Repression of the Type I Interferon pathway underlies MYC & KRAS-dependent evasion of NK & B cells in Pancreatic Ductal Adenocarcinoma

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Summary of Supplementary Materials:

Supplementary Figures 1-6

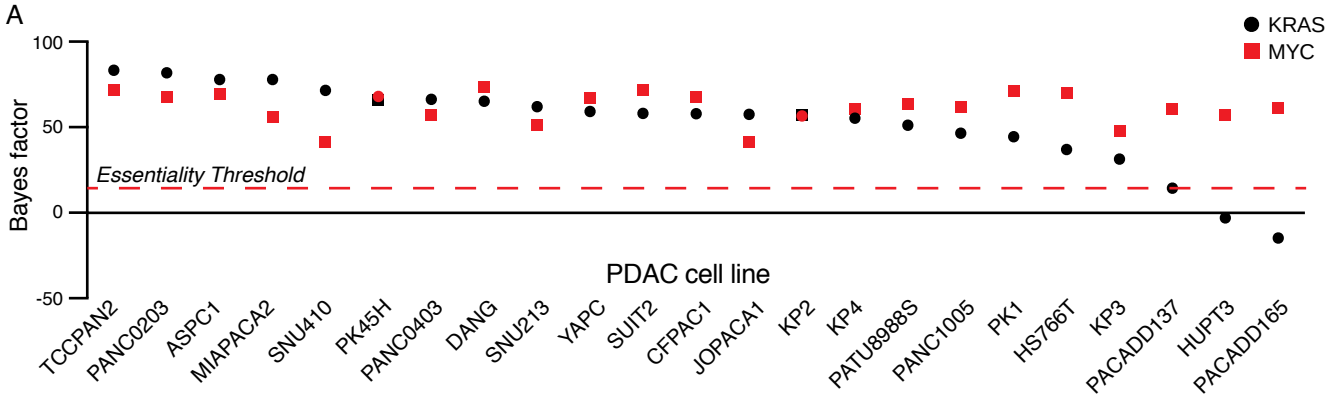
Supplementary Tables 1-3

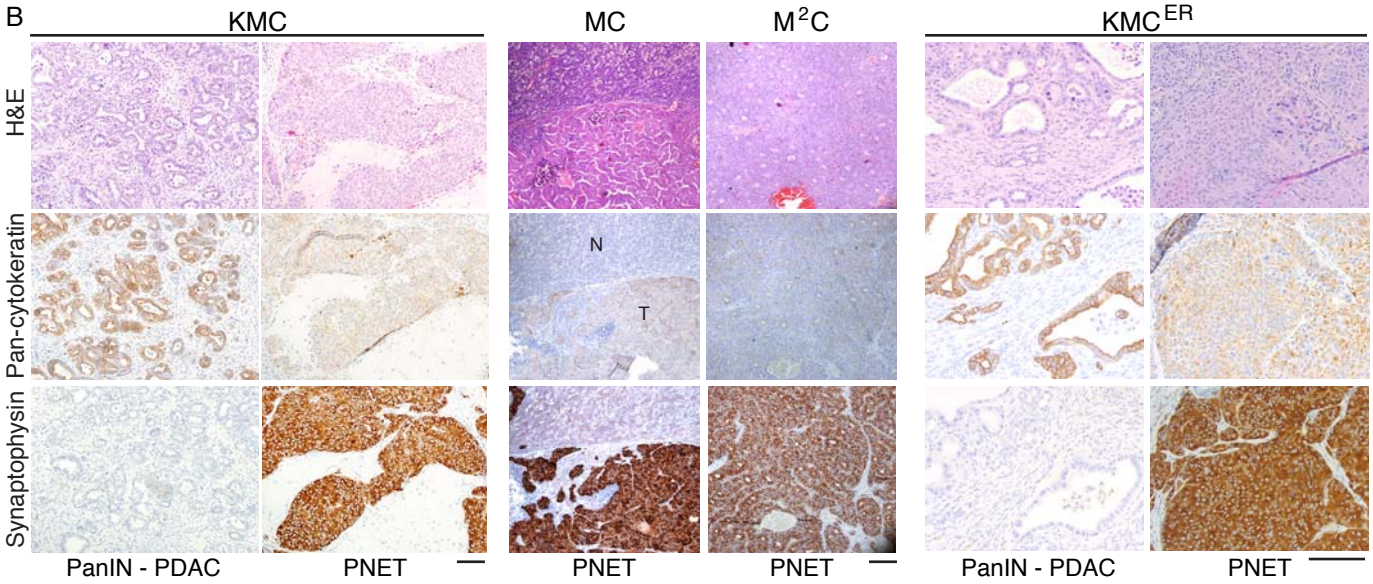
Supplementary Table S4 (provided as a separate Excel file)

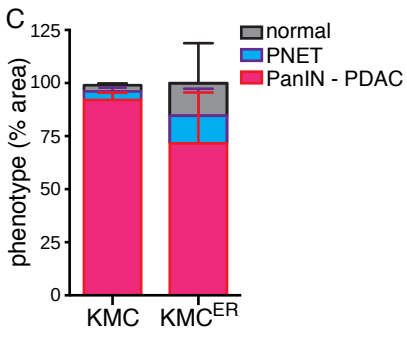
Detailed RNA-SEQ Materials & Methods

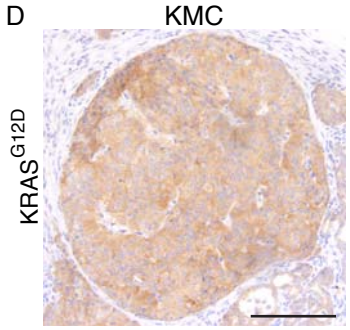
Supplementary References

Figure S1


A
KRAS
MYC
Bayes factor
100
50
0
-50
Essentiality Threshold
PDAC cell line
TCCPAN2
PANC0203
ASPC1
MIAPACA2
SNU410
PK45H
PANC0403
DANG
SNU213
YAPC
SUIT2
CFPAC1
JOPACA1
KP2
KP4
PATU8988S
PANC1005
PK1
HS766T
KP3
PACADD137
HUPT3
PACADD165


B
KMC
MC
M2C
KMCER
H&E
Pan-cytokeratin
Synaptophysin
N
T
PanIN - PDAC
PNET
PNET
PNET
PanIN - PDAC
PNET


C
normal
PNET
PanIN - PDAC
phenotype (% area)
125
100
75
50
25
0
KMC
KMCER


D
KMC
KRASG12D

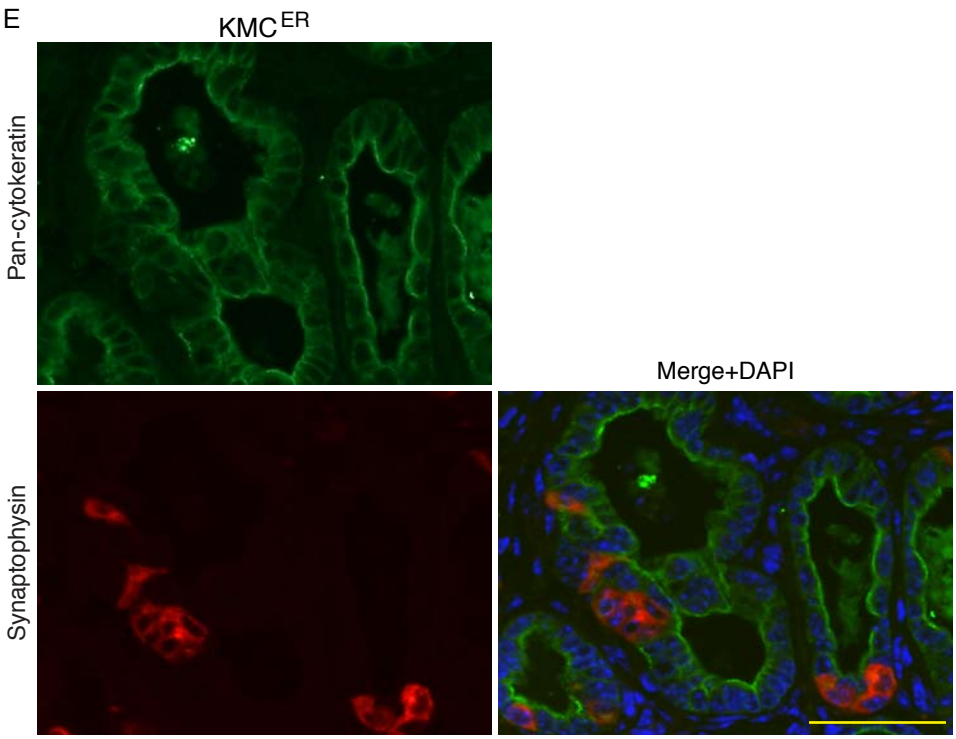

E
KMCER
Pan-cytokeratin
Merge+DAPI
Synaptophysin

Figure S1: Characterization of MYC-driven pancreatic tumour phenotypes

A) Meta-analysis of CRISPR deletion of *KRAS* or *c-MYC* in human PDAC cell lines using the Avana 2018 gRNA library, generated using the PICKLES database: https://hartlab.shinyapps.io/pickles/. A Bayes factor >5 indicates essentiality of each gene for the indicated cell line. **B)** Histological characterization of tumours in mice of the indicated genotypes. For KMC and KMC^{ER}, representative images of both ductal epithelial lesions and neuroendocrine tumour areas are shown. N = normal pancreas; T = tumour. Scale bars = 100μm. **C)** Quantification of tumour area showing features of PanIN-PDAC versus PNET or normal tissue in KMC (N=11) and KMC^{ER} (N=10) mice. **D)** IHC staining with $KRas^{G12D}$-specific antibody shows expression of mutant KRAS in an area of PNET. Scale bar = 1000 μM. **E)** Immunofluorescent detection of cytokeratin and synaptophysin double-positive cells in KMC^{ER} tumour tissue. Scale bar = 50 μM.

Figure S2

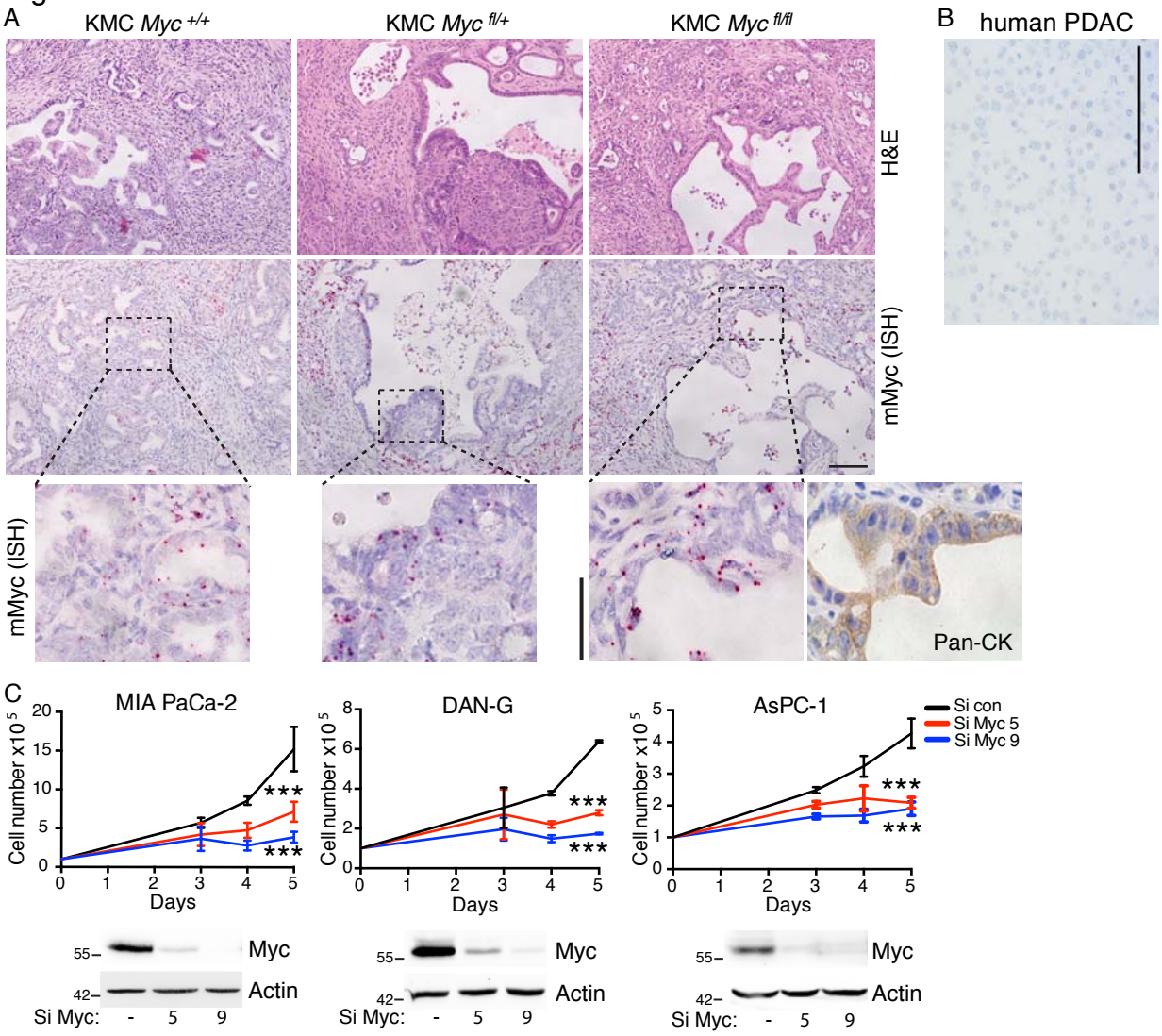

A
KMC Myc +/+
KMC Myc fl/+
KMC Myc fl/fl
H&E
mMyc (ISH)
mMyc (ISH)
B
human PDAC
Pan-CK
C
MIA PaCa-2
DAN-G
AsPC-1
Cell number x10 5
Days
Si con
Si Myc 5
Si Myc 9
***
Myc
Actin
55–
42–
Si Myc: - 5 9

Figure S2: Expression of endogenous c-Myc is required for KMC PDAC development

A) *In situ* hybridization for murine *c-Myc* in PDAC from KMC, KMC-*cMyc*$^{fl/+}$ and KMC-*cMyc*$^{fl/fl}$ mice (red stain). Note the retention of expression in ductal epithelium of KMC-*cMyc*$^{fl/fl}$ tumours. Scale bars = 100μm (main panels) and 50μm (zoomed insets). **B)** ISH staining of a section of human PDAC using the same murine *c-Myc*-specific probe as in (A). Scale bar = 100μm. **C)** Cell propagation measurement upon depletion of MYC in 3 human PDAC cells lines using either of 2 siRNAs. N=3 independent experiments. *** denotes $p<0.001$ (ANOVA, Dunnett's multiple comparisons test). Lower panels: Immunoblots confirm MYC depletion.

Figure S3

A

Most significantly downregulated pathways, KMC v KC

#	Maps	0 | 2 | 4 | 6 | 8 -log(pValue)	pValue	FDR
1	Immune response_Role of DPP4 (CD26) in immune regulation		7.997e-10	9.263e-7
2	CHDI_Correlations from Replication data_Causal network (positive correlations)		1.998e-9	9.263e-7
3	Role of CD8+ Tc1 cells in COPD		2.021e-9	9.263e-7
4	Immune response_IFN-gamma in macrophages activation		2.589e-9	9.263e-7
5	T follicular helper cell dysfunction in SLE		3.101e-8	7.776e-6
6	Immune response_IFN-alpha/beta signaling via JAK/STAT		3.572e-8	7.776e-6
7	Immune response_T cell subsets: cell surface markers		3.804e-8	7.776e-6
8	SLE genetic marker-specific pathways in T cells		7.114e-8	1.272e-5
9	Immune response_NF-AT in immune response		1.834e-7	2.916e-5
10	Aberrant production of IL-2 and IL-17 in SLE T cells		2.303e-7	2.996e-5
11	Immune response_TCR alpha/beta signaling pathway		2.303e-7	2.996e-5
12	Inflammatory mechanisms of pancreatic cancerogenesis		4.351e-7	4.753e-5
13	B-regulatory cells and tumor cells intercellular interaction		4.351e-7	4.753e-5
14	Apoptosis and survival_Anti-apoptotic TNFs/NF-kB/Bcl-2 pathway		4.650e-7	4.753e-5
15	Immune response_T cell co-signaling receptors, schema		5.843e-7	5.574e-5

Figure S3: MYC levels modulate the immune landscape in PDAC

Metacore GeneGO analysis of RNA SEQ data from KC (N=6) versus KMC (N=6) tumours, showing the top 15 most significantly downregulated pathways in KMC, relative to KC PDAC tumours. FDR = false discovery rate. Pathway annotations current as of January 2020.

Figure S4

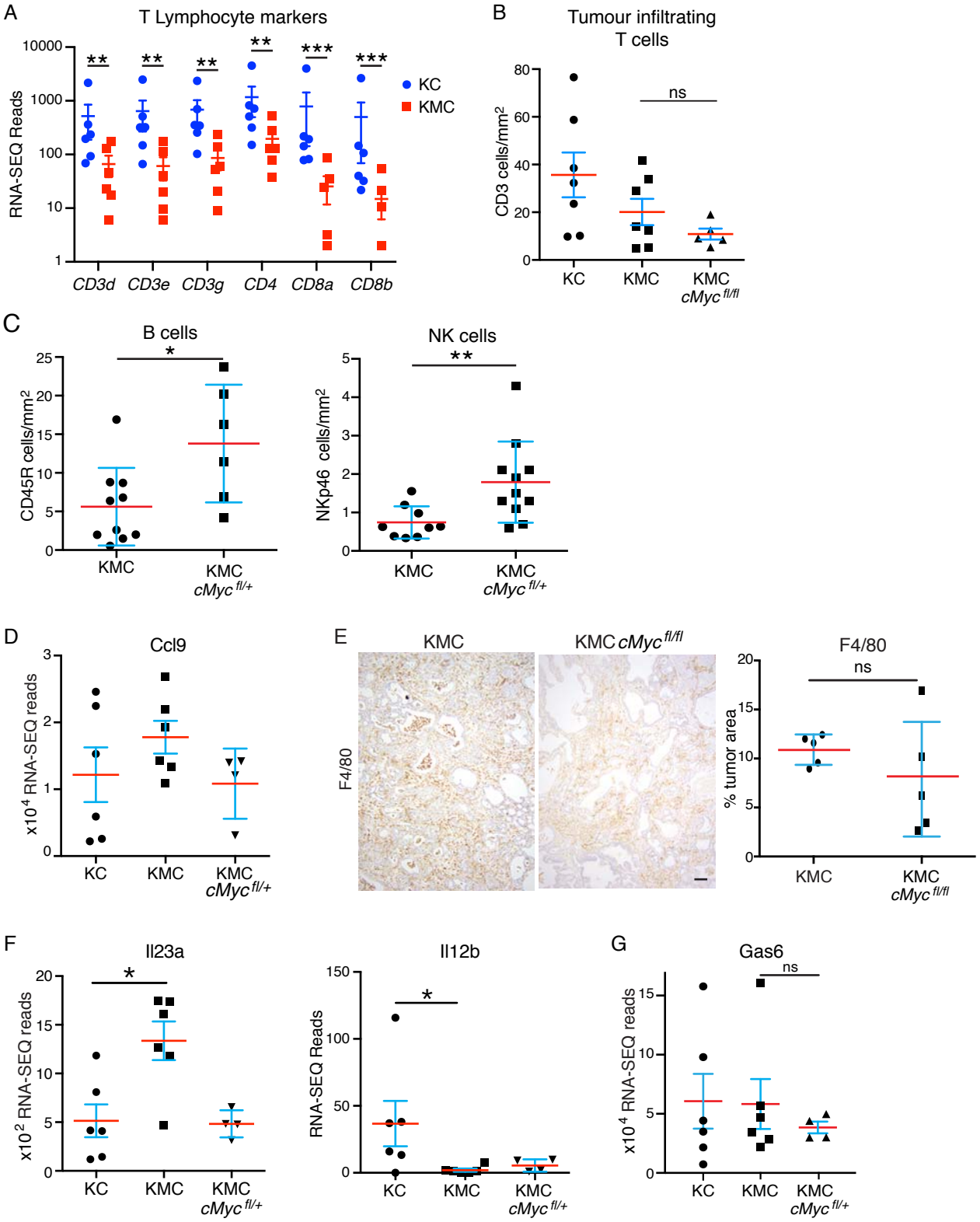
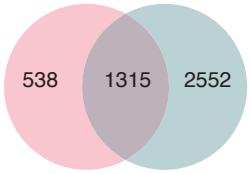

A
T Lymphocyte markers
RNA-SEQ Reads
KC
KMC
CD3d
CD3e
CD3g
CD4
CD8a
CD8b
B
Tumour infiltrating T cells
CD3 cells/mm2
ns
KC
KMC
KMC cMyc fl/fl
C
B cells
CD45R cells/mm2
KMC
KMC cMyc fl/+
NK cells
NKp46 cells/mm2
KMC
KMC cMyc fl/+
D
Ccl9
x10 4 RNA-SEQ reads
KC
KMC
KMC cMyc fl/+
E
KMC
KMC cMyc fl/fl
F4/80
F4/80
ns
% tumor area
KMC
KMC cMyc fl/fl
F
Il23a
x10 2 RNA-SEQ reads
KC
KMC
KMC cMyc fl/+
Il12b
RNA-SEQ Reads
KC
KMC
KMC cMyc fl/+
G
Gas6
ns
x10 4 RNA-SEQ reads
KC
KMC
KMC cMyc fl/+

Figure S4: Previously described mechanisms fail to account for long-term effects of MYC on the immune landscape of PDAC

A) RNA-SEQ analysis of T lymphocyte gene expression in KC (N=6) versus KMC tumours (N=6). Mean and SEM values shown (panels A-G). Note that samples with zero reads are omitted from log scale graphics (pertains only to KMC samples). **B)** IHC quantification of CD3-positive T cell tumour infiltration in KC (N=7), KMC (N=7) and KMC-*Myc*$^{fl/fl}$ (N=5) PDAC; ns = not significant, Kruskal-Wallis and Dunn's multiple comparison test. **C)** IHC quantification of and CD45R-positive B and NKp46-positive NK cell tumour infiltration in KMC versus KMC-*Myc*$^{fl/+}$ PDAC. * denotes P<0.05, ** denotes P<0.01. Mann-Whitney test. **D)** RNA-SEQ analysis of *Ccl9* expression in KC (N=6), KMC (N=6) and KMC-*Myc*$^{fl/+}$ (N=4) PDAC. **E)** IHC quantification of F4/80 positive macrophage infiltration, measured as % tumour area, in KMC (N=5) versus KMC-*Myc*$^{fl/fl}$ (N=5) PDAC. Scale bar = 100µm; ns = not significant, Mann-Whitney test. **F)** RNA-SEQ analysis of IL23 subunit expression in KC (N=6), KMC (N=6) and KMC-*Myc*$^{fl/+}$ (N=4) PDAC. * denotes P<0.05. ANOVA and post-hoc Tukey test. **G)** RNA-SEQ analysis of Gas6 expression in KC (N=6), KMC (N=6) and KMC-*Myc*$^{fl/+}$ (N=4) PDAC. ANOVA and post-hoc Tukey test; ns = not significant.

Figure S5

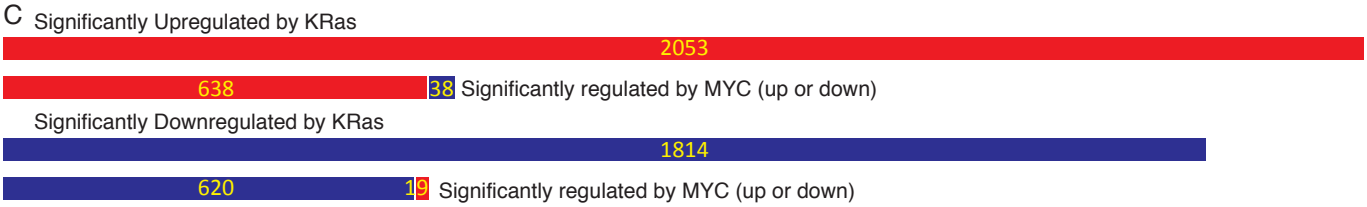
A



all significantly regulated genes

B

C



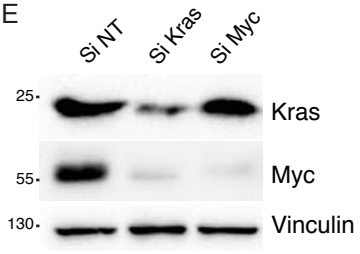
D

Most significantly downregulated pathways, acute activation of MYC, KRas or both in MEFs

#	Maps	0 2 4 6 8 -log(pValue)	pValue	min(pValue)	FDR
1	Immune response_IFN-alpha/beta signaling via JAK/STAT		3.272e-11 2.061e-8 3.207e-2	3.272e-11	4.646e-8 2.756e-5 3.366e-1
2	Immune response_Antigen presentation by MHC class I: cross-presentation		1.154e-10 7.553e-4 3.771e-6	1.154e-10	8.191e-8 5.315e-2 4.748e-3
3	Ubiquinone metabolism		3.393e-8 4.291e-3 5.275e-1	3.393e-8	1.414e-5 1.282e-1 7.773e-1
4	Immune response_Induction of the antigen presentation machinery by IFN-gamma		3.983e-8 1.015e-5 2.244e-5	3.983e-8	1.414e-5 4.525e-3 1.413e-2
5	Immune response_Antigen presentation by MHC class I, classical pathway		3.231e-7 6.382e-5 1.429e-1	3.231e-7	8.351e-5 1.422e-2 4.996e-1
6	Immune response_IFN-alpha/beta signaling via MAPKs		3.529e-7 1.639e-4 7.954e-3	3.529e-7	8.351e-5 3.130e-2 2.173e-1

lsl-KRas^{G12D}+ Rosa26-lsl-MYC lsl-KRas^{G12D} Rosa26-lsl-MYC

E



F

Most significantly upregulated pathways upon KRAS or MYC depletion in KMC PDAC cells

#	Maps	0 2.5 5 7.5 10 12.5 15 17.5 20 -log(pValue)	pValue	min(pValue)	FDR
1	Cytoskeleton remodeling_Regulation of actin cytoskeleton organization by the kinase effectors of Rho GTPases		1.931e-12 3.230e-24	3.230e-24	4.067e-10 4.700e-21
2	Chemotaxis_Lysophosphatidic acid signaling via GPCRs		1.941e-13 6.642e-22	6.642e-22	9.505e-11 4.832e-19
3	Neurophysiological process_Receptor-mediated axon growth repulsion		3.904e-12 1.145e-17	1.145e-17	7.168e-10 5.552e-15
4	Apoptosis and survival_NGF/ TrkA PI3K-mediated signaling		2.501e-9 1.614e-17	1.614e-17	1.361e-7 5.871e-15
5	Immune response_IFN-alpha/beta signaling via MAPKs		3.193e-16 1.298e-9	3.193e-16	4.691e-13 5.105e-8
6	Neurophysiological process_Dynein-dynactin motor complex in axonal transport in neurons		4.027e-7 8.841e-16	8.841e-16	6.800e-6 2.573e-13
7	Immune response_IFN-alpha/beta signaling via JAK/STAT		2.854e-15 4.451e-9	2.854e-15	2.097e-12 1.295e-7

siMYC siKRas

Figure S5: Coordinate regulation of gene expression by KRAS and MYC in PDAC & MEFs

A) Summary of RNA-SEQ analysis showing overlap in significantly altered gene expression (irrespective of direction or magnitude change), relative to control infected MEFs, upon acute CRE-mediated activation of *lsl-KRas*^{G12D} and *Rosa26*$^{DM\text{-}lsl\text{-}MYC}$ alleles. Numbers indicate the number of significantly regulated genes. **B)** Co-directional gene regulation of MYC-regulated genes upon activation of KRas^{G12D}, as per (A). Numbers indicate the number of significantly regulated genes. **C)** Co-directional regulation of KRas^{G12D}-regulated genes upon expression of Rosa26-driven MYC, as per (B). **D)** Metacore GeneGO analysis of the most significantly down-regulated pathways upon activation of *lsl-KRas*^{G12D}, *Rosa26*$^{DM\text{-}lsl\text{-}MYC}$ or both in MEFs. Coloured bars indicate p values (-log). **E)** Immunblots showing depletion of MYC and KRAS in KMC PDAC tumour cells treated with the indicated siRNAs. **F)** Metacore GeneGO analysis of RNA-SEQ data from KMC PDAC tumour cells upon siRNA-mediated depletion of MYC (N=4) or KRas (N=4) *in vitro*. MYC was depleted using pooled siRNA targeting both human and murine *c-MYC*.

