Identification of additional Ser/Thr protein kinases in the genome of Streptococcus thermophilus using structural homology detection.

Gwenaëlle André-Leroux, Veronique Monnet, Samantha Samson

To cite this version:

Gwenaëlle André-Leroux, Veronique Monnet, Samantha Samson. Identification of additional Ser/Thr protein kinases in the genome of Streptococcus thermophilus using structural homology detection.. JOBIM : Journées Ouvertes Biologie Informatique Mathématiques, Jul 2018, Marseille, France. , 2018. hal-02733931

HAL Id: hal-02733931
https://hal.inrae.fr/hal-02733931

Submitted on 2 Jun 2020

Identification of additional Serine/Threonine protein kinases in the genome of *Streptococcus thermophilus* using structural homology detection

Samantha Samson[1], Véronique Martin[1], Lucia Haller[2], Véronique Monnet[2], Gwenaëlle André-Leroux[1]

[1] MaIAGE, INRA, Université Paris-Saclay, 78350, Jouy-en-Josas, France [2] MICALIS, INRA, Université Paris-Saclay, 78350, Jouy-en-Josas, France

Introduction

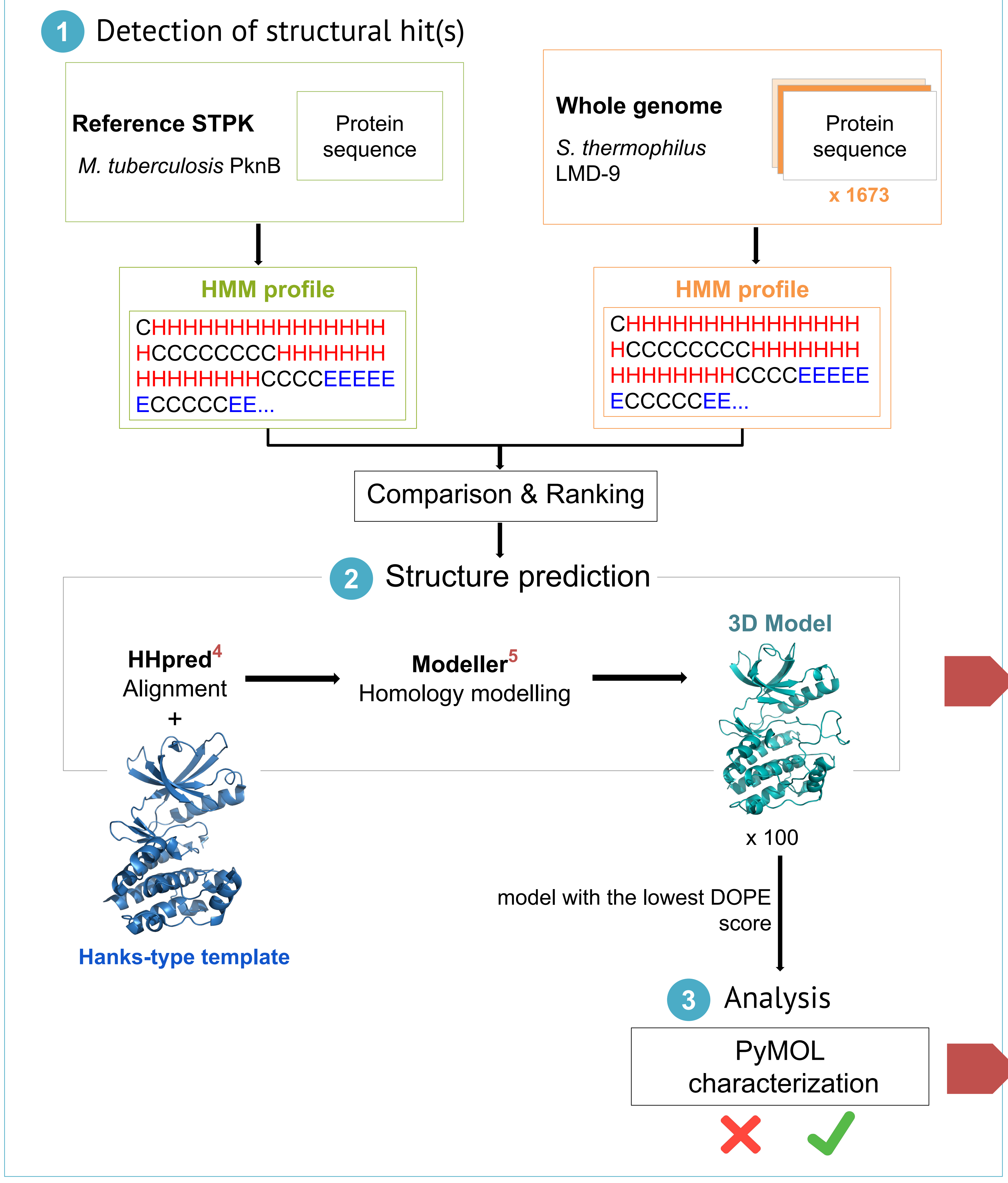
The Ser/Thr/Tyr phosphoproteome of *S. thermophilus* LMD-9 shows the phosphorylation of 106 proteins. When the one of and only Ser/Thr Protein Kinase (STPK) identified in this genome -named PknB- is depleted, 96 out of those 106 proteins are still phosphorylated, hence evidencing the presence of at least another STPK[1].
Knowing that the catalytic domain of STPK all shares a similar 3D fold, called Hanks-type kinase, we hypothesized that the unidentified kinase(s) should share a similar fold[2]. Thus, we screen the whole genome of *S. thermophilus* LMD-9, predicting the structural profil of each of the 1673 fasta sequences and comparing it with to a STPK of reference.

