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Genomewide characterization of

messenger RNA alternative

polyadenylation

조선대학교 대학원

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선택적 mRNA polyadenylation의

유전체 특성 분석

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CONTENTS

| I. KOREAN ABSTRACTv |
|--|
| II. INTRODUCTION 1 |
| III. MATERIALS AND METHODS |
| 1. Cell culture······6 |
| 2. Plasmid constructs |
| 3. RNA interference 7 |
| 4. RNA-seq and alignments8 |
| 5. ApA analysis |
| 6. Scatter plot for differential expression and ApA analysis10 |
| 7. Measurement of RSI10 |
| 8. Real-time quantitative PCR (RT-qPCR) analysis12 |
| 9. Western blot analysis and antibodies |
| 10. Cell cycle analysis 14 |
| 11. Bromodeoxyuridine (BrdU) incorporation assay15 |
| 12. Chromatin immunoprecipitation (ChIP) assay15 |
| IV. RESULTS 17 |





| V. DISCUSSION | 4 |
|-----------------|---|
| VI. ABSTRACT | 8 |
| VII. REFERENCES | 0 |



CONTENTS OF FIGURES AND TABLES

| Figure 1 |
|-------------|
| Figure 223 |
| Figure 324 |
| Figure 427 |
| Figure 5 |
| Figure 6 |
| Figure 737 |
| Figure 840 |
| Figure 943 |
| Figure 1045 |
| Figure 1147 |
| Figure 12 |
| Table 1 |
| Table 2 |
| Table 376 |





| Table 4. ····· | 77 |
|----------------|----|
| Table 5. ····· | |
| Table 6. ····· | |
| Table 7 | |





국문초록

선택적 mRNA polyadenylation의 유전체 특성 분석

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메신저 RNA (mRNA)는 번역하기 전에 일련의 전사 후 처리 공정을 거쳐야 한다. 전사 후 단계 중 하나는 cleavage 및 polyadenylation 으로 이루어진 3 '말단 처리이며, 이 과정은 mRNA의 3' 말단을 구분하여 전사 및 번역 후 조절을 위한 조절 요소를 결정하는 데 중요하다. 3 ' 말단 처리는 mRNA의 3' 말단의 다양한 *trans* 작용 인자 및 *cis* 작용 인자에 의해 조절된다. 최근 polyadenylation 이 일어나는 부분의 조절로 같은 유전자에서 서로 다른 3' 말단을 가지는 mRNA 아형의 발현에 관하여 점점 밝혀지고 있다. 더 중요한 것은, 3' 말단에서 발생하는 전반적인 길이의 변화가 암 세포에서 주로 관찰

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되고 있다. 또 다른 중요한 polyadenylation 현상은 히스톤 polyadenylation 이다. 다른 mRNA와 달리 히스톤 mRNA는 polyadenylation 대신 3 '말단에 stemloop 구조를 가진다. 최근 연구에 의하면 히스톤 polyadenylation 은 DNA 대한 감수성을 높여 결손 염색체의 빈도를 손상에 증가시키고, 게놈 불안정성을 유발한다. 본 연구에서는 선택적 polyadenylation 을 분석하기 위해 종양 형성, 세포 사멸 및 게놈 안정성에 중요한 역할을 하는 종양 단백질 p53과 p53 결합 단백질로 알려져 있는 53bp1의 새로운 기전으로 mRNA 3' polyadenylation 을 조절함을 확인하였다. 또한 53bp1는 히스톤 말단의 polyadenylation 을 조절하는 중요한 인자임을 확인하였다. 이러한 연구 결과는 mRNA의 3' 말단의 선택적 polyadenylation 조절에서 p53 또는 53bp1의 이전 알려져 있지 않은 새로운 중요 인자임을 시사한다.







INTRODUCTION

In the nucleus of eukaryotic cells, the 3' end of most protein-coding genes is cleaved and polyadenylated [1-4]. A recent study shows that over 50% of human genes have multiple polyadenylation sites. For that reason, increasing the potential diversity in mRNA transcript length [5]. The formation of mRNA transcripts using these distinct polyadenylation sites (PASs) is carried out by Alternative polyadenylation (APA) with the most common form involving the differential use of APA sites located within the same transcript [6]. Processing at a most proximal polyadenylation sites in the 3' untranslated region (3' UTR) produces different-length of mRNA isoform even though encodes the same protein which removed negative regulatory elements that reduce mRNA stability or repress translation efficiency such as AU-rich elements (AREs) [7] and microRNA (miRNA) targeting sites [8, 9]. It has been reported that both rapidly proliferating cells and/or transformed cells preferentially express mRNAs with shortened 3' UTRs [4, 10-12]. In spite of these observations, the mechanisms that control the large-scale polyadenylation switch observed in proliferative, differentiation, and/ or transformed cells, as well as the





target genes subject to this regulation, are unclear [13].

Tumor protein p53 is one of the most frequently deleted or mutated tumor suppressor genes in diverse cancers, presence as a germ-line mutation in cancer-prone families with the Li-Fraumeni syndrome, and highly penetrant tumorigenic phenotype in p53 null mice [14]. The classical function of tumor suppressor gene p53 is to act as a transcription factor to regulate its downstream protein coding genes in response to various growth conditions and cellular stresses [14, 15]. Recently, over 80% of DNA binding domain mutation in p53 were found in variety cancers [16] and over 125 protein-coding genes and noncoding RNAs were direct transcriptional targets of p53 [17], which contain specific sequences for p53 binds leading to activation of their transcription activity [15]. Although originally characterized only as a transcriptional coactivator, today function of p53 have been expanded to include transcriptional repression [18], regulation of translation [19], homologous recombination [20], and transcription-independent apoptotic response [21].

p53 binding protein 1 (53bp1) was originally identified as a protein that binds to the DNA-binding domain of p53 [22]. It contains two tudor domains and C-terminal tandem







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BRCT domains [23, 24]. BRCT domain is thought to be protein-protein interaction domain and is found in many DNA damage response proteins [23, 25]. A recent report shows that 53bp1 is a key mediator of the cell's response to DSBs [26]. Upon the induction of DSB lesions, 53BP1 rapidly relocates to the sites of breaks and is believed to promote the stabilization of additional DNA damage response factors at DSBs [26]. The recognition of histone H4 dimethylated at lysine 20 (H4K20me2) by the 53bp1 has been shown to be important for 53bp1 localization to DSBs: linking chromatin structure, lysine methylation, and DSB signaling [27]. Another recent study reported that 53BP1 recognizes p53 dimethylated at lysine 370 through its Tudor domain and modulates p53 transactivation at several target genes [28]. Thus, 53BP1 might also have functions in transcription regulation. Also, 53bp1 knockout mice are radio-sensitivity, growth retardation, cell cycle defects, chromosomal instability, and cancer-prone [29].

Histone proteins are essential components of eukaryotic chromosomes. In mammalian cells, as cells enter S phase, most of the histone synthesis are increased and they are rapidly degraded at the end of S phase. In metazoan, replication-dependent histone



mRNAs are the unique mRNAs that lack poly(A) tails. Instead, they have a conserved stem-loop structure in 3' UTR, which is the *cis*-acting element required for regulation of histone mRNA half-life [30]. A small number of histone proteins (replication-independent histone) such as H1°, H2a.Z, macroH2a, H3-cid, and H3.3 are encoded by polyadenylated mRNAs [30]. This type of mRNAs encodes the variant histories used for DNA repair and chromatin remodeling [31]. They are expressed throughout the cell cycle [32]. The expression of replication-dependent histone is regulated by histone-specific pre-mRNA processing factors and universal polyadenylation associated factors. The stem-loop binding protein (Slbp) binds to 26 nucleotide stem-loop sequence, which is evolutionarily conserved in metazoans and is involved in all aspects of histone mRNA metabolism [33]. Slbp has an RNA-binding domain (RBD) that is dissimilar to the RBD of any other RNAbinding protein [33-35]. In the first step of histone mRNA 3' end processing, Slbp binds the stem-loop and then the histone downstream element (HDE) binds with a U7 small nuclear ribonucleoprotein (U7 snRNP), including the U7 small nuclear RNA (snRNA) and the Sm-like proteins Lsm10, Lsm11, [36-40]. Together, Slbp and U7 snRNP recruit a universal 3' end of polyadenylated mRNAs cleavage factor that contains cleavage and





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polyadenylation specific factor 3 (CPSF3), which has endonuclease activity and Symplekin (Sympk) which serve as a scaffold for recruiting regulatory factors [41]. Histone levels are strictly regulated to prevent deleterious effects such as genomic instability and chromosome abnormality [42, 43].

In this study, we used isogenic non-cancerous mouse embryonic fibroblast (MEF) cell lines to understand changes of molecular features on p53 or 53bp1 knockout. We employed RNA sequencing (RNA-seq) approaches to investigate the changes at high resolution and found an unexpected link between p53 or 53bp1 and APA. We show p53 or 53bp1 knockout induces global mRNA 3' UTR-APA. Our results suggest that p53 and 53bp1 have a new role in post-transcriptional gene regulation as well as pre-transcriptional regulation. We also investigated the 53bp1 knockout cells showed that decrease the expression of replication-dependent histories. We show depletion of 53bp1 increased polyadenylated histone mRNA via epigenetic regulation of well-known histone 3' end processing factor Slbp. This result indicates that 53bp1 regulate histone gene expression, which is an uncharacterized role for 53bp1 in the genomic instability.



MATERIALS AND METHODS

1. Cell culture

The human cervical adenocarcinoma cell line HeLa, obtained from the American Type Culture Collection (ATCC, Rockville, MD, USA), was cultured in Minimum Essential Media (Gibco-BRL, Grand Island, NY, USA). p53 wild-type ($p53^{+/+}$), p53 knockout ($p53^{-/-}$), 53bp1 wild-type ($53bp1^{+/+}$), 53bp1 knockout ($53bp1^{-/-}$) MEFs were cultured in Dulbecco's modified Eagle's medium (Gibco-BRL). In all cases, the media was supplemented with 10% heat-inactivated fetal bovine serum (Gibco-BRL), 100 units/ml penicillin, and 100 mg/ml streptomycin sulfate (Invitrogen, Carlsbad, CA, USA). All cells were maintained in a humidified incubator containing 5% CO₂ at 37 °C.

2. plasmid constructs

The full-length mouse p53 cDNA was amplified from addgene (pLCRcala-53) by Reverse-transcription Polymerase chain reaction (RT-PCR) using the p53 primers 5'-





AATAAGCTTATGACTGCCATGGAGGAGGAGTCACAGT-3' (forward) and 5'-AATGAATTCGTCTGAGTCAGGCCCCACTTTCTTG-3' (reverse). The amplified p53 cDNA construct was cloned into the mammalian expression vector pcDNA3 in-frame with the hemagglutinin (HA) tag. The human 53bp1 constructs were gifted from Prof. Stanley Fields laboratory and siRNA-resistant full-length 53bp1 construct prepared by the GeneTailor Site-Directed Mutagenesis kit (Invitrogen) according to the manufacturer's instructions.

3. RNA interference

| The target sequences were 5'- GGACA | AGUC | UCUCA | GCUAUd | TdT-3' | for 53bp1 | , 5'- |
|-------------------------------------|---------|-----------|-------------|--------|-----------|-------|
| GCCAGAACUUGGUAGUCAAdTdT-3' | for | ATP | citrate | lyase | (Acl), | 5'- |
| GUUCUGAUUCAAAGGAGUCUAdTdT- | 3' | for | Slbp | | and | 5'- |
| ACGAAAUUGGUGGCGUAGGdTdT-3' v | vas use | d as a ne | egative cor | ntrol. | | |





4. RNA-seq and alignments.

To evaluate transcriptome features under p53 knockout and 53bp1 knockout at nucleotide-wise resolution, we performed RNA-seq analysis of poly (A+) RNAs isolated from p53^{+/+}, p53^{-/-}, 53bp1^{+/+} and 53bp1^{-/-} MEFs. In total, 65,339,058 paired-end reads for p53^{+/+}, 72,206,840 paired-end reads for p53^{-/-} 76,292,696, paired-end reads for 53bp1^{+/+} and 79,341,754 paired-end reads for 53bp1^{-/-} MEF cells were produced from Hi-Seq pipeline with length of 50 bp of each end. The short reads were aligned to the mm10 reference genome by TopHat [44], with up to two mismatches allowed. The unmapped reads were first trimmed to remove poly-A/T tails (repeats of [A/N] s or [T/N] s) from read ends/starts and then aligned to the reference genome. It is worth noting that we only retained the reads with at least 30 bp in both ends after trimming.

5. ApA analysis

To detect the potential alternative PAS of a transcript between each wild-type and





knockout MEFs, we evaluated candidate PAS motifs (AATAAA, ATTAAA, AGTAAA, CATAAA, TATAAA, GATAAA, ACTAAA, AATACA, AATATA, AAGAAA, AATAGA, AATGAA, TTTAAA, AAAATA, TATATA, AGATAA, ATTACA, AGAATA) [5, 45] in the 3' UTR of the transcript by contrasting the short-read coverage up/downstream of the site across each wild-type and knockout samples with χ^2 -test. Specifically, we first scanned the 3' UTR of a transcript (by mm10 annotation) to identify PAS motifs as candidates of alternative PAS. For each candidate PAS, we calculated the mean coverage upstream of the site (N and M) and downstream of the site (n and m) with (N, n) denoting the coverage in wild-type and (M, m) denoting the coverage in knockout. In the calculation, the upstream region starts at the beginning of the last coding exon adjacent to the 3' UTR of the transcript and ends at the beginning of the PAS motif site. Next, a canonical 2 x 2 χ^2 -test was applied to report a P-value for each candidate site. The candidate PAS with the most significant P-value ≤ 0.05 was considered for further analysis. It is noteworthy that the χ^2 test will report shortening events in both wild type (when N/n > M/m) and knockout (when N/n < M/m).





6. Scatter plot for differential expression and ApA analysis

To select candidate transcripts with sufficient signal for reliable differential expression analysis and 3' UTR-shortening identification, we first analyzed the short-read alignments of the RNA-seq data against mouse mm10 reference genome using Cufflink [46]. We further filtered out the transcripts with positional short-read coverage ≤ 25 in the entire 3' UTR in both cell lines. In addition, transcripts with 3' UTR overlapping exons in the strand in opposite direction were removed to avoid mingled short-read signals that might lead to inaccurate 3' UTR-shortening identification. Finally, to allow precise PAS analysis, only transcripts with at least two occurrences of the 18 PAS motifs in the 3' UTR are retained in the study.

7. Measurement of RSI

A numerical presentation of 3' UTR shortening was developed by calculating the RSI of





a given transcript. A relative expression of total or longer 3' UTR-containing transcripts was measured by normalizing to the total amount of RNAs used in RT–qPCR analysis. The following equation was used to determine the RSI.

LI = [normalized expression of longer 3' UTR-containing transcript]/ [normalized expression of total (long + short) transcript]

 $RSI = -Log_2$ (LI/ [LI in reference cell line]); thus, RSI = 0 for a reference cell line. If RSI < 0 in a target cell line, then there is a 3' UTR shortening.

If RSI > 0 in a target cell line, then there is a 3' UTR lengthening. The RSI contains the information about the changes in the proportion of a longer 3' UTR-containing transcript in a given cellular context compared with a reference cell. For example, a value of 1 in the RSI of a transcript indicates that the proportion of the longer 3' UTR-containing transcript of the total (long + short) transcript decreases by 50% compared with that of the reference cell line, indicating an enrichment of 3' UTR-shortened transcript (that is, 3' UTR shortening).





8. Real-time quantitative PCR (RT-qPCR) analysis

Total RNA was isolated from cells using the Trizol Reagent (Invitrogen) according to the manufacturer's protocol. 1 µg total RNA was reverse transcribed using Superscript III reverse transcriptase (Invitrogen). Real-time quantitative PCR was performed using SYBR Premix Ex Taq (Clontech, Mountain View, CA, USA) on a Stratagene Mx3000P (Stratagene, Amsterdam, Netherlands). 20ng cDNA were amplified as follows: 94°C for 2 min, 94 °C for 5 s, 60°C for 10 s, and 72°C for 20 s. Steps 2 through 4 were repeated for 40 cycles. Each reaction was performed in triplicate, and results of 3 independent experiments were used for statistical analysis. Relative mRNA expression levels were quantified using the $\Delta\Delta C(t)$ method [47]. Data are presented as means ±standard error (s.e.). Statistical comparisons were carried out using unpaired t- test and values of p < 0.05were considered to be statistically significant. Primer sequences can be found in Tables 1-3.







9. Western blot analysis and antibodies

Cells were lysed in RIPA buffer [50mM Tris-HCl (pH 7.5), 150mM NaCl, 1% Nonidet P-40, 0.5% sodium deoxycholate, 0.1% sodium dodecyl sulfate (SDS)] containing ethylenediaminetetraacetic acid (EDTA)-free protease inhibitor cocktail (Roche, Basel, Switzerland). Equal amounts of proteins were then resolved on 8–15% SDS-PAGE gels, followed by electrotransfer to polyvinylidene difluoride membranes (Millipore, Bedford, MA, USA). The membranes were blocked for 1 h in TBS-T [10mM Tris-HCl (pH 7.4), 150 mM NaCl, 0.1% Tween 20] containing 5% fat-free milk at room temperature and then incubated with the indicated primary antibodies overnight at 4°C. After incubation for 2 h with peroxidase-conjugated secondary antibodies appropriate [1:4000, Jackson ImmunoResearch Inc., West Grove, PA. USA], developed using enhanced chemiluminescence detection system. The following antibodies were used in this study; anti-mouse p53 (FL-393, Santa Cruz Biotechnology, Santa Cruz, CA, USA), anti-human p53 (DO-1, Santa Cruz Biotechnology), anti-53bp1 (Origene, Rockville, MD, USA), anti-Acl (Cell Signaling, Beverly, MA, USA), anti-mouse Slbp (H-10, Santa Cruz





Biotechnology), anti-human Slbp (EPR12673, Abcam, Cambridge, MA, USA), anti-Cpsf3 (Bethyl, Montgomery, TX, USA), anti-Lsm10 (Origene), anti-Lsm11 (Bethyl), anti-Symplekin (Bethyl), anti-HistoneH1 (Abcam, Cambridge, MA, USA), anti-HistoneH2a (Abcam), anti-HistoneH2b (Abcam), anti-HistoneH3 (Abcam), anti-HistoneH4 (Abcam), anti-α-tubulin (Neomarkers, Fremont, CA, USA) and anti-β-actin (Santa Cruz Biotechnology).

