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行政院國家科學委員會專題研究計畫成果報告

DnaD 與 DNA 結合性質之研究

Study of DNA binding properties of DnaD

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一、中文摘要

DnaD是引子合成體的成員之一，參與了在革蘭氏陽性菌的染色體DNA複製的起始和重啟的過程中。DnaD蛋白質主要是由兩個結構區域所組成：分別為N端的聚合區域(oligomerization domain)和C端的單股DNA結合區域 (ssDNA binding domain)。此研究報告了從嗜高溫細菌 *Geobacillus kaustophilus* HTA426來的DnaD的N端區域(胺基酸1-128; DnaDn)於解析度2.3Å的結晶結構。DnaDn的結構顯示它具有一個伸延的翼型螺旋摺疊，即暗示DnaD是個具有典型雙股DNA結合區域的蛋白質。DnaDn在結晶狀態下形成四套體，但是在凝膠過濾法的結果中指出DnaD的此區域在溶液中是可以形成穩定的二套體。因此，DnaDn的結構分析可能提出了可以與DNA和DnaB的結合位置，與相關的革蘭氏陽性菌DNA複製引子合成體的聚集機制。

關鍵字：引子合成體、DnaD、PriB、DNA鍵結、DNA複製

Abstract

The DnaD is one of the primosomal proteins that are required for initiation and re-initiation of chromosomal DNA replication in Gram-positive bacteria. The DnaD protein is composed of two major structural domains: an N-terminal oligomerization domain and a C-terminal ssDNA binding domain. Here, we report the crystal structure of the N-terminal domain (aa 1–128) of DnaD (DnaDn) of *Geobacillus kaustophilus* HTA426 at 2.3 Å resolution. The structure of DnaDn reveals an extended winged-helix fold, a typical double-stranded DNA binding motif as winged-helix proteins. DnaDn formed tetramers in the crystalline state, but the results of gel filtration chromatography further indicated that this domain of DnaD was a stable dimer in solution. The structural analysis of DnaDn may suggest the binding sites for DNA and DnaB, and an assembly mechanism for Gram-positive bacterial DNA replication primosome.

Keywords: Primosome, DnaD, PriB, DNA binding, DNA replication

二、Introduction

Initiation and re-initiation of chromosomal DNA replication in bacteria for genome

duplication is a complex process that relies on divergent multi-protein assembly for entry of the replicative DNA helicase at the replication origin [1]. In *Escherichia coli*, loading the replicative DNA helicase DnaB for *oriC* -dependent DNA replication is carried out with the aid of two other proteins, DnaA and DnaC [1]. The replication forks could accidentally be arrested anywhere along the DNA, so reloading DnaB helicase for *oriC*-independent DNA replication is required [2,3]. To date, there are two DnaB helicase-recruiting pathways: PriA-PriB-DnaT-DnaC dependent reactions are most robust on fork structures with no gaps in the leading strand, while the PriC-DnaC dependent system preferentially utilizes fork structures with large gaps in the leading strand [4].

The mechanisms of DNA replication in bacteria have mainly been studied at the Gram- negative *E. coli* and, to a lesser extent, at the Gram-positive bacteria. In Gram-positive *Bacillus subtilis*, both DNA replication initiator proteins DnaA and PriA have homologs of *E. coli*, suggesting that at least the initial stages of DNA replication are similar. Nevertheless, essential helicase-loading components such as PriB, PriC, DnaT, and DnaC proteins are not found in *B. subtilis*. Instead, three other proteins, DnaD, DnaB, and DnaI, have been genetically and biochemically proven to be required for replication initiation of *B. subtilis* [5–10]. Based upon weak sequence homology, DnaI is believed to be the Gram-positive functional counterpart of *E. coli* DnaC [7]. The DnaD and DnaB proteins have no homologs in Gram-negative bacteria and their functions for DnaA-dependent and PriA-dependent initiation of DNA replication are thus needed to be addressed.

DnaD interacts with DnaA [11], PriA [8], DnaB [8], and DnaD itself [11,12]. DnaD binds with PriA on forked DNA and stimulates DnaB loading activities [8]. DnaD also functions as a global regulator of DNA architecture [10]. DnaD interacts with linear DNA and forms a nucleoprotein structure with a round DnaD scaffold in an open circle around the periphery [13]. DnaD is present mainly as a stable dimer but also may form higher-order oligomers [11]. DnaD consists of two domains with distinct activities: an N-terminal domain with oligomerization activity, and a C-terminal domain (DnaDc) with weak ssDNA-binding activity [12]. Although DnaD can bind to DNA and form large nucleoprotein complexes in the absence of the N-terminal domain, it does not exhibit the DNA remodeling activity [12].

