

Supplementary Appendix

This appendix has been provided by the authors to give readers additional information about their work.

Supplement to: Goldbach-Mansky R, Dailey NJ, Canna SW, et al. Neonatal-onset multisystem inflammatory disease responsive to interleukin-1 β inhibition. N Engl J Med 2006;355:581-92.

Supplementary Table 1. Changes in Bone Conduction (Sensorineural Threshold) after 3 and 6 Months of Treatment with Anakinra

Measure[†]	Hearing Changes (n= 18)	
Audiogram*	Mo 3	Mo 6
No change	10 (56)	9 (50)
Improved	6 (33)	6 (33)
Diminished	0 (0)	0 (0)
Both [‡]	0 (0)	1 (6)
Incomplete	2 (11)	2 (11)
TOTAL	18	18

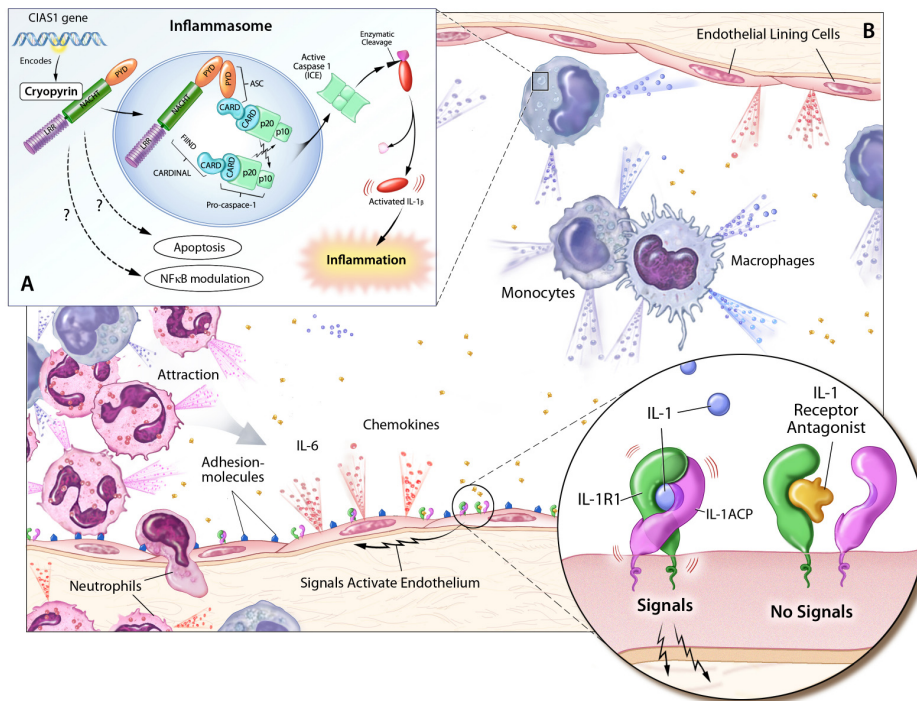
[†] Changes in hearing are based on pure tone thresholds at 500, 1000, 2000 and 4000 Hz or Speech Awareness Thresholds when reliable pure tone thresholds could not be established. Improvement is defined as an increase of 20 dB in hearing threshold at one or more frequencies, or as an increase of 10 dB in hearing threshold at two or more consecutive frequencies (ASHA, 1994). No change in either ear is recorded as “stable”, change in one or both ears recorded as appropriate change (improved or diminished). Bone conduction data sets were incomplete on 2 patients (4 ears) due to inability to establish reliable behavioral responses.

*Measures represent absolute patient numbers (percent)

[‡] One of the ears in one patient had improved hearing at 2000 & 4000 Hz and diminished hearing at 500 and 1000 Hz.

Ref.: ASHA (1994) Guidelines for the audiologic management of individuals receiving ototoxic drug therapy. Definition of ototoxicity-induced hearing loss. ASHA 36 (Suppl. 12), 11-19

Supplementary Figure 1. Mechanism of IL-1 β Activation and Blockade by IL-1 Receptor Antagonist.



Panel A shows interleukin-1 β (IL-1 β) activation through upstream inflammasome assembly:

The inflammasome is a molecular complex of several proteins that, upon assembly, activates caspase-1 (also called IL-1 converting enzyme (ICE)). The association of cryopyrin (also called NALP3, PYPAF1, CATERPILLER 1.1) and ASC through homotypic interaction of the 2 pyrin domains (PYD) on one side and cryopyrin and CARDINAL through NACHT domain-FIIND domain interaction on the other side allows the recruitment of 2 CARD domain containing caspase-1 molecules. The close proximity of the 2 pro-caspase-1 molecules results in their autoactivation and the activated complex can then cleave the inactive form of pro-IL1 β into its bioactive form, a potent cytokine causing inflammation. The inflammasome can assemble in monocytes and macrophages. These cells can produce and release IL-1 β .

