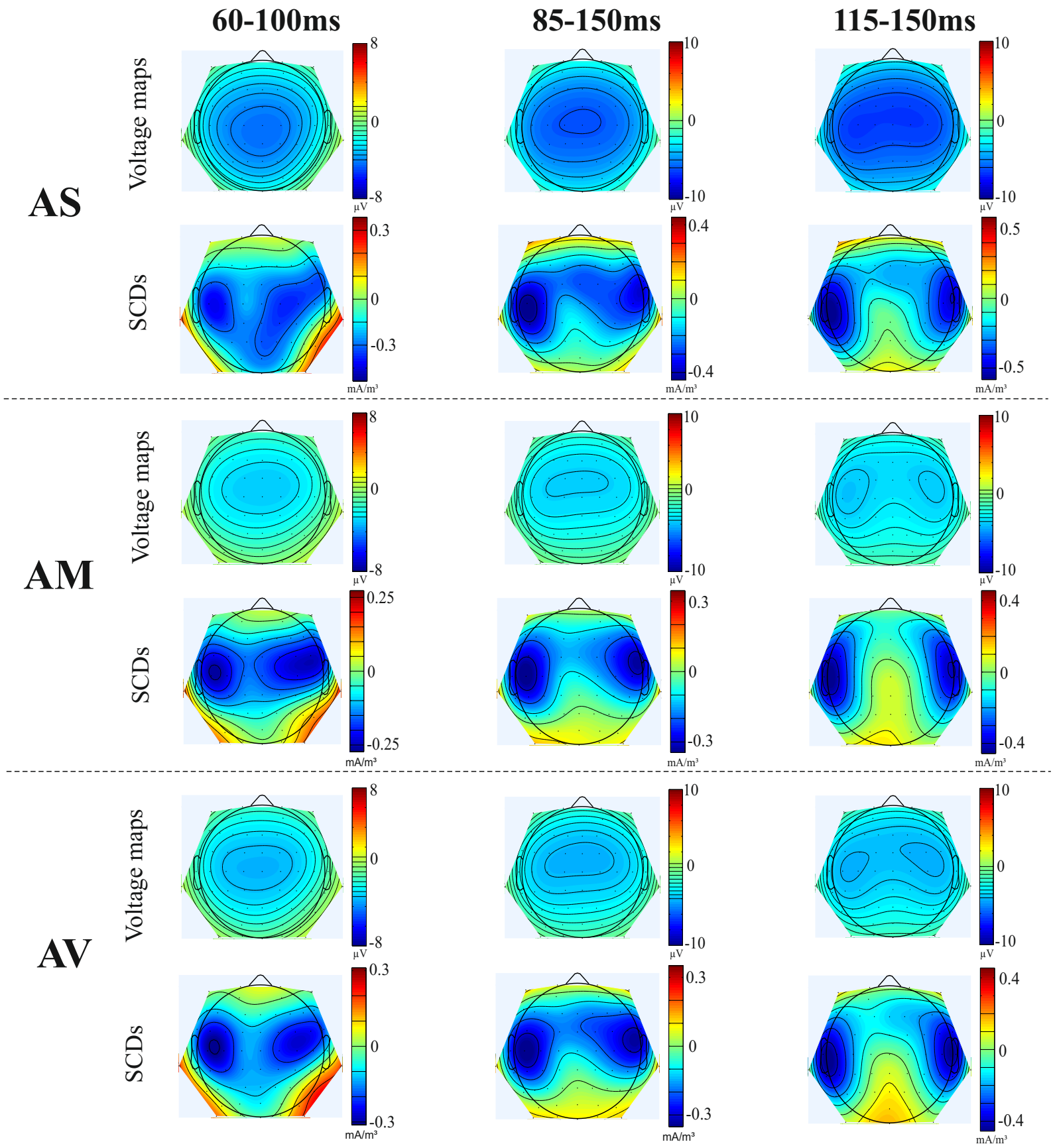Additional file 2: