

# Clinical efficacy of different BCG strains in the treatment of NMIBC- myth or reality

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# Conflicts of Interest

- Sanofi, Merck, Janssen, Ferring, AstraZeneca, Verity

1. Why while millions of patients have been treated with BCG for prevention of NMIBC no clinical difference has been shown among studies despite the use of various strains worldwide ?

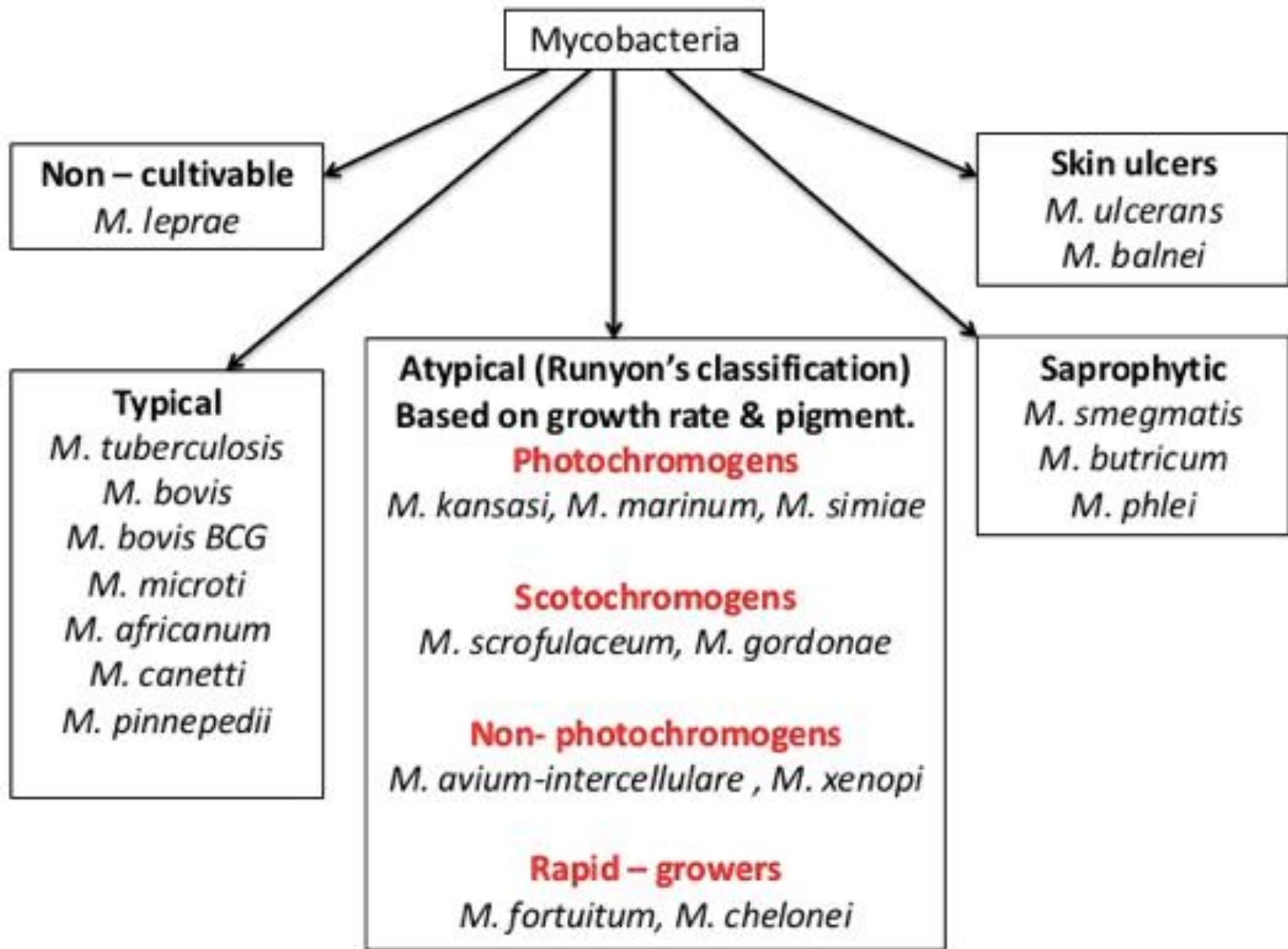
2. What can we learn from BCG used as an anti-tuberculosis vaccine  
?

3. How can we explain the absence of difference in clinical efficacy for BCG in NMIBC throughout the globe ? An insight into its mechanism of action

**Tan GH, Kuk C, Zlotta AR.** Are there differences among Bacillus Calmette-Guérin (BCG) strains regarding their clinical efficacy in the treatment of non-muscle invasive bladder cancer? The jury is still out but the answer is likely no.

Can Urol Assoc J. 2019 Jul 23. doi:  
10.5489/cuaj.5923.