Very recently, the crystal structure of DnaDn from *B. subtilis* (BsDnaD) has been reported, which reveals an extended winged helix fold and a unique tetramerization motif for the DnaD-mediated scaffold formation [14]. Nevertheless, we found that most residues critical for the tetramer formation of BsDnaD, if not all, are not conserved among *bacillus*- related species. This finding raises an interesting question as to whether forming tetramer or oligomer is functionally important for the initiation and re-initiation of chromosomal DNA replication in Gram-positive bacteria. In this study, we have cloned, expressed, purified, and crystallized DnaDn protein from *Geobacillus kaustophilus* HTA426 (*Gk*DnaDn), and determined its structure at 2.3 A resolution. Based on its structural features and the results of gel filtration chromatography, the structure-function relationships of DnaDn are discussed.

三、Result and Discussion

The structure of the N-terminal domain of DnaD monomer - We crystallized a selenomethionine-incorporated variant of DnaDn (Fig. 1A) and determined its structure to 2.3 A resolution. MAD phasing techniques were used to generate an electron density map that was readily interpretable. Briefly, the

overall structure of each one DnaDn (Fig. 1B) consists of six α-helices, three β-sheets, one wing, and a metal ion. α-helices of DnaDn are at residues 3–12 (α1), 19–28 (α2), 32–47 (α3), 54–60 (α4), 65–78 (α5), and 99–117 (α6). One turn is found between α2 and α3. β-sheets of DnaDn are at residues 14–18 (β1), 82–84 (β2) and 96–98 (β3), where β2 and β3 form the antiparallel strands. A metal ion is coordinated with the residues at α2 and α6. A β2-loop-β3 motif, or a "wing", formed at residues 85–95, is a highly flexible region. The majority of the electron density for DnaDn is of good quality, but a discontinuity is observed for residues 85–95, suggesting that this region is dynamic.

Four identical polypeptide chains of DnaDn form a tetramer (dimer of dimer) with a non-crystallographic 4-fold symmetry. The structures of the four crystallographically independent DnaDn were superimposed with a root mean square deviation of 0.5 for C-α atoms of secondary structural elements and showed no distinct differences. A search of the PDB using the program DALI revealed close similarity of the structure of DnaDn monomer to that of the DNA binding domain in the winged-helix proteins for which structural information exists, such as LexA repressor (Fig. 1C) and MotA transcription factor ($Z > 7.0$), but DnaDn shares no similarity with them at the level of amino acids sequence. Despite no sequence identity with the wingedhelix proteins, structure of DnaDn displays a winged-helix fold (α3-α4-turn-α5-wing) topologically similar to that of DNA binding domain of winged-helix proteins [22,23]. Three structural differences are found among DnaDn and the winged-helix proteins. First is that DnaDn does not have the C-terminal wing found in the winged-helix domain of HNF-3γ [24]; the wing of the canonical winged-helix proteins is replaced by an α-helix (α6) in DnaDn. Second, extra region α1-β1-α2 motif is found in DnaDn and makes its dimerization forces stronger. Third, the electron density map of the DnaDn structure shows a metal ion at α2 and α6; for which it is only found at the C terminus of recognition helix of winged-helix protein ILF [23]. We assumed that it was a magnesium ion because that metal was used for crystallization of DnaDn. While the structure of DnaDn monomer is dominated primarily by hydrophobic core, the solvent-accessible surfaces shows two positively charged regions found at α1 (Lys3, Lys4, and Lys5) and α5 (Arg73, Arg74, and Lys78). Since Arg73, Arg74, and Lys78 of DnaDn located at the typical recognition helix of DNA binding motif HTH, they may play a role for DNA interaction (see below).

