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小分子-靶点蛋白富集及质谱筛选 (LFQ)

SM-PD Enrichment Kit (Azide tag, MS grade)

Catalog Number: MG07

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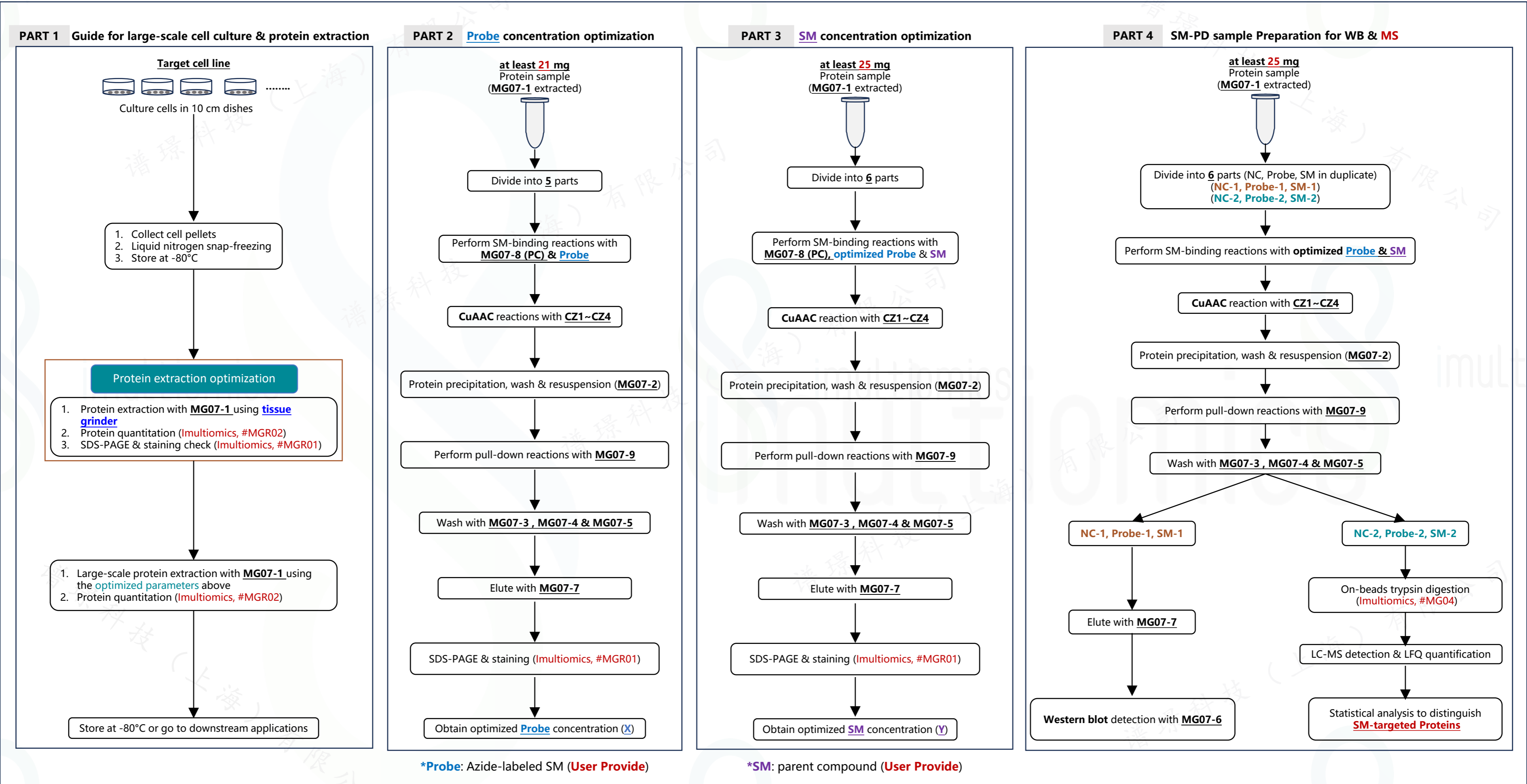
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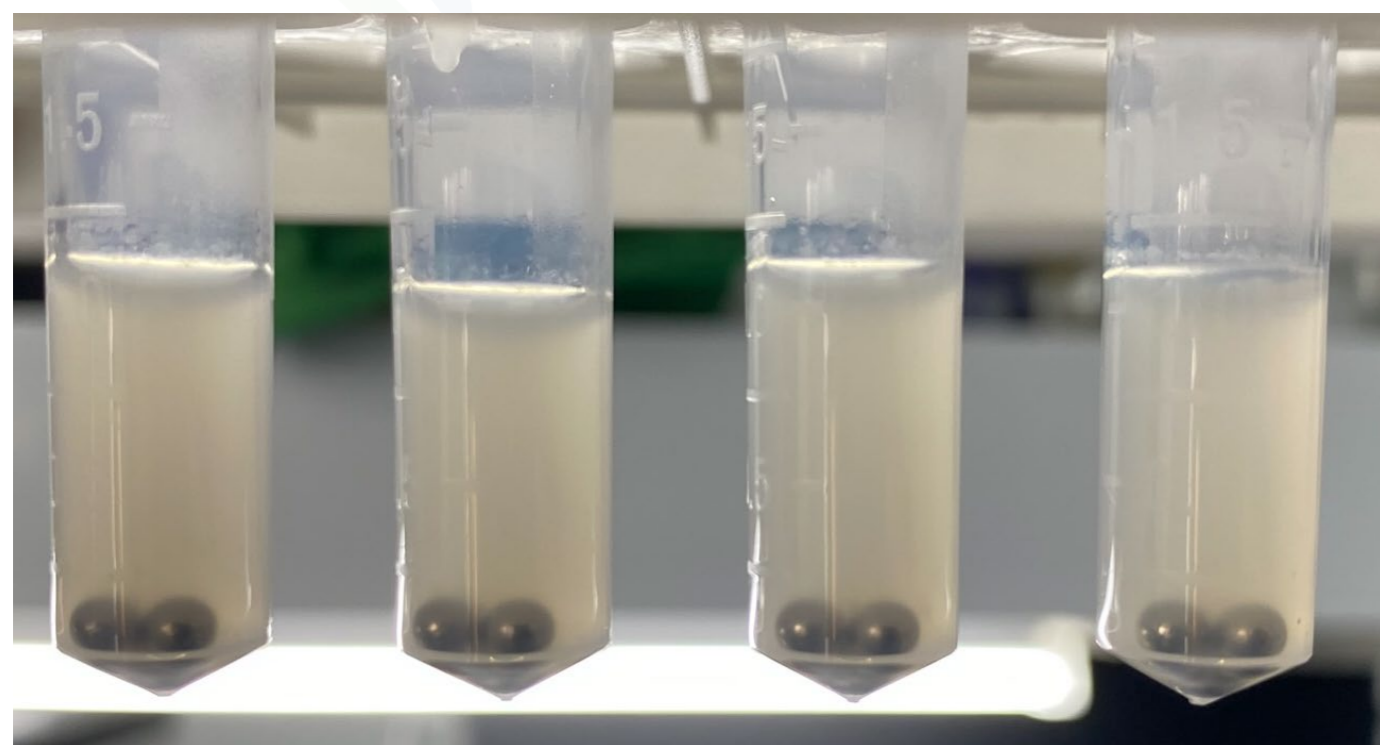
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Workflow of SM-PD Enrichment Kit (Azide tag, MS grade) Catalog Number: #MG07