# History of BCG: Bladder Cancer

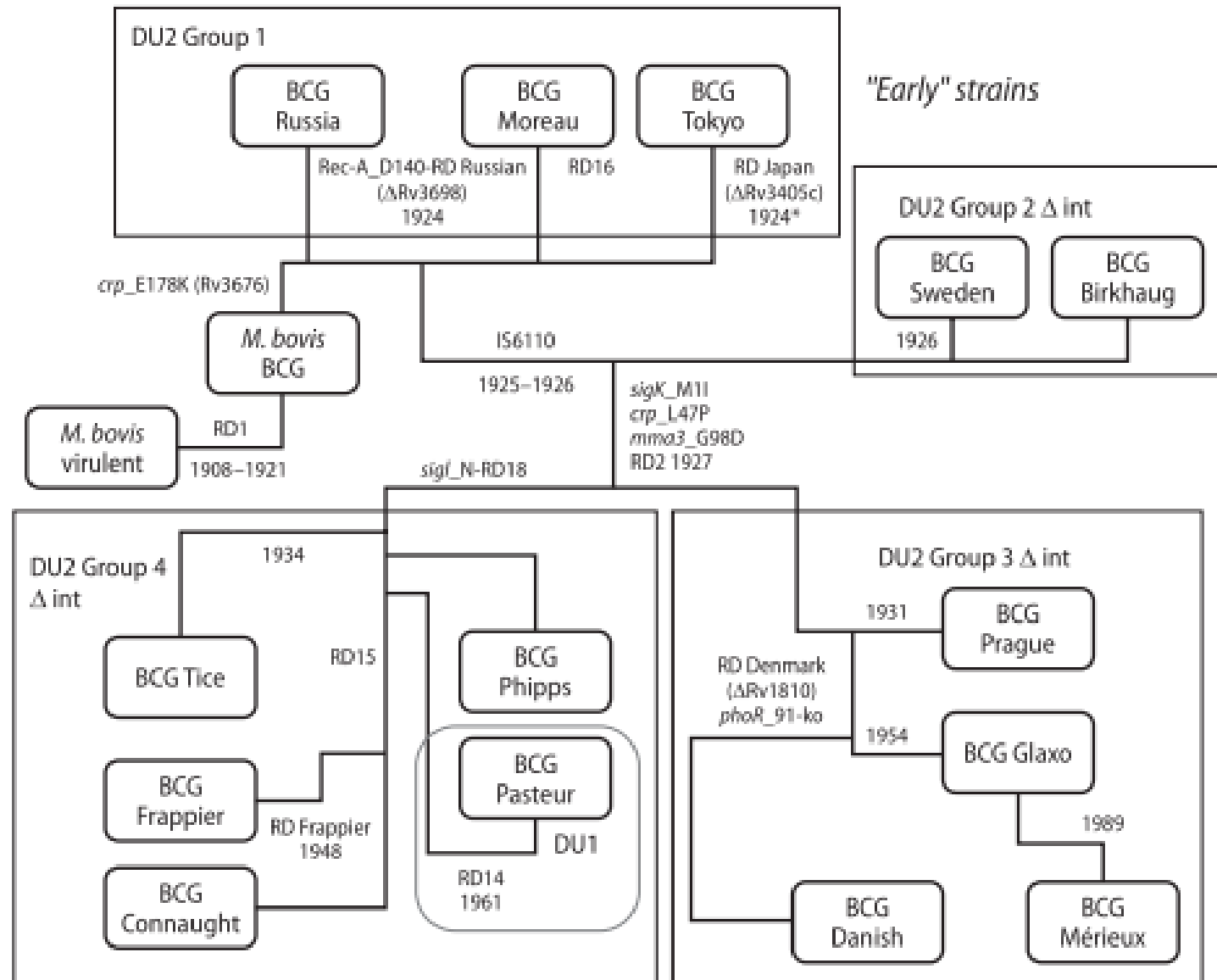
- Morales et al, J Urol 1976
- First use of intravesical BCG (with Martinez Pineiro, Spain 1975)
- 120 mg in 50cc of saline via urethral catheter into the bladder
- Strain: Frappier (Montreal); packaged in vials of 6
- Noted at least 3-6 weeks needed to mount delayed hypersensitivity reaction
- Side effects lasted 1 week
- Regimen → weekly dosing (minimize side effects) x 6 weeks (due to packaging convenience and time to mount immune response)
- 7 of 10 patients with recurrent tumors demonstrated response (decrease recurrence and or eradication of tumor)

# BCG schedule is empiric

- Schedule for BCG intravesical instillations very empiric!
- 6 weekly instillations used for 30 years but any rationale? Because indeed A Morales received six vials from the pharma company in 1976.....



