Seiichi Katayama, J Bacteriol Parasitol 2014, 5:4
http://dx.doi.org/10.4172/2155-9597.S1.006

OMICS Group Conferences
Accelerating Scientific Discovery

2nd International Congress on
Bacteriology & Infectious Diseases
November 17-19, 2014 DoubleTree by Hilton Hotel Chicago-North Shore, USA

Function of the phased A-tracts upstream of the phospholipase C gene promoter in *Clostridium perfringens*

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Clostridium perfringens is a Gram-positive spore-forming anaerobic bacterium and a pathogen causing gas gangrene and food poisoning. Gas gangrene is mainly caused by α-toxin (phospholipase C) produced by *C. perfringens*. Three phased A_{5-6}-tracts (–66 to –40) lie upstream of the promoter for plc gene encoding the α-toxin. The results of gel retardation assays and hydroxyl radical footprintings revealed that the α subunit of *C. perfringens* RNA polymerase binds to the minor grooves of the phased A-tracts through its C-terminal domain (αCTD). The affinity of the α subunit for the phased A-tracts was estimated by surface plasmon resonance (SPR) [$K_d$was 6.1(α0.3)α10^{-8} M]. To identify the amino acid residues involved in the binding of the α subunit to the phased A-tracts, 27 amino acids residues in the α CTD were substituted with alanine. SPR analyses revealed that Arg261, Asn264, Gly292 and Lys294 in the α CTD were critical for the binding to the phased A-tracts. The topology of these amino acid residues on the predicted structure of *C. perfringens* α CTD indicated a contact path with the phased A-tracts that was similar to that of Escherichia coli α CTD with the upstream (UP) element. SPR analyses at different temperatures (15, 25 and 37°C) indicated that the affinity of *C. perfringens* α subunit for the phased A-tracts increased atlower temperatures, whereas that of *E. coli* α subunit for UP element did not. These results suggested that the phased A-tracts directly enhanced the *plc* gene expression in a low-temperature-dependent manner.

Biography

Seiichi Katayama has completed his PhD at the age of 32 years from Kagawa Medical School in Japan. He has been studying on an anaerobe, *Clostridium perfringens*. He learned genetic molecular bacteriology at Pasture Institute in France for about two years (1993 to 1995). He is an Associate Professor in the department of life science, Okayama University of Science. He has published more than 37 papers in some journals.

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