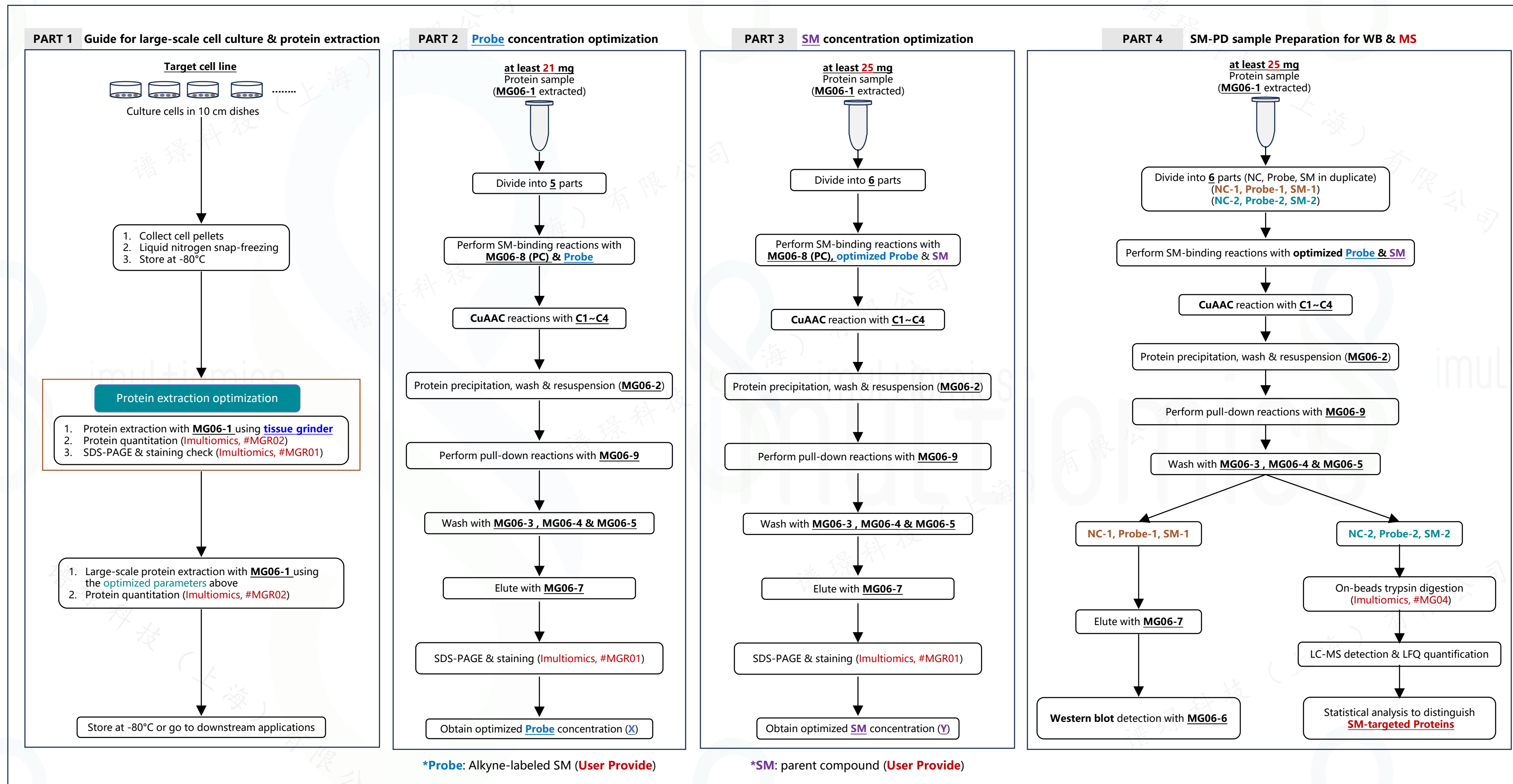


小分子-靶点蛋白富集及质谱筛选 (LFQ)

SM-PD Enrichment Kit (**Alkyne** tag, MS grade)

