Appendix

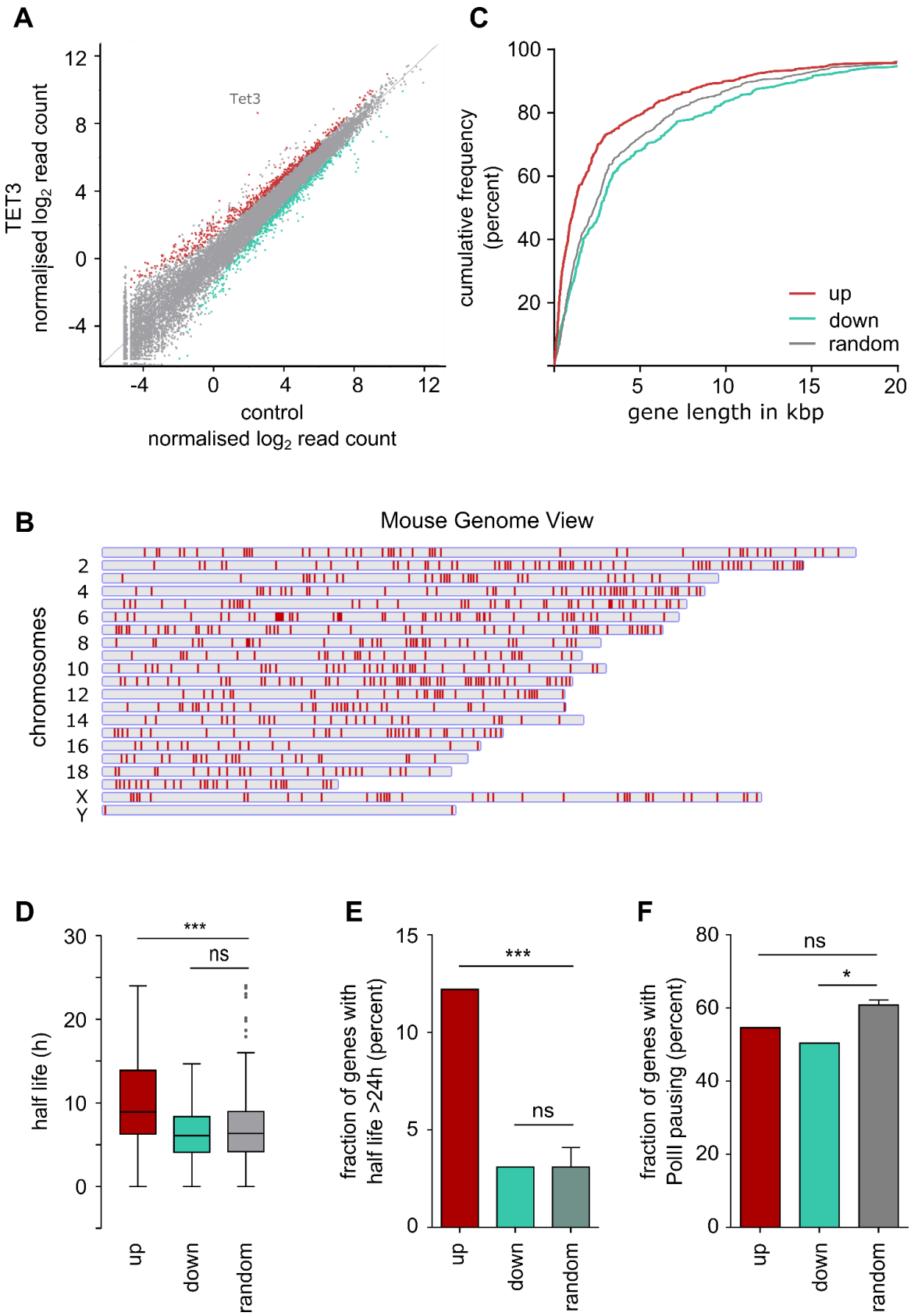
Text S1

In the analysis of RNA-seq experiments different statistical filters are routinely used to identify differentially expressed genes, and the numbers of genes called differentially expressed can vary dramatically between filters. There is not necessarily one 'correct' list of differentially expressed genes, and the meaningfulness of the filter depends on the question asked. For the analysis shown in Figure 2A we have used the intersection of DESeq2 and an Intensity Difference Filter. This constitutes very stringent filtering for differential gene expression. To carefully explore the data, we also assessed differential expression using a combination of DESeq2 and a Dynamic Fold Change filters (for details see Methods) which resulted in over 1000 genes being called as differentially regulated. Interestingly, even in this larger group of genes functional enrichment analysis did not reveal any overrepresented gene ontology groups or biological pathways.

