SUPPORTING INFORMATION MATERIALS AND METHODS

FACS-sorting and RNA isolation

MECs were isolated from twenty 10-week old C57BL/6 wildtype mice and stained with antibodies that recognize cell surface stem/progenitor cell markers (Table S2) as described in Materials and Methods. Cells were sorted on a FACS Aria Cell Sorting Flow Cytometer (BD Biosciences) and RNA was isolated from each subpopulation using the Arcturus Pico Pure RNA isolation kit (Molecular Devices, Sunnyvale, CA). A post-sort analysis was performed to assess the purity of the cell populations and was estimated to be 99.6% for LIN$^-$ cells, 98.6% for LIN$^-$CD24$^+$CD29^{hi} cells, 97.9% for LIN$^-$CD24^{hi}CD29^{lo}CD61$^+$ cells, 98.1% for LIN$^-$CD24^{hi}CD29^{lo}CD61$^-$ cells, 98.9% for LIN$^-$CD24^{hi}Sca1^{hi} cells, 98.3% for LIN$^-$CD24^{hi}Sca1^{lo} cells, 97.4% for LIN$^-$CD24^{lo}Sca1^{lo} cells, and 97.1% for LIN$^-$CD24^{lo}Sca1$^-$ cells. The mRNA was then amplified using a NuGEN FFPE amplification kit (NuGEN, San Carlos, CA) and qPCR was performed as described in Materials and Methods. The experiment was performed three times.

Affymetrix microarray analysis

MECs were isolated from 2 paired sets of wildtype and germline C/EBPβ$^{-/-}$ glands (10 mice per group), FACS-sorted based on CD24, CD29 and LIN expression, and total RNA was isolated using the Arcturus Pico Pure RNA isolation kit (Molecular Devices). A post-sort analysis was performed to assess the purity of the cell populations and was estimated to be 98.4% ± 0.3 for LIN$^-$CD24^{hi}CD29^{lo} cells and 98.7% ± 0.4 for LIN$^-$CD24$^+$CD29^{hi} cells. The mRNA was then amplified using a NuGEN FFPE amplification kit (NuGEN), followed by DNA fragmentation, biotinylation and hybridization onto Affymetrix Mouse 430 2.0 array chips. CEL files were processed using dChip (PM-MM model, quantile normalization) [55]. Probe sets with an

average signal <40 units were omitted. We performed ANOVA analysis for differential

expression among any of the 4 experimental groups. Within each of the 2 subpopulations

profiled (LIN⁻CD24^{hi}CD29^{lo}, LIN⁻CD24^{+}CD29^{hi}), log-transformed gene expression values were

centered on the average of the corresponding wildtype group. Using this centered version of the

dataset, significant genes were clustered using the method described by Creighton *et al* [56].

Expression array data can be accessed at the National Center for Biotechnology Information

Gene Expression Omnibus (GEO) Database, accession number GSE16380.

SUPPORTING INFORMATION FIGURE LEGENDS

Figure S1. C/EBPβ mRNA is enriched in CD24^{lo}Sca1^{-} progenitors. MECs were isolated from

10-week old C57BL/6 wildtype mice and RNA was isolated from FACS-sorted stem/progenitor

cell populations. **(A)** Dot/contour plots depict specific gates drawn for each subpopulation that

was sorted. For all sorted cell populations, lineage-positive cells were excluded using a mouse

lineage panel kit plus biotin-conjugated CD31 and CD140a antibodies. **(B)** qPCR showed that

C/EBPβ was enriched in the LIN⁻CD24^{lo}Sca^{-} as compared to all other subpopulations analyzed

(n=3).

Figure S2. Differentially expressed genes in C/EBPβ$^{-/-}$ stem/progenitor cell subpopulations. **(A)**

Dot plots depict specific gates drawn for each subpopulation that was sorted. For all sorted cell

populations, lineage-positive cells were excluded. **(B)** Heat map illustrates 181 differentially

expressed transcripts that were significantly changed (p<0.001, ANOVA) in C/EBPβ$^{-/-}$ MECs as

compared to wildtype in either LIN⁻CD24^{hi}CD29^{lo} or LIN⁻CD24^{+}CD29^{hi} subpopulations (fold

change >1.4). Among this group, 46 probe sets (ps) represented genes that appeared

differentially expressed in the LIN⁻CD24^hiCD29^lo subpopulation alone (19 up and 27 down), 58

ps were altered in the LIN⁻CD24⁺CD29^hi subpopulation, and 77 ps were significantly changed in

both subpopulations. Each row represents a transcript, and each column depicts the

subpopulation analyzed in wildtype or C/EBPβ^-/- cells. The yellow color indicates a high level of

expression as compared to wildtype, the blue color indicates a low level of expression, and black

depicts no change. The numbers of gene transcripts (i.e. Affymetrix probe sets) for each gene

cluster are indicated.

