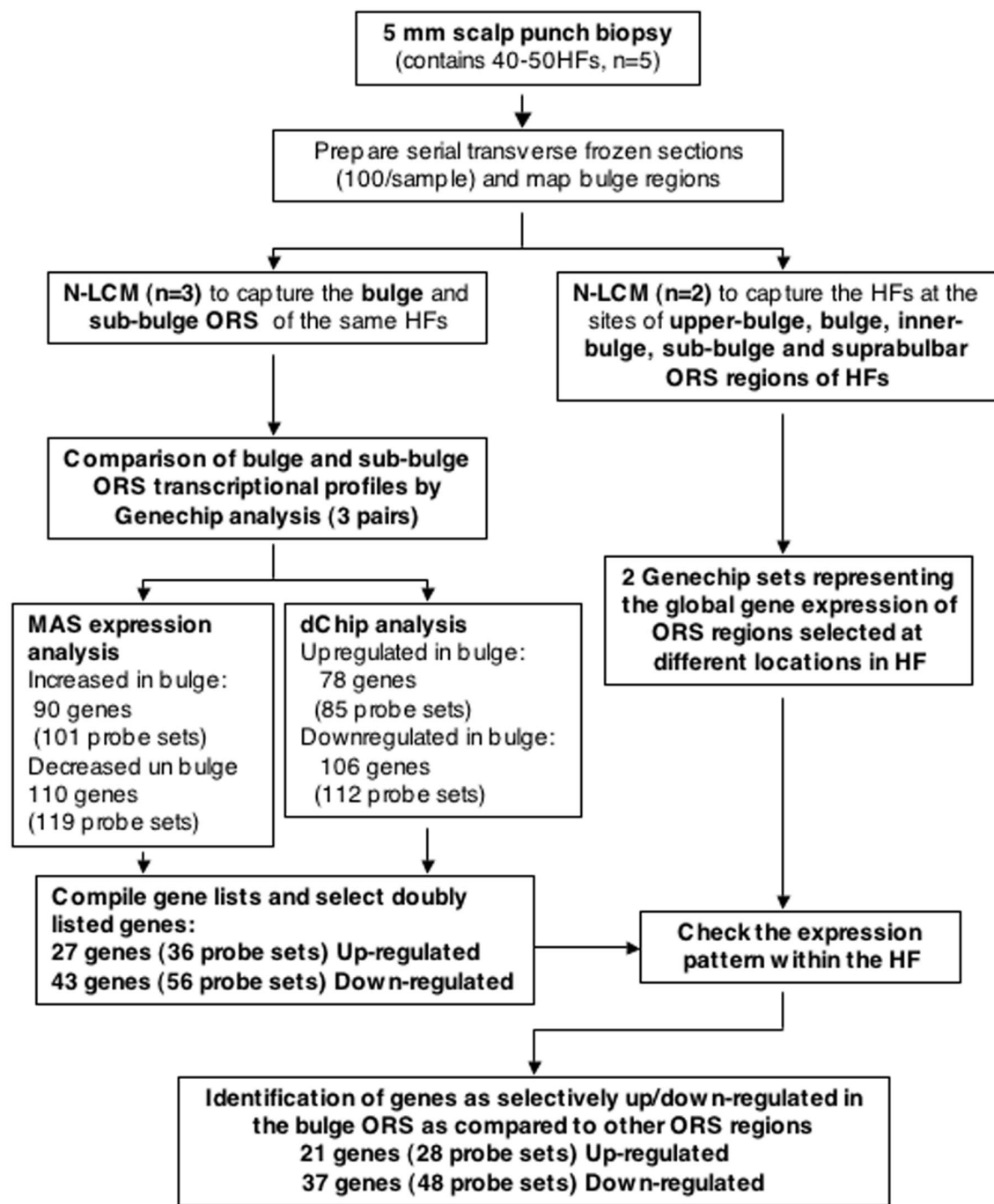
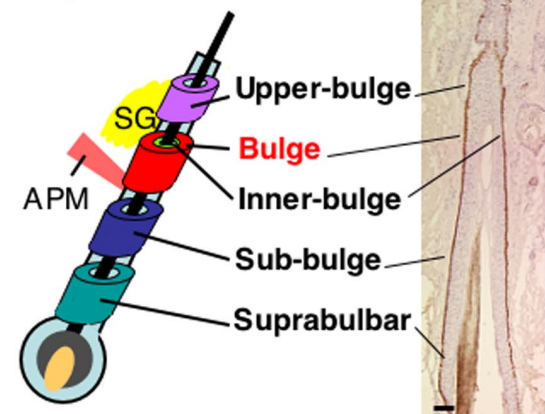


