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Perceived thickness and creaminess modulates the short-term satiating effects of high protein drinks

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ABSTRACT

 Previous research suggests that increasing beverage protein content enhances subsequent satiety, but whether this effect is entirely attributable to post-ingestive effects of protein or is partly caused by the distinct sensory characteristics imparted by the presence of protein remains unclear. To try and discriminate nutritive from sensory effects of added protein, we contrasted effects of three higher energy (c. 1.2MJ) and one lower energy (LE: 0.35MJ) drink preloads on subsequent appetite and lunch intake. Two higher energy drinks had 44% of energy from protein, one with the sensory characteristics of a juice drink (HP-) and the second thicker and more creamy (HP+). The high-carbohydrate preload (HC+) was matched for thickness and creaminess to the HP+ drink. Participants (healthy male volunteers, n=26) consumed significantly less at lunch after the HP+ (566g) and HC+ (572g) than after HP- (623g) and LE (668g) drinks, although the compensation for drink energy accounted for only 50% of extra energy at best. Appetite ratings indicated that participants felt significantly less hungry and more full immediately before lunch in HP+ and HC+ compared to LE, with HP- intermediate. The finding that protein generated stronger satiety in the context of a thicker creamier drink (HP+ but not HP-), and that an isoenergetic carbohydrate drink (HC+) matched in thickness and creaminess to the HP+ drink generated the same pattern of satiety as HP+ both suggest an important role for these sensory cues in the development of protein-based satiety.

Introduction

 It has been widely reported that meals with a higher proportion of energy as protein are more satiating than isoenergetic meals lower in protein content both in acute tests of satiety using 53 short-term measures of rated appetite and/or intake $(1-10)$ and longer-term studies on manipulated 54 protein content of the diet^{$(11-14)$}. However, there remains some uncertainty about the mechanisms underlying the enhanced satiating efficiency of protein-based foods and drinks. Although there is clear evidence that protein ingestion results in a different profile of satiety-57 related hormonal signals compared to other macronutrients^{$(15-17)$} that has been interpreted as the 58 basis of protein-based satiety⁽¹⁸⁾, a confounding issue in interpretation of many short-term studies of protein-based satiety is the difficulty in fully disguising the addition of protein. This often results in orosensory differences between protein and control conditions that could also contribute to the behavioural effects of these foods and drinks. It is well established that orosensory cues are an important component of short-term satiety. For example, high-energy preloads have been shown to be more satiating when ingested by the participant than when 64 infused directly into the stomach or intestine (19) . Observations like this add weight to the 65 satiety-cascade model (20) , where learned and sensory cues from food are suggested to be critical components of the short-term satiating effects of nutrients. Several recent studies provide additional evidence to support this view. Firstly, sensory characteristics that were consonant with the presence of energy (thickness and creaminess) enhanced the satiating effects 69 of energy in a drink context (21) . Secondly, the sensory characteristics, but not protein content, 70 of a snack preload altered subsequent selection of protein-rich foods (22) . The present study extends these findings to ask whether perceived thickness and creaminess imparted by addition of protein in a beverage may at least in part explain why protein-enriched foods and drinks are found to be more satiating than are other macronutrients in short-term tests of satiety.

 A key driver for the present study was an earlier investigation in our laboratory that found that a drink preload containing 50% of additional energy as protein was more satiating than an 77 isoenergetic drink enriched with carbohydrate only⁽²³⁾. Indeed in that study there was no evidence of satiety, either through reduced intake at a test lunch or in altered appetite ratings, after the high-energy (1250 kJ) carbohydrate-enriched drink compared to the low-energy (327 kJ) control drink. This finding is consistent with broader suggestions that energy consumed in 81 beverage form generates weak satiety⁽²⁴⁾. In this previous study we attempted to disguise the nutritional differences between the two high-energy drinks, however evaluations by participants clearly reported subtle sensory differences, with the high-protein drink rated as slightly more creamy, slightly thicker in texture and less pleasant than the carbohydrate drink. Therefore, sensory differences may have contributed to the short-term satiating effects of the protein drink rather than simply post-ingestive effects. More recent studies suggest a key role for sensory 87 characteristics in determining the satiating effects of beverages^{(21)}.