A. Protein extraction & quantitation



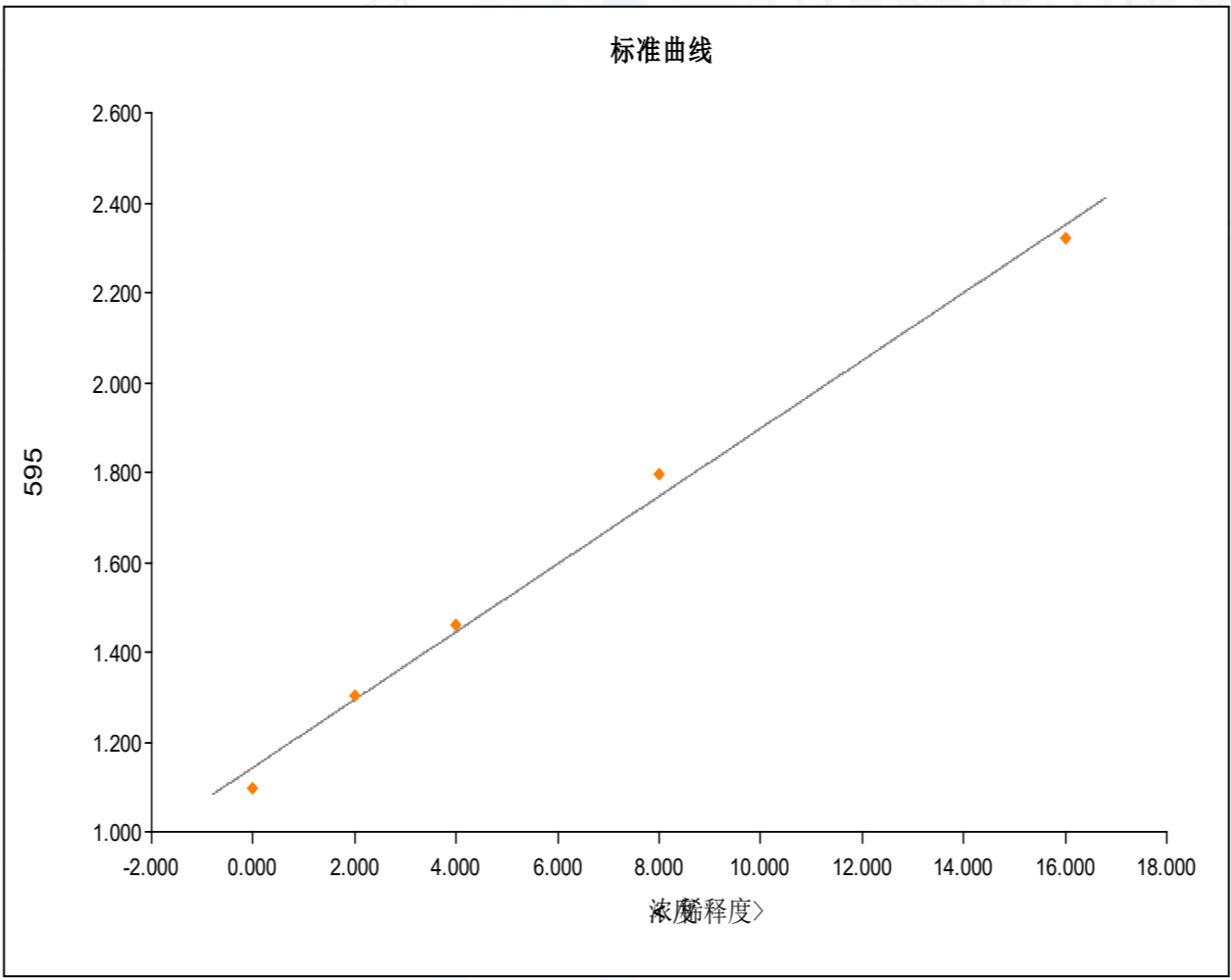
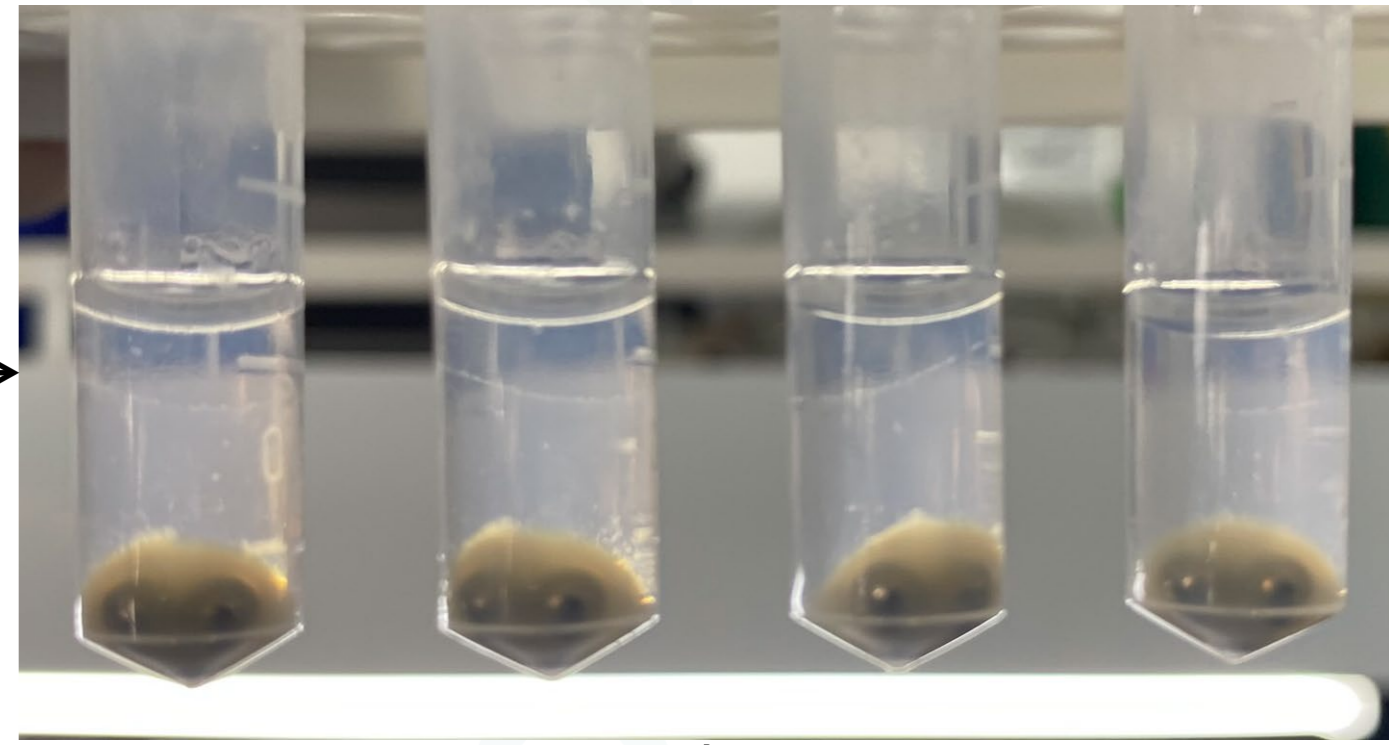
Cell pellet of 9 × 10 cm dishes was resuspended into **3.6mL** of MG07-1 (Cell lysis buffer) in #MG07 Kit, allocated to 2ml tubes, and **3×3mm (diameter)** of zirconia balls were added into each tubes.