We thus asked whether the observed differential expression of the larger, less stringently filtered group was due to particular features of the genes or transcripts. Expression differences occurred at all expression levels (Suppl. Fig. S1A) and all genomic locations (Suppl. Fig. S1B). Interestingly, genes that showed higher expression in the TET3 expressing samples ('up' group) were significantly shorter than a random set of genes. Those which showed lower expression in the TET3 overexpressing samples ('down' group) tended to be longer although the difference was not statistically significant (Supp. Fig. S1C). Notably, using transcript half-life data from Sharova et al. 2009, transcripts in the 'up' group were found to be more stable (Sup. Fig. S1D), and were highly enriched for transcripts with very long half-lives (>24 h, Supp. Fig. S1E). We also examined if the groups of differentially expressed genes showed differences in their regulation by RNA polymerase pausing. Using a genome-wide nuclear run-on assay, Min and co-workers sorted ES cell promoters into four groups: transcribed and paused, transcribed and not paused, not transcribed and paused or not transcribed and not paused. We took their list of transcribed genes and intersected this with the 'up' and 'down' groups of Tet3 differentially regulated genes. Interestingly, the 'down' group was depleted for paused genes (Supp. Fig. S1F).

Taken together, we conclude that TET3 does not specifically regulate a functionally related set of genes. Rather, TET3's global effects on transcription and total RNA levels affect genes differently depending on their transcriptional features. The observed expression differences are likely caused by different susceptibility of RNA steady-state levels to a global alteration.

Supplementary Figure S1



Supplementary Figure S1 legend

A) Scatter plot showing RNA-seq data quantified as $\log_2$ read count per mRNA normalised to the total number of reads. Each dot represents one transcript. Red dots: Significantly higher in TET3 expressing cells; turquoise dots: significantly less in TET3 expressing cells (intersection of DESeq2 and dynamic fold change filters, see methods).

B) Genome View showing the localisation of differentially regulated genes between TET3 expressing and control cells. Chromosomes are displayed as grey bars and location of genes by red stripes.

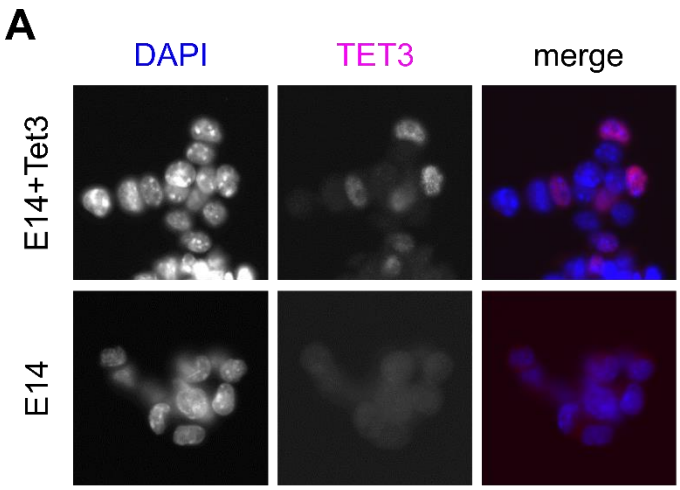
C) Cumulative frequency of lengths of genes that are relatively higher (up, red, n=431) or relatively lower (down, turquoise, n=410) in TET3 expressing cells, and randomly sampled genes (random, grey, n=500). 'Up' are significantly shorter than random genes (Kruskal-Wallis with Dunn's multiple comparisons post test, p=0.0004).