S Figure 1

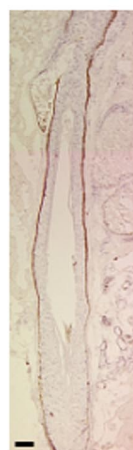


S Figure 2



KRT15
(204734_at)

Upper-bulge	5776.5 P	5301.7 P
Bulge	5969.4 P	5013.0 P
Inner-bulge	149.5 A	301.0 P
Sub-bulge	5653.7 P	4846.8 P
Suprabulbar	3157.0 P	3524.6 P
Epidermis*	2633.6 P	



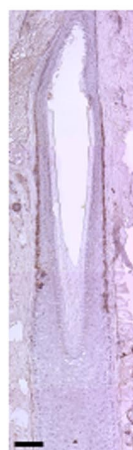
FST
(204948_s_at)

Upper-bulge	298.8 P	331.9 P
Bulge	252.4 A	179.8 P
Inner-bulge	391.4 A	163.9 A
Sub-bulge	163.9 A	122.6 A
Suprabulbar	115.2 A	103.2 A
Epidermis*	107.7 A	



FZD1
(204451_at)

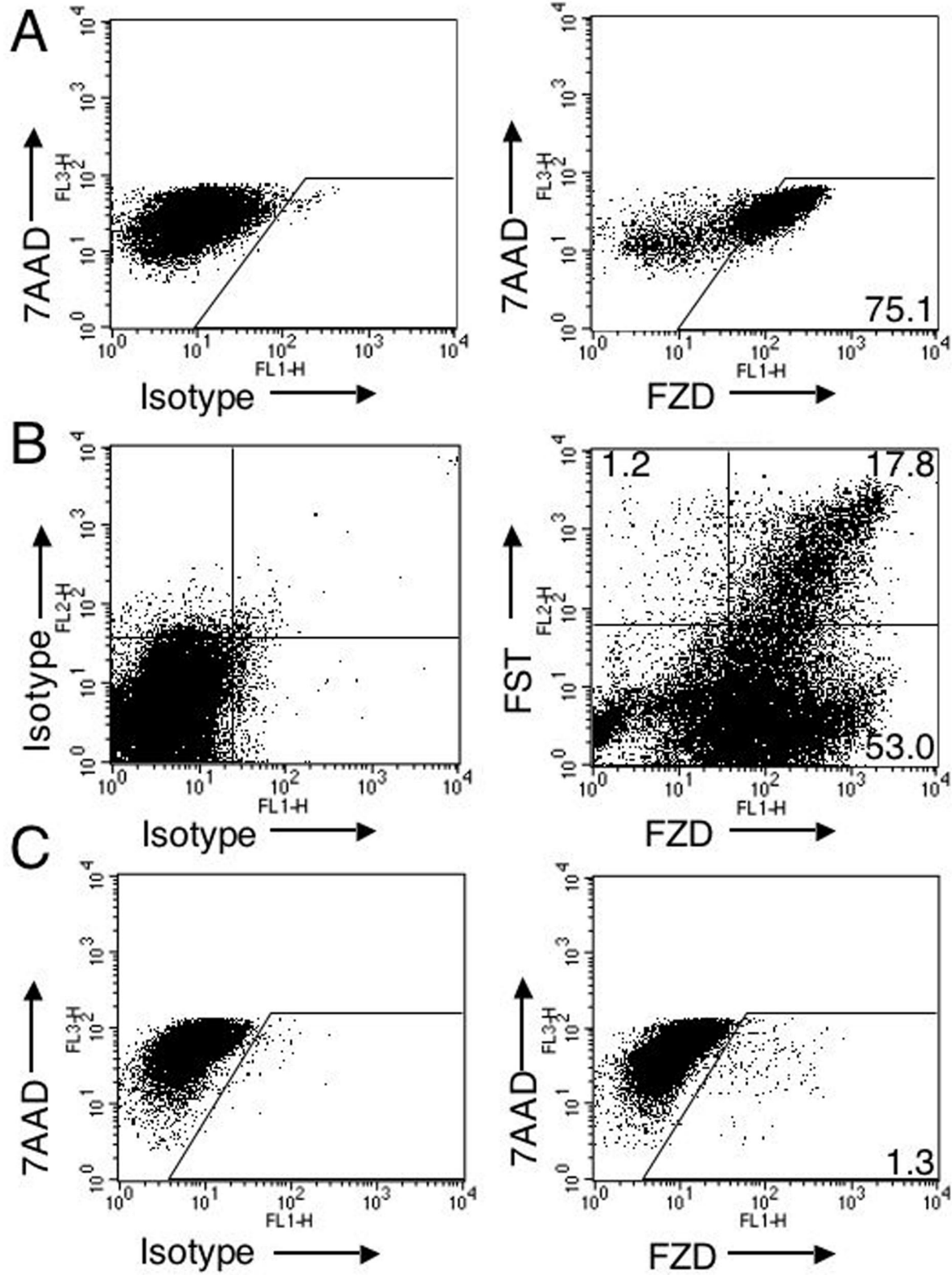
Upper-bulge	438.8 P	732.9 P
Bulge	385.1 P	692.6 P
Inner-bulge	219.6 A	224.2 P
Sub-bulge	394.3 P	435.9 P
Suprabulbar	280.0 P	258.3 A
Epidermis*	284.9 P	



