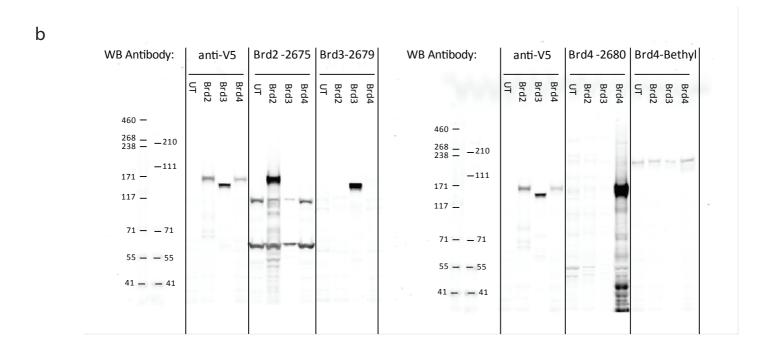
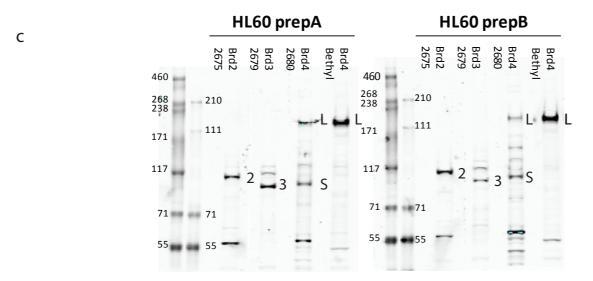
Supplementary Figure 1

a GSK BRD2 2675 epitope: aa192-aa211, PKNSHKKGAKLAALQGSVTS GSK BRD3 2679 epitope: aa153-aa169, KGKGRKPAAGAQSAGTQ GSK BRD4 2680 epitope: aa2-aa16, SAESGPGTRLRNLPV Bethyl BRD4 A301-985A epitope: aa1312-1362 in NP 490597.1





Supplementary Figure 1: Antibody epitopes and western blotting. a. To generate selective polyclonal antibodies against human BRD2, BRD3 and BRD4 specific non homologous peptides were synthesized (GSK BRD2 2675: aa192-aa211, CPKNSHKKGAKLAALQGSVTS; GSK BRD3 2679: aa153-aa169, CKGKGRKPAAGAQSAGTQ; GSK BRD4 2680: aa2-16, SAESGPGTRLRNLPVC) and used for immunization of rabbits and affinity purification. b. Specificity of binding and absence of cross-reactivity was confirmed using GFP and V5 epitope tagged Bet constructs. Whole cell lysates from untransfected HEK293 cells (UT) or HEK293 cells transfected with plasmids containing V5-GFP tagged human BRD2, BRD3 or BRD4 (short isoform - aa 1-722), were separated by Tris-acetate SDS PAGE and used to test the antibodies by Western-blotting. An alternative antibody against BRD4-L from Bethyl Laboratories (A301-985A) was also included, along with an antibody used to detect the V5 tagged constructs (Abcam Ab9116). Each of the Bet antibodies specifically recognised proteins corresponding to the appropriate transfected sample and each aligned to bands in samples probed with the anti-V5 antibody. The Bethyl Laboratories antibody only detected a band equivalent to the molecular weight of full length endogenous long BRD4 isoform, due to its epitope at the C-terminus of FL BRD4 (aa1312-1362). c. Selective recognition of endogeneous BET proteins was tested by Western-blotting of nuclear extracts from HL-60 cells. An additional antibody against BRD4 from Bethyl Laboratories (A301-985A) was included. Numbers 2, 3 and L or S indicate the location of bands corresponding to the expected molecular weight of endogenous untagged BET proteins BRD2, BRD3, BRD4-long and BRD4-short respectively. Replicate experiments from two independent HL-60 preparations are shown. Each antibody recognised a protein of the appropriate molecular weight but not at the molecular weight of the other BET proteins.