 The present study directly assessed the importance of sensory properties by contrasting the satiating effects of three isocaloric high energy drinks relative to a low energy control. Two versions of the high-energy drinks were enriched with protein but differed sensorially: one high-sensory protein drink (HP+) was created to taste slightly thicker and creamier than the other (HP-). The third high energy drink (HC+) was enriched purely by carbohydrate and had its flavour adjusted to match that of the high-sensory (HP+) protein drink. Since the same high carbohydrate formulation in the absence of sensory cues was not satiating in our previous 96 study⁽²³⁾, any evidence that the sensory-enhanced HC+ drink resulted in satiety would be clear evidence that sensory characteristics such as thicker texture and creamy flavour may be a key element of the generation of satiety by nutrients in a beverage context. Thus, if the enhanced satiating effects of addition of protein are only a consequence of post-ingestive actions, the prediction would be that the HP- and HP+ drinks would have similar effects on subsequent

- rated appetite and intake at a test meal. In contrast, if protein-induced satiety is dependent on
- the sensory characteristics imparted by the added protein, then the two sensory-enhanced drinks
- (HP+ and HC+) would be predicted to be more satiating than the high-protein low-sensory (HP-
-) drink. Thus the present design provided a clear means of dissociating the potential roles of
- sensory and post-ingestive effects of the satiating effects of protein.
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Method

Design

 A repeated measures design contrasted satiety (changes in rated appetite and test lunch intake) following consumption of four preload drinks. Three preloads had a higher energy content, two with 44% of energy added as protein either with (HP+) or without (HP-) enhanced creaminess and thickness, and the third (HC+) had energy added as carbohydrate but thickness and creaminess matched to the HP+ condition. The fourth preload was a low-energy control (LE).

Participants

 Potential participants were recruited from participant databases held by the School of Psychology, University of Sussex, on the basis that they were participating in a study about mood and food. Inclusion criteria were young men aged 18-35 years of age whose body mass 121 index (BMI) was within the normal range (18-25 kg/m²). Healthy normal weight men were tested to minimise demand effects generated by the laboratory testing setting. Exclusion criteria included smoking more than 5 cigarettes a week, an eating, metabolic or respiratory disorder, any athletes in training, and those having a restrained eating style defined as individuals scoring seven or more on the restraint scale score from the Three Factor Eating 126 Ouestionnaire (TFEO)⁽²⁵⁾. Participants gave written informed consent and the protocol was approved by the Sussex University Ethics Committee. Two participants failed to attend all sessions and their data were excluded. The 26 male participants who completed all sessions had a mean age of 21.1 years (SD: 2.3), a mean TFEQ restraint of 2.7 (SD:2.4) and normal BMI 130 of 21.9 kg/m² (SD:1.6). Participants received £40 for participation.

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Test preload drinks

 Drinks were developed iteratively using taste tests with volunteers to create two high protein drinks (HP+ and HP-) with similar energy content, one resembling a juice drink, and the other perceived by volunteers to be a creamy drink. The HC+ drink was developed to match the HP+ in terms of perceived thickness and creaminess but with the additional energy added as carbohydrate only. The final prototype drinks were assessed by an untrained panel of 10 male volunteers who were provided with 20ml samples of each of the high-energy preloads, served in 50ml containers covered in foil to obscure visual cues. They were instructed to take a sufficient mouthful to allow completion of a series of sensory ratings, and were provided with water to cleanse the palate between mouthfuls. Sensory evaluations were made using 100mm pen and paper visual analogue scales (VAS). Ratings confirmed that the two high-energy high- sensory drinks (HP+ and HC+) were significantly thicker [F(1.1, 8.8) = 9.74, *p<*0.05] (HP+: 73 146 \pm 6; HC+: 72 \pm 5), and had higher "dairy-like" characteristics [F(1.1, 9.1) = 8.16, *p*<0.05] 147 (HP+: 59 ± 8 ; HC+: 66 ± 8) than the HP- beverage (dairy: 32 ± 9 ; thickness: 38 ± 10). HP+ and 148 HC+ also tended $[F(2,16)=2.42, NS]$ to be perceived as creamier (HP+: 59 \pm 8; HC+: 66 \pm 8) 149 than the HP- drink (32 \pm 9). The overall pattern of data confirmed that HP+ and HC+ were reasonably well matched on the sensory characteristics we were interested in, and both were perceived as thicker and more creamy than was HP-.

