Bioscience
Innovation for Health and Security

Beryllium Alters Lipopolysaccharide-Mediated Intracellular Phosphorylation and Cytokine Release in Human Peripheral Blood Mononuclear Cells

Anu Chaudhary, Ph.D.

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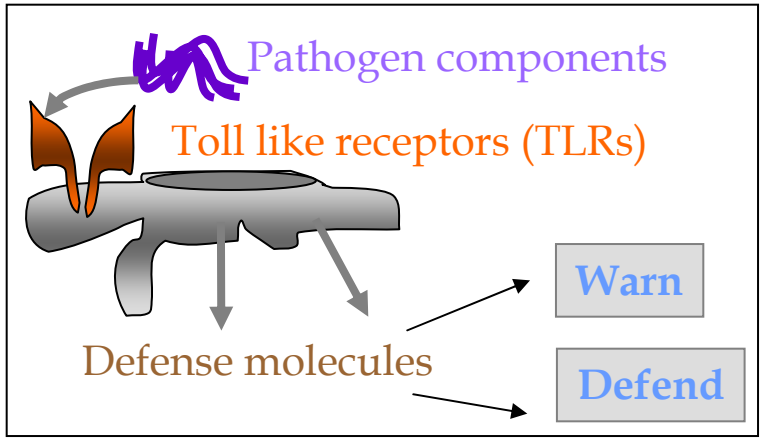
Hypothesis

Beryllium exposure may alter the innate immune response to bacterial components such as endotoxin

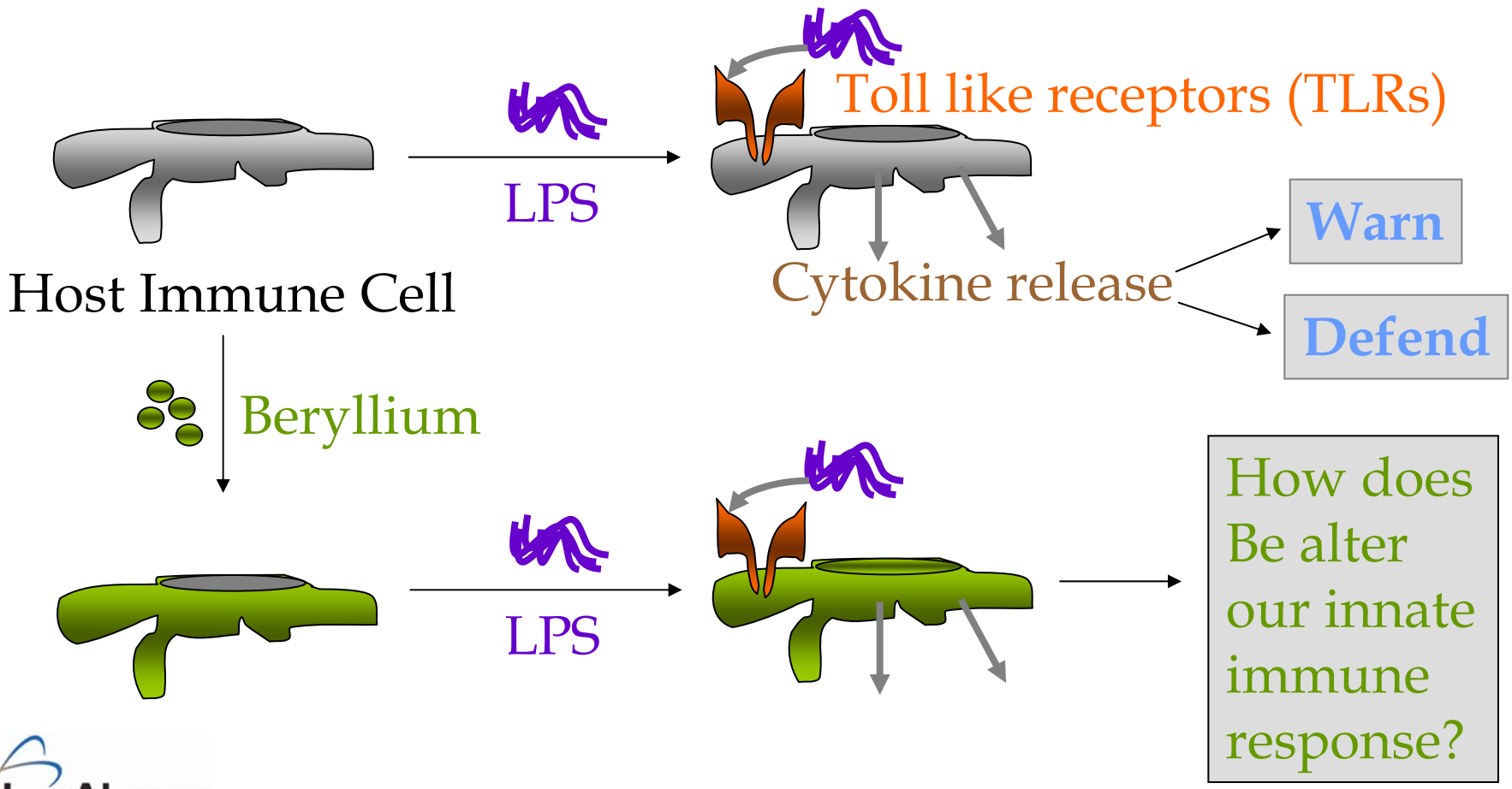


- Innate immunity implicated in host response to atmospheric pollutants *via* Toll-like receptors.
- ROFA (transition metals, particle associated microbial matter) mediates responses *via* TLRs 2 and 4
- Interplay between microbes and particulates

