Expanded View Figures

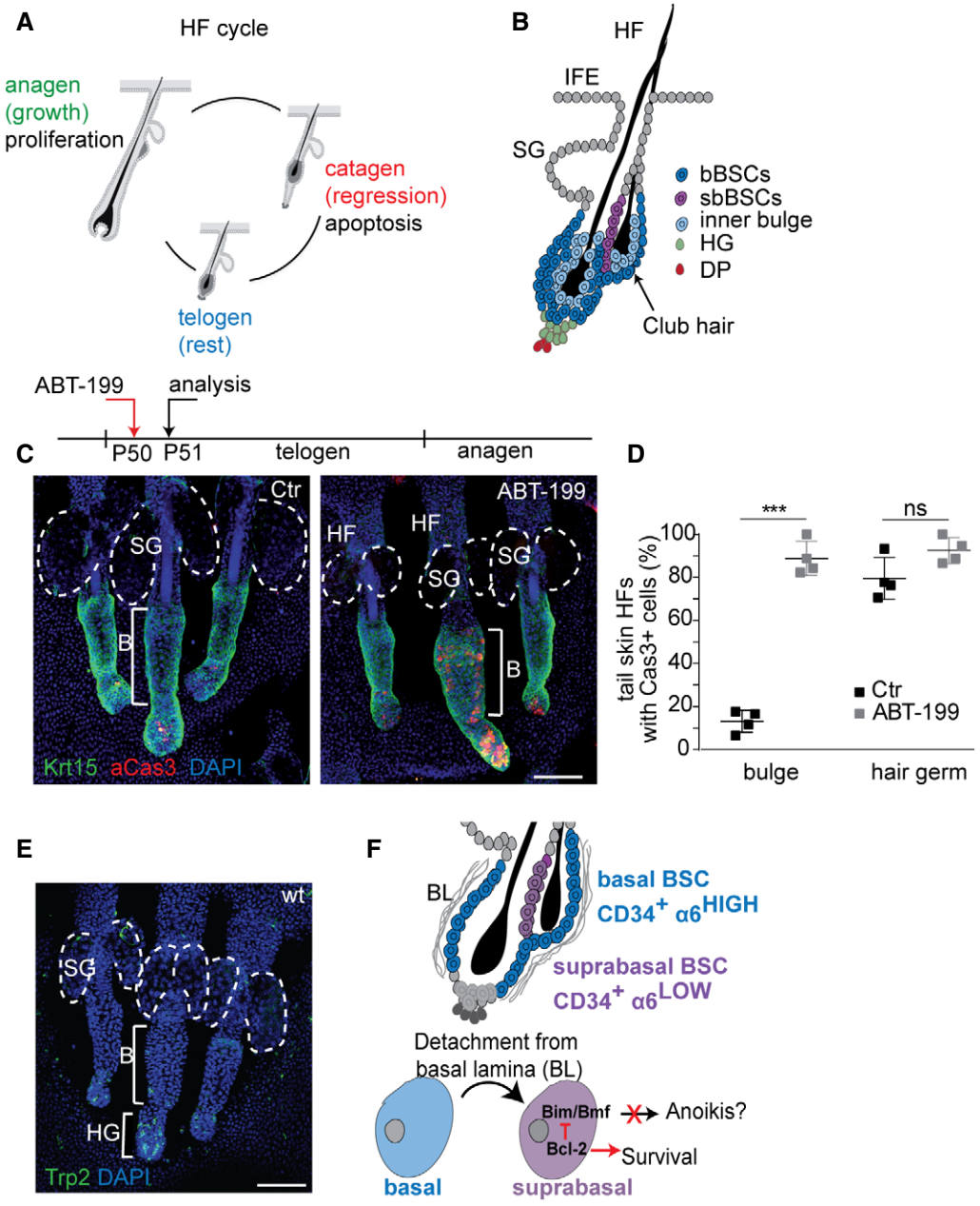

Figure EV1. Bcl-2 inhibition causes SC-specific cell death and loss of suprabasal BSCs.

A Schematic of the hair cycle phases anagen (growth), catagen (regression) and telogen (rest).

B HF Schematic focusing on basal and suprabasal bulge SCs (bBSCs, sbBSCs), the hair germ (HG) and the dermal papilla (DP). IFE, interfollicular epidermis; SG, sebaceous glands.