Figure S6

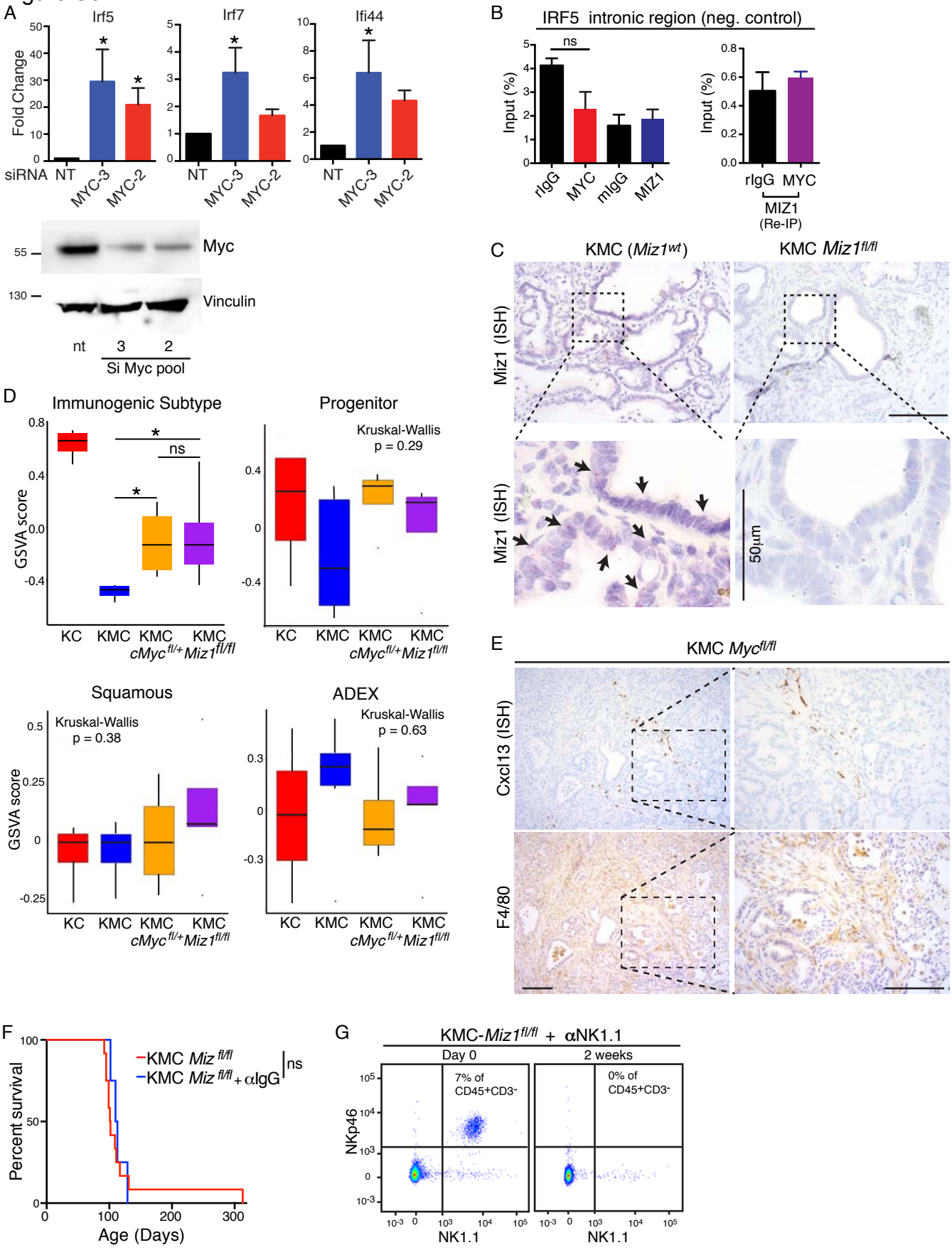

A
Irf5
Irf7
Ifi44
Fold Change
siRNA NT MYC-3 MYC-2
Myc
Vinculin
55
130
nt 3 2
Si Myc pool
B
IRF5 intronic region (neg. control)
Input (%)
ns
rIgG MYC mIgG MIZ1
rIgG MYC
MIZ1
(Re-IP)
C
KMC (Miz1wt)
KMC Miz1fl/fl
Miz1 (ISH)
50µm
D
Immunogenic Subtype
Progenitor
Squamous
ADEX
GSVA score
Kruskal-Wallis
p = 0.29
p = 0.38
p = 0.63
KC KMC KMC KMC
cMycfl/+Miz1fl/fl
E
KMC Mycfl/fl
Cxcl13 (ISH)
F4/80
F
Percent survival
KMC Miz fl/fl
KMC Miz fl/fl + αIgG
ns
Age (Days)
G
KMC-Miz1fl/fl + αNK1.1
Day 0
2 weeks
7% of CD45+CD3-
0% of CD45+CD3-
NKp46
NK1.1

Figure S6: The role of Miz1 in MYC-dependent suppression of the immune landscape

A) Q-PCR analysis of expression of indicated genes in KMC PDAC cells upon depletion of MYC (human & murine) with either of 2 pools of siRNA. Mean and SEM of 3 independent experiment shown. * denotes P<0.05, ANOVA and Fisher's LSD test. Immunoblot (right panel) confirms MYC depletion. **B)** Chromatin immunoprecipitation (ChIP) and ChIP-re-ChIP analysis of MYC and MIZ1 binding to an intronic region of the human IRF5 locus in DAN-G cells. Mean and SEM of technical replicates from 1 of 3 independent experiments shown; ns= not significant, ANOVA and Tukey's multiple comparison test. **C)** ISH analysis showing effective deletion of *Miz1* in KMC-*Miz1*$^{fl/fl}$ PDAC ductal epithelium. **D)** PDAC subtype analysis performed as per Bailey et al. (1) showing the impact of *c-Myc* or *Miz1* deletion in KMC tumours upon the murine equivalent of the human "immunogenic" subtype but none of the other three subtype gene signatures. Data derived from RNA-SEQ analysis of tumours from the indicated genotypes. * denotes P<0.05, Kruskal-Wallis and Dunn's multiple comparison test. **E)** Expression of *Cxcl13* detected in KMC-*Myc*$^{fl/fl}$ PDAC tumours by ISH. Lower panels show F4/80 IHC. Scale bars = 50μm. **F)** Survival analysis upon treatment of KMC-*Miz1*$^{fl/fl}$ tumour-bearing mice with Goat IgG isotype control for anti-Cxcl13 (N=4) versus no treatment (N=12). **G)** Representative FACS analysis of whole blood from KMC *Miz1*$^{fl/fl}$ mouse before or after 2 weeks treatment with NK1.1 depleting antibody. N = 3.

Muthalagu et al. Table 1: Significantly regulated by Rosa26-DM-MYC in MEFs			Rosa26-DM-MYC				KRasG12D		
gene_name	symbol	Mean WT	Mean Myc	FC	padj	Mean WT	Mean Kras	FC	padj
eukaryotic translation initiation factor 3, subunit J1	Eif3j1	64.1565504	945.188481	10.8716963	3.598E-112	64.1565504	60.1759299	-1.1443306	0.40091837
predicted gene 14432	Gm14432	1.24245536	123.98403	6.16533384	1.9113E-31	1.24245536	97.1798448	5.22077466	3.6592E-26
predicted gene 14305	Gm14305	1.24245536	123.98403	6.16533384	1.9113E-31	1.24245536	97.1798448	5.22077466	3.6592E-26
aggrecan	Acan	43.2824021	376.76798	5.2658549	5.1975E-48	43.2824021	436.760769	5.94672458	4.4571E-56
early growth response 1	Egr1	119.989019	729.317897	3.85840791	3.0658E-23	119.989019	397.382522	2.34765373	1.6979E-09
ribosomal protein L3	Rpl3	536.873861	2483.33373	3.79185428	2.2424E-34	536.873861	659.173956	1.18025614	0.24500707
BarH-like homeobox 1	Barx1	1.82270046	57.4601183	3.70709579	2.2197E-16	1.82270046	15.3932528	1.45319361	0.05272342
elastin	Eln	45.1426709	372.678443	3.59148383	7.5892E-16	45.1426709	130.867446	1.72471394	0.00218212
chemokine (C-X-C motif) ligand 12	Cxcl12	1184.25058	4386.28773	3.38163554	8.7207E-51	1184.25058	995.073229	-1.1822431	0.1063643
glucosaminyl (N-acetyl) transferase 1, core 2	Gcnt1	27.7489748	136.998363	3.23764255	4.0153E-18	27.7489748	44.2134855	1.25508137	0.19325662
S-adenosylhomocysteine hydrolase	Ahcy	154.157749	553.717283	3.09480144	1.5807E-27	154.157749	131.478128	-1.1737524	0.25124495
paired-like homeodomain transcription factor 1	Pitx1	57.4823739	229.02654	3.0843358	4.2328E-19	57.4823739	159.184687	2.19408866	1.6825E-09
predicted gene 14295	Gm14295	32.9264444	156.832298	2.95663638	1.0867E-12	32.9264444	151.158669	2.95008579	4.0681E-13
neuronal regeneration related protein	Nrep	155.534414	522.560415	2.78379788	8.1159E-20	155.534414	696.427153	3.58187666	1.0854E-31
cholesterol 25-hydroxylase	Ch25h	84.5404353	289.027589	2.56281105	1.7956E-11	84.5404353	57.9064128	-1.4071476	0.0378792
complement component 1, s subcomponent 1	C1s1	20.503079	88.0221809	2.49764992	5.228E-09	20.503079	36.5537735	1.30111996	0.15941118
gap junction protein, beta 2	Gjb2	466.955423	1234.05857	2.45019492	5.0056E-24	466.955423	1377.76121	2.69949858	4.5552E-30
solute carrier family 4 (anion exchanger), member 4	Slc4a4	416.774414	1065.45994	2.43012358	4.7564E-34	416.774414	738.968161	1.7047928	2.5064E-12
aryl hydrocarbon receptor nuclear translocator-like 2	Arntl2	29.5339372	105.703248	2.33810372	3.5958E-09	29.5339372	126.094713	2.71534529	2.5073E-13
ribosomal protein L35A	Rpl35a	262.829783	670.286858	2.26949274	1.1171E-14	262.829783	254.907265	-1.0399124	0.8063984
leucine rich repeat and Ig domain containing 1	Lingo1	48.1246734	138.946181	2.23817066	8.2454E-08	48.1246734	120.975807	1.94377644	9.5754E-06
testis expressed gene 11	Tex11	4.51576857	45.121904	2.20051265	1.4716E-05	4.51576857	25.6048323	1.59905966	0.01356919
uncoupling protein 2 (mitochondrial, proton carrier)	Ucp2	367.949283	904.553843	2.18734334	4.1476E-12	367.949283	943.493742	2.25492896	1.5434E-13
RNA binding protein, fox-1 homolog (C. elegans) 1	Rbfox1	48.1253524	116.083316	2.1603512	3.7038E-07	48.1253524	21.3792537	-1.7502863	0.00072943
chemokine (C-X-C motif) ligand 3	Cxcl3	8.98697059	44.1748296	2.15511234	3.4268E-06	8.98697059	97.4518957	4.12312019	1.1957E-22
collagen, type XIV, alpha 1	Col14a1	356.822992	892.645982	2.13239673	7.1291E-09	356.822992	422.44105	1.13258895	0.45366735