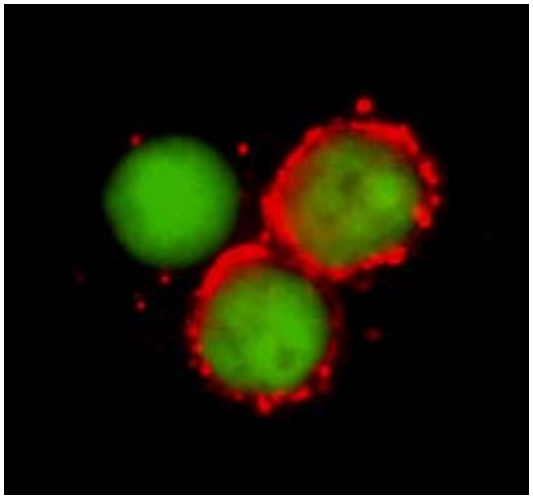
Various Metals Impact Host Immunity

- Lead chloride strongly enhances susceptibility of rodents to endotoxin shock
- Nickel chloride amplifies LPS-triggered IL6 and IL1β release by activating Nrf2 antioxidants
- Mercury potentiates LPS-mediated TNF expression, decreases IL6, NE on IL1β
- Zinc is protective against LPS induced lethality in mice
- Mice sensitized with Ni and LPS later react to Ni, Cr, Pd and Ag

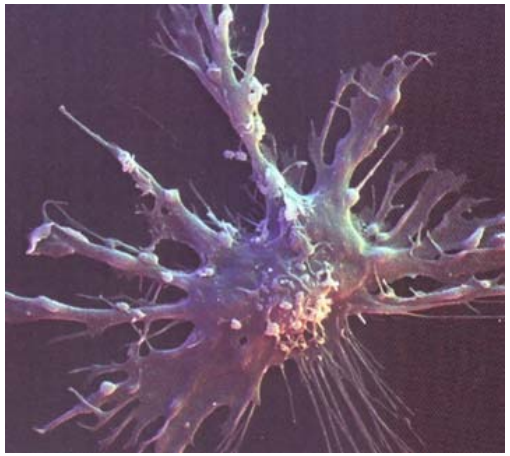
Experimental Design



Cell Model

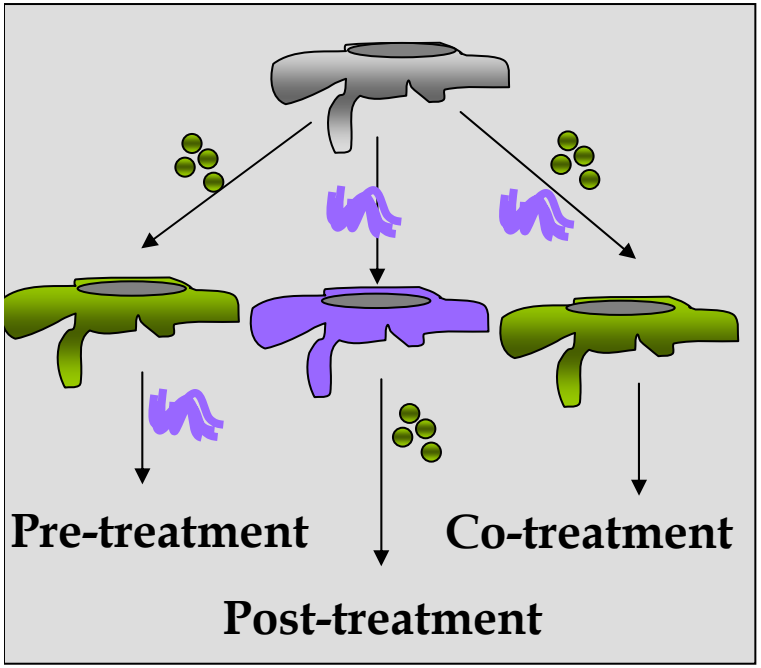


PBMC+ DC (20:1)
70% T cells
Monocytes, B cells
Dendritic cells (DC) from same donor