C Timeline experimental ABT-199 treatment (P50) and analysis (P51) in telogen and Immunofluorescence staining of telogen tail whole mounts for Keratin 15 (Krt15) and active caspase-3 (aCas3) in control (Ctr) and Bcl-2 inhibitor (ABT-199)-treated mice. B, bulge; HF, hair follicle; SG, sebaceous gland; dashed lines, sebaceous glands. Scale bar, 100 μm.

D Quantification of tail HF with $aCas3^{+}$ cells in bulge and hair germ of control (Ctr) and ABT-199-treated mice (n = 4 biological replicates; mean ± standard deviation (SD); ***$P < 0.001$, ns=not significant, two-tailed unpaired Student's t-test.

E Immunofluorescence staining of telogen tail whole mounts for tyrosinase-related protein-2 (Trp2) in wild-type (wt) animals (n = 3 mice). SG, sebaceous gland; B, bulge; HG: hair germ; dashed lines: sebaceous glands. Scale bar, 100 μm.

F Model for Bcl-2-mediated protection of suprabasal BSCs from apoptosis.

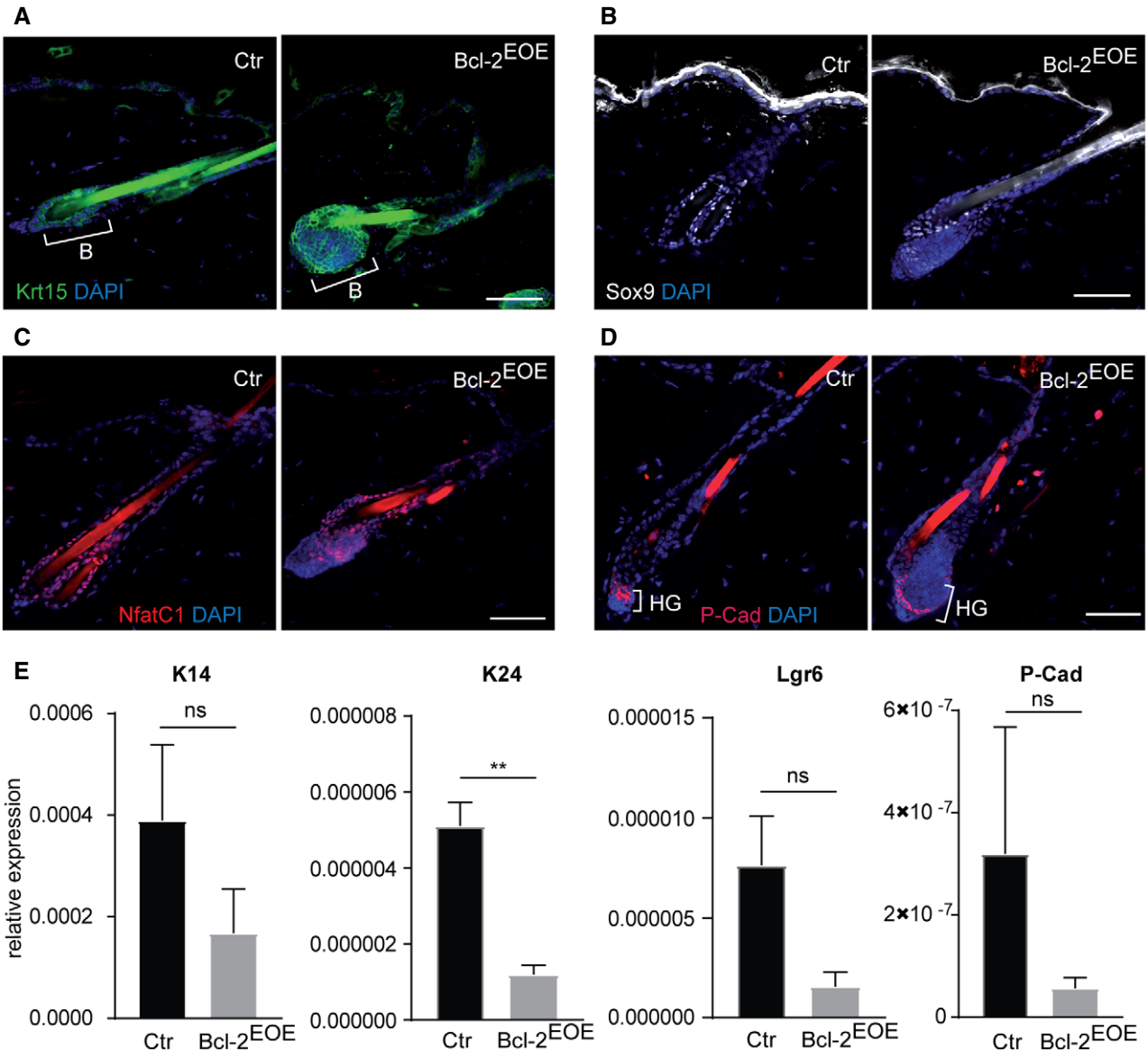

Figure EV2. Hair follicle markers in Bcl-2^{EOE} mice.

A–D Immunofluorescence staining of telogen back skin hair follicle for Keratin 15 (Krt15, green) (A), Sox9 (white) (B), NfatC1 (red) (C) and hair germ marker P-cadherin (P-Cad, red) (D) in control (Ctr) and Bcl-2^{EOE} mice. B: bulge, HG: secondary hair germ. Scale bars, 50 μm.

E qRT–PCR for HF markers K14, K24, Lgr6 and P-Cad mRNA expression in telogen back skin epidermis of control (Ctr) and Bcl-2^{EOE} mice. (n = 4 biological replicates; mean ± standard error of mean (SEM); $^{**}P < 0.01$, ns, not significant, two-tailed unpaired Student's t-test).