10. Cell cycle analysis

Trypsinized cells were washed twice with PBS and fixed in 70% ethanol. The fixed cells were washed with PBS, incubated with 100 mg/ml RNase at 37°C for 30 min, stained with PI (50 mg/ml) and analyzed on FACSCalibur flow cytometer (BD Biosciences, San Jose, CA, USA). The percentages of cells in different phases of the cell cycle were analyzed using the Cell Quest Pro (BD Biosciences).





11. Bromodeoxyuridine (BrdU) incorporation assay

BrdU incorporation was using the BrdU (colorimetric) kit (Roche Diagnostics, Germany) according to the manufacturer's instructions.

12. Chromatin immunoprecipitation (ChIP) assay

ChIP was performed using a commercially available SimpleChIP Assay Kits (Cell Signaling) according to manufacturer's instructions. DNA-bound protein was immunoprecipitated using an anti-acetyl-Histone H3 (Millipore, Billerica, MA, USA) antibody or rabbit IgG (Active Motif, Rixensart, Belgium) as a negative control. For quantification of co-precipitated DNA, samples were then subjected to amplification by employing primers (mSlbp forward: 5'-TTCACTCGGCAACAGCACT-3', mSlbp reverse: 5'hSlbp 5'-GTCTCATGGTTTCGCCCTTC-3', forward: GCGACTGCGCTCAGATTAAC-3', hSlbp reverse: 5'-CGCTTTTTCTGCGTAGCCTG-

3', mCpsf3 forward: 5'-ATAACGTCGCCAAGATGCTC-3', mCpsf3 reverse: 5'-





GCTTTGGAAGCCTGATCTTG-3', hCpsf3 forward: 5'-CGCAGTCCTGACGTCCTAC-

| 3', h | Cpsf3 | reverse: | 5'-AAGGA | AGAACO | CCCATTC | ACC-3', | mLsm10 | forward: | 5'- |
|-------|-------|----------|-----------|--------|---------|---------|----------|----------|-----|
| GAG | GGCGT | CAGATO | CTCGTTA3' | , | mLsm1 | 0 | reverse: | | 5'- |
| ACG | FGGTG | GCAAA | СТССТТА-3 | , , | hLsm1 | 0 | forward | : | 5'- |
| AGA | GACAC | GGTCT | CCCCATT-3 | °, 8 | and | hLsm10 | reve | erse: | 5'- |

CATGAAGCCAAGCTTGTGAA-3') which amplified the promoter region of each gene.





RESULT

1. Deficient of p53 induces global mRNA 3' UTR alternative polyadenylation

To determine the role of p53 in mRNA 3' UTR-APA on a genome-wide level, we performed RNA sequencing (RNA-Seq) to compare APA events in p53^{-/-} and p53^{+/+} MEFs [48]. The RNA-Seq results show, as expected, that p53^{-/-} promoted APA in some mRNA 3' UTR (Figure 1). Based on the RNA-Seq results, there are several patterns in APA event. For example, several genes in p53^{-/-} MEF cells showed decreased signal (Ocel1 and Gdf10) (Figure 1A) or increased signal (Gng2 and Igfbp5) (Figure 1D) in the 3' most exon, although the signal from upstream exons increased. Tsc22d2 and Trmt5 showed decreased signal (Figure 1B) and Elk1 and Slc30a4 (Figure 1E) increased signal in 3' most exon, however, upstream exons signal was similar. Aen and Alcam showed decreased both upstream exons signal and 3' most exon signal (Figure 1C). On the other hand, Scoc and Dpt have increased both signals (Figure 1F). As the signals from upstream exons reflecting the amount of transcripts varied among the 3' UTR-shortened and lengthened transcripts, we analyzed whether 3' UTR-APA in the transcriptome of p53^{-/-} MEF cells correlates with





differential gene expression. To this end, we enriched 6,779 transcripts in our data set that are eligible for combined analysis of 3' UTR-APA and differential expression. Next, each transcript was plotted by fold changes in the differential gene expression (y-axis in Figure 1G) and the significance of 3' UTR shortening (x-axis in Figure 1G). This approach identified 793 3' UTR-shortened transcripts (Table 4) or 3' UTR-lengthened transcripts (Table 5) (about 11.7%) out of 6,779 transcripts in p53^{-/-} MEF cells. Although 7% (56/793) of the 3' UTR shortened transcripts either increased (16/793) or decreased (40/793) their expression level, 6.7% (53/793) of the 3' UTR lengthened transcripts either increased (22/793) or decreased (31/793) their expression level. A significant proportion (88.3%) of them in the p53^{-/-} MEF transcriptome remained unchanged, indicating no significant correlation between the differential gene expression (DEG) and the 3' UTR-APA in the p53^{-/-} MEF transcriptome.









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Figure 1. p53 knockout induces global mRNA 3' UTR alternative polyadenylation

A-F. RNA-seq reads from p53^{+/+} and p53^{-/-} MEFs are aligned to mouse genome mm10 RefSeq. Representative examples of transcripts with 3' UTR ApA are presented. Annotated gene structures are at the bottom of the alignment. **G.** Scatter plot of RNA-seq data. Red dots represent individual transcripts in the analysis. Horizontal blue dashed lines represent the cutoff values for two-fold changes in differential gene expression. Vertical green dashed lines represent the cutoff values for $-\log_{10}$ (P-value) of 3' UTR shortening (1.3 corresponds to P=0.05) in p53^{-/-} and p53^{+/+} MEFs, which was determined by χ^2 -test. The green boxes highlight the aligned reads in 3' UTRs.





2. p53 regulates 3' UTR alternative polyadenylation

To validate these RNA-Seq results, we analyzed APA using the 'relative shortening index (RSI)' which was previously developed a method to determine the increase or decrease in the level of the short isoform relative to the long isoform [1]. We generated pairs of primers were used to detect a total (short + long) or a long-specific transcript (Figure 2A) and RT-qPCR validation of the 3' UTR-APA of randomly selected genes showed APA changes consistent with the RNA-Seq dataset (Figure 2B). We also examined the change in expression level of 3' UTR polyadenylation and splicing factors. Interestingly, most of 3' UTR polyadenylation were up-regulated in p53^{-/-} MEF cells except Cpeb1 (Figure 2C), and U4 and 7sk splicing factor were up-regulated, however, other splicing factors were no significantly changed (Figure 2D). Collectively, these data suggest that p53 knockout in cells leads to expression change of 3' UTR polyadenylation factors and transcriptome-wide APA in the 3'-most exon of mRNAs.

To show a direct function of p53 in APA, p53^{-/-} MEF cells were transfected with fulllength p53 expression construct. We test shortened transcripts and lengthened transcripts in





p53^{-/-} MEF cells. As shown in Figure 3A, overexpression of p53 resulted in the length of all tested transcripts are recovered. We also checked 3' UTR polyadenylation, most of the factors, we tested, are recovered except Cpsf3 and Cpeb1. These results suggested that p53 regulates APA via several 3' UTR polyadenylation factors through an unknown mechanism(s).







Figure 2. p53 regulation of mRNA alternative polyadenylation

A. A schematic presenting primer sets for RT–qPCR. Pairs of primers were used to detect a total (short + long) or a long-specific transcript. The RSI was calculated to determine the 3' UTR ApA in a target cell line by RT–qPCR. **B.** Validation of RNA-seq data. Error bars represent s.e. from three repeats of experiments. Student's t-tests are done for statistical significance (P<0.05). **C.** RT-qPCR analysis of polyadenylation factor mRNA levels in p53^{-/-} MEF cells were expressed relative to p53^{+/+} MEF cells. **D.** RT-qPCR analysis of splicing factor mRNA levels.







Figure 3. Introduction of wild-type p53 leads to restoration of alternative

polyadenylation

A. The RSI was measured using total RNAs isolated from $p53^{-/-}$ MEF cells with p53 overexpression (P<0.05). B. RT-qPCR analysis of polyadenylation factor mRNA levels in p53 over-expressed p53^{-/-} MEF cells were expressed relative to p53^{+/+} MEF cells.





3. Knockout of 53bp1 leads to global mRNA 3' UTR lengthening

To determine the role of 53bp1 in mRNA 3' UTR-APA on a genome-wide level, we performed RNA-Seq to compare APA events in 53bp1^{-/-} and 53bp1^{+/+} MEFs. The RNA-Seq results show, as expected, that 53bp1^{-/-} promoted APA in some mRNA 3' UTR (Figure 3). Based on the RNA-Seq results, there are several patterns in APA event similar as p53^{-/-} MEF cells. For example, several genes in 53bp1^{-/-} MEF cells showed decreased signal (Ctss and Lum) (Figure 4A) or increased signal (Klhl24 and Gpf64) (Figure 4D) in the 3' most exon, although the signal from upstream exons increased. Kdma3a and Hypk showed decreased signal (Figure 4B) and Prrc1 and Reep5 increased signal in 3' most exon (Figure 4E), however, upstream exons signal was similar. Pole and Kif23 showed decreased both upstream exons signal and 3' most exon signal (Figure 4C). On the other hand, Anp32e and Krt19 have increased both signals (Figure 4F). As the signals from upstream exons reflecting the amount of transcripts varied among the 3' UTR-shortened and lengthened transcripts, we analyzed whether 3' UTR-APA in the transcriptome of 53bp1^{-/-} MEF cells correlates with differential gene expression. To this end, we enriched 5,674 transcripts in




our data set that are eligible for combined analysis of 3' UTR-APA and differential expression. Next, each transcript was plotted by fold changes in the differential gene expression (y-axis in Figure 4G) and the significance of 3' UTR shortening (x-axis in Figure 4G). This approach identified 483 3' UTR-shortened transcripts (Table 6) or 3' UTR-lengthened transcripts (Table 7) (about 8.5%) out of 5,674 transcripts in 53bp1^{-/-} MEF cells. As contrasted with p53^{-/-} MEF cells, 3' UTR lengthened transcripts (60.7%, 293/483) were more than shortened transcript (39.3%, 190/483) in 53bp1^{-/-} MEF cells. Although 4.7% (23/483) of the 3' UTR shortened transcripts either increased (6/483) or decreased (17/483) their expression level, 4.7% (23/483) of the 3' UTR lengthened transcripts either increased (15/483) or decreased (8/483) their expression level. A significant proportion (90.6%) of them in the 53bp1^{-/-} MEF transcriptome remained unchanged, indicating no significant correlation between the differential gene expression (DEG) and the 3' UTR-APA in the 53bp1^{-/-} MEF transcriptome.













Figure 4. 53bp1 knockout induces global mRNA 3' UTR alternative polyadenylation

A-F. RNA-seq reads from 53bp1^{+/+} and 53bp1^{-/-} MEFs are aligned to mouse genome mm10 RefSeq. Representative examples of transcripts with 3' UTR ApA are presented. Annotated gene structures are at the bottom of the alignment. **G.** Scatter plot of RNA-seq data. Red dots represent individual transcripts in the analysis. Horizontal blue dashed lines represent the cutoff values for two-fold changes in differential gene expression. Vertical green dashed lines represent the cutoff values for $-\log_{10}$ (P-value) of 3' UTR shortening (1.3 corresponds to P=0.05) in 53bp1^{-/-} and 53bp1^{+/+} MEFs, which was determined by χ^2 -test. The green boxes highlight the aligned reads in 3' UTRs.





5. 53bp1 controls 3' UTR alternative polyadenylation

To validate these RNA-Seq results, we analyzed APA using the RSI method to determine the increase or decrease in the level of the short isoform relative to the long isoform same as p53^{-/-} MEF cells. RT-qPCR validation of the 3' UTR-APA of randomly selected genes showed APA changes consistent with the RNA-Seq dataset (Figure 5A). We also examined the change in expression level of 3' UTR polyadenylation and splicing factors. Interestingly, on the contrary to p53^{-/-} MEFs, most of 3' UTR polyadenylation factors were down-regulated except Cpeb1 (Figure 5B) and all of the tested splicing factors were upregulated in 53bp1^{-/-} MEF cells (Figure 5C). These results demonstrate that 53bp1 knockout in cells leads to the change of 3' UTR polyadenylation factor levels and transcriptome-wide APA in the 3'-most exon of mRNAs, contrariwise p53 knockout.







Figure 5. 53bp1 regulation of mRNA alternative polyadenylation

A. Validation of RNA-seq data. The RSI was calculated to determine the 3' UTR ApA in a target cell line by RT–qPCR. Error bars represent s.e. from three repeats of experiments. Student's t-tests are done for statistical significance (P<0.05). **B.** RT-qPCR analysis of polyadenylation factor mRNA levels in 53bp1^{-/-} MEF cells were expressed relative to 53bp1^{+/+} MEF cells. **C.** RT-qPCR analysis of splicing factor mRNA levels.







6. Knockout of 53bp1 results in increase of replication-dependent histone mRNA polyadenylation

We performed the differential gene expression (DEG) analysis using the differentially expressed transcripts in 53bp1^{-/-} MEF cells (Figure 6A). The DEG analysis result showed that most replication-dependent histone transcripts were increased (Figure 6B). However, we performed western blot for detect histone protein expression levels. Interestingly, all of the histone protein expressions, we tested, were decreased (Figure 6C). Our RNA-seq data only represented polyadenylated RNAs; therefore, we hypothesize that decrease of histone proteins by histone 3' UTR polyadenylation. To confirm our hypothesis, we performed RTqPCR validation of 3' UTR polyadenylation of two well-known histone genes (Hist1h2ac and Hist1h2bg). During reverse transcription, random hexamers prime randomly along the RNAs, whereas oligo(dT) primers only prime at polyadenylated mRNA. Thus, utilizing PCR amplifications using cDNAs resulting from random hexamers and oligo(dT) primers, we were able to measure the relative amount of total mRNA and polyadenylated mRNA. RT-qPCR results show, polyadenylated histone transcripts were increased, although total





histone transcripts were decreased (Figure 6E). These results support the hypothesis that

53bp1 knockout down-regulate histone expression via histone 3' UTR polyadenylation.







Figure 6. Expression of the polyadenylated histone genes in 53bp1^{-/-} MEF cells





A. Scatter plots confront 53bp1^{+/+} MEF and 53bp1^{-/-} MEF RNA-seq data on the *x* and *y*-axes, respectively. Absolute expression levels (53bp1^{+/+} MEF cells versus 53bp1^{-/-} MEF cells RNA-seq FPKM units). The red dot indicates higher expressed transcript in 53bp1^{+/+} MEF cells. A black dot indicate and blue dot indicate higher expressed transcript in 53bp1^{+/+} MEF cells. A black dot indicates unchanged transcript. **B.** Individual histone transcript level in 53bp1^{-/-} MEF RNA-seq data compared with 53bp1^{+/+} MEF. **C.** Western blot analysis of histones in 53bp1^{+/+} and 53bp1^{-/-} MEFs. **D.** A schematic presenting primer sets for RT–qPCR. Pairs of primers were used to detect a total (short + polyadenylated) or a polyadenylated-specific transcript. **E.** RT-qPCR analysis of 3' UTR polyadenylated histone mRNAs in 53bp1^{+/+} and 53bp1^{-/-} MEFs (P<0.05).





7. Expression of Slbp is decreased in 53bp1 knockout MEF cells

We next examined which histone 3' UTR processing factor affect histone polyadenylation. To performed screening a subset of histone 3' UTR processing factors by RT-qPCR and western blot analysis. The RT-qPCR result shows that only Slbp mRNA expression level is significantly down-regulated in 53bp1^{-/-} MEF cells. However, other histone 3' UTR processing factors such as Lsm10, Lsm11, Cpsf3, and U7 were no significant change (Figure 7A). Only Sympk was up-regulated. We also checked protein expression level of histone 3' UTR processing factors by western blot. Only Slbp protein expression is decreased in 53bp1^{-/-} MEF cells same as an RT-qPCR result (Figure 7B). Other histone 3' UTR processing factors were unchanged, even though mRNA level was increased. Previous research shows that Slbp expression has an effect on cell cycle, and when cells enter S phase, Slbp is synthesized and rapidly degrades at the end of S phase [49-51]. Accordingly, we performed cell cycle analysis and did not show significant cell cycle change, also confirmed no change S phase accumulation by BrdU incorporation assay (Figure 7C). These results proved that 53bp1 regulate histone polyadenylation via





Slbp expression and without change cell cycle.







Figure 7. Slbp protein levels are decreased in 53bp1^{-/-} MEF cells

A. RT-qPCR analysis of histone 3' UTR processing factors in $53bp1^{+/+}$ and $53bp1^{-/-}$ MEFs (P<0.05). B. Western blot analysis of histone 3' UTR processing factors in $53bp1^{+/+}$ and





53bp1^{-/-}MEFs. **C.** Cell cycle analysis was performed on 53bp1^{+/+} and 53bp1^{-/-}MEFs (Left panel). Quantification of cells in G0/G, S-phase (intermediate), and G2 of the cell cycle is shown in the each histogram. Quantification of BrdU incorporation in 53bp1^{+/+} and 53bp1^{-/-}MEFs (Right panel).





8. Replication-dependent histone mRNA polyadenylation is decreased in 53bp1 depleted human cells

To further investigated whether suppression of 53bp1 in the human cell line causes replication-dependent histone mRNA polyadenylation, histone 3' UTR polyadenylation and protein expression level were analyzed by RT-qPCR and western blot analysis using 53bp1 depleted HeLa cells. The depletion of 53bp1 with siRNA led to increased level of polyadenylated histone mRNAs (Hist1h2ac and Hist1h2bd) and decreased protein level of histones same as MEF cells (Figure 8A, B). In addition, we measured histone 3' UTR processing factors using RT-qPCR and western blot analysis. The demonstrated that depletion of 53bp1 result showed also decreased only Slbp mRNA and protein level (Figure 8C, D). We performed cell cycle analysis and BrdU incorporation assay did not show significant changes of S phase same as 53bp1^{-/-} MEF cells (Figure 8D). These results showed that 53bp1 regulate histone polyadenylation via Slbp expression is not only MEFs specific phenomenon but also human cells.







Figure 8. Replication-dependent histone mRNA polyadenylation decreased in 53bp1

depleted cells

A. RT-qPCR analysis of 3' UTR polyadenylated histone mRNAs in 53bp1 depleted HeLa cells (P<0.05). **B.** Western blot analysis of histone in 53bp1 depleted HeLa cells. **C.** RT-





qPCR analysis of histone 3' UTR processing factors in 53bp1 depleted HeLa cells (P<0.05). D. Western blot analysis of histone 3' UTR processing factors in 53bp1 depleted HeLa cells. E. Cell cycle analysis was performed on 53bp1 depleted HeLa cells (Left panel). Quantification of cells in G0/G, S-phase (intermediate), and G2 of the cell cycle is shown in the each histogram. Quantification of BrdU incorporation in 53bp1 depleted HeLa cells (Right panel).





