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SA-B-Gal Activity

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We use this protocol and it's working

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Abstract

To measure cellular senescence



Senescence associated SA- β -Galactosidase Activity

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1.1 The cells were washed twice with ice cold PBS and then incubated with 60 μ L of cell lysis buffer provided with the cellular senescence assay kit (Fluorometric format) (Cell Biolabs, USA) for 10 minutes at 4 °C.



1.2 The whole cell lysates were centrifuged 15 minutes at 14000 g at 4 °C. Total protein concentrations were determined by Qubit Protein Assay Kit (ThermoFisher, [Q33211](#)) using the Qubit 4 Fluorometer (Thermo Fisher Scientific, Singapore, shipped from Germany) according to the manufacturer's instructions.



1.3 Following protein quantification, the assay buffer (Cell Biolabs, Inc., CBA-231, San Diego, USA) was added 1:1 to the cell lysates and incubated for 3h in the dark at 37 °C.



1.4 The reaction was stopped by adding 120 μ L of the Stop solution (provided with the cellular senescence assay kit) per 30 μ L of the reaction mixture.

1.5 Fluorescence was measured using a plate reader at 360 nm (Excitation) / 465 nm (Emission). SA-b-Gal activity was normalized to total protein concentrations.

