Final report – August 2010

Grant number 31/06 The use of a biological marker, 5-HT2C genotype, as a predictor of motivation, adherence and weight loss in participants of an obesity intervention programme.

Project summary

The project started in April 2007 and was completed in April 2010. The budget was 49,000 euros, which was originally estimated to provide £30,000 British pounds, however, due to the advantageous exchange rate the project budget provided £45,000 British pounds. This significant increase in funding allowed the expansion of the aims of the project and the genotyping aspect of the study was extended to include the study of additional genotypes: 5-HT1A, 5-HT2A, (serotonin receptors), DRD2 and DRD4 (dopamine receptors). A questionnaire measuring eating behaviours was also added to the study (Three-factor eating questionnaire) and an additional questionnaire measuring motivation in relation to exercise (BREQ-2) was added at the end of the intervention, as well as the start of the intervention to allow the measurement of changes in motivation to exercise. The original project proposal included carrying out a Motivational Interview (MI) with each participant, however this plan was modified to allow the study of the efficacy of Motivational Interviewing as part of a diet and exercise intervention, therefore the study group was divided into 2 groups with half of the participants receiving the normal intervention (questionnaires, baseline measurements and buccal swab only) and the other half receiving a motivational interview in addition to the normal intervention.

Original aims of the project

To determine whether there is a correlation between 5-HT2C genotype and extent of motivation to make behavioural changes, prior to a diet and exercise intervention programme.

To determine whether 5-HT2C genotype predicts success in:
- adherence to a diet and exercise intervention programme
- actual weight loss

To determine whether extent of motivation prior to the study correlates with success in:
- adherence to a diet and exercise intervention programme
- actual weight loss

Extended aims of the project

To determine whether there is a correlation between 5-HT1A, 5HT2A, 5-HT2C, DRD2 or DRD4 genotype and extent of motivation to make behavioural changes, prior to a diet and exercise intervention programme.

To determine whether there is a correlation between 5-HT1A, 5HT2A, 5-HT2C, DRD2 or DRD4 genotype and changes in motivation to make behavioural changes after participating in a diet and exercise intervention programme.

To determine whether 5-HT1A, 5HT2A, 5-HT2C, DRD2 or DRD4 genotype predicts success in:
- adherence to a diet and exercise intervention programme
- actual weight loss

To determine whether extent of motivation prior to the study correlates with success in:
adherence to a diet and exercise intervention programme
actual weight loss

To determine whether there is a correlation between 5-HT1A, 5HT2A, 5-HT2C, DRD2 or DRD4 genotype and eating behaviours measured by the Three-factor eating questionnaire.

To determine whether eating behaviours measured by the Three-factor eating questionnaire correlates with success in:
actual weight loss

To determine the efficacy of Motivational interviewing as part of a diet and exercise intervention programme by determining the influence of MI on:
adherence to the diet and exercise intervention programme
actual weight loss

Methodology

100 obese participants of a community-based exercise and diet intervention programme took part in the study. At the start of the programme the participants were measured to calculate their Body Mass Index (BMI) and a buccal swab was taken for genotyping. They filled in two questionnaires, one to measure motivation related to exercise, the Behavioural Regulation in Exercise Questionnaire (BREQ-2) and the other to measure eating behaviours the Three-Factor Eating questionnaire (TFEQ). The BREQ-2 questionnaire gives a Relative Autonomy Index (RAI) value which is a measure of motivation to exercise. The TFEQ questionnaire measures Restraint, Hunger and Disinhibition eating behaviours. Adherence to the programme was measured by whether the participants completed the programme or dropped out before the end of the study. DNA extracted from the buccal swabs was genotyped for polymorphisms in 5-HT1A, 5HT2A, 5-HT2C, DRD2 and DRD4 genes using Taqman assays (5-HT1A, 5HT2A and 5-HT2C) and PCR followed by restriction enzyme digest (DRD2 and DRD4). The following polymorphisms were investigated:

5-HT1A - 1019 G/C
5HT2A -1438 G/A
5-HT2C -759 T/C
DRD2 – TaqI A1/A2 allele
DRD4 – +/- seven repeat allele

48 participants received a Motivational Interview (MI) intervention and 52 received the normal intervention only. The MI consisted of a one-to-one session with a trained counsellor to discuss behavioural change, readiness to change and barriers to change.

At the end of the programme the participants were contacted and their BMI measurements taken and they repeated the BREQ-2 questionnaire, this was usually between 12-16 weeks after the start of the programme.

Results

	Male	Female
Number of participants	30	70
Average age	50.3 +/- 13.9	46.9 +/- 13.6
Average BMI at the start of the study	39.4 +/- 7.1	39.7 +/- 8.8
Number of participants who withdrew (%)	4 (13.3)	24 (34.3)
Participants who received a Motivational Interview	13	35
Average change in BMI measured at the end of the study	0.72 +/- 1.73	0.46 +/- 1.33
Average RAI measured at the start of the study using BREQ-2	21.0 +/- 23.0	30.6 +/- 27.6
Average change in RAI measured at the end of the study	5 +/- 13.6	4.7 +/- 18.8
TFEQ restraint measured at the start of the study	8.2 +/- 4.3	9.7 +/- 3.5
TFEQ disinhibition measured at the start of the study	7.7 +/- 3.7	8.2 +/- 3.8
TFEQ hunger measured at the start of the study	5.6 +/- 3.9	4.3 +/- 2.9

Table 1 The data collected during the study from the participants. 28 participants withdrew from the study before the end of the programme; most of the participants who withdrew were female. The most common reason stated for withdrawing from the study was that the participant was too busy to carry on with the programme. On average both males and females reduced their BMI, the reduction was greater for males than females. Females scored higher than males on the Relative Autonomy Index measured by the BREQ-2 questionnaire, this index increased for both males and females by the end of the programme indicating that the intervention increased the participants' motivation to participate in exercise. The increase was similar for males and females. The Three-Factor Eating Questionnaire scores for restraint and disinhibition were higher in females than males, however the score for hunger was higher in men.

Gene	Genotype	Frequency %
HT1A	CC	21
	GC	55.6
	GG	23.4
HT2A	AA	18.2
	GA	40.4
	GG	41.4
HT2C *	TT/T	3.8
	TC	12.4
	CC/C	83.8
DRD2	A2/A2	66.7
	A1/A2	27.1
	A1/A1	6.2
DRD4	<7R/<7R	67.5
	7R/<7R	28.8
	7R/7R	3.8
DRD2/DRD4	0 or 1 risk alleles	83.8
	+ 2 risk alleles	16.2

Table 2 The genotype frequencies observed in the study. * HT2C is on the X chromosome therefore in males there are only 2 genotypes, T or C. The frequencies for all the genes are similar to the frequencies observed in previous studies. As the frequencies for *DRD2* A1 allele and *DRD4* 7-repeat allele were uncommon these genes were also considered together and expressed as +2 risk alleles. Participants were considered to have 2+ risk alleles if they were homozygous for either gene or were heterozygous for both genes.

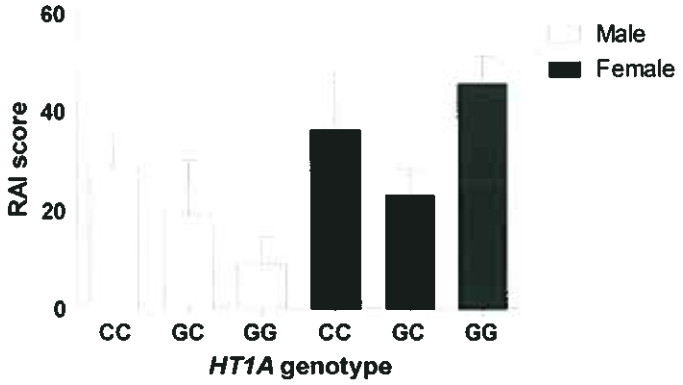


Figure 1a The relationship between the Relative Autonomy Index scores measured using the BREQ-2 questionnaire at the beginning of the study and *HT1A* genotype. There was no significant difference observed between the different genotypes.