SUPPORTING INFORMATION TABLES

Table S1: Primer sequences for real-time PCR

Gene Name	Sense Primer (5′-3′)	Antisense Primer (5′-3′)
C/EBPβ[a]	TGATGCAATCCGGATCAA	CACGTGTGTTGCGTCAGTC
Pbx-1[a]	CAACAGTGCGCTGCTAAGG	CAGTTGGAGGTATCAGAGTGAACA
Krt15[a]	GGAAGAGATCCGGGACAAA	TGTCAATCTCCAGGACAACG
Krt5[a]	TGGAGATCGCCACCTACAG	TGACTGGTCCAACTCCTTCC
Sca-1[a]	CCCCTACCCTGATGGAGTCT	TGTTCTTTACTTTCCTTGTTTGAGAA
Tagln[a]	CCTTCCAGTCCACAAACGAC	GTAGGATGGACCCTTGTTGG
Eya-1[a]	GGGTCTTTAGACAGTTTCTCAGGT	TGGGTATGATCTGTTGGAAGG
Notch3[c]	AGATCAATGAGTGTGCATCC	GCAGACTCCATGACTACAGG
ΔNp63	GAAAACAATGCCCAGACTCAA	TGTGCGTGGTCTGTGTTG
18S rRNA[b]	GTAACCCGTTGAACCCCATT	CCATCCAATCGGTAGTAGCG

[a] Sequences were designed using the Universal ProbeLibrary Assay Design Center, Roche Applied

Biosciences (http://qpcr.probefinder.com/roche3.html)

[b] Sequences were derived from Asselin-Labat *et al* [1]

[c] Sequences were derived from Bouras *et al* [43]

Table S2: List of Antibodies

Antibody	Vendor	Catalog Number
CD24-PE	BD Biosciences	553262
CD24-FITC	BD Biosciences	553261
CD29-FITC	Biolegend	102206
CD61-Alexa Fluor® 647	Biolegend	104314
Sca1(Ly6A/E)-PE	BD Biosciences	553336
LIN-biotin[a]	BD Biosciences	559971
CD140a-biotin[a]	eBioscience	13-1401
CD31-biotin[a]	BD Biosciences	553371
Streptavidin-APC	BD Biosciences	554067
Streptavidin-PE-Cy7	BD Biosciences	557598

[a] Included in lineage exclusion (LIN)

Table S3: Microarray results

Probe Set	Gene Symbol	Entrez Gene	Fold Change in LIN⁻CD24ʰⁱCD29ˡᵒ subpopulation[a]	Fold Change in LIN⁻CD24⁺CD29ʰⁱ subpopulation[a]	P-value (ANOVA)
1430014_at	5430419D17Rik	71395	+ 9.64		1.6E-04
1449542_at	Pbx-1	18514	+ 3.30		5.1E-04
1455188_at	Ephb1	270190	+ 2.49		5.9E-04
1422667_at	Krt15	16665	+ 2.12		3.5E-05
1434964_at	X83328	13929	+ 1.89		6.3E-04
1420960_at	Fancg	60534	+ 1.86		1.4E-04
1424282_at	Pet112l	229487	+ 1.82		8.7E-04
1437696_at	BC049807	381066	+ 1.82		2.3E-04
1435085_at	Crebl2	232430	+ 1.74		1.7E-04
1460347_at	Krt14	16664	+ 1.73		1.9E-04
1417787_at	Dkkl1	50722	+ 1.70		1.3E-04
1451844_at	Prlr	19116	+ 1.65		5.8E-04
1440330_at	Hist1h2be	319179	+ 1.62		2.4E-04
1424096_at	Krt5	110308	+ 1.50		2.6E-05
1450928_at	Id4	15904	+ 1.48		7.7E-04
1441206_at	Synpo2	118449	+ 1.47		8.7E-04
1430651_s_at	Zfp191	59057	+ 1.46		1.3E-04
1448897_at	Mkrn2	67027	+ 1.43		4.7E-04
1423935_x_at	Krt14	16664	+ 1.42		4.9E-04
1419671_a_at	Il17rc	171095	- 5.71		6.8E-04