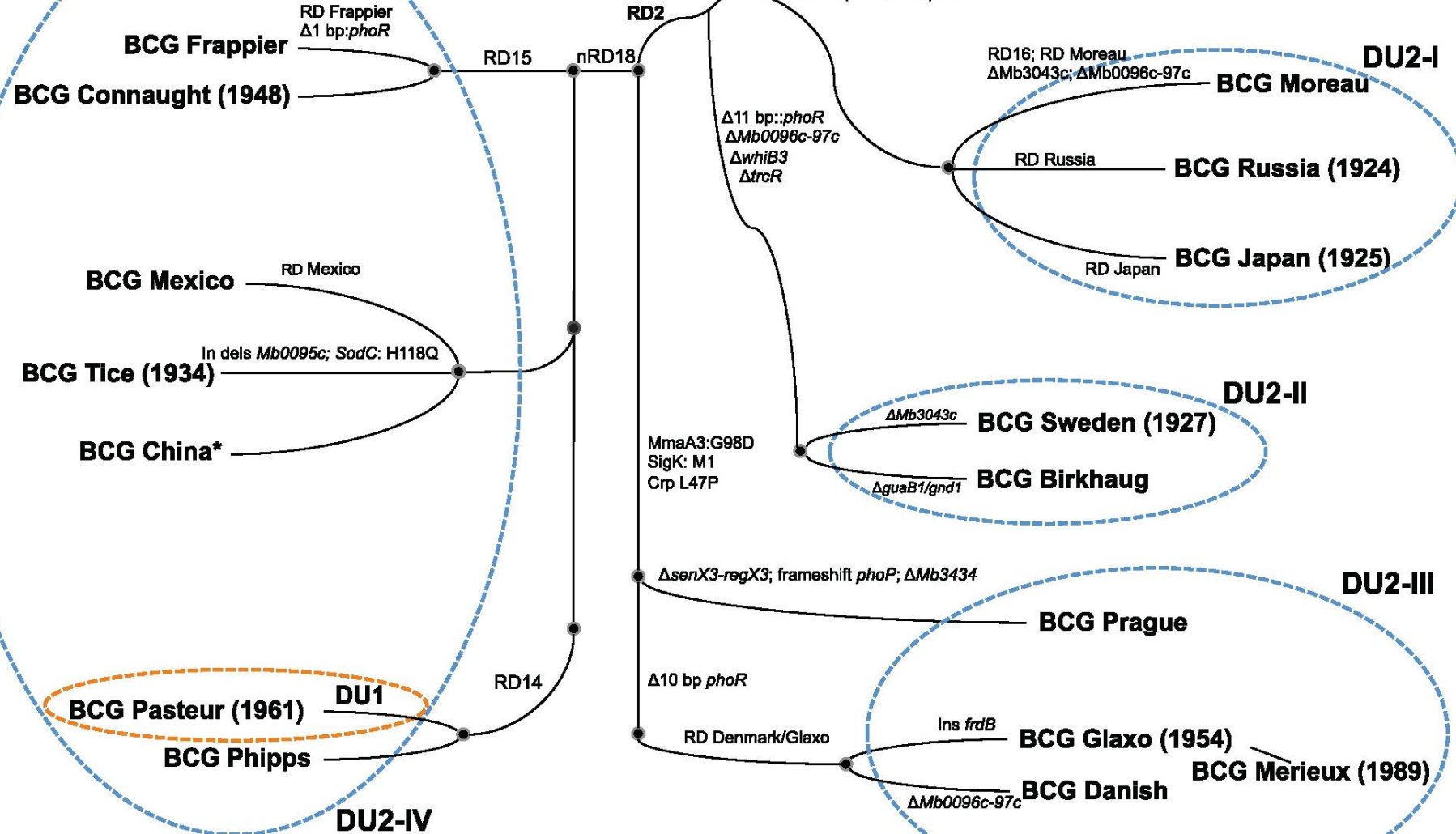
# History and genealogy of BCG substrains



"Late" strains

*Mycobacterium bovis*

RD1 | RD3  
**BCG (1908-1921)**



# RD1

- Common to all BCG vaccine strains is the deletion of region of difference one (RD1) that is preserved in *M. bovis* and *M. tuberculosis*.
- Loss of this region known to have been the critical event in the attenuation of the initial *M. bovis* strain.

# Gan et al (2013). BCG immunotherapy for bladder cancer- the effects of substrain differences Nat Rev Urol

Strain	<i>n</i> *	Mean CRR % (range)*	Commercial product	Weight (mg)	Recommended dose (cfu) <sup>‡</sup>	Secretion of lipid virulence factors? <sup>61</sup>	Secretion of MPB64/MPB70 and MPB83 <sup>76</sup>
Moscow <sup>§</sup>	103	90.5	SII-ONCO-BCG <sup>®</sup> (Serum Institute, India)	120	3–57 × 10 <sup>8</sup>	Yes	Present/High
Moreau RdJ	100	90	ImmunoBCG (FAP, Brazil)	80	0.04 × 10 <sup>8</sup>	No	Present/High
Connaught	450	79 (70–92)	Immunocyst <sup>®</sup> (Sanofi-Aventis, France)	81	1.8–15.9 × 10 <sup>8</sup>	NT	NT
Tokyo	111	77 (63–84)	Tokyo 172 (QSMI, Thailand)	80	0.4–0.5 × 10 <sup>8</sup>	No	Present/High
Pasteur	230	74 (40–80)	None	NA	NA	Yes	Absent/Present
Tice	277	71 (56–82)	OncoTice <sup>®</sup> (Merck, USA)	12.5	2–8 × 10 <sup>8</sup>	Yes	Absent/Present
Glaxo	180	65 (53–88)	None	NA	NA	No	Absent/Present
A. Frappier	145	60 (39–100)	None	NA	NA	Yes	Absent/Present
S. African	13	69	None	NA	NA	NT	NT
Copenhagen	42	67	None	NA	NA	Yes	Absent/Present
Romanian	33	64	None	NA	NA	NT	NT
RIVM/1	15	60	BCG-Medac <sup>®</sup> (Medac, Germany)	80	2–30 × 10 <sup>8</sup>	NT	NT