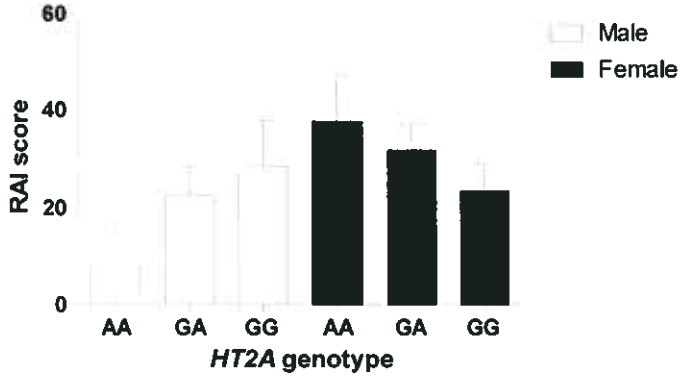


Figure 1b The relationship between the Relative Autonomy Index scores measured using the BREQ-2 questionnaire at the beginning of the study and *HT2A* genotype. There was no significant difference observed between the different genotypes.

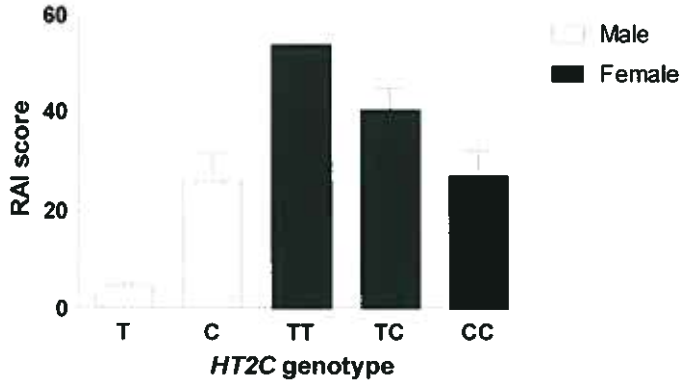


Figure 1c The relationship between the Relative Autonomy Index scores measured using the BREQ-2 questionnaire at the beginning of the study and *HT2C* genotype. There was no significant difference observed between the different genotypes.

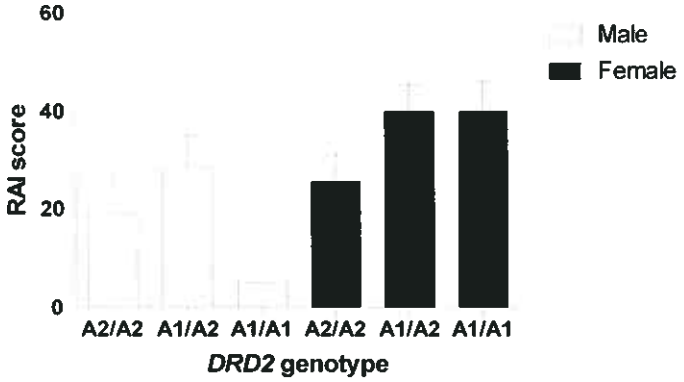


Figure 1d The relationship between the Relative Autonomy Index scores measured using the BREQ-2 questionnaire at the beginning of the study and *DRD2* genotype. There was no significant difference observed between the different genotypes.

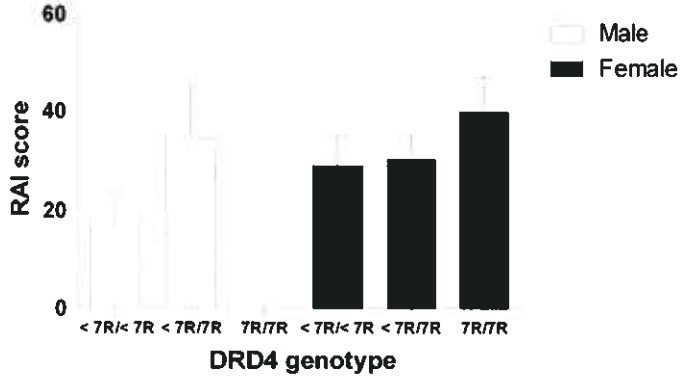


Figure 1e The relationship between the Relative Autonomy Index scores measured using the BREQ-2 questionnaire at the beginning of the study and *DRD4* genotype. There was no significant difference observed between the different genotypes. Note there were no male participants with the 7R/7R genotype.

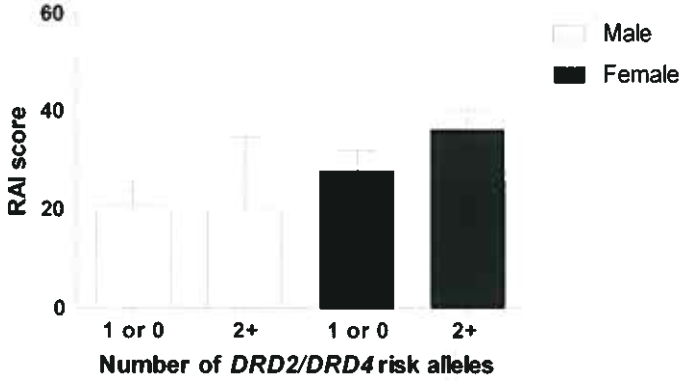


Figure 1f The relationship between the Relative Autonomy Index scores measured using the BREQ-2 questionnaire at the beginning of the study and *DRD2*/*DRD4* composite genotype. There was no significant difference observed between the different genotypes.

Figures 1a-1f The relationships between the Relative Autonomy Index scores measured using the BREQ-2 questionnaire at the beginning of the study and the genotypes studied. The Relative Autonomy Index is a measure of motivation to take part in exercise. There was no significant difference observed between the different genotypes, the lack of significance is likely to be due to the high variability observed in the RAI scores of participants within the groups.

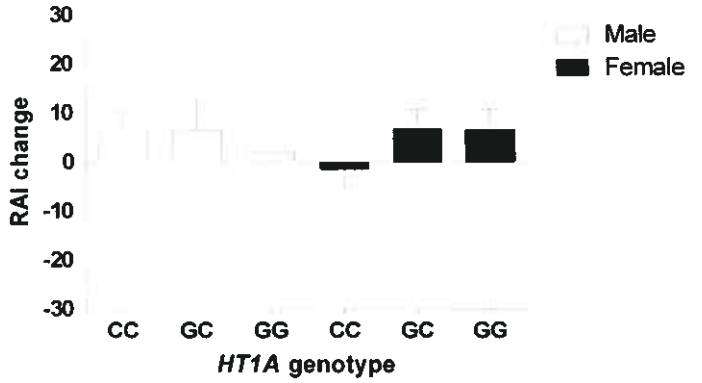


Figure 2a The relationship between the change in Relative Autonomy Index scores over the course of the study and *HT1A* genotype. There was no significant difference observed between the different genotypes.

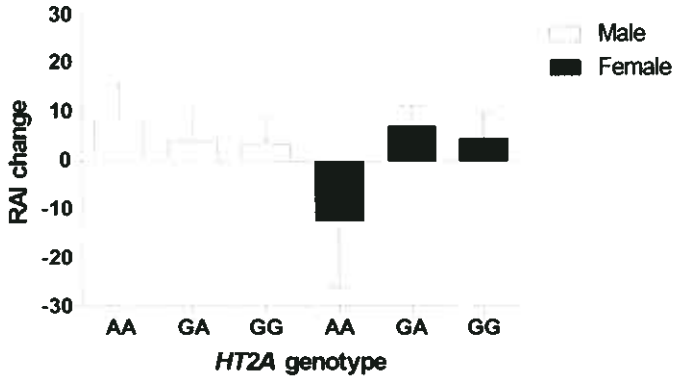


Figure 2b The relationship between the change in Relative Autonomy Index scores over the course of the study and *HT2A* genotype. There was no significant difference observed between the different genotypes.

