Supplemental Figure 1

Confirmation of -1189 ERE mutation

Mutation of the WT ERE results in the formation of an EcoNI restriction site. Eight colonies were screened by amplification across the target region and subsequent digest of the PCR reaction with EcoNI. WT band is 332bp, whereas in modified constructs (including plasmid control from [9]) digestion resulted in two bands of 191 and 141bp. Sequencing confirmed these clones were correctly mutated (a) Southern blots and Pulsed Field Gel electrophoresis were used to confirm the overall BAC structure was maintained of the 8 clones tested. Probes targeting 5' region of the BAC (probe I), the central region (probe II) and the 3' region (probe III) confirmed the presence of these regions in all BACs, as in the WT BAC. (b)(i) SalI and NotI digestion and PFGE confirms BAC overall structure is maintained (b)(ii).