Catalog Number: MG06

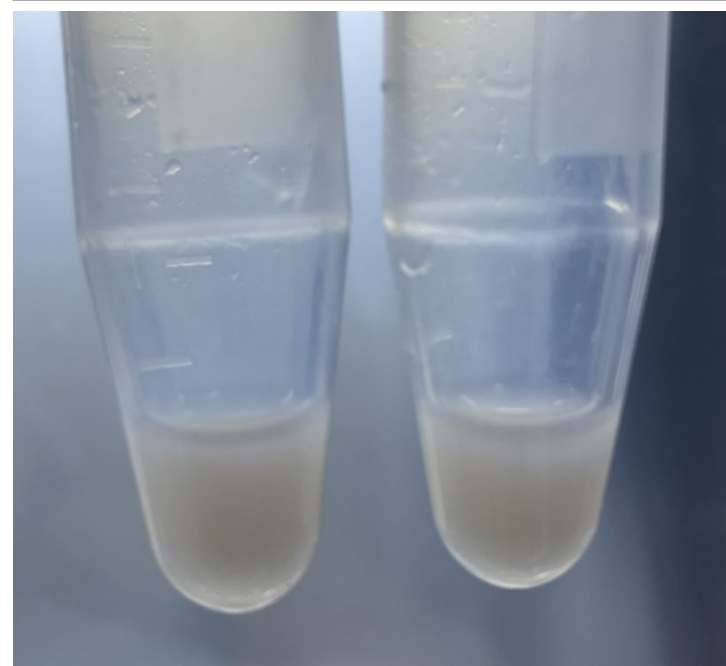
Workflow of SM-PD Enrichment Kit (Alkyne tag, MS grade) Catalog Number: #MG06