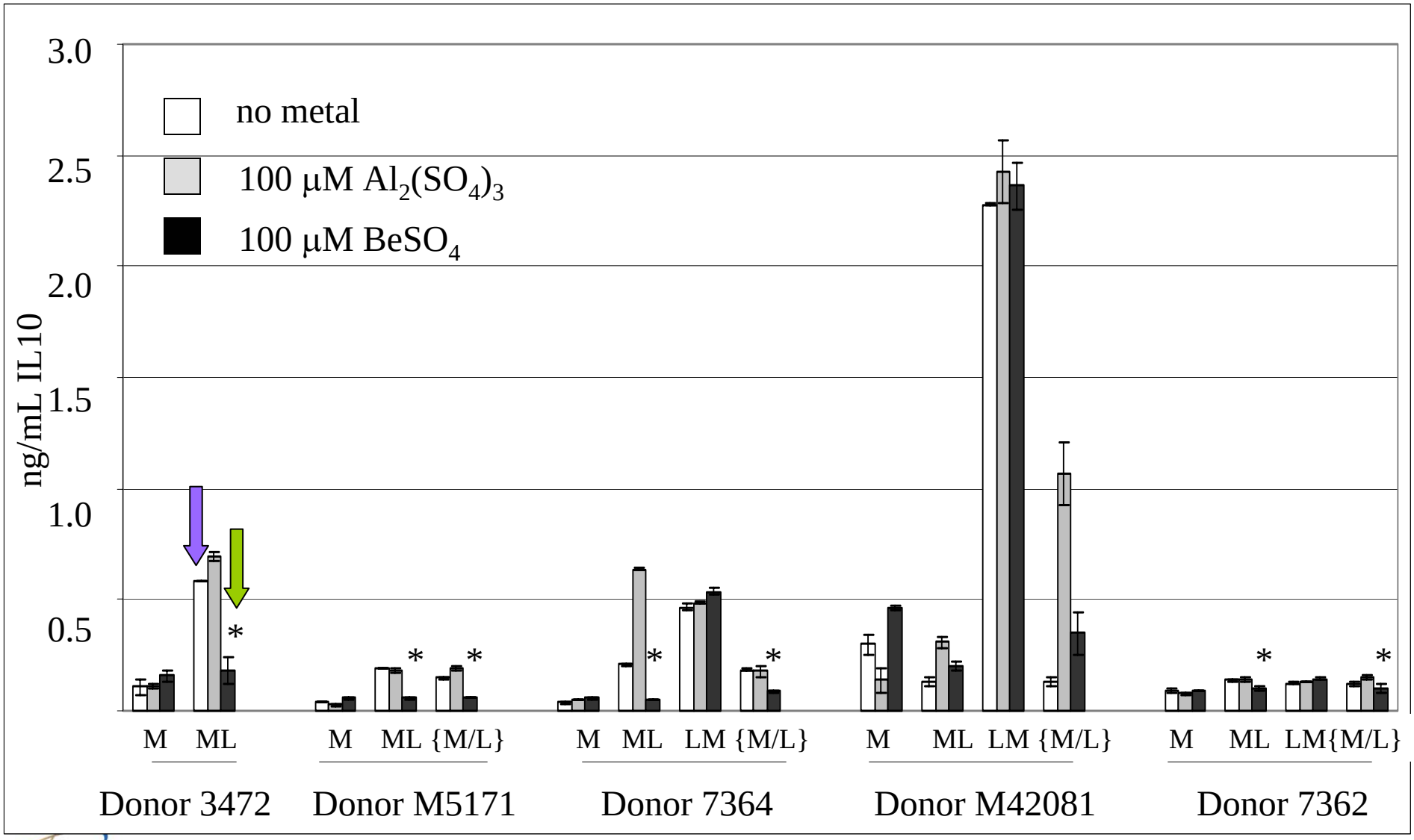
- ✓ Primary cells.
- ✓ Target cells for CBD are alveolar macrophages and WBCs
- ✓ Combining PBMC with APC (DC) from WBC of the same donor, one can elicit immune response *in vitro*.
- ✓ Maier *et al* have shown that PBMCs from CBD subjects mimic the Be-stimulated cytokine production. (*Eur Respir J*. 2001; 17:403-15).

Beryllium Induces IL1β Release in Cells Stimulated with LPS



IL1β mean pg/mL ± SD	Donor 3472	Donor M5171	Donor 7364	Donor M42081
control	65 ± 16.4	28 ± 5.5	16.3 ± 0.04	79 ± 10.4
$Al_2(SO_4)_3$	69 ± 7.5	33 ± 0.9	17.12 ± 1.2	95 ± 4.9
$BeSO_4$	82 ± 9.7	42 ± 1.6	16 ± 1.2	62 ± 6.7
Control → LPS	436 ± 0	48 ± 0.7	106 ± 2.5	50 ± 8.9
$Al_2(SO_4)_3$ → LPS	138 ± 5.2	43 ± 1.7	66 ± 0.44	88 ± 12.8
$BeSO_4$ →LPS	524 ± 4.2*	75 ± 0.8*	155 ± 10.7*	86 ± 0.07*
LPS → Control	802 ± 10.8	222 + 1.12	424 ± 1.08	301.67 ± 1
LPS → $Al_2(SO_4)_3$	799 ± 15.5	213 + 5.24	412 ± 1.25	322.9 ± 4.7
LPS → $BeSO_4$	820 ± 21.03	208 + 6.32	422 ± 2.911	303.5 ± 5.08
{LPS/Control }	444 ± 35.9	150.95 ± 3.9	106 ± 2.5	50 ± 9
{LPS/$Al_2(SO_4)_3$}	402 ± 15.12	201 ± 2.2	115 ± 10.3	274 ± 1.13
{LPS/$BeSO_4$}	485 ± 20.91*	256 ± 5.8*	335 ± 7.13*	107 ± 1.9*

