Loss of cyclin A2 in murine colonic epithelial cells disrupts colon homeostasis by triggering DNA damage and dysplasia and high cyclin A2 expression is a good-prognosis factor in patients with colorectal cancer

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The authors declare to have no conflicts of interest

Keywords: cyclin A2, murine colitis-associated cancer model, colorectal cancer (CRC), DNA damage, regenerative pathways, dysplasia, Consensus Molecular Subtypes (CMS), prognosis.

Abstract

To clarify the function of cyclin A2 in colon homeostasis and colorectal cancer (CRC) we generated mice deficient for cyclin A2 in colonic epithelial cells (CEC). Colons of those mice displayed architectural changes in the mucosa, and signs of inflammation as well as an increased proliferation of CEC associated with the appearance of low- and high-grade dysplasia. The main initial events triggering those alterations in cyclin A2 deficient CEC appear to be abnormal mitoses and DNA damage. Cyclin A2 deletion in CEC promoted the development of dysplasia and adenocarcinomas in the murine colitis-associated cancer model. We next explored the status of cyclin A2 expression in clinical CRC samples at the mRNA and protein level and found higher expression in tumors of stage I and II patients compared to those of stage III and IV. A meta-analysis of 11 transcriptome datasets comprising 2,239 primary CRC tumors displayed different *CCNA2* (the mRNA coding for cyclin A2) expression levels among the CRC tumor subtypes with highest in CMS1 and lowest in CMS4. Moreover, high expression of *CCNA2* was found to be a good prognosis factor independently from other prognostic factors for the CMS1, CMS3 and CMS4 subtypes.

Cyclin A2 deficiency in colonic epithelial cells leads to dysplasia

Cyclin A2 expression levels predict the outcome in colorectal cancer

**Better
Prognosis**

**Bad
Prognosis**

**in Consensus Molecular
Subtypes:**
CMS1
CMS3
CMS4

~~CCNA2~~

Abnormal
mitoses

DNA damage

Elevated β -catenin
activation

Crypts disruption
Inflammation
 $\uparrow$ Regeneration

Dysplasia

Colorectal cancer (CRC) is one of the leading causes of malignancy-related death worldwide with increasing incidence (International Agency for Research on Cancer, WHO; <http://gco.iarc.fr/today/fact-sheets-cancers>). Disease status of patients can be classified according to histologic tumor grade (differentiation) and anatomic extent of disease (*i.e.*, (TNM, tumors/nodes/metastases, stages I–IV), which describes tumor growth into the wall of rectum or colon (stage II), involvement of regional lymph nodes (stage III) and metastatic spread to other organs (stage IV)(2). Once metastasis has developed, 5-year survival rates following surgery are below 10% (3). Also, adjuvant chemotherapy after surgical resection for stage III CRC and high-risk stage II tumor (*i.e.*, tumors with poor differentiation, T4 classification, lymphovascular invasion, perineural invasion) increased the patients survival rates, but many patients will relapse or develop distant metastases (4, 5). Thus, there is an urgent need to better understand the molecular changes leading to the development of CRC to identify novel therapeutic strategies.

An important risk factor for CRC development is inflammation. Early-onset of chronic inflammatory bowel disease and widespread manifestation (*i.e.*, pancolitis) have a higher probability of developing CRC (6). Besides, inherited mutations have been identified for 5% of CRC patients, with diseases traditionally divided into polyposis syndromes, such as the Familial Adenomatous Polyposis (FAP), and non-polyposis syndromes like the Lynch Syndrome, also known as Hereditary Non-Polyposis Colorectal Cancer (HNPCC) (7). Sporadic CRC is a molecularly highly heterogeneous disease. Research teams from different institutions performed recently a meta-analysis on the different reported sub-classifications to define the existence of four distinct gene expression–based subtypes (Consensus Molecular Subtypes or CMS), *i.e.* immune (CMS1), canonical (CMS2), metabolic (CMS3) and mesenchymal (CMS4) subtype (8). Each CMS has a unique biology and gene expression pattern (8) providing now a frame for designing subtype-specific and personalized treatments. Indeed, CMS display different clinical features including prognosis and response to therapies, with CMS4 having the worst prognosis.