60 Hz, 45s/cycle, 6 cycles



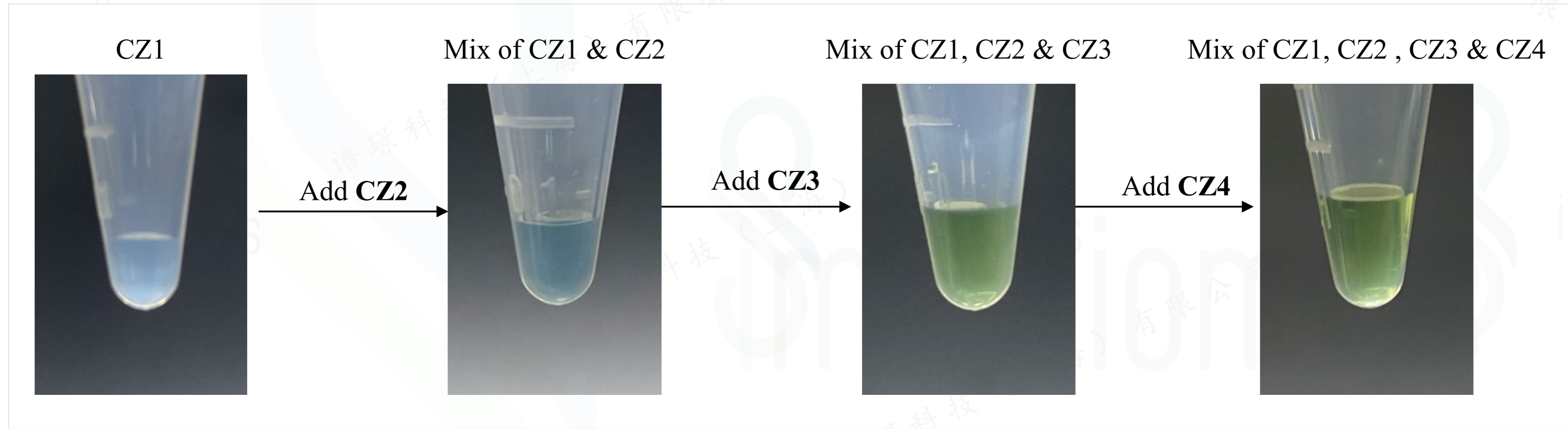
(SCIENTZ-48, Ningbo Scientz Biotechnology Co., Ltd)

