

Isolation of Splenic Microvesicles in a Murine Model of Intraperitoneal Bacterial Infection

 Hanine El Itawi¹, Fareeha Batool¹, Céline Stutz¹, Abdul Wahid Qureshi¹, Fatiha El Ghazouani^{1,2}, Olivier Huck^{1,2}, Florence Toti^{1,2}
¹Université de Strasbourg ²Regenerative Nanomedicine, Centre de Recherche en Biomédecine de Strasbourg, INSERM (French National Institute of Health and Medical Research)

Corresponding Author

 Florence Toti
 toti@unistra.fr

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Materials

Name	Company	Catalog Number	Comments
2 mL tubes type Eppendorf	Dutscher	54816	Conical bottom stéril microtubes
Allegra 64 R Centrifuge	Beckman Coulter		
Automatic cell counter	Biorad		
Bovine serum albumin	Euromedex	04-100-812-E	Prepared, filtered with 0.22 µm sieve and stored at 4 °C under sterile conditions by using the following formulas: 2 mM EDTA, 0,5% BSA and sterile PBS
CD11 (Mac-1)	e-Biosciences	45-00112-80	Conjugated to eFluor 450; λ _{max} excitation 405 nm λ _{max} emission 445 nm
CD16/32	BD Biosciences	553142	unconjugated
EDTA	Calbiochem	Calbiochem	S 6381-92-6
Falcon tube	Cell star	227261	50 mL
Fetal Bovine serum	Dutscher	S1810-500	Batch number = S14028S1810
Fortessa Aria	BD Biosciences		for cell sorting
Fortessa flow cytometer	Becton-Dickinson.		
Fungizone	PAN biotech	P06-01050	
HBSS	Gibco	14175-053	Without phenol red, without Ca ⁺² and Mg ⁺²
ICAM-1	abcam	ab171123	
LYG-6 (Gr-1)	BD Biosciences	566218	Conjugated to BUV395; λ _{max} excitation 348 nm, λ _{max} emission 395 nm
Lysis buffer erythrocytes (ACK)	Sigma		Prepared, filtered with 0.22 µm sieve and stored at 4°C under sterile conditions by using the following formulas: NH ₄ Cl, 0.15 M (molarity), 53.491 (mw) 4 g KHCO ₃ 1 mM (molarity) 100.115 (mw), 50 mg

			EDTA 0.1 mM (molarity), 292.24 (mw), 14.6 g pH: 7.2–7.4
NanoDrop 1000 spectrophotometer	Thermoscientific		
PBS	Lonza	17-516F	Without Ca ⁺² and Mg ⁺²
Plastic petri dish			100 mm
Polystyren tube	Falcon	352070	
q-Nano Gold	iZON science		
RPMI 1640 culture medium: 2 g/L glucose	PAN biotech	p04-18047	Supplemented withsupplemented with Streptomycin (100 U/mL) / Penicillin (100 U/mL), Fungizone (250 mg/mL), L-glutamine (2 mM) and FBS 10%.
Scalpels			
Sieve Nylon	Falcon USA	352360	100 µm
Streptomycin/Penicillin	PAN biotech	P06-07100	
Syringe			2 mL
Trypan Blue	Biorad	1450013	
VCAM1	abcam	ab215380	