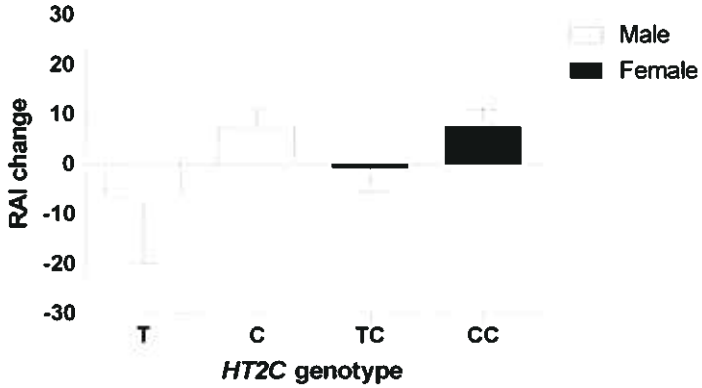


Figure 2c The relationship between the change in Relative Autonomy Index scores over the course of the study and *HT2C* genotype. There was no significant difference observed between the different genotypes.

Figure 2d The relationship between the change in Relative Autonomy Index scores over the course of the study and *DRD2* genotype. There was no significant difference observed between the different genotypes. Note there were no male participants with the A1/A1 genotype who completed the study.

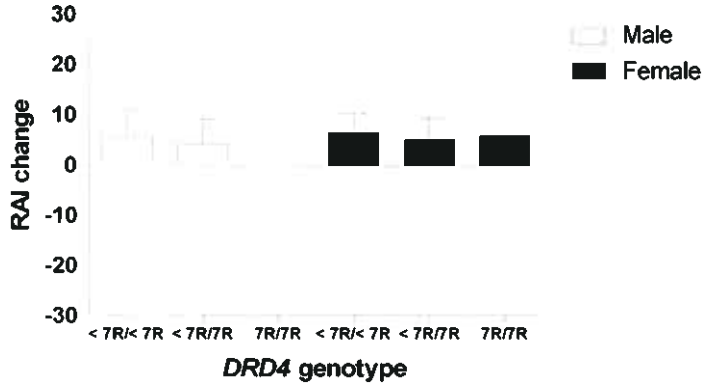


Figure 2e The relationship between the change in Relative Autonomy Index scores over the course of the study and *DRD4* genotype. There was no significant difference observed between the different genotypes. Note there were no male participants with the 7R/7R genotype.

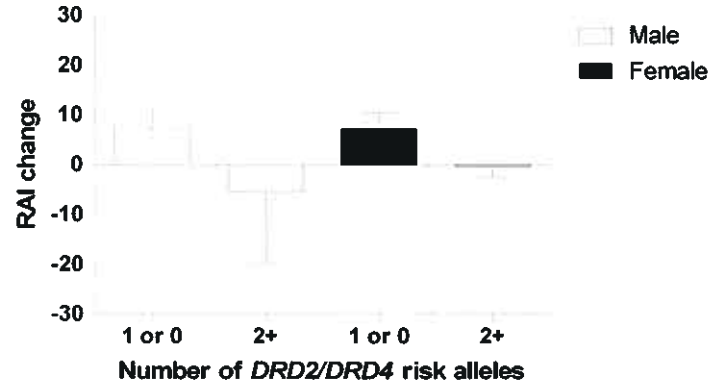


Figure 2f The relationship between the change in Relative Autonomy Index scores over the course of the study and *DRD2/DRD4* composite genotype . There was no significant difference observed between the different genotypes.

Figures 2a-2f The relationships between the Relative Autonomy Index (RAI) scores measured over the course of the study and the genotypes studied. The RAI is a measure of motivation to take part in exercise. There was a trend indicating that participants with the T allele of the *HT2C* gene or 2+ risk alleles for the *DRD2/DRD4* composite genotype had negative changes in RAI over the course of the study, but these results were not significant due to the wide variability of the RAI scores and the small number of participants with *DRD2* or *DRD4* risk alleles who completed the programme. Participants with these risk alleles were significantly more likely to drop out of the programme (Table 3), making the numbers in these groups even smaller. However, if this trend is a true effect this would indicate that people with these genotypes find a diet and exercise intervention demotivating and that they are less motivated at the end of the intervention. This has implications for developing personalised interventions which address the needs of this group of people.

Gene	Genotype	Completed programme (% total subjects)	Dropped out of programme (% total subjects)
HT1A	CC	64.7	35.3
	GC	86.7	13.3
	GG	73.7	26.3
HT2A	AA	66.7	33.3
	GA	80	20
	GG	71.8	28.2
HT2C	TT/T	66.7	33.3
	TC	70	30
	CC/C	80.1	19.4
DRD2	A2/A2	81.5	18.5
	A1/A2	77.3	22.7
	A1/A1	60	40
DRD4	<7R/<7R	77.4	22.6
	7R/<7R	82.6	17.4
	7R/7R	66.7	33.3
DRD2/DRD4	0 or 1 risk alleles	83.6	16.4
	+ 2 risk alleles	50	50 * $p = 0.016$

Table 3 The level of adherence to the study grouped by genotype. Participants who completed the study were considered to have adhered to the study, participants who dropped out of the study were considered at non-adherent. The *DRD2/DRD4* 2+ risk alleles composite genotype correlated significantly with a lack of adherence (Chi-squared test, p=0.016). Participants with this composite genotype had a 50% chance of dropping out of the study compared to an average drop out rate for the other genotypes of 16.4. The *DRD2/DRD4* TaqIA and 7-repeat genotypes are associated with blunted responses to dopamine; these data indicate that dopamine receptor genotype may influence the participants motivation to engage with obesity intervention programmes.

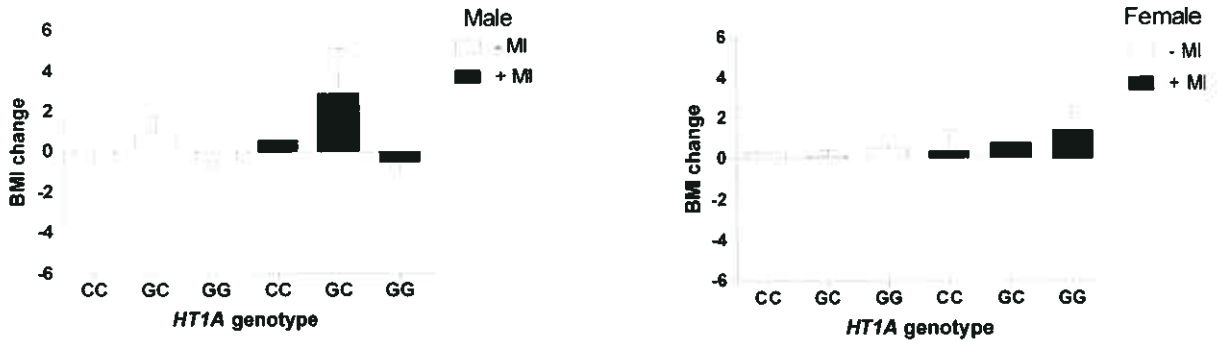


Figure 3a i and 3a ii The relationship between BMI change and HT1A genotype divided into participants who received a motivational interview (shaded columns) and participants who did not receive a motivational interview (open columns) and male (i) and female (ii). There was no significant difference observed between the genotypes.

