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Integrative approach to structure of huge protein complex in Kai-clock protein system

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Kai-clock system is one of the simplest biological clocks: the system is composed with only three proteins, KaiA, KaiB and KaiC, and they repeat association-dissociation with 24-hrs period as follows, $A_2+B_4+C_6 \rightarrow A_2C_6+B_4 \rightarrow A_2+B_6C_6 \rightarrow A_{12}B_6C_6 \rightarrow A_2+B_4+C_6 \rightarrow \dots$. To understand this system, it is essential to reveal the complex structures in every phase. For this purpose, we have been focused on the largest complex, $A_{12}B_6C_6$, which could appear at the turning point from the association phase to the dissociation phase. Basic structure of $A_{12}B_6C_6$ was reported with cryo-EM observation by J. Snijder, et al.. However, the reported structure missed the N-term domains of KaiA. Here, we introduced the whole structural model of $A_{12}B_6C_6$ in solution. To analysis this huge complex in solution, we utilized two state-of-art solution scattering techniques, SEC-SAXS and SEC-inversed Contrast Matching-SANS (SEC-iCM-SANS). In addition, we combined computational modeling and molecular dynamics simulation to build three-dimensional structure and to clarify its dynamics. We should note that the iCM-SANS selectively observing the KaiA protomers in the complex played crucial role in the analysis. The detailed will be shown in the presentation.

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