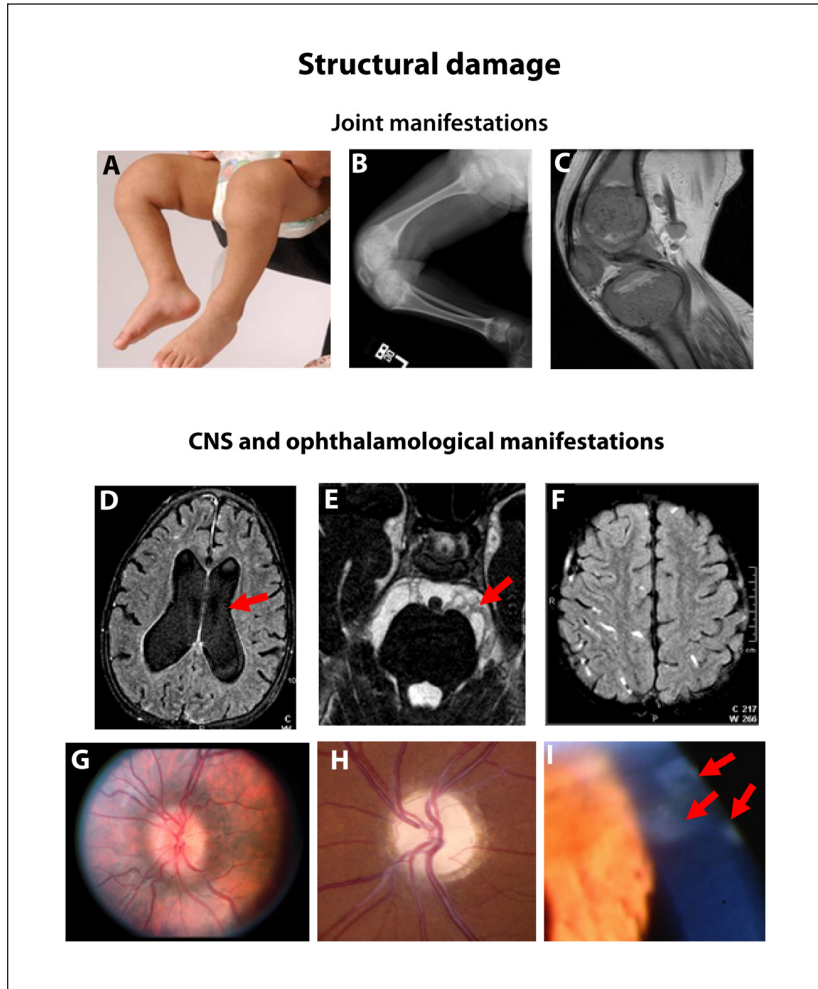
Panel B shows downstream IL-1 β induced mechanisms of inflammation:

Endothelial cells express IL-1 receptor complexes. The IL-1 Type 1 receptor (IL-1R1) binds IL-1 β and then interacts with the IL-1 receptor accessory protein (IL-1AcP) to form an active heterodimeric signaling complex. Endogenous or exogenous IL-1 receptor antagonist (anakinra) can bind to the IL-1 receptor and prevent assembly of the signaling complex. Signaling through the IL-1 receptor complex can activate downstream inflammatory processes in the adjacent endothelial cells, including the production of adhesion molecules, chemokines, and IL-6.

Abbreviations:

PYD	pyrin domain
LRR	leucine-rich repeat domain
FIIND	domain with function to find
NACHT	domain present in NAIP, CIITA, HET-E, and TP1
ICE	interleukin-1 β converting enzyme
ASC	apoptosis-associated specklike protein with a CARD domain
CARD	caspase recruitment domain
CARDINAL	CARD- inhibitor of NF-kappaB- activating ligand
IL-1R1	IL-1 receptor Type 1
IL-1AcP	IL-1 receptor accessory protein

Supplementary Figure 2. Structural Damage in NOMID.

