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# Co-administration of SR141716 with peptide $YY_{3–36}$ or oxyntomodulin has additive effects on food intake in mice

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**Background:** SR141716 has been shown to significantly inhibit food intake and reduce body weight by antagonizing $CB_1$ receptors. The gut hormones peptide $YY_{3–36}$ ($PYY_{3–36}$) and oxyntomodulin (OXM) inhibit food intake through $Y_2$ and Glucagon-Like-Peptide (GLP)-1 receptors respectively.
**Objective:** To determine the effects of co-administration of SR141716 with either $PYY_{3–36}$ or OXM in mice on food intake.
**Methods:** Mice (n = 14 per group) were fasted for 16 h prior to study days and given two intraperitoneal injections: study 1, vehicle–saline, SR141716–saline, vehicle–PYY3–36 or SR141716–PYY3–36; study 2: vehicle–saline, SR141716–saline, vehicle–OXM or SR141716–OXM.
Food was returned and measured following injections.
**Results:** Co-administration of SR141716–$PYY_{3–36}$ or SR141716–OXM showed greater inhibition in food intake when compared with administration of SR141716, $PYY_{3–36}$ or OXM alone.
**Conclusion:** Our data show that SR141716 in combination with $PYY_{3–36}$ or OXM reduces food intake additively in mice.

Keywords: food intake, oxyntomodulin, $PYY_{3–36}$, Rimonabant, SR141716
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## Introduction

A number of agents have been shown to reduce food intake including the cannabinoid-1 ($CB_1$) receptor antagonist SR141716 (Rimonabant) and the gut hormones peptide $YY_{3–36}$ ($PYY_{3–36}$) and oxyntomodulin (OXM). The endocannabinoid system contributes to the physiological regulation of energy balance, food intake and lipid and glucose metabolism through the $CB_1$ receptors in the brain [1,2]. In animal models and humans, $CB_1$ blockade results in weight loss, decreased waist circumference, resistance to diet-induced obesity and associated dyslipidaemia [2,3].

The gut-derived hormones $PYY_{3–36}$ and OXM are produced in the L cells of the gastrointestinal tract. Acute and chronic administration has been shown to reduce food intake in rodents. In humans, peripheral administration of OXM and $PYY_{3–36}$ has been shown to reduce food intake compared with vehicle control groups [4–6].

SR141716, $PYY_{3–36}$ and OXM reduce food intake through different appetite circuits, namely $CB_1$, $Y_2$ and GLP-1 receptors. In the present study, we demonstrate

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that co-administration of SR141716 with $PYY_{3–36}$ or OXM has additive effects on decreasing food intake, following co-administration peripherally to mice.

## Methods

All procedures were approved by the British Home-Office Animals (Scientific Procedures) Act 1986 (project licence number 70/5281). For each study, singly housed adult male C57BL6 mice (Harlan, Bicester, UK) were fasted for 16 h and received an intraperitoneal (i.p.) injection (100 μl) in the early light phase. Following injection, food intake was measured at 1, 2, 4, 8 and 24 h postinjection. $PYY_{3–36}$ (Phoenix Pharmaceuticals, Belmont, CA, USA) and OXM (Bachem UK, Merseyside, UK) were dissolved in saline. SR141716 (Vernalis, Wokingham, Buckinghamshire, UK) was dissolved in 10% dimethyl sulphoxide solution (Sigma, Poole, UK) and sonicated for 2 min. The doses of SR141716, $PYY_{3–36}$ and OXM used in these studies have previously been shown to inhibit food intake when injected intraperitoneally into mice [2,7].

### Study 1: Effect of Co-administration of SR141716 and $PYY_{3–36}$ on Food Intake in Fasted Mice

Animals each received two i.p. injections of vehicle–saline, SR141716 (0.3 mg/kg)–saline, vehicle–$PYY_{3–36}$ (50 μg/kg) or SR141716 (0.3 mg/kg)–$PYY_{3–36}$ (50 μg/kg).