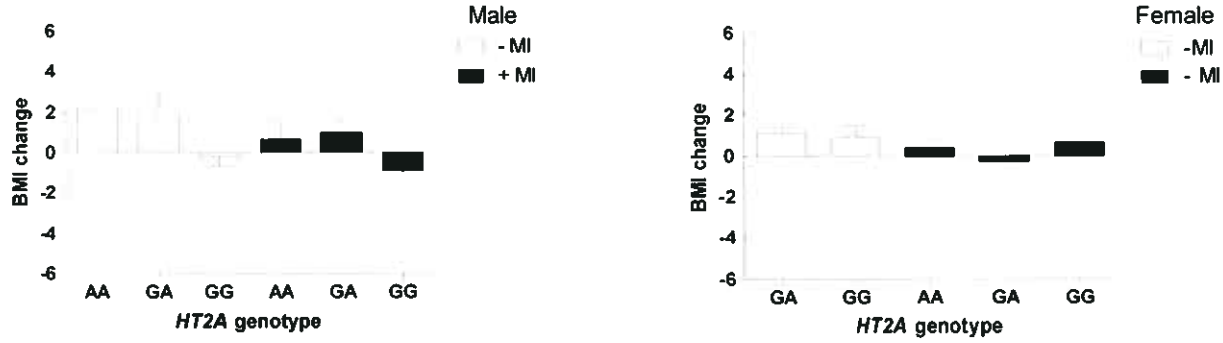


Figure 3b i and 3b ii The relationship between BMI change and *HT2A* genotype divided into participants who received a motivational interview (shaded columns) and participants who did not receive a motivational interview (open columns) and male (i) and female (ii). Female participants with the GA genotype who did not have a motivational interview had a significantly greater drop in their BMI compared to participants who had a motivational interview (p=0.04). Note there were no female participants with the AA genotype who received an MI and completed the study.

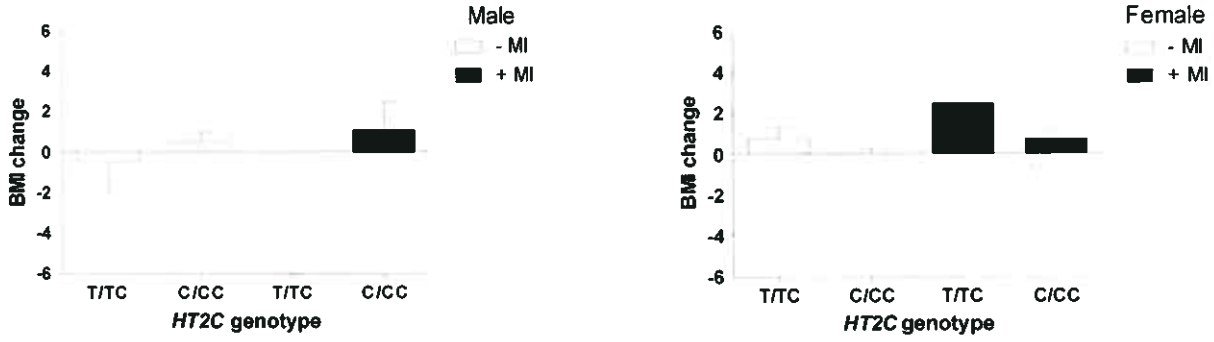


Figure 3c i and 3c ii The relationship between BMI change and *HT2C* genotype divided into participants who received a motivational interview (shaded columns) and participants who did not receive a motivational interview (open columns) and male (i) and female (ii). There was no significant difference observed between the genotypes. There were no male participants who had the T/TC genotype, did not receive an MI and completed the study.

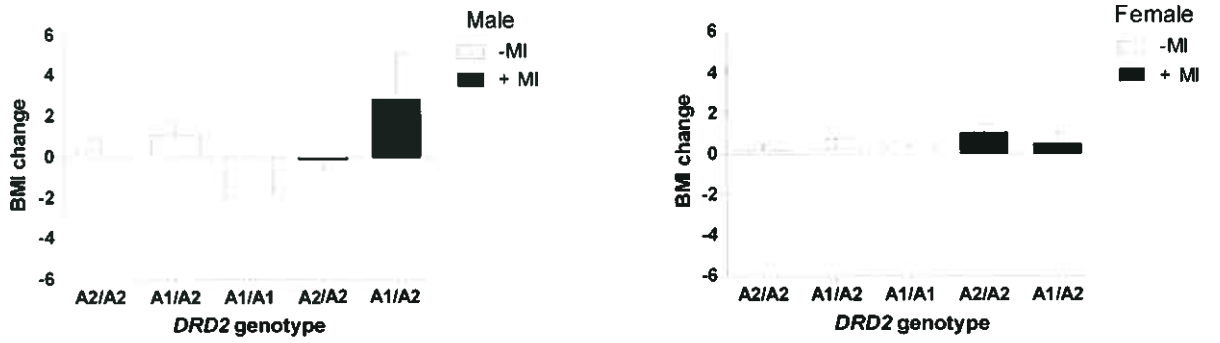


Figure 3d i and 3d ii The relationship between BMI change and *DRD2* genotype divided into participants who received a motivational interview (shaded columns) and participants who did not receive a motivational interview (open columns) and male (i) and female (ii). There was no significant difference observed between the genotypes. Note there were no male or female participants with the A1/A1 genotype who received a motivational interview who completed the study.

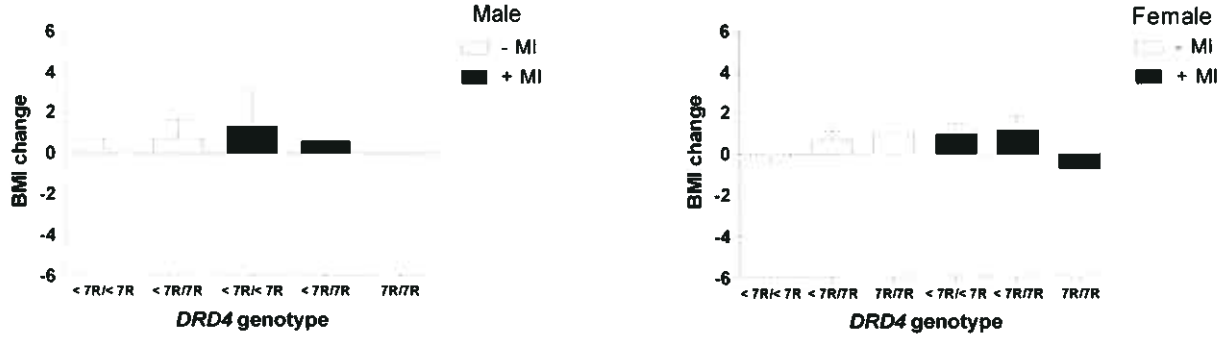


Figure 3e i and 3e ii The relationship between BMI change and *DRD4* genotype divided into participants who received a motivational interview (shaded columns) and participants who did not receive a motivational interview (open columns) and male (i) and female (ii). There was no significant difference observed between the genotypes. Note there were no male participants with the 7R/7R genotype who received an MI who completed the study.