A. Protein extraction & SDS-PAGE check

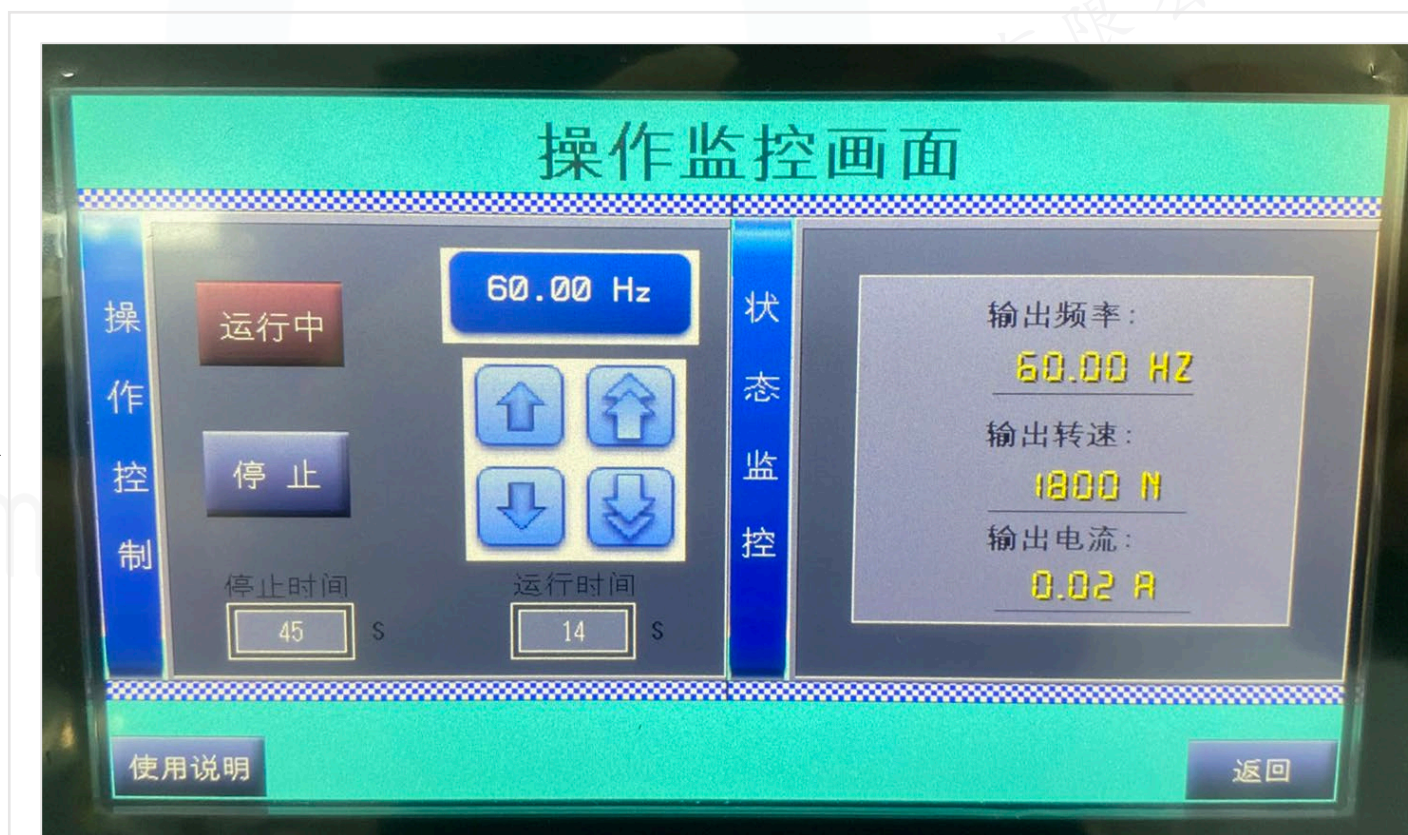
Cell pellets

HEK-293T



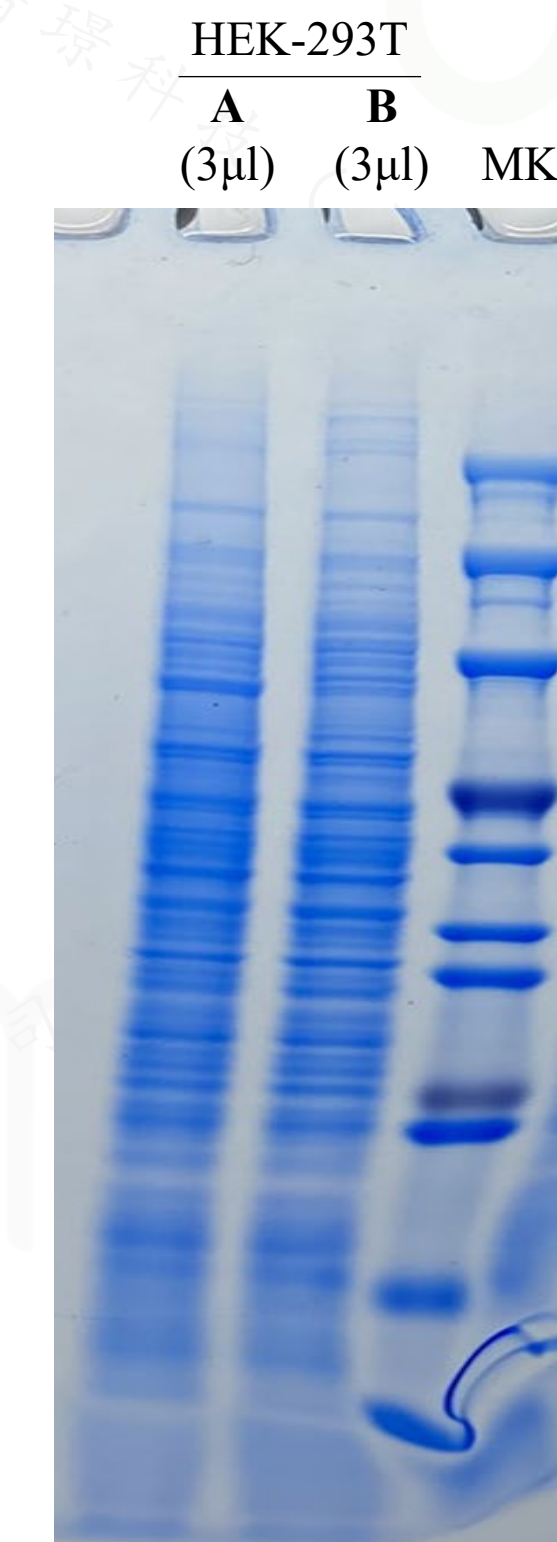
1. Cell pellet: **3×10cm** dishes/tube.
2. Total volume: **~400μl/tube**

Tissue grinder



(SCIENTZ-48, Ningbo Scientz Biotechnology Co., Ltd)

