

Using QRT-PCR to Determine Gene Expression in Plants (*Arabidopsis thaliana*)

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Introduction

In the antennae complex of plants, Photosystem II, which is surrounded by LHCB and ELIP complexes (Fig. 1), is where electrons are excited by the energy of light. When a plant is exposed to high light, the amount of LHCB complexes decreases while the amount of ELIP complexes increases. Both of these chloroplast proteins are coded by nuclear DNA; however, when the chloroplast is bleached, the production of LHCB and ELIP decreases, raising the question of what signal from the damaged chloroplast reaches the nucleus to cease the transcription of these proteins. In the production of chlorophyll in the plant *Arabidopsis thaliana* (Fig. 2), several steps can be inhibited by proteins such as GUN-5. Using a gun-5 mutant, the effects of chloroplast production on the expression of ELIP are being investigated to determine potential intermediates that could serve as chloroplast signals to the nucleus (Fig. 3).

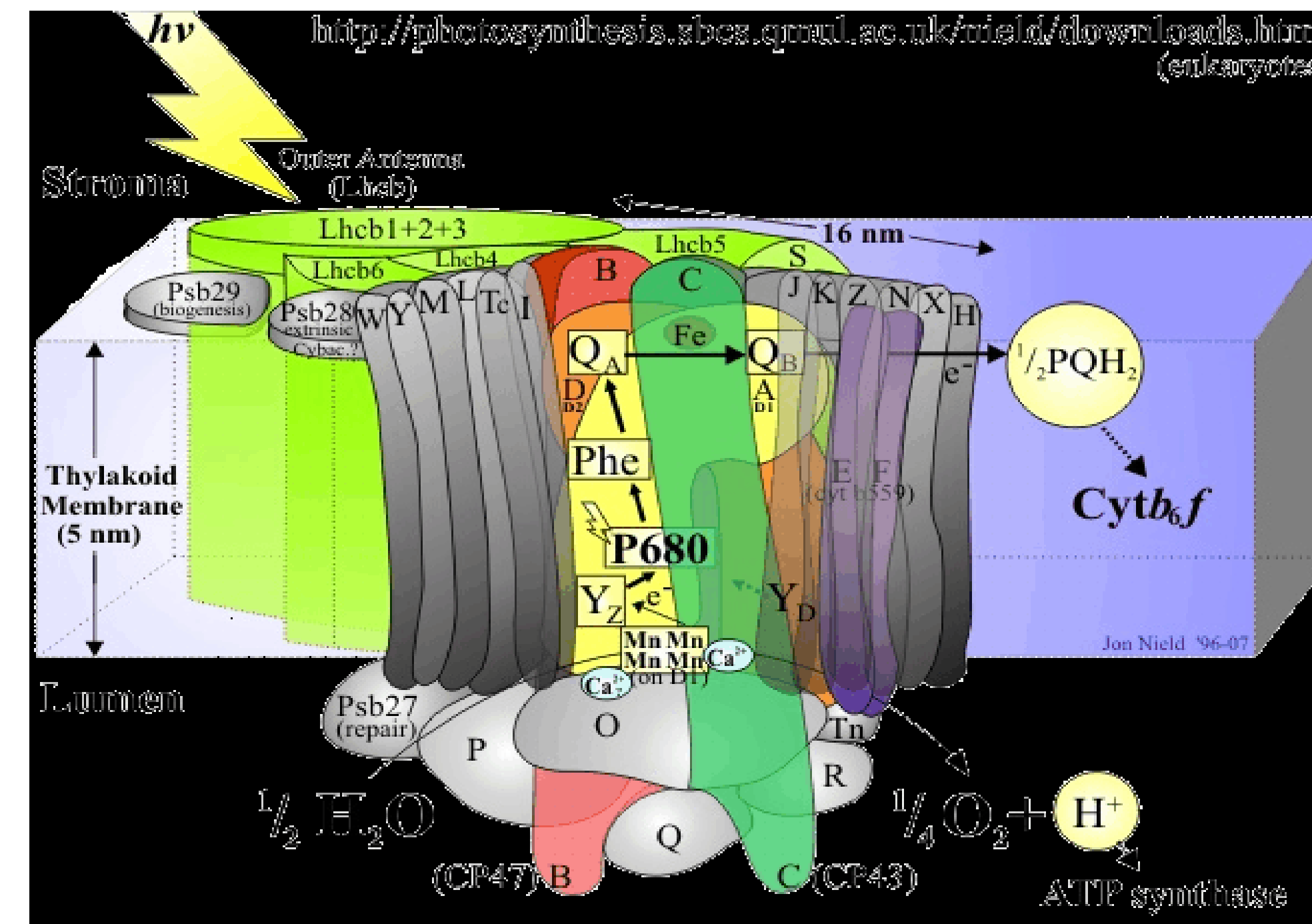


Figure 1: Photosystem II

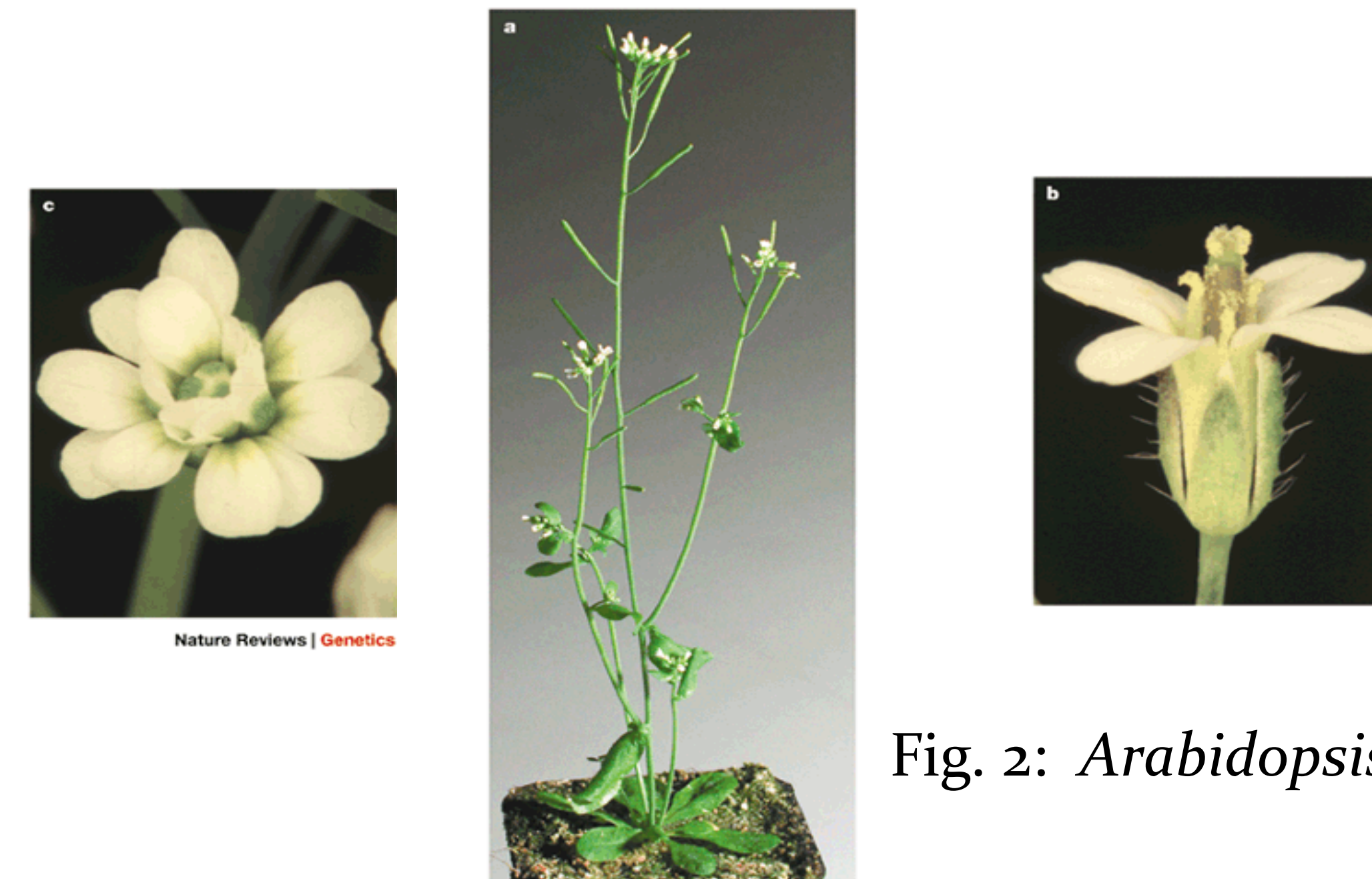


Fig. 2: *Arabidopsis thaliana*