9. Slbp regulates histone polyadenylation

Recent reports showed that Slbp regulates histone polyadenylation [30, 33-35, 50]. We transiently knockdown Slbp on HeLa cells for confirming that histone polyadenylation. We performed RT-qPCR to check histone polyadenylation of two histone genes (Hist1h2ac and Hist1h2bd). RT-qPCR results showed that decreased total histone mRNA expression and increased polyadenylated histone expression same as previously reported (Figure 9A) [35, 51-53]. We also tested other histone 3' UTR processing factors mRNA level. RT-qPCR results show that Lsm10 and Cpsf3 are no significant changed; however, Lsm11 and Sympk mRNA level are increased (Figure 9B), however in western blot analysis, no significant change of histone 3' UTR processing factors even though RNA level were changed, and decreased histone expression level (Figure 9C).







Figure 9. Slbp regulates histone polyadenylation

A. RT-qPCR analysis of 3' UTR polyadenylated histone mRNAs in Slbp depleted HeLa cells (P<0.05). **B.** RT-qPCR analysis of histone 3' UTR processing factors in Slbp depleted HeLa cells (P<0.05). **C.** Western blot analysis of histone 3' UTR processing factors and histone in Slbp depleted HeLa cells.





10. Slbp controls 53bp1 dependent histone polyadenylation

To elucidate whether the expression of Slbp is dependent on 53bp1, 53bp1^{-/-} MEF cells and 53bp1 depleted HeLa cells were transfected with siRNA-resistant full-length 53bp1 expression construct and measuring Histone 3' UTR processing factors by RT-qPCR and western blot analysis. As expected, overexpression of full-length 53bp1 in both 53bp1^{-/-} MEF cells and 53bp1 depleted HeLa cells led to increased Slbp mRNA expression (Figure 10A and Figure 11A) and expression of Slbp protein recovered (Figure10B and Figure 11B). Also, RT-qPCR and western blot analysis result showed that polyadenylated Histone mRNAs are decreased and protein expression level of histones is recovered (Figure 10B, C and Figure 11B, C). These results support that histone expression regulated by 53bp1 via Slbp.







Figure 10. Slbp expression depends on 53bp1 in MEF cells

A. Histone 3' UTR processing factor was measured using total RNAs isolated from 53bp1⁻ ^{/-} MEF cells with 53bp1 overexpression by RT-qPCR (P<0.05). **B.** Western blot analysis of histones and histone 3' UTR processing factors in 53bp1 overexpressed 53bp1^{-/-} MEF cells.





C. RT-qPCR analysis of 3' UTR polyadenylated histone mRNAs in 53bp1 overexpressed

53bp1^{-/-} MEF cells (P<0.05).







Figure 11. 53bp1 regulate Slbp expression in HeLa cells

A. Histone 3' UTR processing factor was measured using total RNAs isolated from 53bp1 depleted HeLa cells with 53bp1 overexpression by RT-qPCR (P<0.05). **B.** Western blot analysis of histones and histone 3' UTR processing factors in 53bp1 overexpressed 53bp1 depleted HeLa cells. **C.** RT-qPCR analysis of 3' UTR polyadenylated histone mRNAs in





53bp1 overexpressed 53bp1 depleted HeLa cells (P<0.05).





11. 53bp1 regulates expression of Slbp through histone acetylation

53bp1 has a new function which regulates most histone acetylation binding with Acl and acetvlation of histones is decreased in the 53bp1^{-/-} MEF cells (not yet published). To examine whether Slbp expression is regulated by Acl, we performed histone 3' UTR processing factors and histone polyadenylation RT-qPCR. Acl knockdown HeLa cells showed that decrease Slbp mRNA expression same as a 53bp1 knockdown, however, other histone 3' UTR processing factors were not a correlation between Acl knockdown and 53bp1 knockdown (Figure 12A). Histone polyadenylation RT-qPCR showed that Acl depleted HeLa cells were increased histone polyadenylation similar as 53bp1 depleted HeLa cells (Figure 12B). In western blot analysis, only Slbp protein was decreased both 53bp1 and Acl depleted cells and no significant change of other histone 3' processing factors. Also, histone protein expressions were decreased in Acl knockdown cells same as 53bp1 knockdown cells (Figure 12C). Histone acetylation is generally associated with activation of transcription via the neutralization of the lysine residues, consequently reducing the affinity of histones for DNA and opening the chromatin [54-56]. It has been





reported that histone acetylation correlates with transcriptional activation and that histone deacetylation correlates in many cases with repression in yeast [57]. To examine whether Slbp expression is regulated by histone acetylation on its promoter, we performed total acetyl histone H3 ChIP assay for Slbp, Cpsf3, and Lsm10 promoter. As the ChIP assay result, total acetyl histone H3 enrichment is dramatically decreased on Slbp promoter region of 53bp1^{-/-} MEF cells and 53bp1 depleted HeLa cells, whereas others do not (Figure 12D). These data suggested that decrease of histone acetylation on Slbp promoter causes reduction of Slbp expression by novel function of 53bp1.







- 51 -





Figure 12. Slbp promoter activation requires 53bp1

A. Histone 3' UTR processing factor was measured using total RNAs isolated from Acl depleted HeLa cells by RT-qPCR (P<0.05). **B.** RT-qPCR analysis of 3' UTR polyadenylated histone mRNAs in Acl depleted HeLa cells (P<0.05). **C.** Western blot analysis of histone 3' UTR processing factors and histone in Acl depleted HeLa cells. **D.** ChIP assays of total acetyl histone H3 in 53bp1^{-/-} MEF cells and 53bp1 depleted HeLa cells. **RT-qPCR** primers located within the promoter region on Slbp, Cpsf3, and Lsm10.





DISCUSSION

In this study, we have utilized genetically defined mouse embryonic fibroblasts to examine the growth regulatory properties of the molecular signatures of p53 or 53bp1 knockout transcriptome and discovered widespread 3' UTR-APA. We showed that knockout of p53 or 53bp1 read to global APA change by RNA-seq and RT-qPCR. These are a novel function for APA regulation of p53 and 53bp1. Although an explicit mechanism of how p53 or 53bp1 knockout leads to the 3' UTR-APA in selected transcripts is unknown. We found that almost known 3' end polyadenylation factors alter their expression on changes in $p53^{-/-}$ MEF cells or $53bp1^{-/-}$ MEF cells compared with wild-type MEF cells. Recent reports showed that embryonic development is associated with reprogramming of APA, which is correlated with the change in the set of genes encoding the polyadenylation factor level [2, 58]. This observation indicated that 3' UTR-APA is likely caused by expression or activity change of mRNA 3' UTR polyadenylation factors. Similarly, our analysis identified expression change of multiple 3' UTR polyadenylation factors in p53^{-/-} or 53bp1^{-/-} MEFs. These suggested that p53 or 53bp1 mediated 3' UTR-APA may occur by





multiple 3' UTR polyadenylation factors. p53 is a well-known transcription factor. However, there are no known p53 regulatory elements in the promoter region of 3' UTR polyadenylation factors. Therefore, p53 may indirectly regulate 3' UTR polyadenylation factors. Previously, De novo motif discovery analysis result showed many promoters of key 3' UTR polyadenylation factors to be bound by the transcription factor, E2f family and E2f enhance APA event and cell proliferation [11]. E2f has nonreciprocal functional interactions with p53 [59]. Summarize of these results, the intricate interactions between E2f and p53 is one of the potential mechanisms. p53 regulate embryonic development and differentiation to diverse environmental stresses [60]. 3' UTR-APA events associated with both proliferation [12] and oncogenic transformation [4]. Recently, Fu et al. [61] found opposing pattern of 3'UTR-APA between transformed breast cancer cell lines and nontransformed cells, further indicating the APA regulation depend on cellular context over the effect of cell proliferation, differentiation and transformation states [11]. Previous studies that reported tumorigenesis and embryonic development have a direct link and relevance to each other [62-65]. The sum of these results, p53 and its binding protein, 53bp1, may regulate cell proliferation, differentiation, and tumorigenesis through a new pathway, 3'



UTR-APA, which is not yet known. Further studies are required to understand how p53 and 53bp1 regulate 3' UTR-APA via 3' UTR polyadenylation factors.

RNA-seq analysis of 53bp1^{-/-} MEF cells displayed a remarkable increase in replicationdependent histone mRNAs. This is the first report 53bp1 knockout increase the expression of the replication-dependent histone genes on a large scale. We showed that the increase in replication-dependent histone gene expression was at least in part due to the aberrant polyadenylation of replication-dependent histone mRNAs. Histone modification and mRNA 3' end formation are necessary for normal cell cycle progression and maintaining genomic stability [33, 53, 66-68]. In this report, we were screening histone 3' UTR processing factors in 53bp1^{-/-} MEF cells and 53bp1 depleted cells by RT-qPCR and western blot analysis. As these results, we identified 53bp1 works to deplete Slbp at the mRNA and protein levels. Several studies have shown the Slbp is a key component of the machinery processing histone mRNA 3' ends and functions in the initiation of cap-dependent translation of histone mRNA [52, 69, 70]. Transient recovery of 53bp1 in 53bp1^{-/-} MEF and 53bp1 depleted cells showed that both Slbp and Histone 3' UTR processing are





recovered. These results revealed that 53bp1 is an up-stream regulator of Slbp. 53bp1 regulate gene expression acts as a transcriptional co-activator of p53 [22] and modulate histone acetylation with Acl, epigenetically (not yet published). The data from total acetyl histone H3 ChIP assay showed that knockout or depletion of 53bp1 decrease acetyl histone H3 on Slbp promoter. This result suggested that decreasing Slbp expression in 53bp1 depleted cells induced by epigenetic changes at the Slbp promoter. These events subsequently lead to an increase in polyadenylated canonical histone mRNA and decrease in histone protein levels. The tight regulation of histone expression is critical for cell viability and chromatin maintains [42]. A recent report showed that decrease histone level by knockdown or mutation of Slbp cause genomic instability and impaired cellular proliferation in Drosophila [53]. 53bp1 is a key transducer of the cell cycle checkpoint control and DNA damage response [71]. It also highly involved in maintaining genomic stability via multiple pathways [71-76]. Our data suggest that 53bp1 regulates histone expression by epigenetic regulation of Slbp, which is a novel and unexpected function of 53bp1 in genome stability pathway.





ABSTRACT

Genomewide characterization of messenger RNA alternative

polyadenylation

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Messenger RNAs (mRNAs) have to go through a series of post-transcriptional processing steps prior to translation. One of the post-transcriptional steps is 3' end processing, which is composed of cleavage and polyadenylation, is important for delimiting the 3' untranslated region (3' UTR) of mRNA and determining the regulatory elements for downstream post-transcriptional/ translational regulation. 3' UTR processing is very flexible because of the variety of trans-acting and cis-acting elements at the 3' end





of the mRNA. In recent years, experimental and computational studies have revealed the differential use of alternative polyadenylation (APA) sites of the same gene leading to different 3 'UTR mRNA. More importantly, overall changes in 3 'UTR length is observed in cancer cells. Another important polyadenylation phenomenon is histone polyadenylation. As contrasted with other metazoan mRNAs, replication-dependent histone transcripts are not polyadenylated and alternatively have a conserved stem-loop structure at the 3'end. Recently, histone polyadenylation increase sensitivity to DNA damage as well as increase the frequency of missing chromosomes and induce genomic instability. In this study, we use well-known Tumor protein p53 and p53 Binding protein 53bp1, which are play a role in tumorigenesis, apoptosis, and genomic stability, for analyze ApA. We identify 3' UTR alternative polyadenylation of mRNAs as an additional molecular signature of p53 or 53bp1. Also, 53bp1 is key-molecule to regulate histone polyadenylation. Together, these findings identify a previously uncharacterized role for p53 or 53bp1 in the modulating 3' UTR length of mRNAs.





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Forward for **Reverse for** Forward for **Reverse for** Gene total total long long name mRpl22 TGGGCATC AAGTTCAC GTGATCTT GCTTGTTGCA CCTGGACTGC GCTCTTGCTG TGGGCTTTTA GACTTGCTCA AC CG GG mTimp2 GACTTCAT CCATCCAG ACCCAGAG CTGCCTGGTG TGTGCCCTGG AGGCACTCAT TGGAATGCAT TGGCTAGAAA GA CC GG mTomm20 GAGAGCTG GGTCGGAA GGCTTTGG GTTTGCAGGC GGCTTTCCAA GCTTGGTCAG CACTGTCACT TGGTTCAAGG GT ΤG AA mAlcam GGAGCGGATT TCCTGTGACA CTCACTCAAG ATAGGGGGTG GGATAGCCTG GCTTGGTAGA AGCACCTCCA AACCGTGAA С mKras' AGACACGA AGAAGGC GTTAGCTC ATTCCCTAGG CAGTGCCCCA AACAGGCTCA ATCGTCAACA TCAGCGCAAC GG CCC AT **CCACTGGA** TACAGCAA TCATCCCAGT mRan TGGGCTTC GAGTGAATGT TATTGCGATG GCCATTCCGT CAGGGGAGA GG GT G GGA mCamk2d TGTCAACAGT AAGAGGAGA GAGCGTTATT GAGCTCCCCA GCCACTTCTT GGACGTCCCA GGGCATGCTG TGGTAAAGCA С G mCol11a1 TATGATGGCT AAGCAAGCT AGAATTCTGT AGTTCCAAAG GTGCGTCTCG GGACCGACTT AACCCTTGCA TCAAAAGTAT

Table 1. Primer sequences for alternative polyadenylation





| | | C | TGT | GAGAC |
|----------|------------|------------|------------|------------|
| mNcam1 A | AACCACAAA | AAGCTCAGA | GTGGGAAGG | GGCCCCCGA |
| (| CCCTTCCCAG | GGCTTGTCCA | TTACCCAGGT | AGTACAGAAT |
| (| Ĵ | С | G | G |
| mElk1 C | GAGATCACCC | GTGCCTGAAG | AGGGTTGTGC | ACACAAACCA |
| I | AACCGCAGA | ACCAGAGCTT | AAAGCAAGT | TTTCTCAAAA |
| I | A | | G | CCAT |
| mUbe2n A | ATGAGAGC | TTTAGGTG | TGTGGACA | AGCTCCTTAA |
| I | AACGCCCGTT | CTGCCATTGG | GTTGGTTGGC | AAGGCGGCAT |
| | АT | GT | TT | |

Table 2. Primer sequences for confirming the expression of genes

| Gene | Forward | Reverse | |
|---------|-----------------------|----------------------|--|
| name | Forward | | |
| mCpsf1 | ACATACCGACGCTTGCTGAT | TAGCGGTTTAGCAGTTCCCC | |
| mCpsf2 | CGGAATTTGTAGGGGGGCGTA | ATCCGATGCGTCCAGTTTCT | |
| mCpsf3 | GCACGTTTACAGCAAGAGGC | TTCTACAGCCCGAGTCTCCA | |
| mCpsf4 | GCACCCTCGATTTGAACTGC | CTGCATGACCCCAATGACCT | |
| mCpsf5 | AAGCCTTGTTTGCAGTCCCTA | AATGATGGGTCCATACCCCG | |
| mCpsf6 | TCACGGGAAAAGAGTCGTCG | CGGTATTCTCGCTCTCGGTC | |
| mCpsf7 | AAACGAATACCTCCACGGGC | GGCTTATCCACACGAGCAGA | |
| mCpeb1 | CAGCAGGACGGTGTTTGTTG | TCACACGACCAGACCCAATG | |
| mPabpn1 | GCCCGCTCTATCTACGTTGG | ACACGGTTGACTGAACCACA | |
| mCstf1 | GACGCTGGTGAGGTACACAG | CAGGCTGATGGTCCTTTCGT | |





| mCstf2 | CAAGTGCCAATGCAAGACCC | TCTCATGTCATGTGGAGGCG | |
|--------|------------------------|-------------------------|--|
| mCstf3 | ACTGTTGAGGAAGCCGTGAG | GGGACTACAGCCCCCTTTTC | |
| U1 | GATACCATGATCACGAAGGTG | CACAAATTATGCAGTCGAGTTT | |
| | GTT | CC | |
| U2 | TTTGGCTAAGATCAAGTGTAGT | AATCCATTTAATATATTGTCCTC | |
| | ATCTGTTC | GGATAGA | |
| U4 | GCGCGATTATTGCTAATTGAAA | AAAAATTGCCAATGCCGACTA | |
| U6 | GCTTCGGCAGCACATATACTAA | ACGAATTTGCGTGTCATCCTT | |
| | AAT | | |
| 5s | CGGCCATACCACCCTGAAC | GCGGTCTCCCATCCAAGTAC | |
| 5.8s | CGGCTCGTGCGTCGAT | CCGCAAGTGCGTTCGAA | |
| 7sk | CACCCCATTGATCGCCAGGGTT | TTGACCGAAGACCGGTCCTCCT | |
| | GA | СТ | |
| mU7 | AAGTGTTACAGCTCTTTTAGAA | AGGGGTTTTCCGACCGAAGTC | |
| | TTTGT | AGA | |
| hU7 | CAGTGTTACAGCTCTTTTAGA | AGGGGCTTTCCGGTAAAAAGC | |
| | TTTG | | |
| mSlbp | TCTGGAAGGTGGCTTTGCAT | GTGCCAGCATACACATCAAAGT | |
| | | Т | |
| hSlbp | AAGTCCCAAGACACCTTCGAC | TCACATCCTTCTTCCGCTGG | |
| mLsm10 | CGGTGAAGGAGCGAACTATTT | CTCGGGCCACACTCTCATC | |
| | | | |





| hLsm10 | GGCATGGTTTCTCCGTCCT | CCTGCTTGCACGCGACATT |
|--------|------------------------|-------------------------|
| mLsm11 | GCAATGTGCTTACGCGGATG | TGAAGGTGCGGATGTGAACG |
| hLsm11 | TGGGTGAACTCCATCGCTGTA | GTCGAATGCAACAAGGAAGCC |
| mSympk | CGGAGTGTGGCATCACAGTTT | CGCACTTCAATGGATTTGTCTG |
| hSympk | GAGATCATCGCATTCCAAGCA | TCACATTCTCGTCCCTCAAGAG |
| m53bp1 | CCTCCTTTGCCTTCAGTGAG | TGCTTGTCATCAGCCACTTC |
| h53bp1 | ATTGAGGATACGGAACCCATGT | TGCTGGATTCATCAGGATACTAT |
| | | СА |
| Acl | TGGATGCCACAGCTGACTAC | GGTTCAGCAAGGTCAGCTTC |
| mp53 | TGAACCGCCGACCTATCCTTA | GGCACAAACACGAACCTCAAA |
| Gapdh | TGCACCACCAACTGCTTAGC | GGCATGGACTGTGGTCATGAG |