### Study 2: Effect of Co-administration of SR141716 and OXM on Food Intake in Fasted Mice

Animals each received two i.p. injections of vehicle–saline, SR141716 (0.3 mg/kg)–saline, vehicle–OXM (6.2 mg/kg) or SR141716 (0.3 mg/kg)–OXM (6.2 mg/kg).

### Statistical Analysis

Results are shown as mean ± s.e.m. and were analysed using a one-way ANOVA with a Dunnett's two-sided *post hoc* test; $p < 0.05$ was considered statistically significant.

## Results

### Study 1: Effect of Co-administration of SR141716 and $PYY_{3–36}$ on Food Intake in Fasted Mice

SR141716–saline, vehicle–$PYY_{3–36}$ and SR141716–$PYY_{3–36}$ administration significantly inhibited food intake in the first hour postinjection when compared with vehicle–saline administration (figure 1). Co-administration of SR141716–$PYY_{3–36}$ led to significantly greater reduction in food intake than SR141716–saline- or vehicle-$PYY_{3–36}$-injected animals [food intake 0–1 h (g): vehicle–saline 0.97 ± 0.04, SR141716–saline 0.87 ± 0.05, vehicle–$PYY_{3–36}$ 0.75 ± 0.07, SR141716–$PYY_{3–36}$ 0.52 ± 0.06 ($p < 0.05$, SR141716–saline vs. vehicle–saline; $p < 0.001$, vehicle–$PYY_{3–36}$ vs. vehicle–saline; $p < 0.0001$, SR141716–$PYY_{3–36}$ vs. vehicle–saline; $p < 0.01$, SR141716–$PYY_{3–36}$ vs. SR141716–saline; $p < 0.01$, SR141716–$PYY_{3–36}$ vs. vehicle–$PYY_{3–36}$)] (figure 1). There were no significant changes in interval food intake at any further time points (table 1). Cumulative food intake was significantly reduced in the vehicle–$PYY_{3–36}$ up to 2-h postinjection and in the SR141716–$PYY_{3–36}$ co-administration group up to 4-h postinjection (table 1).

### Study 2: Effect of Co-administration of SR141716 and OXM on Food Intake in Fasted Mice

SR141716–saline, vehicle–OXM and SR141716–OXM significantly reduced food intake in the first hour post-administration compared with vehicle-saline-injected animals. Co-administration of SR141716–OXM decreased food intake greater than either SR141716–saline- or vehicle-OXM-injected animals [food intake 0–1 h (g):

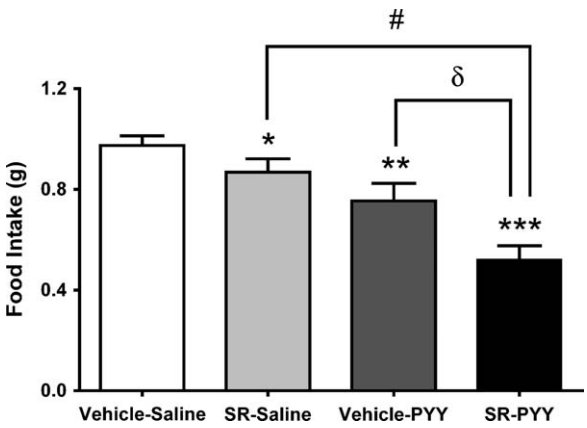


**Fig. 1** The effect of co-administration of vehicle–saline, SR141716 (0.3 mg/kg)–saline, vehicle–$PPY_{3–36}$ (50 μg/kg) or SR141716 (0.3 mg/kg)–$PYY_{3–36}$ (50 μg/kg) on food intake in fasted mice in the first hour postinjection. SR, SR141716; $PYY_{3–36}$, peptide $YY_{3–36}$. *$p < 0.05$, SR141716–saline vs. vehicle–saline; **$p < 0.001$, vehicle–$PYY_{3–36}$ vs. vehicle–saline; ***$p < 0.0001$, SR141716–$PYY_{3–36}$ vs. vehicle–saline; #$p < 0.01$, SR141716–$PYY_{3–36}$ vs. SR141716–saline; δ$p < 0.01$, SR141716–$PYY_{3–36}$ vs. vehicle–$PYY_{3–36}$.