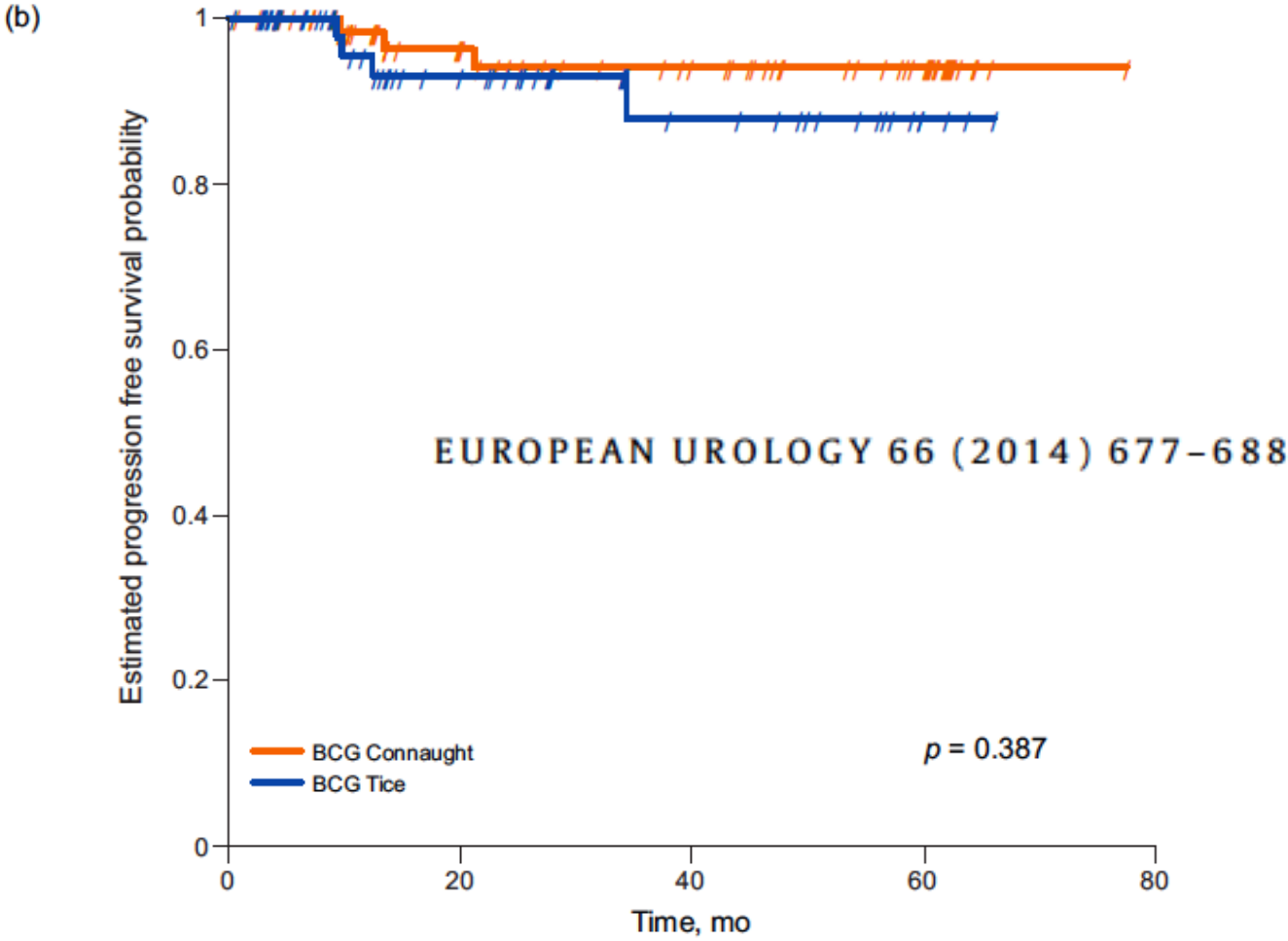
# CONCLUSION

- No meaningful correlations between BCG strain and survival outcomes (RFS, CSS, and OS)

