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NucLib: A chemical library of non-canonical (deoxy-) ribonucleotides produced by a versatile enzyme cascade.

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PURPOSE OF THE ABSTRACT
Ribonucleotides and their corresponding deoxyribonucleotides are central to a myriad of biological processes such as storage and expression of genetic information, energy metabolism and signal transduction. Due to the biological omnipresence of this class of compounds, analogues of the canonical nucleotides are widely used to manipulate biological systems, leading to application in fields ranging from medicine to synthetic biology. The actual mode of action relies highly on the form of the respective nucleotide in terms of 3'-oxygenation and phosphorylation level.
In the project NucLib, we aim for the establishment of a versatile and flexible enzymatic cascade for the biosynthesis all relevant forms of non-canonical nucleotides. This includes the ribose and deoxyribose forms as nucleoside mono-, do-, and triphosphates.
In its core, the cascade consist of the enzymes phosphoribosyl transferase(1), polyphosphate kinase(2, 3) and ribonucleotide reductase(4, 5). We investigate the substrate specificities of the enzymes for the conversion of non-canonical substrates and improve the promiscuity by enzyme engineering. We also investigate the possibilities to produce each relevant compound either separately or in one complete mixture for each non-canonical nucleotide.
The cascade was already successfully applied for the biosynthesis of 2Cl-adenosine triphosphate, the active metabolite of the anti-cancer drug Mavenclad®(6). Part of the cascade was used to synthesize the active metabolites of the drug 5-Aminoimidazole-4-carboxamide ribonucleotide (AICAR)(7). Further extension of the substrate scope will enable the creation of a chemical library of non-canonical (deoxy-) ribonucleotides to evaluate and facilitate their biotechnological application.

FIGURES

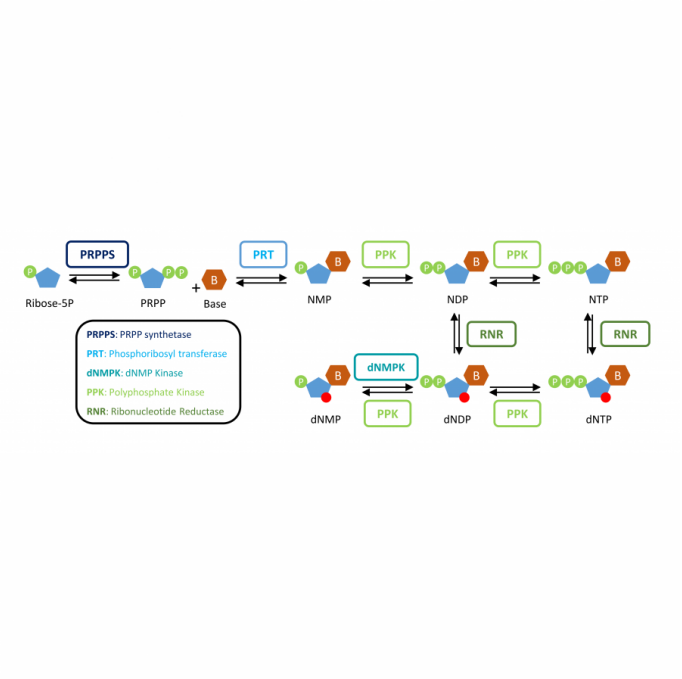

FIGURE 1

Figure 1: NucLib Cascade:
Concept of the enzymatic cascade for biosynthesis of non-canonical (deoxy-) ribonucleotides for the creation of the NucLib chemical library.

FIGURE 2

KEYWORDS

Ribonucleotide reductase | Non-canonical nucleotides | Nucleotide modfying enzymes | Enzyme cascades

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