

AperTO - Archivio Istituzionale Open Access dell'Università di Torino

**Hepatic macrophage activation is associated with adipose tissue insulin resistance in non-diabetic patients with Non-Alcoholic Fatty Liver Disease**

**This is a pre print version of the following article:**

*Original Citation:*

*Availability:*

This version is available <http://hdl.handle.net/2318/1606917> since 2017-02-10T11:56:09Z

*Terms of use:*

Open Access

Anyone can freely access the full text of works made available as "Open Access". Works made available under a Creative Commons license can be used according to the terms and conditions of said license. Use of all other works requires consent of the right holder (author or publisher) if not exempted from copyright protection by the applicable law.

(Article begins on next page)

## **Hepatic macrophage activation is associated with adipose tissue insulin resistance in non-diabetic patients with Non-Alcoholic Fatty Liver Disease**

C. Rosso<sup>1</sup>, K. Kazankov<sup>2</sup>, M. Marietti<sup>1</sup>, M. Gaggini<sup>3</sup>, E. Buzzigoli<sup>3</sup>, H.J. Møller<sup>4</sup>, G.P. Caviglia<sup>1</sup>, M.L. Abate<sup>1</sup>, A. Smedile<sup>1</sup>, G.M. Saracco<sup>1</sup>, H. Vilstrup<sup>2</sup>, A. Gastaldelli<sup>3</sup>, J. George<sup>5</sup>, H. Grønbaek<sup>3</sup>, E. Bugianesi<sup>1</sup>

<sup>1</sup>*Division of Gastroenterology and Hepatology, Department of Medical Sciences, University of Torino, Torino, Italy;* <sup>2</sup>*Department of Hepatology and Gastroenterology, Aarhus University Hospital, Aarhus, Denmark;* <sup>3</sup>*Cardiometabolic Risk Unit, Institute of Clinical Physiology, CNR, Pisa, Italy;* <sup>4</sup>*Department of Clinical Biochemistry, Aarhus University Hospital, Aarhus, Denmark;* <sup>5</sup>*The Storr Liver Centre, University of Sydney and Westmead Hospital, Westmead, Australia*

**Background & Aim:** The onset and progression of liver damage in Non-Alcoholic Fatty Liver Disease (NAFLD) is tightly associated with insulin resistance (IR) in a dysfunctional adipose tissue (AT). Macrophage activation is a key step for both the chronic low inflammatory state of IR and for hepatic damage. The aim of this study is to elucidate the pathways linking IR in the AT, circulating and hepatic macrophage activation and liver damage in 40 non-diabetic patients with biopsy-proven NAFLD.

**Material and Methods.** [<sup>2</sup>H<sub>5</sub>]glycerol was infused in all study subjects to evaluate Glycerol-Ra and lipolysis. Adipose tissue-IR (AT-IR) was calculated as FFAs\*insulin(INS) (AT-IR1) and as Glycerol-Ra\*INS (AT-IR2). Soluble CD163 (sCD163), a marker of hepatic macrophage activation, was measured by an enzyme-linked immunosorbent assay (ELISA). CD163 mRNA expression in the liver was evaluated by qPCR using the CFX96 (Bio-Rad), SSoFast™ EvaGreen® Supermix (BioRad). Histology was scored according to Kleiner. Hepatic fat was assessed by liver biopsy. Visceral fat (VF) and subcutaneous fat (SF) were measured with standard nuclear magnetic resonance (NMR).

**Results.** AT-IR showed significant associations with features of liver damage at liver biopsy, including hepatic fat (AT-IR1: r=0.50, p=0.001; AT-IR2: r=0.44, p=0.004), NAS score (r=0.43, p=0.006 and r=0.31, p=0.05 respectively) and fibrosis (AT-IR1: r=0.51 and AT-IR2: r=0.34, p=0.001 for both). Plasma levels of sCD163 were significantly associated with fasting plasma levels of FFAs (r=0.35, p=0.026), with lipolysis (r =0.35, p=0.028) and with AT-IR (AT-IR1 r=0.38, p=0.016 and AT-IR2 r=0.31, p=0.005). Circulating sCD163 increased proportionally to liver fat (r=0.53; p=0.005) but not to visceral or subcutaneous fat (p=NS for both). The hepatic expression of CD163 (n=20) had a linear correlation with plasma levels of sCD163 (r=0.44, p=0.05). NAFLD subjects with more than two-fold hepatic expression of CD163 had significantly higher hepatic fat content (p=0.028). The hepatic expression of CD163 was higher than two-fold in 75% of NAFLD patients with moderate (F2) and in 100% of those with severe fibrosis (F3).

**Conclusions.** We speculate that in NAFLD patients hepatic macrophages activation can be directly stimulated by an increased flux of FFA due to AT-IR, thus directly linking IR, dysfunctional adipose tissue and liver damage.

*Funded by: FP7/2007-2013 under grant agreement no. HEALTH-F2-2009-241762, FLIP; PRIN2009ARYX4T; Horizon2020 under grant agreement no.634413, EPoS*

**Keywords.** NAFLD, Adipose tissue insulin resistance, sCD163, macrophage, fibrosis