 The composition of the preloads is summarised in Table 1, and all were prepared from a base of low-energy fruit-yoghurt drink (Apricot and Peach drink Danao®, Danone). HP+ and HP- were developed to provide 44% of energy as protein and HC+ contained 87% of added energy as carbohydrate and 13% as protein. Protein content was varied through use of different amounts of virtually fat free fromage-frais (Waitrose brand) and a whey isolate (CMC Whey®, Fast Research, Staffordshire, UK), which at the concentrations used had reduced bitterness compared with other whey sources and so was easier to disguise. Carbohydrate was added as a

combination of maltodextrin (Cerostar) and sucrose. HP+ and HC+ had added yoghurt and

vanilla flavours (IFF) to enhance perceived creaminess and a small amount of guar gum

(Meyprodor, a water soluble fibre) to enhance perceived thickness. The LE condition used the

base drink diluted with water.

Test meals

Participants consumed a standardised breakfast in the laboratory on each test day consisting of

breakfast cereal (either Crunchy-nut cornflakes or Special K cereal, both Kellogg's UK),

orange juice and semi-skimmed milk (1710.2 KJ). The test lunch comprised *ad libitum*

consumption of pasta (fusilli variety, Sainsbury's UK) mixed with commercial tomato-based

herb sauce (Napoletana, Sainsbury's UK) and served in bowls at a ratio of 250g cooked pasta to

250g sauce. The test meal provided 500KJ (3.7g protein; 19.8g carbohydrate; 1.5g fat) per

100g.

Assessment of rated appetite, mood and food intake at the test lunch

 Data were collected using the Sussex Ingestion Pattern Monitor (SIPM: University of Sussex), 176 a computer-based Universal Eating Monitor^{(26)} for measuring food intake and recording rated 177 appetite (27) . This ensured minimal monitoring or disturbance from the experimenter. SIPM consisted of a disguised electronic balance (Sartorius BP 4100-S, Sartorius, Goettingen, Germany) fitted into the desktop and connected to an Apple Macintosh G3 computer, with the balance surface obscured by a placemat. The system was custom programmed using FutureBasic (Staz Software) to read the balance weight on stability to 0.1g accuracy during the test meal. At the start of the lunch session a 500g plate of pasta was placed on the balance and the experimenter left the cubicle. The computer instructions were to "Eat as much as you want". A separate side plate was provided to place cutlery on when not eating so that the weight of cutlery did not interfere with weighing. The SIPM system prompted participants to

 call the experimenter for a refill after the sixth interruption to their meal, by which time 300- 400g had been consumed, which ensured that participants could not use an empty bowl as an external cue to end their meal. This process was repeated until the participants indicated that 189 they had "finished" their meal.

 Before and after each preload and meal, participants completed computerised ratings of hunger, fullness, thirst, clear-headed, happy, friendly, jittery, nauseous, energetic, relaxed, presented in the form "How <descriptor> do you feel?". Mood ratings were included as distractors. Ratings were made by electronic VAS end-anchored with "Not at all" (scored zero) and "Extremely" (scored 100). Sensory and hedonic ratings (familiar, sweet, pleasant, sour, bitter, creamy, fruity, refreshing, thick, novel, dairy, fatty) of the preload were made using the same style of VAS when the drink was first tasted and once it had been consumed in full, and participants also rated the lunch when first tasted and at the end of the meal. Polarity of all computerised ratings was randomised to minimise carry-over effects.

