Nimacimab, A Peripherally Restricted CB1 Inhibitor, Promotes Metabolic Homeostasis In A Diet-Induced Obesity (DIO) Mouse Model As Demonstrated By Weight Loss, Restored Hormonal Regulation, And Reduced Inflammatory Biomarkers

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Background

The endocannabinoid system, specifically cannabinoid receptor 1 (CB1), plays a critical role in obesity by impacting:
- Body weight
- Lipid metabolism
- Food intake

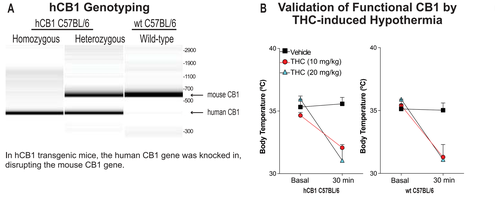
Nimacimab is a novel antibody-based CB1 inhibitor:
- IgG4 negative allosteric modulating inverse agonist of human CB1 (hCB1)
- Acts through a non-incretin pathway
- Antibody-based inhibitor with nearly complete peripheral restriction
- Excellent Ph1 safety profile with promising preclinical efficacy
- Ph2 obesity clinical trial is expected to be completed in Q3 2025 (NCT06577090)

Key question: Can nimacimab promote effective body weight loss and restore metabolic homeostasis in a diet-induced obesity (DIO) mouse model?

steatosis
high cholesterol
high blood glucose
insulin resistance
obesity-induced inflammation

normal fat deposition in the liver
normal cholesterol
normal blood glucose
restored insulin homeostasis
reduced inflammation

Functional Knock-in and Signaling of Human CB1 Receptor (hCB1) in Transgenic Mice



In hCB1 transgenic mice, the human CB1 gene was knocked in, disrupting the mouse CB1 gene.

Figure 1. Characterization of hCB1 transgenic mice. (A) Representative qPCR genotyping result of hCB1 transgenic mice. Wild-type mice (wt C57BL/6) were used as a negative control for human CB1 gene and a positive control for mouse CB1 gene. (B) Wild-type and homozygous hCB1 animals were given two doses of THC to induce a CB1-mediated transient hypothermia (n=3 per group). (C) hCB1 mice gained body weight as expected when fed a high-fat diet (HFD). Wt C57BL/6 data was generated by The Jackson Laboratory.

Objectives

Primary goals:
- Develop a DIO model using hCB1 transgenic mice
- Evaluate the efficiency of nimacimab in promoting body weight loss and restoring metabolic homeostasis in obese mice
- Investigate the mechanisms of action of nimacimab
- Confirm the rigor and reproducibility of nimacimab as an effective drug to reverse obesity-induced metabolic dysfunction

Initial DIO Study

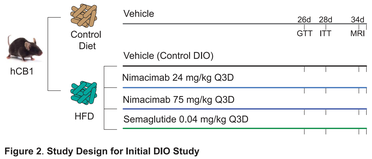


Figure 2. Study Design for Initial DIO Study

6-8 weeks old hCB1 mice were single housed and fed ad libitum with either regular chow (Control Diet) or a high-fat diet (HFD, 60 kcal % fat, D12492) under constant ambient conditions of 22 ± 2 °C with constant humidity (30-70%) and 12h/12h light/dark cycle. On Day -1, mice were randomized to the different groups based on body weight. After 14 weeks of HFD feeding, treatment started as indicated in Figure 2. Daily body weights were recorded. A glucose tolerance test (GTT) was performed on day 26, an insulin tolerance test (ITT) on day 28, and on day 34, body composition was analyzed by EchoMRI.

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Conflicts of interest: The authors are consultants[1] or employees[2] of Skye Bioscience, a biotech company developing therapies for obesity and metabolic diseases.

