

Increased Expression of LRRK2, a Susceptibility Gene of IBD Results in Enhanced Pro-Inflammatory Response and Severe Experimental Colitis

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Disclosure

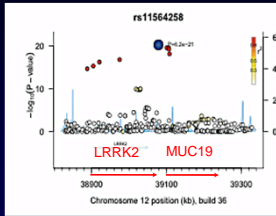
Patent: Janssen Pharmaceuticals:
The use of anti-IL-12p40 mAb for the treatment of Crohn's disease patients

LRRK2/MUC19 Locus has the Second Strongest Association with IBD in GWAS Studies

Crohn's disease		Ulcerative colitis		Total IBD	
Gene	Odds Ratio	Gene	Odds Ratio	Gene	Odds Ratio
1. IL23R	2.371	1. IL23R	1.682	1. IL23R	2.013
2. Nod2	1.557	2. HLA region	1.444	2. LRRK2/MUC19	1.334
3. LRRK2/MUC19	1.463	3. IL-10	1.277	3. Nod2	1.295
4. IRGM	1.324	⋮	⋮	4. IRGM	1.249
5. PTGER4	1.294	⋮	⋮	5. TAB1	1.209
6. TAB1	1.247	⋮	⋮	⋮	⋮
7. ATG16L1	1.233	17. LRRK2/MUC19	1.163	⋮	⋮
⋮	⋮	⋮	⋮	⋮	⋮

Jostins L. et al. 2012 Nature

Many SNPs are associated with LRRK2 gene region

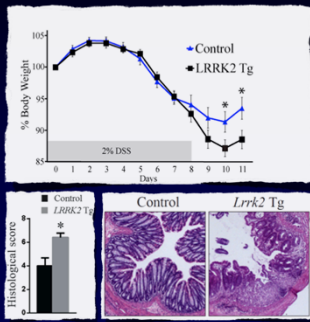


rs11564258 is located downstream of LRRK2 gene and in intron1 of MUC19 gene

Gardet et al., 2010 J Immunol

Franke et al., 2010 Nat Genet

LRRK2 Transgenic Mice Exhibit Severe Experimental Colitis



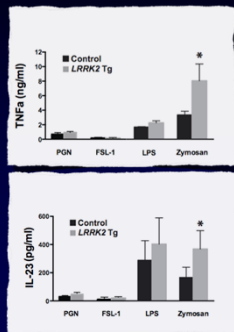
Methods

- Lrrk2 transgenic mice (Lrrk2 Tg)
- 2% Dextran Sodium Sulfate (DSS)-induces colitis

Results

- Lrrk2 Tg mice show more severe colitis compared to Wt littermate controls.

Only Zymosan Stimulation Induces Significant Increases in Cytokines by LRRK2 Tg Mice



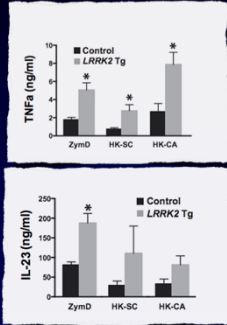
Methods

- Bone marrow-derived dendritic cells (BMDC)
- in vitro Stimulation with TLR ligands
- ELISA

Results

- LPS (TLR4), PGN and FSL-1 (TLR2) stimulation didn't show significant difference
- Zymosan which can stimulate Dectin-1 and TLR2 shows significant increase in Lrrk2 Tg

LRRK2 Positively Regulates Dectin-1, an innate immune Receptor of β -glucans



Methods

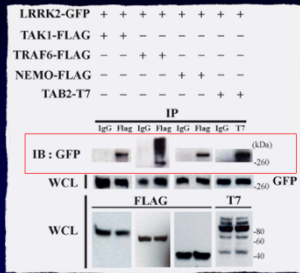
- BMDC in vitro Stimulation with Dectin-1 ligands Zymosan depleted (ZymD) Heat-killed *S. Cerevisiae* (HK-SC) Heat-killed *Candida albicans* (HK-CA)

- ELISA

Results

- Dectin-1 agonists induced increased response in *Lrrk2* Tg

LRRK2 Interacts with TAK1 Complex



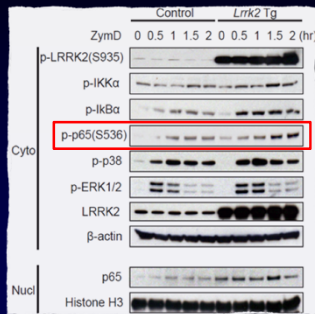
Methods

- HEK293T cells were transfected with LRRK2 and TAK1, TRAF6, NEMO or TAB2 vector
- Immunoprecipitation and Western Blotting

Results

- LRRK2 binds to TAK1, TRAF6, NEMO and TAB2

LRRK2 Activates NF- κ B and MAP Kinase Signaling upon Dectin-1 Stimulation



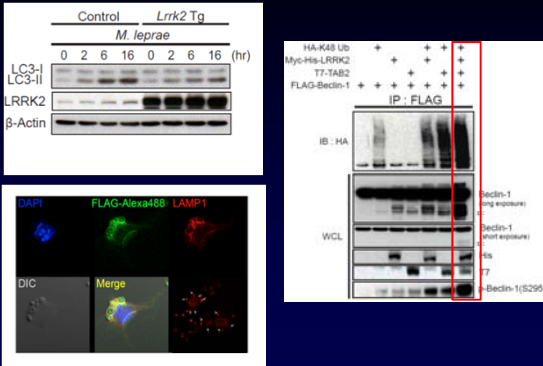
Methods

- BMDC
- in vitro Stimulation with ZymD
- Western Blotting

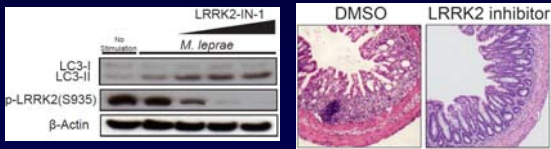
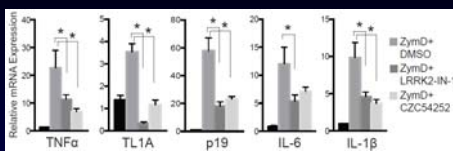
Results

- BMDC from *Lrrk2* Tg show increased NF- κ B and MAP kinase in response to ZymD.

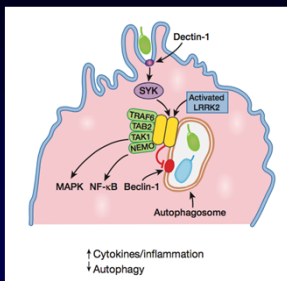
LRRK2 Causes Decreased Autophagy through Inhibition of Beclin-1



LRRK2 Inhibition Results in Amelioration of Inflammation



Summary



- Lrrk2 Tg mice display increased severity of colitis.
- LRRK2 thru Dectin-1 signaling interacts with TAK1 complex and activates NF-κβ signaling.
- LRRK2 can bind and inactivate Beclin-1 resulting in decreased Autophagy.
- Inhibitors of LRRK2 can ameliorate experimental inflammation.

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Warren Strober