Procedure

 Participants were instructed to eat as normal on the day before testing, but consume only water from 11pm the prior evening. On each test day, breakfast was served between 08.30 and 10.00h, and participants left the laboratory after breakfast before returning for their later appointments, but were restricted to drinking water only during this period. A 500ml bottle of water was provided to encourage water consumption throughout the morning. To encourage compliance with instructions not to eat or drink anything other than water, participants were warned that random samples of saliva could be collected at any time during the study (this was not followed up). Participants returned to the laboratory 180 minutes after breakfast and consumed the relevant preload in a small, ventilated cubicle where they also completed the mood and appetite ratings. Preloads were served in a 400ml polystyrene cup with an opaque lid and straw, and participants were instructed to consume all the drink within 10 minutes. To monitor compliance, each preload was weighed before and after consumption and preload session duration recorded. Once they had consumed the preload and completed the associated ratings they rested in an adjacent waiting room until lunch, which was served 30 minutes after the preload session began. The delay between preload and lunch was selected based on an earlier study, where similar drinks had the same impact on subsequent appetite regardless of 218 whether they were consumed 30 or 120 minutes prior to the test meal⁽²³⁾. Once they had consumed as much of the lunch as they wanted and had completed all ratings, they were free to leave except on the final session, when they had a structured debriefing where they were asked about the purpose of the study. Participants were also asked if they had noticed differences between the preloads, breakfast or lunch meals across the test days and were asked: "Have you ever tasted a high protein shake – otherwise known as body building drinks?" to judge familiarity with products like the drinks under test.

Data analysis

 Intake data were contrasted between the four preload conditions using one-way repeated measures ANOVA, with the prediction that all three higher energy preloads would reduce intake but that HP+ and HC+ would have a larger effect than HP-. Total energy intake was calculated as the sum of energy consumed at breakfast, preload and test meal, and these were contrasted using ANOVA. The degree of compensation at the *ad libitum* meal for the energy consumed in the preloads was calculated as the energy difference between each high energy test 233 preload and the LE, expressed as a fraction of the reduction $(28, 29)$. Computer failure meant all rating data were lost for one participant on one day, and initial analysis of changes in hunger after preload consumption identified one participant as a significant outlier (data more than 2 standard deviations from the mean) in two preload conditions and his data were excluded from further analysis. After confirming there were no spurious baselines differences, changes in

 hunger and fullness immediately after consuming the preload and at the start of lunch were calculated and contrasted using 2-way ANOVA. Similarly, sensory and hedonic ratings before and after preload consumption were contrasted between preloads to confirm the expected sensory differences were evident and that these did not generate confounding differences in liking. Within-subjects contrasts were used to test specific predictions and Bonferonni post hoc corrections applied when making post-hoc comparisons. Data were analysed using SPSS 18 for Macintosh.

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Results

Intake

Lunch intake varied significantly between preload conditions (F(3,75) =6.26, *p<*0.01: Figure

250 1a), with intake following the two thicker and more creamy drinks $(HP+$ and $HC+$

significantly less than after the LE control (*p*<0.01, *p*<0.001 respectively). Critically, intake

after the thick/creamy high protein HP+ drink was significantly less than after the high protein

253 drink without thick/creamy sensory characteristics (HP-, p<0.05), and intake after the HP- drink

did not different significantly from that after LE (Figure 1a). Short-term total energy intake

(Figure 1b) also differed significantly between conditions (F(3,75)=11.13, *p<*0.001), with

significantly greater energy intake in all three high-energy conditions compared to LE although

257 total energy intake was significantly lower in the HP+ than HP- condition $(F(1,25)=5.46,$

*p<*0.05). Overall compensation for preload energy was 22.4% in the HP- condition compared

with 50.2% in the HC+ and 52.6% in HP+ conditions.

