Superior Performance Gel Documentation and Fluorescence Detection by Multi-purpose KETA ML system

INTRODUCTION

KETA multi-purpose imaging system (ML) is designed for most imaging experiments like gel documentation, fluorescence and chemiluminescence detection. Due to KETA ML equipped with 2/3" high quality CCD and coupled with 2 stage cooling device, it has super high sensitivity on chemiluminescence signal detection, which had been proven in Technical Bulletin #28. In this article, the performances on both gel documentation and fluorescence detection in KETA ML were examined.

MATERIALS

- KETA ML Imaging System (Wealtec)
- V-GES and Mini-GES for electrophoresis (Wealtec)
- Coomassie Brilliant Blue R-250 (Sigma)
- SYPRO Red, SYPRO Orange and SYPRO Ruby for protein staining (Invitrogen)
- SYBR Green I and SYBR Safe for DNA staining (Invitrogen)
- Ethidium Bromide (Sigma)

PROCEDURES

1. DNA and protein samples were diluted into proper concentration.
2. After loading, DNA and protein samples were separated by using 1% agarose and 12 % SDS-PAGE gels in mini-GES and V-GES, respectively.
3. After electrophoresis, gels were stained with Coomassie Brilliant Blue, EtBr, SYBR Green I, SYBR Safe, SYPRO Red, SYPRO Orange, SYPRO Ruby dye according to the instructions.
4. Gels were observed with KETA ML imaging system by choosing proper excitation light source and filter.

RESULT

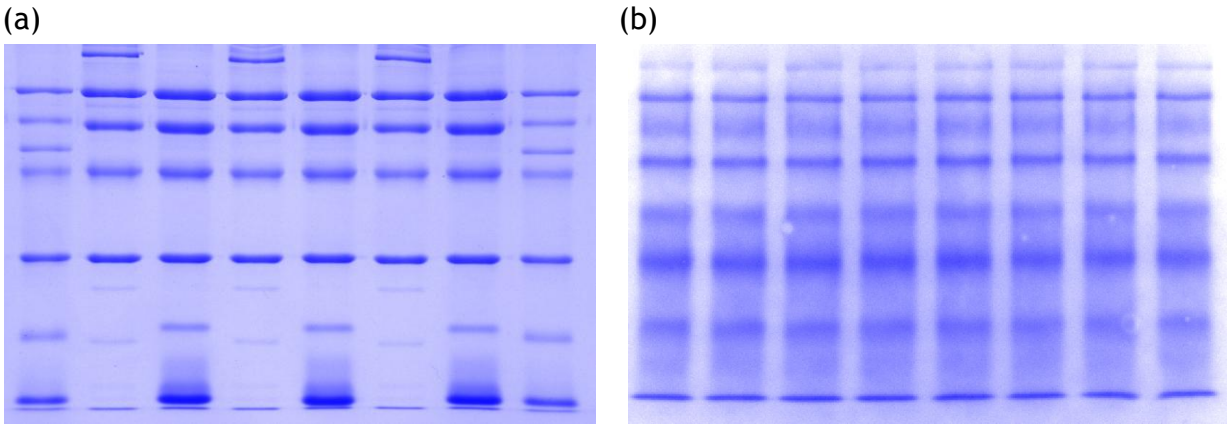

Figure 1. *General gel documentation. Protein markers on (a) SDS-PAGE and (b) PVDF membrane and stained by Coomassie Brilliant Blue.*

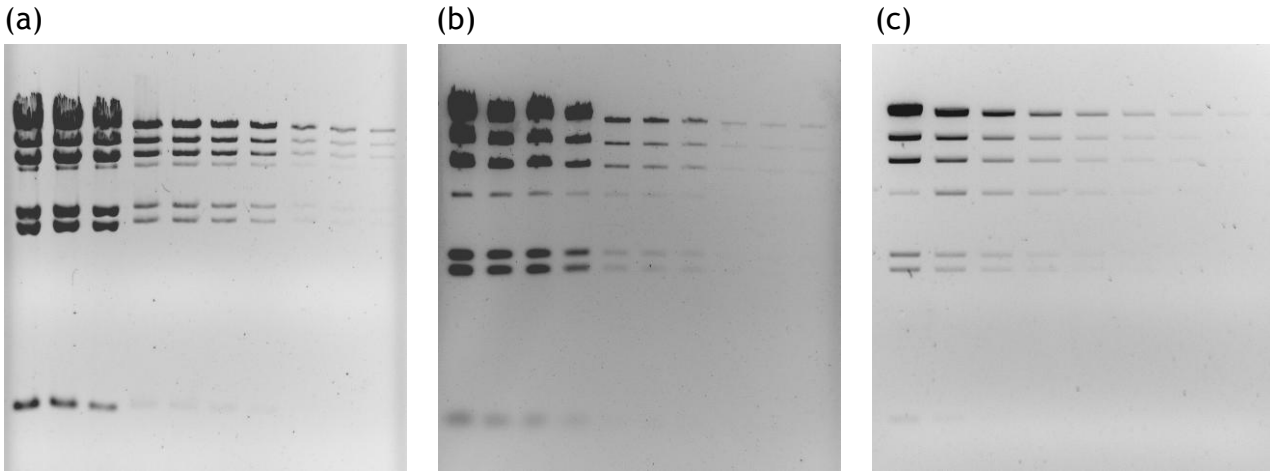

Figure 2*(A)***.** *Fluorescent signals detection of KETA ML.*

λDNA/HindIII ladders were stained with different dyes, and excited by trans-UV (312 nm) and observed through different filters.

