Bioinformatics Investigation of the Twin-Arginine Translocation pathway in Cyanobacteria

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Protein translocation systems are essential in living organisms to transport proteins to specific locations for required cellular functions, as correct localization is essential for different cellular activities. One of the significant translocation systems is the twin-arginine translocation (TAT) system, which transports fully folded proteins in plants and bacteria, thus making it important in biotechnology and pathogenic studies. The *Synechocystis sp. PCC 6803*, a model photosynthetic organism, possesses two distinct membranes with single genes that code for the TAT transport system components. Evidence has shown the presence of the activity of twin-arginine transporters in both the thylakoid and the plasma membrane. However, the difference in the protein sorting process between the two membranes and what guides protein translocation to the thylakoid or plasma membranes using the same TAT system is still unknown. Using predictive algorithms and bioinformatic tools, we have identified Synechocystis TAT proteins that are predicted to be localized to the plasma and thylakoid membranes, respectively. Across several cyanobacteria species, we also observed the occurrence of one of the TAT components (TatA) almost invariably in the plasma membrane. At the same time, the other component (extendend length version we dub $TatA_L$) was present in both the plasma and thylakoid membranes. There was also a major difference in the amino acids in the transmembrane region in the two of the three components that make up the TAT system in cyanobacteria species. Other results also show a close relationship between plants (e.g., *Pisum sativum* or *Arabidopsis thaliana*), *Bacillus subtilis*, and the *Synechocystis* TAT systems. The *B. subtilis* TAT is interesting because it is composed of only two components (TatA and a cognate TatC); whereas, the other TAT systems have three distinct components (e.g., TatA, TatB, and TatC). These results suggest that *Synechocystis* uses a unique process by which specific proteins are directed and transported across the thylakoid and plasma membranes and involves a dual TAT system that combines elements of the plants and *Bacillus subtilis* TAT system. This dual system could be represented by the plasma membrane primarily utilizing a plant-type 3-membered TAT system, while the thylakoid membrane employs a gram-positive 2-membered TAT system. The unique TAT system allow *Synechocystis* to transport different precursors across the thylakoid or plasma membranes, respectively, with the individual TAT components playing a significant role in distinguishing translocation to the two distinct membranes.