Table 3. Primer sequences for histone polyadenylation

| Concentration | Forward for | Reverse for | Forward for | Reverse for |
|---------------|-------------|--------------------|-------------|--------------------|
| Gene name | total | total | PolyA | PolyA |
| mHist1h2ac | CAACGACGA | TTCTGTTGCT | AAGCCTACC | GGAACAACC |
| | GGAGCTCAA | TATTTCCCCT | ACCTACTCCG | AAGGAGCTT |
| | CAAG | TGG | Т | GC |
| mHist1h2bg | TGTTTCTACC | CTTGGTCAC | ACGCTGGGA | CGTTGGTTAC |
| | ATGCCCGAG | GGCCTTCTTG | AATGACACA | ACTGAGGCA |
| | CC | G | ATA | А |





| hHist1h2ac | GACGAGGAG | ACCTGTCAA | CCTGTCCACT | TTCACTTACC |
|------------|-----------|------------|------------|------------|
| | CTCAACAAA | ATCACTTGCC | GTTGGTAGG | ACCATTCCAG |
| | CTG | С | С | С |
| hHist1h2bd | ACGATGCCT | AGCCTTAGTC | CCAACTCATC | TCCCCTCGGT |
| | GAACCTACC | ACCGCCTTCT | CTGGTTTGCT | AACCTTCTTT |
| | AA | | | |

Table 4. 3' UTR shortened transcripts in p53^{-/-} MEF

| Gene Name | Transcript Name | Shortening P-values | log2 fold change $(p53^{-/-}/p53^{+/+})$ |
|-----------|-----------------|---------------------|---|
| Nadk | NM_138671 | 0.002270635 | -9.473843295 |
| Mme | NM_008604 | 0.039689234 | -4.22985422 |
| Ccdc80 | NM_026439 | 0.028259053 | -2.717481141 |
| Fzd8 | NM_008058 | 0.003401358 | -2.710499863 |
| Rgs5 | NM_009063 | 0.047370497 | -2.626426757 |
| Ccng1 | NM_009831 | 0.001156014 | -2.503682711 |
| Sgk1 | NM_001161850 | 0.014280375 | -2.439432382 |
| Serpine2 | NM_009255 | 0.015643642 | -2.32291134 |
| Npr3 | NM_001039181 | 0.012876631 | -2.193612874 |
| Ssbp1 | NM_212468 | 0.02320321 | -2.107699987 |
| Col11a1 | NM_007729 | 3.97E-09 | -1.986279716 |
| Hr | NM_021877 | 0.041641943 | -1.982100367 |
| Lox14 | NM_001164311 | 0.003528497 | -1.928070828 |
| Abi3bp | NM_001014399 | 0.003597091 | -1.859927169 |
| Fam20c | NM_030565 | 0.015743623 | -1.740494419 |





| Ppap2b | NM_080555 | 0.028688744 | -1.721269809 |
|----------|--------------|-------------|--------------|
| Aspn | NM_001172481 | 2.37E-06 | -1.707855641 |
| Alcam | NM_009655 | 0.000212505 | -1.685117913 |
| Ogn | NM_008760 | 0.019546534 | -1.663179098 |
| Maob | NM_172778 | 0.000462135 | -1.602119709 |
| Edil3 | NM_001037987 | 0.049792193 | -1.575732255 |
| Pqlc3 | NM_172574 | 0.012045374 | -1.547778463 |
| Klf2 | NM_008452 | 0.017666514 | -1.537670055 |
| Rpl39 | NM_026055 | 8.35E-11 | -1.460933717 |
| Osr1 | NM_011859 | 0.001071898 | -1.451358689 |
| Slc12a2 | NM_009194 | 0.028259009 | -1.441115128 |
| Timp2 | NM_011594 | 2.30E-07 | -1.348461997 |
| Nrk | NM_013724 | 0.007515853 | -1.262736177 |
| Get4 | NM_001163316 | 0.049347735 | -1.25286215 |
| Vim | NM_011701 | 0.000382295 | -1.242988117 |
| Gnb1 | NM_001160016 | 0.004953171 | -1.240412477 |
| Qars | NM_001168270 | 0.029583518 | -1.227485533 |
| Fam53b | NM_175268 | 0.026881018 | -1.11396985 |
| Med15 | NM_001040683 | 0.028619358 | -1.112636378 |
| Aen | NM_001162939 | 0.001334537 | -1.103462096 |
| Adamts5 | NM_011782 | 0.001358686 | -1.087638086 |
| Megf9 | NM_172694 | 0.027694652 | -1.066939035 |
| Higd1a | NM_019814 | 0.014955162 | -1.063743198 |
| BC056474 | NM_001001493 | 0.002810255 | -1.052332827 |
| Cd9 | NM_007657 | 0.005589512 | -0.965737427 |
| Itm2a | NM_008409 | 0.040574259 | -0.958678337 |
| Rcn2 | NM 011992 | 0.045081884 | -0.937683946 |





| Keap1 | NM_001110305 | 0.01187661 | -0.928497888 |
|----------|--------------|-------------|--------------|
| Tmem106b | NM_027992 | 0.000102891 | -0.872129703 |
| E112 | NM_138953 | 0.031954297 | -0.848522392 |
| Lmbrd1 | NM_026719 | 0.010606945 | -0.830633441 |
| Fam114a1 | NM_026667 | 0.00739316 | -0.808094642 |
| Pias1 | NM_019663 | 0.042840394 | -0.798573242 |
| Rpl23 | NM_022891 | 0.000477042 | -0.79365085 |
| Itm2b | NM_008410 | 0.013010648 | -0.781458976 |
| Sh3bgrl | NM_019989 | 0.014133455 | -0.778573237 |
| Cd109 | NM_153098 | 0.008481515 | -0.774296029 |
| Anxa7 | NM_001110794 | 0.019670252 | -0.720114241 |
| Tmed10 | NM_026775 | 2.37E-05 | -0.702409598 |
| Tmem134 | NM_001078649 | 0.015478416 | -0.679994881 |
| Slc35e1 | NM_177766 | 0.011363311 | -0.67812888 |
| Chmp3 | NM_025783 | 0.033592441 | -0.62752048 |
| Vma21 | NM_001081356 | 0.01069244 | -0.599513485 |
| Stk3 | NM_019635 | 0.016625496 | -0.599209871 |
| Asah1 | NM_019734 | 3.62E-08 | -0.58962975 |
| Srsf3 | NM_013663 | 0.045667405 | -0.58712236 |
| Oraov1 | NM_028184 | 0.046116991 | -0.574000635 |
| Zfp747 | NM_175560 | 0.019523255 | -0.573997797 |
| Sgms2 | NM_028943 | 0.047220116 | -0.571035528 |
| Fto | NM_011936 | 0.003415505 | -0.56975872 |
| Tmtc3 | NM_001110013 | 0.016876892 | -0.569525837 |
| Atp11c | NM_001037863 | 0.002442142 | -0.568475837 |
| Aldh6a1 | NM_134042 | 0.037652442 | -0.55963897 |
| Usp47 | NM_177249 | 0.043545707 | -0.551236068 |





| Rab28 | NM_027295 | 0.014843403 | -0.540663888 |
|---------|--------------|-------------|--------------|
| Rnf217 | NM_001146349 | 0.0416157 | -0.539068014 |
| Exoc3 | NM_177333 | 0.013651305 | -0.532401015 |
| Fads3 | NM_021890 | 0.039976689 | -0.524551812 |
| Sparc | NM_009242 | 0.049992789 | -0.506335524 |
| Tvp23b | NM_026210 | 0.015352185 | -0.503918083 |
| Tomm20 | NM_024214 | 0.000137571 | -0.499372763 |
| Sfrs18 | NM_025669 | 0.007096247 | -0.498387866 |
| Haus8 | NM_001163042 | 0.003148997 | -0.487749703 |
| Wdr41 | NM_172590 | 0.043931696 | -0.484014187 |
| Dnajc3 | NM_008929 | 0.049597794 | -0.481169574 |
| Cthrc1 | NM_026778 | 0.004516954 | -0.475654166 |
| Nt5c3 | NM_001252374 | 0.000238394 | -0.468949452 |
| Zfp654 | NM_028059 | 0.009081687 | -0.463299147 |
| Rdh14 | NM_023697 | 0.015254968 | -0.445681088 |
| Ebf1 | NM_007897 | 0.010144121 | -0.445457625 |
| Rcn1 | NM_009037 | 0.046506833 | -0.442709407 |
| Map3k7 | NM_009316 | 0.013770916 | -0.428348623 |
| Serinc1 | NM_019760 | 0.003414556 | -0.42741452 |
| Eif4g3 | NM_001256195 | 0.012991964 | -0.425997708 |
| Marcks | NM_008538 | 0.004626708 | -0.421370639 |
| H3f3a | NM_008210 | 9.27E-14 | -0.416080693 |
| Sec24d | NM_027135 | 0.000397174 | -0.415239196 |
| Cmpk1 | NM_025647 | 0.000241398 | -0.409868714 |
| Ggcx | NM_019802 | 0.007381093 | -0.39912198 |
| Col4a5 | NM_001163155 | 0.017392051 | -0.39218664 |
| Nr2f6 | NM_010150 | 0.001361575 | -0.390069731 |





| Zdhhc20 | NM_029492 | 0.029795155 | -0.388555173 |
|---------|--------------|-------------|--------------|
| Fbxo31 | NM_133765 | 0.031402531 | -0.386437505 |
| Lman2 | NM_025828 | 0.00110839 | -0.384334981 |
| Scp2 | NM_011327 | 0.034467407 | -0.383644742 |
| Rnf11 | NM_013876 | 0.023819497 | -0.380282569 |
| Calm1 | NM_009790 | 0.000246143 | -0.379491505 |
| Sec24a | NM_175255 | 0.043767731 | -0.375642337 |
| Slc25a3 | NM_133668 | 0.010588607 | -0.374910207 |
| Dynlt3 | NM_025975 | 0.007271182 | -0.368324937 |
| Fopnl | NM_025345 | 0.045477989 | -0.362397419 |
| Sec22b | NM_011342 | 0.023691609 | -0.361340359 |
| Angel2 | NM_001199020 | 0.026159151 | -0.360303166 |
| Erbb2ip | NM_001005868 | 0.007226404 | -0.345779301 |
| Prrc1 | NM_028447 | 0.000755571 | -0.344666155 |
| Rpl22 | NM_009079 | 0 | -0.343499014 |
| Ccdc90b | NM_001162918 | 0.029007238 | -0.339773193 |
| Wls | NM_026582 | 0.001045896 | -0.335312065 |
| Ubl3 | NM_011908 | 0.012133738 | -0.335122725 |
| Serinc3 | NM_012032 | 0.009267082 | -0.334665161 |
| B3galtl | NM_001081204 | 0.047108805 | -0.333902648 |
| Slain2 | NM_153567 | 0.024569068 | -0.332538101 |
| Rab21 | NM_024454 | 0.035779507 | -0.33074985 |
| Zc3h11a | NM_144530 | 0.005271498 | -0.326635418 |
| Smim14 | NM_133697 | 0.021871589 | -0.324624555 |
| Btbd1 | NM_146193 | 0.017675494 | -0.304896458 |
| Rab1 | NM_008996 | 0.023404919 | -0.304051165 |
| Rac1 | NM_009007 | 9.19E-11 | -0.301545994 |





| Tpm4 | NM_001001491 | 0.01235255 | -0.300554816 |
|----------|--------------|-------------|--------------|
| Mtpn | NM_008098 | 0.030293811 | -0.292921059 |
| Zfp503 | NM_145459 | 0.016270064 | -0.288965264 |
| Map11c3b | NM_026160 | 2.02E-08 | -0.285381372 |
| Rpl15 | NM_025586 | 2.10E-05 | -0.284904213 |
| Sec63 | NM_153055 | 0.004351316 | -0.281263687 |
| Pdia3 | NM_007952 | 0.008229022 | -0.269917879 |
| Mbnl2 | NM_207515 | 3.93E-05 | -0.268891135 |
| Oaz2 | NM_010952 | 0.034208111 | -0.266210287 |
| Ripk1 | NM_009068 | 0.018887723 | -0.25482088 |
| Ufd11 | NM_011672 | 0.049565234 | -0.249043696 |
| Idua | NM_008325 | 0.022569929 | -0.247915796 |
| Pskh1 | NM_173432 | 0.035467907 | -0.246253802 |
| Srp9 | NM_012058 | 0.003142255 | -0.241329995 |
| Неса | NM_001033432 | 0.023296961 | -0.230244671 |
| Tm9sf3 | NM_133352 | 4.95E-07 | -0.228914497 |
| Rps6 | NM_009096 | 0.000514993 | -0.22537355 |
| Gxylt2 | NM_198612 | 0.022162291 | -0.221433386 |
| Reep5 | NM_007874 | 2.65E-08 | -0.217497478 |
| Copg | NM_017477 | 0.036479855 | -0.212992389 |
| Srsf5 | NM_001079695 | 0.001767923 | -0.212634182 |
| Cxcl14 | NM_019568 | 0.003596061 | -0.212436762 |
| Cast | NM_009817 | 0.017789142 | -0.212161574 |
| Yipf5 | NM_023311 | 0.043196013 | -0.211512546 |
| B2m | NM_009735 | 0.026445494 | -0.201187325 |
| Dlgap4 | NM_001042487 | 0.012569503 | -0.199289216 |
| Hnrpdl | NM_016690 | 0.022302966 | -0.197675759 |





| Aamp | NM_146110 | 0.029659338 | -0.192303279 |
|----------|--------------|-------------|--------------|
| Sacm11 | NM_030692 | 0.008545754 | -0.190898005 |
| Slc25a12 | NM_172436 | 0.045043758 | -0.188676137 |
| Strn3 | NM_001172098 | 0.020273494 | -0.18845981 |
| Pik3c3 | NM_181414 | 0.018407644 | -0.183576025 |
| Actb | NM_007393 | 0.036358838 | -0.18146677 |
| Rint1 | NM_177323 | 0.044244722 | -0.178839324 |
| Ddx5 | NM_007840 | 8.81E-09 | -0.178676755 |
| Top2b | NM_009409 | 0.049400421 | -0.173125603 |
| Sbf2 | NM_177324 | 0.023965774 | -0.17151966 |
| Tjp1 | NM_009386 | 0.046047916 | -0.170198164 |
| Mdp1 | NM_023397 | 0.010845482 | -0.168166305 |
| Brwd3 | NM_001081477 | 0.043764473 | -0.166513554 |
| Ipo8 | NM_001081113 | 0.033790967 | -0.16320578 |
| Col3a1 | NM_009930 | 0.000680545 | -0.163127849 |
| Tsc22d2 | NM_001081229 | 0.038452472 | -0.159670789 |
| Egln1 | NM_053207 | 0.022028786 | -0.159613204 |
| Nedd4 | NM_010890 | 0.001034673 | -0.157512186 |
| Nxn | NM_008750 | 0.004647204 | -0.155996436 |
| Hcfc2 | NM_001081218 | 0.000876552 | -0.153218626 |
| Limch1 | NM_001001980 | 0.006747461 | -0.15201797 |
| Vps13a | NM_173028 | 0.042992808 | -0.150405122 |
| Ube2b | NM_009458 | 0.008945722 | -0.148965737 |
| Cisd2 | NM_025902 | 0.000995992 | -0.147117768 |
| Man1a | NM_008548 | 0.034337954 | -0.146663207 |
| Bmpr2 | NM_007561 | 0.002161634 | -0.146629944 |
| Vta1 | NM_025418 | 0.011210986 | -0.138804035 |





| C330007P06Rik | NM_029951 | 0.010118437 | -0.132613819 |
|---------------|--------------|-------------|--------------|
| Dr1 | NM_026106 | 0.037691015 | -0.124427573 |
| Prpf4b | NM_013830 | 0.025111653 | -0.119665403 |
| Ywhab | NM_018753 | 0.031672522 | -0.114011246 |
| Tfdp2 | NM_001184711 | 0.039888955 | -0.1139567 |
| Zmpste24 | NM_172700 | 0.043311663 | -0.113399455 |
| Pgap2 | NM_145583 | 0.035050915 | -0.113049611 |
| D8Ertd738e | NM_001007571 | 0 | -0.1114535 |
| Col5a1 | NM_015734 | 0.037066945 | -0.109655338 |
| Mapre2 | NM_153058 | 0.043501244 | -0.109527559 |
| Chtop | NM_023215 | 0.041136362 | -0.107403013 |
| Mbd2 | NM_010773 | 0.014066949 | -0.107209715 |
| Sucla2 | NM_011506 | 0.001954328 | -0.107103061 |
| Ankrd39 | NM_026241 | 0.011647288 | -0.10314828 |
| Nxf1 | NM_016813 | 0.004380856 | -0.102962376 |
| Snapc5 | NM_183316 | 0.01995693 | -0.097254307 |
| Ccnl1 | NM_019937 | 0.019621475 | -0.094984025 |
| Ccdc115 | NM_027159 | 0.028572661 | -0.09471836 |
| Setd3 | NM_028262 | 0.043170637 | -0.093400937 |
| Ralgapb | NM_177658 | 0.023390568 | -0.091437147 |
| Rsph3a | NM_025789 | 0.045573084 | -0.083569959 |
| Fxr2 | NM_011814 | 8.87E-06 | -0.081625066 |
| Guf1 | NM_172711 | 0.044532902 | -0.081042381 |
| Snapc1 | NM_178392 | 0.03872605 | -0.079358848 |
| Tial1 | NM_009383 | 0.001348663 | -0.075388442 |
| Zfr | NM_011767 | 0.009399516 | -0.072917391 |
| Itgb1 | NM_010578 | 0.02027728 | -0.064430649 |