Beryllium sulfate inhibits LPS- mediated IL10 release

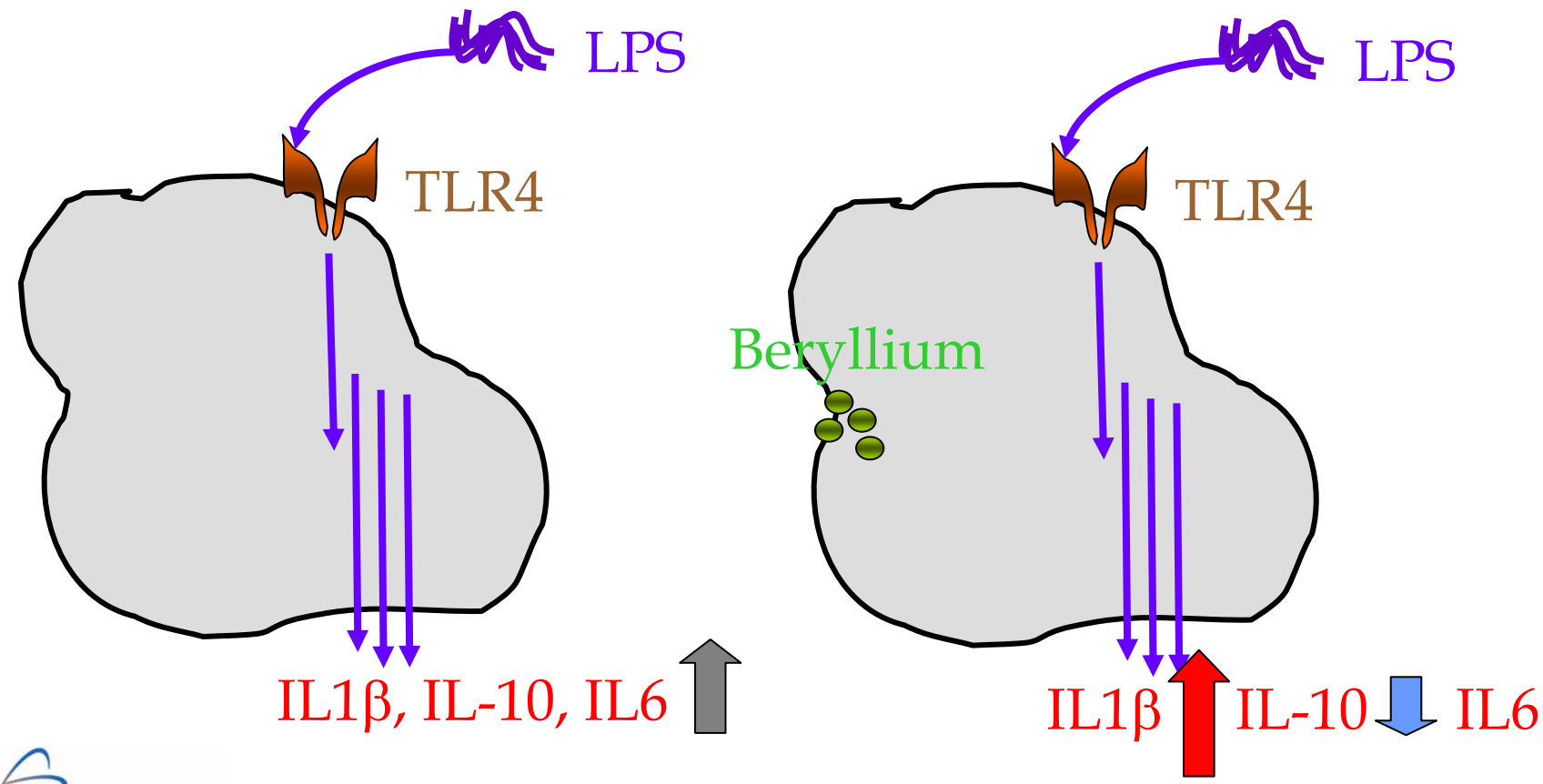


There is No Effect on LPS-Mediated IL6 Release in Cells Stimulated with $BeSO_4$

Beryllium does not sequester LPS

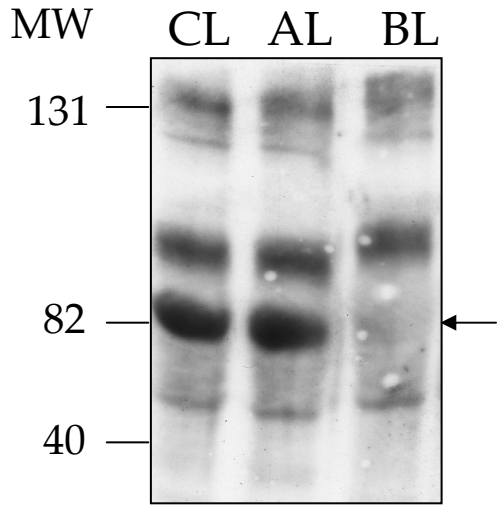
IL6 mean pg/mL ± SD	Donor M5171	Donor 7364	Donor M42081
control	81 ± 3.2	166 ± 9	62 ± 4.6
$Al_2(SO_4)_3$	87 ± 5.2	117 ± 1.6	118 ± 14.4
$BeSO_4$	67 ± 2*	51.8 ± 2.9*	40 ± 1.9*
Control → LPS	412 ± 3.15	669 ± 0	90.5 ± 8.7
$Al_2(SO_4)_3$ → LPS	436 ± 0.7	669 ± 0	244 ± 17.8
