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	2019
	Carotenoid in the eyespot apparatus is required for triggering phototaxis in <i>Euglena gracilis</i> .
	Plant Journal, 20 October 2019 https://doi.org/10.1111/tpj.14576
	Shota Kato* (1, 2), Kazunari Ozasa (3), Mizuo Maeda (3), Yuri Tanno** (4), Shun Tamaki (1), Mieko Higuchi-Takeuchi (3), Keiji Numata (3), Yutaka Kodama (4), Mayuko Sato (3), Kiminori Toyooka (3), Tomoko Shinomura (1) (1) Institute for Basic Science, DGIST, Republic of Korea (3) 4 (* **H3O)
	(Euglena gracilis) Plant Journal
	(C)
	Carotenoids are the most universal and most widespread pigments in nature. They have played pivotal roles in the evolution of photosensing mechanisms in microbes and of vision in animals. Several groups of phytoflagellates developed a photoreceptive organelle called the eyespot apparatus (EA) consisting of two separable components: the eyespot, a cluster of carotenoid-rich globules that acts as a reflector device, and actual photoreceptors for photobehaviors. Unlike other algal eyespots, the eyespot of Euglenophyta lacks reflective properties and is generally considered to act as a shading device for the photoreceptor (paraflagellar body, PFB) for major photomovements. However, the function of the eyespot of Euglenophyta has not yet been fully proven. Here, we report that the blocking carotenoid biosynthesis in <i>Euglena gracilis</i> by suppressing the phytoene synthase gene (<i>crtB</i>) caused a defect in eyespot function resulting in a loss of phototaxis. Raman spectroscopy and transmission electron microscopy suggested that <i>EgcrTB</i> -suppressed cells formed eyespot globules but had a defect in the accumulation of carotenoids in those packets. Motion analysis revealed the loss of phototaxis in <i>EgcrTB</i> -suppressed cells: a defect in the initiation of turning movements immediately after a change in light direction, rather than a defect in the termination of cell turning at the appropriate position due to a loss of the shading effect on the PFB. This study revealed that carotenoids are essential for light perception by the EA for the initiation of phototactic movement by <i>E. gracilis</i> , suggesting one possible photosensory role of carotenoids in the EA for the phototaxis.