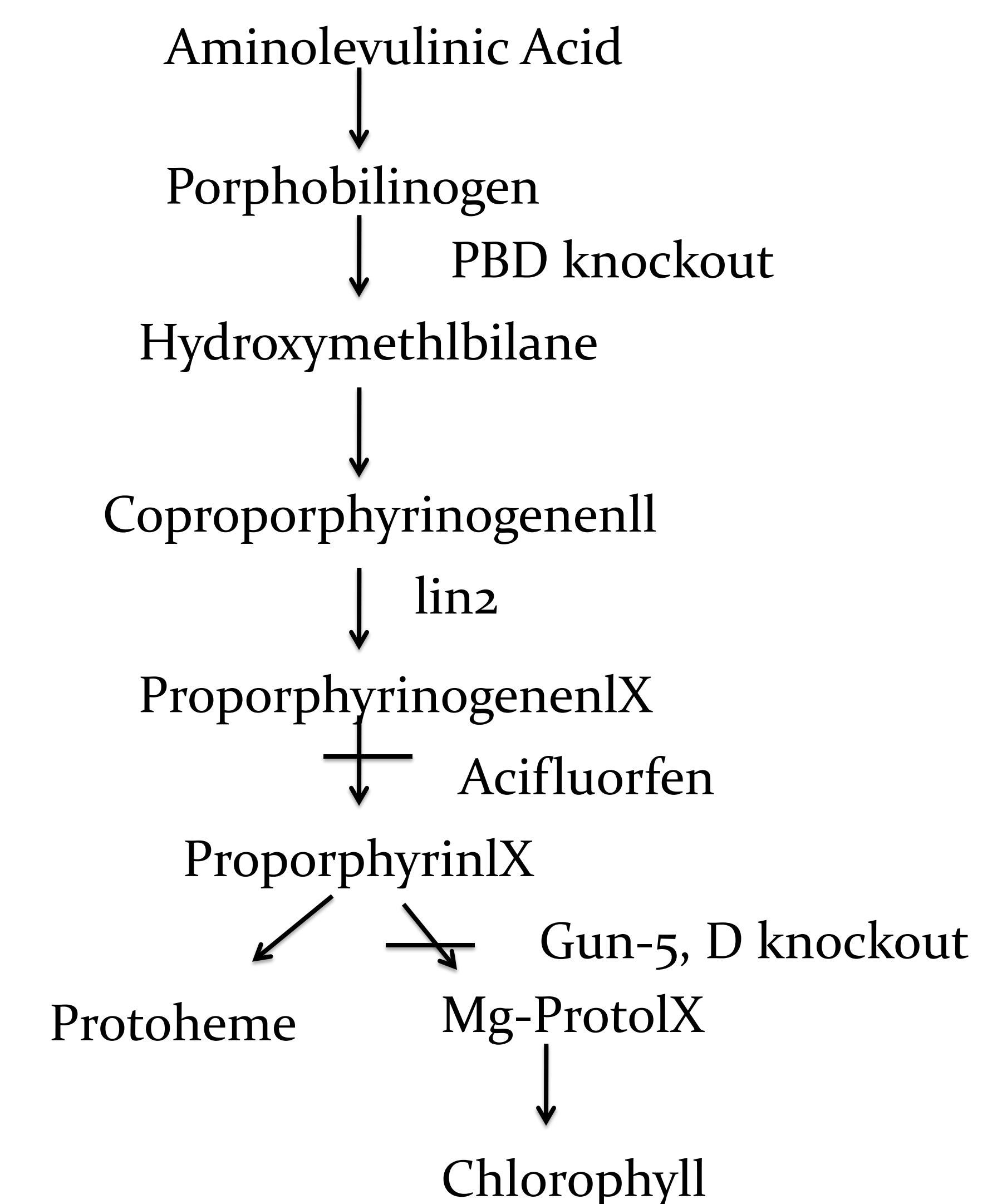


Figure 3: Synthesis of Chlorophyll

Objectives & Procedures

- Establish a set of standards for ELIP expression (Fig. 4)
 - Run gel electrophoresis of cDNA (Fig.
 - Sequence cDNA clones
 - Test primers for Real Time PCR of cDNA clones
 - Make dilution sequence for standards (Fig. 6)
- Measure *ELIP 1* and *2* mRNA expression in plant tissue
 - Treatment of plants
 - Isolation of total RNA
 - Reverse Transcription of mRNA to make cDNA
 - Real Time PCR of cDNA

Citations:

Figure 1: Nield, John. Mechanical and Structural Biology, 2009.