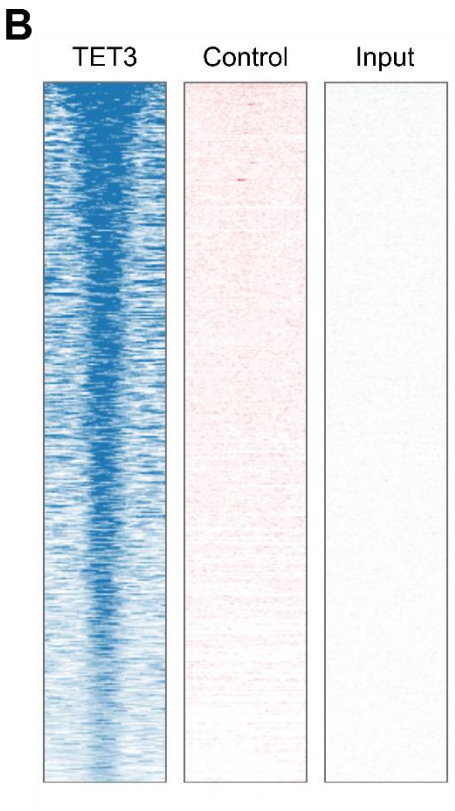
D) Distribution of half-lives for genes that are expressed and are relatively higher (up, red, n=431) or relatively lower (down, turquoise, n=410) in TET3 expressing cells, and randomly sampled genes (random, grey, n=500) shown as Tukey box whisker plots. 'Up' genes have significantly longer half-lives than a random sample (Kruskal-Wallis with Dunn's multiple comparisons post test, *** p<0.001, ns: not significant). Transcript stability data was taken from (Sharova et al. 2009).

E) Fraction of genes per group (up, down, random) which have half-lives of 24 h or longer (data from (Sharova et al. 2009)). 'Up' genes were significantly enriched for transcripts with long half-lives (>24h, Chi-square test with Bonferroni correction, *** p=0.0004, ns: not significant).

F) Fraction of genes per group (up, down, regulated) which are transcribed and paused. Data for RNA polymerase II pausing was obtained from (Min et al. 2011): Class I genes are transcribed and not paused, class II genes are transcribed and paused. According to this classification 339 'up' genes, 343 'down' genes and 413 random genes are transcribed (class I + class II). Paused fractions of those were calculated. The 'down' group is depleted for paused genes (Chi-square test with Bonferroni correction, * p=0.0344, ns: not significant).

Supplementary Figure S2





C

PROTEIN	ACCESSION	SCORE	COVERAGE	# UNIQUE PEPTIDES
TET3	Q8BG87	3767.00	43.35	52
OGT	Q8CGY8	1989.21	48.57	38
TRAP1	Q9CQN1	1191.07	53.68	29
PKD2	O35245	978.28	26.71	18
EMSY	Q8BMB0	911.31	30.06	16

Supplementary Figure S2 legend

A) Immunofluorescence imaging using anti-TET3 antibody (ABE290) in cells stably transfected with a Tet3 overexpression plasmid (pIG414, see Table S6) and untransfected E14 cells as described in Material and Methods, Nascent Transcription Assay. Images were taken on an Olympus BX61 epifluorescence microscope. Nuclear TET3 signal is only present in transfected cells.

B) Aligned probes plot covering MACS peaks and 1 kb upstream and downstream regions. MACS peaks were called on Tet3 overexpressing ES cells. Each horizontal line corresponds to one genomic region and ChIP-seq reads are represented by density of colour. Regions are sorted by intensity of TET3 signal. TET3 (blue lines): TET3 ChIP-seq signal (antibody Millipore ABE290) in cell transfected with a Tet3 expression construct, Control (red lines): TET3 ChIP-seq signal (ABE290) in cells transfected with a control construct (same vector backbone, containing a small part of the Tet3 gene excluding the epitope encoding region), Input (green lines): ChIP input material before antibody pull-down. Regions showing an enrichment for ABE290 pull-down in Tet3 overexpressing cells only show background signal in cells overexpressing a control construct. Input material shows hardly any signal.

C) Table of the five most frequently found proteins by rapid immunoprecipitation mass spectrometry of endogenous proteins (RIME, Mohammed et al. 2016) with anti-TET3 antibody (Millipore ABE290) but not with normal rabbit IgG (Santa Cruz Biotechnology sc-2027). TET3 is the most frequent protein found, followed by its known interactor OGT.

