocessed FabulessTM test drink compared to control, but not following processed FabulessTM vs. control (Ps>0.1).

Accordingly, food intake at dinner was also reduced by the unprocessed FabulessTM test drink compared to control [t(42)=211; P=0.021]. In contrast, neither energy or food intake at dinner were reduced by the processed FabulessTM test drink compared to control (P=0.2). The results were similar for total (lunch + dinner) energy intake.

INSERT TABLE 2 HERE

DISCUSSION

The present study found that processing of Fabuless[™] in a yogurt beverage eliminated the modest, but statistically significant effects on food intake observed when the ingredient was added at the end of manufacture. The significant effects of the unprocessed ingredient were found at eight hours after treatment consumption (dinner), and generally speaking not at four hours post-treatment (lunch).

An ingredient that reliably leads to significant and meaningful reductions in energy intake could be highly beneficial in a weight management context and therefore highly desirable to consumers who want to achieve an ideal weight. FabulessTM is marketed as such an ingredient based on data obtained from an initial series of studies reported by Burns *et al.* (2000; 2001; 2002), which suggested that FabulessTM could reduce 24h energy intake by up to an impressive 30%. However, subsequent studies by the same and other groups (Logan *et al.*, 2006; Diepvens *et al.*, 2008) have not replicated these initial results, and the nature of the ingredient (a structured emulsion) suggests it could be sensitive to common processes used in food production.

The sample size and sensitivity of measures in the current study were appropriate for this type of study design (Flint *et al.*, 2000); Gregersen *et al.*, 2008). However, the magnitude of food intake effects for unprocessed FabulessTM seen here (~10% energy intake reduction compared to control) was much less than in the Burns *et al.* (2000; 2001; 2002) studies (average = 23% reduction). The reasons for this difference are not entirely clear. However, compared to other studies, the magnitude of effect reported by Burns et al. is unusually large, especially considering that anorectic drugs have achieved similar magnitudes of effect. For example, Rogers & Blundell (1979) found that fenfluramine reduced food intake by 26%, whilst Rolls *et al.* (1998) showed a maximum energy intake reductive effect of 26%, but only after a 14-day sibutramine course.

Our findings may partially replicate results from Burns *et al.* (2000; 2001; 2002) for the unprocessed Fabuless[™] used here, but show that thermal and shear processing eliminates the functionality of this food ingredient. Indeed, high-shear mixing and homogenising may destabilise emulsified structures by means of coalescence or flocculation of the dispersed phase, whilst subsequent pasteurising may provide and additional destabilising risk through protein denaturisation (McClements, 2005). Note that it is unclear exactly what type or level of processing and storage of Fabuless[™] was applied in the 'Burns' trials. Commercial products containing Fabuless[™] will typically be exposed to the same or other basic food manufacturing processes as used in the 'processed' treatments in our study. Nevertheless, micrographic images of the products used in this trial (not shown here) indicate a finer dispersion of the lipid fraction in Fabuless[™] after processing. However, we are not aware of any structural assessment of Fabuless[™] that definitively predicts integrity of its functional efficacy. Without such a test, it is difficult to predict when functionality is retained, other than by empirical clinical testing. Therefore, evidence for the effectiveness of Fabuless[™] is only relevant where there is explicit evidence that functionality (and not just a particular structure) is retained in products when processed, stored and prepared as

relevant to the market product. While it is certainly possible that different or milder processes may have less effects on efficacy, the effects and limits of process-sensitivity need to be shown.

One remarkable aspect of the study presented in this paper is the general lack of effects on selfreported appetite and satiety measures. Methodologically, the VAS measures were sound as clear effects were found in a subsequent work using the same participants, measures and instructions (not related to the current hypothesis and/or Fabuless[™]; data not published). Note, however, that Burns *et al.* (2002) also reported no effects of Fabuless[™] on appetite related ratings, regardless of test conditions.