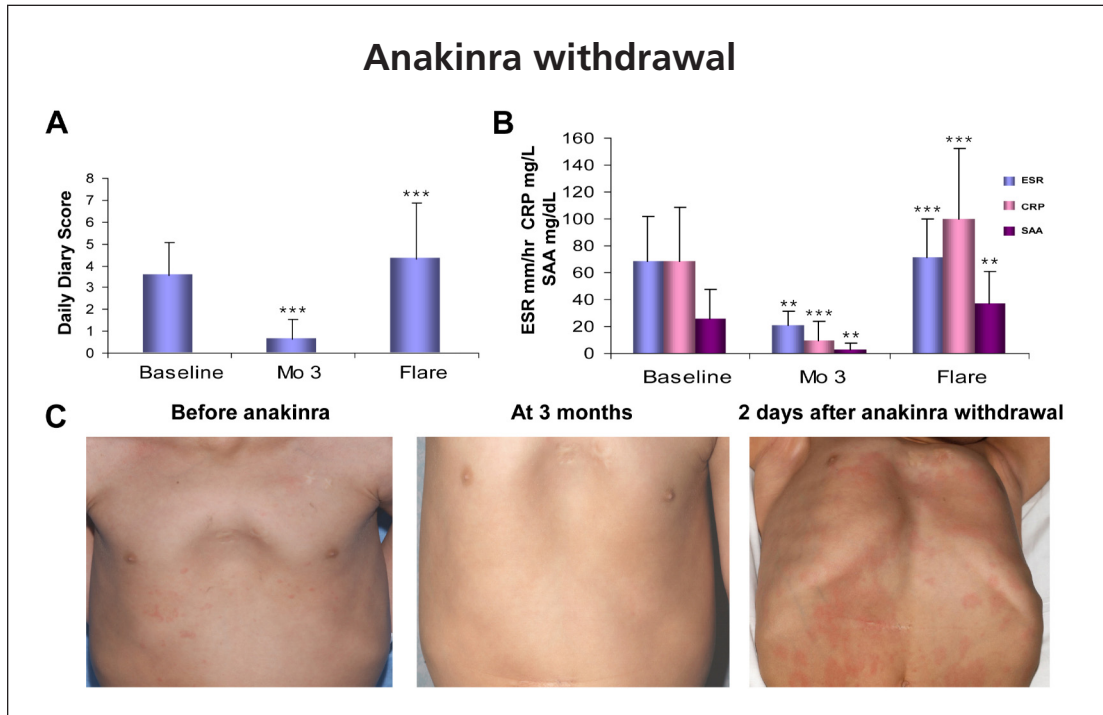


Panels A-C show baseline bony overgrowth of the same knee, on photograph, x-ray and MRI.

Panels D-F show ventriculomegaly, arachnoid adhesions and dural enhancement on contrast enhanced MRI images.

Panels G-I show papilledema, optic nerve atrophy and corneal opacification.

Supplementary Figure 3. Disease Flare after Intentional Anakinra Withdrawal.



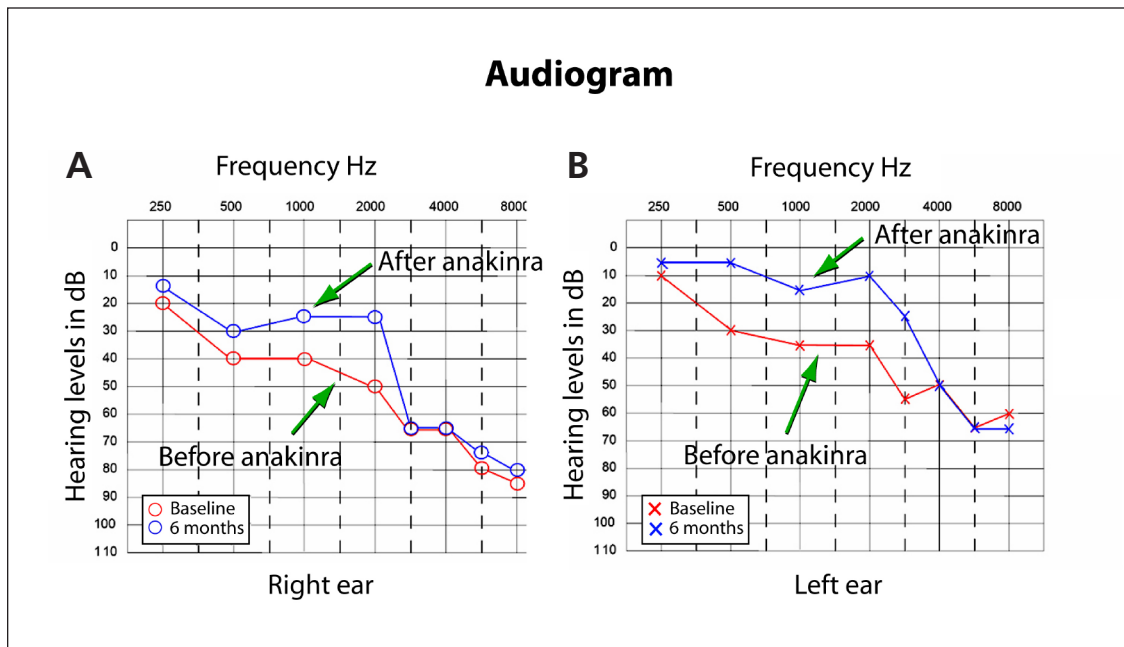
Panel A shows the mean baseline daily NOMID specific diary score before therapy, at 3 months after therapy, and after 2-7 days of intentional anakinra withdrawal in 11 patients with NOMID.

Panel B shows mean levels of ESR, CRP and SAA levels at baseline, at 3 months after anakinra therapy, and after 2-7 days after anakinra withdrawal in 11 patients with NOMID.

Panel C shows pictures of exaggerated rash after anakinra withdrawal compared to baseline, and 3 months post therapy with anakinra.

p<0.01, *p<0.001

Supplementary Figure 4. Audiogram Showing Hearing Improvement post Anakinra Therapy.



Panels A and B show audiograms of the right and left ear respectively of one representative patient. The red line indicates bone threshold hearing levels before initiation of treatment with anakinra, the blue line indicates improvement over several frequencies at 3 months after treatment initiation.