Results

Nimacimab promotes effective body weight loss with preserved lean mass - initial DIO study

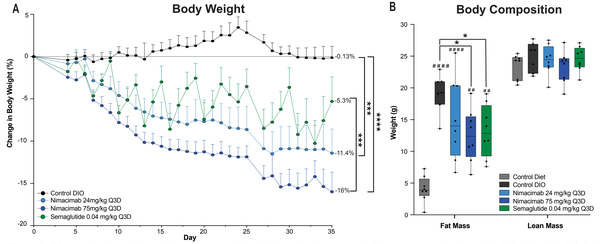

Figure 3. Effect on body weight and body composition of nimacimab or semaglutide treatments in mice with DIO. Mice with DIO fed a high-fat diet were dosed with nimacimab (24 mg/kg, or 75 mg/kg, IP, Q3D), semaglutide (0.04 mg/kg, SC, Q3D), or vehicle for 35 days. (A) Percentage of body weight change over time from day 0. Average % body weight change reported at day 35 of treatment. Two-way ANOVA followed by Tukey's multiple comparisons test, ***p<0.001, ****p<0.0001. (B) Fat mass and lean mass were measured by MRI on day 34. One-way ANOVA followed by Tukey's multiple comparisons test. *p<0.05, **p<0.01. # # # # p< 0.0001, # # p< 0.01 vs control diet. Data are expressed as mean ± SEM. n=8 per group.

Nimacimab improves metabolic homeostasis - initial DIO study

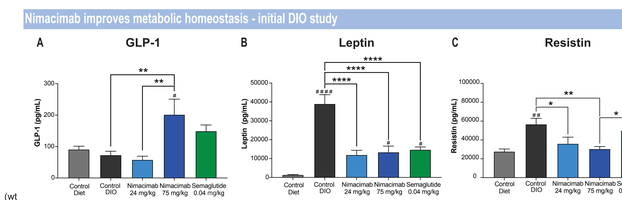

Figure 4. Nimacimab treatment improves key hormones in a DIO model. Serum was collected on day 35, and GLP-1 (A), Leptin (B), and Resistin (C) levels were determined with Bio Plex Multi-Plex immunoassay. Data are expressed as mean ± SEM. n=7-8 per group. One-way ANOVA followed by Tukey's multiple comparisons test. *p<0.05, **p<0.01, ****p<0.0001, # p< 0.05, # # p< 0.01 vs control diet.

Nimacimab improves glycemic control - initial DIO study

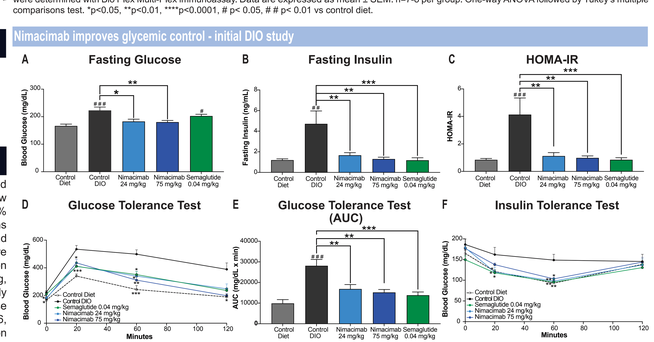

Figure 5. Nimacimab treatment improves glycemic control in a DIO model. On day 26 of treatment, animals were fasted for 4 h and glucose levels were measured before they were injected 2g/kg glucose. Effect of nimacimab and semaglutide treatments on fasting blood glucose (A), fasting insulin levels (B), HOMA-IR (C), and intraperitoneal GTT (D). Individual baseline levels of glucose were subtracted to calculate the area under the curve (AUC) (E). On day 28, animals were fasted for 4 h before receiving an intraperitoneal insulin injection (dose 0.75 U/kg at 5 mL/kg) and blood glucose was checked via tail prick to run an ITT (F). For (A), (B), (C), and (E) One-way ANOVA followed by Tukey's multiple comparisons test. For (D) and (F), two-way repeated measurements ANOVA analysis with time and treatment as main factors, followed by Tukey's multiple comparisons test. Data are expressed as mean ± SEM. n=7-8 per group. * p<0.05, **p<0.01, p<0.001, ****p<0.0001 vs control DIO and # p< 0.05, # # p<0.01, # # # p<0.001, # # # # p<0.0001 vs control diet.