$BeSO_4$ → LPS	405 ± 9	669 ± 40	335 ± 22.6
LPS → Control	436 ± 0	669 ± 0	385 ± 2
LPS → $Al_2(SO_4)_3$	436 ± 0	669 ± 0	463 ± 1.3
LPS → $BeSO_4$	436 ± 0	669 ± 0	347 ± 21.1
{LPS/Control }	436 ± 0	669 ± 0	90.5 ± 8.7
{LPS/$Al_2(SO_4)_3$}	436 ± 0	669 ± 0	353 ± 17.3
{LPS/ $BeSO_4$}	436 ± 0	669 ± 0	398 ± 22.3

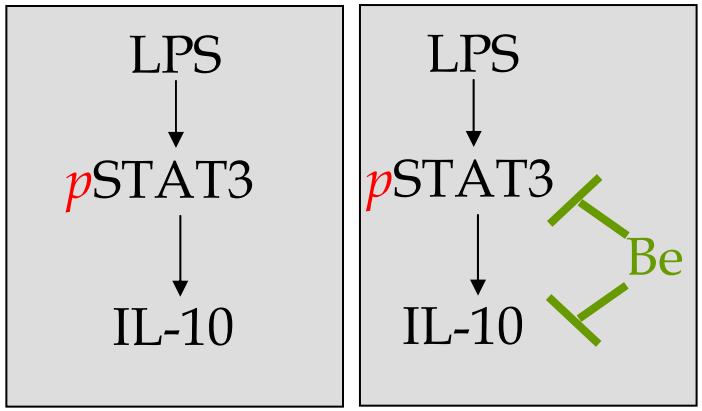
Cytokine Release: Summary

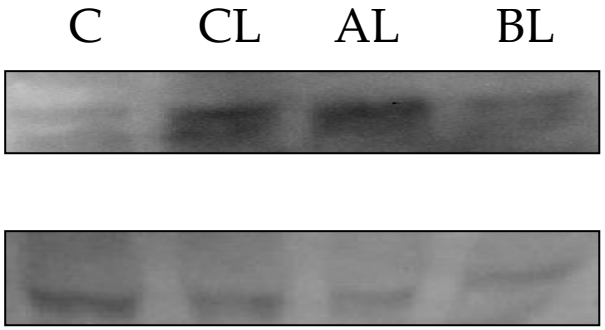


Tyrosine Phosphorylation Mediated Intracellular Signaling Is Altered in Cells Treated with $BeSO_4$

WCL
IB: αpY99 (pTyr)