<<http://www.queenmaryphotosynthesis.org/nield/psIIimages/oxygenicphotosynthmodel.html>>.

Figure 2: Page, Damian & Ueli Grossniklaus. The Art and Design of Genetic Screens: Arabidopsis thaliana. Nature Review s Genetics, 2002.

<http://www.nature.com/nrg/journal/v3/n2/fig_tab/nrg730_F1.html>.

Figure 5: East Tennessee State University. Dept. Molecular Biology. 2009. <<http://www.etsu.edu/com/mbcf/Services/RealTimePCR.aspx>>.

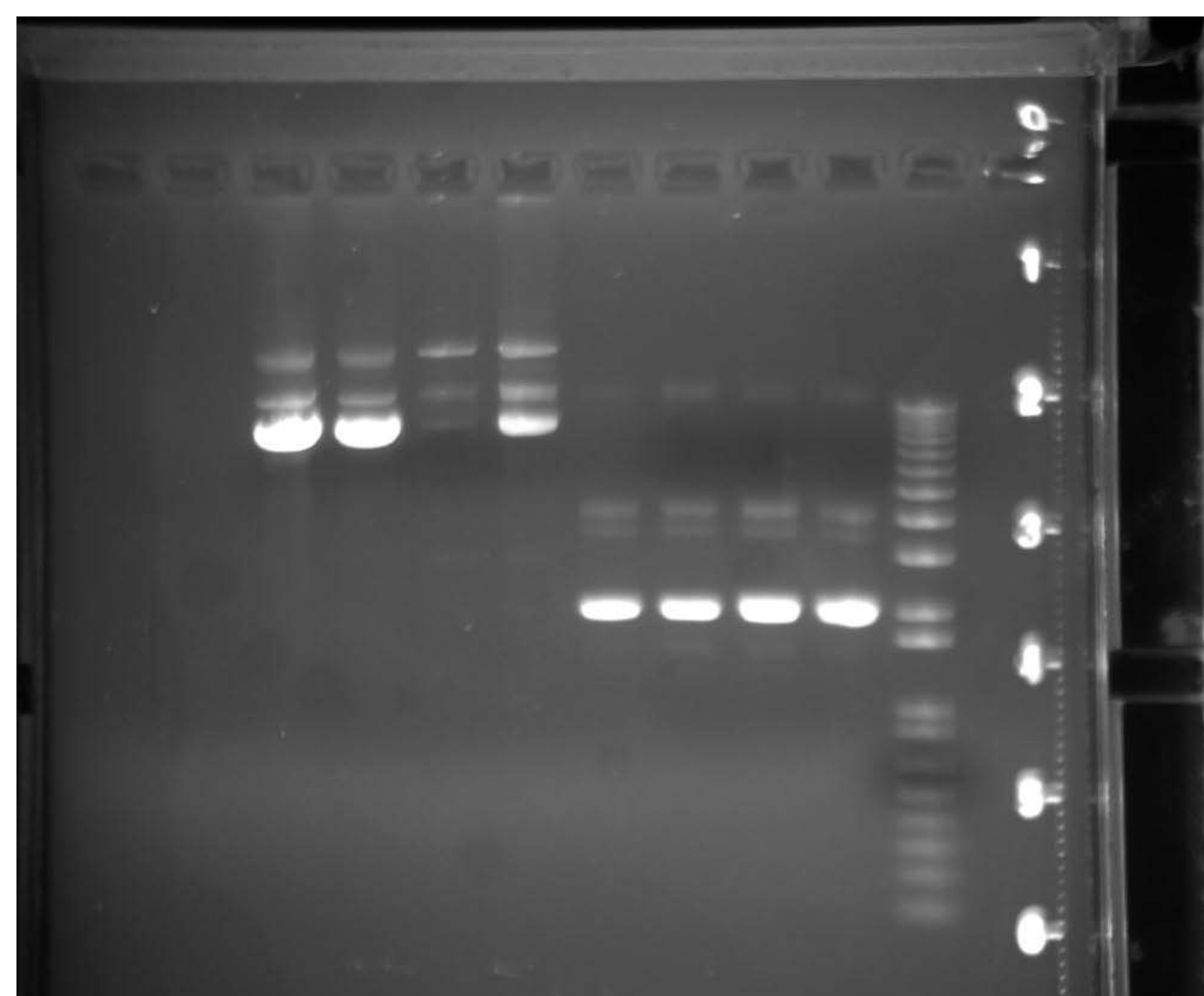
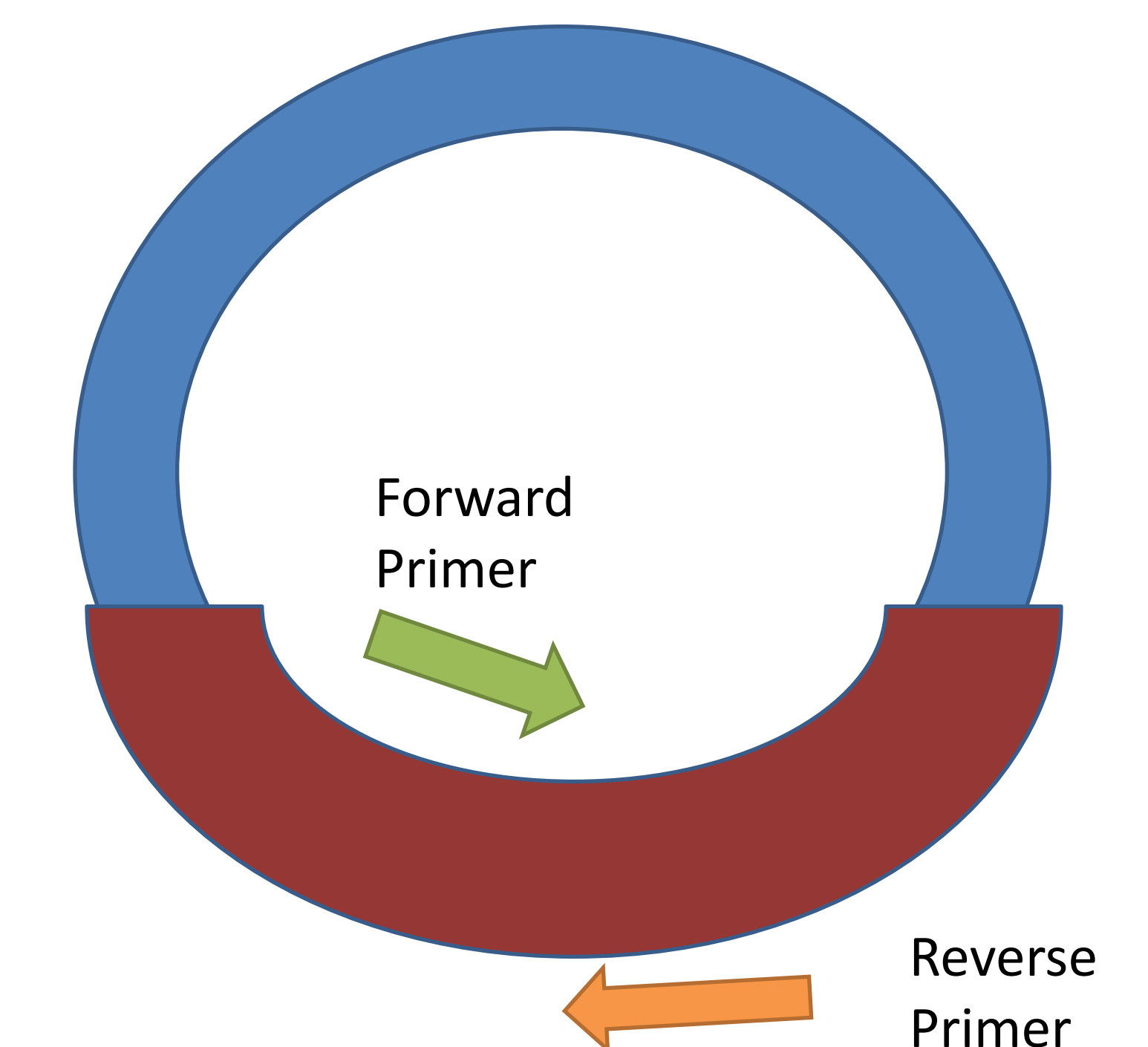