stathmin-like 2	Stmn2	471.56321	1016.58154	2.10452857	3.0332E-29	471.56321	687.522212	1.44121678	3.1912E-07
ATP-binding cassette, sub-family C (CFTR/MRP), member 9	Abcc9	6.00327074	30.650801	2.03165881	4.693E-05	6.00327074	2.21132711	-1.2135332	0.32620283
dehydrogenase/reductase (SDR family) member 9	Dhrs9	6914.53855	3354.34643	-2.0211656	5.7421E-44	6914.53855	7308.90491	1.05534366	0.43322324
phosphoglucomutase 2-like 1	Pgm2l1	910.103983	415.647965	-2.0382757	4.1851E-15	910.103983	459.163966	-1.8870619	1.4332E-12
killer cell lectin-like receptor subfamily G, member 1	Klrg1	46.1382124	15.3356311	-2.0467463	6.0546E-05	46.1382124	49.3708324	1.09736102	0.67235079
keratin 8	Krt8	49.2037077	15.9919905	-2.0677079	6.4585E-05	49.2037077	33.87599	-1.2570359	0.25766885
complement component 1, q subcomponent, alpha polypeptide	C1qa	96.0403408	30.5885461	-2.0909054	5.9834E-05	96.0403408	32.5811736	-1.9597613	0.00016737
cytochrome b-245, beta polypeptide	Cybb	58.2279072	14.8957366	-2.1035549	6.0558E-05	58.2279072	21.7643382	-1.7170517	0.00396098
keratin 18	Krt18	738.346417	331.654806	-2.1069968	2.6175E-14	738.346417	453.155335	-1.5577085	1.2553E-05
cytochrome P450, family 4, subfamily a, polypeptide 12B	Cyp4a12b	249.648925	105.317142	-2.1279639	5.5532E-12	249.648925	92.4436468	-2.3665844	4.752E-16
macrophage expressed gene 1	Mpeg1	142.382428	49.0587781	-2.1749934	5.8298E-07	142.382428	62.4450648	-1.841547	7.7255E-05
adhesion G protein-coupled receptor E1	Adgre1	48.4593625	12.9144933	-2.2020817	1.5083E-05	48.4593625	19.085029	-1.8045674	0.00131003
lysozyme 2	Lyz2	640.693301	263.986704	-2.2120653	6.9373E-13	640.693301	245.467467	-2.3190661	4.9062E-15
solute carrier family 35, member G2	Slc35g2	60.5033026	14.0595059	-2.2588671	6.1808E-06	60.5033026	13.8580472	-2.2474515	3.1288E-06
interferon induced transmembrane protein 1	Ifitm1	56.5303805	15.4004012	-2.2731793	2.6693E-06	56.5303805	38.3979449	-1.2349948	0.29193915
membrane-spanning 4-domains, subfamily A, member 6D	Ms4a6d	39.2208561	8.33826325	-2.2920645	4.9134E-06	39.2208561	14.4880989	-1.7390771	0.00314315
chemokine (C-C motif) ligand 9	Ccl9	157.866027	47.5159776	-2.3150343	2.8988E-07	157.866027	55.793623	-2.0830693	4.8946E-06
membrane-spanning 4-domains, subfamily A, member 7	Ms4a7	29.9055157	4.77486953	-2.3428454	1.2793E-06	29.9055157	7.89980619	-1.9708875	0.00013605
lipase, member O2	Lipo2	138.035551	47.1696106	-2.4353373	7.4298E-11	138.035551	65.734296	-1.8087044	1.6292E-05
vomeronasal 2, receptor 79	Vmn2r79	39.5989236	3.63851515	-2.5094738	6.4409E-08	39.5989236	8.33888486	-2.04784	3.6636E-05
ribosomal protein L34	Rpl34	1239.37663	217.545527	-2.5834357	7.7989E-08	1239.37663	174.965987	-2.8285385	8.606E-10
ribosomal protein L34, pseudogene 1	Rpl34-ps1	1239.37663	217.545527	-2.5834357	7.7989E-08	1239.37663	174.965987	-2.8285385	8.606E-10
predicted gene 4705	Gm4705	1239.37663	217.545527	-2.5834357	7.7989E-08	1239.37663	174.965987	-2.8285385	8.606E-10
complement receptor 2	Cr2	51.2716247	9.29053789	-2.7435041	4.8133E-09	51.2716247	13.4174428	-2.2339336	3.3442E-06
X-prolyl aminopeptidase (aminopeptidase P) 2, membrane-bound	Xpnpep2	74.384279	17.9084636	-2.7459391	1.0542E-09	74.384279	6.92768851	-4.2069541	1.2075E-19
cathepsin S	Ctss	108.592105	29.6013711	-2.7596857	2.5516E-12	108.592105	40.804823	-2.1524198	8.9845E-08
ribonuclease T2B	Rnaset2b	64.4920881	12.2418811	-2.8114219	1.296E-09	64.4920881	15.2920443	-2.5494434	2.426E-08
processing of precursor 4, ribonuclease P/MRP family, (S. cerevisiae)	Pop4	2438.43211	724.896465	-3.2081646	3.5388E-65	2438.43211	2035.86019	-1.1850538	0.03830884
OTU domain containing 7A	Otud7a	398.537016	101.550901	-3.3796888	3.2416E-39	398.537016	438.780185	1.12340111	0.29669589
ATPase, H+ transporting, lysosomal V0 subunit C	Atp6v0c	339.649275	68.728053	-3.6296803	1.6274E-24	339.649275	70.9875122	-3.7110289	5.3365E-26
ribosomal protein S18	Rps18	1824.50163	430.196317	-3.7769031	1.2814E-45	1824.50163	374.079424	-4.2349863	2.8546E-54
interferon-induced protein 44	Ifi44	454.625564	77.0224674	-4.1633693	2.8874E-24	454.625564	37.1164803	-6.8057643	2.2414E-43
family with sequence similarity 181, member B	Fam181b	140.393082	19.4097118	-4.2198874	7.3269E-22	140.393082	38.8164246	-2.6120159	1.5574E-10
tripartite motif-containing 30A	Trim30a	62.3710788	4.47305267	-4.3955581	2.228E-19	62.3710788	7.26441419	-3.5807343	1.317E-14
carbonic anhydrase 8	Car8	267.801302	44.3196309	-4.5777753	1.7173E-32	267.801302	37.9569594	-4.8094997	3.4467E-35
histocompatibility 2, T region locus 23	H2-T23	302.049111	37.0606673	-6.0641773	1.0389E-48	302.049111	39.1900652	-5.5851865	6.068E-46
ribosomal protein S2	Rps2	4790.88714	546.523211	-7.7903453	1.223E-162	4790.88714	534.763925	-7.8809918	3.646E-167
predicted gene 29376	Gm29376	157.641087	4.84987028	-8.5912582	1.9744E-48	157.641087	4.05053933	-8.9604317	1.4617E-50
histocompatibility 2, T region locus 22	H2-T22	1002.51961	78.4156284	-10.680361	1.381E-177	1002.51961	89.8157478	-9.6073631	4.656E-172
histocompatibility 2, T region locus 9	H2-T9	1002.51961	78.4156284	-10.680361	1.381E-177	1002.51961	89.8157478	-9.6073631	4.656E-172
tripartite motif-containing 30D	Trim30d	149.791915	0.89340609	-11.922393	5.2908E-56	149.791915	0	-12.750712	3.2846E-59
ribosomal protein L29	Rpl29	2104.72968	68.0201294	-17.094978	1.76E-128	2104.72968	54.7644598	-19.429614	2.63E-141
predicted pseudogene 8210	Gm8210	2104.72968	68.0201294	-17.094978	1.76E-128	2104.72968	54.7644598	-19.429614	2.63E-141
tripartite motif-containing 12A	Trim12a	207.539651	0.37681761	-19.698875	1.0291E-83	207.539651	0	-20.88215	3.5986E-87