Dimer and oligomer in crystalline state -
Crystal structure of DnaDn shows that the two subunits are tightly associated (Fig. 1D). The contacts of DnaDn monomers are made through many hydrophobic interactions. As shown in Fig. 1E, the monomer–monomer interface of DnaDn is stabilized by the hydrophobic core: Typ9 (α1), Ile15 (β1), Val17 (β1), Leu20 (α2), Leu22 (α2), Tyr25 (α2), Leu35 (α3), Val36 (α3), Leu39 (α3), Met61 (α4), Leu110 (α6), and Tyr111 (α6). When using a watersized probe of radius 1.4Å, the loss of solvent-accessible surface area upon dimerization of DnaDn was calculated to be 3386 $Å^2$, where over two-thirds (2320 $Å^2$) of the buried surface in DnaDn dimer was contributed by hydrophobic interactions.

In the crystal of DnaDn, the four molecules formed two pairs of dimers, A-B and C-D, respectively (Fig. 1F). Since the two dimers of DnaDn associate via slight contact to create the tetramer, it was thought that the tetrameric state may be possibly due to crystalpacking forces (Fig. 1F). We noted that in crystal, another crystallographically related tetramer A-B-A′-B′ was formed and further

stabilized via strong hydrophobic interactions (Fig. 1G). The hydrophobic residues dominated the interface that buried 2885 Å^2 of surface area per dimer. This unique tetramerization motif in the structure of GkDnaDn has been found in that of BsDnaDn [14]. These structural findings may suggest that if oligomerized, the N-terminal region of DnaD dictated the self- association.

Oligomerization of GkDnaDn in solution - Since the structure implied that *Gk*DnaDn could form tetramers in the crystalline, it needed biochemical verification to confirm its oligomerization state. We carried out gel filtration chromatography on this protein, and the results revealed that only single peak was found (Fig. 2). Based on the assumptions of similar shape and partial specific volume as compared with standard proteins, the native molecular weight of *Gk*DnaDn was estimated as ~ 28 kDa. Thus, *Gk*DnaDn was a stable dimer in solution.

Conflicting reports exist on the oligomeric status of *Bs*DnaDn and *Gk*DnaDn. Although glycerol density-gradient centrifugation studies and cross-linking analysis with glutaraldehyde have showed that full-length *Bs*DnaD forms as a stable dimer in solution [11], *Bs*DnaDn has been reported to be a tetramer examined by sedimentation velocity studies and cross-linking analysis with succinimidyl octaester (SOXL) [12]. In contrast to *Bs*DnaDn, this study shows that *Gk*DnaDn was a dimer in solution. Are these due to inherent differences between the two species? Then we went back to check the tetrameric interactions reported by the structure of *Bs*DnaDn [14]. According to the structural information, 3 polar/electrostatic interaction- pairs Lys3(B)-Glu108(A′), Tyr107(B)-Met1(A′), and Met1(A)-Met1(A′), and 12 van der Waals interaction- pairs Lys3(B)-Gly104(A′), Lys3(B)-Glu108(A′), Phe6(B)-Tyr107(A′), Phe6(B)-Ile110(A′), Ile7(B)-Gln100(A′), Ile7(B)-Gly104(A′), Gln10(B)-Trp103(A′), Glu11(B)-Gln 100(A′), Thr16(B)-Thr16(A′), Met1(A)-Met1(A′), Phe6(A)-Met9(A′), and Met9(A)-Gln10(A′) are found to be crucial for a tetramer formation of *Bs*DnaDn [14]; however, all of them are not conserved between *Gk*DnaDn and *Bs*DnaDn, except for the first amino acidMet1. This may be the reason that *Gk*DnaDn could not form a tetramer as *Bs*DnaDn since these critical residues are not found in *Gk*DnaDn.

Possible roles of the potential tetramer formation surface of GkDnaDn - DnaD interacts with PriA, DnaA, DnaB, and DnaD itself. It has been reported that the C-terminal domain of DnaD may play a role not only in ssDNA binding, but also interacts with DnaA and PriA. The yeast two-hybrid analysis has showed that the residues 133–140 of DnaD are an essential region for interacting with DnaA [11], and the dnaD2021 mutation (Glu141 mutated to Lys) of DnaD may result in a defect in the interaction with PriA [25]. The DnaB–DnaD interactions are still unknown yet. Despite GkDnaDn forming a dimer, the dimer–dimer interface caused by crystal-packing forces contain a strongly hydrophobic patch that may mediate protein–protein interactions like *Bs*DnaDn tetramer. We found that a distinct hydrophobic patch of *Gk*DnaDn at residues 4–30 has a strong similarity to the N-terminal domain of *Gk*DnaB protein (residues 27-58). This spans a 32-residue stretch with 32% identity and 41% similarity, suggesting a potential region of DnaD for DnaB binding. However, how DnaD interacts with DnaB has not been reported, and this possibility requires further investigations.