Table S1: Genes differentially expressed between Tet3 expressing and control cells

GENE	ID	DESCRIPTION	LOG_2 FC	FDR
GM10222	ENSMUSG00000067736	predicted gene 10222	-2.6	6.38E-11
SFRP2	ENSMUSG00000027996	secreted frizzled-related protein 2	-2.5	3.14E-07
PHLDA2	ENSMUSG00000010760	pleckstrin homology-like domain, family A, member 2	-2.2	3.01E-07
BAHCC1	ENSMUSG00000039741	BAH domain and coiled-coil containing 1	-2.1	2.53E-07
PTCH1	ENSMUSG00000021466	patched homolog 1	-1.9	4.92E-11
GM4027	ENSMUSG00000092019	predicted gene 4027	1.9	0.04659
RPL39L	ENSMUSG00000039209	ribosomal protein L39-like	2.4	3.56E-07
AC140240.1	ENSMUSG00000096528	Putative uncharacterized protein	2.9	1.26E-16
PRAMEL7	ENSMUSG00000025839	preferentially expressed antigen in melanoma like 7	3.1	2.06E-13
TET3	ENSMUSG00000034832	tet methylcytosine dioxygenase 3	6.1	0

Log_2 fold-change (FC) for each gene was calculated from the mean reads per million (RPM) for three replicates of Tet3 overexpressing versus control cells. Negative fold-change reflects downregulation upon Tet3 overexpression. False discovery rate (FDR) is taken from DESeq2 statistics.

Table S2: Proteins identified by TET3 RIME

Proteins identified by mass spectrometry after immunoprecipitation with anti-TET3 antibody but not with unspecific rabbit IgG. The table is given in a separate file.

Table S3: Amino acid sequences of TET3 variants

TET3 (1714 aa)

MFLPETPQQYAVEINAREGTGPWAQGATVKTGSELSPVDGPVPGQMDSGPVYHGDSRQLSTSGAPVNGAREPAGPGLLGA
AGPWRVDQKPDWEAASGPTHAARLEDAHDLVAFSAVAEAVSSYGALSTRLYETFNREMSREAGSNGRGPRPESCSEGSEDLD
TLQTALALARHGMKPPNCTCDGPECPDFLEWLEGKIKSMAMEGGQGRPRLPGALPPSEAGLPAPSTRPPLLSSEVPQVPPLEG
LPLSQSALSIAKEKNISLQTAIAIEALTQLSSALPQPSHSTSQASCPLPEALSPSAPFRSPQSYLRAPSWPVVPPEEHPSFAPDSPAF
PPATPRPEFSEAWGTDTPPATPRNSWPVPRPSPDPMAELEQLLGSASDYIQSVFKRPEALPTKPKVKVEAPSSSPAPVPSPISQR
EAPLLSSEPDTHQKAQTALQQHLHHKRNLFLEQAQDASFPTSTEPQAPGWWAPPGSPAPRPPDKPPKEKKKKPPTPAGGPVG
AEKTTPGIKTSVRKPIQIKKSRSRDMQPLFLPVRQIVLEGLKPQASEGQAPLPAQLSVPPPASQGAASQSCATPLTPEPSLALFAP
SPSGDSLLPPTQEMRSPSPMVALQSGSTGGPLPPADDKLEELIRQFEAEFGDSFGLPGPPSVPIQEPENQSTCLPAPESPFATRS
PKKIKIESSGAVTVLSTTCFHSEEGGQEATPTKAENPLTPTLSGFLESPLKYLDTPTKSLLDTPAKKAQSEFPTCDCVEQIVEKDEGP
YYTHLGSGPTVASIRELMEDRYGEKGKAIRIEKVIYTGKEGKSSRGCPIAKWVIRRHTLEEKLLCLVRHRAGHHCQNAVIVILILAW
EGIPRSLGDTLYQELTDTLRKYGNPTSRRCGLNDDRTCACQGKDPNTCGASFSFGCSWSMYFNGCKYARSKTPRKFRLTGDNP
KEEEVLRNSFQDLATEVAPLYKRLAPQAYQNQVTNEDVAIDCRLGLKEGRPFSGVTACMDFCAHAHKDQHNLYNGCTVVCTL
TKEDNRCVGQIPEDEQLHVLPLYKMASTDEFGSEENQNAKVSSGAIQVLTAFPREVRRLPEPAKSCRQRQLEARKAAAEKKKLQ
KEKLSTPEKIKQEALELAGVTTDPGLSLKGGLSQQSLKPSLKVEPQNHFSSFKYSGNAVVESYSVLGSCRPSDPYSMSSVYSYHSR
YAQPGLASVNGFHSKYTLPSFGYYGFPSSNPVFPSQFLGPSAWGHGGSGGSFEKKPDLHALHNSLNPAYGGAEFAELPGQAVA