On this basis, our project aims at identifying the missing STP kinase(s).

Objectives

- **Structure/Function analysis** of PknB identified in *S. thermophilus* LMD-9 genome as regard to the solved reference in *Mycobacterium tuberculosis*[3].
- **Screening of structural homologues of PknB**, using profile-profile comparison[4], within the 1673 protein sequences of the *S. thermophilus* LMD-9 genome.
- **Identification and analysis** of the best structural hit(s) once homology modeled[5].

Method: Metafoldscan pipeline



① Detection of structural hit(s)

Reference STPK
M. tuberculosis PknB
Protein sequence

Whole genome
S. thermophilus LMD-9
Protein sequence
x 1673

HMM profile
CHHHHHHHHHHHHHHHH
HCCCCCCCCHHHHHHH
HHHHHHHHCCCCEEEEE
ECCCCCEE...

HMM profile
CHHHHHHHHHHHHHHHH
HCCCCCCCCHHHHHHH
HHHHHHHHCCCCEEEEE
ECCCCCEE...

Comparison & Ranking

② Structure prediction

HHpred[4]
Alignment
+
Hanks-type template

Modeller[5]
Homology modelling

3D Model
x 100

model with the lowest DOPE score

③ Analysis

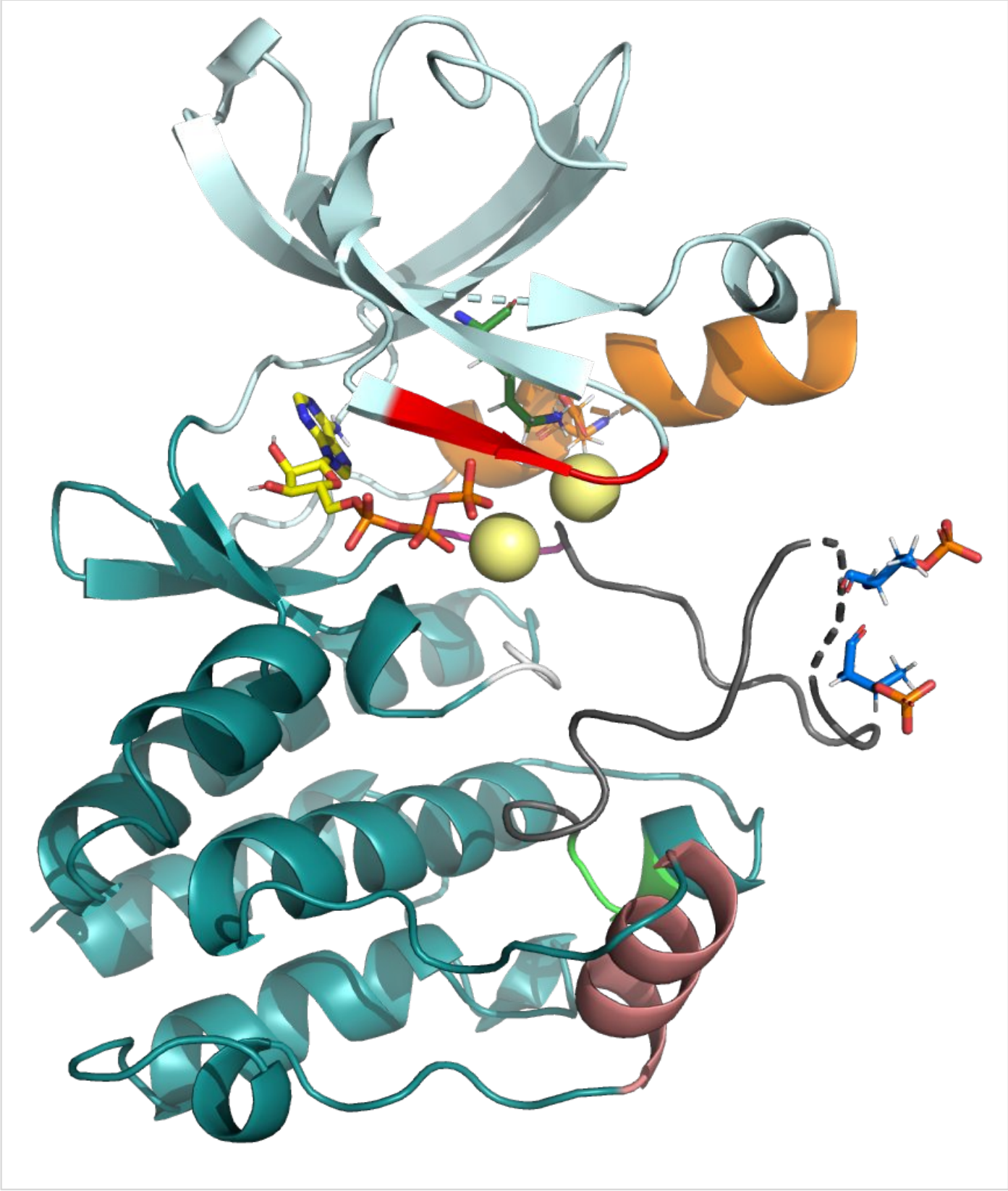
PyMOL characterization

Results: analysis of the structural hits

➔ Structural homologues to the reference PknB catalytic domain in *M. tuberculosis*[3] (PDBid: 1O6Y - length: 332 amino acids):

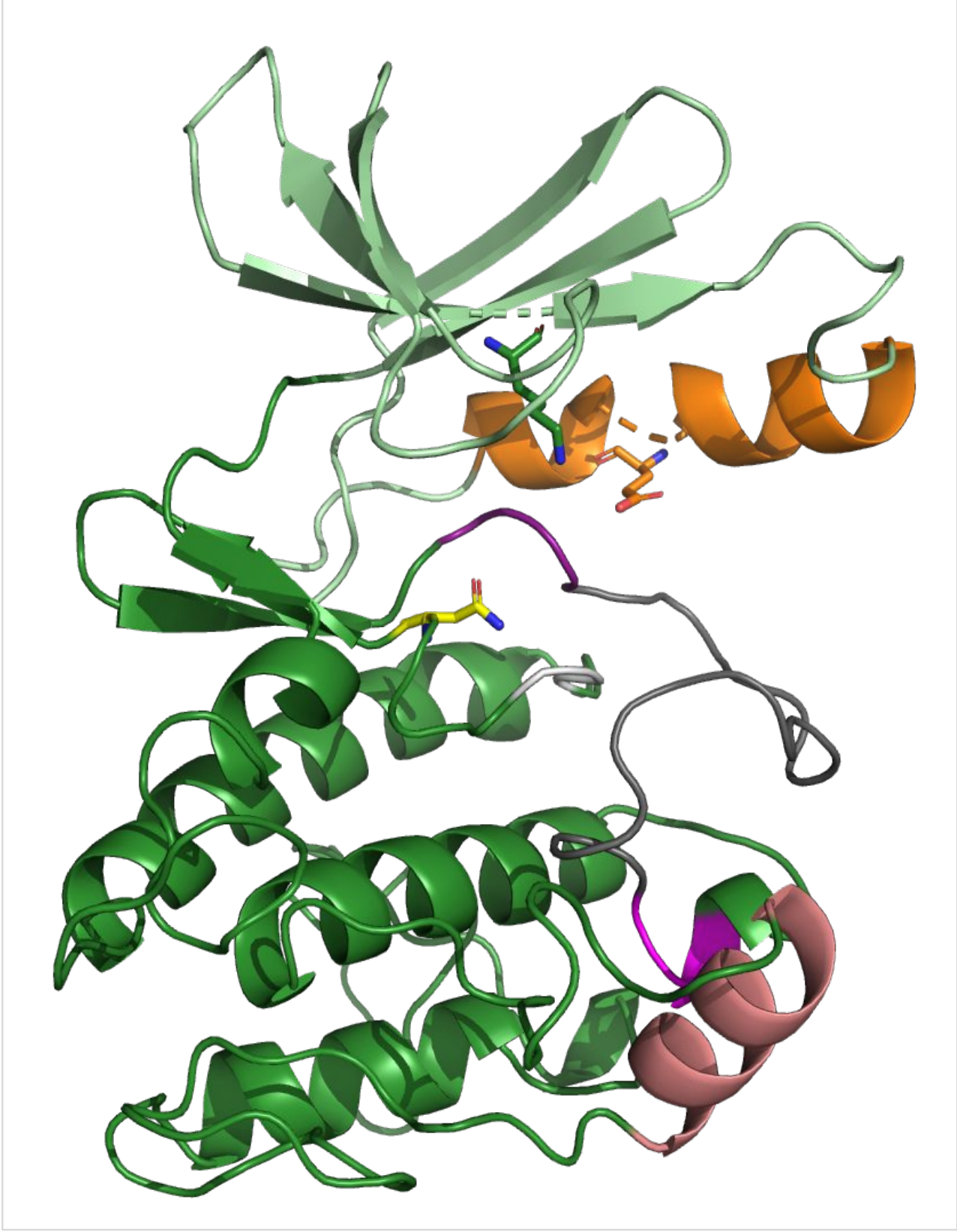
Rank	Annotation of the hits	Homology (%)	Identity (%)	Aligned residues
1	Serine/Threonine Protein Kinase PknB	100	40	271
2	Hypothetical protein	99.9	19	251
3	AarF/ABC1/UbiB kinase family protein	99.4	25	138
4	aminoglycoside phosphotransferase	94.2	24	29
5	membrane protein	13.4	43	14

