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Transcriptome sequencing and metabolic pathways of astaxanthin accumulated in *Haematococcus pluvialis* mutant under 15% CO_2

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Abstract:

Transcriptome sequencing and annotation was performed on *Haematococcus pluvialis* mutant red cells induced with high light under 15% CO_2 to demonstrate why astaxanthin yield of the mutant was 1.7 times higher than that of a wild strain. It was found that 56% of 1,947 differentially expressed genes were upregulated in mutant cells. Most significant differences were found in unigenes related to photosynthesis, carotenoid biosynthesis and fatty acid biosynthesis pathways. The pyruvate kinase increased by 3.5-fold in mutant cells. Thus, more pyruvate, which was beneficial to carotenoids and fatty acid biosynthesis, was generated. Phytoene synthase, zeta-carotene desaturase, lycopene beta-cyclase involved in β-carotene biosynthesis in mutant cells were upregulated by 10.4-, 4.4-, and 5.8-fold, respectively. Beta-carotene 3-hydroxylase catalyzing conversion of β-carotene into astaxanthin was upregulated by 18.4-fold. The fatty acid biosynthesis was promoted because of the upregulation of acetyl-CoA synthetase and acetyl-CoA carboxylase, thus increasing astaxanthin esterification and accumulation in mutant cells.

Keywords: *Haematococcus pluvialis*, astaxanthin, mutant cells, transcriptome sequencing, metabolic pathways

1. Introduction

Microalgae production is widely considered for removing CO_2 from the flue gas of coal-fired power plants, and its biomass can be a feedstock for biodiesel and various high valued products, such as astaxanthin (Cheng et al., 2016a; Williams and Laurens, 2010; Zheng et al., 2016). *Haematococcus pluvialis* with the highest astaxanthin content in nature is the only commercialized microalgae in astaxanthin production. Astaxanthin accumulation is generally assumed to be a survival strategy adopted by *H. pluvialis* grown in stressful environments, which is important in energy and carbon storage, and in quenching the reactive oxygen species generated during stress response (Lemoine and Schoefs, 2010; Li et al., 2008; Yuan et al., 2011). However, the mechanism underlying the enhanced astaxanthin yield of *H. pluvialis* mutant under high concentration of CO_2 has not been fully documented.

Transcriptome sequencing is an efficient approach for obtaining microalgae functional genomics information. This method has identified the key pathways and important genes for biofuel production, molecular changes promoting or accompanying triacylglycerol accumulation, and molecular mechanism of carbon sequestration(Rismani-Yazdi et al., 2011; Cheng et al., 2014; Fan et al., 2016; Miller et al., 2010). Many studies have also discussed the gene transcript and metabolic changes of *H. pluvialis* responding to various stress (e.g., light, salinity, temperature, hormone, iron and inorganic carbon) (Gao et al., 2015; Hong et al., 2015; Li et al., 2010; Wen et al., 2015). Protein-encoding genes involved in the