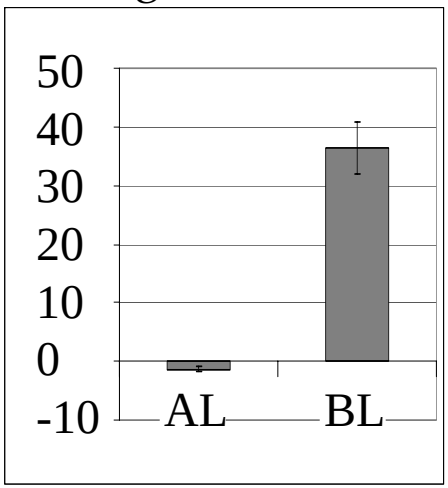




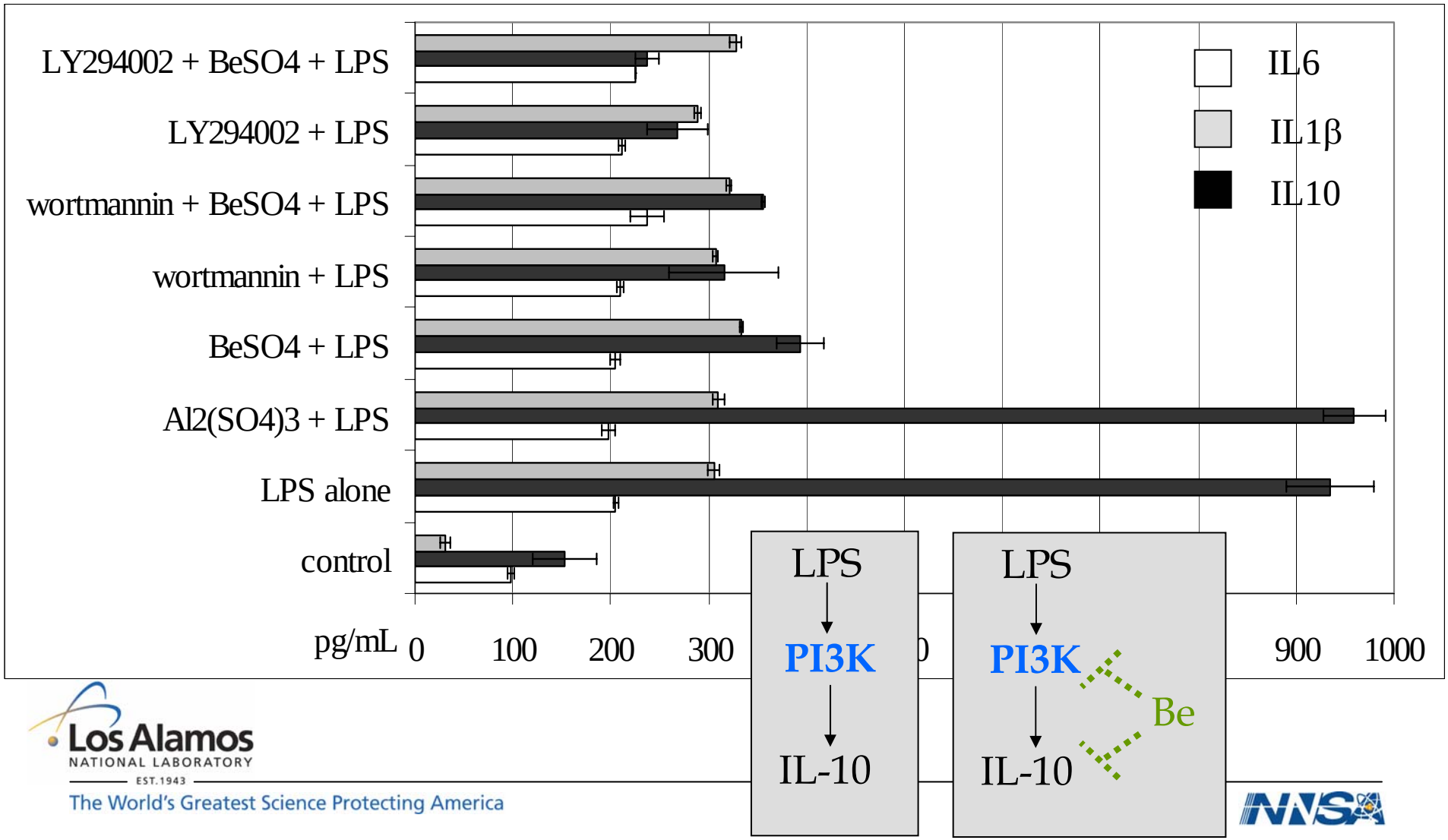
WCL
IB: αpSTAT3(pY705)