SDS-PAGE gel staining

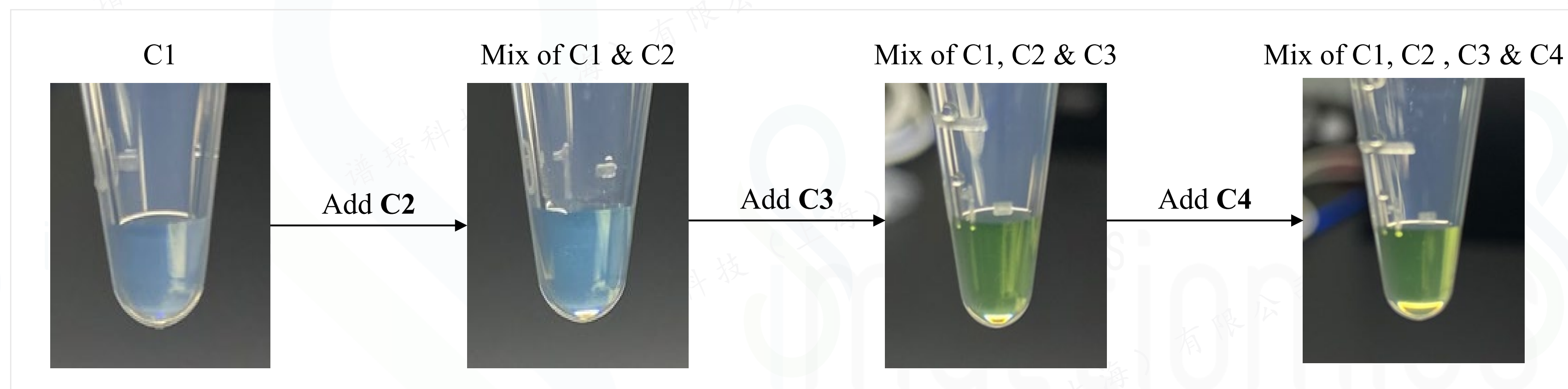


- A:**
1. Cell pellet: **6×10cm** dishes.
 2. Protein concentration: **6.9 mg/ml**.
 3. Total volume: **~2.4ml**

- B:**
1. Cell pellet: **4×10cm** dishes.
 2. Protein concentration: **6.0 mg/ml**.
 3. Total volume: **~1.6ml**

1. Cell pellet was resuspended into **MG06-1** (cell lysis buffer) in #MG06 Kit, and protein was extracted using **tissue grinder** (SCIENTZ-48, Ningbo Scientz Biotechnology Co., Ltd).
2. Protein concentration was measured with Bradford Assay Kit (**Imultiomics**, #MGR02).
3. Protein samples were treated with **MG06-7** (5 × sample loading buffer)) in #MG06 Kit, separated by SDS-PAGE, and stained with Coomassie blue staining reagent (**Imultiomics**, #MGR01).

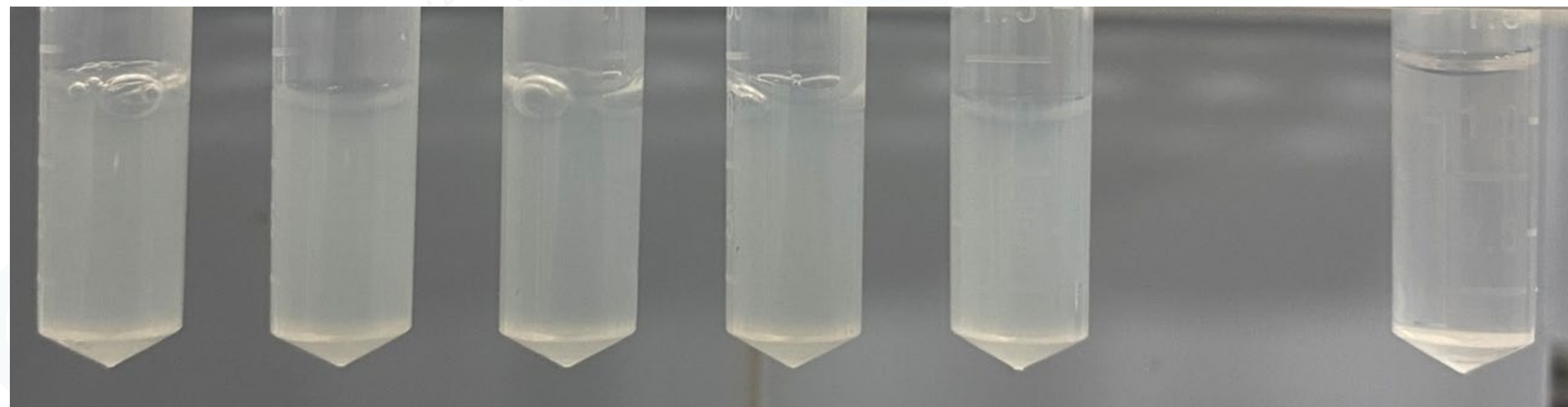
B. Solution color changes during CuAAC mix preparation