Figure 5: Gel Electrophoresis of cDNA Clones

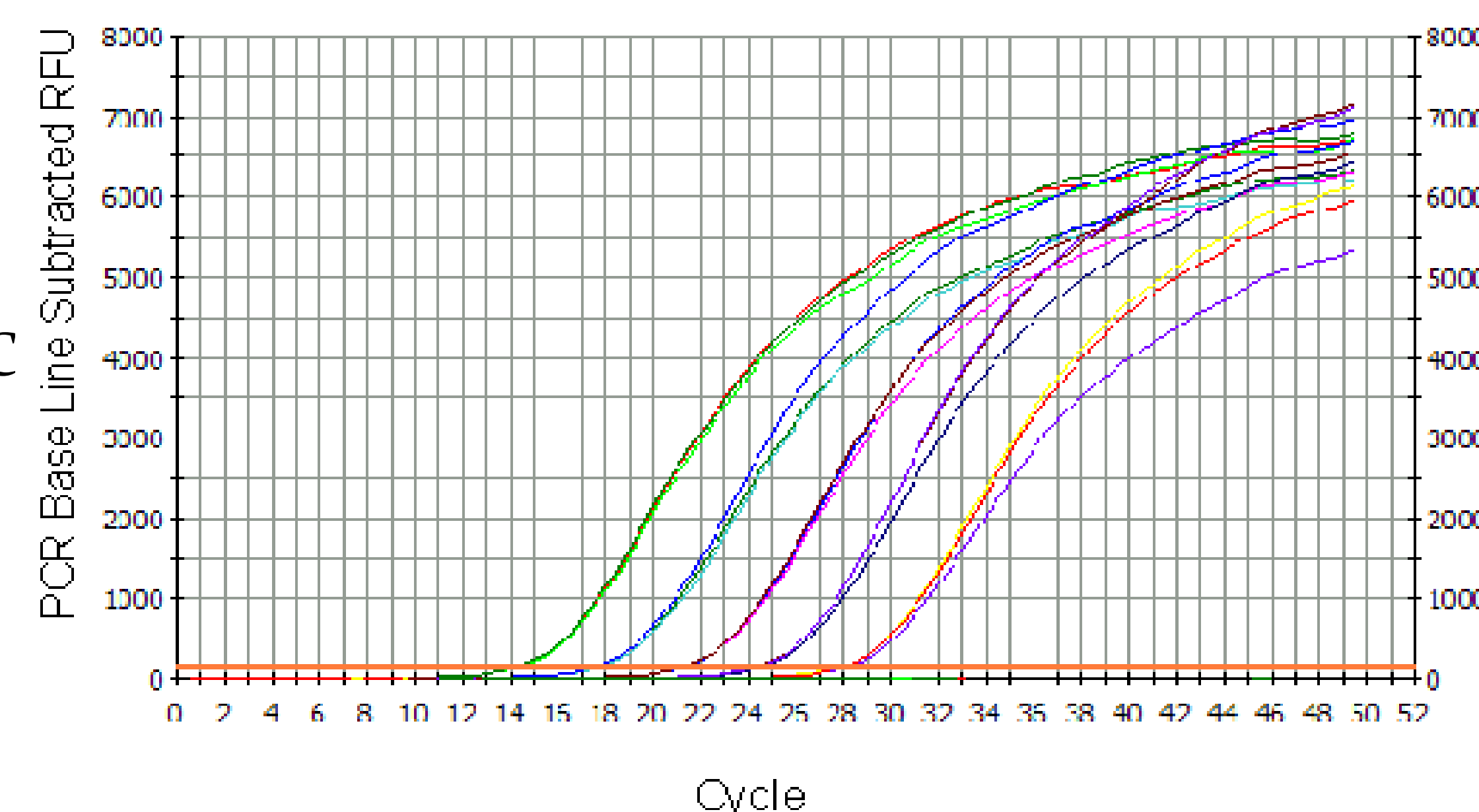


Figure 6: RT-PRC Results

Figure 4: Standards of ELIP 1 and 2