Rated hunger and fullness

 Rated hunger and fullness immediately before preload consumption did not differ significantly 263 between preload conditions [hunger: $F(3,72) = 2.23$, NS; fullness $F(3,72) = 2.48$, NS]. As

264 expected, changes in hunger depended on time of rating $[F(1,72) = 14.07, p<0.001]$, with a larger initial decrease in hunger immediately after preload consumption and some recovery of 266 hunger by the lunch test. There was a trend for a significant overall effect of preload $[F(3,72) =$ 267 2.67, p=0.056], but the interaction between time and preload was not significant $[F(3,72) =$ 0.86, NS]. As can be seen (Table 2), hunger decreased immediately after consuming all four preloads but this decrease was only sustained in the HP+ and HC+ conditions. The decrease in hunger in both the HP+ and HC+ conditions immediately before lunch was significantly greater 271 than that in the LE control condition (both p<0.05) with changes after HP- intermediate and not significantly different from other preloads. A similar pattern was seen with fullness ratings 273 (Table 2), and here the effects of time $[F(1,72) = 14.87, p \le 0.001]$, preload $[F(2,72 = 8.37, p \le 0.001]$ 274 p<0.001) and the preload x time interaction $[F(3,72) = 3.09, p \le 0.05]$, were all significant. Rated fullness increased in all four conditions immediately after consuming the drinks, although this increase was significantly greater in the HC+ than in the other three conditions (LE p<0.001, HP- p<0.05, HP+ p<0.01). However, the initial increase in fullness was not sustained in the LE condition, and immediately before lunch the largest increases in fullness were seen in the HP+ and HC+ conditions.

Rated thirst and nausea

 Protein-elicited thirst presented a possible confound for interpretation of this study (Table 2). As baseline first did not differ significantly between conditions, change data were used to 284 contrast effects of preloads. Thirst varied with time $(F(1,72) = 6.88, p<0.05)$, with the expected large decrease immediately after drink consumption, but although the main effect of preload 286 condition was not significant $(F(3,72) = 1.33, NS)$ there was a significant interaction between 287 Preload and Time $(F(3,72) = 3.22, p<0.05)$. Surprisingly thirst was reduced more after the two high protein preloads relative to the LE control and HC+ preloads prior to lunch.

 Differences in lunch intake could also have been confounded by any gastric discomfort from consuming these drinks. However, if so then we would have expected differences in nausea ratings between preloads however there was no significant difference in baseline nausea 293 [F(3,72) = 1.66, NS], and no significant effects of preload [F(3,72) = 0.29, NS], time [F(1,72) = 294 1.43, NS or time x preload interaction $[F(3,72) = 2.39, NS)$ for changes in nausea immediately and 30 minutes after preload ingestion.

Sensory and hedonic ratings of the test meal and preloads

 To assess whether the sensory differences evident during pilot work were detectable during the satiety tests, evaluations of the four preloads at the start and end of ingestion were examined. To allow comparisons between pilot and test data, only ratings at the initial taste test are shown 301 (Table 3). As expected, preloads differed significantly in perceived creaminess $[F(3,75) =$ 37.00, p*<*0.001], thickness [F(3,75) = 23.82, p*<*0.001], fattiness, [F(3,75) = 16.39, p*<*0.001] and perceptions of dairy [F(3,75) = 17.01, p*<*0.001]. HP- was rated as significantly less thick and less fatty than were the HP+ and HC+, but (in contrast to pilot data) was rated similarly on creaminess and dairy-like characteristics. Sensory ratings did not differ between the start and end of preload ingestion, with only one significant interaction arising from evaluation of ratings of the "dairy-like" characteristics [F(2.0,47.6) = 2.80, *p<*0.05], although within-subjects contrasts did not identify the cause of that interaction which may be spurious. The drinks did 309 not differ significantly in sweetness $[F(3,75) = 1.10, NS]$, bitterness $[F(3,75) = 0.47, NS]$ or novelty (F(3,75) = 1.93, NS). As expected, rated novelty declined significantly between the start and end of ingestion [F(1,25) = 10.48, p*<*0.01].