| Mrpl33 | NM_025796 | 0.00365216 | -0.062335557 |
|-----------|--------------|-------------|--------------|
| Recq15 | NM_130454 | 0.029004152 | -0.057992191 |
| Rwdd4a | NM_203507 | 0.012333694 | -0.056914476 |
| Vps37a | NM_033560 | 2.96E-05 | -0.056157297 |
| Gatad2a | NM_001113346 | 0.006058894 | -0.048724896 |
| Mapkap1 | NM_177345 | 0.029629464 | -0.040189778 |
| Slc27a1 | NM_011977 | 0.000426317 | -0.036078644 |
| Gm2a | NM_010299 | 0.017267182 | -0.036016763 |
| Vapa | NM_013933 | 0.027914895 | -0.033977416 |
| Rps3 | NM_012052 | 0.020393516 | -0.028250204 |
| Tmem101 | NM_029649 | 0.034622694 | -0.026220468 |
| Map4k5 | NM_201519 | 0.001364672 | -0.023184795 |
| Cwc15 | NM_023153 | 0.021568962 | -0.018910274 |
| Spast | NM_016962 | 0.037175399 | -0.017472603 |
| Actr2 | NM_146243 | 0.006641268 | -0.016889042 |
| Ywhae | NM_009536 | 0.008612403 | -0.01607104 |
| Zfp617 | NM_133358 | 0.003422274 | -0.01546712 |
| Npc2 | NM_023409 | 0.047088441 | -0.013882119 |
| Hnrnpa2b1 | NM_016806 | 6.15E-06 | -0.012257713 |
| Pdhb | NM_024221 | 0.02774968 | -0.004454767 |
| Ccdc117 | NM_134033 | 0.03819422 | -0.002463985 |
| Thbs1 | NM_011580 | 0.026614163 | 0 |
| Dimt1 | NM_025447 | 0.01456881 | 0.00457307 |
| Tcf4 | NM_001083967 | 0.006839863 | 0.006750245 |
| Fam32a | NM_026455 | 0 | 0.012757463 |
| Idh3a | NM_029573 | 0.002760123 | 0.016360305 |
| Cggbp1 | NM_178647 | 0.015831461 | 0.018257631 |





| Hnrnpk | NM_025279 | 0.010462743 | 0.020909379 |
|------------|--------------|-------------|-------------|
| Rala | NM_019491 | 0.00132815 | 0.023715425 |
| Nvl | NM_026171 | 5.55E-05 | 0.023788788 |
| Ccdc122 | NM_175369 | 0.046895509 | 0.024388074 |
| Nek4 | NM_011849 | 0.019263417 | 0.025058596 |
| Gpr137 | NM_207220 | 0.033769477 | 0.025308742 |
| Mapk1ip11 | NM_178684 | 0.000790582 | 0.025661598 |
| Smc6 | NM_025695 | 0.008085776 | 0.027912075 |
| Ankrd26 | NM_001081112 | 0.032389295 | 0.034538052 |
| Ddx46 | NM_145975 | 0.004448333 | 0.037433992 |
| Nkrf | NM_029891 | 0.021062394 | 0.039922784 |
| Acta2 | NM_007392 | 0.0403986 | 0.044196923 |
| Ccnh | NM_023243 | 0.039808794 | 0.044311228 |
| Rap1b | NM_024457 | 0.033208056 | 0.044992076 |
| R3hcc11 | NM_177464 | 0.03993453 | 0.048362137 |
| D1Bwg0212e | NM_028043 | 0.013765235 | 0.051587011 |
| Ube2d3 | NM_025356 | 0.001442091 | 0.052200509 |
| Ywhaq | NM_011739 | 0.006772115 | 0.053389243 |
| Tsen15 | NM_025677 | 0.038991132 | 0.054703047 |
| Rap1a | NM_145541 | 0.009773009 | 0.055168276 |
| Fgd6 | NM_053072 | 0.031181574 | 0.055679677 |
| Trmt5 | NM_029580 | 0.000343831 | 0.062044636 |
| Ranbp2 | NM_011240 | 0.00438552 | 0.064102351 |
| Kpnb1 | NM_008379 | 0.026614163 | 0.067885046 |
| Tpp2 | NM_009418 | 0.012446278 | 0.072179756 |
| Rnf128 | NM_023270 | 0.008815635 | 0.074046346 |
| Vcan | NM_001134475 | 0.030381034 | 0.075107857 |





| Utp20 | NM_175158 | 0.0355848 | 0.076370878 |
|---------|--------------|-------------|-------------|
| Tmem38a | a NM_144534 | 0.018460546 | 0.079294532 |
| Rps8 | NM_009098 | 7.42E-07 | 0.09350564 |
| Mtmr2 | NM_023858 | 0.015333594 | 0.094567698 |
| Iqgap1 | NM_016721 | 0.024728862 | 0.096678151 |
| Anapc16 | NM_025514 | 0.000686254 | 0.097231684 |
| Faim | NM_011810 | 0.01514966 | 0.105386761 |
| Ketd5 | NM_027008 | 0.003207622 | 0.107637355 |
| Gulp1 | NM_028450 | 0.005944957 | 0.107671156 |
| Nif311 | NM_022988 | 0.048600256 | 0.113855994 |
| Kif5b | NM_008448 | 0.046753615 | 0.114513412 |
| Obfc1 | NM_175360 | 0.009346365 | 0.114595738 |
| Sdccag3 | NM_026563 | 0.004054888 | 0.115157816 |
| Wtap | NM_001113533 | 0.025077647 | 0.115247763 |
| Smg1 | NM_001031814 | 0.011075461 | 0.117054767 |
| Spcs2 | NM_025668 | 0.008743046 | 0.117749372 |
| Insr | NM_010568 | 0.004501137 | 0.118022576 |
| Hmgb1 | NM_010439 | 5.22E-05 | 0.120366326 |
| Hnrnpu | NM_016805 | 0.032104149 | 0.120659022 |
| Ide | NM_031156 | 0.005639452 | 0.120799232 |
| Tra2a | NM_198102 | 0.018192869 | 0.121741356 |
| Usp36 | NM_001033528 | 0.021827654 | 0.128597376 |
| Bclaf1 | NM_001025392 | 0.001507186 | 0.131094654 |
| Josd1 | NM_028792 | 0.003776795 | 0.131746538 |
| Pak1 | NM_011035 | 0.045251777 | 0.133250969 |
| Dnajc11 | NM_172704 | 0.02267077 | 0.133732035 |
| Ldha | NM_001136069 | 1.51E-07 | 0.138310221 |



| Uba3 | NM_001111106 | 0.036118301 | 0.142559577 |
|-----------|--------------|-------------|-------------|
| Prdx4 | NM_016764 | 0.030382278 | 0.154406194 |
| Eif4g2 | NM_001040131 | 0.031788879 | 0.157913855 |
| Rsl24d1 | NM_198609 | 0.030615168 | 0.161460609 |
| Csnk1a1 | NM_146087 | 0.000332975 | 0.162688258 |
| Eln | NM_007925 | 0.003316025 | 0.166552604 |
| Pnkp | NM_021549 | 0.044136594 | 0.169546896 |
| Apmap | NM_027977 | 0.015714342 | 0.169684504 |
| Hnrnph1 | NM_021510 | 0.012732088 | 0.170081531 |
| Eif3a | NM_010123 | 0.000335866 | 0.172717782 |
| Zfp71-rs1 | NM_145622 | 0.012372335 | 0.175816663 |
| Wdr5 | NM_080848 | 0.019287318 | 0.183450489 |
| Snrpd3 | NM_026095 | 0.024066098 | 0.183846311 |
| Snapc3 | NM_029949 | 0.005857036 | 0.191634048 |
| Psmd11 | NM_178616 | 0.017070785 | 0.192575393 |
| Sh3glb1 | NM_019464 | 0.042885299 | 0.193720378 |
| Rbl1 | NM_011249 | 0.035830043 | 0.19399814 |
| Atp6v1d | NM_023721 | 0.019386874 | 0.196397513 |
| Gzf1 | NM_028986 | 0.026181581 | 0.198523753 |
| Srsf7 | NM_001195487 | 0.000178153 | 0.201294559 |
| Rdh13 | NM_175372 | 0.024984607 | 0.201783591 |
| Pla2g4a | NM_008869 | 0.013588485 | 0.20613882 |
| Tmem60 | NM_177601 | 0.003428297 | 0.206309492 |
| Serbp1 | NM_001113565 | 0.041752671 | 0.208600199 |
| Ptma | NM_008972 | 0.001670593 | 0.209803213 |
| Zfyve20 | NM_030081 | 0.042069948 | 0.21311167 |
| Mtch2 | NM_019758 | 0.001726418 | 0.214194955 |





| Ppp4r2 | NM_182939 | 0.002018311 | 0.216288547 |
|----------|--------------|-------------|-------------|
| Yap1 | NM_009534 | 0.035881207 | 0.220446979 |
| Fnbp4 | NM_018828 | 0.013664294 | 0.222072993 |
| Tstd3 | NM_029840 | 0.002781 | 0.230111458 |
| Scaf11 | NM_028148 | 0.032724929 | 0.232313285 |
| Kif11 | NM_010615 | 0.048639731 | 0.244113255 |
| Nf1 | NM_010897 | 0.011238265 | 0.24546468 |
| Ube2i | NM_001177609 | 4.88E-08 | 0.250599769 |
| Kras | NM_021284 | 0.003470009 | 0.251410889 |
| Frg1 | NM_013522 | 0.018630212 | 0.253301425 |
| Aaed1 | NM_025370 | 0.004726932 | 0.254665881 |
| Ran | NM_009391 | 1.21E-05 | 0.257127852 |
| Nedd1 | NM_008682 | 0.018356549 | 0.257880381 |
| Zfp277 | NM_178845 | 0.031812247 | 0.259620562 |
| Grsf1 | NM_178700 | 0.048136623 | 0.260838449 |
| Srek1ip1 | NM_026075 | 0.000712871 | 0.266620192 |
| Stag1 | NM_009282 | 0.00185379 | 0.267819297 |
| Slit3 | NM_011412 | 0.017873668 | 0.269020071 |
| C2cd3 | NM_001017985 | 0.04999238 | 0.271237258 |
| Anapc1 | NM_008569 | 0.012333269 | 0.27235959 |
| Myo1b | NM_010863 | 0.027734904 | 0.284570019 |
| Proser1 | NM_173382 | 0.023289959 | 0.285328825 |
| Cep110 | NM_030000 | 0.04108657 | 0.291839335 |
| Pdap1 | NM_001033313 | 0.024165509 | 0.292519545 |
| Zmynd11 | NM_144516 | 0.043047082 | 0.292525996 |
| Dnpep | NM_016878 | 0.046471776 | 0.296992079 |
| Spry2 | NM_011897 | 0.007922037 | 0.298706166 |





| Dnmt3a | NM_007872 | 0.00298667 | 0.30422008 |
|---------------|--------------|-------------|-------------|
| Lss | NM_146006 | 0.032610689 | 0.305306453 |
| Mpzl1 | NM_001001880 | 0.038667207 | 0.311313702 |
| Rbm34 | NM_172762 | 0.047401088 | 0.3139419 |
| Tmem209 | NM_178625 | 0.010063317 | 0.316570285 |
| Hmgn5 | NM_016710 | 0.021232074 | 0.329958725 |
| Hspa4 | NM_008300 | 0.003483676 | 0.33079122 |
| Etf1 | NM_144866 | 0.037471376 | 0.340492492 |
| Tceb1 | NM_026456 | 0.04019729 | 0.340887634 |
| Iars | NM_172015 | 0.021717263 | 0.343280723 |
| Gnb4 | NM_013531 | 0.009106455 | 0.345563651 |
| Psma3 | NM_011184 | 0.009922233 | 0.347579778 |
| 9430038I01Rik | NM_029886 | 0.017975572 | 0.349990226 |
| Galm | NM_176963 | 0.008261626 | 0.351757206 |
| Rbm33 | NM_028234 | 0.006807743 | 0.353826427 |
| Orc6 | NM_019716 | 0.012640187 | 0.353952369 |
| Dhodh | NM_020046 | 0.032371932 | 0.366462829 |
| Atp6v1b2 | NM_007509 | 7.52E-10 | 0.367645044 |
| Rfc2 | NM_020022 | 0.029070084 | 0.368574343 |
| Tex2 | NM_198292 | 0.002184619 | 0.369635533 |
| Mtap | NM_024433 | 0.022491829 | 0.377745203 |
| Usp1 | NM_146144 | 0.009543113 | 0.379404631 |
| Nsg1 | NM_010942 | 0.017824929 | 0.386129674 |
| Ghitm | NM_078478 | 6.30E-06 | 0.390301275 |
| Acn9 | NM_001077713 | 0.033618427 | 0.390949103 |
| Tfpi | NM_011576 | 0.001867215 | 0.394637576 |
| Ckap2l | NM_181589 | 0.03320224 | 0.398970792 |





| Set | NM_023871 | 0.003356913 | 0.403325015 |
|----------|--------------|-------------|-------------|
| Lrch2 | NM_001081173 | 0.012895919 | 0.403506779 |
| Pcm1 | NM_023662 | 8.23E-05 | 0.413826328 |
| Ptar1 | NM_028208 | 0.018642171 | 0.416590164 |
| Zfp330 | NM_145600 | 0.033643412 | 0.418468036 |
| Pdcd6ip | NM_001164677 | 0.031551534 | 0.437059519 |
| Pus1 | NM_001025561 | 0.013352887 | 0.448173813 |
| Farsb | NM_011811 | 0.024568538 | 0.46585601 |
| Wbp5 | NM_011712 | 0.000108246 | 0.466335197 |
| Rsrc1 | NM_025822 | 0.019637378 | 0.467700075 |
| Ranbp1 | NM_011239 | 0.000631013 | 0.472452186 |
| Ubqln1 | NM_026842 | 0.02878865 | 0.472685769 |
| Exosc8 | NM_027148 | 4.31E-07 | 0.480144804 |
| Rnaseh2b | NM_026001 | 0.042133212 | 0.491095657 |
| Cbfb | NM_022309 | 0.042995223 | 0.51208926 |
| Mtx2 | NM_016804 | 0.046885554 | 0.536482693 |
| Hmgcs1 | NM_145942 | 0.003279854 | 0.56541287 |
| Rbm28 | NM_133925 | 0.030058202 | 0.566220885 |
| Trim37 | NM_197987 | 0.045557463 | 0.568333019 |
| Prpf38a | NM_172697 | 0.01811914 | 0.568691628 |
| Bmp2k | NM_080708 | 0.025041637 | 0.576889442 |
| Pwwp2a | NM_001164231 | 0.001665638 | 0.579980848 |
| Lbr | NM_133815 | 0.034337299 | 0.605327338 |
| Irx3 | NM_008393 | 0.005687504 | 0.619146058 |
| Cep76 | NM_001081073 | 0.040122412 | 0.62399453 |
| Rassf8 | NM_027760 | 4.90E-06 | 0.628135049 |
| Psmc6 | NM_025959 | 0.000892871 | 0.630263644 |





| Zfp60 | NM_009560 | 0.00674271 | 0.653705973 |
|--------|--------------|-------------|-------------|
| Hn1 | NM_008258 | 0.000103425 | 0.66046758 |
| Lin9 | NM_001103182 | 0.026358838 | 0.665029632 |
| Dis3 | NM_028315 | 0.019480302 | 0.692832384 |
| Ick | NM_019987 | 0.044618087 | 0.736524339 |
| Naa25 | NM_172722 | 0.001489937 | 0.753418478 |
| Sms | NM_009214 | 0.048996516 | 0.757853556 |
| Zdhhc2 | NM_178395 | 0.021251135 | 0.768430052 |
| Atp11c | NM_001001798 | 0.002021255 | 0.791825014 |
| Ssbp3 | NM_023672 | 0.001037956 | 0.828542271 |
| Rcc2 | NM_173867 | 9.88E-06 | 0.871253249 |
| Abca1 | NM_013454 | 0.026535299 | 0.920976172 |
| Chek2 | NM_016681 | 0.035238995 | 0.92662375 |
| Vcam1 | NM_011693 | 0.044087321 | 0.94690625 |
| Snx10 | NM_001127348 | 0.002090973 | 0.976906537 |
| Rbpms2 | NM_028030 | 0.039063037 | 0.987883433 |
| Tcf12 | NM_001253864 | 0.000878164 | 1.034816346 |
| Klhl2 | NM_178633 | 0.025185545 | 1.079327973 |
| Scarb2 | NM_007644 | 0.010809188 | 1.132588129 |
| Dgkh | NM_001081336 | 0.027222482 | 1.190717873 |
| Limd2 | NM_172397 | 0 | 1.204983844 |
| Ocel1 | NM_029865 | 2.94E-09 | 1.332293257 |
| Anp32e | NM_001253757 | 0.037195985 | 1.860350672 |
| Ankrd1 | NM_013468 | 0.005379162 | 2.047986902 |
| Gbp2 | NM_010260 | 0.007773691 | 2.187512423 |
| Thbs2 | NM_011581 | 0.007420898 | 2.235048033 |
| Gdf10 | NM 145741 | 0.028788625 | 2.433620991 |





| Slc16a3 | NM_001038653 | 0.011909358 | 2.434540474 |
|---------|--------------|-------------|-------------|
| Ptprz1 | NM_001081306 | 0.036722378 | 3.152268748 |
| Rcan2 | NM_030598 | 0.036471931 | 3.969189382 |
| Clca1 | NM_009899 | 0.049871938 | 4.01891501 |
| | | | |