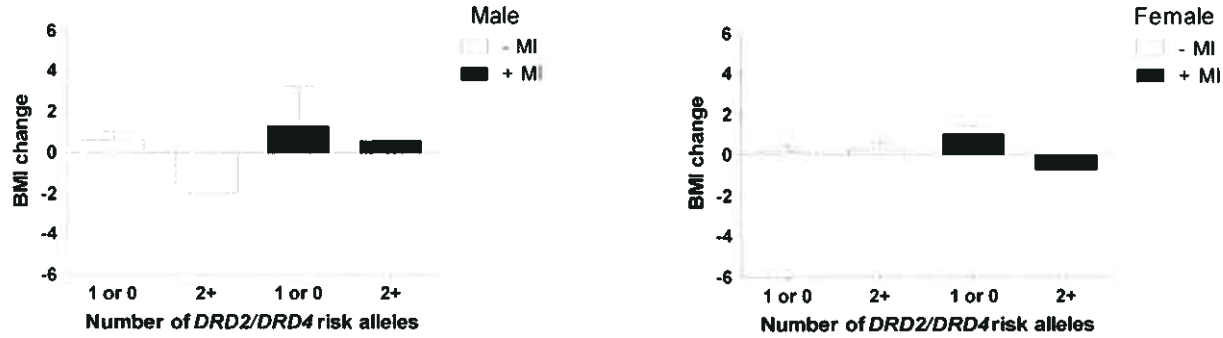


Figure 3f i and 3f ii The relationship between BMI change and *DRD2/DRD4* composite genotype divided into participants who received a motivational interview (shaded columns) and participants who did not receive a motivational interview (open columns) and male (i) and female (ii). There was no significant difference observed between the genotypes.

Figures 3a-3f The relationship between BMI change and the genotypes investigated. There was no significant effect of genotype on BMI change except for females with the GA genotype who had a significantly greater BMI loss when they did not receive a motivational interview. Although this lack of significance is disappointing it is partly due to the high drop out rate (28%). This meant that for some genotypes there were only a few participants in the group. The effect on the *DRD2* and *DRD4* genotypes was particularly pronounced; the TaqIA and 7-repeat alleles had a significant influence on whether participants dropped out of the study (Table 3). In addition these alleles were comparatively rare in the population (Table 2).

	Male		Female	
	Completed programme	Dropped out of programme	Completed programme	Dropped out of programme
RAI scores at start of study	23.1 +/- 23.6	6.0 +/- 12.1	29.7 +/- 26.6	32.6 +/- 30.7

Table 4 The Relative Autonomy Index (RAI) scores recorded at the beginning of the study and the level of adherence to the programme. The trend indicated that for male participants a low RAI score was associated with an increased risk of dropping out of the study, this effect was not observed in females, however the results was not significant due to the wide variability in the RAI scores.

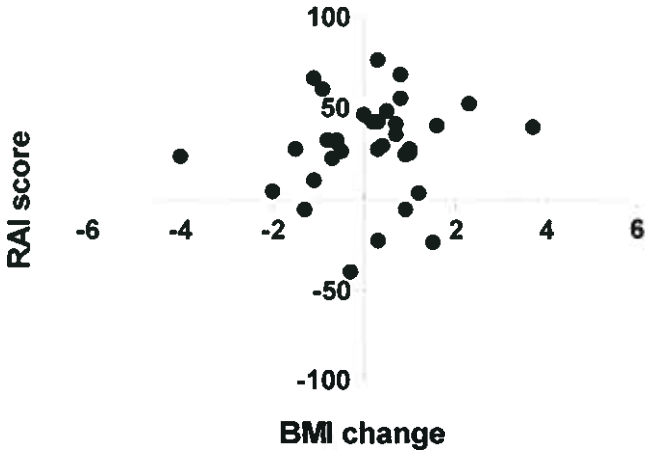


Figure 4a The relationship between the Relative Autonomy Index scores measured at the start of the study and the subsequent Body Mass Index change achieved by the participants who did not have a Motivational Interview. . The r value was 0.116 which did not indicate a significant correlation.

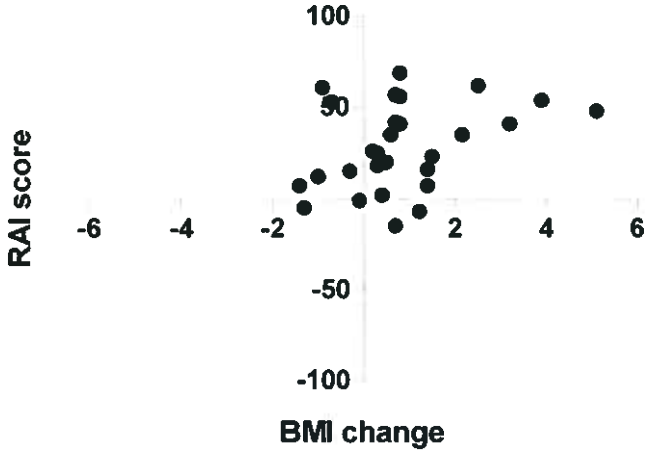


Figure 4b The relationship between the Relative Autonomy Index scores measured at the start of the study and the subsequent Body Mass Index change achieved by the participants who had a Motivational Interview. . The r value was 0.378 which indicated a significant correlation (p=0.006).

Figures 4a and 4b The relationship between Relative Autonomy Index (RAI) scores measured at the beginning of the programme and BMI change. There was a significant relationship between RAI and BMI change in the group who received a motivational interview (p=0.006), but in the group which did not receive a motivational interview there was no significant relationship between the measures. One explanation for this observation may be that the motivational interview includes discussion of a person's readiness to change their behaviour. The RAI index measures motivation to exercise and the motivational interview which follows the questionnaire may help clarify the issues involved and the barriers to change for the participant. This may mean that the outcome (BMI change) is more closely related to the motivation to change measured by the RAI in participants who received a motivational interview, whereas the participants who did not receive a motivational interview did not consider their motivation in the same level of detail.

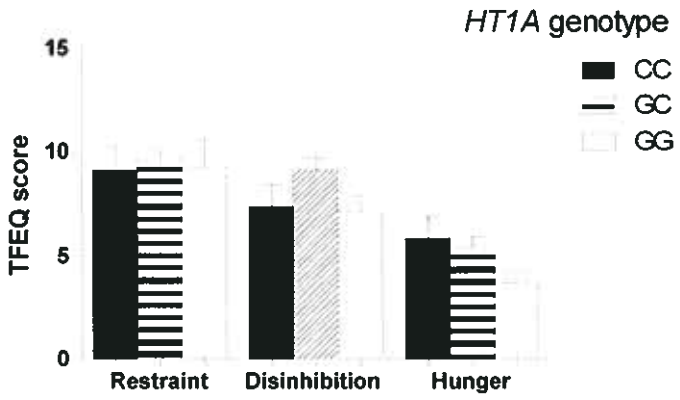


Figure 5a The relationship between the scores for restraint, disinhibition and hunger measured using the Three Factor Eating questionnaire (TFEQ) and HT1A genotype. Participants with the GG genotype had significantly lower scores for hunger than those with the CC or GC genotypes (p=0.04). There were no significant differences between the genotypes for the scores for restraint or disinhibition.

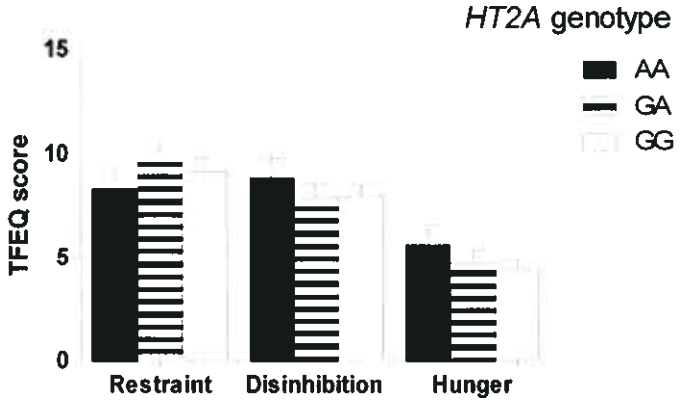


Figure 5b The relationship between the scores for restraint, disinhibition and hunger measured using the Three Factor Eating questionnaire (TFEQ) and *HT2A* genotype. There were no significant differences between the genotypes for the scores for restraint, disinhibition or hunger.