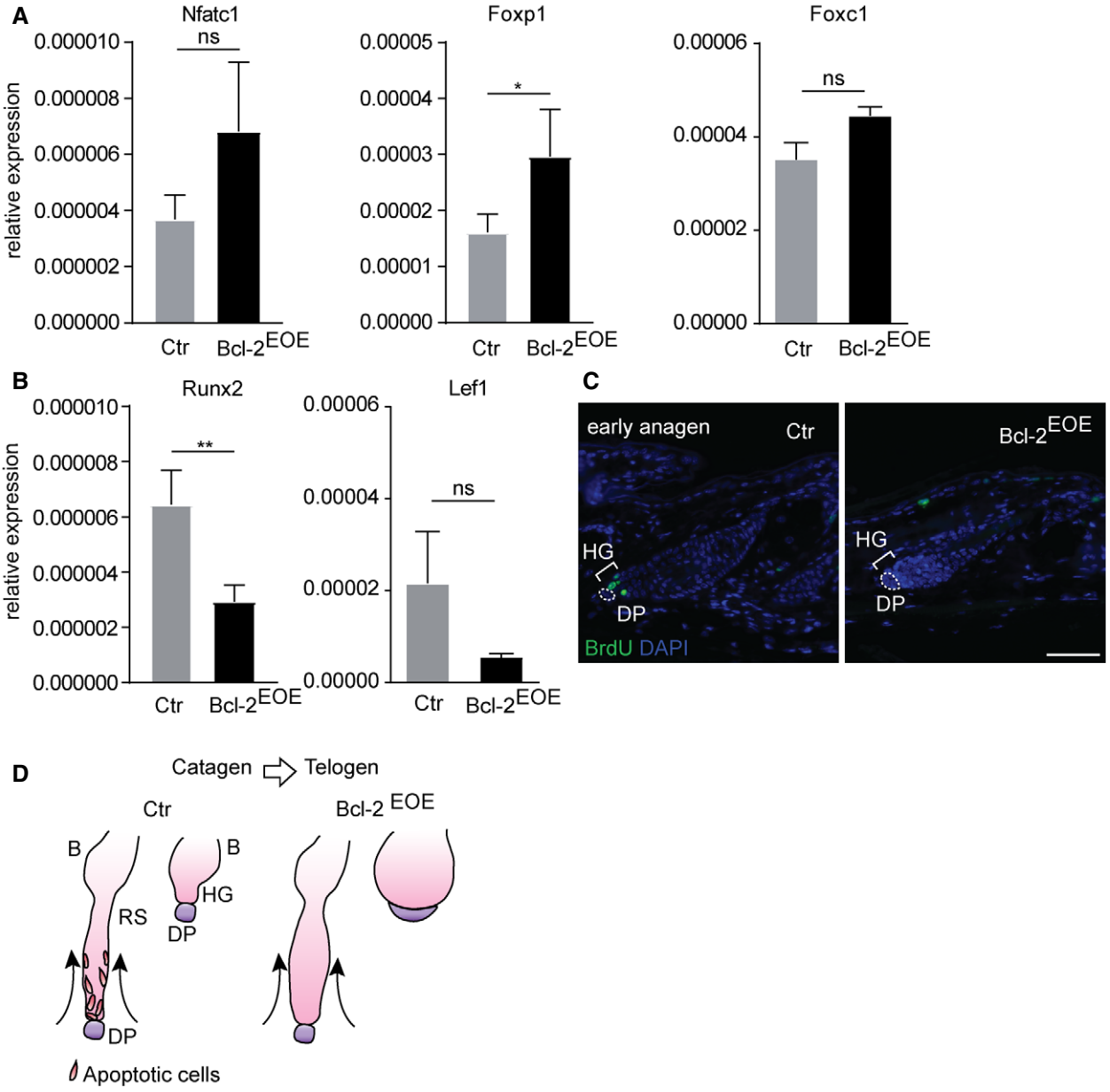

Figure EV3. Increase in bulge and abnormal hair germ compartment impairs HF growth induction in Bcl-2^{EOE} mice.

A, B qRT–PCR for mRNA expression of SC quiescence-related markers NfatC1, Foxp1 Foxc1 (A) and HF activation-related markers Runx2, Lef1 (B) in epidermis of control (Ctr) and Bcl-2^{EOE} mice (P27), (n = 4 biological replicates; mean ± standard error of mean [SEM]; *P < 0.05, **P < 0.01; ns, not significant, two-tailed unpaired Student's t-test).

C Immunofluorescence staining of back skin hair follicles for BrdU incorporation (1 h pulse) in control (Ctr) and Bcl-2^{EOE} mice for hair growth initiation (n = 3 biological replicates). Dashed lines and DP, dermal papilla; HG, hair germ. Scale bar, 50 μm.

D Schematic model of hair follicle retraction in catagen hair follicles and bulge architecture comparing control and Bcl-2^{EOE} mice.