Table 5. 3' UTR lengthened transcripts in p53^{-/-} MEF

| Gene Name | Transcript Name | Shortening P-values | log2 fold change $(p53^{-/-}/p53^{+/+})$ |
|-----------|-----------------|---------------------|--|
| Xaf1 | NM_001037713 | -0.00016 | 4.327928 |
| Ncam1 | NM_001113204 | -0.00064 | 3.613468 |
| Bche | NM_009738 | -0.04685 | 2.953972 |
| Rpp25 | NM_133982 | -0.01322 | 2.904772 |
| Rasl10b | NM_001013386 | -0.01726 | 2.886988 |
| Hoxb7 | NM_010460 | -0.03115 | 2.873238 |
| Bcam | NM_020486 | -0.01841 | 2.864856 |
| Gng2 | NM_001038637 | -0.01402 | 2.533142 |
| Gdpd1 | NM_025638 | -0.04108 | 2.23576 |
| Tgif1 | NM_001164074 | -0.04673 | 2.218879 |
| Mogat2 | NM_177448 | -0.02717 | 2.12754 |
| Igfbp5 | NM_010518 | -0.00048 | 1.703104 |
| Frmd4a | NM_001177843 | -0.02157 | 1.579885 |
| Uba7 | NM_023738 | -0.02897 | 1.578136 |
| Ermp1 | NM_001081213 | -0.0212 | 1.513295 |
| Bcl2l11 | NM_207680 | -0.02827 | 1.294806 |
| Nhsl1 | NM_001163592 | -0.03434 | 1.294688 |
| Frmd4a | NM_001177844 | -0.02666 | 1.231694 |
| Slc7a11 | NM_011990 | -0.00071 | 1.230798 |





| Glrb | NM_010298 | -0.02881 | 1.193454 |
|------------|--------------|-----------|----------|
| Hs6st1 | NM_015818 | -0.02276 | 1.086756 |
| Trip13 | NM_027182 | -0.01563 | 1.004429 |
| Cldn12 | NM_022890 | -0.04996 | 0.967975 |
| Cdkn2aipnl | NM_029976 | -0.01235 | 0.95889 |
| Fbln2 | NM_001081437 | -3.30E-05 | 0.923297 |
| Tspan5 | NM_019571 | -0.00781 | 0.900928 |
| Grem2 | NM_011825 | -0.03522 | 0.882022 |
| Alg11 | NM_183142 | -0.01495 | 0.871699 |
| Tmeff1 | NM_021436 | -0.00951 | 0.855545 |
| Gas7 | NM_001109657 | -0.02047 | 0.83369 |
| Tnk2 | NM_001110147 | -0.01989 | 0.808891 |
| Dtl | NM_029766 | -0.03608 | 0.80428 |
| Gm13212 | NM_001205101 | -0.00445 | 0.795427 |
| Hmgcr | NM_008255 | -0.03363 | 0.788314 |
| Aldh112 | NM_153543 | -0.03273 | 0.767937 |
| Pdgfra | NM_001083316 | -0.04936 | 0.746243 |
| Scly | NM_016717 | -0.00893 | 0.728569 |
| Dus41 | NM_028002 | -0.0156 | 0.688201 |
| Tubb2b | NM_023716 | -0.00123 | 0.683731 |
| Rab6a | NM_001163663 | -0.03364 | 0.647368 |
| Zfp930 | NM_001013379 | -0.04297 | 0.643135 |
| Sc4mol | NM_025436 | -0.01925 | 0.618494 |
| Basp1 | NM_027395 | -0.00549 | 0.604799 |
| Sertad2 | NM_001038625 | -0.02208 | 0.583754 |
| Zfp827 | NM_178267 | -0.03209 | 0.581935 |
| Hbs11 | NM_001042593 | -0.03454 | 0.575888 |





| Sulf1 | NM_172294 | -0.00078 | 0.569318 |
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| Lman21 | NM_001013374 | -0.03393 | 0.553003 |
| Zranb3 | NM_027678 | -0.01108 | 0.551464 |
| Lipa | NM_021460 | -0.03293 | 0.533458 |
| Smc5 | NM_001252684 | -0.0298 | 0.532682 |
| Gm14295 | NM_001205057 | -0.04519 | 0.523637 |
| Chchd4 | NM_133928 | -0.00976 | 0.520132 |
| Stk38 | NM_134115 | -0.0044 | 0.495112 |
| Gsr | NM_010344 | -0.02519 | 0.48663 |
| Myd88 | NM_010851 | -0.02464 | 0.473021 |
| Slc39a8 | NM_026228 | -0.02588 | 0.472095 |
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| Cdon | NM_021339 | -0.00673 | 0.461677 |
| Il13ra1 | NM_133990 | -0.03352 | 0.459726 |
| Rlim | NM_011276 | -0.01635 | 0.445735 |
| Grb10 | NM_001177629 | -0.00017 | 0.444548 |
| Cic | NM_001110132 | -0.03734 | 0.442394 |
| Mrpl13 | NM_026759 | -0.02968 | 0.428897 |
| Fbxl12 | NM_001002846 | -0.04617 | 0.422862 |
| 110059E24Rik | NM_025423 | -0.01473 | 0.411543 |
| Rrs1 | NM_021511 | -0.02847 | 0.404517 |
| Arl8b | NM_026011 | -0.01278 | 0.403303 |
| Slc43a3 | NM_021398 | -0.02533 | 0.395432 |
| Rbms2 | NM_001039080 | -6.18E-05 | 0.38527 |
| Ep400 | NM_029337 | -0.00725 | 0.370048 |
| Midn | NM_021565 | -0.00169 | 0.363895 |
| Rnf4 | NM_011278 | -0.01519 | 0.363393 |



1



| Papd5 | NM_001164499 | -0.04606 | 0.357361 |
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| Rarg | NM_001042727 | -0.0007 | 0.342488 |
| Patl1 | NM_172635 | -0.03868 | 0.333632 |
| Trmt6 | NM_175113 | -0.03821 | 0.328875 |
| Topbp1 | NM_176979 | -0.03315 | 0.323678 |
| Elf2 | NM_023502 | -0.03343 | 0.315205 |
| 2310044G17Rik | NM_173735 | -0.03144 | 0.315019 |
| Hmbs | NM_013551 | -0.03815 | 0.313382 |
| Aff4 | NM_033565 | -0.01294 | 0.308015 |
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| Mllt6 | NM_139311 | -0.03166 | 0.300566 |
| Spcs3 | NM_029701 | -0.00728 | 0.298784 |
| Pgam5 | NM_028273 | -0.00087 | 0.297133 |
| Ankrd28 | NM_001024604 | -0.01328 | 0.275482 |
| Papola | NM_011112 | -0.01524 | 0.272971 |
| Zadh2 | NM_146090 | -0.011 | 0.272847 |
| Hoxa11 | NM_010450 | -0.01503 | 0.268993 |
| Wapal | NM_001004436 | -0.0373 | 0.26629 |
| Atp5f1 | NM_009725 | -0.04946 | 0.254822 |
| Prdm4 | NM_181650 | -0.00438 | 0.254528 |
| Pa2g4 | NM_011119 | -0.01287 | 0.250461 |
| Myl12b | NM_023402 | -0.0072 | 0.249665 |
| Thoc3 | NM_028597 | -0.04467 | 0.249382 |
| Cep5711 | NM_001243074 | -0.02475 | 0.241969 |
| Smarcd2 | NM_001130187 | -0.00057 | 0.234736 |
| Snx6 | NM_026998 | -0.03193 | 0.233404 |
| Rad50 | NM_009012 | -0.01959 | 0.230442 |





| Pnrc2 | NM_026383 | -0.01819 | 0.22468 |
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| Chmp4b | NM_029362 | -0.03478 | 0.216905 |
| Tars2 | NM_027931 | -0.00208 | 0.216884 |
| Dld | NM_007861 | -0.00234 | 0.216405 |
| Ube2f | NM_026454 | -0.01067 | 0.213627 |
| Wbscr16 | NM_033572 | -0.0005 | 0.212182 |
| Gcsh | NM_026572 | -0.02573 | 0.211645 |
| Sirt5 | NM_178848 | -0.02756 | 0.206625 |
| Eif1 | NM_011508 | -2.66E-05 | 0.20625 |
| Rbm22 | NM_025776 | -0.00611 | 0.205249 |
| Ttl | NM_027192 | -0.02697 | 0.199918 |
| Ubxn7 | NM_177633 | -0.03577 | 0.19833 |
| Mmachc | NM_025962 | -0.01426 | 0.197534 |
| Cltc | NM_001003908 | -0.00027 | 0.190379 |
| Spsb1 | NM_029035 | -0.02364 | 0.186202 |
| Abcc1 | NM_008576 | -0.04617 | 0.181197 |
| Gja1 | NM_010288 | -0.01933 | 0.180818 |
| Csde1 | NM_144901 | -0.00808 | 0.180291 |
| Lamc1 | NM_010683 | -2.78E-06 | 0.174213 |
| Usp54 | NM_030180 | -0.02913 | 0.173065 |
| Psmc4 | NM_011874 | -0.04363 | 0.170862 |
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| Ppfibp1 | NM_001170433 | -0.01524 | 0.16537 |
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| Cotl1 | NM_028071 | -0.04924 | 0.152192 |
| Lemd2 | NM_146075 | -0.04227 | 0.15054 |
| Alg6 | NM_001081264 | -0.01042 | 0.146803 |




| Clic4 | NM_013885 | -1.77E-07 | 0.142635 |
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| Rhobtb2 | NM_153514 | -0.03232 | 0.1419 |
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| Chordc1 | NM_025844 | -0.00555 | 0.132184 |
| Aven | NM_028844 | -0.03119 | 0.130698 |
| Sik3 | NM_027498 | -0.03488 | 0.128049 |
| Col16a1 | NM_028266 | -0.02411 | 0.127216 |
| Mrpl36 | NM_053163 | -0.00561 | 0.124579 |
| Ccdc25 | NM_145944 | -0.00017 | 0.12312 |
| Zbtb2 | NM_001033466 | -0.03622 | 0.122466 |
| Dffb | NM_007859 | -0.01798 | 0.118907 |
| Dnajc8 | NM_172400 | -0.01263 | 0.118122 |
| Rpl13a | NM_009438 | -0.03217 | 0.116105 |
| Foxj3 | NM_172699 | -0.00233 | 0.115492 |
| Tns3 | NM_001083587 | -0.04896 | 0.114099 |
| Specc1 | NM_001029936 | -0.04943 | 0.109117 |
| Fkbp1a | NM_008019 | -0.00381 | 0.103215 |
| Lgalsl | NM_173752 | -0.0311 | 0.09893 |
| Exoc1 | NM_027270 | -0.00216 | 0.095754 |
| Hsp90ab1 | NM_008302 | -0.00879 | 0.092975 |
| Coq2 | NM_027978 | -0.01089 | 0.091674 |
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| Txnip | NM_001009935 | -0.01534 | 0.07085 |





| Kpna6 | NM_008468 | -0.02554 | 0.069753 |
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| Zfp825 | NM_146231 | -0.01894 | 0.069181 |
| Cd164 | NM_016898 | -0.00629 | 0.064302 |
| Hdgf | NM_008231 | -0.01127 | 0.063592 |
| Gltpd1 | NM_024472 | -0.0206 | 0.043996 |
| Cfl1 | NM_007687 | -6.93E-08 | 0.0437 |
| Casp7 | NM_007611 | -0.00694 | 0.039861 |
| Spty2d1 | NM_175318 | -0.04507 | 0.034125 |
| Slc7a6 | NM_178798 | -0.00094 | 0.033462 |
| M6pr | NM_010749 | -0.01948 | 0.031739 |
| Marf1 | NM_001081154 | -0.03847 | 0.03098 |
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| Hnrnpul2 | NM_001081196 | -0.00024 | 0.028341 |
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| Ctr9 | NM_009431 | -0.02258 | 0.015458 |
| Msn | NM_010833 | -0.03509 | 0.013507 |
| Sf3b3 | NM_133953 | -0.00172 | 0.004574 |
| Vhl | NM_009507 | -0.01068 | 0.003767 |
| Rabif | NM_145510 | -0.00355 | 0.003479 |
| Hibch | NM_146108 | -0.04871 | 0.002295 |
| E2f8 | NM_001013368 | -0.03129 | -0.00127 |
| Ccnt2 | NM_028399 | -0.02145 | -0.00484 |
| Cpsf7 | NM_001164272 | -0.03611 | -0.00545 |
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| Elk3 | NM_013508 | -0.03122 | -0.01663 |
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| Glipr2 | NM_027450 | -0.03071 | -0.01786 |
| Tmem165 | NM_011626 | -0.00016 | -0.01947 |
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| Elk1 | NM_007922 | -0.04764 | -0.04158 |
| Pgpep1 | NM_023217 | -0.00142 | -0.04238 |
| Ftl1 | NM_010240 | -0.04652 | -0.04638 |
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| Tmed4 | NM_134020 | -0.02679 | -0.04703 |
| Prune | NM_173347 | -0.00371 | -0.04913 |
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| Trim23 | NM_030731 | -0.00099 | -0.06765 |
| Golga7 | NM_001042484 | -0.00211 | -0.07129 |
| BC030336 | NM_001164580 | -0.01859 | -0.077 |
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| Mdm4 | NM_008575 | -0.01233 | -0.08139 |
| Ano6 | NM_175344 | -0.03102 | -0.08271 |
| Elf1 | NM_007920 | -0.01686 | -0.08776 |
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| Actr1a | NM_016860 | -0.02369 | -0.09698 |
| Atxn3 | NM_029705 | -0.02534 | -0.09751 |
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| Zbtb9 | NM_001005916 | -0.04781 | -0.10907 |
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| Krtcap3 | NM_027221 | -0.00781 | -0.15054 |
| Hp1bp3 | NM_001122897 | -0.01093 | -0.15254 |
| Plin3 | NM_025836 | -0.00092 | -0.15959 |
| Dnajc5 | NM_016775 | -0.00058 | -0.1597 |
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| Pak2 | NM_177326 | -0.00399 | -0.17429 |
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| Myh9 | NM_022410 | -0.03668 | -0.20178 |
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| Flrt2 | NM_201518 | -0.00779 | -0.20445 |





| Gns | NM_029364 | -0.00998 | -0.20586 |
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| Eef2 | NM_007907 | -0.00011 | -0.20781 |
| Fut11 | NM_028428 | -0.00713 | -0.21071 |
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| Fem1a | NM_010192 | -0.00035 | -0.2152 |
| Sec31a | NM_026969 | -0.02087 | -0.21629 |
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| Cap1 | NM_007598 | -0.01439 | -0.23344 |
| Smarca2 | NM_026003 | -0.01245 | -0.23673 |
| Asb7 | NM_080443 | -0.01556 | -0.24102 |
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| Aff1 | NM_001080798 | -0.02875 | -0.24655 |
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| Tmem19 | NM_133683 | -0.03051 | -0.25607 |
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| Cep350 | NM_001039184 | -0.00184 | -0.27821 |
| Rfx1 | NM_009055 | -0.00762 | -0.28368 |
| Dhx33 | NM_178367 | -0.00935 | -0.28636 |





| Oxct1 | NM_024188 | -0.00895 | -0.29469 |
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| Uhmk1 | NM_010633 | -0.01263 | -0.29616 |
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| Echdc2 | NM_001254754 | -0.00499 | -0.31636 |
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| Slc25a37 | NM_026331 | -0.01977 | -0.32239 |
| Prnp | NM_011170 | -0.00024 | -0.33423 |
| Pigk | NM_025662 | -0.00279 | -0.33836 |
| Stx18 | NM_026959 | -0.0231 | -0.34112 |
| 3110001D03Rik | NM_025849 | -0.01368 | -0.34952 |
| Figf | NM_010216 | -0.02241 | -0.35244 |
| Canx | NM_001110500 | -0.04635 | -0.37117 |
| Mob3a | NM_172457 | -0.01317 | -0.37142 |
| Myo10 | NM_019472 | -0.04466 | -0.37373 |
| Rnf38 | NM_001038993 | -0.02283 | -0.37658 |
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| Twist1 | NM_011658 | -0.02407 | -0.39811 |
| Pja2 | NM_001025309 | -2.82E-06 | -0.40333 |
| Tnpo1 | NM_001048267 | -0.00807 | -0.41176 |
| Ankrd13a | NM_026718 | -0.03478 | -0.41472 |
| Arpc5 | NM_026369 | -0.00483 | -0.41476 |
| Golga4 | NM_018748 | -0.01812 | -0.41853 |
| Nono | NM_001252518 | -0.00266 | -0.42946 |
| Ap2b1 | NM_001035854 | -0.01659 | -0.43596 |
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| Cnn2 | NM_007725 | -0.02603 | -0.46481 |
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| B4galt6 | NM_019737 | -0.01214 | -0.47704 |
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| Nckipsd | NM_030729 | -0.03277 | -0.50596 |
| Rassf3 | NM_138956 | -0.00892 | -0.51872 |
| Myadm | NM_016969 | -2.87E-08 | -0.52206 |
| Ptpn1 | NM_011201 | -0.0329 | -0.52744 |
| Sun2 | NM_001205345 | -0.03522 | -0.52754 |
| Timp3 | NM_011595 | -0.00525 | -0.54591 |
| Adamts15 | NM_001113548 | -0.00934 | -0.54781 |
| Actn1 | NM_134156 | -0.02329 | -0.5709 |
| Smim7 | NM_172396 | -0.00696 | -0.57664 |
| Ndrg3 | NM_013865 | -0.00548 | -0.58199 |
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| Ddr1 | NM_001198831 | -0.03676 | -0.60141 |
| Fbxo34 | NM_030236 | -0.02953 | -0.60291 |
| Flnb | NM_134080 | -0.03126 | -0.60772 |
| 2610305D13Rik | NM_145078 | -0.04672 | -0.61003 |
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| Tram1 | NM_028173 | -0.02249 | -0.64947 |
| Fam3a | NM_025473 | -0.00585 | -0.6733 |
| Mfge8 | NM_001045489 | -0.02027 | -0.67582 |
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| Sh3bp2 | NM_011893 | -0.01315 | -0.68499 |
| Csgalnact2 | NM_030165 | -0.03076 | -0.68671 |
| Plxna3 | NM_008883 | -0.03317 | -0.69796 |





| Sft2d3 | NM_026006 | -0.04694 | -0.70981 |
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| Fbxo25 | NM_025785 | -0.01726 | -0.7156 |
| Msrb3 | NM_177092 | -6.61E-05 | -0.73303 |
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| Rgs3 | NM_134257 | -0.00024 | -0.73703 |
| Tgfb2 | NM_009367 | -0.0017 | -0.73897 |
| Sptbn1 | NM_175836 | -0.0007 | -0.73935 |
| Six2 | NM_011380 | -0.01117 | -0.74756 |
| Man1a2 | NM_010763 | -0.01781 | -0.75464 |
| Wnt5a | NM_001256224 | -0.01982 | -0.76339 |
| Ankrd13d | NM_026720 | -0.03987 | -0.78065 |
| Dag1 | NM_010017 | -0.00126 | -0.80168 |
| Tprgl | NM_026388 | -0.01143 | -0.80509 |
| Hist1h1c | NM_015786 | -0.01906 | -0.81004 |
| 2310045N01Rik | NM_001145552 | -0.0009 | -0.84263 |
| Nfat5 | NM_133957 | -0.01037 | -0.84313 |
| Atf5 | NM_030693 | -0.03071 | -0.85838 |
| Mgat1 | NM_010794 | -0.00232 | -0.86173 |
| Robo1 | NM_019413 | -0.03969 | -0.88133 |
| Tspan12 | NM_173007 | -0.02226 | -0.90545 |
| H2afj | NM_177688 | -0.00667 | -1.00523 |
| Rpl37a | NM_009084 | -0.00376 | -1.03046 |
| Hist1h2bc | NM_023422 | -0.00898 | -1.03994 |
| Rb1 | NM_009029 | -0.04381 | -1.0485 |
| Sec16b | NM_033354 | -0.04518 | -1.06245 |
| Tcf712 | NM_001142923 | -0.0362 | -1.08551 |
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| Rap2a | NM_029519 | -0.01164 | -1.15385 |
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| Pphln1 | NM_175363 | -0.01858 | -1.20049 |
| Rnpc3 | NM_001038696 | -0.04121 | -1.20102 |
| Dlc1 | NM_015802 | -0.00044 | -1.25027 |
| Nrbp2 | NM_144847 | -0.0037 | -1.27865 |
| Fat4 | NM_183221 | -0.01929 | -1.38881 |
| Sdc2 | NM_008304 | -0.00015 | -1.4805 |
| Dpt | NM_019759 | -0.00819 | -1.48814 |
| Ids | NM_010498 | -0.02071 | -1.56982 |
| Agtr2 | NM_007429 | -0.02564 | -1.57914 |
| Tom112 | NM_001039092 | -0.01828 | -1.63318 |
| Pik3r1 | NM_001024955 | -0.00103 | -1.67609 |
| Myo1c | NM_001080774 | -0.03771 | -1.90426 |
| Col8a1 | NM_007739 | -0.00015 | -1.93285 |
| Ltbp2 | NM_013589 | -0.02366 | -2.03732 |
| Lynx1 | NM_011838 | -0.04623 | -2.05528 |
| Mgp | NM_008597 | -0.0279 | -2.26817 |
| Scoc | NM_001039137 | -1.45E-10 | -2.40506 |
| Cox7a2l | NM_001159529 | -0.02475 | -2.43993 |
| Thbd | NM_009378 | -2.91E-05 | -2.52601 |
| Chodl | NM_139134 | -0.04544 | -2.72301 |
| Perp | NM_022032 | -0.02791 | -3.75153 |
| S100b | NM_009115 | -0.02397 | -4.18927 |