TDNHHPIPHHQQPAYPGPKEYLLPKVPQLHPASRDPSPFAQSSSCYNRSIKQEPIDPLTQAESIPRDSAKMSRTPLPEASQNGGP
SHLWGQYSGGPSMSPKRTNSVGGNWGVFPPGESPTIVPDKLNSFGASCLTPSHFPESQWGLFTGEGQQSAPHAGARLRGKP
WSPCKFGNGTSALTGPSLTEKPWGMGTGDFNPALKGGPGFQDKLWNPVKVEEGRIPTPGANPLDKAWQAFGMPLSSNEKL
FGALKSEEKLWDPFSLEEGTAEEPPSKGVVKEEKSGPTVEEDEEELWSDSEHNFLDENIGGVAVAPAHCSILIECARRELHATTPL
KKPNRCHPTRISLVFYQHKNLNQPNHGLALWEAKMKQLAERARQRQEEAARLGLGQQEAKLYGKKRKWGGAMVAEPQHKE
KKGAIPTRQALAMPTDSAVTVSSYAYTKVTGPYSRWI

TET3$_{trunc}$ (749 aa)

MFLPETPQQYAVEINAREGTGPWAQGATVKTGSELSPVDGPVPGQMDSGPVYHGDSRQLSTSGAPVNGAREPAGPGLLGA
AGPWRVDQKPDWEAASGPTHAARLEDAHDLVAFSAVAEAVSSYGALSTRLYETFNREMSREAGSNGRGPRPESCSEGSEDLD
TLQTALALARHGMKPPNCTCDGPECPDFLEWLEGKIKSMAMEGGQGRPRLPGALPPSEAGLPAPSTRPPLLSSEVPQVPPLEG
LPLSQSALSIAKEKNISLQTAIAIEALTQLSSALPQPSHSTSQASCPLPEALSPSAPFRSPQSYLRAPSWPVVPPEEHPSFAPDSPAF
PPATPRPEFSEAWGTDTPPATPRNSWPVPRPSPDPMAELEQLLGSASDYIQSVFKRPEALPTKPKVKVEAPSSSPAPVPSPISQR
EAPLLSSEPDTHQKAQTALQQHLHHKRNLFLEQAQDASFPTSTEPQAPGWWAPPGSPAPRPPDKPPKEKKKKPPTPAGGPVG
AEKTTPGIKTSVRKPIQIKKSRSRDMQPLFLPVRQIVLEGLKPQASEGQAPLPAQLSVPPPASQGAASQSCATPLTPEPSLALFAP
SPSGDSLLPPTQEMRSPSPMVALQSGSTGGPLPPADDKLEELIRQFEAEFGDSFGLPGPPSVPIQEPENQSTCLPAPESPFATRS
PKKIKIESSGAVTVLSTTCFHSEEGGQEATPTKAENPLTPTLSGFLESPLKYLDTPTKSLLDTPAKKAQSEFPTCDCV

TET3$_{CXXC}$ (1804 aa)