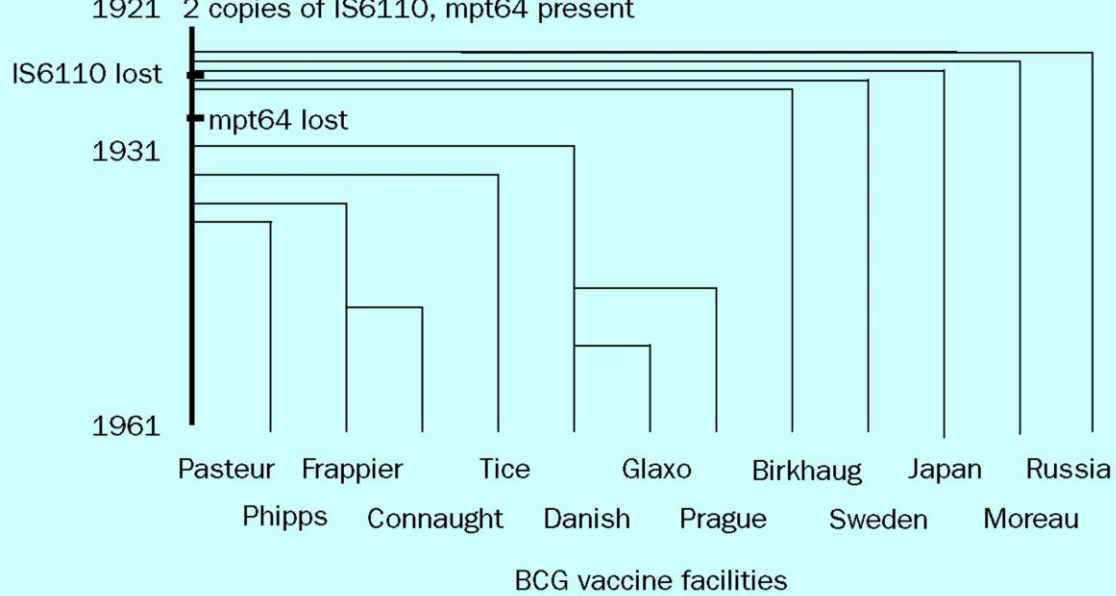
# Progression-free survival

## NO DIFFERENCE!

Rentsch et al  
Eur Urol 2014



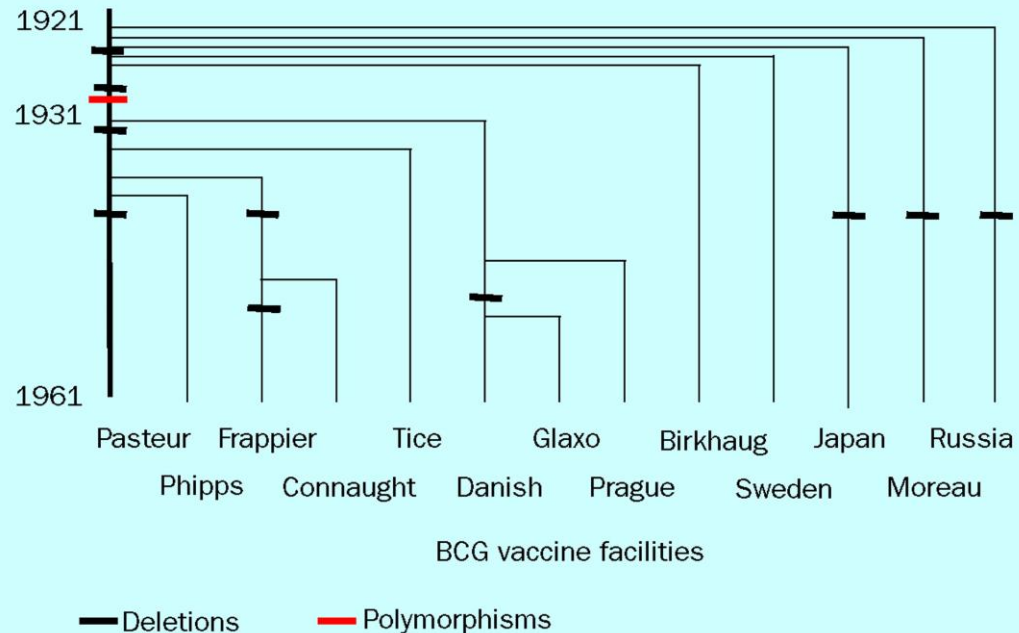
	No. at risk	0	20	40	60	80
BCG Connaught	71	45	32	18	0	0
BCG Tice	60	31	16	3	0	0



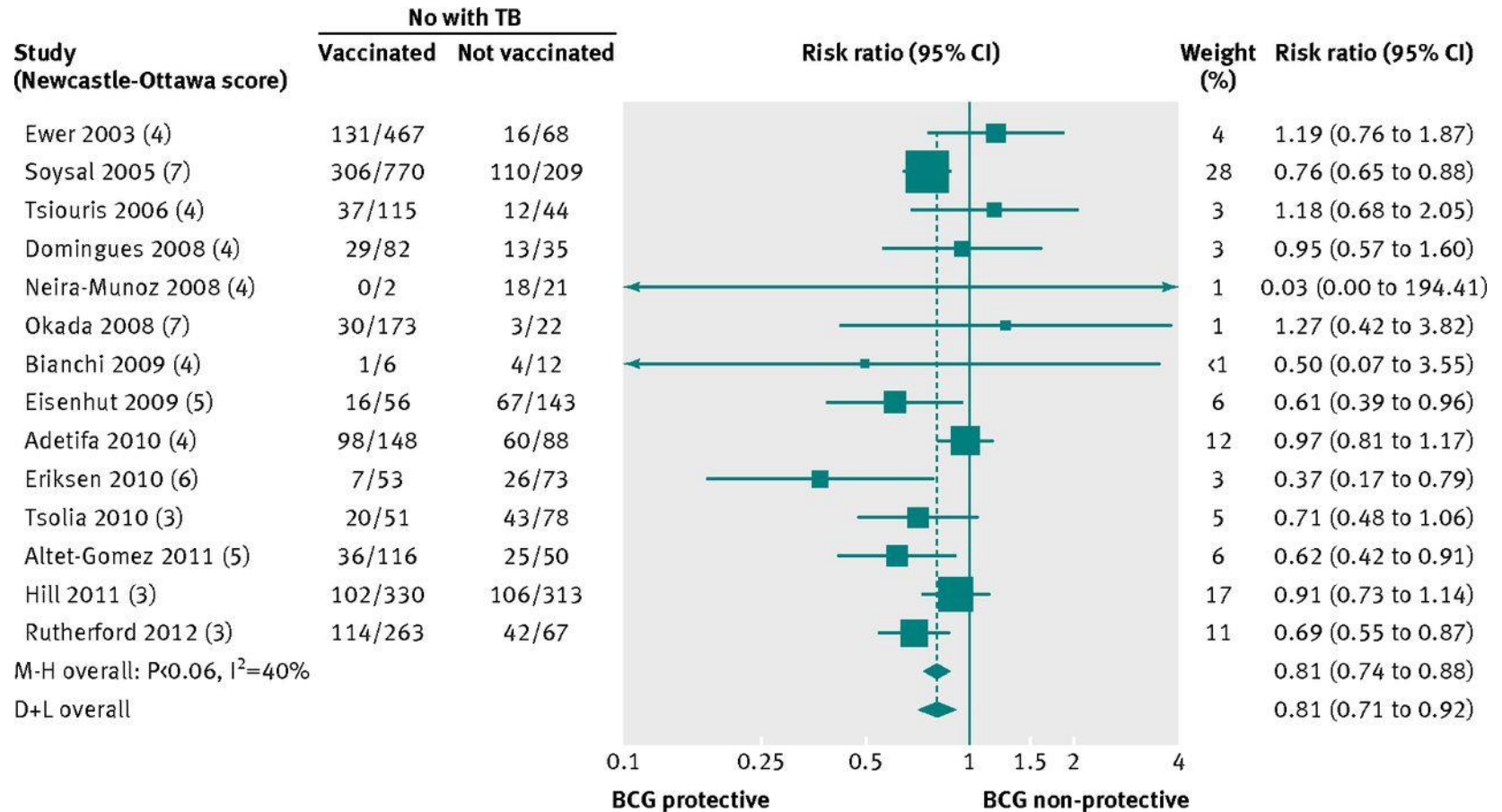
IS6110	1	1	1	1	1	1	1	1	1	1	2	2	2
mpt64	-	-	-	-	-	-	-	-	+	+	+	+	+

