Supplemental Tables S1 and S2.

Table S1. Composition of pre-incubation buffer.

Chemical name	3x conc. solution*[a]
Tris-acetate (pH 8.2)	300 mM
Mg acetate	9.3 mM
ATP adjusted pH 7.0 by KOH	13 mM
DTT	4 mM
19 amino acids, without L-tyrosine	each 0.04 mM
L-tyrosine	0.04 mM
phosphoenolpypuvate	84 mM
pyruvate kinase	6.67 U/ml

a. 30% volume of cell extract is added and incubated for 80 min at 37 °C.

Table S2. List of chemicals and concentration used in the cell-free protein synthesis presented in this paper by micro dialysis method (30-μl inner solution dialyzed against 1 ml feeding solution).

CAS No.	Chemical name	Concentration	Reaction solution	Feeding solution
7365-45-9	HEPES	59.25 mM	+	+
19473-49-5	L-Glutamic Acid Pottasium Salt Monohydrate	197.8 mM	+	+
922-32-7	Creatine Phosphate Disodium salt	79.2 mM	+	+
1492-18-8	Folinic acid-Ca	35.5 mg/L	+	+
987-65-5	ATP-Na_2-$3H_2O$	1.28 mM	+	+
36051-31-7	GTP-Na_2-$3H_2O$	0.89 mM	+	+
36051-68-0	CTP-Na_2-$3H_2O$	0.89 mM	+	+
19817-92-6	UTP-Na_2-$3H_2O$	0.89 mM	+	+
631-61-8	Ammonium Acetate	27.4 mM	+	+
3483-12-3	DTT	1.85mM	+	+
60-18-4	L-Tyr	1.53 mM	+	+
37839-81-9	3,5-cAMP-Na	0.66 mM	+	+
16674-78-5	Magnesium Acetate Tetrahydrate	9.28 mM	+	+
	19 amino acids mix without L-Tyr[*a]	each 1.5 mM	+	+
	NaN_3	0.050 %	+	+
	tRNA (*E. coli*)	175 μg /ml	+	−
	Creatine Kinase	250 μg/ml	+	−
	T7 RNA polymerase	67 μg /ml	+	−
	Template DNA (plasmid)	2 μg/ml	+	−
	S30 extract	9 μl (30%)	+	−
	S30 buffer[*b]	300 μl (30%)	−	+

a. 19 amino acids mix contains 10 mM DTT (0.75 mM final concentration).
b. S30 buffer is composed of 10 mM Tris–acetate (pH 8.2), 14 mM magnesium acetate, 60 mM potassium acetate, and 1 mM DTT.