WCL
IB: αactin

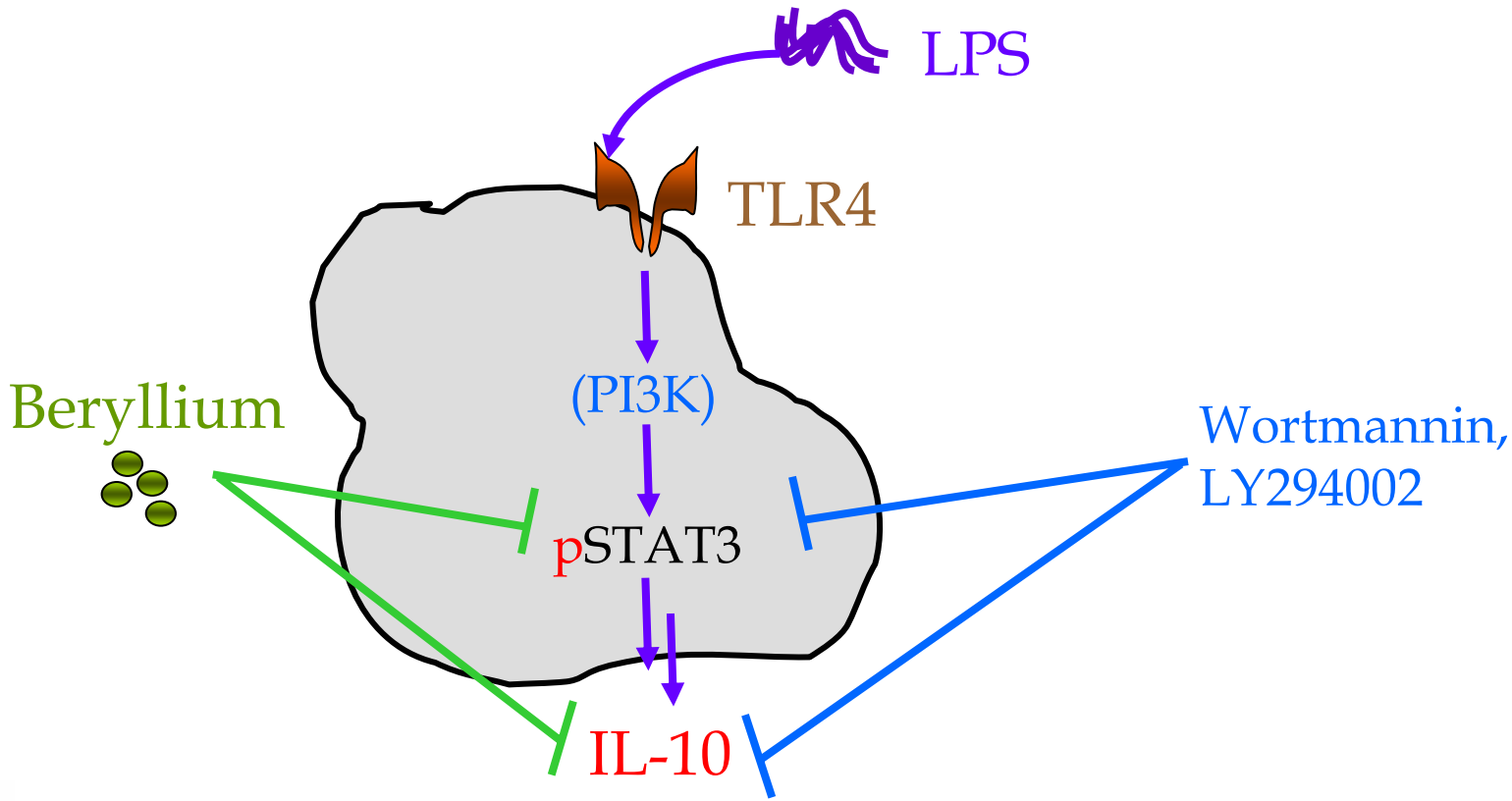
Average % inhibition



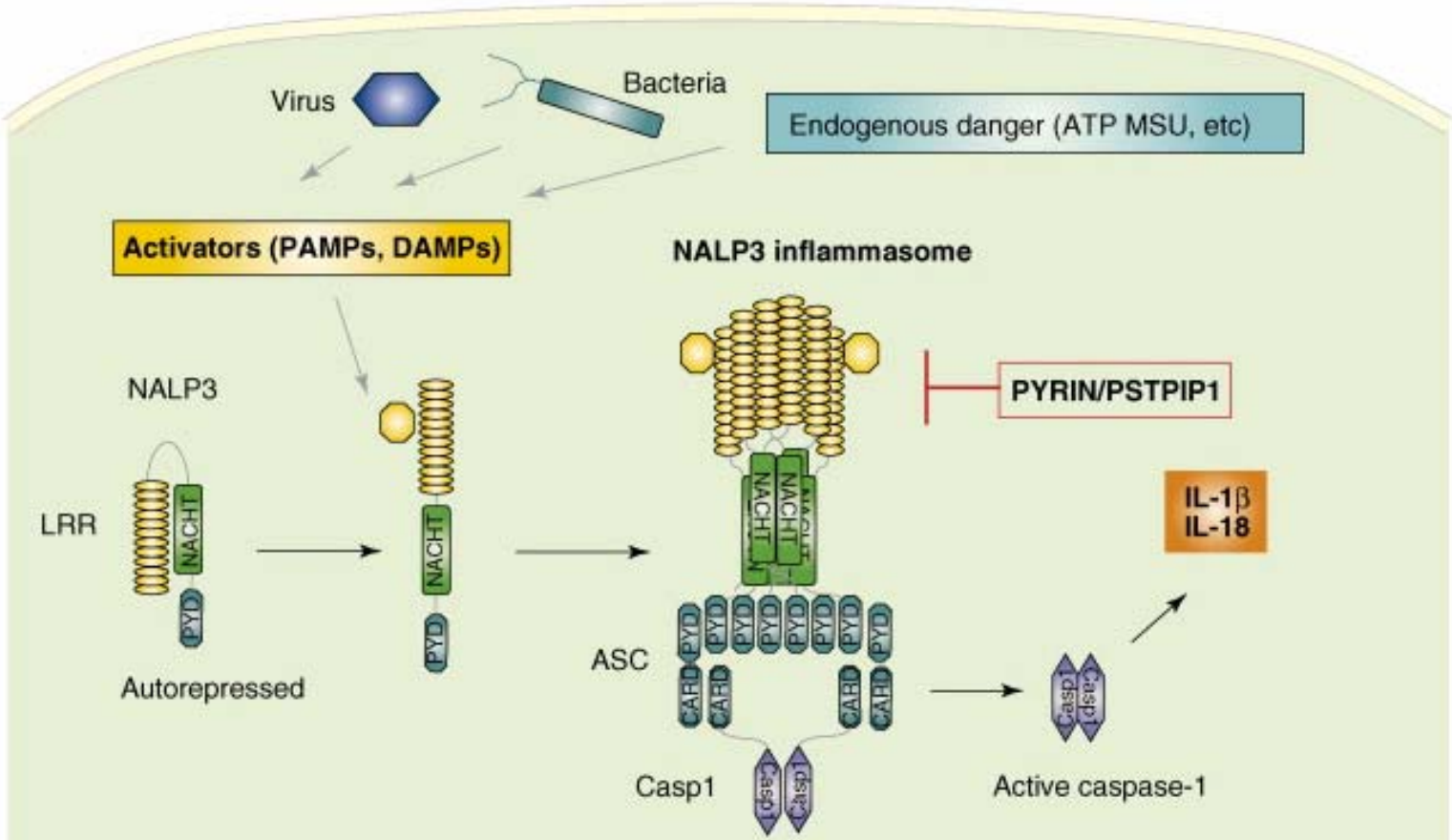
Inhibitors of PI3K Inhibit LPS Mediated IL10 Release but have no Effect on IL1β or IL6.



