

Table 3. Reduction of the SOR-Fe(CN)₆ complex by cell extracts in the presence of NADPH or NADH as electron donors

Electron donor	Redox state of the SOR-FeCN ₆ complex, units/mg*	
	[SOR-Fe ³⁺ -CN-Fe ²⁺ (CN) ₅], [†]	[SOR-Fe ³⁺ -CN-Fe ³⁺ (CN) ₅], [‡]
NADH	40	55
NADPH	82	90
None	0	0

Reductions were followed aerobically at 20°C in a spectrophotometric cuvette (120 µl final volume) containing 10 mM Tris·HCl (pH 7.6), 300 µM NADPH or NADH and 100 µM of the SOR-FeCN₆ complex. The reaction was initiated by adding 43 µg of a DH5α *Escherichia coli* soluble extracts. UV-visible spectra (250-1000 nm) were taken every 3 seconds using a HP 8453 diode array spectrophotometer. Initial velocities were calculated from the decrease of the absorptions at 645 nm (SOR-Fe³⁺ absorption band) and at 420 nm (Fe³⁺CN₆ absorption band). In all cases, the reduction of the SOR- FeCN₆ complexes was found to be reversible.

*One unit of activity is defined as the amount of cell extract catalyzing the reduction of 1 nmol of SOR Fe³⁺ by min.

[†]The SOR-Fe³⁺-CN-Fe²⁺(CN)₅ complex was prepared by mixing 100 µM SOR (as isolated in the ferrous state) with 100 µM of ferricyanide.

[‡]The SOR-Fe³⁺-CN-Fe³⁺(CN)₅ complex was prepared by mixing 100 µM SOR (as isolated, in the ferrous state) with 100 µM ferricyanide and then oxidized with 100 µM ammonium persulfate. The initial velocity was calculated from the decrease of the absorption at 420 nm (ferricyanide absorption band). It was found identical to that calculated from the decreased of the absorption at 645 nm (SOR Fe³⁺ absorption band).