Poster #: 3066-LB

# Activating the Adipose Thermogenic Switch: A First-in-Class *Ucp1* saRNA Drives Durable, High-Quality Weight Loss in Diet-Induced Obese Mice

Jie Wang, Jun-nan Chen, Fei-fei Xu, Xiao-jie Zhou, Wei-Hsiang Lin, Moorim Kang and Long-Cheng Li
Ractigen Therapeutics, Suzhou, China

Ractigen THERAPEUTICS

## Financial Disclosures

All authors are employees of Ractigen Therapeutics and may hold stock or stock options. This study was sponsored by Ractigen Therapeutics.

## Background

- Obesity is a global epidemic associated with type 2 diabetes (T2D), metabolic dysfunction-associated steatotic liver disease (MASLD), and cardiovascular disease (CVD). While glucagon-like peptide-1 receptor agonists (GLP-1 RAs, e.g., semaglutide) achieve 15–25% weight loss, they are limited by gastrointestinal (GI) side effects, lean mass loss, and rapid weight regain after discontinuation.
- **Uncoupling protein 1 (*UCP1*)** is a key mediator of non-shivering thermogenesis in brown and beige adipose tissue and a validated target for increasing energy expenditure. However, it has remained "undruggable" by conventional therapeutics due to off-target toxicity and poor specificity.
- **saRNA** is a class of synthetic duplex RNAs designed to target gene promoters to induce gene expression at the transcriptional level via the **RNAa** mechanism.
- **LiCO** (lipid-conjugated oligonucleotide) is a lipid-RNA conjugate which can deliver saRNA into adipose tissue by subcutaneous (SC) dosing.

## Objectives

To evaluate the feasibility of Ucp1 activation by LiCO conjugated Ucp1 saRNA (LiCO-saUcp1) in wild-type mice and its therapeutic benefits, alone or in combination with semaglutide, in diet-induced obesity (DIO) mice.

## Materials and Methods

- **Animals:** Wild-type (C57BL/6J) mice were used for pharmacodynamic (PD) assessment; DIO mice (10–17 weeks on high-fat diet, HFD) were used for efficacy studies.
- **Treatments:** LiCO-saUcp1 (SC, 1–30 mg/kg), semaglutide (IP, 0.04–0.1 mg/kg), or their combination.
- **Key Endpoints:** Body weight (BW), body composition, energy expenditure markers, and hepatic histology were monitored for up to 2 months post-withdrawal to assess durability.

## Results

### LiCO-saUcp1 induced *Ucp1* in wild-type mice

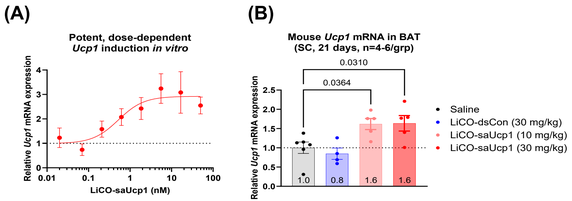


**Figure 1. LiCO-saUcp1 induced *Ucp1* mRNA expression both *in vitro* and *in vivo*.** **(A)** *In vitro* transfection of mouse NCTC 1469 cells with LiCO-saUcp1 resulted in a potent, dose-dependent induction of *Ucp1* mRNA ($EC_{50}$ = 1.4 nM at 72 h). **(B)** A single SC injection (10 or 30 mg/kg) in C57BL/6J mice resulted in significant and sustained *Ucp1* upregulation in brown adipose tissue (BAT) for 21 days, confirming robust target engagement. Data were analyzed by one-way ANOVA.

### Efficacy study #1: LiCO-saUcp1 suppressed body weight gain in DIO mice

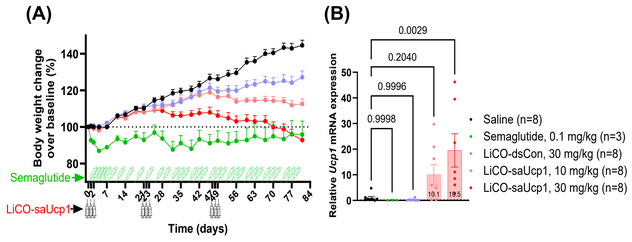


**Figure 2. LiCO-saUcp1 dose-dependently attenuated body weight gain and demonstrated on-target activity in the inguinal white adipose tissue (iWAT) of DIO mice following 10 weeks of HFD**. DIO mice received 3 daily SC injections (QD × 3) every 3 weeks for 3 cycles. **(A)** LiCO-saUcp1 (10 and 30 mg/kg) attenuated BW gain over an 81-day period compared to saline controls. **(B)** On day 83, target engagement was further validated by robust *Ucp1* mRNA upregulation in iWAT, demonstrating precise and potent *in vivo* activity. Data were analyzed by one-way ANOVA.