Model for Inhibition of IL10 Release



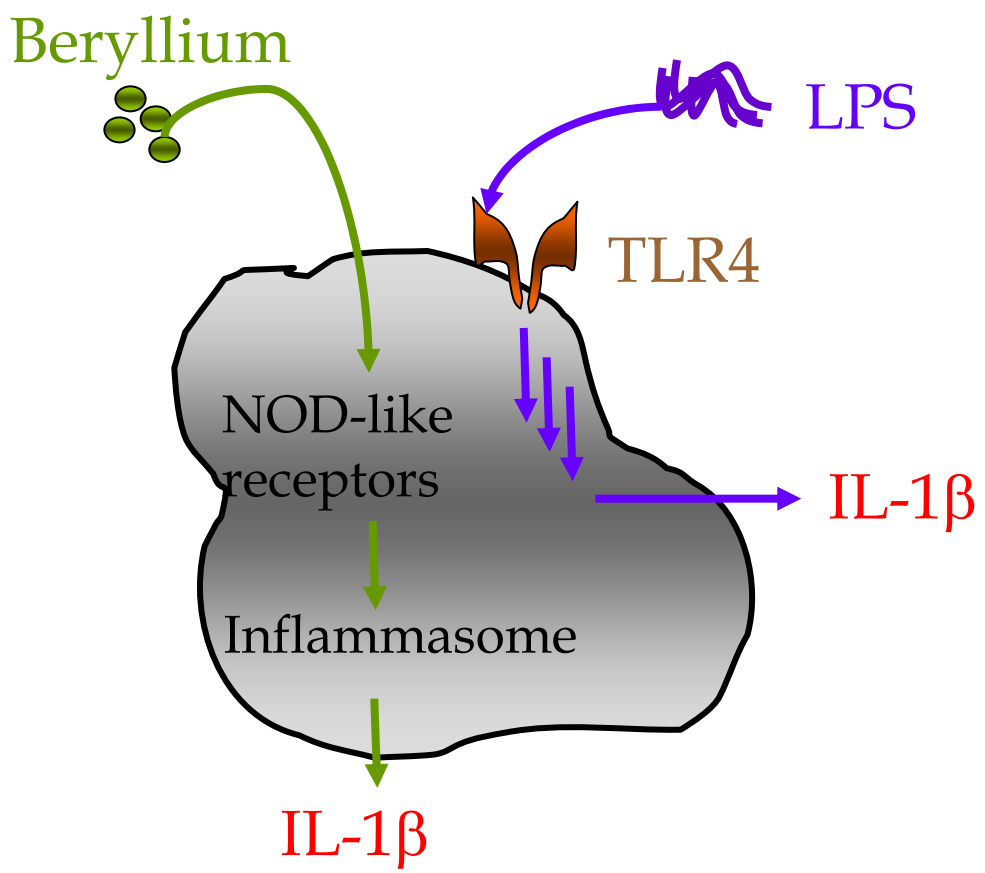
Processing of IL1β Through NALP3 Inflammasome



McDermott et al, Trends in Molecular Medicine 13(9) 2007, 381-388

TRENDS in Molecular Medicine

Potential Mechanism for the Induction of LPS-Stimulated IL1β Release



Summary & Future Directions

- Beryllium alters the innate immune response
- LPS-mediated IL10 is inhibited and IL1β is induced
- It is likely that Beryllium triggers the inflammasome to stimulate IL1β release from cells
- Mechanistically,
  - Beryllium inhibits LPS-mediated pSTAT3
  - Beryllium mimics effects of inhibition of PI3K

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