4°C, 20000g, 15min

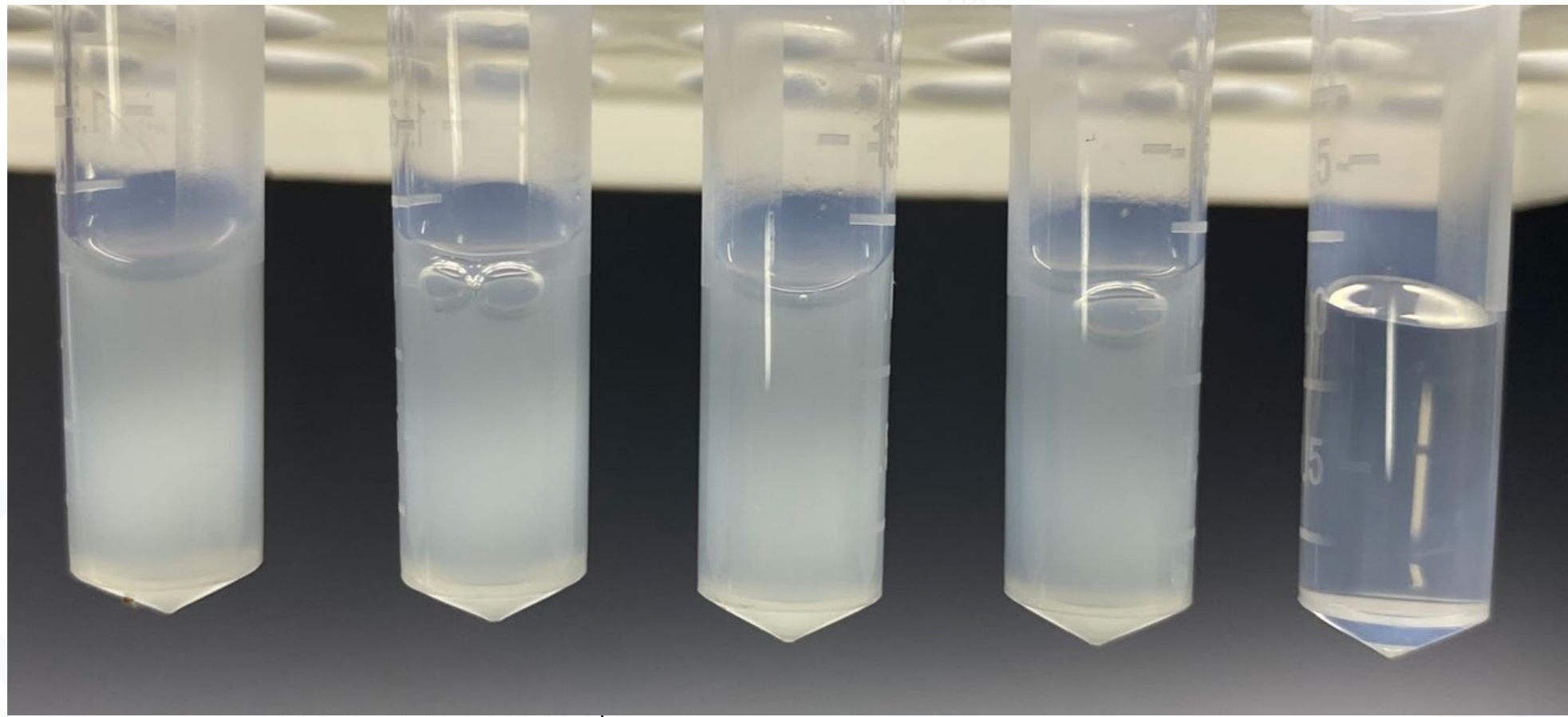
