Figure S2. A kinetic assay was used to determine the percentage of leucine in isoleucine samples.

A, Representative plateau formation of Leu-tRNA\textsuperscript{Leu} at various concentrations of isoleucine – 20 mM (●), 40 mM (▲), 60 mM (♦). WT LeuRS (additionally purified to remove Leu-AMP bound in the enzyme’s active site) was present at 4 µM concentration and tRNA\textsuperscript{Leu} was present at 20 µM concentration.

B, Replot of Leu-tRNA\textsuperscript{Leu} plateaus vs isoleucine concentration. The slope of the line represents the fraction of leucine present in L-Ile Sigma BioUltra lot 1 (♦), L-Ile Roth Cellpure (◊), L-Ile Serva Analytical grade (▼), and L-Ile Sigma BioUltra lot 2 unpurified (○) or purified (●). The inset shows Leu-[\textsuperscript{32}P]-tRNA\textsuperscript{Leu} formation in dependence of L-Ile BioUltra lot 1 concentration on a plot with a narrower x-axis scale.