

Sensitivity of KETA ML Imaging System

INTRODUCTION

Newly launched Ketagalan series imaging systems (KETA G/M/C series) were equipped with variable cameras according to the using purpose of the system. In the M series, ML system has the most sensitive camera, the K16C II, which is designed especially for chemiluminescence detection. K16C II camera with 2/3" 2-stage Peltier cooled CCD element and 1.4 mega-pixels resolution provides the lowest cooling temperature (-25°C) of the CCD and the best detection limit in chemiluminescence detection. With the 2-stage Peltier cooled system, the signal to noise ratio will be significantly enhanced and have the best sensitivity.

EQUIPMENTS AND MATERIALS

- Mouse cell lysate obtain from Graduate Institute of Physiology in National Taiwan University College of Medicine.
- 1st antibody: B-Actin (Sigma), 2nd antibody: anti-mouse-IgG-HRP.
- ECL Enhanced Chemiluminescence reagent (Millipore)
- KETA ML imaging system (Wealtec)

PROCEDURES

1. Western blot experiment was performed by the laboratory in Graduate Institute of Physiology in National Taiwan University College of Medicine.
2. Serial dilutions of mouse cell lysate were separated with SDS-PAGE with the total protein amount of 1250, 625, 313, 156, 78, 36, 19, and 9 ng.
3. After electrophoresis, transfer the protein onto the PVDF membrane.
4. Hybridization with the 1st and then the 2nd antibody.
5. The result was presented with ECL Enhanced Chemiluminescence reagent.
6. Detect the result through KETA ML image system by using batch capture method.

RESULTS

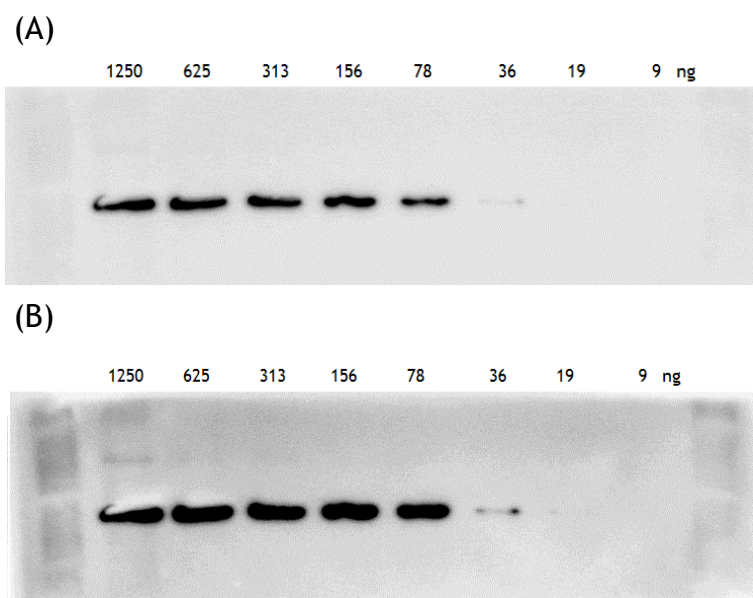


Figure 1. Detection of B-actin in series diluted mouse cell lysate via KETA ML image system. (a) With 10 seconds exposure. (b) With 40 seconds exposure.

DISCUSSION

As detecting ECL Enhanced chemiluminescence result with KETA ML image system, due to the high sensitivity of the 2-stage Peltier CCD camera, it takes only few seconds to capture the image. In the figure 1(A), when exposure over than 10 seconds, because of the high quantum efficiency of 16 bit K16C II camera, the detection of high amount total protein samples will get saturated easily. Users should lower down the amount of total protein samples or replace the camera which with higher pixel depth. About the detection limit, sample with 36 ng total protein can be observed after 10 seconds exposure. After prolonged the exposure time to 40 seconds, even for the 19 ng sample could be detected as in figure 1(B). No binning function was activated in this experiment.