

## Phenylalanyl-tRNA Synthetase from Baker's Yeast: Specificity and Quantitation of Affinity Elution with tRNA

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**Summary:** tRNA<sup>Phe</sup> is able to elute phenylalanyl-tRNA synthetase from cation exchangers in a 1:1 ratio. Elution of phenylalanyl-tRNA synthetase in a 1:1 ratio is also observed for four non-cognate tRNAs investigated, specific for valine, serine, isoleucine and tyrosine. If protein mix-

tures are subjected to affinity elution the cognate pair [tRNA<sup>Phe</sup> · phenylalanyl-tRNA synthetase] is eluted first, followed by noncognate pairs. The unspecific elution is not influenced by complexation of phenylalanyl-tRNA synthetase with an analog of phenylalanyl-adenylate.

### *Phenylalanyl-tRNA-Synthetase aus Bäckerhefe: Spezifität und Quantifizierung der Affinitäts-Elution mit tRNA*

**Zusammenfassung:** tRNA<sup>Phe</sup> vermag Phenylalanyl-tRNA-Synthetase von Kationenaustauschern im Verhältnis 1:1 auszuwaschen. Die gleiche 1:1-Stöchiometrie ergibt sich, wenn Phenylalanyl-tRNA-Synthetase mit den tRNAs für Valin, Serin, Isoleucin und Tyrosin eluiert wird. Bei präparativer Affinitätselution von Proteingemischen wird

zunächst das [tRNA<sup>Phe</sup> · Phenylalanyl-tRNA-Synthetase]-Paar ausgewaschen, gefolgt von den nicht zusammengehörenden Paaren [tRNA<sup>Xxx</sup> · Phenylalanyl-tRNA-Synthetase]. Die unspezifische Elution wird durch Komplexbildung zwischen dem Enzym und einem Phenylalanyl-AMP-Analogen nicht beeinflusst.

**Key words:** Aminoacyl-tRNA synthetase, isolation, equilibrium, tRNA-enzyme interaction.

Specific elution of aminoacyl-tRNA synthetases from cation exchangers with tRNA, called "affinity elution", is a useful method for the purification of this group of enzymes<sup>[1-3]</sup>. The method makes use of the difference in charge between aminoacyl-tRNA synthetase and [aminoacyl-tRNA

synthetase · tRNA] complex to specifically desorb the enzyme from the ion exchanger. It is used here to demonstrate that unspecific interactions between noncognate tRNA and phenylalanyl-tRNA synthetase do occur.

#### *Enzyme:*

Phenylalanyl-tRNA synthetase, L-phenylalanine:tRNA<sup>Phe</sup> ligase (AMP-forming) (EC 6.1.1.20).

#### *Abbreviations:*

tRNA<sup>Phe</sup>-C-C-A = tRNA<sup>Phe</sup>.