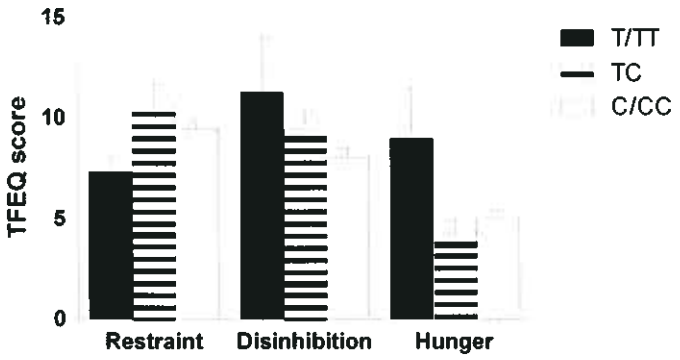


Figure 5c The relationship between the scores for restraint, disinhibition and hunger measured using the Three Factor Eating questionnaire (TFEQ) and *HT2C* genotype. Participants with the TC or C/CC genotypes had significantly lower scores for hunger than those with the T/TT genotype (p=0.03).There were no significant differences between the genotypes for the scores for restraint or disinhibition.

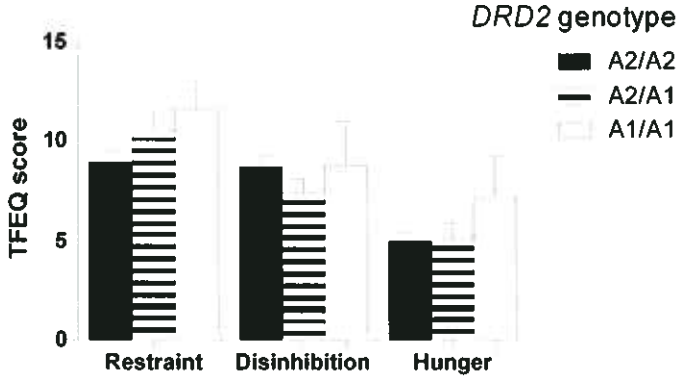


Figure 5d The relationship between the scores for restraint, disinhibition and hunger measured using the Three Factor Eating questionnaire (TFEQ) and *DRD2* genotype. There were no significant differences between the genotypes for the scores for restraint, disinhibition or hunger.

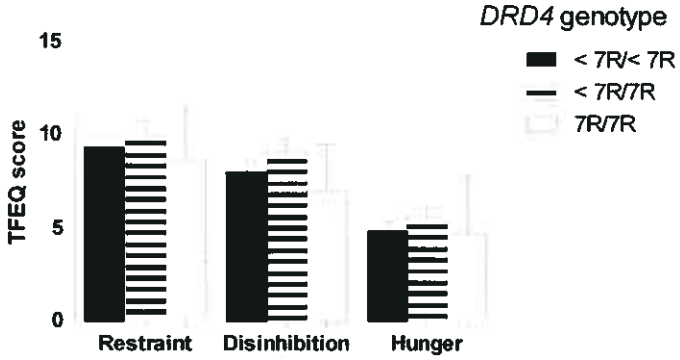


Figure 5e The relationship between the scores for restraint, disinhibition and hunger measured using the Three Factor Eating questionnaire (TFEQ) and *DRD4* genotype. There were no significant differences between the genotypes for the scores for restraint, disinhibition or hunger.

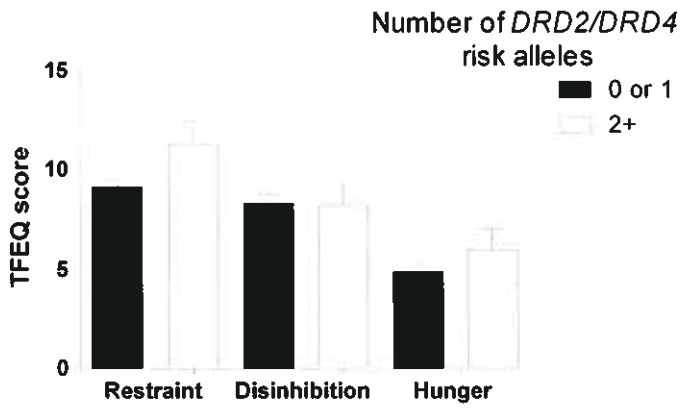
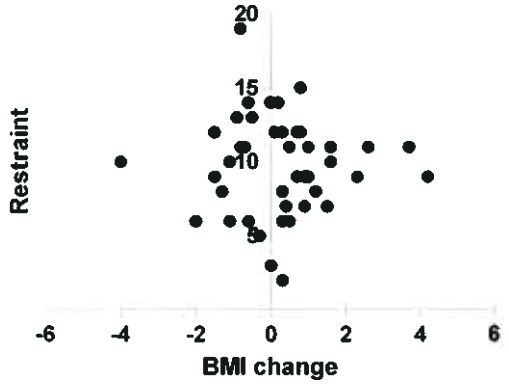
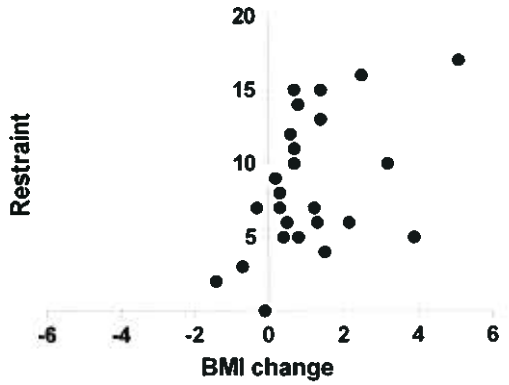


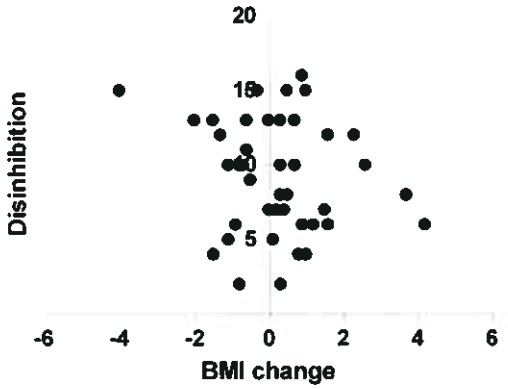
Figure 5f The relationship between the scores for restraint, disinhibition and hunger measured using the Three Factor Eating questionnaire (TFEQ) and *DRD2/DRD4* composite genotype. There were no significant differences between the genotypes for the scores for restraint, disinhibition or hunger when all participants were considered together, however when females were considered alone scores for restraint for the 2+ genotypes were significantly higher than the scores for the 0 or 1 genotypes (p=0.05).

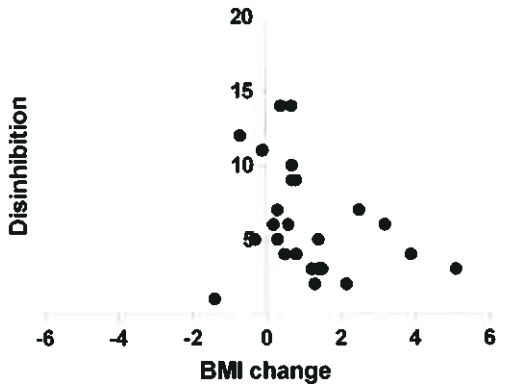
Figures 5a-5f The relationship between the scores for restraint, disinhibition and hunger measured using the Three Factor Eating questionnaire (TFEQ) and genotype. There were significant relationships between *HT1A* genotype and hunger, *HT2C* genotype and hunger and *DRD2/DRD4* composite genotype and restraint (in female participants only). *HT1A* and *HT2C* code for serotonin receptors and it is known that serotonin is involved in the control of appetite, these data confirm that serotonin receptors are involved in mediating the sensation of hunger. The risk alleles TaqIA and 7-repeat of the *DRD2* and *DRD4* dopamine receptors have been associated with impulsive behaviour; high scores for restraint in the TFEQ questionnaire are associated with binge-eating, considered an impulsive behaviour. These data confirm that the association between dopamine receptor risk alleles and impulsive behaviour also applies to eating behaviours.