# BCG as anti-tuberculosis vaccine

***BCG — different strains, different vaccines? Behr M. The Lancet Infectious Diseases 2002: 86-92***



# Protection against *Mycobacterium tuberculosis* infection (TB) as determined by interferon $\gamma$ release assay (QuantiFERON) in children vaccinated with BCG.



A Roy et al. *BMJ* 2014;349:bmj.g4643





# Conclusions

- Evidence from animal and human studies shows that there are significant differences in the immune response induced by different BCG vaccine strains.
- **However: lack of data demonstrating the superiority of individual BCG vaccine strains!**

FEMS Microbiol Rev 32 (2008) 821–841



World Health  
Organization

Global Vaccine Safety,  
Immunization, Vaccines and Biologicals  
20, avenue Appia, CH-1211 Geneva 27

## INFORMATION SHEET

OBSERVED RATE OF VACCINE REACTIONS

# *BACILLE CALMETTE-GUÉRIN (BCG) VACCINE*

April 2012

- Pasteur 1173 P2, the Danish 1331, the Glaxo 1077 (derived from the Danish strain), the Tokyo 172-1, the Russian BCG-I, and the Moreau RDJ strains (NIBSC and WHO, 2004).
- The concentration of live particles in the vaccines ranges from 50,000 to 3 million per dose, according to the strains.
- Each strain has a different immunological profile
- There is no standardized production of BCG vaccine between manufacturers

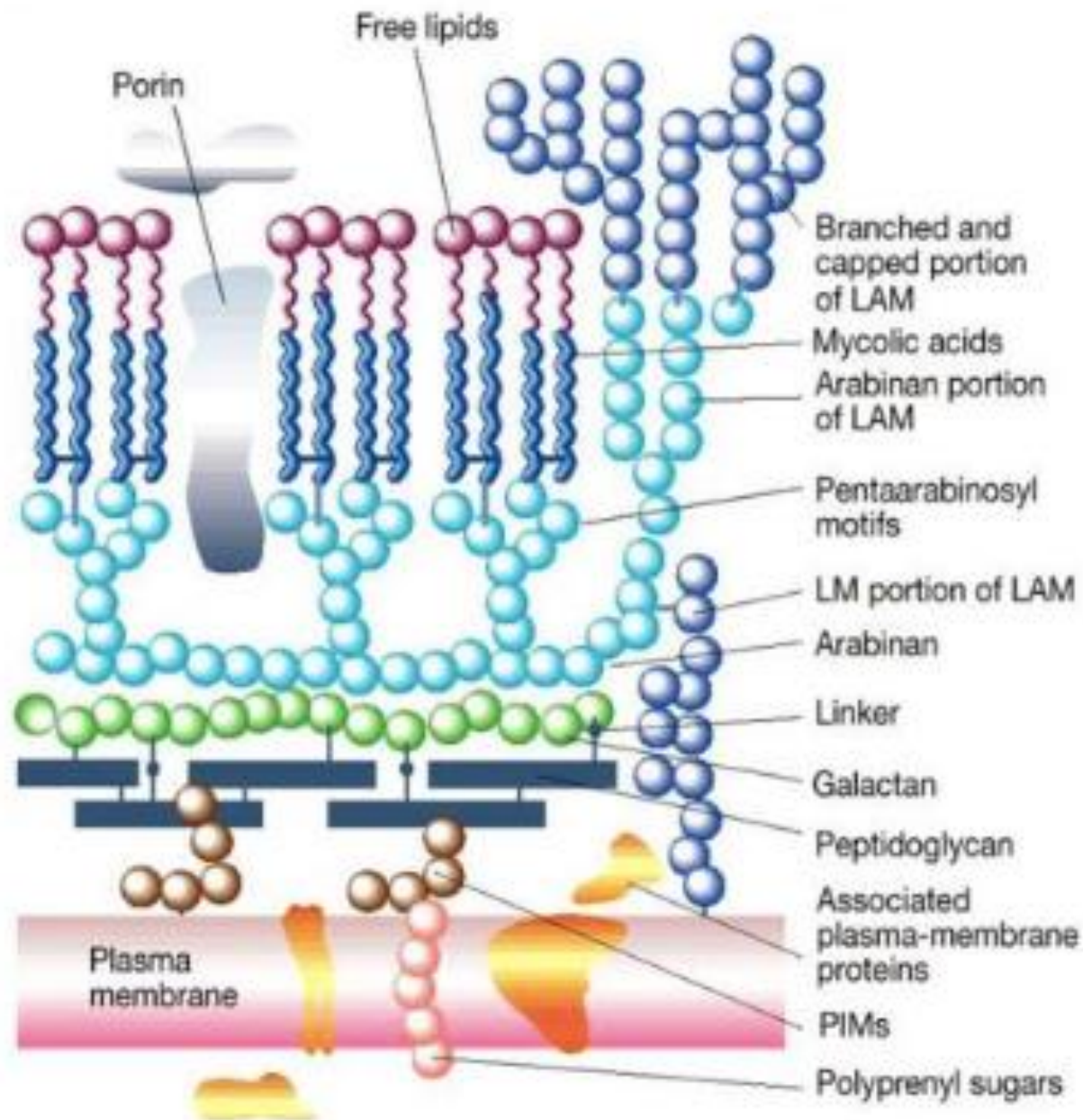
**WHY?**



# BCG very complex live attenuated micro-organism

- Cell wall | Culture filtrate
- Inner membrane | AG 85 (32kd, fibronectin-binding)
- Insoluble cell wall | P64 (~ hsp 65)
- Glucane | 22 kd
- Arabinomannans | Pst2
- Fatty acids | Pst3
- Cytoplasm