1433972_at	Camta1	100072	- 5.40	2.5E-04
1447608_x_at	Btbd14a	67991	- 3.40	6.2E-04
1453055_at	Sema6d	214968	- 3.25	1.3E-04
1423505_at	Tagln	21345	- 3.19	1.5E-04
1418569_at	Fblim1	74202	- 3.05	4.9E-05
1416882_at	Rgs10	67865	- 2.80	6.3E-04
1425505_at	Mylk	107589	- 2.40	3.9E-04
1429206_at	Rhobtb1	69288	- 2.25	6.6E-04
1454632_at	6330442E10Rik	268567	- 2.15	7.1E-04
1448552_s_at	2310028N02Rik	66950	- 2.13	8.8E-04
1425826_a_at	Sorbs1	20411	- 2.06	3.2E-04
1434936_at	Hirip3	233876	- 2.01	1.9E-04
1426887_at	Nudt11	58242	- 2.00	7.7E-04
1426677_at	Flna	192176	- 1.86	6.8E-04
1423587_a_at	Exosc10	50912	- 1.85	7.7E-04
1417022_at	Slc7a3	11989	- 1.82	2.1E-05
1416454_s_at	Acta2	11475	- 1.80	7.7E-06
1417329_at	Slc23a2	54338	- 1.78	4.9E-04
1449085_at	Phf10	72057	- 1.73	1.7E-04
1417984_at	Ube2v2	70620	- 1.62	6.6E-04
1428471_at	Sorbs1	20411	- 1.49	3.8E-04
1427476_a_at	Trim32	69807	- 1.50	2.5E-04
1431375_s_at	Parva	57342	- 1.48	1.1E-04
1437194_x_at	1200011O22Rik	71735	- 1.45	6.3E-04

1458963_at	1810054D07Rik	69863	- 1.43		7.5E-04
1436069_at	Ing5	66262	- 1.43		8.5E-04
1429761_at	Rtn1	104001	+ 4.33	+ 1.95	1.7E-04
1452426_x_at	Zfp236	329002	+ 2.73	+ 2.16	1.5E-04
1452508_x_at	Ptms	69202	+ 2.26	+ 2.04	1.8E-04
1458813_at	Scn5a	20271	+ 2.17	+ 2.72	2.9E-04
1424976_at	Rhov	228543	+ 2.10	+ 1.55	1.4E-04
1455404_at	Jph2	59091	+ 2.02	+ 1.41	7.8E-06
1426296_at	Rad52	19365	+ 1.90	+ 1.71	1.4E-04
1460431_at	Gcnt1	14537	+ 1.83	+ 3.33	8.7E-04
1438211_s_at	Dbp	13170	+ 1.63	+ 2.44	1.0E-04
1452367_at	Coro2a	107684	+ 1.61	+ 1.40	3.7E-04
1432919_at	4921511E18Rik	70879	+ 1.59	+ 2.63	9.2E-04
1418174_at	Dbp	13170	+ 1.57	+ 1.93	1.3E-04
1426230_at	Sphk2	56632	+ 1.51	+ 1.90	1.3E-05
1448297_a_at	Tnk2 /// LOC672286	51789 /// 672286	+ 1.49	+ 1.61	8.7E-05
1422678_at	Dgat2	67800	+ 1.49	+ 1.39	2.8E-04
1417812_a_at	Lamb3	16780	+ 1.49	+ 1.27	7.6E-06
1453371_at	4930535B03Rik	75137	+ 1.46	+ 1.56	7.9E-04
1424175_at	Tef	21685	+ 1.39	+ 1.72	6.2E-04
1455678_at	Sema4b	20352	+ 1.38	+ 1.49	5.2E-04
1428067_at	Rasl12	70784	+ 1.38	+ 1.73	3.5E-04
1422869_at	Mertk	17289	+ 1.35	+ 1.46	1.8E-04