 There were no overall significant differences in rated pleasantness of the four preloads [F(3,75)=2.70, NS], but there was a significant interaction between preload and rating time [F(3,75)=6.27, p*<*0.001]. Ratings before ingestion did not differ significantly between

316 conditions (F(3,75) = 0.86, NS). However, pleasantness decreased significantly for the HP+ and HP- preloads but did not change in HC+ or LE conditions (Figure 2). There were no significant differences in overall rated pleasantness of the pasta between conditions [F(3,75)=1.92, NS] nor any interaction between Preload and Taste [F(2.4,59.7)=1.59, NS]. Rated pleasantness of the pasta declined significantly from start to end of the meal in all conditions [F(1,25)=26.60, *p<*0.001]. **Participant awareness** The majority of participants (20/26) believed the experiment was investigating "food and mood" in line with the explanation provided during recruitment. Two participants correctly identified: "effects of the drink upon appetite/the meal". Ten participants correctly said they received different drinks each test day, while nine participants recalled noticing only two different drinks. Overall these responses indicate that many participants were not overtly aware of the purpose of the experiment. None of the participants reported regularly consuming commercially available protein drinks. **Discussion** In this study the addition of protein to a beverage only resulted in short-term satiety when the addition of protein was combined with small increases in thickness and creamy flavour. Thus the sensory-enhanced HP+ drink was more satiating than the same level of protein added in the absence of sensory cues (HP-). Moreover, whereas the addition of extra energy purely as 340 carbohydrate was previously found to be ineffective at generating satiety in this context⁽²³⁾.

when the same carbohydrate was added alongside increased creamy flavour and thickness (the

 HC+ preload), the drink was as satiating as was the HP+ drink. Together both the difference in satiety response between protein drinks which differed in sensory characteristics and similarity of response to drinks that were perceived as similarly thick and creamy but which differed in macronutrient content (HP+ and HC+) suggest that the sensory characteristics of beverages are critical in determining short-term satiety.

 The key question is what explains the difference in satiety between HP+ and HP- conditions. This effect cannot easily be attributed to nutritional differences since these preloads had similar amounts of added protein, both chiefly through different extracted versions of whey protein. Many studies suggest that whey protein is more satiating than other forms of protein based on 352 both greater compensatory eating responses⁽³⁰⁾, greater suppression of rated appetite^(17, 31) and 353 increased release of satiety hormones^{$(17, 31)$} after consuming preloads enriched in whey protein, 354 although some studies failed to confirm whey as more satiating than other protein sources (16) . However, as HP+ and HP- had similar levels of whey protein, it is difficult to attribute the difference in effects on appetite to small differences in the type of protein. A more consistent finding in the literature is that preloads enriched with carbohydrate are less satiating than are 358 energy-matched protein preloads^{$(2, 4, 23, 32, 33)$}. Thus the prediction, based on nutrient composition would be that the HC+ preload would have been less satiating than the HP+ preload. The finding that altering the thickness and creamy flavour of the HC+ preload to make it more similar to the HP+ preload resulted in similar satiety responses to the two drinks implies that may be sensory rather than macronutrient differences which are critical in determining different short-term satiety responses between carbohydrate and protein-enriched beverages. This finding fits well with a recent study in our laboratory that also found that making drinks thicker in texture and creamier in flavour enhanced the degree to which added protein was 366 satiating⁽²¹⁾. In relation to the present study, the HC+ drink was more satiating than was a similar carbohydrate drink without added thickness or creaminess in an earlier study⁽²³⁾. It

 would have been useful to have included this HC- (the high carbohydrate without added sensory quality) in the present study. However, conditions equivalent to the HC+/HC- contrasts 370 were included in our recent study⁽²¹⁾, and again altering thickness and creamy flavour enhanced satiety.