MSQFQVPLAVQPDLSGLYDFPQGQVMVGGFQGPGLPMAGSETQLRGGGDGRKKRKRCGTCDPCRRLENCGSCTSCTNRR
THQICKLRKCEVLKKKAGLLKEVEINAREGTGPWAQGATVKTGSELSPVDGPVPGQMDSGPVYHGDSRQLSTSGAPVNGAR
EPAGPGLLGAAGPWRVDQKPDWEAASGPTHAARLEDAHDLVAFSAVAEAVSSYGALSTRLYETFNREMSREAGSNGRGPR
PESCSEGSEDLDTLQTALALARHGMKPPNCTCDGPECPDFLEWLEGKIKSMAMEGGQGRPRLPGALPPSEAGLPAPSTRPPL
LSSEVPQVPPLEGLPLSQSALSIAKEKNISLQTAIAIEALTQLSSALPQPSHSTSQASCPLPEALSPSAPFRSPQSYLRAPSWPVVP
PEEHPSFAPDSPAFPPATPRPEFSEAWGTDTPPATPRNSWPVPRPSPDPMAELEQLLGSASDYIQSVFKRPEALPTKPKVKVE
APSSSPAPVPSPISQREAPLLSSEPDTHQKAQTALQQHLHHKRNLFLEQAQDASFPTSTEPQAPGWWAPPGSPAPRPPDKPP
KEKKKKPPTPAGGPVGAEKTTPGIKTSVRKPIQIKKSRSRDMQPLFLPVRQIVLEGLKPQASEGQAPLPAQLSVPPPASQGAAS
QSCATPLTPEPSLALFAPSPSGDSLLPPTQEMRSPSPMVALQSGSTGGPLPPADDKLEELIRQFEAEFGDSFGLPGPPSVPIQEP
ENQSTCLPAPESPFATRSPKKIKIESSGAVTVLSTTCFHSEEGGQEATPTKAENPLTPTLSGFLESPLKYLDTPTKSLLDTPAKKAQ
SEFPTCDCVEQIVEKDEGPYYTHLGSGPTVASIRELMEDRYGEKGKAIRIEKVIYTGKEGKSSRGCPIAKWVIRRHTLEEKLLCLVR
HRAGHHCQNAVIVILILAWEGIPRSLGDTLYQELTDTLRKYGNPTSRRCGLNDDRTCACQGKDPNTCGASFSFGCSWSMYFN
GCKYARSKTPRKFRLTGDNPKEEEVLRNSFQDLATEVAPLYKRLAPQAYQNQVTNEDVAIDCRLGLKEGRPFSGVTACMDFC
AHAHKDQHNLYNGCTVVCTLTKEDNRCVGQIPEDEQLHVLPLYKMASTDEFGSEENQNAKVSSGAIQVLTAFPREVRRLPEP
AKSCRQRQLEARKAAAEKKKLQKEKLSTPEKIKQEALELAGVTTDPGLSLKGGLSQQSLKPSLKVEPQNHFSSFKYSGNAVVESY
SVLGSCRPSDPYSMSSVYSYHSRYAQPGLASVNGFHSKYTLPSFGYYGFPSSNPVFPSQFLGPSAWGHGGSGGSFEKKPDLHA
LHNSLNPAYGGAEFAELPGQAVATDNHHPIPHHQQPAYPGPKEYLLPKVPQLHPASRDPSPFAQSSSCYNRSIKQEPIDPLTQ
AESIPRDSAKMSRTPLPEASQNGGPSHLWGQYSGGPSMSPKRTNSVGGNWGVFPPGESPTIVPDKLNSFGASCLTPSHFPES
QWGLFTGEGQQSAPHAGARLRGKPWSPCKFGNGTSALTGPSLTEKPWGMGTGDFNPALKGGPGFQDKLWNPVKVEEGRI
PTPGANPLDKAWQAFGMPLSSNEKLFGALKSEEKLWDPFSLEEGTAEEPPSKGVVKEEKSGPTVEEDEEELWSDSEHNFLDE
NIGGVAVAPAHCSILIECARRELHATTPLKKPNRCHPTRISLVFYQHKNLNQPNHGLALWEAKMKQLAERARQRQEEAARLGL
GQQEAKLYGKKRKWGGAMVAEPQHKEKKGAIPTRQALAMPTDSAVTVSSYAYTKVTGPYSRWI

Table S4: Proteins identified by TET3 SILAC RIME

DESCRIPTION	LOG_2 RATIO ($TET3/TET3_{TRUNC}$)
C-Jun-amino-terminal kinase-interacting protein 3 [JIP3_MOUSE]	6.014306
UDP-N-acetylglucosamine--peptide N-acetylglucosaminyltransferase 110 kDa subunit [OGT1_MOUSE]	4.438169
ATP-dependent RNA helicase DDX3Y [DDX3Y_MOUSE]	2.639674
Serine/arginine repetitive matrix protein 1 [SRRM1_MOUSE]	2.55985
Serine/arginine repetitive matrix protein 2 [SRRM2_MOUSE]	2.341055
Small ubiquitin-related modifier [SUMO2_MOUSE]	2.257824
Histone H2A type 1-H [H2A1H_MOUSE]	2.180717
Serine/arginine-rich splicing factor 3 [SRSF3_MOUSE]	2.167112
Histone H1.2 [H12_MOUSE]	2.107027
Plasminogen activator inhibitor 1 RNA-binding protein [PAIRB_MOUSE]	2.060282
Serine/arginine-rich splicing factor 2 [SRSF2_MOUSE]	2.048302
Insulin-like growth factor 2 mRNA-binding protein 1 [IF2B1_MOUSE]	2.040219
Probable ATP-dependent RNA helicase DDX5 [DDX5_MOUSE]	1.991832
Nucleophosmin [NPM_MOUSE]	1.975577
Polycystin-2 [PKD2_MOUSE]	1.9628
Serine/arginine-rich splicing factor 5 [SRSF5_MOUSE]	1.956321
Histone H2AX [H2AX_MOUSE]	1.939331
Peroxiredoxin-1 [PRDX1_MOUSE]	1.922404
Serine/arginine-rich splicing factor 7 [SRSF7_MOUSE]	1.863931
Nucleolin [NUCL_MOUSE]	1.82439
Histone H2A.Z [H2AZ_MOUSE]	1.791866
Histone H3.3C [H3C_MOUSE]	1.783167
Caprin-1 [CAPR1_MOUSE]	1.772848
RNA-binding protein 8A [RBM8A_MOUSE]	1.769339
T-complex protein 1 subunit theta [TCPQ_MOUSE]	1.766596
Histone H4 OS=Mus musculus [H4_MOUSE]	1.752685
Nuclease-sensitive element-binding protein 1 [YBOX1_MOUSE]	1.738551
T-complex protein 1 subunit beta [TCPB_MOUSE]	1.719965
Glyceraldehyde-3-phosphate dehydrogenase [G3P_MOUSE]	1.713721
Eukaryotic initiation factor 4A-I [IF4A1_MOUSE]	1.680657
Probable tRNA(His) guanylyltransferase [THG1_MOUSE]	1.627772
Histone H2B type 1-F/J/L [H2B1F_MOUSE]	1.619335
Transcription intermediary factor 1-beta [TIF1B_MOUSE]	1.601965
Heat shock cognate 71 kDa protein [HSP7C_MOUSE]	1.599317
T-complex protein 1 subunit zeta [TCPZ_MOUSE]	1.57674
Elongation factor 1-alpha 1 [EF1A1_MOUSE]	1.553169
Guanine nucleotide-binding protein subunit beta-2-like 1 [GBLP_MOUSE]	1.51742
Heat shock protein HSP 90-alpha [HS90A_MOUSE]	1.465976
Elongation factor 1-delta [EF1D_MOUSE]	1.454056
Tubulin beta-5 chain [TBB5_MOUSE]	1.409618
Tubulin alpha-1C chain [TBA1C_MOUSE]	1.403313
Protein NipSnap homolog 2 [NIPS2_MOUSE]	1.399752

Poly(rC)-binding protein 1 [PCBP1_MOUSE]	1.377707
T-complex protein 1 subunit delta [TCPD_MOUSE]	1.337047
Heat shock protein HSP 90-beta SV=3 - [HS90B_MOUSE]	1.315124
Protein NipSnap homolog 1 [NIPS1_MOUSE]	1.309429
Peptidyl-prolyl cis-trans isomerase A [PPIA_MOUSE]	1.246036
Pyruvate kinase PKM [KPYM_MOUSE]	1.231414
Alpha-enolase [ENOA_MOUSE]	1.217688
Histone H1.5 [H15_MOUSE]	1.214584
Elongation factor 2 [EF2_MOUSE]	1.151521
Methylcytosine dioxygenase TET3 [TET3_MOUSE]	0
Tubulin beta-4B chain [TBB4B_MOUSE]	-0.26751
TBC1 domain family member 15 SV=1 - [TBC15_MOUSE]	-0.8505
Desmoglein-1-alpha [DSG1A_MOUSE]	-1.30234
Stress-70 protein, mitochondrial [GRP75_MOUSE]	-1.58131
Cofilin-1 [COF1_MOUSE]	-1.65336
Elongation factor 1-gamma [EF1G_MOUSE]	-2.17254
Serum albumin [ALBU_MOUSE]	-3.94989

For every protein the number of peptides found in the TET3 sample was divided by the number of peptides found in the $TET3_{trunc}$ samples, and normalised for the ratio of found TET3 peptides. The list was manually curated to exclude actins, keratins and ribosomal proteins.