1446856_at	D3Wsu167e	28044	+ 1.35	+ 1.53	4.7E-04
1418412_at	Tpd52l1	21987	+ 1.34	+ 1.58	2.6E-04
1418749_at	Psd3	234353	+ 1.32	+ 1.77	1.8E-05
1434548_at	Serinc3	26943	- 4.83	- 3.59	6.8E-05
1428875_at	Golph4	73124	- 2.70	- 1.65	4.3E-04
1449047_at	Hacl1	56794	- 2.45	- 3.02	5.5E-04
1419549_at	Arg1	11846	- 2.40	- 3.97	3.6E-04
1422643_at	Moxd1	59012	- 1.92	- 1.85	4.3E-05
1457273_at	LOC669490	669490	- 1.86	- 2.20	1.5E-04
1455117_at	Mcmdc1	71567	- 1.69	- 2.95	6.9E-04
1449184_at	Pglyrp1	21946	- 1.63	- 2.09	8.7E-04
1420543_at	ORF28	246738	- 1.63	- 2.79	9.2E-04
1458268_s_at	Igfbp3	16009	- 1.62	- 1.71	1.8E-04
1449329_at	Zfp235	56525	- 1.57	- 1.24	5.9E-04
1424692_at	2810055F11Rik	67217	- 1.55	- 1.26	3.3E-04
1417580_s_at	Selenbp1	20341	- 1.54	- 1.17	7.8E-04
1448881_at	Hp	15439	- 1.44	- 1.86	4.2E-04
1436600_at	Tnrc9	244579	- 1.44	- 1.43	2.1E-04
1415822_at	Scd2	20250	- 1.40	- 1.22	9.6E-04
1428117_x_at	Tmem181	77106	- 1.37	- 1.60	5.7E-04
1424035_at	Rora	19883	- 1.36	- 1.79	8.5E-04
1434606_at	Erbb3	13867	- 1.34	- 1.77	1.1E-05
1421812_at	Tapbp	21356	- 1.31	- 1.44	6.9E-04
1415882_at	Ghitm	66092	- 1.31	- 1.47	8.1E-04

1419739_at	Tpm2	22004	+ 2.98	- 1.50	7.3E-04
1424603_at	Sumf1	58911	+ 2.23	- 1.76	5.9E-05
1421761_a_at	Barx2	12023	+ 1.94	- 2.56	4.2E-04
1458496_at	Cyb561	13056	+ 1.69	- 1.43	8.3E-06
1428378_at	Zc3hav1	78781	+ 1.58	- 1.28	3.8E-04
1422092_at	Pfkfb2	18640	+ 1.57	- 2.12	7.0E-04
1444500_at	Ahsa1	217737	+ 1.51	- 1.20	4.1E-04
1427527_a_at	Pthlh	19227	+ 1.48	- 1.94	8.0E-04
1452027_a_at	Trp63	22061	+ 1.46	- 1.59	5.0E-04
1436236_x_at	Cotl1	72042	+ 1.45	- 1.82	7.4E-04
1441407_at	Rod1	230257	+ 1.39	- 2.17	9.5E-04
1423323_at	Tacstd2	56753	+ 1.38	- 1.46	9.9E-04
1436556_at	Tmem139	109218	+ 1.34	- 1.43	5.8E-04
1459733_at	Pea15	18611	+ 1.31	- 1.47	7.5E-04
1426859_at	Inhbb	16324	+ 1.29	- 1.71	6.2E-04
1454254_s_at	1600029D21Rik	76509	+ 1.27	- 1.44	8.4E-04
1425163_at	AI661453	224833	+ 1.25	- 1.56	2.4E-04
1435576_at	AW413774	106046	+ 1.22	- 1.60	8.9E-04
1441464_at	A630035D09Rik	319627	+ 1.17	- 1.45	2.2E-05
1448946_at	Kif3c	16570	- 1.20	+ 1.51	6.7E-05
1455420_at	Rad23b	19359	- 1.20	+ 1.49	9.7E-04
1430521_s_at	Cpne8	66871	- 1.37	+ 1.88	5.8E-04
1424470_a_at	Rapgef3	223864	- 1.37	+ 1.64	5.7E-04