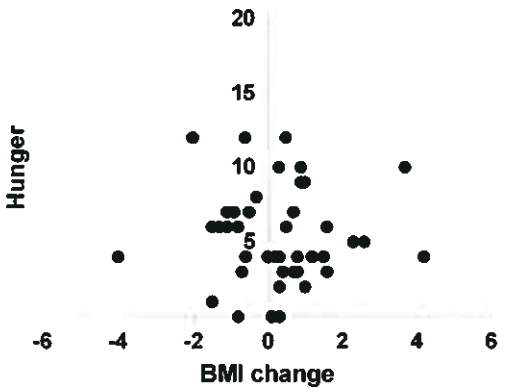


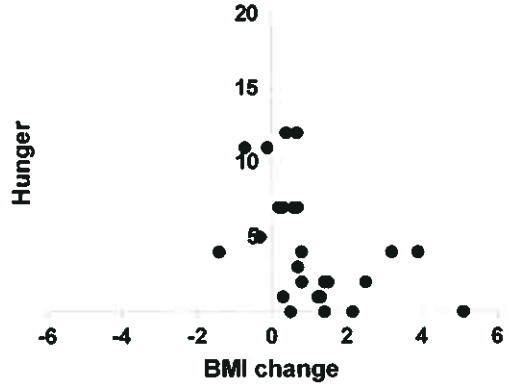
Figures 6a (left) and 6b (right) Correlation between score for restraint measured using the TFEQ and BMI change over the study. Participants who did not have a motivational interview (MI) are shown in Figure 6a, participants who had a motivational interview are shown in Figure 6b. There was a significant relationship between restraint score and BMI change in participants who had a motivational interview (r=0.463, p=0.02), however in participants who did not receive a motivational interview there was no correlation (r=-0.015).





Figures 6c (left) and 6d (right) Correlation between score for disinhibition measured using the TFEQ and BMI change over the study. Participants who did not have a motivational interview (MI) are shown in Figure 6c, participants who had a motivational interview are shown in Figure 6d. There was no significant relationship between disinhibition score and BMI change either group (r= -0.296, non-MI group, r= -0.188, MI group).





Figures 6e (left) and 6f (right) Correlation between score for hunger measured using the TFEQ and BMI change over the study. Participants who did not have a motivational interview (MI) are shown in Figure 6e, participants who had a motivational interview are shown in Figure 6f. There was a significant inverse relationship between hunger score and BMI change in participants who had a motivational interview (r= -0.447, p=0.03), however in participants who did not receive a motivational interview there was no correlation (r= 0.006).

Figures 6a-6e Relationships between scores for restraint, disinhibition and hunger and BMI change over the study. Both restraint and hunger showed a significant relationship with BMI change, but only for the groups receiving the motivational interview, in the case of hunger the relationship was an inverse correlation. These results reflect the correlation between RAI score and BMI change which was also only present in the motivational interview group. As with the RAI data this may indicate that the motivational interview helps the participant to clarify their feelings about diet and exercise. This leads to a greater relationship between the eating behaviour scores and BMI change. In the case of restraint it appears that the ability to restrain from eating correlates with greater BMI loss. In the case of hunger, lower reported sensations of hunger correlate with greater BMI change.

	All participants		Male		Female	
	+ MI	- MI	+ MI	- MI	+ MI	- MI
BMI change	0.99 +/- 1.41	0.25 +/- 1.47	1.19 +/- 1.76	0.4 +/- 1.70	0.17 +/- 1.36	0.88 +/- 1.21
% drop out	37.5	19.2	15.4	11.8	45.7	22.9

Table 5 Participants who received a motivational interview (MI) had a significantly larger drop in BMI compared to participants who did not receive a motivational interview (p=0.04). Twice as many participants dropped out from the MI group compared to the non-MI group. As part of the motivational interview process the participant is encouraged to discuss their readiness to make behavioural changes. It may be that the participants in the MI group were more realistic about whether they had the commitment to make changes to their lifestyle. Those that decided they were not ready dropped out, but those that continued were significantly more successful at reducing their BMI than those participants who did not receive an MI. This also indicates that Motivational Interviewing may be a useful addition to standard diet and exercise interventions.

Significant outcomes, discussion and conclusions.

The overall aim of this study was to investigate the influence of serotonin and dopamine receptor gene polymorphisms on various parameters relevant to the successful participation of obese people taking part in a diet and exercise obesity intervention. The participants' motivation to take part in exercise was measured at the beginning and end of the study. Eating behaviours were investigated, drop-out rate was monitored, and weight-loss measured using Body Mass Index change. The study group was divided into two groups with one group receiving a motivational interview, whilst the other group did not. Finally *HT1A*, *HT2A* and *HT2C* serotonin receptor genotypes were determined along with *DRD2* and *DRD4* dopamine receptor genotypes.

There were no significant correlations between any genotype and extent of motivation to make behavioural changes. Likewise there were no significant correlations between any genotype and the change in motivation over the study. One reason for this lack of significance was the wide variation in scores for the Relative Autonomy Index (RAI). There appeared to be a relationship between *DRD2*/*DRD4* risk alleles and a reduction in RAI over the course of the study, participants with all other genotypes had an average increase in RAI score, however so many participants with the *DRD2*/*DRD4* risk alleles dropped out of the study that the remaining small number did not allow significance to be achieved.

The influence of genotype on adherence and on weight loss measured by change in BMI was investigated. The *DRD2*/*DRD4* composite genotype had a significant influence on the likelihood of participants dropping out from the study. Participants with this genotype had a 50% drop out rate compared with a drop out rate of 16%. However there were no significant correlations between any genotype and weight loss measured by change in BMI.

There appeared to be a relationship between RAI scores and drop out rate in males, the males who subsequently dropped out had lower RAI scores for motivation compared to participants who did not drop out (6.0 vs 23.1), however the variability in RAI scores meant that this difference was not significant. An interesting result was observed when the relationship between RAI score and BMI change was considered. There was a significant correlation between RAI scores and BMI change, but only in the group who received a motivational interview. This pattern was repeated when the correlation between eating behaviours measured by the Three Factor Eating questionnaire (TFEQ) and BMI change were considered. There were significant correlations between scores for restraint and BMI change and between scores for hunger and BMI change (inverse correlation), but again this effect was only seen in the group who received a motivational interview.

The serotonin receptor genes *HT1A* and *HT2C* influence extent of hunger measured by TFEQ. The GG genotype of the *HT1A* gene and the C allele of the *HT2C* gene were associated with reduced scores for hunger. Lower scores for hunger using this questionnaire have been associated with obesity and indicate a loss of the ability to sense true hunger. Both the GG genotype of the *HT1A* gene and the C allele of the *HT2C* gene are associated with obesity and the novel data presented here indicates that this risk of obesity is due to a genotype-influenced loss of the perception of hunger.

The application of motivational interviewing to this obesity intervention programme had somewhat contradictory results. The participants who received a motivational interview (MI) lost significantly more weight, measured by BMI change, than participants who did not receive a motivational interview. This finding demonstrated the efficacy of this technique in a weight-loss situation. The effect of motivational interviewing on drop-out rate was more unexpected, as the group who received an MI had a drop out rate twice as high as the drop out of the group who did not receive an MI. It appears that the MI helps the participants to decide how serious they are about attempting to

make lifestyle changes, and those participants who realise they are not ready to change drop out rather than persist with the programme.