Possible roles of winged-helix fold of DnaDn - It has been reported that *Bs*DnaDc cannot untwist duplex DNA [26]. The structures of DnaDn reveal a winged-helix fold as winged-helix proteins. Winged-helix proteins are almost DNA binding proteins via its recognition helix and the wing to interact either with the major grooves or with the minor grooves of DNA. Recently we solved the DNA-complexed structures of two winged-helix proteins, FOXK1a [23] and FOXO3a [22], which showed that the DNA-binding elements of a winged-helix fold are located on the most basic electrostatic surface of a protein [22–24]. As shown in Fig. 3, three positively charged residues Arg73, Arg74, and Lys78 of *Gk*DnaDn may be directly involved in DNA binding because they are exposed on the surface of α5, like the recognition helix of winged-helix proteins. In addition, these positions are highly conserved. Since the DNA remodeling activity of DnaD is sum of the N-terminal oligomerization domain and the C-terminal DNA binding domain [12,26], the role for the DnaDn-DNA interactions may not be ruled out. To date, there is only one DNA-complexed structure of a dimeric winged-helix protein, the replication terminator protein (RTP), found in the PDB [27]. The structure of the RTP-DNA complex revealed that while the two recognition helices of RTP are in close contact with the DNA major grooves, the wings and N-termini of RTP do not form intimate contacts with the DNA. The overall structure of a *Gk*DnaDn dimer is similar to that of RTP. According to the structure of RTP-DNA complex, we manually superimpose the location of duplex DNA with our DnaDn structure, and propose a putative model for GkDnaDn-DNA interactions (Fig. 3).

The DNA binding region proposed here on a winged-helix fold of GkDnaDn partially overlaps the binding sites for DnaB mentioned above. The Gram-negative bacterial primosome assembly provides a good example of how protein–protein interaction can affect protein–DNA interaction. A dynamic primosome assembly process in which single-stranded DNA is handed off from one primosome protein to another as a repaired replication fork is reactivated [28]. Like that of PriB, DnaD is also second to assemble the Gram-positive bacterial primosome [8], suggesting that a general DNA replication restart mechanism was used between Gram-positive and Gram-negative bacteria.

Proposed mechanism of DnaD for its supercoiled DNA opening activity - DnaD is a potential modulator for global superhelical density [26]. Although Soultanas et al. have provided sufficient data supporting the formation of scaffolds of *Bs*DnaDn, it was thought that this characteristic may be possibly due to its non-specific DNA binding activity like that of PriB [10,12,14]. Our previous EM and crystal structural studies of PriB showed that PriB molecules can cooperatively bind to φX174 DNA. Nevertheless, the precise function of PriB in vivo seems unlikely to form a high-density nucleoprotein complex [16]. It is attractive to speculate that, DNA remodeling activity of DnaD may be required to stabilize the supercoiled DNA in the earlier stage of DNA replication initiation. The crystal structure of *Gk*DnaDn present here may suggest an interaction with duplex DNA via the residues Arg73, Arg74, and Lys78 of *Gk*DnaDn (Fig. 3). In order to open supercoiled DNA, the first step of DnaD action is to guide and recognize DNA. This may be a driving force

to deform supercoiled DNA and then rotate it to convert to an open circular form.

Submission of atomic coordinates and related structure factors of DnaDn - Atomic coordinates and related structure factors of DnaDn have been deposited in the Protein Data Bank (PDB) with identification code 2vn2.