### Efficacy study #2: LiCO-saUcp1 enhanced therapeutic benefits of semaglutide monotherapy

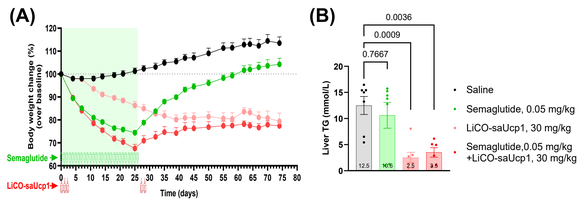


**Figure 3. Co-administration of LiCO-saUcp1 with semaglutide synergistically enhanced body weight loss and reduced hepatic triglycerides (TG).** DIO mice (17-week HFD, n=6–7 per group) were treated with daily semaglutide (0.05 mg/kg) and/or LiCO-saUcp1 (30 mg/kg SC on days 0–2 and 27–28). **(A)** Combination therapy produced the most pronounced and durable BW reduction over 74 days compared with either monotherapy. **(B)** On day 78, LiCO-saUcp1 (alone and in combination) significantly reduced liver TG levels compared to saline and semaglutide monotherapy, indicating profound hepatic benefits. Data were analyzed by one-way ANOVA.

### Efficacy study #3: LiCO-saUcp1 prevented post-withdrawal weight rebound

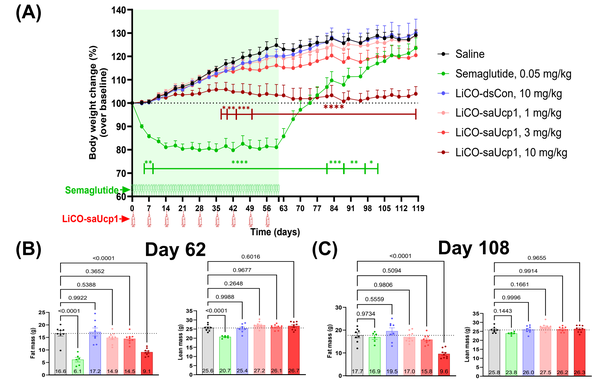


**Figure 4. LiCO-saUcp1 selectively reduced fat mass, preserved lean mass, and prevented post-withdrawal weight rebound.** DIO mice (11-week HFD, n=7-9 per group) received either daily semaglutide (0.05 mg/kg) for 61 days or weekly LiCO-saUcp1 (1–10 mg/kg) for 56 days. **(A)** Following treatment withdrawal, LiCO-saUcp1-treated mice exhibited significantly attenuated weight rebound compared with rapid regain observed in semaglutide group. Data were analyzed by two-way ANOVA. Significance levels are denoted as $^{*}P < 0.05$, $^{**}P < 0.01$, $^{***}P < 0.001$, and $^{****}P < 0.0001$. **(B-C)** Body composition analysis at day 62 (during treatment) and day 108 (post-withdrawal) demonstrated dose-dependent reduction in fat mass with complete preservation of lean mass, indicating selective and durable weight loss. Data were analyzed by one-way ANOVA.

### Efficacy study #3: LiCO-saUcp1 alleviated hepatic steatosis in a dose-dependent manner

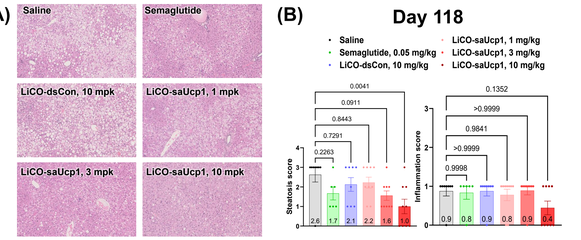


**Figure 5. LiCO-saUcp1 mediated durable, dose-dependent amelioration of hepatic steatosis and inflammation.** Livers were harvested on day 118 after treatment cessation for histological evaluation. **(A-B)** H&E staining showed that LiCO-saUcp1 treatment produced significant, dose-dependent improvements in both steatosis and inflammation scores relative to saline controls, with effects that were durably maintained after treatment withdrawal. Data are presented as mean ± SEM (n=6-9 per group) and were analyzed by one-way ANOVA.

### Efficacy study #4: Co-treatment enabled effective weight management with low-dose semaglutide

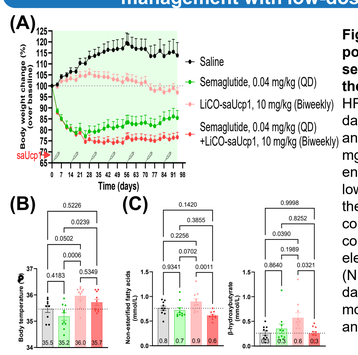


**Figure 6. Combination therapy potentiated the efficacy of lower-dose semaglutide via Ucp1-mediated thermogenesis.** DIO mice (15-week HFD, n=10 per group) were treated with daily low-dose semaglutide (0.04 mg/kg) and/or biweekly LiCO-saUcp1 (10 mg/kg). **(A)** Co-treatment markedly enhanced BW reduction compared with low-dose semaglutide alone throughout the study. On-target Ucp1 activation was confirmed by a significant increase in core body temperature on day 91 **(B)** and elevated serum non-esterified fatty acid (NEFA) and β-hydroxybutyrate levels on day 94 **(C)**, indicating enhanced lipid mobilization and oxidation. Data were analyzed by one-way ANOVA.

## Conclusion

LiCO-saUcp1 represents a first-in-class therapeutic that drives potent, durable weight loss via Ucp1-mediated thermogenesis and browning of white adipose tissue. Its ability to selectively reduce fat mass while preserving lean mass—and critically, to prevent post-withdrawal weight rebound—addresses key limitations of current anti-obesity therapies. Furthermore, its capacity to alleviate hepatic steatosis and synergize with semaglutide represents a highly differentiated therapeutic mechanism with strong complementary potential alongside incretin-based therapies.

Contact: bd@Ractigen.com