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**When Crystal Structure Does Not Seem to Depict Physiologically Relevant Conformation - A Novel Calmodulin Conformation in Calmodulin and Calcineurin-Peptide Complex.** Zongchao Jia, Qilu Ye, Xin Li, Andrew Wong, Qun Wei, Biochemistry, Queen's Univ., 18 Stuart St., Kingston ONTARIO K7L 3N6, CANADA.

Calcineurin is a calmodulin-binding protein in brain and the only serine/threonine protein phosphatase under the control of Ca<sup>2+</sup>/calmodulin (CaM) which plays a critical role in coupling Ca<sup>2+</sup> signals to cellular responses. CaM up-regulates the phosphatase activity of calcineurin by binding to CaM-binding domain (CBD) of calcineurin subunit A. We have determined crystal structures of CaM bound to a CBD peptide in two forms and different space groups. The first one is a chimeric construct containing CaM and the CBD peptide covalently fused through a glycine linker; the second form is a conventional complex structure between CaM and the peptide obtained through co-crystallization. Unexpectedly, both structures display an intimate homodimer, in which CaM possesses a native-like extended conformation and the CBD peptide shows  $\infty$ N-helical structure. The N-terminal lobe from one CaM and the C-terminal lobe from the second molecule form a combined binding site to trap the peptide. Thus the dimer provides two binding sites, each of which is reminiscent of the fully collapsed conformation of CaM commonly observed in complex with, for example, MLCK peptide. The interaction between the peptide and CaM is highly specific and similar to MLCK. However, the novel CaM binding conformation is “deemed” physiologically irrelevant because the full-length calcineurin and CaM forms 1:1 complex as observed in early studies and because of the fact that CaM-peptide construct exists predominantly as a monomer in solution. It is thus extremely intriguing that three independent crystal structures obtained from different constructs and under different conditions persistently reveal a physiologically “irrelevant” conformation. Investigating the cause of this unique observation would be interesting and informative.