Bronchoalveolar lavage fluid collection and analysis

The mouse lungs are lavaged three times with 1 ml of PBS containing 1% FCS (PBS-1%). The retained BALF is centrifuged at 400 x g for 5 min at 4°C. The recovered supernatants are collected and stored at -70°C until assessed for cytokine concentration, and the cell pellet are resuspended in 200 µl of PBS-1%. Total cell numbers were counted using a hemacytometer, and cytospins were prepared (1 x 10^5 cells/slide), stained with the Hema 3 Staining System (Fisher Diagnostics, Middletown, VA) and differential cell counts were determined.