Table S1: Significantly regulated by Rosa26$^{DM\text{-}MYC}$ in MEFs

Table shows RNA-SEQ reads for each gene significantly regulated by more than 2 fold upon activation of Rosa26$^{DM\text{-}lsl\text{-}MYC}$ allele for 24hrs in primary MEFs. Comparison is with identically treated passage-matched wild-type MEFs. FC = fold change, numbers >2 in red, <-2 in dark blue. Adjusted p values <0.05 shown in red. Also shown are RNA-SEQ reads for the same genes upon similar activation of KRas^{G12D}, compared with wt MEFs, using the same colour scheme, plus FC>1.5 in pink; <-1.5 in light blue, to highlight convergent and divergent gene expression.

Muthalagu et al. Table S2: Significantly regulated by KRasG12D in MEFs			KRasG12D				Rosa26-DM-MYC		
gene_name	symbol	Mean WT	Mean Kras	FC	padj	Mean WT	Mean Myc	FC	padj
eukaryotic translation initiation factor 2, subunit 3, structural gene Y-linkec	Eif2s3y	0.95216308	1998.55476	129.833744	5.664E-278	0.95216308	0	-1.4988728	0.06062855
ubiquitously transcribed tetratricopeptide repeat gene, Y chromosome	Uty	0	884.316627	67.1207876	3.626E-195	0	0	-1	1
lysine (K)-specific demethylase 5D	Kdm5d	0	662.304932	50.5755893	6.282E-165	0	0	-1	1
aggrecan	Acan	43.2824021	436.760769	5.94672458	4.4571E-56	43.2824021	376.76798	5.2658549	5.1975E-48
predicted gene 14432	Gm14432	1.24245536	97.1798448	5.22077466	3.6592E-26	1.24245536	123.98403	6.16533384	1.9113E-31
predicted gene 14305	Gm14305	1.24245536	97.1798448	5.22077466	3.6592E-26	1.24245536	123.98403	6.16533384	1.9113E-31
angiotensin II receptor, type 2	Agtr2	15.7793228	133.695559	4.56150761	2.9217E-27	15.7793228	38.2355497	1.59033617	0.01254336
matrix metallopeptidase 3	Mmp3	136.749548	667.348859	4.35665303	2.1302E-52	136.749548	97.2113552	-1.3470612	0.032642
integrin alpha 2	Itga2	403.501296	2391.97884	4.27609074	1.3578E-32	403.501296	781.743469	1.68247665	0.00032938
chemokine (C-X-C motif) ligand 3	Cxcl3	8.98697059	97.4518957	4.12312019	1.1957E-22	8.98697059	44.1748296	2.15511234	3.4268E-06
CAP-GLY domain containing linker protein family, member 4	Clip4	47.8791174	264.656114	3.96828509	1.05E-32	47.8791174	92.3223762	1.56649213	0.00243501
neuronal regeneration related protein	Nrep	155.534414	696.427153	3.58187666	1.0854E-31	155.534414	522.560415	2.78379788	8.1159E-20
matrix metallopeptidase 13	Mmp13	155.399526	613.931216	3.57228269	1.4617E-50	155.399526	72.2221062	-1.9663024	1.3468E-10
angiopoietin-like 7	Angptl7	16.4852509	114.20065	3.42064825	3.3096E-16	16.4852509	24.5246848	1.06183508	0.84438158
protein tyrosine phosphatase, non-receptor type 22 (lymphoid)	Ptpn22	18.5545258	99.0121901	3.34221124	8.6456E-16	18.5545258	13.3297182	-1.2176134	0.41396645
5' nucleotidase, ecto	Nt5e	364.391148	1201.91286	3.09341307	2.2774E-69	364.391148	561.656865	1.46623046	3.4366E-07
UDP glucuronosyltransferase 1 family, polypeptide A6A	Ugt1a6a	8.03480752	61.453962	3.04804551	1.8526E-11	8.03480752	6.46019745	-1.137722	0.63886701
UDP glucuronosyltransferase 1 family, polypeptide A7C	Ugt1a7c	8.03480752	61.453962	3.04804551	1.8526E-11	8.03480752	6.46019745	-1.137722	0.63886701
predicted gene 14295	Gm14295	32.9264444	151.158669	2.95008579	4.0681E-13	32.9264444	156.832298	2.95663638	1.0867E-12
aryl hydrocarbon receptor nuclear translocator-like 2	Arntl2	29.5339372	126.094713	2.71534529	2.5073E-13	29.5339372	105.703248	2.33810372	3.5958E-09
gap junction protein, beta 2	Gjb2	466.955423	1377.76121	2.69949858	4.5552E-30	466.955423	1234.05857	2.45019492	5.0056E-24
solute carrier organic anion transporter family, member 4a1	Slco4a1	47.7162055	169.127353	2.6755642	1.5302E-13	47.7162055	92.288298	1.58286893	0.00504209
SRY (sex determining region Y)-box 2	Sox2	1.7407353	44.0829915	2.66988929	5.5642E-09	1.7407353	14.6161948	1.43883641	0.09856763
platelet derived growth factor receptor, alpha polypeptide	Pdgfra	3115.15488	8509.45825	2.60761287	1.8624E-47	3115.15488	2774.2256	-1.1163749	0.27015438
BTB (POZ) domain containing 11	Btbd11	395.548323	1128.86745	2.60321898	1.2837E-30	395.548323	468.368597	1.1561862	0.24393558
chemokine (C-X-C motif) ligand 5	Cxcl5	326.105364	1068.75976	2.56915347	1.373E-12	326.105364	716.431248	1.83901036	3.7567E-05
teneurin transmembrane protein 4	Tenm4	518.878455	1542.75415	2.56766402	1.6525E-17	518.878455	777.588656	1.41571372	0.01235578
dynactin associated protein	Dynap	135.395406	399.70383	2.55615329	2.249E-19	135.395406	168.717532	1.19011059	0.26569057
LIM homeobox transcription factor 1 beta	Lmx1b	9.27675368	56.3178892	2.54476058	1.9901E-08	9.27675368	36.7096318	1.93926558	0.00036214
interleukin 1 receptor-like 1	Il1rl1	6517.2516	16370.8782	2.47472727	8.005E-112	6517.2516	6903.2188	1.05721003	0.3904014
S100 calcium binding protein A16	S100a16	194.164971	540.017815	2.46650158	2.3791E-20	194.164971	337.147226	1.62386457	1.3547E-05
pro-platelet basic protein	Ppbp	281.783911	752.639986	2.45158995	2.7229E-21	281.783911	555.733427	1.83568095	2.4042E-09
SLIT and NTRK-like family, member 6	Slitrk6	101.338872	267.280842	2.45012761	2.2342E-15	101.338872	94.7034135	-1.0364693	0.87798622
stanniocalcin 1	Stc1	570.960696	1458.39492	2.44517734	1.1939E-33	570.960696	525.12974	-1.0652864	0.63215101
tetraspanin 11	Tspan11	191.547889	487.030946	2.43931661	9.1394E-29	191.547889	194.105123	1.01341935	0.94267674
protein tyrosine phosphatase, non-receptor type 13	Ptpn13	2319.43332	6180.97857	2.43657515	5.0084E-22	2319.43332	4800.04713	1.93583462	9.0976E-12
inositol polyphosphate-4-phosphatase, type II	Inpp4b	38.647779	139.048752	2.42500458	9.7424E-10	38.647779	47.5440336	1.05411227	0.85307414
mesothelin	Msln	94.8803598	252.687251	2.42313404	2.0679E-16	94.8803598	115.97148	1.209853	0.2302722
plakophilin 1	Pkp1	247.243603	629.508284	2.40847809	2.0998E-24	247.243603	377.973505	1.5070381	4.7237E-05
solute carrier organic anion transporter family, member 2a1	Slco2a1	706.396386	1953.21767	2.39182498	4.8339E-14	706.396386	1008.55147	1.3569393	0.04063668
SH2 domain containing 5	Sh2d5	21.5814343	80.8246867	2.39138088	1.9957E-08	21.5814343	18.1514119	-1.268931	0.29530018
matrix metallopeptidase 10	Mmp10	194.158143	483.545105	2.37584089	2.3765E-22	194.158143	92.2319447	-1.9644836	9.1886E-11
uridine phosphorylase 1	Upp1	98.7345971	254.8278	2.37436284	6.9582E-16	98.7345971	89.8885642	-1.1023956	0.60433384
RIKEN cDNA 1700012B09 gene	1700012B09	35.7824245	107.99172	2.36826389	2.9262E-09	35.7824245	21.6987941	-1.4116178	0.083757
receptor (calcitonin) activity modifying protein 3	Ramp3	447.507127	1120.29098	2.35989618	9.1843E-28	447.507127	455.554789	1.01030688	0.95498973
transmembrane protein 74	Tmem74	47.4629724	137.920668	2.35626597	3.0627E-10	47.4629724	81.2260883	1.43963792	0.03756677
neurofilament, light polypeptide	Nefl	9.35888857	43.5187591	2.34788232	6.0851E-07	9.35888857	18.6723294	1.29500272	0.27452285
insulin-like growth factor binding protein 4	Igfbp4	1334.59241	3296.60021	2.34769627	1.311E-29	1334.59241	1457.61627	1.09259709	0.46545598
early growth response 1	Egr1	119.989019	397.382522	2.34765373	1.6979E-09	119.989019	729.317897	3.85840791	3.0658E-23
solute carrier family 29 (nucleoside transporters), member 2	Slc29a2	231.60392	632.245992	2.3183837	3.1354E-12	231.60392	385.083991	1.53094683	0.00308132
amphiregulin	Areg	152.497791	382.731677	2.30326765	3.7321E-20	152.497791	165.94577	1.03291954	0.86492337
ATP-binding cassette, sub-family B (MDR/TAP), member 1A	Abcb1a	35.5826231	124.818208	2.28134437	2.2317E-07	35.5826231	38.7777996	1.00917617	0.97953635
B cell CLL/lymphoma 6, member B	Bcl6b	31.3120711	96.696222	2.26954406	3.3943E-08	31.3120711	49.925942	1.37748957	0.10462077
heparan sulfate (glucosamine) 3-O-sulfotransferase 1	Hs3st1	33.797491	104.19539	2.25918022	2.1877E-08	33.797491	50.8797892	1.30844891	0.17897592
uncoupling protein 2 (mitochondrial, proton carrier)	Ucp2	367.949283	943.493742	2.25492896	1.5434E-13	367.949283	904.553843	2.18734334	4.1476E-12
HtrA serine peptidase 3	Htra3	128.55064	382.721851	2.24632399	6.0845E-08	128.55064	169.765412	1.22762119	0.33681013
apelin	Apln	569.051448	1317.49437	2.23683648	3.2331E-29	569.051448	606.711944	1.05429775	0.68517776
solute carrier family 14 (urea transporter), member 1	Slc14a1	43.1917414	121.953647	2.23560386	5.0422E-08	43.1917414	53.1698142	1.18979449	0.43518206
ATPase, class V, type 10D	Atp10d	376.995665	899.667831	2.21237516	3.8224E-21	376.995665	396.820193	1.03614497	0.83370112
paired-like homeodomain transcription factor 1	Pitx1	57.4823739	159.184687	2.19408866	1.6825E-09	57.4823739	229.02654	3.0843358	4.2328E-19
E2F transcription factor 8	E2f8	446.019925	1159.18301	2.1633259	6.0871E-09	446.019925	597.202328	1.26460883	0.19310259
laminin, beta 3	Lamb3	50.6992264	136.619835	2.15123375	3.3114E-09	50.6992264	57.3894466	1.01663897	0.9541412
angiogenin, ribonuclease A family, member 2	Ang2	29.9063644	87.7223263	2.1490838	4.241E-07	29.9063644	29.2347842	-1.1239732	0.64560433
TNF receptor-associated factor 1	Traf1	7.37259726	36.0484944	2.12357566	1.5914E-05	7.37259726	22.2050312	1.499192	0.05942895
integrin beta 3	Itgb3	2448.54278	5620.54075	2.11933605	1.3723E-14	2448.54278	2467.03178	1.00802702	0.9707814
tetratricopeptide repeat domain 9	Ttc9	799.476058	1754.10484	2.11832459	1.7587E-31	799.476058	1038.45759	1.28665642	0.00152108
isthmin 1, angiogenesis inhibitor	Ism1	15.6973576	50.9379937	2.1065506	7.13E-06	15.6973576	22.2536692	1.14077684	0.61939252
mitogen-activated protein kinase 13	Mapk13	222.005625	519.942271	2.09430824	5.0517E-12	222.005625	254.33016	1.11898335	0.50688134
anoctamin 1, calcium activated chloride channel	Ano1	510.991344	1161.79934	2.09143296	1.505E-16	510.991344	679.464037	1.28265708	0.03189815
podoplanin	Pdpn	2980.07429	6344.91877	2.0821372	5.7474E-39	2980.07429	2988.93788	1.00692782	0.9572059
neuronal pentraxin 1	Nptx1	62.2079972	151.826694	2.0784333	1.0855E-08	62.2079972	67.1941784	1.06417609	0.79558775
neurofilament, medium polypeptide	Nefm	18.1817591	64.2705451	2.05337145	3.7349E-05	18.1817591	35.2186143	1.41319178	0.11691875
high mobility group AT-hook I, related sequence 1	Hmga1-rs1	5094.28082	10800.3102	2.04521667	2.572E-25	5094.28082	8458.92736	1.62202378	3.3472E-11
sema domain, transmembrane domain (TM), and cytoplasmic domain, (se	Sema6d	887.862884	1886.72582	2.0290313	3.3596E-20	887.862884	726.739774	-1.2047693	0.07788857
sema domain, immunoglobulin domain (Ig), transmembrane domain (TM)	Sema4b	752.251683	1680.51681	2.02136634	2.2393E-10	752.251683	835.278732	1.09964859	0.59792531
ecotropic viral integration site 2a	Evi2a	93.3930274	200.169126	2.01617271	9.5531E-11	93.3930274	75.5067802	-1.2226447	0.20739795
sema domain, immunoglobulin domain (Ig), TM domain, and short cytopla	Sema4f	117.377747	257.487274	2.01328174	9.6713E-11	117.377747	80.205827	-1.3777804	0.02605644
cytohesin 1 interacting protein	Cytip	61.6794865	163.575385	2.0098007	1.788E-06	61.6794865	67.47863	-1.0442752	0.87691826
superoxide dismutase 3, extracellular	Sod3	66.5152689	152.332354	2.00159899	2.0007E-08	66.5152689	106.159516	1.46433519	0.01154312
hyaluronan synthase1	Has1	46.3023124	110.057383	2.00104476	3.9855E-07	46.3023124	66.8727224	1.30317024	0.14950723
XIAP associated factor 1	Xaf1	277.225443	107.258552	-2.0046858	8.8233E-06	277.225443	173.301598	-1.4397292	0.05679382
guanylate binding protein 7	Gbp7	370.466121	168.491492	-2.0116289	9.2475E-13	370.466121	232.961891	-1.5113832	0.00015269
coiled-coil serine rich 1	Ccser1	73.5955371	28.1216916	-2.012506	8.7827E-06	73.5955371	50.2063058	-1.3355778	0.14513573
solute carrier family 26, member 7	Slc26a7	38.1858792	11.2198487	-2.021993	6.9703E-05	38.1858792	28.4222804	-1.1957479	0.47584582
synaptonemal complex protein 1	Sycp1	165.556361	71.5323763	-2.0270965	1.471E-07	165.556361	148.884831	-1.1239521	0.56800656
asporin	Aspn	5380.67646	2439.26952	-2.0296133	5.194E-12	5380.67646	4421.51946	-1.194272	0.21123721
nucleoporin 62 C-terminal like	Nup62cl	1177.71357	528.88007	-2.0311254	3.4058E-11	1177.71357	724.497662	-1.5519636	0.00025166
endothelial cell-specific molecule 1	Esm1	25.9698225	1.5405459	-2.0344982	1.289E-06	25.9698225	2.78339568	-1.8974466	3.4929E-05
G protein-coupled receptor 1	Gpr1	242.399464	102.043485	-2.0377494	1.8824E-08	242.399464	144.046764	-1.5544241	0.00233405
gap junction protein, alpha 5	Gja5	31.103744	6.06440862	-2.0386482	5.9143E-05	31.103744	13.0423404	-1.572524	0.03024052
vomeronasal 2, receptor 79	Vmn2r79	39.5989236	8.33888486	-2.04784	3.6636E-05	39.5989236	3.63851515	-2.5094738	6.4409E-08
tubulin polymerization promoting protein	Tppp	102.669951	41.0315664	-2.0540433	3.4607E-07	102.669951	50.8054898	-1.756856	0.00023368
cytidine monophosphate (UMP-CMP) kinase 2, mitochondrial	Cmpk2	213.873498	91.4555944	-2.0552819	6.7337E-10	213.873498	176.662457	-1.1495069	0.40886708