HIT 1 : Ser/Thr Protein Kinase PknB



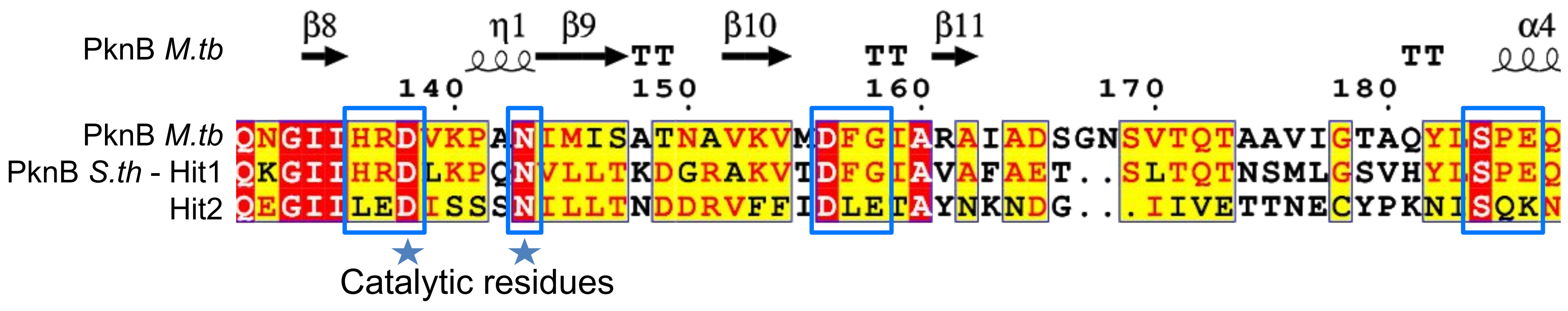
Hanks-type kinase
☑ Structure
☑ **Conserved** functional motifs

HIT 2 : Hypothetical protein



Pseudokinase ?
☑ Structure
☑ **Degenerated** functional motifs

ESPript alignment between PknB *M. tb* and *S. th* + Hit2



Catalytic residues

Conclusion & Perspectives

- Structure/Function analysis of *S. thermophilus* LMD-9 PknB.
- Identification of two promising structural hits called Hits 2 & 3.
- Among them, characterization of Hit 2 as a potential pseudokinase.
- Phenotypical experimental validation in progress of Hit 2 and Hit 3.

Bibliography

1. Haller L., et al. **The Hanks-type kinase PknB, targeting the divisome, is not the single player in the *Streptococcus thermophilus* protein phosphorylation process** (*To be published*).
2. Stancik A., et al. **Serine/Threonine Protein Kinases from Bacteria, Archea and Eukarya Share a Common Evolutionary Origin Deeply Rooted in the Tree of Life.** J. Mol. Bio. 2008. 430(1): 27-32
3. Ortiz-Lombardía M., et al. **Crystal structure of the catalytic domain of the PknB serine/threonine kinase from *Mycobacterium tuberculosis*.** J. Biol. Chem. 2003. 278 (15): 13094-100
4. Söding J, Biegert A, Lupas N. **The HHpred interactive server for protein detection and structure prediction.** Nucleic Acids Res. 2005. 33(Web Server issue): W244–W248.
5. Sali A. & Blundell. T.L. **Comparative protein modelling by satisfaction of spatial restraints.** J. Mol. Biol 1993. 234, 779-815
6. DeLano, W.L. **The PyMOL Molecular Graphics System** 2002. http://www.pymol.org.