(a) Ethidium bromide with WK101, exposed for 0.5 seconds.
(DNA amount: 1666.7, 1100, 550, 166.7, 125, 83.3, 41.7, 16.7, 11 and 5.5 ng/lane),

(b) SYBR Green I with WK102, exposed for 1.5 seconds.
(DNA amount: 1666.7, 1000, 800, 500, 100, 80, 50, 10, 8 and 5 ng/lane)

(c) SYBR Safe dye with WK102, exposed for 2 seconds.
(DNA amount: 200, 100, 50, 25, 12.5, 6.25, 3.125 and 1.5625 ng/lane).

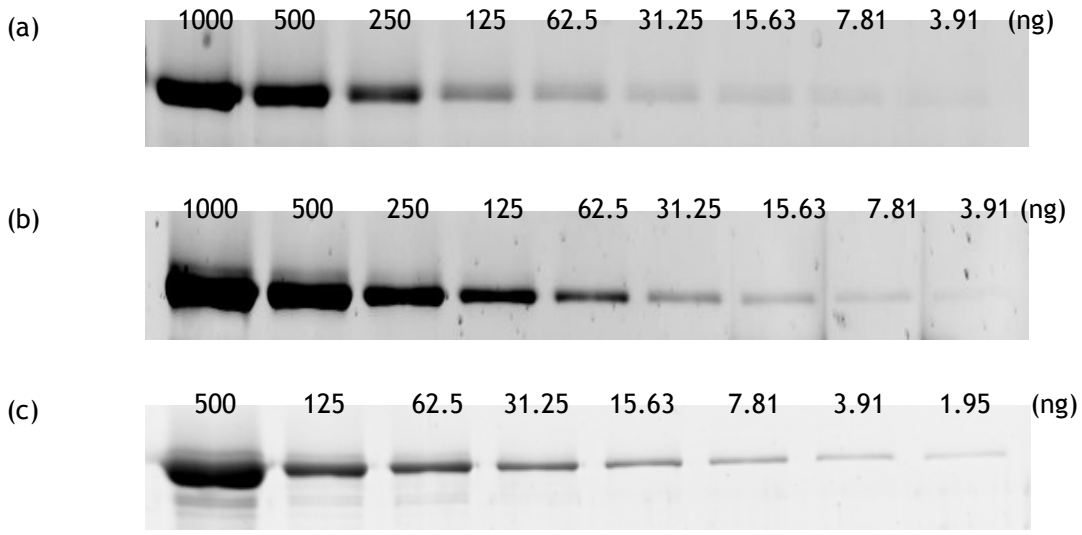

Figure 2*(B)*. *Fluorescent signals detection of KETA ML.*

Purified transferrin protein samples were stained with (a) SYPRO Red, (b) SYPRO Orange and (c) SYPRO Ruby dye, excited by Trans-UV (312 nm) UV transilluminator and observed through WK101 filter with 8, 2 and 0.2 seconds exposure time, respectively.

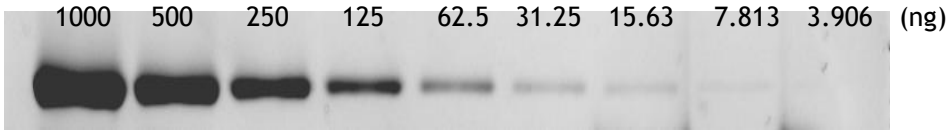

Figure 3. *SYPRO Orange signal excitation through MC100 Epi-Blue LED light source.*

Image was taken through WK101 filter with 2 seconds exposure time.

DISCUSSION

Designed specifically for multi-purpose experiments, KETA ML was not only performed well on chemiluminescence detection, this note also proved that it could be applied to the general gel documentation such as stained gel and PVDF membrane (*fig. 1*). For fluorescent signal excitation, provided with 312 nm UV transilluminator, KETA ML is sufficient for most commercial dyes. Also coupled with the multi-color Epi RGB LED lights, MC100, it provides more choices with longer wavelength for fluorescence excitation. For example, SYPRO Orange signal can be excited by a safer, non-UV Epi blue light source (*fig. 3*). Equipped with motorized filter wheel which have maximal five sets of emission filter, KETA ML can be applied to observe various dye staining, such as EtBr, SYBR Green I and

SYBR Safe for nucleic acid staining, and SYPRO Orange, SYPRO Red and SYPRO Ruby for protein staining (*fig. 2*).

Besides performed excellently on the chemiluminescence detections in the previous articles, KETA ML was proved to have outstanding detection on both gel documentation and florescent observation. If users are targeting among all kinds of illumination samples, KETA ML is exactly the right model for them to file their result. Even if users are trying to have *in vivo* images, capability to upgrade with XE300, KETA ML can be also a good option for them.

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