sortilin-related VPS10 domain containing receptor 1	Sorcs1	181.571017	77.2459441	-2.0570683	3.526E-09	181.571017	105.72441	-1.6263767	0.00031333
testis development related protein	Tdrp	268.371493	114.520002	-2.0573469	3.1779E-10	268.371493	154.601624	-1.6312265	0.00010611
LIM and senescent cell antigen like domains 2	Lims2	1166.22946	530.190266	-2.0627428	3.0309E-17	1166.22946	749.284015	-1.4972607	2.4607E-05
involucrin	Ivl	206.469652	91.3391273	-2.0651303	7.1774E-11	206.469652	115.718422	-1.6590023	2.9479E-05
interferon regulatory factor 7	Irf7	291.734456	127.485779	-2.0653718	8.4593E-12	291.734456	179.56405	-1.5333282	0.00032442
histocompatibility 2, M region locus 9	H2-M9	58.8530583	20.5155926	-2.0686468	1.1685E-05	58.8530583	24.6501373	-1.7496574	0.00233405
Eph receptor A3	Epha3	3711.16703	1728.01398	-2.0791244	1.2837E-30	3711.16703	2981.59216	-1.2361518	0.0077546
mitogen-activated protein kinase-activated protein kinase 3	Mapkapk3	1196.3894	535.996267	-2.0792671	3.9002E-16	1196.3894	620.511739	-1.8337123	1.0085E-10
DDB1 and CUL4 associated factor 12-like 1	Dcaf12l1	293.555459	130.367125	-2.0822128	1.2565E-14	293.555459	216.488091	-1.3157787	0.01811377
chemokine (C-C motif) ligand 9	Ccl9	157.866027	55.793623	-2.0830693	4.8946E-06	157.866027	47.5159776	-2.3150343	2.8988E-07
F-box and WD-40 domain protein 10	Fbxw10	68.4549569	17.7160878	-2.0995134	2.5164E-05	68.4549569	39.4612232	-1.4002193	0.12952221
RIKEN cDNA 1500009L16 gene	1500009L16F	297.401171	125.204523	-2.1029186	1.6134E-12	297.401171	195.759518	-1.436469	0.00325372
phospholipid phosphatase 1	Plpp1	16317.5589	7468.93312	-2.1038299	6.1291E-27	16317.5589	11234.0797	-1.4281075	4.5503E-06
transmembrane protein 196	Tmem196	42.2479344	11.9530157	-2.1074944	1.8142E-05	42.2479344	42.5380189	1.02120296	0.95134113
collagen, type XIX, alpha 1	Col19a1	154.72298	60.1284205	-2.1098575	1.3469E-08	154.72298	92.1945292	-1.4631603	0.01496104
peripheral myelin protein 2	Pmp2	24.9734323	4.01515012	-2.1139431	2.3824E-05	24.9734323	18.4975376	-1.10011	0.7464419
membrane metallo endopeptidase	Mme	158.258391	66.0405917	-2.1286568	3.1779E-10	158.258391	115.607529	-1.3144042	0.07161736
G protein-coupled receptor, family C, group 5, member C	Gprc5c	65.9722527	19.8535852	-2.1341544	6.1655E-06	65.9722527	47.3293827	-1.173329	0.51198315
small proline-rich protein 2K	Sprr2k	487.34406	190.083757	-2.1357686	9.1527E-09	487.34406	668.869559	1.31657308	0.10764691
chemokine (C-C motif) ligand 20	Ccl20	35.1205537	4.71750589	-2.1421154	1.2572E-05	35.1205537	36.8526193	1.03417608	0.92118752
cathepsin S	Ctss	108.592105	40.804823	-2.1524198	8.9845E-08	108.592105	29.6013711	-2.7596857	2.5516E-12
WD repeat and FYVE domain containing 1	Wdfy1	6202.96489	2848.10636	-2.1602454	1.338E-110	6202.96489	5430.77814	-1.1419423	0.0018195
LIM and calponin homology domains 1	Limch1	1538.97305	670.684752	-2.1628342	2.6871E-19	1538.97305	1838.48405	1.18226171	0.15615772
chemokine (C-X3-C motif) receptor 1	Cx3cr1	74.0067207	22.7915945	-2.1954943	1.8048E-06	74.0067207	33.3251177	-1.7240478	0.00336981
aminoadipate-semialdehyde synthase	Aass	142.75778	53.1828032	-2.1959594	1.0155E-08	142.75778	114.815487	-1.1750949	0.41367661
regulator of G-protein signaling 5	Rgs5	104.819154	36.8212474	-2.2036969	4.7239E-08	104.819154	73.3329198	-1.2900937	0.17682104
CD40 antigen	Cd40	404.591682	170.051112	-2.2077396	2.331E-19	404.591682	199.982148	-1.924526	6.8471E-13
complement component 1, q subcomponent, C chain	C1qc	78.6027571	23.0335965	-2.2311581	1.51E-06	78.6027571	30.4300071	-1.8900291	0.00045791
complement receptor 2	Cr2	51.2716247	13.4174428	-2.2339336	3.3442E-06	51.2716247	9.29053789	-2.7435041	4.8133E-09
solute carrier family 35, member G2	Slc35g2	60.5033026	13.8580472	-2.2474515	3.1288E-06	60.5033026	14.0595059	-2.2588671	6.1808E-06
C1q and tumor necrosis factor related protein 3	C1qtnf3	14233.5801	5769.41321	-2.2540865	3.1856E-16	14233.5801	7958.05111	-1.6904832	1.3049E-06
fatty acid binding protein 7, brain	Fabp7	140.567065	49.8483516	-2.2687132	5.7447E-10	140.567065	96.9173744	-1.3354689	0.08504039
protease, serine 46	Prss46	59.7956772	11.7817921	-2.2777885	2.6594E-06	59.7956772	16.3586981	-1.9533107	0.00043849
endomucin	Emcn	94.2639042	32.6683551	-2.2816884	5.2142E-09	94.2639042	70.1445299	-1.2736098	0.19183354
R-spondin 3	Rspo3	326.862857	130.198276	-2.2880013	8.7853E-18	326.862857	322.289946	-1.009352	0.96377728
testis expressed gene 15	Tex15	674.60122	273.839763	-2.3017465	3.3829E-21	674.60122	450.977231	-1.4589255	0.00016925
EF hand domain containing 1	Efhd1	75.3367816	23.9992294	-2.3149305	2.3121E-08	75.3367816	66.2709025	-1.0864536	0.73935139
lysozyme 2	Lyz2	640.693301	245.467467	-2.3190661	4.9062E-15	640.693301	263.986704	-2.2120653	6.9373E-13
activating transcription factor 3	Atf3	479.706989	187.800977	-2.3343292	6.367E-21	479.706989	312.41742	-1.4722059	0.00015105
KN motif and ankyrin repeat domains 4	Kank4	126.438836	37.8960993	-2.3357158	4.1279E-08	126.438836	77.3202456	-1.3637929	0.1206519
interferon gamma induced GTPase	Igtp	211.828135	69.4773953	-2.3556111	1.338E-09	211.828135	109.482476	-1.664147	0.00165354
gamma-aminobutyric acid (GABA) A receptor, subunit beta 3	Gabrb3	220.921799	79.8529502	-2.3596382	2.3079E-12	220.921799	159.292374	-1.3079011	0.09039602
cytochrome P450, family 4, subfamily a, polypeptide 12B	Cyp4a12b	249.648925	92.4436468	-2.3665844	4.752E-16	249.648925	105.317142	-2.1279639	5.5532E-12
fibronectin type III domain containing 5	Fndc5	79.8001367	19.0119616	-2.3671225	3.8332E-07	79.8001367	31.8017151	-1.773855	0.00301077
potassium channel tetramerisation domain containing 12b	Kctd12b	120.935712	37.4048471	-2.4034568	1.3355E-09	120.935712	96.2192214	-1.1912154	0.40047094
regulator of G-protein signaling 4	Rgs4	4211.73911	1638.11148	-2.429843	4.2914E-32	4211.73911	3660.15539	-1.1416179	0.22286187
thymine DNA glycosylase	Tdg	223.550502	77.7386347	-2.4535568	2.0961E-13	223.550502	339.344885	1.406731	0.02048491
thymine DNA glycosylase, pseudogene	Tdg-ps	223.550502	77.7386347	-2.4535568	2.0961E-13	223.550502	339.344885	1.406731	0.02048491
solute carrier family 5 (choline transporter), member 7	Slc5a7	1077.10637	398.396013	-2.5418031	9.4366E-34	1077.10637	634.038623	-1.6395199	1.9341E-09
ribonuclease T2B	Rnaset2b	64.4920881	15.2920443	-2.5494434	2.426E-08	64.4920881	12.2418811	-2.8114219	1.296E-09
neurocalcin delta	Ncald	481.119015	164.708807	-2.5887519	7.3333E-23	481.119015	313.38718	-1.4541002	0.00086616
guanylate binding protein 2	Gbp2	575.172315	184.543788	-2.6071234	5.5649E-15	575.172315	282.350323	-1.8577676	3.6346E-06
family with sequence similarity 181, member B	Fam181b	140.393082	38.8164246	-2.6120159	1.5574E-10	140.393082	19.4097118	-4.2198874	7.3269E-22
syntrophin, gamma 1	Sntg1	94.6426507	25.4473406	-2.6248673	8.8435E-11	94.6426507	68.3075073	-1.3215711	0.15390077
cell adhesion molecule 1	Cadm1	176.891997	56.3297143	-2.6403214	7.5561E-16	176.891997	94.2252323	-1.7082655	5.0547E-05
serine/threonine kinase 26	Stk26	169.936902	50.5050187	-2.72659	1.1956E-15	169.936902	120.705258	-1.3698124	0.04444912
collagen, type VIII, alpha 2	Col8a2	685.604242	236.908151	-2.7401064	1.5535E-48	685.604242	471.566154	-1.4377001	1.1646E-06
transmembrane protein 88	Tmem88	263.720837	73.9612908	-2.7404852	1.1299E-14	263.720837	175.477031	-1.3533977	0.07433453
ribosomal protein L34	Rpl34	1239.37663	174.965987	-2.8285385	8.606E-10	1239.37663	217.545527	-2.5834357	7.7989E-08
ribosomal protein L34, pseudogene 1	Rpl34-ps1	1239.37663	174.965987	-2.8285385	8.606E-10	1239.37663	217.545527	-2.5834357	7.7989E-08
predicted gene 4705	Gm4705	1239.37663	174.965987	-2.8285385	8.606E-10	1239.37663	217.545527	-2.5834357	7.7989E-08
RIKEN cDNA 3830417A13 gene	3830417A13	83.6630695	18.1996235	-2.8412797	1.6515E-11	83.6630695	47.6334734	-1.5182997	0.02818962
late cornified envelope 1G	Lce1g	126.35704	32.0333443	-2.84619	1.8506E-14	126.35704	75.5736056	-1.4476986	0.02683728
myosin VIIA	Myo7a	424.88928	119.354271	-2.9089239	5.9193E-19	424.88928	280.459851	-1.4071497	0.02391029
ectonucleotide pyrophosphatase/phosphodiesterase 5	Enpp5	392.159112	117.618555	-2.9314538	1.5658E-28	392.159112	195.551711	-1.8765374	6.4671E-10
late cornified envelope 1H	Lce1h	590.719809	173.241424	-2.9847001	8.1118E-28	590.719809	438.814193	-1.3047304	0.03869838
ubiquitin specific peptidase 18	Usp18	314.747545	75.8766217	-3.1058899	5.337E-18	314.747545	159.585725	-1.733819	0.00020504
netrin 4	Ntn4	2496.98282	727.578491	-3.1998381	7.3592E-57	2496.98282	2065.89059	-1.1927594	0.07787436
tripartite motif-containing 30A	Trim30a	62.3710788	7.26441419	-3.5807343	1.317E-14	62.3710788	4.47305267	-4.3955581	2.228E-19
ATPase, H+ transporting, lysosomal V0 subunit C	Atp6v0c	339.649275	70.9875122	-3.7110289	5.3365E-26	339.649275	68.728053	-3.6296803	1.6274E-24
X-prolyl aminopeptidase (aminopeptidase P) 2, membrane-bound	Xpnpep2	74.384279	6.92768851	-4.2069541	1.2075E-19	74.384279	17.9084636	-2.7459391	1.0542E-09
ribosomal protein S18	Rps18	1824.50163	374.079424	-4.2349863	2.8546E-54	1824.50163	430.196317	-3.7769031	1.2814E-45
NADH dehydrogenase (ubiquinone) 1 alpha subcomplex, 12	Ndufa12	785.07962	127.469081	-4.2790027	2.2371E-31	785.07962	680.260649	-1.1314531	0.53845054
carbonic anhydrase 8	Car8	267.801302	37.9569594	-4.8094997	3.4467E-35	267.801302	44.3196309	-4.5777753	1.7173E-32
spermatogenesis associated glutamate (E)-rich protein 3	Speer3	53.5482083	0.20127248	-4.8806973	1.1654E-22	53.5482083	30.4018304	-1.3921246	0.14173074
proline rich 9	Prr9	129.839529	12.6150231	-5.1441226	9.636E-33	129.839529	74.3158864	-1.5792341	0.00345635
histocompatibility 2, T region locus 23	H2-T23	302.049111	39.1900652	-5.5851865	6.068E-46	302.049111	37.0606673	-6.0641773	1.0389E-48
interferon-induced protein 44	Ifi44	454.625564	37.1164803	-6.8057643	2.2414E-43	454.625564	77.0224674	-4.1633693	2.8874E-24
ribosomal protein S2	Rps2	4790.88714	534.763925	-7.8809918	3.646E-167	4790.88714	546.523211	-7.7903453	1.223E-162
predicted gene 29376	Gm29376	157.641087	4.05053933	-8.9604317	1.4617E-50	157.641087	4.84987028	-8.5912582	1.9744E-48
histocompatibility 2, T region locus 22	H2-T22	1002.51961	89.8157478	-9.6073631	4.656E-172	1002.51961	78.4156284	-10.680361	1.381E-177
histocompatibility 2, T region locus 9	H2-T9	1002.51961	89.8157478	-9.6073631	4.656E-172	1002.51961	78.4156284	-10.680361	1.381E-177
tripartite motif-containing 30D	Trim30d	149.791915	0	-12.750712	3.2846E-59	149.791915	0.89340609	-11.922393	5.2908E-56
ribosomal protein L29	Rpl29	2104.72968	54.7644598	-19.429614	2.63E-141	2104.72968	68.0201294	-17.094978	1.76E-128
predicted pseudogene 8210	Gm8210	2104.72968	54.7644598	-19.429614	2.63E-141	2104.72968	68.0201294	-17.094978	1.76E-128
tripartite motif-containing 12A	Trim12a	207.539651	0	-20.88215	3.5986E-87	207.539651	0.37681761	-19.698875	1.0291E-83

Table S2: Significantly regulated by KRas^{G12D} in MEFs

Table shows RNA-SEQ reads for each gene significantly regulated by more than 2 fold upon activation of lsl-KRas^{G12D} allele for 24hrs in primary MEFs. Comparison is with identically treated passage-matched wild-type MEFs. FC = fold change, numbers >2 in red, <-2 in dark blue. Adjusted p values <0.05 shown in red. Also shown are RNA-SEQ reads for the same genes upon similar activation of Rosa26$^{DM\text{-}lsl\text{-}MYC}$, compared with wt MEFs, using the same colour scheme, plus FC>1.5 in pink; <-1.5 in light blue, to highlight convergent and divergent gene expression.

Muthalagu et al. Table S3: Significantly regulated by KRasG12D+Rosa26-DM-MYC in MEFs