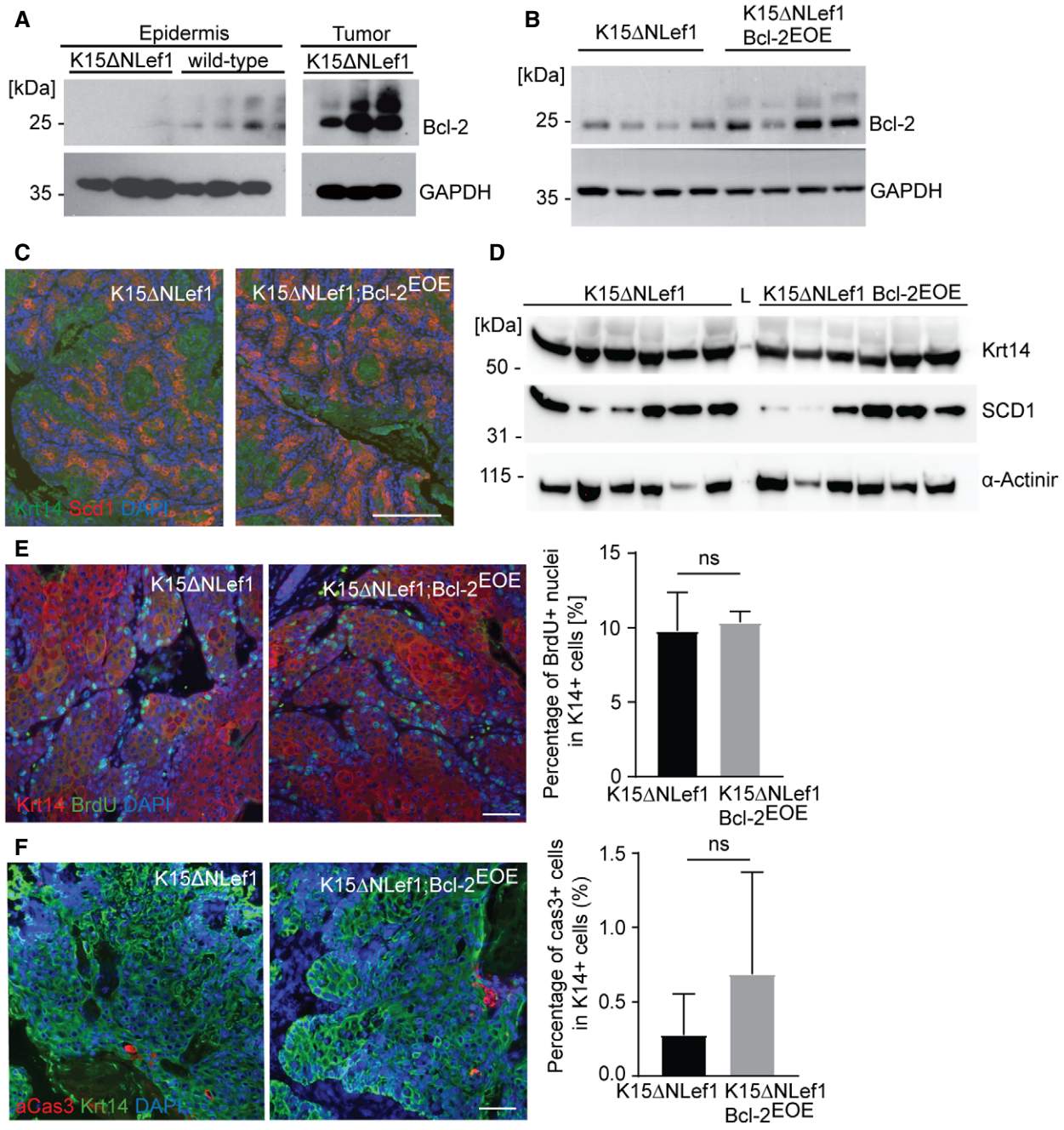

Figure EV4. BSCs expressing high levels of Bcl-2 are selected for tumour initiation.

A, B Western blot analysis for Bcl-2 protein levels in K15ΔNLef1 and wild-type epidermis and K15ΔNLef1 tumours (n = 3 biological replicates) (A) and Bcl-2 levels in tumours of K15ΔNLef1 and K15ΔNLef1;Bcl-2^{EOE} mice (n = 4 biological replicates) (B).

C Immunofluorescence staining of K15ΔNLef1 and K15ΔNLef1;Bcl-2^{EOE} tumours, stained for Krt14 (green) and the sebaceous differentiation marker Scd1 (red). Scale bar, 100 μm.

D Western blot analysis for Scd1 and Krt14 protein levels in tumours of K15ΔNLef1 and K15ΔNLef1;Bcl-2^{EOE} mice (n = 6 biological replicates).

E Immunofluorescence staining of Krt14 (red) and BrdU (green) in K15ΔNLef1 and K15ΔNLef1;Bcl-2^{EOE} tumours and quantification of the proliferation rate in K14^{+} cells (n = 3 biological replicates; mean ± standard error of mean (SEM); ns, not significant, two-tailed unpaired Student's t-test).

F Immunofluorescence staining of aCas3 (red) and Krt14 (green) in K15ΔNLef1 and K15ΔNLef1;Bcl-2^{EOE} tumours and quantification of the apoptosis rate in K14^{+} cells (n = 3 biological replicates; mean ± standard error of mean (SEM); ns, not significant, two-tailed unpaired Student's t-test).

