

This is an Open Access document downloaded from ORCA, Cardiff University's institutional repository: <https://orca.cardiff.ac.uk/id/eprint/89723/>

This is the author's version of a work that was submitted to / accepted for publication.

Citation for final published version:

Zhou, You , Llauradó, Gemma, Oresic, Matej, Hyötyläinen, Tuulia, Orho-Melander, Marju and Yki-Järvinen, Hannele 2015. Circulating triacylglycerol signatures and insulin sensitivity in NAFLD associated with the E167K variant in TM6SF2. *Journal of Hepatology* 62 (3) , pp. 657-663. 10.1016/j.jhep.2014.10.010

Publishers page: <http://dx.doi.org/10.1016/j.jhep.2014.10.010>

Please note:

Changes made as a result of publishing processes such as copy-editing, formatting and page numbers may not be reflected in this version. For the definitive version of this publication, please refer to the published source. You are advised to consult the publisher's version if you wish to cite this paper.

This version is being made available in accordance with publisher policies. See <http://orca.cf.ac.uk/policies.html> for usage policies. Copyright and moral rights for publications made available in ORCA are retained by the copyright holders.



**Circulating triacylglycerol signatures and insulin sensitivity in NAFLD associated with the E167K variant in TM6SF2**

You Zhou<sup>1\*</sup>, Gemma Llauradó<sup>1,2\*</sup>, Matej Oresic<sup>3</sup>, Tuulia Hyötyläinen<sup>3</sup>, Marju Orholm<sup>4</sup>, Hannele Yki-Järvinen<sup>1,5</sup>

<sup>1</sup>*Minerva Foundation Institute for Medical Research, Helsinki, Finland.*

<sup>2</sup>*CIBERDEM, Endocrinology Unit, Joan XXIII University Hospital, IISPV Pere Virgili Health Research Institute, Rovira i Virgili University, Tarragona, Spain*

<sup>3</sup>*Steno Diabetes Center, Niels Steensens Vej 2, 2820 Gentofte, Denmark*

<sup>4</sup>*Department of Clinical Sciences, Diabetes and Cardiovascular Disease Genetic Epidemiology, Skånes University Hospital, Lund University Diabetes Center, Lund University, Malmö, Sweden*

<sup>5</sup>*Department of Medicine, University of Helsinki, Helsinki, Finland*

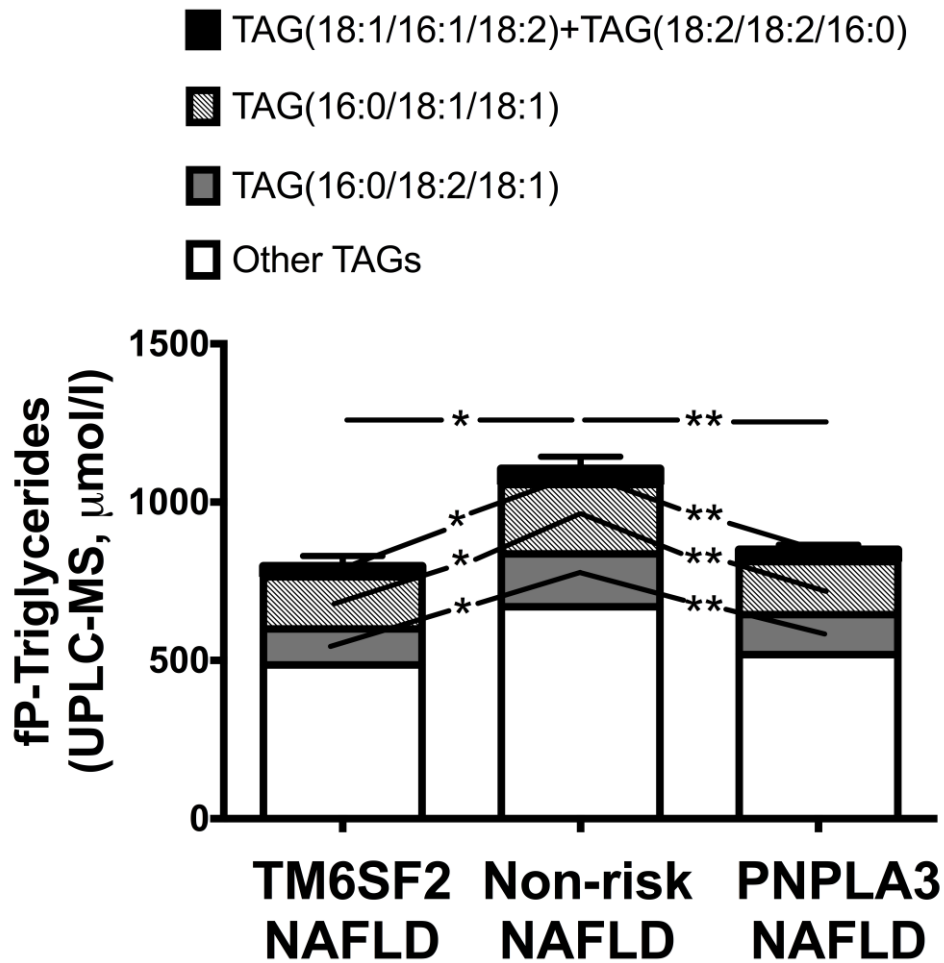
**Table of Contents**

Supplementary Methods	2
Supplementary Fig. 1.	3
Supplementary Table 1. Composition of circulating lipid clusters	4

