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The Structural Characterization of the Putative DNA-Binding Protein BldB from *Streptomyces Lividans*

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Mutants blocked at the earliest stages of morphological development in *Streptomyces* species are called *bld* mutants.

We have cloned *bldB* gene ORF from *S.lividans*. Genomic Southern blot analysis for main strains *S.lividans*, *S.seoulensis*, *S.coelicolor A3(2)*, and *S.griseus* indicated that *bldB* gene is conserved in all main *Streptomyces* strains. BldB protein expression was started from earliest stage of differentiation and finished at the stage of aerial mycelium.

The molecular weight of the BldB protein was determined using mass spectroscopy, SDS-PAGE, gel filtration column and analytical ultracentrifuge. The NMR data analysis shows that the full sequential assignment of BldB impossible. NOE values were poorly dispersed and negative NOE values appear only at the NH chemical shift region of 7.5 – 8.5 ppm and negative NOE values compose approximately 50%. It shows that the majority of BldB is unstructured and its conformation is flexible. Far and near-UV CD spectra of BldB were sensitive to protein solution pH, suggesting a proton induced conformation change. Thermal denaturation and renaturation profiles monitored by ellipticity at 220 nm show pH dependent specific curves, indicating a low cooperative and reversible one-step structural transition. BldB protein solution has a higher melting temperature, lower temperature-denaturation sensitivity, and more helix content at pH 5.0, indicating that BldB has relatively stable structure in acid environment. The ellipticity of BldB was decreased with increasing temperature, but far-UV CD spectra at high temperature of 80°C were remained with identical shape to native CD spectra without wavelength shift at 208 and 222 nm. It shows that BldB contains a thermostable structural content (“nucleus”).