# Mycobacterial (acid-fast) cell wall



▲ Cytoplasmic proteins

▲ Cell wall proteins

■ Secreted proteins

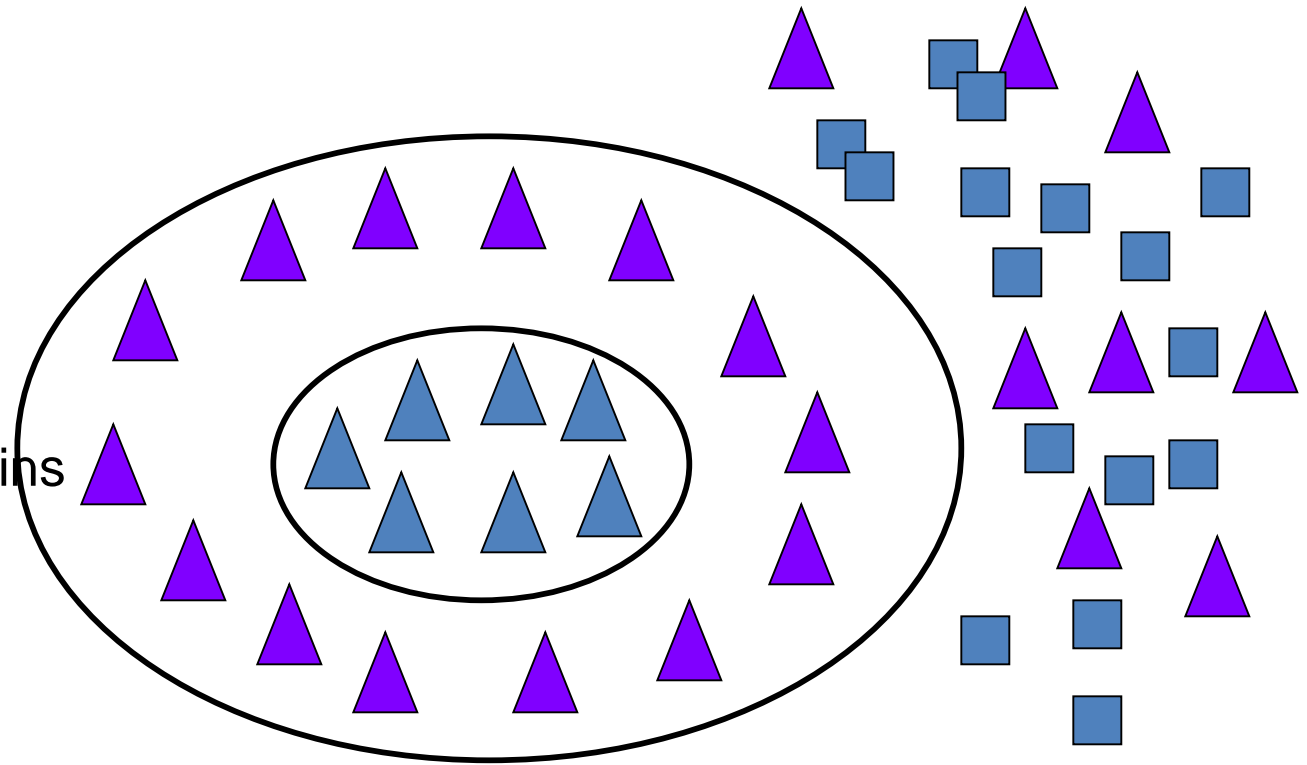
**Ag 85**

22 kDa

PstS-2 and -3

Heat Shock proteins

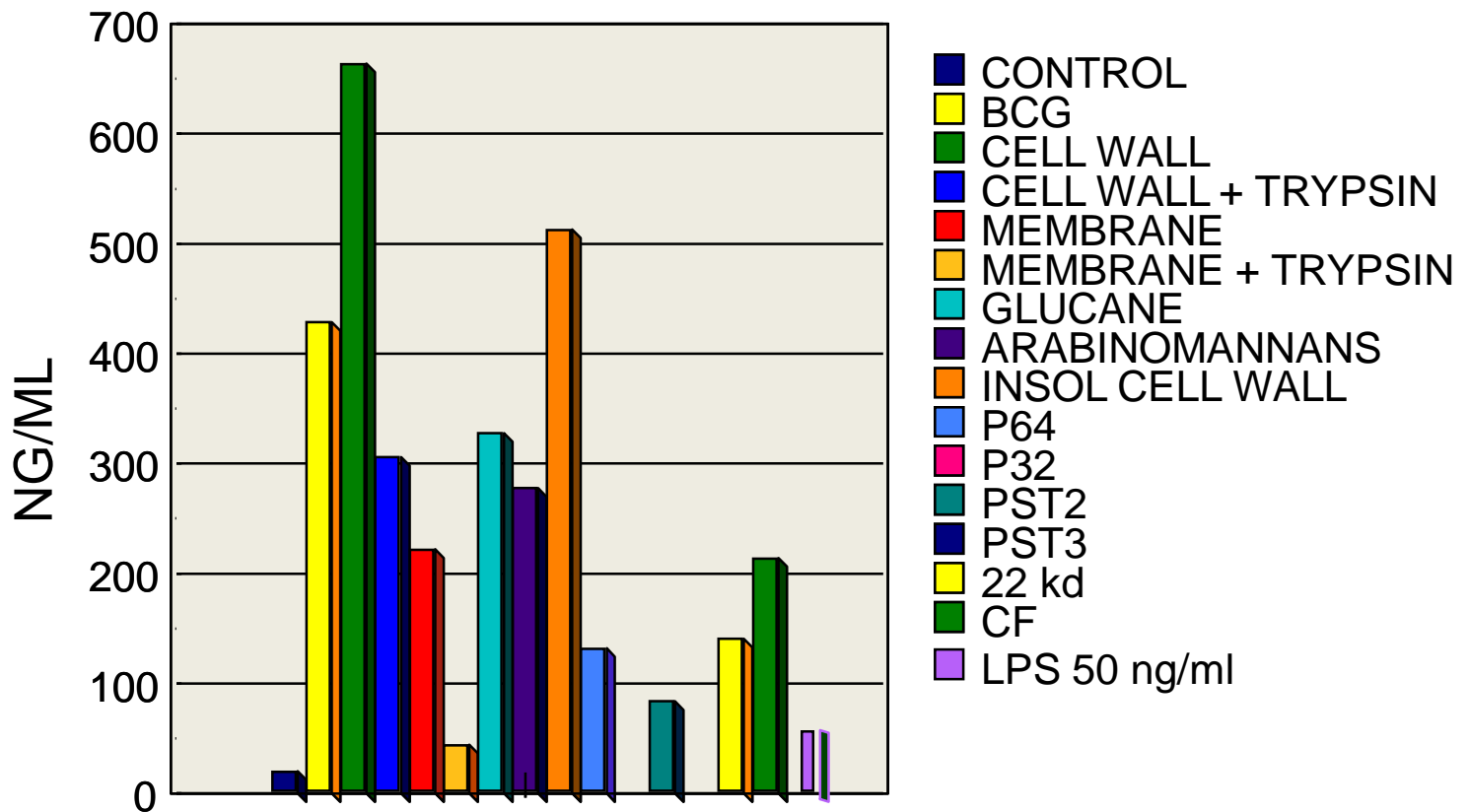
ESAT-6



# AG 85 complex

- Central role in synthesizing major components of the inner and outer leaflets of the mycobacterial outer membrane
- Binds Fibronectin!
- BCG attaches to urothelium of the bladder through Fibronectin. (Zlotta et al. Evolution and Clinical Significance of the T Cell Proliferative and Cytokine Response Directed Against the Fibronectin Binding Antigen 85 Complex of Bacillus Calmette-Guerin During Intravesical Treatment of Superficial Bladder Cancer. J Urol 1997: 492–498, Zhao et al JBC, 1999)
- Safety and efficacy of MVA85A, a new tuberculosis vaccine, in infants previously vaccinated with BCG: a randomised, placebo-controlled phase 2b trial. Lancet. 2013 381(9871): 1021–1028.

# IL-12 PRODUCTION



Zlotta et al, 2001



**Many other parameters than the strain account for BCG efficacy in Bladder Cancer!!!!**

# Toll-like receptor (TLR)

- Toll-like receptor (TLR) pathways are of particular interest in cancer immunotherapy.
- TLRs: family of receptors that bind to common components of many pathogens as well as signals released by damaged cells.
- Expressed on many innate immune cells, including dendritic cells. The most potent of all antigen presenting cells, dendritic cells play a pivotal role in bridging the innate and adaptive responses.
