



Figure S2. Analyses of shark-LA single-cell germline sequences. **Top.** Primers in the V-D1 and D2-JH regions, specific for subfamily, generate PCR products of 1.1-1.2 kb (see Fig. 1, line 4). **Bottom.** The five PCR reactions for each B cell are examined for restriction enzyme sites characteristic to the particular genes cloned from shark-LA. G1 is ascertained by the Pvu II site in the V-D1 region. G2A is differentiated from G2B by separate Nde I and EcoR I digests. G3 contains an ApaI site 3' of D1. G5 contains an EcoR V site in the D1-D2 interval. The G4 fragment is subjected to a series of restriction enzyme analyses to determine those G4 members present. Gene-specific analyses are as follows. G4A is characterized by EcoR I/Ssp I fragment of 605 bp, G4CG by a Sca I/EcoR I fragment of 185 bp. The distance between the 5' terminus and the Sca I site is 64 bp, so that G4A can be differentiated by a 249 bp band in this reaction (no Sca I). G4D is characterized by Hinc II fragment of 500 bp, differentiating it from G4D2, which has a single Hinc II site. The combination of Sca I and Hinc II generates the G4D2-specific fragment of 710 bp.