Cyclin A2 is an established regulator of cell proliferation and has been used for molecular diagnostics as a proliferation marker (9). However, several studies suggest that cell cycle regulators can exert additional functions, well beyond cell cycle control (10–12, 12–16). For example, cyclin-CDK inhibitors, p21, p27 and p57, as well as cyclin D1, have been involved in the control of apoptosis, transcription, cytoskeletal dynamics as well as cell migration (12, 13, 13, 16)

We described a role for cyclin A2 in the regulation of cell migration and invasion (17–

19). More specifically, we found that deletion of cyclin A2 in a murine mammary gland epithelial cell line (NMuMG) induces epithelial to mesenchymal transition (EMT) and increases invasiveness *in vitro* as well as *in vivo* in the avian embryo model (18, 19). This concurs with a recent report that identified cyclin A2 as a key regulatory component in EMT and metastasis of lung cancer cells by modulating the TGFbeta signaling pathway (20).

Few studies investigated the role of cyclin A2 in tumor development *in vivo* using genetically altered mice. One study described the generation of mutant mice with reduced cyclin A2 expression. These mice displayed spontaneous tumor development, in particular in the lung, and were also more susceptible to chemically-induced skin cancer (21). On the other hand, tissue-specific cyclin A2 inactivation in the liver was shown to impair hepatocellular carcinoma development and recapitulated the phenotype observed in absence of cyclin-dependent kinase 2 (CDK2) (22). These observations point to a tissue-specific role of cyclin A2.

We previously reported lower cyclin A2 expression in hepatic metastases compared to primary tumors derived from the same CRC patients (17). This prompted us to investigate the role of cyclin A2 in colon homeostasis and carcinogenesis using cell type-specific knockout mice.

Results

Generation of mice deficient for cyclin A2 in the colonic epithelium

We established two mouse models to study the function of cyclin A2 in colon homeostasis and colorectal cancer development by employing the previously established conditional cyclin A2 (*Ccna2^{fl/fl}*) strain that is based on the Cre/LoxP system (23). Constitutive deletion of cyclin A2 in the intestinal epithelium was obtained by crossing cyclin A2 conditional knock-out (KO) mice with the *villin-Cre* mouse strain (*VilCre*). Expression of Cre under the villin promoter was shown to be active in intestinal epithelial cells (24). In addition, we crossed *Ccna2^{fl/fl}* mice with the inducible *Cre* mouse strain, *villin-CreERT²* (*VilCreERT2*), in which *Ccna2* is excised upon intraperitoneal injection of tamoxifen (24).

Efficient inactivation of cyclin A2 in purified colonic epithelial cells (CEC) of *VilCreCcna2^{fl/fl}* mice was confirmed in a RNA sequencing experiment validating the deletion of exons 2 to 7 of the *Ccna2* gene (Supplemental Fig 2A) as well as WB analysis (Fig 1A, left panel). Two tamoxifen injections at day 0 and 2 were sufficient to abrogate cyclin A2 expression in *VilCreERT2Ccna2^{fl/fl}* mice (Fig 1A, right panel).

We also confirmed cyclin A2 loss in colonic crypts of *VilCreCcna2^{fl/fl}* mice by immunohistochemistry (Fig 1B). For this, we analyzed the distal region, close to the anus, as well as the proximal region, close to the caecum (Supplemental Fig 1A), since biological and clinical differences for both segments are well established (25). In both regions we detected cyclin A2 expression in epithelial cells of the lower part of the crypts in control mice, whereas cyclin A2 expression was undetectable in colonic crypts of cyclin A2 KO mice (Fig 1B).

Cyclin A2 depletion in the colon induces epithelial changes in the mucosa and inflammation

During the analysis of cyclin A2 expression we noted epithelial changes in the colonic crypts as well as architectural changes in the mucosa of cyclin A2-deficient mice, but not in control animals (Fig 1B and Supplemental Fig 1C). We also detected immune cell infiltration in cyclin A2-deficient colons. The immune cell infiltrates exhibited morphological characteristics of macrophages and were immunostained using the macrophage marker F4/80. This revealed an increased number of F4/80-positive cells, about 2-fold in constitutive (Fig 1C and Supplemental Fig 1B, left panel) and 1.5-fold in induced (Supplemental Fig 1B, right panel) cyclin A2-deficient colons versus normal ones. Macrophages can be important producers of the pro-inflammatory cytokine IL6 (26) and in fact IL6 was detectable in the stroma of cyclin A2-deficient colons, but not in those of control mice (Fig 1D).

