

Electric supplementary information (ESI)

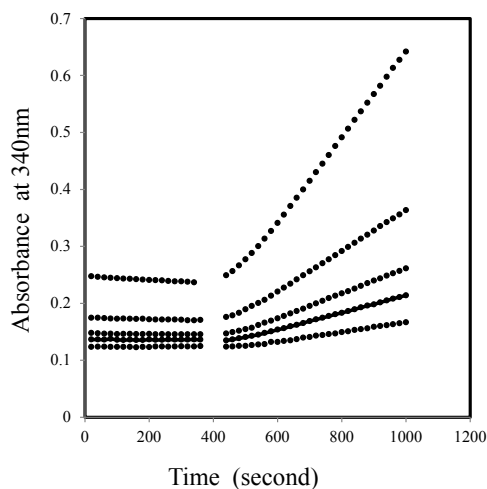
Development of an enzyme cycling method by a purine nucleoside phosphorylase for assaying inorganic phosphate

Information for principal enzymes

Purine nucleoside phosphorylase (PNP) (EC 2.4.2.1) and xanthine dehydrogenase (XDH) (EC 1.17.1.4) were obtained from Diagnostics Department, Asahi Kasei Pharma Corporation, Tokyo, Japan (<http://www.asahi-kasei.co.jp/shindan/en/>). The abbreviation and Product number of PNP and XDH are XDHII, T-134 and PNPLII, T-69, respectively. Lot.1001A (PNPLII) and Lot.0801A (XDHII) were used for the study.

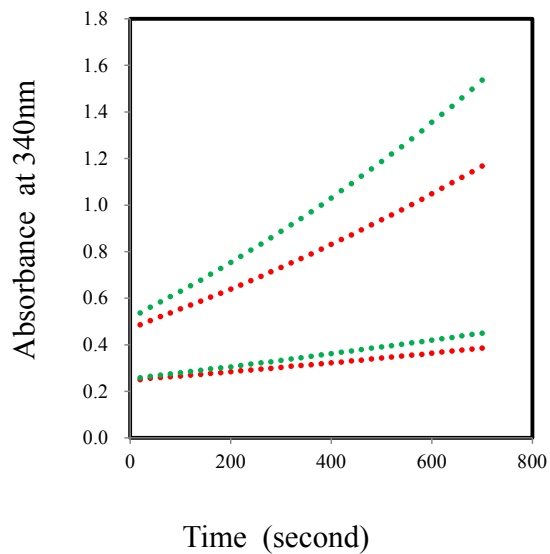
Typical reaction time course on the quantitative determination of dipotassium hydrogen phosphate

The raw chart of time course of the absorbance at 340 nm was shown. The slope after adding 0.5mM guanine at around 400 sec was increased, depending on the amount of dipotassium hydrogen phosphate (0, 0.1, 0.2, 0.4, 1.0 μ M in the reagent with 1 U/ml PNP).



Reaction time course on 2 and 5 U/ml PNP with or without the substrate

Both time course on 2 U/ml and 5 U/ml PNP with and without 50 nM dipotassium hydrogen phosphate were indicated. The lower couple of line was the results on 2 U/ml PNP and the upper on 5 U/ml PNP, respectively.



Dependency of the sensitivity and the reagent blank on various kinds of buffers

Bicine, POPSO, TAPS and Tricine were obtained from Dojindo Laboratories (Kumamoto, Japan). The pH and concentration of each buffer was pH7.5 and 50 mM. The reagent blank value on TAPS was much greater. We speculated this was because of the non-negligible contamination of inorganic phosphate in TAPS.