Table S5: Published TET3 loss of function models

STUDY	DELETED EXONS
(GU ET AL., 2011)	8-9 (correspond to exon 9-10 in ensembl Tet3-003 GRCm38.p5)
(SANTOS ET AL., 2013)	5 (corresponds to exon 6 in ensembl Tet3-003 GRCm38.p5)
(SHEN ET AL., 2014)	7-9 ((correspond to exon 8-10 in ensembl Tet3-003 GRCm38.p5)
(KANG ET AL., 2015)	2 (corresponds to exon 4 in ensembl Tet3-003 GRCm38.p5)
(TSUKADA ET AL., 2015)	3 (corresponds to exon 4 in ensembl Tet3-003 GRCm38.p5)
(IQBAL ET AL., 2011)	siRNA knockdown

Table S6: Plasmids

PLASMID	PROMOTER	TET3 VARIANT	REPORTER	SELECTION MARKER
PIG201	P_{TRE}	oocyte (Tet3)	IRES GFP	puromycin
PIG205	P_{TRE}	somatic ($Tet3_{CXXC}$)	IRES GFP	puromycin
PIG207	P_{TRE}	oocyte truncated ($Tet3_{trunc}$)	IRES GFP	puromycin
PIG208	P_{TRE}	catalytic domain	IRES GFP	puromycin
PIG301	CAG	oocyte (Tet3)	none	blasticidin
PIG305	CAG	somatic ($Tet3_{CXXC}$)	none	blasticidin
PIG307	CAG	oocyte truncated ($Tet3_{trunc}$)	none	blasticidin
PIG409	CAG	oocyte truncated ($Tet3_{trunc}$)	GFP	blasticidin
PIG410	CAG	exon 4	GFP	blasticidin
PIG414	CAG	oocyte (Tet3)	GFP	blasticidin
PIG415	CAG	none	GFP	blasticidin

P_{TRE} promoter is a CMV minimal promoter fused to tetO tetracycline-responsive elements which will drive high expression levels when bound by rtTA (reverse tetracycline transactivator). rtTA binds to its recognition sites in the presence of doxycycline. CAG is an artificial promoter made from the cytomegalovirus early enhancer element, the chicken beta-actin promoter, its first exon and intron and the rabbit beta-globin splice acceptor which drives high ubiquitous expression (Niwa et al., 1991; Okabe et al., 1997). Tet3 variants: oocyte isoform – denoted Tet3 in this study; somatic isoform – $Tet3_{CXXC}$; oocyte truncated – $Tet3_{trunc}$: truncated downstream of exon 4 producing a protein without a CXXC or a catalytic domain (aa 1-149 of TET3); catatlytic domain – aa 750-1714 of TET3. GFP as reporter denotes a C-terminal GFP fusion, and IRES GFP indicates that GFP is present on the same transcript as the Tet3 variant, but translated from an internal ribosome entry site. pIG415 which does not contain the Tet3 gene but expresses GFP was used to establish control cell lines.

Table S7: Total mapped read counts for sequencing samples

TYPE	SAMPLE	READ COUNT
CHIP	301-1 (TET3)	39395631
CHIP	301-3 (TET3)	40954550
CHIP	305-1 ($TET3_{CXXC}$)	27415291
CHIP	305-2 ($TET3_{CXXC}$)	27903005
CHIP	305-3 ($TET3_{CXXC}$)	31638682
CHIP	307-1 ($TET3_{trunc}$)	32318079
CHIP	307-2 ($TET3_{trunc}$)	31777293
CHIP	307-3 ($TET3_{trunc}$)	37155694
CHIP	Input	27143389
RNA	409-1 ($TET3_{trunc}$)	107046381
RNA	409-2 ($TET3_{trunc}$)	96215158
RNA	409-3 ($TET3_{trunc}$)	177757906
RNA	414-1 (TET3)	42078130
RNA	414-2 (TET3)	74766189
RNA	414-3 (TET3)	134326098
RNA	415-1 (CTR)	28318593
RNA	415-2 (CTR)	31997962
RNA	415-3 (CTR)	28557159
ATAC	414-1 (TET3)	31037942
ATAC	414-2 (TET3)	44266617
ATAC	414-3 (TET3)	25623861
ATAC	415-1 (CTR)	44837751
ATAC	415-2 (CTR)	44594574
ATAC	414-3 (CTR)	26622771