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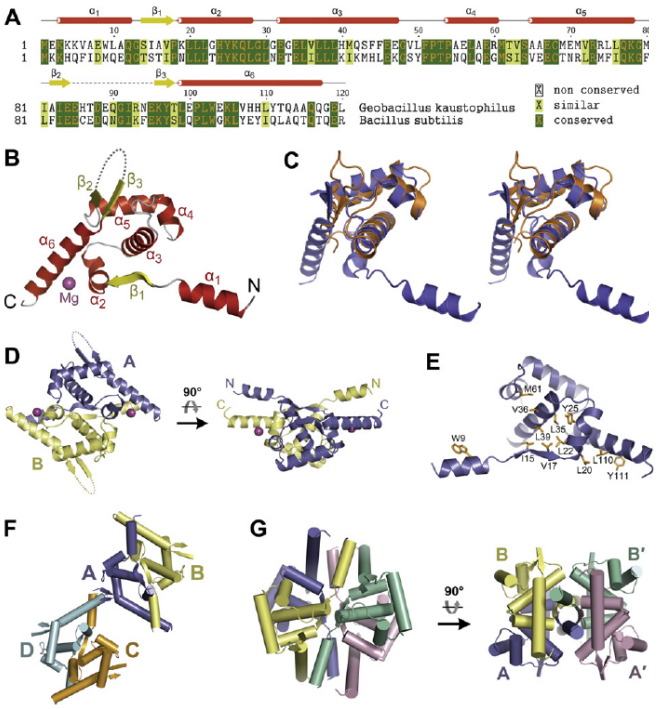

Fig. 1. The structural analysis of *Gk*DnaDn. **(A)** Structure-based sequence alignment of DnaDn proteins from *G. kaustophilus* HTA426 and *B. subtilis*. The labeled secondary structural elements derived from this work are shown above the alignment. **(B)** Ribbon diagram of a DnaDn monomer with the secondary structures labeled. **(C)** Stereo view of structural superimposition of DnaDn (in *blue*) and the DNA binding domain of LexA repressor (PDB code 1LEA). This superimposition indicates a structurally conserved core helix-turn-helix- wing motif with additional N-terminal α1-β1-α2 motif and C-terminal helix (α6) region. **(D)** Ribbon diagram of a DnaDn dimer. Each DnaDn monomer is color-coded. **(E)** The crucial residues of DnaDn for the hydrophobic core of the dimer interface. **(F)** An asymmetric unit contains four crystallographically independent DnaDn monomers A-B-C-D. **(G)** Crystallographically related tetramer A-B-A′-B′ is formed via strong hydrophobic interactions.

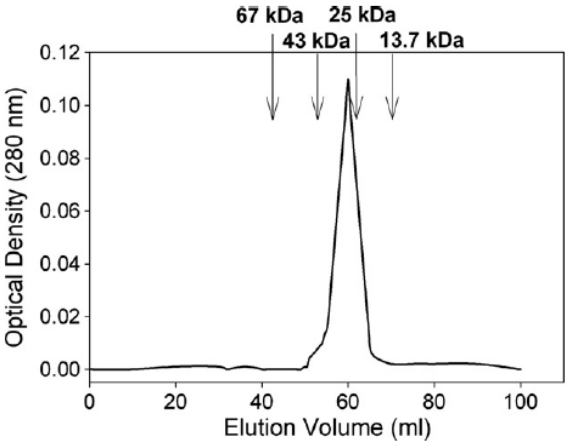

Fig. 2. The gelfiltration chromatographic analysis of DnaDn. The sizes of the molecular mass markers are indicated on the top in kDa. The corresponding peak shows the eluting DnaDn protein.

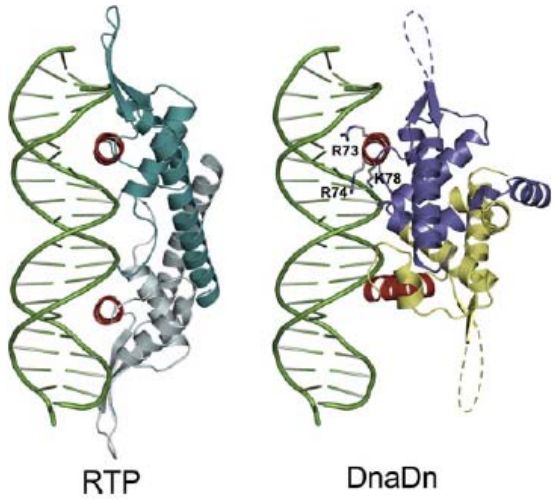

Fig. 3. Proposed model for a DnaDn dimer-DNA interaction. According to the structurally conserved helix-turn-helix-wind motif of DnaDn with that of a dimeric winged-helix protein RTP (PDB code, 1f4k), and three positively charged residues Arg73, Arg74, and Lys78 are exposed on the surface of α5 (in *red*) of DnaDn as the recognition helix (in *red*) of winged-helix proteins, we proposed a putative model based on these observation.