Table 6. 3' UTR shortened transcripts in 53bp1^{-/-} MEF

| Gene Name | Transcript Name | Shortening P-values | log2 fold change |
|-----------|-----------------|---------------------|---|
| | | | (53bp1 ^{-/-} /53bp1 ^{+/+}) |
| | | | |





| Ctss | NM_001267695 | 0.032188 | 13.7838 |
|---------------|--------------|----------|----------|
| Nxt2 | NM_001161430 | 0.024999 | 11.6861 |
| Lum | NM_008524 | 0.006338 | 2.24696 |
| Col10a1 | NM_009925 | 0.017829 | 1.62176 |
| 9930013L23Rik | NM_030728 | 0.035751 | 1.24607 |
| Arvcf | NM_001272032 | 0.011206 | 1.00654 |
| Casc4 | NM_001205370 | 0.003422 | 0.869332 |
| Mxd4 | NM_010753 | 0.011644 | 0.837635 |
| Pros1 | NM_011173 | 0.016754 | 0.790169 |
| Epha1 | NM_023580 | 0.020217 | 0.760779 |
| Prnp | NM_011170 | 0.001043 | 0.683845 |
| Cnksr3 | NM_172546 | 0.020111 | 0.637525 |
| Cog6 | NM_026225 | 0.025452 | 0.610895 |
| B2m | NM_009735 | 1.35E-12 | 0.606737 |
| Sdc2 | NM_008304 | 0.019268 | 0.56431 |
| Tmem230 | NM_001141971 | 0.022413 | 0.563902 |
| Trib3 | NM_175093 | 0.025109 | 0.527591 |
| Nono | NM_001252518 | 4.52E-05 | 0.519716 |
| Wdr37 | NM_001039388 | 0.030728 | 0.505438 |
| Itch | NM_001243712 | 0.044734 | 0.505081 |
| Tprgl | NM_026388 | 0.034804 | 0.501038 |
| Pfn2 | NM_019410 | 0.043104 | 0.461953 |
| Ppt1 | NM_008917 | 8.47E-09 | 0.454568 |
| Coa4 | NM_183270 | 0.01093 | 0.419251 |
| Rdh14 | NM_023697 | 0.043009 | 0.403955 |
| Kdm3a | NM_001038695 | 0.012137 | 0.400004 |
| Cic | NM_001110132 | 0.043454 | 0.384998 |





| Twist1 | NM_011658 | 0.037977 | 0.381642 |
|----------|--------------|----------|----------|
| Prickle1 | NM_001033217 | 0.020052 | 0.381443 |
| Ppfibp1 | NM_001170433 | 0.004987 | 0.371478 |
| Lyrm1 | NM_029610 | 0.049371 | 0.365536 |
| Slc39a1 | NM_013901 | 0.00545 | 0.35977 |
| Tmsb4x | NM_021278 | 0.035571 | 0.358407 |
| Zfp503 | NM_145459 | 0.008543 | 0.345096 |
| Lrp1 | NM_008512 | 0.000123 | 0.337137 |
| Mospd2 | NM_029730 | 0.001105 | 0.336132 |
| Hmgcs1 | NM_145942 | 9.42E-06 | 0.327239 |
| Frk | NM_010237 | 0.018005 | 0.311759 |
| Cln5 | NM_001033242 | 0.016095 | 0.309533 |
| Bloc1s5 | NM_139063 | 0.027366 | 0.304952 |
| Pla2g15 | NM_133792 | 0.049435 | 0.299736 |
| Mocs1 | NM_020042 | 0.031244 | 0.294514 |
| Vipas39 | NM_001142580 | 0.018948 | 0.29159 |
| Zbtb33 | NM_001079513 | 0.028965 | 0.269057 |
| Ttf1 | NM_009442 | 0.048412 | 0.264814 |
| Ralbp1 | NM_009067 | 0.030482 | 0.25936 |
| Dynlt3 | NM_025975 | 0.023263 | 0.255262 |
| Rusc1 | NM_028188 | 0.012758 | 0.25301 |
| Tbc1d20 | NM_024196 | 0.010765 | 0.246077 |
| Rbm39 | NM_133242 | 0.033203 | 0.242756 |
| Derl1 | NM_024207 | 0.026869 | 0.228186 |
| Rpl13a | NM_009438 | 0.000112 | 0.226174 |
| Snx18 | NM_130796 | 0.000483 | 0.219026 |
| Rbbp9 | NM_015754 | 0.029447 | 0.211185 |





| Dennd5a | NM_021494 | 0.042329 | 0.200475 |
|---------|--------------|----------|----------|
| Pja2 | NM_001025309 | 0.001041 | 0.19206 |
| Pik3c3 | NM_181414 | 0.048599 | 0.191803 |
| Capn7 | NM_009796 | 0.036466 | 0.190784 |
| Rnd3 | NM_028810 | 0.032376 | 0.180532 |
| Nptn | NM_009145 | 0.015367 | 0.168717 |
| Ltbp2 | NM_013589 | 0.014271 | 0.168233 |
| Hps3 | NM_080634 | 0.049503 | 0.163076 |
| Gfer | NM_023040 | 1.35E-06 | 0.158488 |
| Plekhf1 | NM_024413 | 0.031158 | 0.132397 |
| Mat2b | NM_134017 | 0.011596 | 0.111132 |
| Slc15a4 | NM_133895 | 0.027246 | 0.099576 |
| Morf411 | NM_001039147 | 0.015913 | 0.092889 |
| Slc39a7 | NM_001077709 | 0.030888 | 0.080452 |
| Commd4 | NM_025417 | 0.013476 | 0.067274 |
| Aplar | NM_145964 | 0.004697 | 0.065553 |
| Nudt3 | NM_019837 | 0.016989 | 0.062057 |
| Mpdz | NM_010820 | 0.034969 | 0.037708 |
| Aplp2 | NM_001102456 | 0.014247 | 0.036381 |
| Chmp1a | NM_145606 | 0.024644 | 0.035668 |
| Haus2 | NM_025475 | 0.039026 | 0.034712 |
| Sgpl1 | NM_009163 | 0.013714 | 0.02999 |
| Hypk | NM_026318 | 0.001556 | 0.025327 |
| Pxn | NM_133915 | 0.043077 | -0.0029 |
| Slc30a4 | NM_011774 | 0.016217 | -0.01177 |
| Pnpo | NM_134021 | 0.036032 | -0.02433 |
| Chp1 | NM_019769 | 0.001979 | -0.02587 |





| Pcyt1a | NM_009981 | 0.047881 | -0.03365 |
|----------|--------------|----------|----------|
| Bmpr2 | NM_007561 | 0.002523 | -0.03499 |
| Cdk2 | NM_183417 | 0.037588 | -0.0411 |
| Setd7 | NM_080793 | 0.025168 | -0.04673 |
| Eif2s3y | NM_012011 | 0.020551 | -0.04899 |
| Snx2 | NM_026386 | 0.040865 | -0.0619 |
| Ano6 | NM_175344 | 0.028569 | -0.07009 |
| Fat1 | NM_001081286 | 0.003854 | -0.07604 |
| Tm2d2 | NM_027194 | 0.018627 | -0.07879 |
| Ptpn9 | NM_019651 | 0.018011 | -0.09067 |
| Ssr2 | NM_025448 | 0.007063 | -0.10059 |
| Pdia3 | NM_007952 | 0.014826 | -0.10534 |
| Nr2f6 | NM_010150 | 0.011125 | -0.1083 |
| Mbd3 | NM_013595 | 0.04066 | -0.1149 |
| Phtf2 | NM_172992 | 0.021272 | -0.11675 |
| Zmpste24 | NM_172700 | 0.042196 | -0.12097 |
| Ccnt2 | NM_028399 | 0.023675 | -0.14228 |
| Kif5b | NM_008448 | 2.90E-06 | -0.15062 |
| Bcap29 | NM_001164090 | 0.047958 | -0.15078 |
| Ubqln2 | NM_018798 | 0.036749 | -0.17722 |
| Capza2 | NM_007604 | 0.010033 | -0.18293 |
| Picalm | NM_001252521 | 0.021973 | -0.18362 |
| Rabif | NM_145510 | 0.029208 | -0.18558 |
| Ktn1 | NM_008477 | 0.008745 | -0.19162 |
| Chtop | NM_023215 | 0.015166 | -0.19317 |
| Gnl31 | NM_198110 | 0.024862 | -0.19457 |
| Pard3 | NM_033620 | 0.044875 | -0.20247 |





| Tctex1d2 | NM_025329 | 0.008409 | -0.2025 |
|----------|--------------|----------|----------|
| Pcbp1 | NM_011865 | 0.000215 | -0.20854 |
| Ehd2 | NM_153068 | 0.002599 | -0.21204 |
| Pxk | NM_145458 | 0.04253 | -0.21308 |
| Lhfp | NM_175386 | 0.003029 | -0.22383 |
| Ppme1 | NM_028292 | 0.005608 | -0.2239 |
| Pigq | NM_011822 | 0.001755 | -0.24241 |
| Kpna1 | NM_008465 | 0.048086 | -0.24445 |
| Pip4k2a | NM_008845 | 0.027845 | -0.25311 |
| Cep85 | NM_144527 | 0.010119 | -0.25377 |
| Sap130 | NM_172965 | 0.04568 | -0.26884 |
| Ndst2 | NM_010811 | 0.01077 | -0.29212 |
| Hp1bp3 | NM_001122897 | 0.009964 | -0.30045 |
| Gp49a | NM_008147 | 0.03248 | -0.3012 |
| Qk | NM_001159517 | 0.01199 | -0.31073 |
| Zc3h15 | NM_026934 | 0.024922 | -0.33077 |
| Tmem131 | NM_018872 | 0.004002 | -0.33227 |
| Fmr1 | NM_008031 | 0.040457 | -0.33805 |
| Ankrd13c | NM_001013806 | 0.007929 | -0.33867 |
| Mier1 | NM_001039081 | 0.035083 | -0.34049 |
| Zfp143 | NM_009281 | 0.045184 | -0.34563 |
| Tars | NM_033074 | 0.039104 | -0.36589 |
| Ngdn | NM_026890 | 0.018268 | -0.36966 |
| Psma5 | NM_011967 | 0.009186 | -0.37978 |
| Cabin1 | NM_172549 | 0.040517 | -0.38153 |
| Fkbp1a | NM_008019 | 2.46E-05 | -0.38178 |
| Parl | NM_001005767 | 0.041615 | -0.392 |





| Mob3a | NM_172457 | 0.038355 | -0.39692 |
|---------------|--------------|----------|----------|
| Snx5 | NM_001199188 | 6.83E-10 | -0.41229 |
| Alg9 | NM_133981 | 0.025112 | -0.41756 |
| Tmem128 | NM_025480 | 0.049133 | -0.42277 |
| Myadm | NM_016969 | 0.030028 | -0.4347 |
| Fam178a | NM_001081225 | 0.002185 | -0.44007 |
| Cfl1 | NM_007687 | 0.000238 | -0.45074 |
| Ubap2 | NM_026872 | 0.001217 | -0.4534 |
| Hnrnpr | NM_028871 | 0.022687 | -0.46063 |
| Fbln2 | NM_001081437 | 0.027843 | -0.46587 |
| Nvl | NM_026171 | 0.019788 | -0.46614 |
| Bmper | NM_028472 | 0.035096 | -0.50731 |
| Smarcc1 | NM_009211 | 0.005003 | -0.5081 |
| Rsl24d1 | NM_198609 | 0.01152 | -0.50838 |
| Rdh13 | NM_175372 | 0.00574 | -0.54425 |
| Sun1 | NM_001256116 | 0.023896 | -0.55531 |
| Tpm4 | NM_001001491 | 0.044388 | -0.55941 |
| Polm | NM_017401 | 0.001029 | -0.57306 |
| Nme1 | NM_008704 | 0.010399 | -0.57775 |
| Glipr2 | NM_027450 | 0.002017 | -0.58123 |
| Ube2f | NM_026454 | 0.021657 | -0.58195 |
| Mlf2 | NM_145385 | 3.01E-05 | -0.58442 |
| Tomm5 | NM_001134646 | 0.005117 | -0.59789 |
| Tmem48 | NM_028355 | 0.022571 | -0.60797 |
| Ptges3 | NM_019766 | 0.046528 | -0.61408 |
| Rfc2 | NM_020022 | 0.0014 | -0.61484 |
| 2810474O19Rik | NM_026054 | 0.004424 | -0.65857 |





| Dstn | NM_019771 | 1.08E-13 | -0.67883 |
|---------|--------------|----------|----------|
| Smim15 | NM_001048250 | 0.003137 | -0.72485 |
| Nup160 | NM_021512 | 0.041827 | -0.73493 |
| Eef1e1 | NM_025380 | 0.024282 | -0.75661 |
| Katnbl1 | NM_024254 | 0.045787 | -0.7653 |
| Bag2 | NM_145392 | 0.033215 | -0.7973 |
| Usp10 | NM_009462 | 0.044516 | -0.80026 |
| Nol10 | NM_001008421 | 0.015644 | -0.85876 |
| Hnrnpm | NM_029804 | 0.029507 | -0.90891 |
| Hes1 | NM_008235 | 0.004753 | -0.92095 |
| Nr1d1 | NM_145434 | 0.027568 | -0.94722 |
| Cdca5 | NM_026410 | 0.031199 | -1.03234 |
| Cyr61 | NM_010516 | 0.017635 | -1.10882 |
| Ddx21 | NM_019553 | 0.044076 | -1.14343 |
| Slc20a1 | NM_015747 | 0.009162 | -1.15787 |
| Hmga2 | NM_010441 | 0.004319 | -1.21498 |
| Ccnb1 | NM_172301 | 0.004775 | -1.25464 |
| H2afy | NM_001159513 | 0.006198 | -1.27879 |
| Kif23 | NM_024245 | 0.037069 | -1.27888 |
| Rrm2 | NM_009104 | 0.007811 | -1.31595 |
| Hmgb3 | NM_008253 | 0.038337 | -1.31905 |
| Aif11 | NM_145144 | 0.00123 | -1.34203 |
| Prr11 | NM_175563 | 0.042414 | -1.41378 |
| Pole | NM_011132 | 0.035284 | -1.56413 |
| Fhl1 | NM_001077362 | 0.011966 | -1.88461 |
| Tpm1 | NM_001164250 | 0.00086 | -2.03308 |
| Ptgs2 | NM_011198 | 0.032098 | -2.77484 |







| Gene Name | Transcript Name | Shortening P-values | log2 fold change (53bp1 ^{-/-} /53bp1 ^{+/+}) |
|-----------|-----------------|---------------------|---|
| Ube2h | NM_001169577 | -0.02395 | -8.343 |
| Arhgef40 | NM_001145922 | -0.01782 | -3.22011 |
| Krt19 | NM_008471 | -0.01163 | -2.2587 |
| Anp32e | NM_001253757 | -0.02219 | -1.2715 |
| Prc1 | NM_145150 | -0.04845 | -1.16398 |
| Slc38a1 | NM_134086 | -0.0071 | -1.10762 |
| Plk2 | NM_152804 | -0.03949 | -1.10463 |
| Cenpq | NM_031863 | -0.02245 | -0.92818 |
| Plau | NM_008873 | -0.01553 | -0.89162 |
| Cenpa | NM_007681 | -0.04633 | -0.88376 |
| Ctgf | NM_010217 | -0.00731 | -0.87979 |
| Exosc8 | NM_027148 | -0.00198 | -0.85992 |
| Vgll3 | NM_028572 | -0.00483 | -0.84936 |
| Idh3a | NM_029573 | -0.01961 | -0.82096 |
| Socs5 | NM_019654 | -0.01344 | -0.8186 |
| Spcs3 | NM_029701 | -0.03582 | -0.78164 |
| Mat2a | NM_145569 | -0.00162 | -0.77796 |
| Ranbp1 | NM_011239 | -0.00075 | -0.77656 |
| Ank2 | NM_001034168 | -0.02209 | -0.77253 |
| Sdad1 | NM_172713 | -0.00329 | -0.76924 |
| Rbpms2 | NM_028030 | -0.01994 | -0.72429 |
| Mrpl18 | NM_026310 | -0.04215 | -0.71352 |
| Zdhhc2 | NM_178395 | -0.00581 | -0.71235 |
| Ppid | NM_026352 | -0.00261 | -0.68298 |