The proposed mechanism in support of the claims for Fabuless[™] has been termed the 'ileal brake', where fat when presented to the distal ileum elicits a strong satiety response (e.g., Lin *et al.*, 1996), an inhibition of upper gastrointestinal functions elicited mainly by the release of GLP-1 in response to the presence of unabsorbed nutrients in the ileum (e.g., Delgado-Aros et al., 2002; Brubaker et al., 2002; Näslund et al., 1999; Giralt & Vergara, 1999). Even the infusion of 3g fat into the ileum delayed small intestinal transit (Symersky et al., 2004, in: Maljaars et al., 2008). Although it is unclear how much undigested fat of the 5g fat in the Fabuless[™] conditions reaches the ileum, it is unlikely to be more than 3g. Moreover, Fabuless[™] is generally added in amounts less than 5g per daily intake equivalent. Finally, although fat is the most commonly researched macronutrient in this respect, also undigested carbohydrates and proteins can induce the ileal brake mechanism (Maljaars et al., 2008), and the mechanism underlying the ileal brake effect is yet to be established (Maljaars et al., 2008). Note also that there is currently no direct evidence in support of the proposed 'targeted delivery' mechanism in FabulessTM (i.e., how the emulsion passes through the stomach into the ileum without being affected). Although a recent report by Haenni et al. (2009) claims that Fabuless[™] leads to a 45 min longer oro-caecal transit time (OCTT), this seems incongruent with the fact that the only significant effects observed in our study were at 8 hours post-ingestion. In addition, Haenni et al. (2009) used an unconventional method of calculating OCTT (using group mean plasma marker values instead of individual marker appearance times; e.g., Staniforth (1989) and Peh & Yuen (1996)), which may have exaggerated individual outlier responses.

In summary, the results of this study show a modest, statistically significant effect only of unprocessed FabulessTM on food and energy intake, while no significant effects were observed when the active ingredient was added prior to homogenisation and pasteurisation. These findings replicate to some extent the earlier demonstrations of inhibitory effects of FabulessTM on food and

energy intake (Burns *et al.*, 2000; 2001; 2002), although the magnitude of effects were much smaller here, and perhaps more realistic. Moreover, no effects of FabulessTM on appetite related ratings were found. Given the clear loss of efficacy when FabulessTM is subjected to normal food processing, functionality after actual processing and storage conditions must be empirically confirmed for any product making claims based on this ingredient.

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Conflict of Interest

Dr Smit, Prof Rogers and Mrs Keenan's work was funded by Unilever. They declare no conflict of interest. Dr Mela, Dr Wiseman, Dr Kovacs and Dr Peters represent the funding company.

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FIGURE 1 – Typical testing day schedule

FIGURE 2 - hunger, fullness and satiety scores collected during a typical testing day. Test drink (yoghurt-based meal replacement drink), lunch and dinner were provided at times '0 min', '240 min' and '480 min', respectively. A = unprocessed vs control; B = processed vs control; *: P<0.05; **: P<0.01

Table 1 Subject characteristics at baseline

	Overall (N=24)	Males (n=8)	Females (n=16)	
Age (years)	23.3 (±5.3)	21.4 (±2.8)	24.2 (±6.1)	
BMI (kg/m2)	22.4 (±3.8) [†]	24.3 (±5.3)	21.5 (±2.4)	
DEBQ-restraint	2.1 (±0.6) [†]	1.8 (±0.6)	2.3 (±0.6)	

Abbreviations: BMI – Body Mass Index; DEBQ – Dutch Eating Behavior Questionnaire. Mean \pm s.d.; † indicates a marginally significant difference between men and women: 0.1>P>0.05.

		control	unprocessed	processed
Energy intake (kJ)	+4h (lunch)	3656 (±129)	3559 (±129)	3625 (±129)
	+8h (dinner)	4339 (±120)	$3921 (\pm 120)^{a}$	4164 (±120)
	Lunch + dinner	7995 (±200)	7480 (±200) ^b	7788 (±200)
Food intake (g)	+4h (lunch)	929 (±23)	865 (±23)	902 (±23)
	+8h (dinner)	1655 (±35)	$1572 (\pm 35)^{b}$	1614 (±35)
	Lunch + dinner	865 (±23)	$929 (\pm 23)^{b}$	902 (±23)

Table 2 Food and energy intake at 4 and 8 hours post-intervention



Measurements:

EI = measurement of energy intake S = questionnaire on satiety feelings and fatigue

T = questionnaire on taste and liking

GI = questionnaire on gastrointestinal complaints

