INTERVENTION WITH THE CCR2 INHIBITOR PROPAGERMANIUM ATTENUATES INSULIN RESISTANCE, ADIPOSE TISSUE INFLAMMATION AND NASH DEVELOPMENT

Petra Mulder
Kanita Salic
Roeland Hanemaaijer
Lars Verschuren
Reinout Stoop
Anita M. van den Hoek
Robert Kleemann

TNO innovation for life

TNO Metabolic Health Research, Leiden, The Netherlands

Background

Obese patients with insulin resistance (IR) and chronic inflammation in white adipose tissue (WAT) have a high risk of developing NASH. The C-C chemokine receptor type 2 (CCR2) has a crucial role in macrophage infiltration in WAT and liver, thereby contributing to chronic inflammation and IR.

Aim

We examined whether intervention with propagermanium, an inhibitor of CCR2, would attenuate: a) IR and WAT inflammation,
b) NASH development.
To mimic the situation in patients, mice with established obesity, IR and WAT inflammation were subjected to treatment.

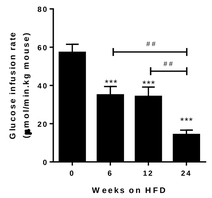
Methods

Experiment 1:
Male C57BL/6J mice on a high-fat diet (HFD) for 0,6,12 or 24 weeks to determine IR development and WAT inflammation for defining optimal time points for intervention with propagermanium. IR was assessed by hyperinsulinemic-euglycemic clamp.

Experiment 2:
Male C57BL/6J were fed a chow diet or HFD for 24 weeks. The intervention groups were pretreated with a HFD and then treated with CCR2 inhibitor propagermanium (0.05% w/w) after 6 weeks (early intervention) or 12 weeks (late intervention). NASH development was examined after a total of 24 weeks of diet feeding.

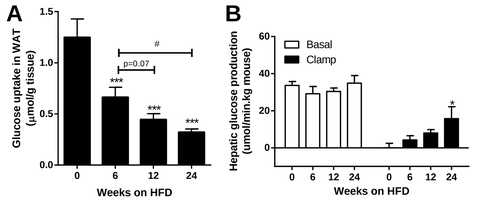
IR in WAT precedes hepatic IR and NASH

Whole-body IR

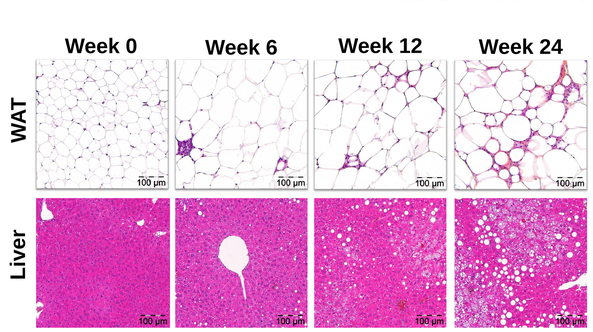


Glucose infusion rate was markedly lowered already after 6 weeks of HFD feeding, indicating development of whole body IR.

Tissue- specific IR



(A) Glucose uptake by WAT was already lowered after 6 weeks of HFD, indicating IR development in WAT.
(B) Hepatic glucose production was suppressed until 12 weeks, but insulins' inhibitory effect became weaker after 24 weeks of HFD and hepatic IR developed.

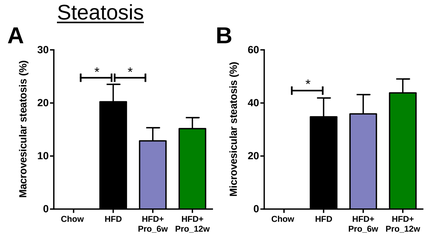


The development of IR in WAT after 6 weeks of HFD feeding was paralleled by increased infiltration of inflammatory cells.

Similar to WAT, hepatic IR coincided with inflammation observed after 24 weeks of HFD feeding.

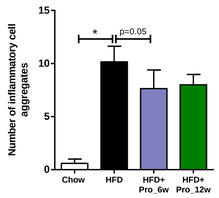
Propagermanium intervention attenuates NASH development

Steatosis

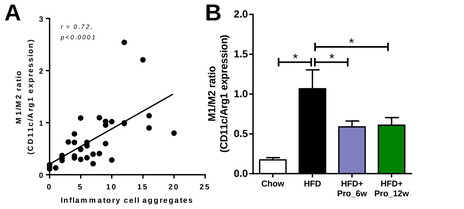


(A) Early propagermanium intervention significantly improves HFD-induced increase in macrovesicular steatosis.
(B) Microvesicular steatosis was comparable between HFD and propagermanium intervention groups.

Hepatic inflammation

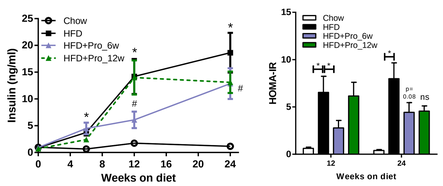


Quantification of inflammatory cell aggregates revealed that lobular inflammation tended to be reduced by early propagermanium intervention.

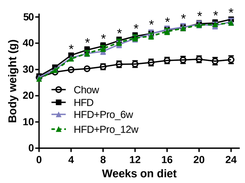


(A) Lobular inflammation strongly correlated with the ratio of M1/M2 macrophage markers. M1/M2 ratio was determined by gene expression of CD11c (pro-inflammatory, M1) and Arg1 (anti-inflammatory, M2).
(B) Propagermanium reduces M1/M2 ratio, suggesting lowering of pro-inflammatory M1 macrophages.

Propagermanium intervention improves IR and WAT inflammation independent of obesity

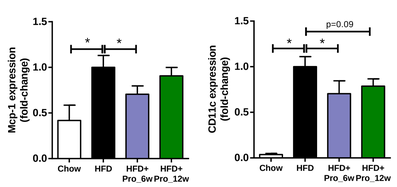


Early propagermanium treatment significantly improved HFD-induced hyperinsulinemia and insulin resistance.



Body weight was comparable between HFD and propagermanium intervention groups.

WAT inflammation



Early propagermanium treatment significantly reduced gene expression of pro-inflammatory markers MCP-1 and CD11c in WAT, indicating reduced WAT inflammation.

Conclusions

- Early risk factors of NASH, i.e. IR and WAT inflammation, can be reduced by propagermanium independently of obesity.
- Propagermanium can attenuate NASH development.
- Early propagermanium intervention was more effective to improve IR, WAT inflammation and NASH development.
- Therapeutic interventions directed at the MCP-1/CCR2 pathway should be initiated early to significantly attenuate NASH development.

A.vandenHoek@tno.nl | www.tno.nl