



DIAPHORASE from *E. coli* (Lot I30203a)

Recombinant

E-DIAEC

06/15

(EC 1.8.1.4) protein-N6-(dihydrolipoyl)lysine:NAD⁺ oxidoreductase

PROPERTIES

1. ELECTROPHORETIC PURITY:

- Single band on SDS-gel electrophoresis (MW ~ 51,700)
- One major bands on isoelectric focusing (pI ~ 6.0)

2. SPECIFIC ACTIVITY:

12 U/mg protein at pH 9.0 and 25°C.

One Unit of diaphorase is defined as the amount of enzyme required to produce one μmole of NAD⁺ from NADH under the following assay conditions:

| | |
|-------------------------------|-------------|
| Tris.HCl buffer, pH 9.0 | 32.3 mM |
| Triton X-100 | 0.65% (v/v) |
| BSA | 0.16 mg/mL |
| Indonotrotetrazolium chloride | 0.5 mM |
| NADH | 0.46 mM |

3. OTHER ACTIVITIES (as a percentage of diaphorase activity):

| Enzyme Measured | Substrate | Activity, % |
|-----------------|-----------|-------------|
| Diaphorase | NADH | 100 |
| NADH oxidase | NADH | ~ 0.1 |

4. PHYSICOCHEMICAL PROPERTIES:

Recommended conditions of use are at pH 9.0 and up to 25°C.

5. STORAGE AND USE CONDITIONS/RECOMMENDATIONS:

The enzyme is supplied as an ammonium sulphate suspension and should be stored at 4°C. For assay, this enzyme should be diluted in 0.5 mM FAD containing 0.5 mg/mL BSA. **Swirl to mix the enzyme suspension immediately prior to use.**