Table 1. Data collection and refinement stastistics of DnaDn.

Data Collection			
Space Group		F222	
a, b, c (Å)	116.51,	124.71,	157.23
	Peak	Inflection	Remote
Wavelength(Å)	0.9794	0.9795	0.9643
Resolution (Å)	2.3	2.3	2.3
Rmerge (%)[a]	10.6 (37.0)[b]	11.0 (55.5)	9.4 (48.4)
$< I/\sigma (I)>$	22.59 (9.20)	12.55 (2.70)	24.55 (5.51)
Completeness	99.3 (100.0)	97.4 (98.9)	97.5 (99.0)
Redundancy	12.3 (12.5)	5.0 (5.0)	10.0 (10.0)
Refinement Statistics			
Resolution (Å)	27.5 ~ 2.3		
No. reflections	46,920		
R_{work}/R_{free} (%)	21.7/22.2		
No. atoms			
Protein	3470		
Ion	4		
Water	178		
B-factor			
Protein	34.52		
Ion	26.05		
Water	45.88		
R.m.s deviations			
Bond lenths (Å)	0.024		
Bond angles (°)	2.6		

[a]Rsym = $\Sigma_h\Sigma_i|I_i-I|\Sigma_h\Sigma_I I$, where I is the mean intensity of the I observations of reflection h.

[b]Numbers in parentheses refer to the highest resolution shell.

赴國外出差或研習心得報告

由於此計畫所需的 DnaD 蛋白質晶體繞射結果已先在本國的同步輻射中心獲得，原核定之欲赴國外差旅費用(至日本 Spring-8 使用同步輻射中心)已流至同會計項目使用。故本次無赴日本出差。

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<tr><td colspan="3">服務單位及職稱
Institute & Position</td><td colspan="5">中山醫學大學生物醫學科學系 助理教授</td></tr>
<tr><td colspan="3" rowspan="2">會議名稱
Title of Meeting</td><td colspan="5">中文：第 6 屆亞太生物物理研討會暨第 27 屆香港神經科學學會年會
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NSC 97-2311-B-040-001</td></tr>
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Nature</td><td colspan="3">□政府機關 Government
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The Biophysical Society of Hong Kong
The Hong Kong Society of Neurosciences</td></tr>
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Assisting Group</td><td colspan="3"></td></tr>
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Importance</td><td colspan="5" rowspan="2">涉及我與會之 Affects Our □會籍 Membership □名稱 Title □權利 Voting Rights □地位 Position
請說明 Remarks ：純學術性會議與政治無關</td></tr>
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Participating Countries</td><td colspan="5"></td></tr>
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□不確知 Don’t Know</td></tr>
<tr><td colspan="2" rowspan="2">發表之論文
Title(s) of Lecture(s) or Poster(s)</td><td colspan="5">中文：DnaD 之 N 端功能性區域蛋白質結晶結構: 以分子的層次了解 DNA 引子合成體的組裝</td></tr>
<tr><td colspan="5">英文：Crystal structure of the N-terminal domain of DnaD protein: a molecular insight into the DNA replication restart primosome assembly.</td></tr>
</table>

＊ 本表請附於會後報告前頁，篇幅如不敷使用，可另備**A4** 紙張橫式繕打。（**Please attach this form to your report**；**use additional A4 blank pages if needed.** ）

填表人簽章 signature 黃晟洋 填表人電話 Tel: 0960521555

行政院國家科學委員會補助國內專家學者出席國際學術會議報告

一、參加會議經過

The 6th Asian Biophysics Association (ABA) Symposium was held from Jan 11 to 15, 2009 in Hong Kong, ROC. As the meeting is based on the biophysics for the main shaft, so lots of the participating researchers are in the fields of macromolecular interactions in which I am rather interested. In the lecture section of the protein-nucleic acid and protein-protein interaction, particularly such as Dr. Iksoo Chang, Dr. Jacqui Matthews, Dr. Julian Lee, Dr. Hanna Yuan, Dr. Dipankar Chatterji and so on, I am deeply attracted to learn more about useful and efficient strategies to develop my research scenario. I also contributed a post presentation (PS1-10). The title is "Crystal structure of the N-terminal domain of DnaD protein: a molecular insight into the DNA replication restart primosome assembly."