## **Supplementary Methods**

### *Lipidomic analysis with UPLC-MS*

For further identification of unknown lipids, fractions collected from UPLC run were infused to a LTQ-Orbitrap (Thermo Fischer Scientific, San Jose, CA) mass spectrometer by a TriVersa Nanomate (Advion Biosciences, Ithaca, NY) using chip-based nanoelectrospray in positive and negative ionization mode. Identifications were based on the exact mass and MS<sup>n</sup> spectra. The instrument was calibrated externally according to the instructions of manufacturer. MS<sup>2</sup> and MS<sup>3</sup> were acquired using either low resolution or high resolution up to target mass resolution R = 60 000 at m/z 400. The normalized collision energies of 30-40% were applied in MS<sup>n</sup> experiments.



**Supplementary Fig. 1. Distribution of plasma TAG species in the ‘TM6SF2 NAFLD’, PNPLA3 NAFLD’ and ‘Non-risk NAFLD’ groups.** Total TAGs are shown as mean  $\pm$  SEM. The significant differences in concentrations of total TAGs, TAG(16:0/18:2/18:1), TAG(16:0/18:1/18:1), TAG(18:1/16:1/18:2)+TAG(18:2/18:2/16:0) are indicated (\* $p < 0.05$ , \*\* $p < 0.01$ ).

**Supplementary Table 1. Composition of circulating lipid clusters**

Cluster name	Size	Representative members
LC1	19	TAG(16:0/18:2/18:1); TAG(16:0/18:1/18:1); TAG(18:1/16:1/18:2)+TAG(18:2/18:2/16:0); TAG(18:1/18:2/18:1); TAG(18:1/18:1/18:1); TAG(14:0/18:1/18:1)+TAG(16:0/16:1/18:1); TAG(16:0/18:1/16:0); TAG(50:3); TAG(54:5); TAG(56:4); TAG(18:1/18:1/20:1)
LC2	22	PC(34:2); PC(36:2); PC(34:1); PC(36:3); PC(38:3); PC(36:1); PC(32:1); PC(38:6); PC(32:0); PC(40:6); PC(34:3); PE(38:1); PC(32:1); PC(38:4)
LC3	19	PC(36:4); PC(38:4); PC(38:6); TAG(56:7); TAG(56:6); PC(40:7); TAG(56:8); 68_TAG(58:8); TAG(58:9); TAG(56:5); PE(38:4)+PC(35:4); PC(38:4).TAG(54:6)
LC4	13	LysoPC(16:0); LysoPC(18:2); LysoPC(18:0); LysoPC(18:1); LysoPC(18:3); LysoPC(20:3); LysoPC(20:4); LysoPC(20:4); LysoPC(22:6); LysoPC(20:3); LysoPC(16:1)
LC5	24	SM(d18:1/24:1); SM(d18:1/16:0); SM(d18:1/22:0); SM(d18:1/24:0); SM(d18:1/22:1); 29_SM(d18:1/18:0); SM(d18:1/20:0); SM(d18:1/23:0); SM(d18:1/16:1); 56_PA(34:0); SM(d18:1/20:1); SM(d18:0/18:0)
LC6	19	TAG(16:0/18:0/18:1); TAG(14:0/16:0/18:1); TAG(46:1);

		TAG(49:1); TAG(16:0/16:0/18:0); TAG(14:0/16:0/16:0)+TAG(16:0/18:0/12:0); TAG(54:1); TAG(44:0); TAG(16:0/18:0/18:0); TAG(44:1); TAG(49:0)*; TAG(42:0); TAG(14:0/16:0/18:0)+TAG(16:0/16:0/16:0)
LC7	8	PC(38:7); PE(38:4); Cer(d18:1/22:6); PC(40:8); PE(40:7); PE(40:4); PC(38:5); PC(40:8)
LC8	16	PC(37:4)/PE(40:4); PC(35:2); PC(p16:0/18:2)+PC(34:3e); PC(34:1e)+PE(37:1e); PE(34:4e); PC(33:2)+PE(36:2); PC(31:1)+PE(34:1); PG(34:0e); PC(33:1)+PE(36:1); TAG(50:2)
LC9	19	PC(38:5e); PC(36:5e)+PE(38:5e); PC(40:7); PC(38:5e); PC(O- 24:1/20:4)+PC(44:5e); PE(38:5e); PE(38:3); PE(38:2); PE(38:3).1; PC(40:5e); PS(38:2); PE(40:6)