Compare the Sensitivity of SYBR Gold Stained Agarose Gel Excited by Epi-blue and 302 UV Transillumination Source

INTRODUCTION

As targeting on epi-blue light excitation stains, there have two systems completed with epi-blue LED among Wealtec molecular imaging systems, including KETA M series and Dolphin View II. Base on the different demands, Dolphin View II was made up as the most convenient stand-alone imaging system where as KETA M series was designed as high end excellent multipurpose system. Also according to the differences on design principle, two systems equipped with two different level cameras, 8 bit 1/2" room temperature CCD for Dolphin View II and 12 bit 2/3" cooled CCD for KETA M series. Both of them have its own advantages and suitable for different customers, respectively. Here are some illustrations to show the differences in both systems and set a guideline for customer to evaluate.

MATERIALS

- KETA ML imaging system (Wealtec)
- Dolphin View II with epi-white/blue/green LED lights (Wealtec)
- SYBR Gold (Invitrogen)
- λDNA/*Hind* III DNA marker(Wealtec)

PROCEDURES

- Prepare the serial diluted Lambda DNA/Hind*III* markers with the following amount: Lane 1 to 10: 1.6, 1.0, 0.8, 0.5, 0.1, 0.08, 0.05, 0.01, 0.008, 0.005 μg total DNA.
- Prepare the 1.2% TAE agarose gel with pH = 8.0.
- Run the DNA samples with 100 V in agarose gel for 1 hour.
- Stain the agarose gel with staining buffer, 10 μL stock (1:10000) in 100 mL TAE buffer (pH 8.0), for 1 hour.
- Observe the result with KETA ML and Dolphin View II imaging system through the same cut off range filter, WK101 filter and high transparent amber filter (550~675 nm).

RESULT

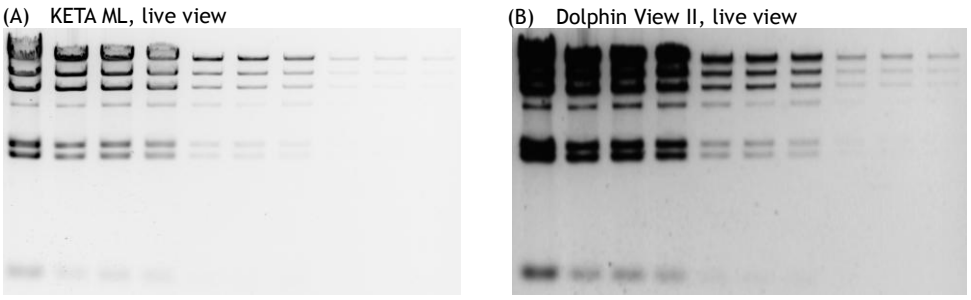

Figure 1. *Trans UV excited SYBR Gold stained agarose gel.*
Observed in (A) KETA ML and (B) Dolphin View II imaging systems with iris fully opened.

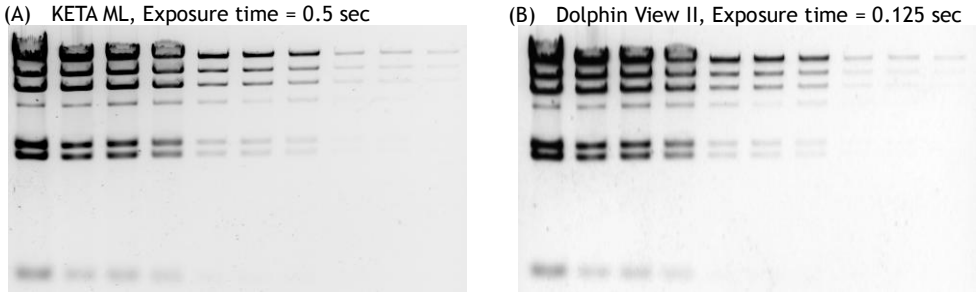

Figure 2. *Blue LED excitated SYBR Gold stained agarose gel.*
Observed in (A) KETA ML and (B) Dolphin View II imaging systems with iris fully opened.

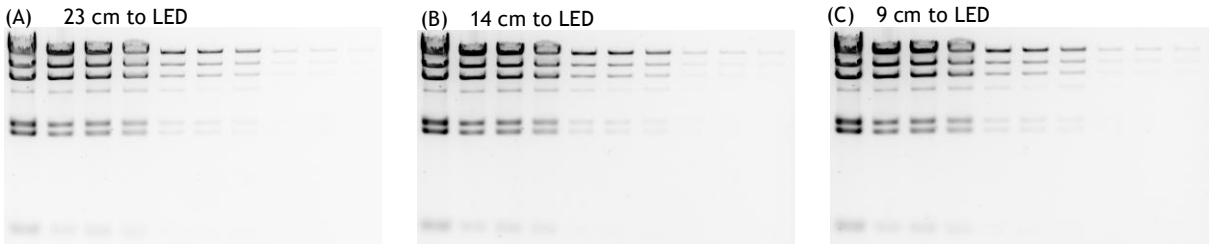

Figure 3. *Epi-Blue LED excitated SYBR Gold stained agarose gel in KETA ML with 0.2 seconds exposure time.*
Place the gel with (A) 23 cm, (B) 14 cm, and (C) 9 cm in distance to LED lights.

DISCUSSION

Despites the differences on the specification, both KETA ML and Dolphin View II shared the same detection limit with 0.676 ng for the SYBR Gold stained DNA agarose gel under the 302 nm UV transilluminator excitation (*fig*. 1). While using epi-blue LED as excitation light source, Dolphin View II can get about 6 times higher signal detection than KETA ML (fig. 2). However, extending the exposure time allows KETA ML to get better signal capture and detection limit without pixel saturations. As shortening the distance from MC 100 LED light to the gel in KETA ML as in *fig*. 3, SYBR Gold signal can be intensified and makes the observation much clearly. Using of WK102 filter in standard package KETA M series systems will be another option to enhance the signal capturing.

KETA M series and Dolphin View II both equipped with MC 100 LED epi-light sources that specifically designed for fluorescence sample observation. When customers are willing to buy the fluorescent imaging system, Dolphin View II will be a cheapest and satisfied option for preliminary fluorescence gel observation. What if users ask for better image quality, detection range, and signal to noise ratio, fully robotic controlled KETA M series imaging system will be the best option in fluorescence detection, especially when placing the sample with correct distance to the epi-LED light source. Additionally, KETA M series can perform very well on chemiluminescent detection which is impossible to do in Dolphin View II system.

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