1452768_at	Tex261	21766	- 1.49	+ 1.26	7.3E-04
1455007_s_at	Gpt2	108682	- 1.55	+ 1.32	4.9E-04
1434542_at	Gpt2	108682	- 1.87	+ 1.35	4.9E-04
1424133_at	Tmem98	103743	- 1.91	+1.28	6.0E-04
1427320_at	Copg2as2	54158	- 2.48	+ 2.25	1.5E-05
1454752_at	LOC380843 /// LOC638266 /// Rbm24	380843 ///638266 ///666794	- 2.97	+ 1.92	9.0E-04
1423222_at	Cap2	67252	- 4.53	+ 1.80	2.0E-04
1419906_at	Hpgd	15446		+ 7.52	9.4E-04
1421727_at	Eya1	14048		+ 3.52	1.4E-05
1420650_at	Atbf1	11906		+ 3.30	6.4E-04
1449106_at	Gpx3	14778		+ 2.76	3.5E-04
1428434_at	Zcchc12	72693		+ 2.66	1.1E-04
1424719_a_at	Mapt	17762		+ 2.65	3.4E-04
1437800_at	Edaradd	171211		+ 2.19	5.3E-04
1453267_at	Atbf1	11906		+ 2.03	3.8E-04
1420351_at	Tnfrsf4	22163		+ 1.90	1.2E-04
1427176_s_at	AI428936	233066		+ 1.78	8.3E-04
1453264_at	Marveld3	73608		+ 1.75	2.4E-04
1424030_at	Grhl1	195733		+ 1.65	5.6E-04
1434044_at	Repin1	58887		+ 1.63	3.4E-04
1436844_at	AW046287	107018		+ 1.59	4.6E-04
1451321_a_at	0610033I05Rik	71684		+ 1.55	6.0E-05

1416125_at	Fkbp5	14229	+ 1.53	5.0E-04
1420138_at	Slc19a1	20509	+ 1.50	2.9E-04
1458707_at	E430016F16Rik	414121	+ 1.43	4.9E-04
1419829_a_at	Gab2	14389	+ 1.43	4.9E-04
1448870_at	Ltbp1	268977	+ 1.40	4.8E-05
1451206_s_at	Pscdbp	227929	- 5.00	7.3E-04
1450395_at	Slc22a5	20520	- 4.24	3.7E-04
1441102_at	Prlr	19116	- 3.21	6.5E-05
1455355_at	6030408C04Rik	217558	- 3.10	3.4E-04
1420718_at	Odz2 ///	23964 ///	- 3.01	5.4E-05
	LOC669490	669490		
1443602_at	Ifngr1	15979	- 2.92	6.3E-04
1456428_at	Cxcl15	20309	- 2.62	3.6E-04
1417089_a_at	Ckmt1	12716	- 1.06	1.6E-04
1424404_at	0610040J01Rik	76261	- 2.48	1.2E-04
1418294_at	Epb4.1l4b	54357	- 2.35	1.3E-04
1427477_at	Tmprss13	214531	- 2.25	2.4E-04
1423952_a_at	Krt7	110310	- 2.06	6.6E-04
1455030_at	Ptprj	19271	- 2.06	4.6E-04
1424351_at	Wfdc2	67701	- 1.92	4.6E-04
1422418_s_at	Supt4h1 ///	20922 ///	- 1.88	2.8E-04
	Supt4h2	20923		
1416389_a_at	Rcbtb2	105670	- 1.85	2.0E-04
1421965_s_at	Notch3	18131	- 1.83	3.3E-04

1417214_at	Rab27b	80718		- 1.79	4.8E-04
1426098_a_at	Cast	12380		- 1.74	3.5E-04
1452670_at	Myl9	98932		- 1.72	9.4E-04
1453392_at	1810054D07Rik	69863		- 1.71	5.7E-04
1417185_at	Ly6a	110454		- 1.67	4.9E-04
1416061_at	Tbc1d15	66687		- 1.67	6.7E-04
1450955_s_at	Sort1	20661		- 1.67	4.1E-07
1448873_at	Ocln	18260		- 1.66	7.2E-04
1424604_s_at	Sumf1	58911		- 1.65	3.9E-04
1436325_at	Rora	19883		- 1.63	5.7E-04
1417896_at	Tjp3	27375		- 1.58	5.6E-04
1460737_at	Igbp1	18518		- 1.57	3.9E-04
1428701_at	Pvrl4	71740		- 1.55	5.2E-05
1436842_at	B230380D07Rik	235461		- 1.54	2.8E-05
1417616_at	St6galnac2	20446		- 1.53	2.4E-04
1423062_at	Igfbp3	16009		- 1.50	2.0E-04
1452391_at	Cxadr	13052		- 1.48	5.9E-04
1426301_at	Alcam	11658		- 1.43	5.2E-04

[a] Mean of the mRNA fold change in the C/EBPβ$^{-/-}$ subpopulations as compared to wildtype.