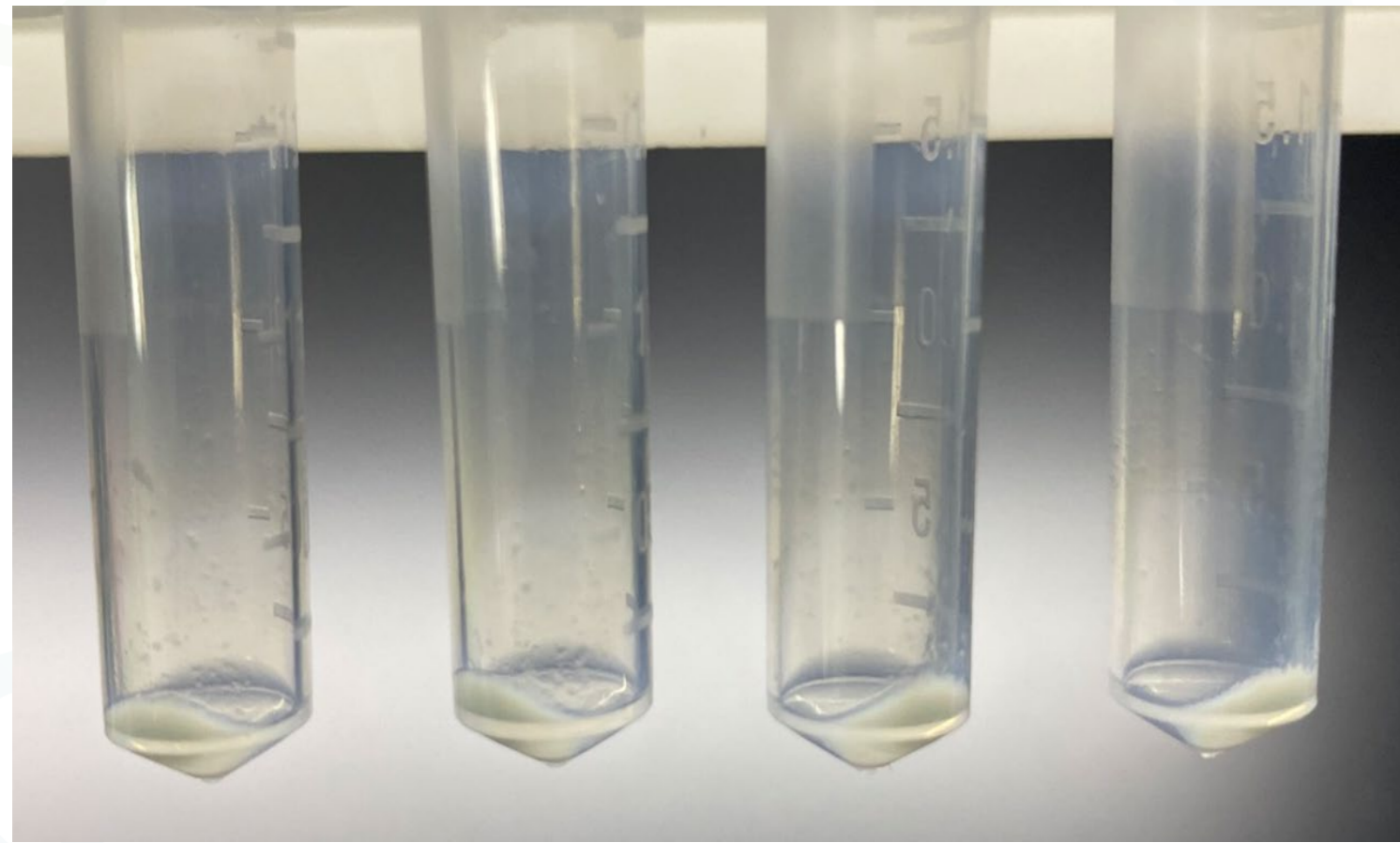


