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'One minute' transformation of competent *E.coli* by plasmid DNA

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In typical currently used transformation procedure (1,2), 100-200 µl of competent cells are mixed with plasmid DNA and the mixture is incubated 10-30 min at 0°C, 2 min at 42°C, and (after dilution in broth) 30-60 min at 37°C (usually with shaking). This procedure promotes a high yield of transformants and should be used when a very small number of plasmid molecules is available for transformation (for example, in cloning experiments). However, in many cases significant amounts of plasmid DNA (> 1 ng) are available. In such cases a sufficient number of transformants can be obtained by the following simple procedure: 3 µl of competent cells are mixed on ice in an eppendorf tube with 1 µl of plasmid DNA (1-100 ng DNA in 10 mM Tris, pH 8.0, 1 mM EDTA). The tube is transferred immediately to 44°C. After 1 min, 100 µl of broth are added and the whole mixture is immediately plated on selective media. The procedure works well with different E. coli strains (for example, W1485F$^-$, DH5α ,SCS1) made competent by simple $CaCl_2$ treatment (3), or a more complicated protocol described by D. Hanahan (4). Both freshly prepared and "old" (preserved for >1 year at -70°C) (2) competent E.coli were transformed equally well. The transformations were carried out with plasmids pBR322, pBR329 or their derivatives pDR1996 (10.6 kb) (5) and pKL1 (15.6 kb) (6). The frequency of transformation obtained using, for example, E.coli W1485F$^-$, made competent by $CaCl_2$ treatment (3), using plasmid pBR329, is $7x10^4$ Ap^R transformants/µg DNA. Plasmid DNA from " minilysates " (7) as well as DNA purified by centrifugation in CsCl gradient (1) can be used. Ampicillin (50 µg/ml), tetracycline (12.5 µg/ml) or chloramphenicol (10 µg/ml) has been used to select transformants on LB agar (1).

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