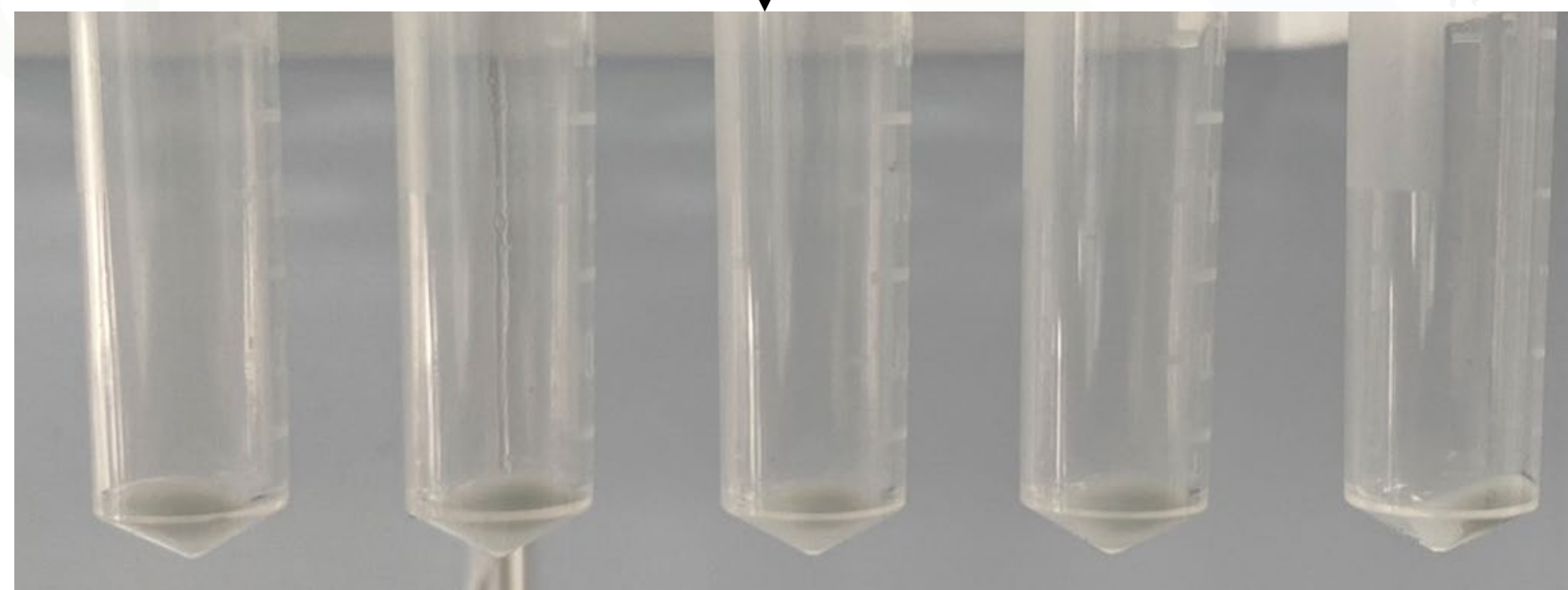
C. CuAAC reaction in Protein samples

Addition of CuAAC mix

No addition of CuAAC mix



Centrifuge (4°C, 7000g)