 How then might altering the thickness and creaminess of a drink enhance the satiating efficiency of ingested nutrients? In line with recent ideas about sensory-nutrient interactions in satiety(34), we hypothesised that products with higher protein content, particularly in a dairy context, have some sensory characteristics in common, including both a thicker texture and creamy flavour. Past experience of both these sensory characteristics and consequent effects of ingestion on appetite of such products should lead to an expectation that drinks with these sensory characteristics would be more filling, so facilitating the consumer to respond to actual nutrient ingestion. Several lines of evidence support this suggestion. Firstly, differences in the profile of release of satiety hormones have been shown between protein and carbohydrate 382 preloads^{$(16, 35)$}. Many of these studies do not report the sensory analysis of the preloads, but it is likely that subtle sensory differences would have existed. It is established that orosensory cues 384 can solicit release of hormones related to appetite control^(36, 37) probably as part of learned 385 preparatory responses which prepare the body to process nutrients^{(38)}. Thus subtle sensory differences between beverages such in thickness and creaminess could modify post-ingestive processing of nutrients by facilitating anticipatory hormone release. Sensory cues also generate 388 explicit expectations about how satiating foods will be⁽³⁹⁾, and recent data from our laboratory confirm that the subtle differences in sensory characteristics between preloads in the present study would have resulted in explicit expectations of satiety(40). This interpretation of the differences in response to the three high energy preloads in the present study relies on subtle sensory differences between stimuli. The analysis of participants' evaluations of the drinks during testing suggest which of these sensory features were most important, but it is possible

 that preloads varied on other dimensions that were not captured by the evaluations used here. HP+ and HP- preloads differed significantly in rated thickness only, with non-significant trends for greater creaminess, fattiness and dairy-like qualities. Although there was a trend for higher creaminess in both HP+ and HC+ conditions relative to HP-, all of these were rated as creamier than was the control. Differences between high energy conditions were less clear in the main study than in the pilot studies, possibly due to contrast effects making this more evident when products were rated alongside each other in the absence of the LE condition, an effect we have 401 seen in other studies⁽²¹⁾, and which fits with more general contrast effects in sensory 402 evaluation⁽⁴¹⁾. Importantly HC+ and HP+ appeared well matched in terms of thickness and creaminess, with only a trend for HC+ having less dairy-like qualities than HP+. The finding that perceived thickness was important fits with other studies that suggest this characteristic is 405 an important orosensory satiety cue^{$(42-44)$}. Studies also suggest viscosity is an important component of the satiating efficiency of beverages, with greater satiety from more viscous drinks⁽⁴⁵⁻⁴⁸⁾, and texture appearing to be more important than flavour in determining satiation in 408 a dairy-context⁽⁴⁹⁾. The current literature implies that textural differences, probably viscosity, may be the most likely explanation for why HC+ was more satiating here than would be expected based on nutrient content alone and why HP- was less satiating than HP+.

 An alternative explanation for differences between preloads, however, could be the small differences in soluble fibre content generated by the use of guar gum as thickening agent. Increased viscosity generated by the addition of insoluble fibres has been shown to enhance 415 satiety^(50, 51), increase release of satiety-related gastric hormones⁽⁵²⁾, and modify gastric 416 emptying^{(53)}. In all of these studies differences in post-ingestive effects of fibre were confounded by likely differences in sensory characteristics through changed viscosity, and the present literature does not allow easy separation of orosensory and post-ingestive effects. However, it has been suggested that the dilution effects of small amounts of added fibre on

420 viscosity in the stomach make orosensory explanations more likely⁽⁵⁴⁾. Most studies exploring effects of fibre use much greater quantities than was used to subtly thicken HP+ and HC+: for 422 example 12g of guar gum was added to explore effects on gastric emptying^{(53)}, and enhanced 423 satiety was reported after addition of 12g of inulin in a protein-rich beverage^{(55)}, compared with 1.2g guar gum used here. No study that we aware of has demonstrated enhanced satiety or physiological response to such small quantities, however the only way to truly isolate sensory versus post-ingestive effects would be to contrast the same preloads when infused into the stomach relative to see whether the apparent sensory/nutrient interactions suggested here persist in the absence of orosensory cues. However, past research suggests that orosensory cues are necessary for the full expression of satiety, with reduced satiety when the same foods are 430 infused into the stomach or intestine than when ingested⁽¹⁹⁾, and although a nutrient effect of the added guar gum or very small differences in fat content between preload cannot be excluded, such explanations are less plausible than would be effects through sensory-nutrient interactions.