Table 7. 3' UTR lengthened transcripts in 53bp1^{-/-} MEF





| Coro1c | NM_011779 | -0.02967 | -0.67901 |
|-----------|--------------|-----------|----------|
| Smg7 | NM_001005507 | -0.03417 | -0.67126 |
| Clca1 | NM_009899 | -0.03327 | -0.66482 |
| Ran | NM_009391 | -0.04276 | -0.64228 |
| Eif1ax | NM_025437 | -0.03331 | -0.63277 |
| Gpd2 | NM_010274 | -0.0376 | -0.58975 |
| Adss | NM_007422 | -0.04849 | -0.58125 |
| Etf1 | NM_144866 | -0.01953 | -0.57841 |
| Jub | NM_010590 | -0.0161 | -0.57609 |
| Hdgf | NM_008231 | -0.01257 | -0.57333 |
| Uhrf1bp11 | NM_029166 | -0.04668 | -0.54562 |
| Cnih4 | NM_030131 | -0.03404 | -0.54502 |
| Hn1 | NM_008258 | -3.55E-05 | -0.54253 |
| Gypc | NM_001048207 | -0.02263 | -0.51924 |
| Usp24 | NM_183225 | -0.04631 | -0.51796 |
| Rbm28 | NM_133925 | -0.01292 | -0.51749 |
| Vasp | NM_009499 | -0.0252 | -0.51693 |
| Nudcd3 | NM_173748 | -0.02025 | -0.50556 |
| Pdlim5 | NM_019808 | -0.00738 | -0.50184 |
| Ube2i | NM_001177609 | -6.35E-06 | -0.47246 |
| Hnrnpu | NM_016805 | -0.00132 | -0.45562 |
| Asns | NM_012055 | -0.0106 | -0.45179 |
| Setd8 | NM_030241 | -0.0409 | -0.45146 |
| Yap1 | NM_009534 | -0.00774 | -0.44329 |
| Lrrc58 | NM_177093 | -4.26E-06 | -0.43383 |
| Tnpo3 | NM_177296 | -0.02611 | -0.42062 |
| Mapk1ip11 | NM_178684 | -0.00461 | -0.4182 |





| Polh | NM_030715 | -0.00454 | -0.41662 |
|---------|--------------|-----------|----------|
| Map3k7 | NM_009316 | -0.03896 | -0.41063 |
| Nt5c3 | NM_001252374 | -0.04135 | -0.4044 |
| Cpne1 | NM_170588 | -1.10E-05 | -0.3992 |
| Ube2d3 | NM_025356 | -0.01627 | -0.39627 |
| Csnk1d | NM_139059 | -0.02516 | -0.39133 |
| Ddx5 | NM_007840 | -3.02E-05 | -0.38899 |
| Limd2 | NM_172397 | -1.90E-07 | -0.37468 |
| Ltbp1 | NM_206958 | -0.03017 | -0.37336 |
| Nemf | NM_025441 | -0.02004 | -0.36238 |
| Wapal | NM_001004436 | -0.02606 | -0.35419 |
| Ptbp3 | NM_144904 | -0.01312 | -0.35198 |
| Hspa4 | NM_008300 | -0.00021 | -0.35192 |
| Ccnc | NM_016746 | -0.01944 | -0.34697 |
| Nek7 | NM_021605 | -0.00478 | -0.34121 |
| Vim | NM_011701 | -0.00319 | -0.33981 |
| Tgs1 | NM_054089 | -0.02401 | -0.32988 |
| Rac1 | NM_009007 | -2.08E-10 | -0.31108 |
| Psmd11 | NM_178616 | -0.04362 | -0.30431 |
| Smarca2 | NM_026003 | -0.0389 | -0.30356 |
| Tcf12 | NM_001253864 | -0.00392 | -0.30323 |
| Tcf19 | NM_025674 | -0.02203 | -0.29986 |
| Cast | NM_009817 | -0.0099 | -0.2976 |
| Sec63 | NM_153055 | -0.0156 | -0.28759 |
| Anapc1 | NM_008569 | -0.00379 | -0.28367 |
| Gabpa | NM_008065 | -0.01676 | -0.28232 |
| Ppp4r2 | NM_182939 | -0.01328 | -0.27973 |





| AI314180 | NM_172381 | -0.03992 | -0.27946 |
|----------|--------------|-----------|----------|
| Edc3 | NM_153799 | -0.03544 | -0.27845 |
| Taf12 | NM_025579 | -0.04768 | -0.26958 |
| Fblim1 | NM_133754 | -0.02047 | -0.2634 |
| Rwdd1 | NM_025614 | -0.03822 | -0.26062 |
| Dbnl | NM_013810 | -0.00835 | -0.25507 |
| Prrx1 | NM_175686 | -0.04099 | -0.23663 |
| Rbm8a | NM_025875 | -0.00904 | -0.23455 |
| Cux1 | NM_198602 | -0.00275 | -0.2345 |
| Med29 | NM_026042 | -0.02419 | -0.23114 |
| Parva | NM_020606 | -0.01106 | -0.22403 |
| Col5a1 | NM_015734 | -0.01322 | -0.21923 |
| Pcmt1 | NM_008786 | -0.00617 | -0.2123 |
| Lox12 | NM_033325 | -7.55E-12 | -0.19853 |
| Mettl6 | NM_025907 | -0.04333 | -0.18838 |
| Immp11 | NM_028260 | -0.02063 | -0.18663 |
| Calm1 | NM_009790 | -6.65E-08 | -0.18457 |
| Rbpms | NM_001042674 | -0.02886 | -0.18401 |
| Ptgr2 | NM_001252626 | -0.02184 | -0.17782 |
| Fam126a | NM_053090 | -0.03262 | -0.17759 |
| Stt3b | NM_024222 | -0.00094 | -0.17675 |
| Lsm14a | NM_025948 | -0.02687 | -0.171 |
| Sdccag3 | NM_026563 | -0.00824 | -0.16887 |
| Eif2s2 | NM_026030 | -6.84E-06 | -0.16498 |
| Brix1 | NM_026396 | -0.00201 | -0.16374 |
| Eif2c2 | NM_153178 | -0.00896 | -0.16368 |
| Blmh | NM_178645 | -0.00419 | -0.1595 |





| Rab14 | NM_026697 | -0.04943 | -0.1559 |
|---------------|--------------|-----------|----------|
| Szrd1 | NM_001025608 | -0.04839 | -0.15536 |
| Ctsc | NM_009982 | -0.01298 | -0.14757 |
| Hnrnpul2 | NM_001081196 | -0.0306 | -0.13921 |
| Wipi2 | NM_178398 | -0.00387 | -0.12334 |
| R3hdm1 | NM_181750 | -0.03698 | -0.12159 |
| Nudcd1 | NM_026149 | -0.01296 | -0.11045 |
| Tm9sf3 | NM_133352 | -1.05E-05 | -0.10797 |
| Pafah1b1 | NM_013625 | -0.00787 | -0.1074 |
| Aggf1 | NM_025630 | -0.02028 | -0.10662 |
| Ube2a | NM_019668 | -0.04829 | -0.10203 |
| Tmem209 | NM_178625 | -0.01182 | -0.09975 |
| Rps3 | NM_012052 | -0.02242 | -0.09838 |
| Tceb3 | NM_013736 | -0.0372 | -0.09742 |
| Lmbrd2 | NM_177178 | -0.01841 | -0.09534 |
| Pafah1b2 | NM_008775 | -0.03301 | -0.08906 |
| Tsc22d2 | NM_001081229 | -0.00233 | -0.0889 |
| Cds2 | NM_138651 | -0.01801 | -0.08781 |
| Cmip | NM_001163262 | -0.04032 | -0.08727 |
| 2810403A07Rik | NM_028814 | -0.01293 | -0.08389 |
| Vapa | NM_013933 | -0.03436 | -0.07439 |
| Sike1 | NM_025679 | -0.03169 | -0.0689 |
| Tex261 | NM_009357 | -0.01497 | -0.0639 |
| Usp45 | NM_152825 | -0.02754 | -0.06382 |
| Spryd7 | NM_025697 | -0.01137 | -0.0616 |
| Pdxk | NM_172134 | -0.01585 | -0.06133 |
| Luc7l | NM_028190 | -0.03903 | -0.05266 |





| Apaf1 | NM_001042558 | -0.03347 | -0.04888 |
|---------|--------------|----------|----------|
| Pmepa1 | NM_022995 | -0.00656 | -0.04279 |
| Csnk1d | NM_027874 | -0.04344 | -0.04195 |
| Mtmr2 | NM_023858 | -0.04763 | -0.03378 |
| Vamp3 | NM_009498 | -0.01857 | -0.03094 |
| Ppm11 | NM_178726 | -0.0206 | -0.02825 |
| Rabl6 | NM_001024616 | -0.00127 | -0.02741 |
| Slc35a1 | NM_011895 | -0.0049 | -0.0271 |
| Cyb5b | NM_025558 | -0.00231 | -0.02695 |
| Zfp106 | NM_011743 | -0.0003 | -0.021 |
| Copg | NM_017477 | -0.03008 | -0.01886 |
| Fam149b | NM_001024512 | -0.02687 | -0.01382 |
| Ldlrad4 | NM_172631 | -0.01424 | -0.00912 |
| Atrnl1 | NM_181415 | -0.04165 | -0.0079 |
| Epn2 | NM_010148 | -0.04064 | -0.00349 |
| Rcbtb1 | NM_027764 | -0.01686 | -0.00188 |
| Dpp8 | NM_028906 | -0.00344 | 0.013792 |
| Hdac4 | NM_207225 | -0.04889 | 0.013885 |
| Snap29 | NM_023348 | -0.02566 | 0.022669 |
| Tmem14c | NM_025387 | -0.01414 | 0.024402 |
| Psmd8 | NM_026545 | -0.00061 | 0.025359 |
| Mc11 | NM_008562 | -0.00833 | 0.029337 |
| Arl14ep | NM_001025102 | -0.0383 | 0.036042 |
| Cggbp1 | NM_178647 | -0.01243 | 0.036539 |
| Timp3 | NM_011595 | -0.00091 | 0.045351 |
| Arl6ip1 | NM_019419 | -0.0358 | 0.051163 |
| Ube213 | NM_009456 | -0.00106 | 0.061472 |





| Ррр3са | NM_008913 | -0.01913 | 0.063797 |
|----------|--------------|-----------|----------|
| Dcaf7 | NM_027946 | -8.27E-05 | 0.068055 |
| Faf2 | NM_178397 | -0.00574 | 0.068576 |
| Ganab | NM_008060 | -0.04934 | 0.072626 |
| Kank2 | NM_145611 | -0.00974 | 0.073747 |
| Rrbp1 | NM_024281 | -0.01421 | 0.074052 |
| Unc5c | NM_009472 | -0.04343 | 0.08587 |
| Ubtd2 | NM_173784 | -0.00449 | 0.085974 |
| Lman2 | NM_025828 | -0.01896 | 0.088069 |
| Rbx1 | NM_019712 | -0.01442 | 0.089079 |
| Camk2d | NM_001025438 | -0.01493 | 0.090691 |
| Pcgf3 | NM_172716 | -0.00176 | 0.092171 |
| Adipor2 | NM_197985 | -0.03369 | 0.092298 |
| Dnajc3 | NM_008929 | -0.03221 | 0.092878 |
| Postn | NM_001198766 | -0.00011 | 0.098695 |
| Trim23 | NM_030731 | -0.00954 | 0.100938 |
| Sacm11 | NM_030692 | -0.02198 | 0.103742 |
| Tomm20 | NM_024214 | -8.00E-06 | 0.105042 |
| Prps2 | NM_026662 | -0.04376 | 0.106142 |
| Cog3 | NM_177381 | -0.01716 | 0.108831 |
| Rpl22 | NM_009079 | -1.39E-09 | 0.116099 |
| Zfand3 | NM_148926 | -0.02784 | 0.11671 |
| Col1a2 | NM_007743 | -3.09E-05 | 0.116721 |
| Rab22a | NM_024436 | -0.02975 | 0.117104 |
| Ier3ip1 | NM_025409 | -0.00267 | 0.12874 |
| AU022252 | NM_001012400 | -0.03553 | 0.134758 |
| Gng12 | NM_001177557 | -0.00022 | 0.135212 |





| Zmat3 | NM_009517 | -0.00023 | 0.139435 |
|---------|--------------|-----------|----------|
| Hook3 | NM_207659 | -0.03335 | 0.145563 |
| Sh3glb1 | NM_019464 | -0.00063 | 0.158699 |
| Ccng1 | NM_009831 | -0.0024 | 0.16093 |
| Tmed10 | NM_026775 | -2.81E-06 | 0.169934 |
| Ghitm | NM_078478 | -0.0021 | 0.171758 |
| Ubxn4 | NM_026390 | -0.00356 | 0.174127 |
| Adam10 | NM_007399 | -0.00713 | 0.181839 |
| Nrf1 | NM_010938 | -0.0407 | 0.182277 |
| Rspry1 | NM_026274 | -0.02411 | 0.189472 |
| Reep5 | NM_007874 | -0.00011 | 0.212931 |
| Ripk1 | NM_009068 | -0.03499 | 0.223971 |
| Cdkn1b | NM_009875 | -0.00675 | 0.224832 |
| Ubfd1 | NM_138589 | -0.00478 | 0.235754 |
| Bnip31 | NM_009761 | -0.00742 | 0.237349 |
| Rpl27a | NM_011975 | -0.02243 | 0.24306 |
| Alcam | NM_009655 | -0.01222 | 0.245407 |
| Rpl15 | NM_025586 | -0.00625 | 0.245799 |
| Ccni | NM_017367 | -0.01066 | 0.251712 |
| Extl3 | NM_018788 | -0.02896 | 0.257441 |
| Btbd1 | NM_146193 | -0.011 | 0.259618 |
| Mpp7 | NM_001081287 | -0.03879 | 0.260221 |
| Thbs2 | NM_011581 | -0.0121 | 0.261684 |
| Clmp | NM_133733 | -0.04449 | 0.267566 |
| Nedd4 | NM_010890 | -0.00055 | 0.268983 |
| Sccpdh | NM_178653 | -0.02042 | 0.271529 |
| Exoc3 | NM_177333 | -0.04308 | 0.274809 |





| Sec22b | NM_011342 | -0.02587 | 0.278451 |
|---------------|--------------|-----------|----------|
| Stard4 | NM_133774 | -0.01299 | 0.291018 |
| Vma21 | NM_001081356 | -0.00736 | 0.295919 |
| Cnih | NM_009919 | -0.00051 | 0.297342 |
| Mfap1a | NM_026220 | -0.01124 | 0.299242 |
| Zdhhc20 | NM_029492 | -0.003 | 0.299684 |
| Ube2b | NM_009458 | -0.017 | 0.301178 |
| Arhgap5 | NM_009706 | -0.00617 | 0.304093 |
| Cacna2d1 | NM_009784 | -0.01144 | 0.315623 |
| Aldh3a2 | NM_007437 | -0.02019 | 0.319175 |
| 2410131K14Rik | NM_001081236 | -0.00027 | 0.320617 |
| Gnpda2 | NM_001038015 | -0.0263 | 0.324312 |
| H3f3a | NM_008210 | -0.02249 | 0.328287 |
| Acox1 | NM_015729 | -0.04231 | 0.33503 |
| Col5a2 | NM_007737 | -0.00043 | 0.359119 |
| Oxct1 | NM_024188 | -0.00168 | 0.374614 |
| Prrc2b | NM_001159634 | -0.02841 | 0.378799 |
| Hsd17b7 | NM_010476 | -0.02278 | 0.389723 |
| Gpr107 | NM_178760 | -0.00683 | 0.391539 |
| Prrc1 | NM_028447 | -8.64E-05 | 0.395095 |
| Sh3pxd2a | NM_008018 | -0.01133 | 0.403305 |
| Ankrd12 | NM_001025572 | -0.04745 | 0.406671 |
| Ebf1 | NM_007897 | -0.00614 | 0.422417 |
| Rcn1 | NM_009037 | -0.03531 | 0.437749 |
| Zbtb7b | NM_009565 | -0.02519 | 0.444149 |
| Pik3r3 | NM_181585 | -0.04182 | 0.446487 |
| Dhcr7 | NM_007856 | -0.03166 | 0.455317 |





| Rab11fip3 | NM_001162868 | -0.01382 | 0.456337 |
|-----------|--------------|-----------|----------|
| Mdfic | NM_175088 | -0.02043 | 0.462409 |
| Atp5s | NM_026536 | -0.00505 | 0.46301 |
| Sc5d | NM_172769 | -0.03315 | 0.48571 |
| AW549877 | NM_145930 | -0.01524 | 0.486831 |
| Pdgfd | NM_027924 | -0.03878 | 0.506482 |
| P4ha1 | NM_011030 | -0.02611 | 0.525364 |
| Dync1li2 | NM_001013380 | -0.00082 | 0.526026 |
| App | NM_001198823 | -0.00943 | 0.533024 |
| Lix11 | NM_001163170 | -0.02478 | 0.541819 |
| Tmem106b | NM_027992 | -0.00055 | 0.546943 |
| Timp2 | NM_011594 | -4.93E-22 | 0.550144 |
| Oaz2 | NM_010952 | -0.0366 | 0.55215 |
| Mlec | NM_175403 | -4.59E-11 | 0.571506 |
| Scp2 | NM_011327 | -0.03601 | 0.575564 |
| Tpp1 | NM_009906 | -0.0019 | 0.58674 |
| Scarb2 | NM_007644 | -0.0001 | 0.589584 |
| Zfp874a | NM_177712 | -0.02027 | 0.594137 |
| Chchd5 | NM_025395 | -9.28E-05 | 0.595418 |
| Map11c3b | NM_026160 | -4.77E-06 | 0.60303 |
| Lox | NM_010728 | -0.01098 | 0.689869 |
| Camk2d | NM_023813 | -0.02327 | 0.723367 |
| Zfp810 | NM_145612 | -0.03626 | 0.727511 |
| Phactr2 | NM_001033257 | -0.03218 | 0.730095 |
| Impact | NM_008378 | -0.04406 | 0.732997 |
| Triqk | NM_001171801 | -0.00706 | 0.736895 |
| Sirt2 | NM_001122766 | -0.00245 | 0.749707 |





| St3gal5 | NM_001035228 | -0.02628 | 0.763436 |
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| Laptm4a | NM_008640 | -0.01025 | 0.766407 |
| Bloc1s6 | NM_019788 | -0.00015 | 0.771208 |
| ORF63 | NM_144854 | -0.04544 | 0.778333 |
| Plxdc2 | NM_026162 | -0.01911 | 0.816558 |
| Spp1 | NM_001204203 | -0.02697 | 0.826694 |
| Ptp4a2 | NM_001164745 | -0.00891 | 0.875069 |
| C1qtnf3 | NM_001204134 | -0.02717 | 0.903337 |
| Col3a1 | NM_009930 | -0.00444 | 0.907479 |
| Dkk3 | NM_015814 | -0.00067 | 0.980646 |
| Gas6 | NM_019521 | -0.04498 | 1.0206 |
| Colec12 | NM_130449 | -0.02781 | 1.05743 |
| Rnase4 | NM_021472 | -0.00222 | 1.0934 |
| Dnmt3a | NM_007872 | -0.0242 | 1.16792 |
| Serpine2 | NM_009255 | -0.00628 | 1.18851 |
| Klhl24 | NM_029436 | -0.01337 | 1.21799 |
| Perp | NM_022032 | -0.02279 | 1.35249 |
| Snx10 | NM_001127348 | -0.04857 | 1.56591 |
| Kif1a | NM_001110315 | -0.04703 | 1.64168 |
| Pdcd6ip | NM_001164677 | -0.0005 | 2.17136 |
| Rasgrp3 | NM_001166493 | -0.00115 | 2.24249 |
| Gpr64 | NM_001079847 | -0.04816 | 2.34329 |
| Eln | NM_007925 | -0.04795 | 2.66854 |
| Fbxo34 | NM_030236 | -0.03188 | 3.58743 |
| Angel2 | NM_001199020 | -0.03461 | 6.02346 |