- TLRs present on a large portion of bladder tumors where higher TLR expression is correlated with less invasive tumors

# Treatment of non-muscle invasive bladder cancer with Bacillus Calmette–Guerin (BCG): Biological markers and simulation studies

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<sup>c</sup> Department of Urology, State Pediatric Medical University, St. Petersburg 194100, Russia

- Some individuals show a higher level of natural resistance than others to infection with certain intracellular pathogens, including *Mycobacterium bovis* BCG (BCG)
- Gene encoding *Nramp* 1 (natural resistance-associated protein 1) exists in two allelic forms, differing for a point mutation.
- [\*NRAMP1\* and \*hGPX1\* gene polymorphisms to BCG response showed that the \*NRAMP1\* D543N G:G genotype displayed decreased CSS.](#)
- Gene polymorphisms that led to reduced RFS or increased recurrence risk post-BCG [\*XPA\*, \*XPC\*, \*XPB\*, \*XPD\*, \*XPG\*, \*XPF\*, \*ERCC1\*, \*ERCC2\*, \*ERCC6\*, \*XRCC1\*, \*XRCC4\*, \*APEX1\*, \*GSTM1\*, \*CCNB1\*, \*PON1\*, and \*SLCO1B1\*](#)

# Conclusions

- Variability in BCG strains not demonstrated to affect clinical outcome in bladder cancer
- Variability in BCG strains not demonstrated to affect efficacy in tuberculosis vaccination
- Mechanism of action of BCG in bladder cancer complex and involving many subcomponents which individually can all stimulate a robust adequate immune response
- Many other parameters than BCG strains account for the clinical efficacy of BCG
- At times of BCG shortage, differences in strains are not a concern