

Nittaya Pitiwittayakul 2014: Molecular Characterization of *groESL* Operon from *Acetobacter pasteurianus* SKU1108 and Its Taxonomic Applications. Doctor of Philosophy (Genetic Engineering), Major Field: Genetic Engineering, Interdisciplinary Graduate Program. Thesis Advisor: Associate Professor Gunjana Theeragool, D.Agr. 201 pages.

The objectives of this study were to clarify the potential roles of the *groESL* gene of acetic acid bacteria (AAB), the effects of the disruption of the *groEL* gene, and the induction of the *groESL* gene expression into the thermotolerant acetic acid bacterium, *Acetobacter pasteurianus* SKU1108. When the *groEL* gene was disrupted, its rate of growth decreased and it exhibited a complete loss of toleration of stressors. The presence of the *A. pasteurianus* SKU1108 *groESL* gene in the *groEL* gene disruptant restored significant acceptance of stressors. A heat shock promoter was induced by stressors, as was shown by a steady increase in the transcription level of the *groESL* gene, along with elevated temperatures and a heightened concentration of acetic acid and ethanol. A CIRCE element which was found in the upstream region was examined and compared with all known genome sequences of AAB. This revealed that the sequences of all of these strains are very well preserved, and have evolved phylogenetically. Moreover, the *groEL* gene, which encodes 60-kDa heat shock proteins, was found to be conserved to a very significant extent among AAB. The *groEL* gene analysis was further used for classification and identification of AAB, together with 16S rRNA gene analysis. Twenty-four strains of AAB, which were isolated from fruits, flowers, mushrooms, and fermented rice products collected in Thailand by an enrichment culture approach and assigned to the genus *Acetobacter*, were taxonomically examined for the 16S rRNA gene and *groEL* gene sequences. Based on analysis of the 16S rRNA gene and *groEL* gene sequences, the strains were divided into ten groups: Group 1 comprised four isolates identified as *A. tropicalis*, Group 2 consisted of three isolates identified as *A. indonesiensis*, Group 3 was made up of two isolates identified as *A. persici*, Group 4 was composed of two isolates (*A. orientalis*), Group 5 possessed one isolate (*A. cibirongensis*), Group 6 contained three isolates (*A. pasteurianus*), Group 7 was constituted of one isolate of *A. papayae*, Group 8 held two isolates identified as *A. fabarum*, Group 9 consisted of one isolate of *A. okinawensis*, and Group 10 was made up of four isolates identified as *A. ghanensis*. Besides the 10 groups, one strain, BCC 15839, constituted an independent cluster which was taken from the type strains of other *Acetobacter* species in phylogenetic trees, based on 16S rRNA and *groEL* gene sequences. The polyphasic approach was implemented. Based on the results obtained in physiological, biochemical and genotypic analysis of the differences between the isolate and the type strains of the genus *Acetobacter*, the isolate was classified as a novel species of *Acetobacter*. The name *A. thailandicus* sp. nov., was introduced.

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Thesis Advisor's signature