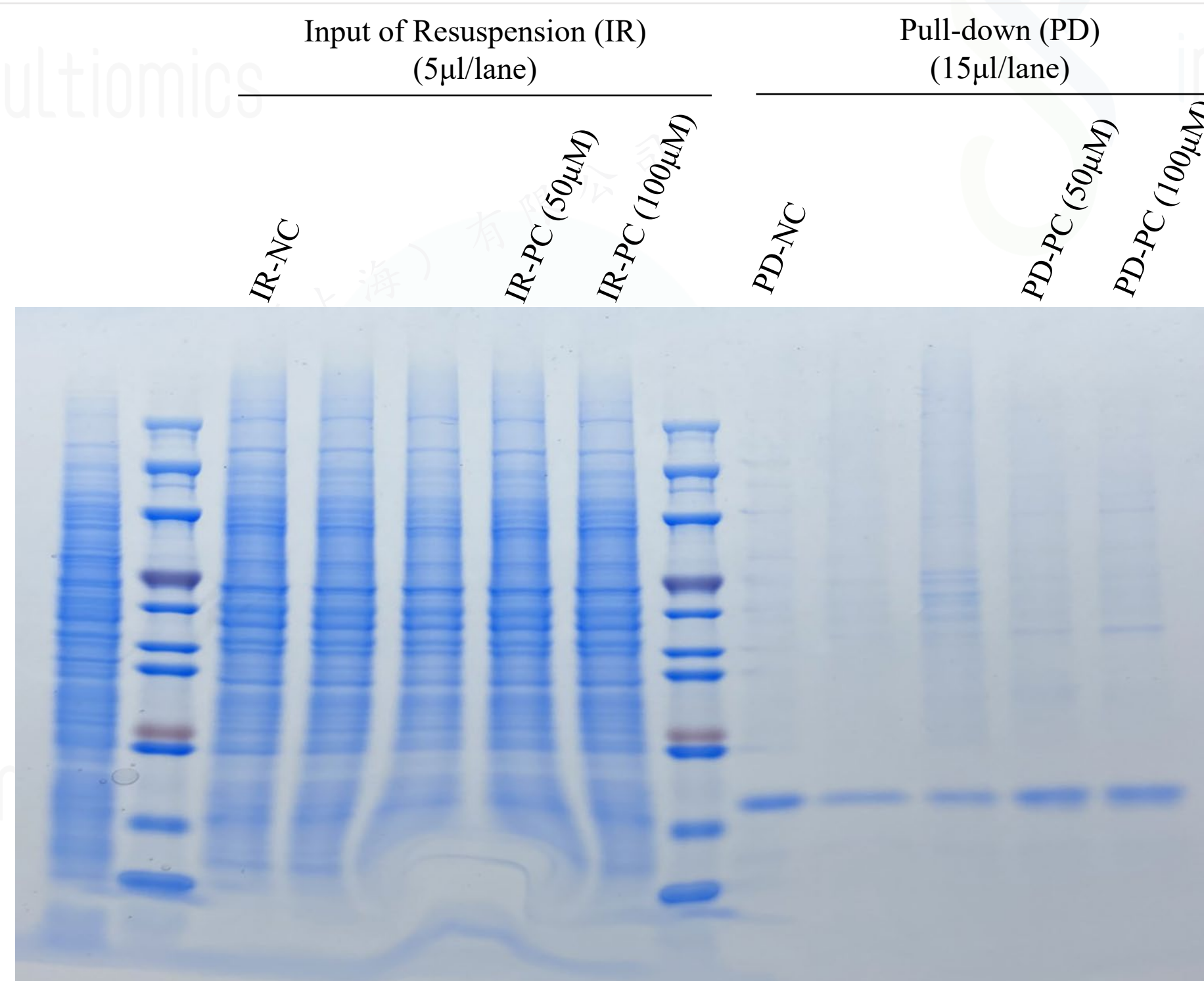


Pellet

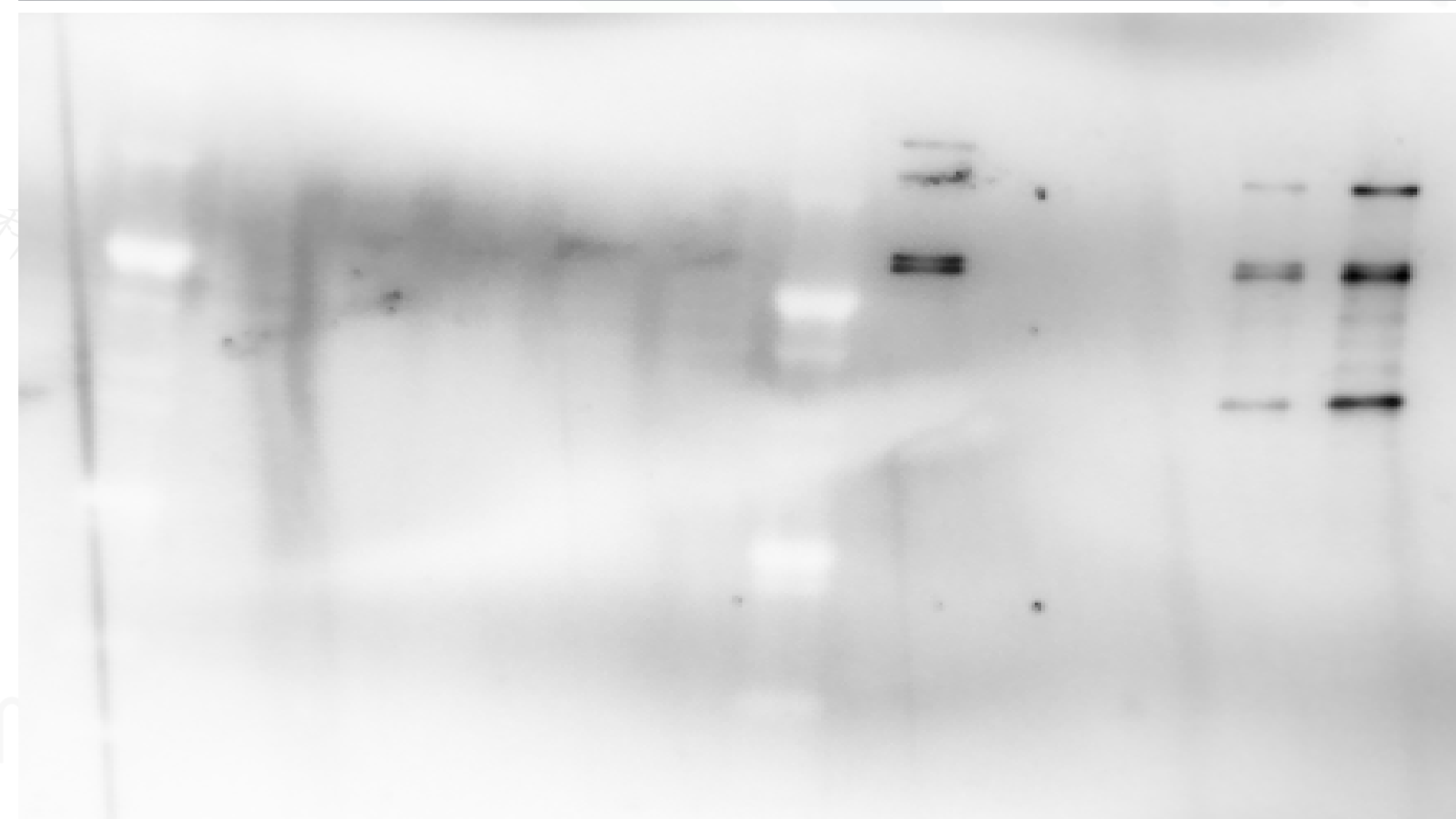
1. Protein samples turned cloudy after incubation with CuAAC mix for 1.5 hours at RT.
2. Pellets were collected after centrifuge for downstream pull-down procedure.

D. Detection of Pulled-down samples by Gel staining & western blot

Comassie Blue Staining
(Imultiomics, #MGR01)



Western blot (Streptavidin-HRP)
(MG06-6 in MG06 Kit)



1. **MG06-8** (Positive Control, PC) in **#MG06 Kit**, the alkyne-labeled SM.
2. The pulled-down samples of **MG06-8** (50, 100μM) using **# MG06 Kit** were detected by gel staining and western blot.
3. Gel staining (up) showed the enriched protein bands compared to NC (negative control) sample.
4. western blot (down) data was consistent with that of gel staining.