Results

Nimacimab modulates lipid metabolism - initial DIO study

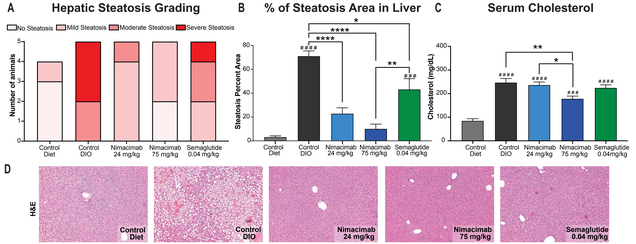

Figure 6. Hepatic steatosis is significantly reduced by nimacimab. (A) H&E-stained liver sections were scored by a pathologist, who was blinded to the treatment groups. 0-3 based on the % of hepatocytes with fat. 0 = no steatosis (<5%), 1 = mild (5-33%), 2 = moderate (>33-66%), and 3 = severe steatosis (>66%). (B) Quantification of steatosis percent area using a computer-aided analysis with Cellprofiler. (C) Cholesterol levels were measured in serum using a commercial kit (Quimica Clinica Aplicada). (D) Representative images of H&E-stained hepatic tissue showing differences in fat deposition among treatment groups. Data are expressed as mean ± SEM. n=4-5 For (B) and (C) One-way ANOVA followed by Tukey's multiple comparisons test. *p<0.05, **p<0.01, ***p<0.001, ****p<0.0001. # # # # p< 0.0001, # # # p< 0.001 vs control diet.

Nimacimab reduces obesity-induced inflammation - initial DIO study

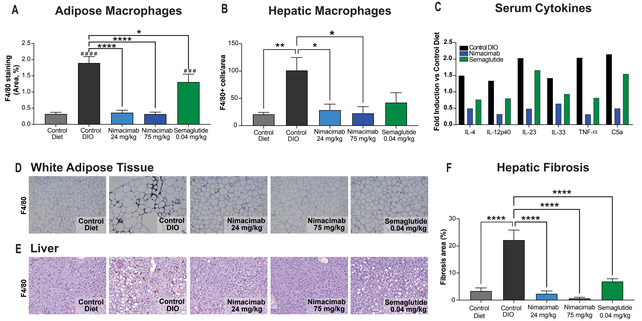

Figure 7. Obesity induced inflammation and fibrosis are significantly reduced by nimacimab. Quantification of F4/80 positive cells (macrophages) in inguinal white adipose tissue (iWAT) (A) and liver (B) was performed with ImageJ. (C) Changes in the expression of key inflammatory cytokines were assayed using the Proteome Profiler Mouse XL Cytokine Array on pooled serum samples (n=7-8 per group). Representative images of F4/80 staining in iWAT (D) and liver (E). (F) Quantification of collagen deposition from Sirius red staining (% area) to assess fibrosis. For (A), (B) , and (F) One-way ANOVA followed by Tukey's multiple comparisons test. *p<0.05, **p<0.01 ***p<0.001, ****p<0.0001, # # # # p< 0.0001, # # # p< 0.001 vs control diet. Data are expressed as mean ± SEM. n=3-7.

Repeat DIO Study

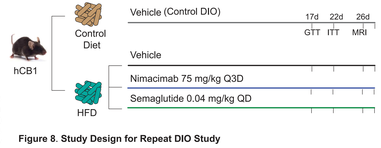

Figure 8. Study Design for Repeat DIO Study

6-8 weeks old hCB1 mice were single housed and fed ad libitum with either regular chow (Control Diet) or a high fat diet (HFD, 60 kcal % fat, D12492) under constant ambient conditions of 22 ± 2 °C with constant humidity (30-70%) and 12h/12h light/dark cycle. After 14 weeks, animals were randomized based on body weight before treatment started, as indicated in Figure 8. Daily body weights were recorded. A glucose tolerance test (GTT) was performed on day 17, an insulin tolerance test (ITT) on day 22, and on day 26, body composition was analyzed by EchoMRI.