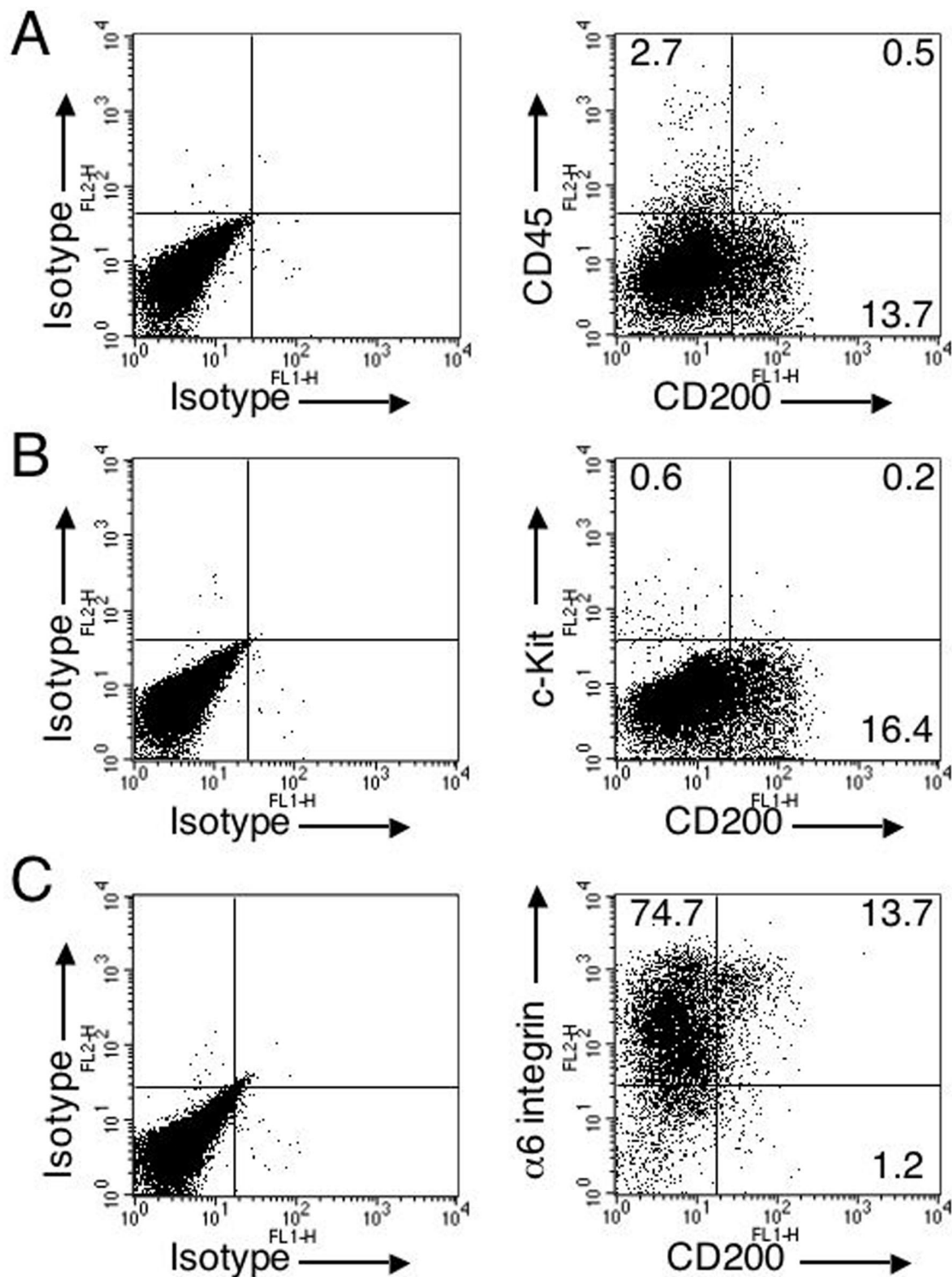
PHLDA1
(217996_at)