gene_name	symbol	Mean WT	Mean MycKras	FC	padj
aggrecan	Acan	43.2824021	660.527155	8.72130989	4.3253E-84
glucosaminyl (N-acetyl) transferase 1, core 2	Gcnt1	27.7489748	196.0805209	4.54473728	1.1057E-31
gap junction protein, beta 2	Gjb2	466.955423	2364.985825	4.4734454	2.0759E-69
chemokine (C-X-C motif) ligand 3	Cxcl3	8.98697059	94.71136401	4.04589344	2.8361E-22
elastin	Eln	45.1426709	439.011641	4.02905949	2.2919E-19
early growth response 1	Egr1	119.989019	767.9263895	4.00565515	6.2083E-25
CAP-GLY domain containing linker protein family, member 4	Clip4	47.8791174	254.6629093	3.83665229	2.5113E-31
BarH-like homeobox 1	Barx1	1.82270046	54.87072145	3.61275007	2.339E-16
integrin alpha 2	Itga2	403.501296	1888.796944	3.50096975	2.8319E-24
paired-like homeodomain transcription factor 1	Pitx1	57.4823739	245.0427559	3.22606801	1.9355E-21
solute carrier family 4 (anion exchanger), member 4	Slc4a4	416.774414	1391.747637	3.13830216	1.9212E-58
pro-platelet basic protein	Ppbp	281.783911	952.7617401	3.05332845	2.3675E-33
nuclear receptor subfamily 1, group H, member 5	Nr1h5	14.0005099	88.86336074	3.02592418	1.2637E-12
ATPase, H+ transporting, lysosomal V0 subunit E2	Atp6v0e2	39.8441402	154.8247926	2.95212193	6.3004E-17
chemokine (C-X-C motif) ligand 12	Cxcl12	1184.25058	3660.38941	2.84973329	1.4587E-37
angiopoietin-like 7	Angptl7	16.4852509	89.58234157	2.80177018	1.5142E-11
plakophilin 1	Pkp1	247.243603	735.1157984	2.79493142	8.5981E-34
S100 calcium binding protein A16	S100a16	194.164971	610.8912967	2.76495743	3.9614E-26
complement component 1, s subcomponent 1	C1s1	20.503079	92.90708547	2.67849947	3.9917E-11
matrix metallopeptidase 3	Mmp3	136.749548	388.1413648	2.61628812	4.5829E-22
insulin-like growth factor 2	Igf2	912.105521	2743.793326	2.6156507	3.0533E-18
solute carrier family 14 (urea transporter), member 1	Slc14a1	43.1917414	142.279444	2.55847704	4.7713E-11
solute carrier family 29 (nucleoside transporters), member 2	Slc29a2	231.60392	693.2518481	2.5161548	7.7344E-15
protein tyrosine phosphatase, non-receptor type 13	Ptpn13	2319.43332	6292.845664	2.47729156	6.2862E-23
chemokine (C-X-C motif) ligand 5	Cxcl5	326.105364	1015.029521	2.46134975	1.2897E-11
5' nucleotidase, ecto	Nt5e	364.391148	948.3364829	2.4534367	2.5556E-43
neuronal cell adhesion molecule	Nrcam	154.069126	405.2908065	2.45275411	1.0552E-22
solute carrier family 17 (sodium-dependent inorganic phosphate cotransporter), member 6	Slc17a6	21.3287104	72.05974685	2.44233379	8.5243E-09
TraB domain containing 2B	Trabd2b	108.71626	323.800266	2.4268601	6.4063E-12
peptidase inhibitor 15	Pi15	300.065366	772.8549345	2.40254069	3.5911E-25
leucine rich repeat transmembrane neuronal 2	Lrrtm2	157.096374	452.4045032	2.39798828	2.4869E-12
angiotensin II receptor, type 2	Agtr2	15.7793228	63.09436434	2.37747528	6.8206E-09
unc-13 homolog C (C. elegans)	Unc13c	166.588113	446.5198225	2.3726152	3.0654E-20
E2F transcription factor 8	E2f8	446.019925	1282.780131	2.35597781	4.5548E-11
aristaless-like homeobox 3	Alx3	11.6796992	51.00408659	2.35214561	1.1618E-07
collagen, type XIV, alpha 1	Col14a1	356.822992	997.6342172	2.3360876	1.1259E-11
neuronal guanine nucleotide exchange factor	Ngef	110.62466	290.5733401	2.31492917	7.6643E-14
HtrA serine peptidase 3	Htra3	128.55064	395.6963846	2.31208268	1.2681E-08
alanyl (membrane) aminopeptidase	Anpep	16.1140118	78.06614769	2.30063755	4.7562E-07
dynactin associated protein	Dynap	135.395406	355.4514782	2.3001447	1.8935E-15
anoctamin 1, calcium activated chloride channel	Ano1	510.991344	1282.927235	2.29657399	3.222E-21
inositol polyphosphate-4-phosphatase, type II	Inpp4b	38.647779	128.9562996	2.29337157	9.4866E-09
interleukin 11	Il11	361.891052	865.667004	2.28880643	9.2992E-26
mesothelin	Msln	94.8803598	235.8996116	2.27843511	1.9754E-14
transcription factor AP4	Tfap4	219.96692	541.1771647	2.27088676	1.5009E-19
BTB (POZ) domain containing 11	Btbd11	395.548323	974.1622865	2.26804115	1.6953E-22
secreted phosphoprotein 1	Spp1	8558.83351	20824.18223	2.2642879	5.0037E-20
uncoupling protein 2 (mitochondrial, proton carrier)	Ucp2	367.949283	932.1181487	2.23283864	2.3911E-13
lumican	Lum	63.0348167	170.3432406	2.21496348	3.8988E-09
RIKEN cDNA 1700012B09 gene	1700012B09	35.7824245	99.24117258	2.20652483	4.9675E-08
collagen, type XV, alpha 1	Col15a1	181.236837	488.6881144	2.20494558	3.3312E-09
aquaporin 1	Aqp1	1619.79319	3686.133335	2.20375796	5.4711E-42
high mobility group AT-hook I, related sequence 1	Hmga1-rs1	5094.28082	11675.17447	2.20318228	7.5195E-31
tropomodulin 2	Tmod2	264.580741	637.6939635	2.20010036	2.8043E-17
expressed sequence AI854703	AI854703	7.08230498	30.85897026	2.18256861	4.8729E-06
potassium voltage-gated channel, subfamily Q, member 5	Kcnq5	215.999978	517.1273532	2.18213624	4.8225E-22
ribosomal protein L35A	Rpl35a	262.829783	628.5910554	2.16756141	1.1815E-13
fibroblast growth factor 10	Fgf10	436.392758	1040.427576	2.15730709	4.9993E-15
protein tyrosine phosphatase, non-receptor type 22 (lymphoid)	Ptpn22	18.5545258	58.72308617	2.15492829	1.1384E-06
SRY (sex determining region Y)-box 2	Sox2	1.7407353	31.27556278	2.13456412	1.0552E-05
treacle ribosome biogenesis factor 1	Tcof1	1426.34483	3515.810541	2.13369332	6.505E-10
neuronal pentraxin 1	Nptx1	62.2079972	154.9921632	2.1292723	1.7548E-09
heparan sulfate (glucosamine) 3-O-sulfotransferase 1	Hs3st1	33.797491	96.35717858	2.11674296	2.4426E-07
solute carrier organic anion transporter family, member 2a1	Slco2a1	706.396386	1682.816281	2.10184339	2.1296E-10
family with sequence similarity 84, member A	Fam84a	57.7738543	138.6790084	2.07393404	9.1553E-08
paired-like homeodomain transcription factor 2	Pitx2	35.3300689	93.12904054	2.07171447	1.786E-07
suprabasin	Sbsn	275.542252	605.1940891	2.06599509	1.5494E-16
cDNA sequence BC048403	BC048403	95.9687685	286.4009379	2.05846599	8.9413E-06
immunoglobulin superfamily, DCC subclass, member 4	Igdcc4	164.432421	392.1430876	2.05313107	3.1167E-09
delta-like 1 homolog (Drosophila)	Dlk1	903.19726	2256.644499	2.04900392	3.7725E-07
autism susceptibility candidate 2	Auts2	184.062926	442.3620265	2.04896119	1.0349E-08
mitogen-activated protein kinase 6	Mapk6	9710.89006	21504.09072	2.03999985	1.6452E-12
histone cluster 2, H4	Hist2h4	548.593784	1238.962513	2.03180415	4.0673E-10
histone H4	LOC1026412	548.593784	1238.962513	2.03180415	4.0673E-10
RIKEN cDNA D630045J12 gene	D630045J12F	125.532936	295.9722922	2.02439498	8.5567E-08
teneurin transmembrane protein 4	Tenm4	518.878455	1169.028293	2.01159066	7.2554E-10
dual specificity phosphatase 5	Dusp5	1871.02166	3908.14032	2.00997903	9.0078E-22
forkhead box G1	Foxg1	178.634599	395.5432177	2.00713522	5.4451E-11
elastin microfibril interfacer 2	Emilin2	1490.22016	3202.722406	2.00332407	4.0013E-13

proline arginine-rich end leucine-rich repeat	Prelp	41.7037301	131.8208943	2.00052337	4.6195E-05
family with sequence similarity 131, member A	Fam131a	137.008591	62.19531699	-2.0037806	1.905E-09
integrin alpha 1	Itga1	1202.2431	573.1027717	-2.0046094	1.3395E-17
colony stimulating factor 3 (granulocyte)	Csf3	97.5283522	34.20659012	-2.0107963	1.7073E-05
sodium channel, voltage-gated, type IX, alpha	Scn9a	26.2155482	3.238997904	-2.0137925	1.6612E-05
apolipoprotein E	Apoe	227.811032	92.7521333	-2.0183322	3.2224E-07
signal transducer and activator of transcription 2	Stat2	516.539366	219.0429329	-2.0206629	6.1262E-08
relaxin family peptide receptor 3	Rxfp3	66.2254858	25.48563631	-2.0218288	4.0325E-06
leucine rich adaptor protein 1-like	Lurap1l	7727.90395	3572.800699	-2.0245457	2.4103E-14
complement receptor 2	Cr2	51.2716247	16.32053798	-2.025147	3.9435E-05
histocompatibility 2, M region locus 9	H2-M9	58.8530583	21.06952806	-2.0274305	1.2697E-05
2'-5' oligoadenylate synthetase-like 2	Oasl2	148.893523	59.46124755	-2.0302615	7.2023E-07
solute carrier family 5 (choline transporter), member 7	Slc5a7	1077.10637	504.8016549	-2.0304789	3.6236E-20
collagen, type XIX, alpha 1	Col19a1	154.72298	63.32499166	-2.039001	3.6006E-08
Fc receptor-like S, scavenger receptor	Fcrls	28.9465242	2.456358016	-2.0406097	1.0841E-05
regulator of G-protein signaling 17	Rgs17	556.418868	257.8992665	-2.0423032	5.1789E-22
poly (ADP-ribose) polymerase family, member 10	Parp10	250.856358	110.1328546	-2.0490297	2.2967E-11
predicted gene 11627	Gm11627	59.9310741	22.46576914	-2.0511706	4.3037E-06
H6 homeobox 1	Hmx1	27.3318114	4.585264129	-2.0546842	3.0596E-05
growth arrest specific 6	Gas6	12192.2078	5621.318529	-2.0577779	1.5017E-18
ubiquitin-like modifier activating enzyme 7	Uba7	154.134888	55.44497327	-2.0589803	2.7643E-06
3-hydroxyacyl-CoA dehydratase 4	Hacd4	1220.58051	576.6124299	-2.0594458	9.2351E-36
solute carrier family 9 (sodium/hydrogen exchanger), member 2	Slc9a2	121.430937	48.14517182	-2.059999	2.4887E-07
calponin 1	Cnn1	23237.2368	10540.96748	-2.0650551	1.2391E-15
potassium inwardly-rectifying channel, subfamily J, member 15	Kcnj15	543.67994	250.4940698	-2.0674358	1.1015E-19
hydroxyprostaglandin dehydrogenase 15 (NAD)	Hpgd	17.2290868	1.801337024	-2.0685058	1.6288E-05
gap junction protein, alpha 5	Gja5	31.103744	6.109006919	-2.0738783	2.5653E-05
membrane bound O-acyltransferase domain containing 2	Mboat2	1141.56361	531.7237451	-2.074678	2.1215E-24
interferon induced transmembrane protein 1	Ifitm1	56.5303805	17.98304468	-2.0757661	1.1737E-05
vanin 1	Vnn1	90.6256712	36.87086744	-2.075802	2.5404E-07
SLAM family member 9	Slamf9	96.3751996	35.03607974	-2.0764284	2.3923E-06
sialic acid binding Ig-like lectin G	Siglecg	3279.74261	1524.628809	-2.0765181	1.3713E-28
leucine zipper, putative tumor suppressor 1	Lzts1	33.0082399	8.104747016	-2.0817596	2.7369E-05
urotensin 2B	Uts2b	30.9775517	5.398581139	-2.0846884	2.4183E-05
colony stimulating factor 1 receptor	Csf1r	155.474493	65.83160968	-2.0869723	5.703E-11
coiled-coil domain containing 109B	Ccdc109b	1763.40645	792.9085818	-2.0929763	2.2324E-17
discs, large homolog 2 (Drosophila)	Dlg2	35.3287111	9.350895571	-2.1043066	1.866E-05
proteasome (prosome, macropain) subunit, beta type 9 (large multifunctional peptidase 2)	Psmb9	33.171661	7.333201368	-2.105112	2.0232E-05
proteasome (prosome, macropain) subunit, beta type 8 (large multifunctional peptidase 7)	Psmb8	209.201646	91.25572378	-2.1095918	1.3462E-12
neuronatin	Nnat	410.077514	188.4459194	-2.1108308	8.412E-24
growth arrest and DNA-damage-inducible 45 beta	Gadd45b	1999.75717	885.6799464	-2.1109689	9.5866E-17
hematopoietic cell specific Lyn substrate 1	Hcls1	26.1716606	3.602674048	-2.1130477	1.2698E-05
solute carrier family 16 (monocarboxylic acid transporters), member 4	Slc16a4	231.979611	93.74835681	-2.114531	9.7551E-09
late cornified envelope 1G	Lce1g	126.35704	48.53386077	-2.1158553	3.6006E-08
serine (or cysteine) peptidase inhibitor, clade B, member 9	Serpinb9	977.128334	425.5520959	-2.1198575	5.5114E-15
phospholipid phosphatase 1	Plpp1	16317.5589	7372.631142	-2.1305622	7.8599E-28
integrin alpha M	Itgam	29.9068736	5.415222501	-2.1308098	1.2918E-05
5-hydroxytryptamine (serotonin) receptor 7	Htr7	169.018404	67.09710674	-2.1314889	6.3519E-09
Fas (TNF receptor superfamily member 6)	Fas	347.992993	149.2047095	-2.1349609	1.0588E-12
myosin IF	Myo1f	21.7004586	2.539666365	-2.1361889	5.2877E-06
RIKEN cDNA E030030I06 gene	E030030I06R	16.693578	0.979816521	-2.1382339	6.5647E-06
tubulin polymerization-promoting protein family member 3	Tppp3	961.243135	426.380101	-2.140408	2.7853E-21
DEAD (Asp-Glu-Ala-Asp) box polypeptide 58	Ddx58	1695.45791	756.1395665	-2.1454868	6.6648E-27
sulfotransferase family 4A, member 1	Sult4a1	312.441789	134.6661757	-2.147663	8.6022E-17
microtubule associated tumor suppressor candidate 2	Mtus2	104.617655	40.77638292	-2.1646201	4.5105E-09
podocan-like 1	Podnl1	590.055731	252.3144858	-2.1672797	1.2074E-16
interferon, alpha-inducible protein 27 like 2B	Ifi27l2b	1080.6444	455.7242859	-2.1725355	7.5655E-15
vomeronasal 2, receptor 79	Vmn2r79	39.5989236	6.977845905	-2.183698	3.7274E-06
nucleoporin 62 C-terminal like	Nup62cl	1177.71357	487.4879327	-2.1854531	1.1448E-13
CD40 antigen	Cd40	404.591682	172.2584967	-2.1871379	1.694E-19
R-spondin 3	Rspo3	326.862857	138.0132603	-2.1873311	1.57E-16
serine (or cysteine) peptidase inhibitor, clade B, member 9e	Serpinb9e	35.2836353	5.900970421	-2.1936269	4.9005E-06
cannabinoid receptor 1 (brain)	Cnr1	233.5057	95.69689561	-2.1955099	8.7026E-14
GLI pathogenesis-related 1 (glioma)	Glipr1	2326.70834	981.1081857	-2.2002966	4.948E-18
G protein-coupled receptor 1	Gpr1	242.399464	92.85191506	-2.2162355	1.1631E-10
heat shock protein 1B	Hspa1b	777.396344	336.4784954	-2.2243304	1.4712E-33
mannosidase, beta A, lysosomal	Manba	652.632213	278.899354	-2.2280622	1.1002E-29
Rho guanine nucleotide exchange factor (GEF) 28	Arhgef28	5442.99473	2368.910884	-2.2282216	7.842E-40
leucine rich repeat containing 17	Lrrc17	5204.48312	2188.569634	-2.2405089	5.2665E-23
TNNI3 interacting kinase	Tnni3k	129.220697	46.11655133	-2.2423737	1.0581E-08
stabilin 1	Stab1	68.7845148	16.2593517	-2.2650219	1.4641E-06
X-prolyl aminopeptidase (aminopeptidase P) 2, membrane-bound	Xpnpep2	74.384279	23.16220994	-2.2693074	3.8133E-07
testis development related protein	Tdrp	268.371493	103.215777	-2.2741044	2.4131E-13
radical S-adenosyl methionine domain containing 2	Rsad2	578.391987	221.7490324	-2.2923461	1.7109E-13
fibronectin type III domain containing 5	Fndc5	79.8001367	20.47851769	-2.2938709	7.686E-07
LIM and senescent cell antigen like domains 2	Lims2	1166.22946	471.6642876	-2.2998673	5.9004E-23
receptor transporter protein 4	Rtp4	170.584947	58.25397961	-2.3013025	9.1413E-10
schlafen 2	Slfn2	222.116803	80.63089328	-2.303594	5.1297E-11
coronin, actin binding protein 1A	Coro1a	109.467774	36.53237207	-2.3170559	1.1157E-08
interferon regulatory factor 9	Irf9	1013.01507	396.1726594	-2.3173809	1.0639E-17
membrane-spanning 4-domains, subfamily A, member 6D	Ms4a6d	39.2208561	8.035306467	-2.3186152	1.0627E-06

