TITLE: PBMC from a subpopulation of celiac patients secrete CXCR3-dependent IL-8 in response to gliadin

AUTHORS: Lammers, Karen [1]; Khandelwal, Sunaina [1]; Kryszak, Debby [1]; Vincenzo, Casolaro [2, 1]; Fasano, Alessio [1]

INSTITUTIONS: 1. Mucosal Biology Research Center, University of Maryland School of Medicine, Baltimore, MD, USA.
2. Asthma & Allergy Center, Johns Hopkins University School of Medicine, Baltimore, MD, USA.

ABSTRACT: We have recently reported the identification and characterization of the chemokine receptor, CXCR3, as a receptor for specific α-gliadin epitopes. Gliadin binding to CXCR3 induces a MyD88-dependent activation of the zonulin pathway and a subsequent increase in intestinal permeability.

Aim: To further explore the role of CXCR3 in the immune response provoked by gliadin in peripheral blood mononuclear cells (PBMC) from healthy donors (HD) and celiac disease (CD) patients.

Methods: PBMC from 21 CD patients on a gluten-free diet and 10 HD were incubated with 1 mg/mL pepsin/trypsin-digested gliadin (PTG) for 24 hours, in the presence or absence of blocking anti-CXCR3 monoclonal antibody (10 µg/mL) or an appropriate isotype control (10 µg/mL). Supernatants were analyzed for their content in IL-6, IL-8, IL-10, TNFα and IFNγ.

Results: All cytokines were produced at higher level in CD patients compared to controls, and production of IL-6, IL-10, TNFα and IFNγ was not influenced by blocking of the CXCR3 chemokine receptor. Conversely, gliadin induced IL-8 production only in a subgroup of individuals, namely 30% of healthy controls and 43% of CD patients. Interestingly, gliadin-induced IL-8 secretion was abrogated when CXCR3 was blocked prior to gliadin stimulation in the CD group, but not in the control group. See Table below:

Conclusions: PBMC from CD patients respond to gliadin with cytokine production at higher level than in controls. A subgroup of individuals responded to gliadin with the production of IL-8 that is CXCR3-dependent only in CD patients but not in healthy controls.

	Non-Responders			Responders		
Group	Medium	PTG	Group	Medium	PTG	αCXCR3+PTG
HD (N=7)	1.4+/-0.1*	2.7+/-0.1	HD (N=3)	0.3+/-0.2	98.8+/-15.2	82.6+/-11.5
CD (N=12)	4+/-2.2	22+/-0.2	CD (N=9)	2.2+/-0.2	179.0+/-30.2	2.7+/-0.1**

*IL-8 values are expressed as ng IL-8/1x10^6PBMC
**P<0.001 compared to responders exposed to PTG