Upper-bulge	235.0 P	196.4 P
Bulge	210.2 P	120.0 P
Inner-bulge	67.3 P	73.4 P
Sub-bulge	67.5 P	61.2 P
Suprabulbar	46.0 P	84.0 P
Epidermis*	189.9 P	

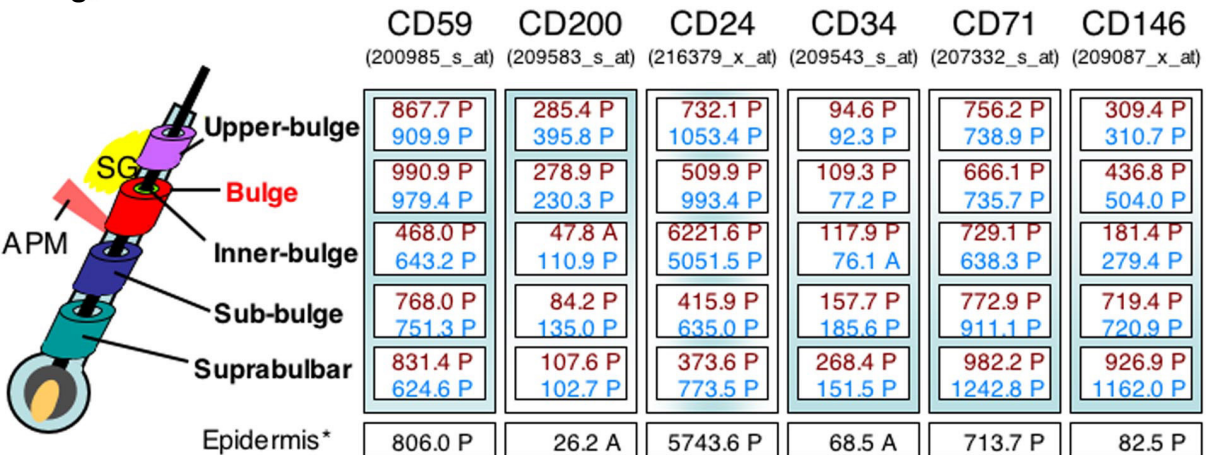
S Figure 3



S Figure 4

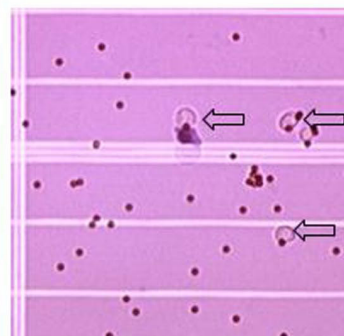
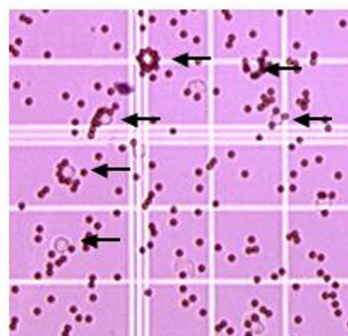
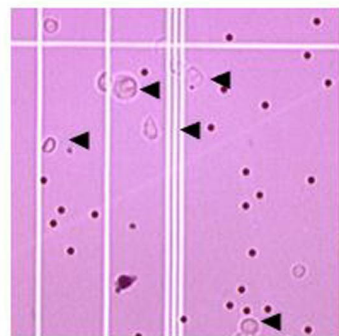


S Figure 5

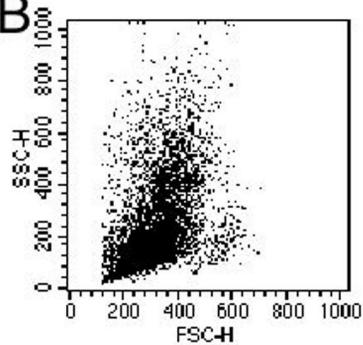


S Figure 6

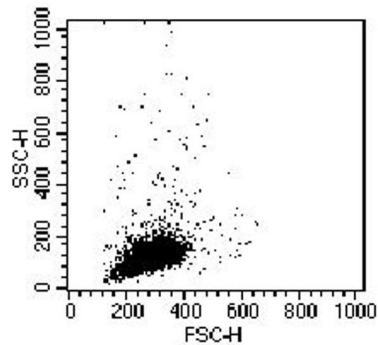
A



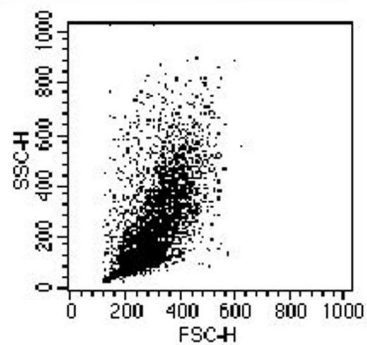
B



mid-HF



CD200^{hi}BNC^{lo}



CD59⁺

Supplemental Figure 1

Genechip data analysis. Three pairs of bulge and sub-bulge ORS Genechips were analyzed and compared by MASv5.0 and dChip 1.3 software algorithms in order to identify gene transcripts that were over-represented or under-represented in the bulge ORS by both algorithms. Two additional sets of Genechips were generated from the ORS regions of upper-bulge, bulge, inner-bulge, sub-bulge and suprabulbar ORS in order to precisely localize gene expression within the hair follicle. Genes that were not selectively and specifically over- or under-represented in the bulge ORS were excluded. Each Genechip pair or set was generated from an individual volunteer.

Supplemental Figure 2

The correlation between mRNA expression patterns and immunohistochemical findings for gene transcripts over-represented in bulge ORS. The upper panels demonstrate human hair follicle vertical sections stained with designated mAbs. Scale bars: 50 μ m. The lower boxes indicate the Genechip signal intensities (mRNA expression levels) detected for individual genes from corresponding ORS subsets. Genechips were normalized to the same parameters. The gene expression patterns correlated well with the immunohistochemical staining patterns of gene products in the ORS. Red and blue numbers indicate individual samples. P: detected as “expressed” or present according to MASv5.0 algorithm. A: detected as “not expressed” or absent according to MASv5.0 algorithm. Parenthesis; Probe set ID of Affymetrix U-133 Genechip sets. *Epidermis; Signal intensities in Genechips generated from human foreskin epidermis are shown.