leukocyte immunoglobulin-like receptor, subfamily B, member 4A	Lilrb4a	25.9261046	1.595956898	-2.3240116	3.2346E-07
fatty acid binding protein 7, brain	Fabp7	140.567065	49.29708574	-2.3340211	6.0849E-11
interferon induced protein with tetratricopeptide repeats 1B like 2	Ifit1bl2	61.2915354	15.08524929	-2.34252	3.0195E-07
CD36 antigen	Cd36	21.536698	2.00949071	-2.3493454	3.5012E-07
basal cell adhesion molecule	Bcam	129.67509	46.92958319	-2.3529653	8.0525E-13
myosin VIIA	Myo7a	424.88928	153.7035558	-2.3544743	1.1509E-12
adhesion G protein-coupled receptor A1	Adgra1	96.8297617	28.12744001	-2.3650885	2.7182E-08
collagen, type VIII, alpha 2	Col8a2	685.604242	275.204883	-2.3775632	1.069E-37
immunity-related GTPase family M member 1	Irgm1	3314.20491	1323.852711	-2.3924187	5.2874E-38
popeye domain containing 2	Popdc2	218.848451	81.62469479	-2.4026294	2.4129E-16
isochorismatase domain containing 2b	Isoc2b	37.1963175	9.264930849	-2.4046983	9.4691E-08
guanylate binding protein 3	Gbp3	203.734903	70.99078971	-2.4111277	1.4109E-12
tripartite motif-containing 34A	Trim34a	63.3314282	15.38785634	-2.4148749	1.4222E-07
lipase, member O2	Lipo2	138.035551	45.84710982	-2.4153821	1.4312E-11
membrane-spanning 4-domains, subfamily A, member 6C	Ms4a6c	21.9909206	0.494068601	-2.4236048	1.945E-08
monoacylglycerol O-acyltransferase 2	Mogat2	382.644819	142.2131286	-2.4717491	3.0321E-23
cDNA sequence BC025446	BC025446	28.744686	3.197460917	-2.5175598	4.2501E-08
V-set immunoregulatory receptor	Vsir	428.620289	158.830207	-2.5191874	3.8613E-29
myomesin 2	Myom2	153.979653	46.06109577	-2.5328666	2.2815E-11
C1q and tumor necrosis factor related protein 3	C1qtnf3	14233.5801	5025.291081	-2.5510381	1.3193E-21
toll-like receptor 13	Tlr13	33.3801578	3.491696513	-2.5545345	2.5013E-08
tumor protein D52-like 1	Tpd52l1	396.10367	141.8797116	-2.5600299	1.0185E-26
neurocalcin delta	Ncald	481.119015	168.480936	-2.5621635	6.8179E-23
activating transcription factor 3	Atf3	479.706989	168.8697578	-2.5732914	2.9985E-26
C-type lectin domain family 5, member a	Clec5a	24.7649355	0.738329341	-2.5785365	1.8876E-09
testis expressed gene 15	Tex15	674.60122	241.1993993	-2.5824382	1.2958E-27
macrophage scavenger receptor 1	Msr1	82.1299824	17.98038831	-2.5841827	6.8423E-09
cell migration inducing protein, hyaluronan binding	Cemip	3938.67541	1386.596372	-2.5977894	3.6703E-26
serine/threonine kinase 26	Stk26	169.936902	53.66883267	-2.6352547	3.9789E-15
ribonuclease T2B	Rnaset2b	64.4920881	14.50798953	-2.6520134	3.4545E-09
aminoadipate-semialdehyde synthase	Aass	142.75778	41.3646705	-2.6613747	2.3247E-13
guanylate binding protein 7	Gbp7	370.466121	123.1087299	-2.6857061	5.7901E-25
regulator of G-protein signaling 4	Rgs4	4211.73911	1468.210273	-2.6950705	5.334E-40
ribosomal protein L34	Rpl34	1239.37663	195.9066733	-2.7009456	4.3832E-09
ribosomal protein L34, pseudogene 1	Rpl34-ps1	1239.37663	195.9066733	-2.7009456	4.3832E-09
predicted gene 4705	Gm4705	1239.37663	195.9066733	-2.7009456	4.3832E-09
C-type lectin domain family 4, member n	Clec4n	24.311901	1.104661858	-2.7187284	1.6862E-09
cell adhesion molecule 1	Cadm1	176.891997	54.17397986	-2.7342009	1.9571E-17
immunity-related GTPase family M member 2	Irgm2	186.244906	52.80559157	-2.7866105	2.6609E-16
poly (ADP-ribose) polymerase family, member 9	Parp9	1074.79941	351.7998527	-2.7900025	4.7634E-34
MX dynamin-like GTPase 2	Mx2	75.8284028	14.63838199	-2.8039933	1.1856E-10
chemokine (C-C motif) ligand 9	Ccl9	157.866027	36.14657386	-2.8120922	1.395E-11
tubulin polymerization promoting protein	Tppp	102.669951	26.95940424	-2.8336009	2.3816E-14
monooxygenase, DBH-like 1	Moxd1	2066.86929	702.783185	-2.864394	4.4897E-97
cytidine monophosphate (UMP-CMP) kinase 2, mitochondrial	Cmpk2	213.873498	61.46241758	-2.8875887	1.0789E-20
TYRO protein tyrosine kinase binding protein	Tyrobp	45.0157997	2.056692005	-2.8913393	1.211E-10
matrix metallopeptidase 12	Mmp12	73.0535392	5.118330533	-3.0086281	3.7734E-11
cytochrome P450, family 4, subfamily a, polypeptide 12B	Cyp4a12b	249.648925	67.34686383	-3.1134707	2.7041E-27
chemokine (C-X3-C motif) receptor 1	Cx3cr1	74.0067207	12.43171464	-3.1206534	5.1736E-13
lymphocyte cytosolic protein 1	Lcp1	110.70374	23.8922186	-3.26211	2.4796E-18
interferon gamma induced GTPase	Igtp	211.828135	44.72298227	-3.264545	6.5627E-18
cytochrome b-245, beta polypeptide	Cybb	58.2279072	5.170961761	-3.2706961	1.0077E-12
XIAP associated factor 1	Xaf1	277.225443	52.98861526	-3.271451	6.0366E-16
adhesion G protein-coupled receptor E1	Adgre1	48.4593625	5.123760465	-3.2875253	6.7451E-13
complement component 1, q subcomponent, beta polypeptide	C1qb	53.7119689	4.998915129	-3.3258967	3.1413E-13
OTU domain containing 7A	Otud7a	398.537016	104.8369304	-3.3393484	1.2395E-40
membrane-spanning 4-domains, subfamily A, member 7	Ms4a7	29.9055157	0.449640866	-3.3779466	4.1648E-14
processing of precursor 4, ribonuclease P/MRP family, (S. cerevisiae)	Pop4	2438.43211	678.2621733	-3.3798818	7.3365E-73
family with sequence similarity 181, member B	Fam181b	140.393082	25.12727291	-3.5182639	9.5681E-18
interferon regulatory factor 7	Irf7	291.734456	66.12520856	-3.6284693	1.0514E-34
guanylate binding protein 2	Gbp2	575.172315	117.2472768	-3.7887929	1.4663E-28
complement component 1, q subcomponent, alpha polypeptide	C1qa	96.0403408	7.260870072	-3.8934177	7.1565E-17
ATPase, H+ transporting, lysosomal V0 subunit C	Atp6v0c	339.649275	64.47130769	-3.9612336	6.6604E-29
macrophage expressed gene 1	Mpeg1	142.382428	20.30606872	-4.0230683	3.1362E-22
complement component 1, q subcomponent, C chain	C1qc	78.6027571	6.639299762	-4.2522126	5.5718E-20
ribosomal protein S18	Rps18	1824.50163	345.4474561	-4.549037	4.5249E-60
ubiquitin specific peptidase 18	Usp18	314.747545	44.34548909	-4.6637025	1.621E-32
spermatogenesis associated glutamate (E)-rich protein 3	Speer3	53.5482083	0	-5.2288727	1.2378E-24
tripartite motif-containing 30A	Trim30a	62.3710788	0.693901606	-5.98552	8.3679E-29
histocompatibility 2, T region locus 23	H2-T23	302.049111	35.03601332	-6.1190391	1.9691E-51
cathepsin S	Ctss	108.592105	6.84178914	-6.3988109	3.8871E-38
lysozyme 2	Lyz2	640.693301	65.33199395	-7.1067361	1.6143E-74
interferon-induced protein 44	Ifi44	454.625564	26.78418172	-8.3869943	4.2967E-53
ribosomal protein S2	Rps2	4790.88714	443.3232484	-9.4032585	6.888E-197
histocompatibility 2, T region locus 22	H2-T22	1002.51961	68.02135051	-12.304463	4.221E-205
histocompatibility 2, T region locus 9	H2-T9	1002.51961	68.02135051	-12.304463	4.221E-205
tripartite motif-containing 30D	Trim30d	149.791915	0	-13.352236	5.0447E-61
tripartite motif-containing 12A	Trim12a	207.539651	0.24426074	-21.544905	8.2444E-89

Table S3: Significantly regulated by the combination of MYC and KRas^{G12D} in MEFs

Table shows RNA-SEQ reads for each gene significantly regulated by more than 2 fold upon combined activation of Rosa26$^{DM\text{-}lsl\text{-}MYC}$ and lsl-KRas^{G12D} alleles for 24hrs in primary MEFs. Comparison is with identically treated passage-matched wild-type MEFs. FC = fold change, numbers >2 in red; <-2 in dark blue. Adjusted p values <0.05 shown in red.

Table S4: Significantly regulated by MYC or Miz1 depletion in KMC PDAC cells

Mean RNA-SEQ values for expressed genes in KMC cells, with and without siRNA depletion of MYC or Miz1, filtered for genes significantly regulated by MYC depletion. Fold change and Adjusted P values for siMYC and siMiz1, relative to control siRNA-treated KMC cells, shown.

SUPPLEMENTARY METHODS

RNA-SEQ analysis

Pancreatic tumours were removed from euthanised subject animals and dissected into 3-5mm fragments to remove all extraneous (eg. normal, gut or lymph node) tissue. Total RNA was isolated using the RNEasy Mini Kit (Qiagen) according to manufacturer's protocol and DNA was depleted with the RNase-Free DNase Set (Qiagen). Quality of the purified RNA was tested on an Agilent 2200 Tapestation using RNA screentape. For analysis of MEF gene expression, libraries were prepared using the TruSeq stranded totalRNA with RiboZero kit (Illumina). For analysis of bulk tumour gene expression, cDNA libraries were prepared as previously (2), using Illumina TruSeq Stranded mRNA LT Kit. Quality and quantity of the cDNA libraries was assessed on an Agilent 2200 Tapestation (D1000 screentape) and Qubit (Thermo Fisher Scientific) respectively. Libraries were run on an Illumina NextSeq500 using the High Output 75 cycles kit (2x36 cycles, paired end reads, single index). Quality checks on the raw RNA-Seq data files were done using FastQC (3) and Fastq Screen (4). Alignment of the RNA-Seq paired-end reads was to the GRCm38 (5) version of the mouse genome and annotation using HiSat2 (6) and TopHat (7). Expression levels were determined and statistically analysed by a workflow combining HTSeq (8), the R environment (9), utilising packages from the Bioconductor data analysis suite (10) and differential gene expression analysis based on the negative binomial distribution using the DESeq2 package (11). Further data analysis and visualisation used R and Bioconductor packages. Pathway modulation analysis was performed using Metacore GeneGO (12). Gene-set enrichment for PDAC subtype gene signatures was performed on the converted mouse identifiers using the 'GSVA' package (13). PDAC subtype gene signatures are as defined in Bailey et al. (1).

Software Versions for pancreatic tumour analysis:

FastQC version 0.11.7; Fastq Screen version 0.12.0; GRCm38.93 version of the mouse genome and annotation; HiSat2 version 2.1.0; HTSeq version 0.9.1; R environment version 3.4.4

Software Versions for KC pancreatic tumours:

GRCm38.75 version of the mouse genome and annotation; TopHat run version 2.0.13

Software Versions for MEF experiments:

GRCm38.75 version of the mouse genome and annotation; TopHat run version 2.0.13; Bowtie version 2.2.4.0; R environment version 3.3.2

RT-PCR

RNA was isolated using TRIZOL (Invitrogen) method according to manufacturer's directions. Quantitect reverse transcription kit (Qiagen 205313) was used for cDNA synthesis followed by real time PCR using SYBR Green method (VWR QUNT95072). Gusb and ACTIN were used for data normalization in mouse and human samples respectively. The following primer sets were used to detect indicated mRNA transcripts:

Mouse

Gusb F: cacttcggcaccacctagag, R: accgcagggtgatttttgt

Irf5 F: tacgaggtctgctccaacg, R: gcctggtagcattctctgga

Irf7 F: cttcagcactttcttccgaga, R: tgtagtgtggtgacccttgc

Ifi44 F: cacacgtggatagcctggat, R: ggcaaaaccaaagactccat

Ifnb1 F: aactccaccagcagacagtg, R: agtggagagcagttgaagac

Myc F: cgcgtccgagtgcattga, R: agcagcgagtccgaggaa

Miz1 F: tctgtggtggggttcgggac, R: ctgctccaagacacgctggc

Cxc13 F: tcacacatataactttcttcatcttgg, R: catagatcggattcaagttacgc

Human

Actin F: ccaaccgcgagaagatga, R: ccagaggcgtacagggatag

IRF5 F: cacactccagcccactttc, R: gcatctgcacctggtaagg

IRF7 F: ctgcagtcacacctgtagcc, R: gtggactgagggcttgtagc

IFI44 F: agcctgtgaggtccaagcta, R: gcagaaagaattagaacatcctttaca

IFNB1 f: gacgccgcattgaccatcta, R: agccaggaggttctcaacaat

Myc F : gcccctggtgctccatga, R: caacatcgatttcttcctcatcttct

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