**Table 1** The effect of co-administration of either vehicle–saline, SR141716 (0.3 mg/kg)–saline, vehicle–$PYY_{3–36}$ (50 μg/kg), SR141716 (0.3 mg/kg)–$PYY_{3–36}$ (50 μg/kg) (study 1) or vehicle–saline, SR141716 (0.3 mg/kg)–saline, vehicle–OXM (6.2 mg/kg), SR141716 (0.3 mg/kg)–OXM (6.2 mg/kg) (study 2) on interval and cumulative food intake in fasted mice

| | Treatment | 0–1 h | 1–2 h | 2–4 h | 4–8 h | 8–24 h | 0–1 h | 0–2 h | 0–4 h | 0–8 h | 0–24 h |
|---|---|---|---|---|---|---|---|---|---|---|---|
| Study 1 | Vehicle–saline | 0.97 ± 0.04 | 0.37 ± 0.05 | 0.55 ± 0.07 | 0.80 ± 0.08 | 4.00 ± 0.08 | 0.97 ± 0.04 | 1.35 ± 0.07 | 1.90 ± 0.10 | 2.70 ± 0.11 | 6.70 ± 0.12 |
| | SR–saline | 0.87 ± 0.05* | 0.34 ± 0.04 | 0.56 ± 0.06 | 0.94 ± 0.06 | 3.89 ± 0.14 | 0.87 ± 0.05* | 1.21 ± 0.06 | 1.77 ± 0.09 | 2.71 ± 0.11 | 6.60 ± 0.16 |
| | Vehicle–$PYY_{3–36}$ | 0.75 ± 0.07** | 0.29 ± 0.05 | 0.66 ± 0.04 | 0.89 ± 0.05 | 4.32 ± 0.08 | 0.75 ± 0.07** | 1.05 ± 0.10* | 1.70 ± 0.09 | 2.59 ± 0.09 | 6.91 ± 0.13 |
| | SR–$PYY_{3–36}$ | 0.52 ± 0.06*** | 0.25 ± 0.04 | 0.65 ± 0.05 | 0.97 ± 0.04 | 4.04 ± 0.13 | 0.52 ± 0.06*** | 0.77 ± 0.07* | 1.41 ± 0.09* | 2.38 ± 0.11 | 6.43 ± 0.14 |
| Study 2 | Vehicle–saline | 0.95 ± 0.04 | 0.30 ± 0.08 | 0.65 ± 0.06 | 1.13 ± 0.07 | 4.01 ± 0.15 | 0.95 ± 0.04 | 1.25 ± 0.10 | 1.90 ± 0.10 | 3.02 ± 0.12 | 7.03 ± 0.15 |
| | SR–saline | 0.71 ± 0.08* | 0.33 ± 0.04 | 0.56 ± 0.09 | 1.09 ± 0.09 | 4.08 ± 0.14 | 0.71 ± 0.08* | 1.04 ± 0.10 | 1.61 ± 0.12 | 2.70 ± 0.16 | 6.78 ± 0.16 |
| | Vehicle–OXM | 0.68 ± 0.06** | 0.30 ± 0.03 | 0.71 ± 0.07 | 1.24 ± 0.06 | 3.96 ± 0.17 | 0.68 ± 0.06** | 0.97 ± 0.08* | 1.68 ± 0.10 | 2.92 ± 0.11 | 6.88 ± 0.18 |
| | SR–OXM | 0.38 ± 0.11*** | 0.46 ± 0.07 | 0.58 ± 0.07 | 1.24 ± 0.10 | 4.24 ± 0.12 | 0.38 ± 0.11*** | 0.84 ± 0.08* | 1.42 ± 0.10* | 2.66 ± 0.12 | 6.90 ± 0.14 |