Protein concentration was measured as **6.5 mg/ml** using Bradford Assay Kit (Imultiomics, #MGR02).

B. Solution color changes during CuAAC mix preparation



C. CuAAC reaction in Protein samples

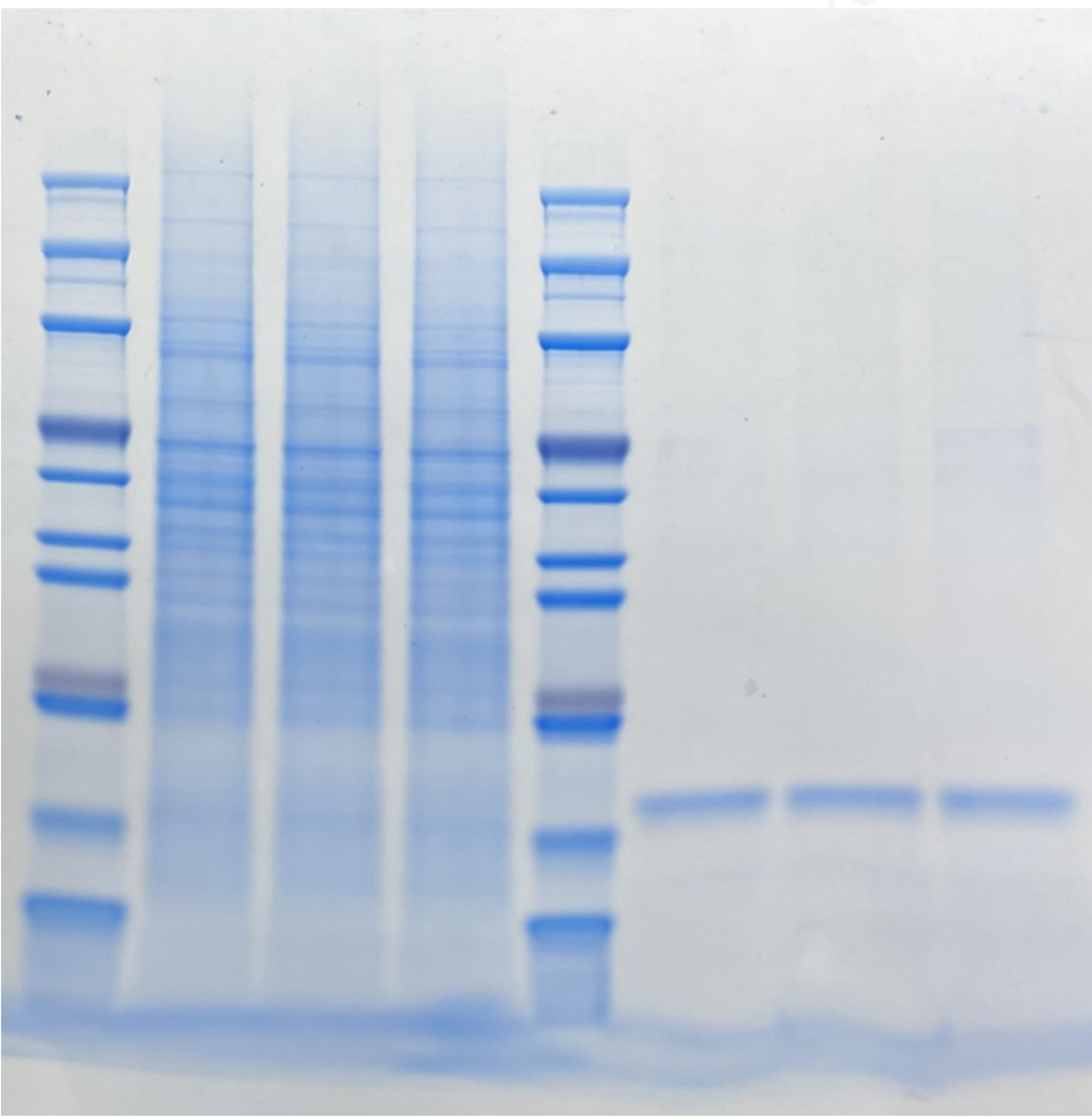
<u>Addition</u> of CuAAC mix			<u>No addition</u> of CuAAC mix	
				
↓ Centrifuge (4°C, 7000g)				
				
Pellet				

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

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

	IR (5μl/lane)			Pull-down (PD)		
	-	50	100	-	50	100
MG07-8 (Positive Control, μM)	-	50	100	-	50	100

Comassie Blue Staining
(Imultiomics, #MGR01)



1. **MG07-8** (Positive Control, PC) in **#MG07 Kit**, the Azide-labeled SM.
2. The pulled-down samples of **MG07-8** (50, 100μM) using **# MG07 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.
5. IR: Input of Resuspension Protein

Note:
MG07-8 is an azide-labeled SM specific targeting cereblon (CRBN) protein widely used in PROTAC (Proteolysis targeting chimera) system. Therefore, the abundance and pattern of protein on SDS-PAGE/western blot enriched by **#MG07 Kit** may be cell/tissue type-dependent.

Western blot
(Streptavidin-HRP)
(MG07-6 in MG07 Kit)