 In this study there was a relatively short delay between beverage consumption and the test meal (minimum of 20 minutes), and this may have exaggerated the effects of sensory quality and reduced the impact of post-ingestive satiety cues. However, the delay we used was chosen since an earlier study found no difference in effect of protein preloads between 30 minute and 438 120 minute delays⁽²³⁾, and other preload studies suggest that short delays are most effective⁽²⁸⁾. However, it may be that some participants treated the drink as a course of the test meal implying the responses were more related to satiation than satiety.

 We did find a decrease in the rated pleasantness of the preload after ingestion in both protein conditions, but not the HC+ or control conditions. This finding is consistent with previous research suggesting that protein foods produce greater sensory-specific satiety (SSS) than do 445 other macronutrients⁽⁵⁶⁾, although SSS effects did not emerge in previous experiments in our

446 . laboratory^{$(1, 23)$}. This difference between protein and non-protein preloads cannot readily explain the differences in intake and appetite at the test lunch since intake and appetite after HC+ and HP+ preloads was similar, and significantly different from that after HP-.

 Overall the critical finding in the present study was that matching high protein and carbohydrate preloads in terms of perceived thickness and creaminess resulted in very similar satiety responses to these drinks, whereas normally protein has been found to be more satiating than carbohydrate. In contrast, there were significant differences in satiety following consumption of protein preloads that were matched in nutritional content but which differed in thickness and creaminess, with the less thick and creamy version (HP-) less satiating. These findings have implications both for the future conduct of human preload studies, where greater care is needed to match stimuli at a sensory level, and in terms of our understanding of the nature of satiety. In particular differences in the satiating effects of different types of foods, such as liquid versus solid etc, may be in part attributed to the role of sensory cues in facilitating post-ingestive satiety.

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- conducted the primary analyses and drafted the initial methods and results for this MS. AL
- provided advice on study design, technical support for the formulation of the test drink preloads
- and provided some of the test ingredients. MY supervised the project, and had primary
- responsibility for production of the final MS and the analysis of rating data.

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620 Table 1. Final nutritional composition of the four test preloads.

622 Table 2. Mean (±SE) changes in hunger, fullness, thirst and nausea immediately and 30

623 minutes after consuming the four test preload drinks.

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- 625

626

627 In each row, data marked with different superscripts differ significantly (p<0.05 or less using Bonferroni protected

628 contrasts).

630 Table 3. Mean (±SEM) sensory and hedonic evaluations of the preloads at the initial taste test.

Preload condition

631

632 For ratings which differed between conditions (thick, creamy and dairy), data marked with different superscripts

633 differ significantly (p<0.05 or less using Bonferroni protected contrasts).

Figure legend

- Figure 1. Test food intake at lunch (panel A) and total energy consumed in the laboratory
- tests (panel B) in the four preload conditions: LE (low energy), HP- (low sensory protein),
- HC+ (high sensory carbohydrate) and HP+ (high sensory protein). All data are mean ±SEM,
- n=26. Letters above each bar indicate significance: within each panel, bars with different letters
- 640 are significantly different (p <0.05 or higher).

- Figure 2. Rated pleasantness of the four test drinks before (Start) and after (End) they had
- been consumed: LE (low energy), HP- (low sensory protein), HC+ (high sensory carbohydrate)
- 644 and HP+ (high sensory protein). All data are mean \pm SEM, n=26. ** denotes significant change
- 645 between start and end ratings, $p<0.01$

Preload condition