Supplementary Figure 2. Adaptation of the protease imaging assay to a fluorescent plate reader format.

HT-1080 fibrosarcoma cells were seeded in a 96-well microtiter plate and treated with 90 nM (5 μg/ml) LF/β-Lac and 26 nM (2 μg/ml) PrAg-L1 (a-c) or PrAg (d-f) in the presence of increasing concentrations of the metalloprotease inhibitors BB-94 (a and d), BB-2516 (b and e), and TIMP-2 (c and f). After addition of 1.5 μM CCF2/AM, the fluorescence emission was recorded with a plate reader using 405 nm excitation and 460 nm emission filters. The data are expressed as mean ± standard error of the mean of triplicate determinations. The specific imaging of metalloproteinase inhibition is demonstrated by the dose-dependent reduction in 460 nM light emission of PrAg-L1, but not in PrAg-treated cells.