二、與會心得

I would briefly elucidate those summaries of my most impressive lectures as following:

1). The talk by Dr, Iksoo Chang, entitled "Cooperative folding kinetics of BBL protein and peripheral subunit-binding domain homologues", emphasizes the folding mechanism of BBL and peripheral subunit-binding domain homologues. His data showed that proteins undergo a gradual folding, and in a meanwhile, folding evens along the continuous folding pathway also could be mapped out from the equilibrium denature experiment. Through a series of the exact calculation of physical and chemical force between amino acids, they would get the nature of a two-state cooperative transition as the kinetic interaction of BBL protein and peripheral subunit-binding domain homologues. In conclusion of his talk, results from their research indicated that an unequivocal resolution to the fundamental examination on protein fast-folding mechanism once presumably cooperative interaction of two proteins.

2). The talk by Dr. Jacqui Matthews, entitled "A structure basis for cell type-specific assembly of complexes recruited by LIM homeodomain transcription factors", describes that various LIM-homeodomain transcription factors selectively enable to exert combinatorial interactions to guide cell type-specific assembly. For example, the combinatorial effects of two different LIM-homeodomain proteins, Lhx3 and Ldb1, specify the formation of V2 interneuron. The addition of Isl1 instead specifies motorneurons via assembly of a tertiary complex in which Isl1 contacts Lhx3 and Ldb1, displacing Lhx3 as the binding partner of Ldb1. Again, they have identified the 30-residue Lhx3-interaction domain (LID) on Isl1 through X-ray structures data. Those data provide a structure basis for the formation of cell type-specific protein-protein interactions, discovering likely prototypic strategy for how competition of protein binding by linear motifs with diverse sequences can regulate important biological events.

3). The talk by Dr. Julian Lee, entitled “Sequence-independent backbone interactions and sequence-dependent side chain interactions on protein folding”, mainly focuses on a molecular insight into protein folding that relies on the effect of sequence-independent backbone interactions and sequence-dependent side chain interactions. In their team work, they has predicted 10 proteins only with Lennard-Jones interaction between backbone atoms and examined their protein folding using fragment assembly and physical energy function. Their results show that native-like structures for proteins of interests are able to be determined mainly by sequence-independent backbone interactions. Furthermore, non-local hydrophobic side-chain interactions in the sequence-dependent manner are required for native-like structure.

4). The talk by Dr. Hanna Yuan, entitled “Structural insights into TDP-43 in nucleic acids binding domain interactions”, elucidates that the specific domain of TDP-43 directly enables to bind DNA/RNA to regulate the alternative expression of related genes. In case of loss or gain of the TDP-43 function, numerous genetic diseases, even lethality will occur. Therefore, their team work mainly finished the resolution to crystal truncated TDP-43 protein in complex with a 10mer DNA at a resolution of 1.65 Á to learn more about biochemical properties and how to interact with DNA/RNA. Their data reveal that TDP-43 is a dimeric protein with two RRM domains, both involved in DNA/RNA binding. The basis of TDP-43’s TG/UG preference in nucleic acids binding has been shown. In addition, C-terminal RRM domains in TDP-43 has an atypical RRM fold that plays a role in protein-protein interactions.

5). The talk by Dr. Dipankar Chatterji, entitled “Protein-protein interaction and its functional implication with respect to DPS protein in mycobacteria”, provides the information about the relationship of the interaction between DNA and Mycobacteria DPS protein that enables to form a trimer, by the treatment of certain conditions. In addition, they also successfully gain the resolution to the crystal DPS protein to explain the important role of N-terminal and C-terminal for dodecamerization and DNA-binding potential ability. Interesting, their results from crystal data of DPS protein show that DPS trimers presumably exist in two forms, DPS-like and Ferritin-like. Only one of them can, however, convert to a dodecamer at 37℃. Furthermore, they has evidenced that the formation of dodecamers resulting from the coordinative interaction of trimers involves some key residues at the interface of DPS proteins, through site-directed mutagenesis method. Similar, they can generate a monomer too by specific mutation opening up new ways to study protein assembly.

三、攜回資料名稱及內容

The 6th Asian Biophysics Association (ABA) Symposium program and abstracts book.