OXM, oxyntomodulin (6.2 mg/kg); $PYY_{3–36}$, peptide $YY_{3–36}$ (50 μg/kg); SR, SR141716 (0.3 mg/kg).
*$p < 0.05$ vs. vehicle–saline.
**$p < 0.001$ vs. vehicle–saline.
***$p < 0.0001$ vs. vehicle–saline.

vehicle–saline 0.95 ± 0.04, SR141716–saline 0.71 ± 0.08, vehicle–OXM 0.68 ± 0.06, SR141716–OXM 0.38 ± 0.11 ($p < 0.01$, SR141716–saline vs. vehicle–saline; $p < 0.005$, vehicle–OXM vs. vehicle–saline; $p < 0.0001$, SR141716–OXM vs. vehicle–saline; $p < 0.01$, SR141716–OXM vs. SR141716–saline; $p < 0.01$ SR141716–OXM vs. vehicle–OXM)] (figure 2). There were no significant differences in interval food intake at any other time points (table 1). However, cumulative food intake was seen to be significantly decreased in the vehicle–OXM co-administration group up to 2-h postinjection and in the SR141716–OXM group up to 4-h postinjection (table 1).

There were no side effects noted in the immediate and post-follow-up period (changes in stool pattern, diarrhoea and tendency to anorexia) in animals in study 1 or 2.

## Discussion

SR141716, $PYY_{3–36}$ and OXM act through different receptor mechanisms. Combined administration of these agents may be expected to produce an additive reduction in food intake through these different appetite pathways. Our data show for the first time that administration of SR141716 in combination with either $PYY_{3–36}$ or OXM in mice reduces food intake additively.

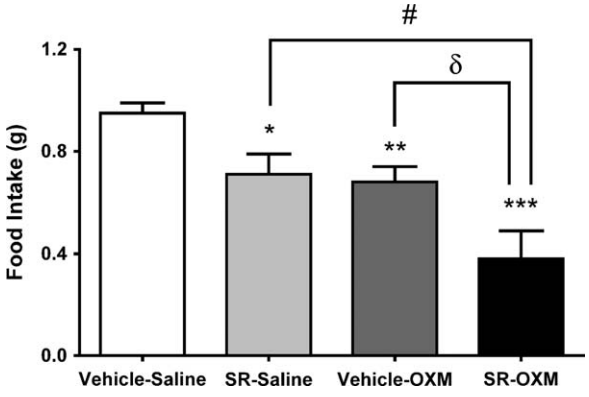


**Fig. 2** The effect of co-administration of vehicle–saline, SR141716 (0.3 mg/kg)–saline, vehicle–OXM (6.2 mg/kg) or SR141716 (0.3 mg/kg)–OXM (6.2 mg/kg) on food intake in fasted mice in the first hour postinjection. SR, SR141716; OXM, oxyntomodulin. *$p < 0.01$, SR141716 (0.3 mg/kg)–saline vs. vehicle–saline; **$p < 0.005$, vehicle–OXM (6.2 mg/kg) vs. vehicle–saline; ***$p < 0.0001$, SR141716–OXM vs. vehicle–saline; #$p < 0.01$, SR141716–OXM vs. SR141716–saline; δ$p < 0.01$, SR141716–OXM vs. vehicle–OXM.

Our findings may have clinical relevance as obese patients are likely to require a number of obesity therapies given in combination to reduce body weight from an obese to a normal body mass index category, reducing the associated morbidity and mortality. An effective clinical therapy for obesity is considered as the one that results in a 5–10% weight loss [3]. Lower doses of SR141716 result in lower incidences of adverse side effects [3]. It is possible that combination therapy of SR141716 with $PYY_{3–36}$ or OXM would allow lower doses of each agent to be used, resulting in an effective antiobesity therapy with minimal side effects.

Further studies are required to determine if SR141716 in combination with either $PYY_{3–36}$ or OXM provides a more effective potential therapy for individuals with obesity than SR141716, $PYY_{3–36}$ or OXM administration alone.

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