Total RNA was isolated from epidermal sheet and RNA probes were generated by single-round amplification. Normalized to the same parameters as ORS Genechips.

Supplemental Figure 3

Anti-FZD1 monoclonal antibody could detect the bulge ORS cells in fixed mid-follicle cell suspension but not living mid-follicle cells. **(A)** Consistent with microarray and immunohistochemistry data, approximately 70% of fixed mid-follicle cells were variably FZD1 positive by FACS analysis. **(B)** The FZD1^{hi}FST^{hi} cells in fixed mid-follicle suspension represented bulge ORS cells. **(C)** Anti-FZD1 antibody could only detect approximately 1% of living mid-follicle cells, suggesting that FZD1 detection by this mAb requires fixation.

Supplemental Figure 4

Mid-follicle CD200^{hi} cells were CD45^{lo} c-Kit^{lo} and α 6 integrin^{hi}. **(A)** A mid-follicle cell suspension was stained for CD200 and CD45. Most of CD200^{hi} cells were distinct from CD45^{hi} bone marrow derived cells. **(B)** A mid-follicle cell suspension was stained for CD200 and c-Kit (CD117). Most of CD200^{hi} cells were distinct from c-Kit^{hi} melanocytes or mast cells. **(C)** A mid-follicle cell suspension was stained for CD200 and α 6 integrin (CD49f). Most CD200^{hi} cells were α 6 integrin^{hi}. Gated to non-fixed living cells based on 7-AAD negative staining.

Supplemental Figure 5

Genechip data sets for the CD markers identified in human hair follicle. Numbers

in boxes indicate the signal intensities for each CD marker (mRNA expression levels) in the indicated ORS subsets. Red and blue numbers indicate individual experiments. P: detected as “expressed” or present according to MASv5.0 algorithm. A: detected as “not expressed” or absent according to MASv5.0 algorithm. Parenthesis; Probe set ID of Affymetrix U-133 Genechip sets. *Epidermis; signal intensities in Genechips generated from human foreskin epidermis. Total RNA was isolated from epidermal sheet and RNA probes were generated by single-round amplification. Normalized to the same parameters as ORS Genechips.

Supplemental Figure 6

Isolated CD200^{hi}BNC^{lo} bulge cells were uniform, relatively compact and smooth when compared to mid-follicle cells or CD59+ hair follicle cells. CD200^{hi}BNC^{lo} bulge cells were isolated from mid-follicle cell suspension by magnetic beads selection. **(A)** By light microscopy, isolated living bulge CD200^{hi} cells (Arrows) possessed compact and smooth morphology compared to mid-follicle (arrowheads) cells or CD59^{hi} cells (open arrows) isolated from mid-follicle suspension. Stained with trypan blue. Original magnification 400x. **(B)** FACS analysis confirmed the microscopic observation. Living 7-AAD negative cells were gated. Magnetic beads were also gated out.

Supplemental table 1. Genes detected as up-regulated in human bulge by MAS 5.0 software

Table with columns: Gene, Probe set, Mouse bulge, Bulge 1, Sub-bulge 1, Fold change 1, Bulge 2, Sub-bulge 2, Fold change 2, Bulge 3, Sub-bulge 3, Fold change 3, Average Fold change. Lists genes such as trinitrocellulose repeat containing 9 (TNRC9), deiodinase, iodothyronine, type II (DIO2), and others with their corresponding expression data.

Numbers indicate signal intensities detected for each probe sets. P: "present" call, detected as expressed, A: "Absent" call, detected as not expressed.

Bold: Up-regulated in both MAS 5.0 and dChip analysis

Underline: "membrane" protein annotated by GOA analysis

Mouse bulge column: asterisks indicate the up-regulation of same or similar genes in mouse bulge transcription profiling data

** Tumber et al. Science, 303 359-363, 2004.

** Morris et al. Nat Biotechnol, 22 411-417, 2004.

** Fuchs et al. Cell 116 769-778, 2004.

parenthesis: gene of similar function was detected as unregulated.