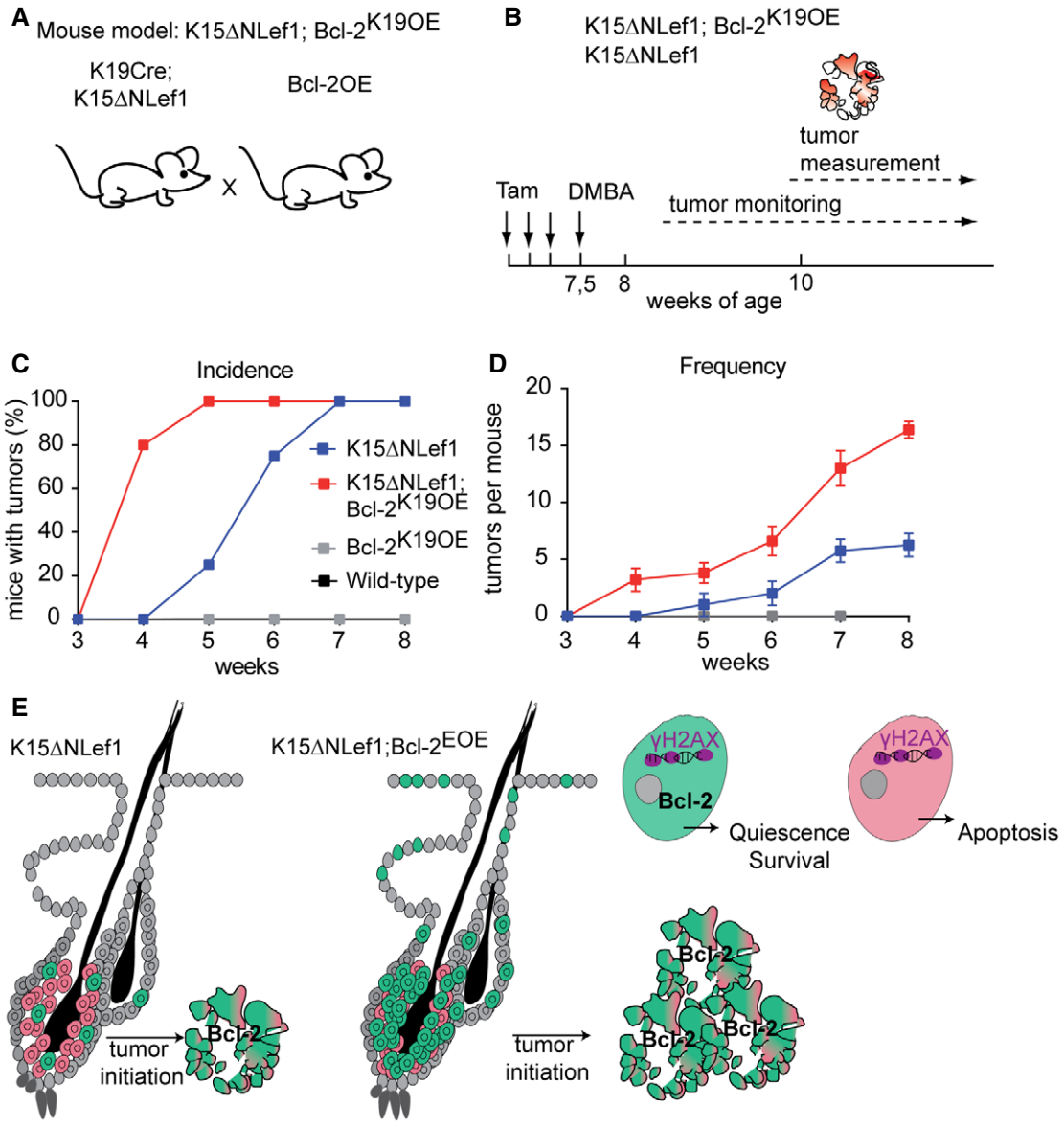

Figure EV5. Bulge-specific Bcl-2 overexpression promotes SC-driven tumour initiation.

A Experimental strategy generating K15ΔNLef1;Bcl-2^{K19OE} mice.

B Schematic of tumour experiment timeline in K15ΔNLef1 and K15ΔNLef1;Bcl-2^{K19OE} genetic background.

C, D Tumour incidence (C) and frequency (D) in wild-type, K15ΔNLef1, Bcl-2^{K19OE} and K15ΔNLef1;Bcl-2^{K19OE} mice ($n \geq 5$ biological replicates; mean ± standard error of mean [SEM]).

E Model presenting selection of Bcl-2 high expressing BSCs for tumour initiation in K15ΔNLef1 mice and demonstrating increase in tumour-initiating BSCs in K15ΔNLef1;Bcl-2^{EOE} mice.