

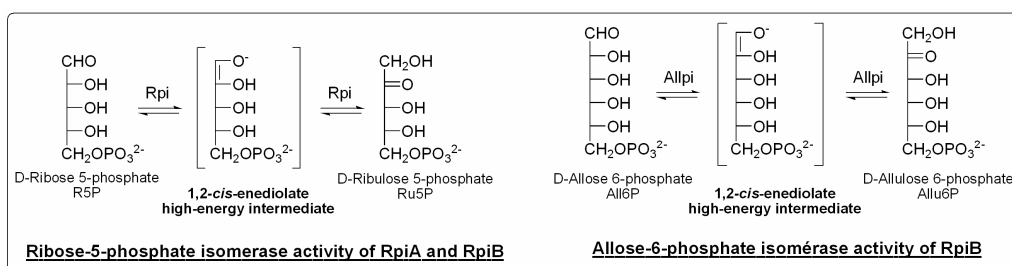
# Evidence of the new allose-6-phosphate isomerase activity of type B ribose-5-phosphate isomerase from *Escherichia coli*. Synthesis and kinetic evaluation of specific inhibitors

S. Mariano<sup>1</sup>, A. K. Roos<sup>2</sup>, S. L. Mowbray<sup>2</sup>, and L. Salmon<sup>1</sup>

<sup>1</sup> Laboratoire de Chimie Bioorganique et Bioinorganique (LCBB), UMR 8182, ICMMO, Bât. 420, Université Paris Sud 91405 Orsay Cedex, France. [smariano@icmo.u-psud.fr](mailto:smariano@icmo.u-psud.fr)

<sup>2</sup> Department of Cell and Molecular Biology, Uppsala University, Biomedical Center, SE-751 24 Uppsala, Sweden.

Ribose-5-phosphate isomerase (Rpi, EC 5.3.1.6), an aldose-ketose isomerase involved in the pentose phosphate pathway, catalyzes the reversible isomerization reaction between D-ribose 5-phosphate (R5P) and D-ribulose 5-phosphate (Ru5P)<sup>[1]</sup>. The reaction is thought to proceed through a proton transfer mechanism and to involve a 1,2-*cis*-enediolate high-energy intermediate. Two unrelated types of enzyme are known to catalyze the isomerization. The most common one, RpiA, is present in almost all organisms like humans and spinach. The second type, RpiB, is found in many bacterial species, like *Mycobacterium tuberculosis* and *Escherichia coli*, and is expected to have an allose-6-phosphate isomerase activity<sup>[2]</sup> (Allpi) in addition to the Rpi activity. The Allpi activity catalyzes the reversible isomerization reaction between D-allose 6-phosphate (All6P) and D-allulose 6-phosphate (Allu6P). This postulated particularity of RpiBs makes them potentially interesting therapeutic targets, particularly for the development of new antibiotics against tuberculosis. Preliminary inhibition assays targeting this second specific activity of RpiBs have proved encouraging<sup>[3]</sup>.



The work that will be presented is based on:

- A new synthesis of D-allose 6-phosphate (All6P) and D-allulose 6-phosphate (Allu6P), substrates of the potential Allpi activity of RpiBs.
- The development of a new appropriate enzymatic test to investigate Allpi activity in RpiBs (employing thiobarbituric acid colorimetric assay).
- The synthesis of new potential high-energy intermediate analogue inhibitors of both Allpi and Rpi activities obtained by oxidative cleavage of the precursor All6P or Allu6P, followed by lactonisation and ring opening by nucleophilic addition.
- Kinetic evaluation of the inhibitors synthesized on Allpi (EcRpiB) and Rpi (RpiA and B of different organisms) activities.
- Cristallographic studies of enzyme-inhibitor and enzyme-substrate complexes and comparison to the kinetics properties observed.

[1]. Woodruff, W. W.; Wolfenden, R. *J. Biol. Chem.*, **1979**, *254*, 5866-5867.

[2]. C. Kim, S. Song, C. Park. *Journal of Bacteriology*, Dec. **1997**, 7631-7637.

[3]. E. Burgos, A. K. Roos, S. L. Mowbray, and L. Salmon, *Tetrahedron Lett.* **2005**, *46*, 3691-3694.