Results

Nimacimab-dependent efficacy and mechanisms of action - DIO repeat study

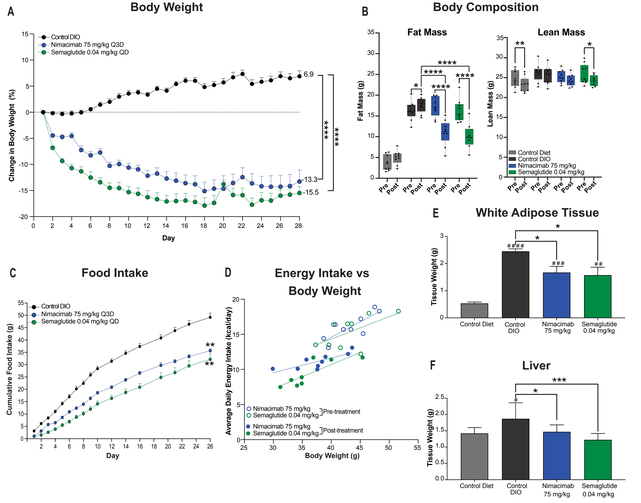

Figure 9. Nimacimab significantly reduces fat mass and improves glucose management in obese mice. hCB1 mice were fed chow or high-fat diet for 14 weeks. (A) Body weight change (%) from day 1 of treatment with nimacimab (75 mg/kg, IP, Q3D), semaglutide (0.04 mg/kg, SC, QD), or vehicle for 28 days. (B) Body composition measured by MRI on day 26 post-treatment (post) and day -1 pre-treatment (pre). (C) Cumulative food intake. (D) Correlation between average daily energy intake and body weight at the beginning and end of treatment. Day 28 necropsy tissue weights for iWAT (E) and liver (F). Effect of nimacimab and semaglutide treatment on oral GTT, day 17 (G) and on intraperitoneal ITT, day 22 (I). In (A), (B), (C), (G), and (I), two-way repeated measurements ANOVA or Mixed-effect analysis with time and treatment as main factors, followed by Tukey's multiple comparisons test. For (E), (F), and (H) One-way ANOVA followed by Tukey's multiple comparisons test. Data are expressed as mean ± SEM. n=8 per group. * p<0.05, **p<0.01, p<0.001, ****p<0.0001 vs control DIO and # p< 0.05, # # p<0.01, # # # p<0.001, # # # # p<0.0001 vs control diet. For (B) all DIO groups had significantly different fat mass to the control diet group pre- and post-treatment (p<0.001).

Conclusions

Skye successfully developed a DIO model:
- Knocked-in human CNR1 (CB1)
- Demonstrated functional CB1 signaling
- Similar rate and magnitude of weight gain

Nimacimab treatment resulted in:
- A dose-dependent body-weight loss
- Improved body composition
- Restored metabolic homeostasis
- Improved hormonal markers
- Reduced inflammation and fibrosis
- Improved lipid metabolism

These studies highlight that peripheral inhibition of CB1 with a non-blood brain barrier-crossing mAb can drive meaningful efficacy while minimizing the potential of CNS-related side effects, a common hurdle with small-molecule CB1 inhibitors. While semaglutide and nimacimab significantly reduced food intake and ultimately achieved similar weight loss, nimacimab further reduced inflammation markers, restored key metabolic hormones, and promoted productive lipid metabolism. These data suggest orthogonal mechanisms of action to incretin-based therapeutics, positioning nimacimab as a strong therapeutic candidate to treat obesity in patients who do not respond to currently available drugs, for weight loss maintenance, or as a combination therapy to enhance weight loss and overall metabolic health.