In summary we have demonstrated that serotonin receptor gene genotype has a significant influence on perception of hunger. We have also shown that dopamine receptor gene polymorphisms significantly influence the drop out rate from an obesity intervention study, and suggest that this is due to the influence of these polymorphisms both on motivation to exercise and on restrained eating behaviours. Finally we have demonstrated the efficacy of motivational interviewing in an obesity intervention programme and have shown that motivational interviews influence the relationship between measures of motivation or eating behaviours and weight loss measured by BMI change

These novel findings indicate that serotonin plays a key role in determining appetite. They also indicate that extent of dopamine signalling may influence an individual's motivation to engage in weight-loss programmes. Further studies are required to confirm these findings and investigate the underlying mechanisms involved.

Budget summary

The project was completed within the budget, as detailed in the introduction and methods significant extra work was also completed within the budget. A final budget summary is included with this report.

Papers presented

Data from this project was presented as a poster at the Bial Foundation conference in Porto, Portugal, April 2010, a copy of the poster is enclosed.

Papers in preparation

Several papers are in preparation to report these results, they will be submitted to peer-reviewed journals within the next few months.

Sheffield Hallam University

Polymorphisms in serotonin and dopamine receptor genes influence weight loss, motivation and eating behaviours

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Introduction

Serotonin (5-HT) and dopamine are modulators of a variety of central nervous system processes including those involved in motivation, depression, control of sleep, and ingestive behaviours. In particular, serotonin has an effect on appetite (Blundell 1984) whilst dopamine is involved in motivation and reward behaviours including those involving food.

These neurotransmitters mediate their effects through a number of receptor subtypes. Polymorphisms in the promoters of the genes that code for some of the serotonin receptors have been associated with an increased risk of depression (*HT1A* gene), obesity (*HT2A* and *HT2C* genes) and diabetes (*HT2C* gene), (Lemonde *et al* 2003, Rosmond *et al* 2002, Yuan *et al* 2000). Polymorphisms in the dopamine receptor genes *DRD2* and *DRD4* have been associated with obesity and bulimia (Epstein *et al* 2000).

The Behavioural Regulation in Exercise questionnaire (Breq-2) measures motivation to exercise using a number of subscales including Intrinsic Regulation, a measure of a person's self-motivation to exercise (Markland & Tobin 2004).

The Three-factor Eating Questionnaire (TFEQ) measures three major factors designated as Cognitive Restraint, Disinhibition and Susceptibility to feelings of Hunger (Stunkard and Messick, 1985). High scores for Cognitive restraint and low scores for Hunger using this questionnaire are associated with raised BMI and disordered patterns of eating (Adami et al 1996).

Aim

To determine whether polymorphisms in serotonin or dopamine receptor genes influence changes in body mass index (BMI) or waist/hip ratio in participants of a diet and exercise programme, the extent of motivation of the participants in relation to exercise, and the participants eating behaviour scores.

Materials and Methods

Subjects and measures: 100 obese participants of a community-based 12-week exercise and diet intervention programme took part in the study. BMI and waist/hip ratio data were collected. The BREQ-2 questionnaire was used to quantify motivation in relation to exercise. The Three-factor Eating Questionnaire (TFEQ) was used to quantify eating behaviours. Measures were repeated at the end of the programme. DNA was extracted from a buccal swab and genotyped for polymorphisms in serotonin and dopamine receptor genes. Serotonin receptor gene polymorphisms were genotyped using Taqman assays (Applied Biosystems). Dopamine receptor genotyping was carried out using PCR and restriction enzyme digestion, the resulting bands were resolved using gel electrophoresis.

Polymorphisms investigated

HT1A gene - (-1019C/G) polymorphism in the promoter
HT2A gene - (-1438G/A) polymorphism in the promoter
HT2C gene - (-759T/C) polymorphism in the promoter
DRD2 gene - Taq 1 allele (A1/A2)
DRD4 gene - Variable Number tandem repeat in exon 3 (2-6 repeats or 7+ repeats)

Results and Discussion

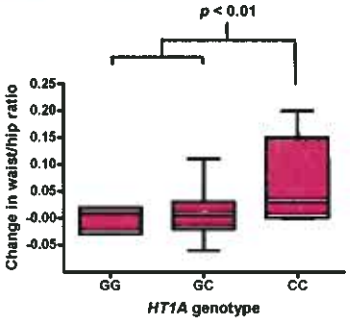

Figure 1: *HT1A* genotype significantly influences the extent of waist/hip ratio change in the participants over the course of the intervention programme. The CC genotype was associated with an increase in waist/hip ratio indicating that these subjects had gained weight despite participating in the programme. The G allele has been associated with an increased risk of depression, itself a risk factor for obesity. Participants with the GG or GC genotype may be more responsive to the support given during the intervention programme compared to those with the CC genotype.

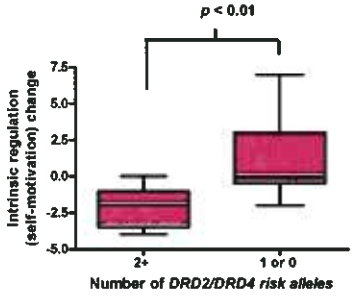

Figure 2: The *DRD2* Taq1 allele and *DRD4* 7R VNTR when considered together were significantly associated with a low or negative change in intrinsic regulation as measured by the BREQ-2 questionnaire . This indicates that people with these genotypes find it harder to become self-motivated when taking part in an exercise programme compared to people with a more favourable genotype. This may be because people with the *DRD2* Taq1 allele or *DRD4* 7R allele experience reduced reward signalling mediated by dopamine receptors in response to exercise.

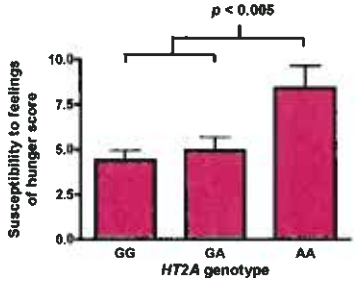

Figure 3: The G allele in the promoter region of the *HT2A* gene was significantly associated with lower scores of hunger measured using the TFEQ questionnaire. The G allele of this gene is associated with obesity. Low hunger scores as measured by the TFEQ have also been associated with obesity (Adami et al 1996). This is because obese people can lose their ability to sense true hunger. These results indicate that the *HT2A* gene may influence perceptions of hunger.

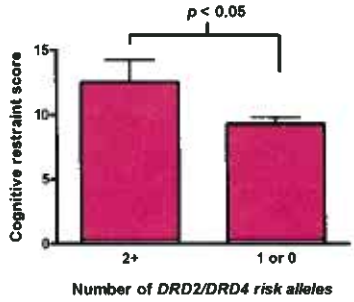

Figure 4: The *DRD2* Taq1 allele and *DRD4* 7R VNTR when considered together were significantly associated with higher cognitive restraint scores as measured by the TFEQ. High cognitive restraint scores can be associated with loss of control of eating, as people who strictly control their diet are more susceptible to binge eating and the associated tendency to obesity. Dopamine receptor genes mediate feelings of reward in relation to food. These results indicate that polymorphisms in dopamine receptor genes may modify reward signalling in response to food which may influence attitudes to dietary control.

Conclusions

These findings suggest that success when attempting to lose weight is influenced by polymorphisms in serotonin and dopamine receptor genes. This influence is mediated by the effect of these genes on eating behaviours and on motivation to participate in exercise. Further studies are required to investigate ways of personalising diet and exercise programmes to take into account the influence of these genetic factors.

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