Finally, about 15% males and 10% females of *VilCreCcna2fl/fl* mice developed rectal prolapses within the first three months of age, which can be considered as an indicator of inflammation (27). Taken together, cyclin A2 deficiency in colonic epithelial cells induces epithelial and architectural changes of the colonic crypts and different signs of inflammation.

RNA sequencing analysis reveals a prominent altered expression of genes involved in cell cycle, mitotic process, chromosome segregation and DNA double-strand break repair

To gain insights into the molecular traits of *Ccna2* mutant mice, RNA sequencing was performed using CEC isolated from the proximal colon of *Ccna2fl/fl* mice (n=4) and *VilCreCcna2fl/fl* mice (n = 4) at four weeks of age. At this age *VilCreCcna2fl/fl* mice already display visible distortion of the crypt architecture that was validated by IHC analysis for the distal region of colons employed for this experiment.

The supervised analysis of the RNAseq data identified 469 upregulated and 640 downregulated genes in *VilCreCcna2fl/fl* mice samples versus *Ccna2fl/fl* mice samples (fold change > 2, adjusted p-value |FDR| >0.05) (Table 1). Pathways analyses of the most altered genes revealed that the most significant biological functions associated with the genes upregulated in *VilCreCcna2fl/fl* mice were related to cell cycle (nuclear division and mitosis, regulation of mitotic cell cycle, $p < 2.79 \times 10^{-12}$), cellular assembly and organization (chromosome segregation, chromatin segregation, $p < 3.01 \times 10^{-9}$), as well as DNA replication, recombination and repair (double-strand break repair and homologous recombination, $p < 3.01 \times 10^{-9}$) (Table 1, Supplemental Table 4 and Supplemental Fig 2B). In line with this, the analysis highlighted increased levels of *Mki67* (coding for Ki-67), *Brca1*, *Brca2*, *Rad18*, *Rad51*, *Blm*, *Exo1* or *Rpa1* (genes involved in the double-strand break repair mechanism) as well as *Cdc45*, *Bub1*, *Mad2* and *Rad21* (genes related to checkpoint controls of the cell cycle and to abnormal mitosis) (Supplemental Fig 2C).

Cyclin A2 loss in colonic epithelial cells leads to increased proliferation of colonic epithelial cells and onset of dysplasia

To validate the RNAseq results described above, we first tested the proliferation status of cyclin A2-deficient colonic epithelial cells by measuring BrdU-uptake. For this, mice received an intraperitoneal injection of the nucleoside analog BrdU and were sacrificed two hours later to isolate colons. The subsequent IHC analysis was performed by dissecting the different regions of the colon as described above (Supplemental Fig 1A) and by separating the bottom, middle and top parts of the crypts. As shown in Figure 2A, BrdU-positive cells were mainly

located in the bottom of the crypts of control mice. In contrast, BrdU incorporation was most prominent in the middle part of the cyclin A2-deficient crypts with an average of more than 4 cells per crypt ($n=60$, $p<0.0001$). Moreover, BrdU-positive cells were detectable in the upper crypt part of *VilCreCna2^{fl/fl}* but not in control mice. These specific profiles of BrdU incorporation of cyclin A2-deficient colonic epithelial cells were observed equally in distal, transverse as well as the proximal region. To confirm the increased proliferation rate of cyclin A2-deficient CEC, we analyzed Ki-67 expression, which is expressed during all active phases of the cell cycle (i.e., G1, S, G2, and M), but is absent in resting cells (28). Ki-67 expression in the distal region of colons was detectable at the bottom of the crypts in colons from control mice but localized to the middle and upper parts of the colonic crypts from *Cna2* knockout animals. A similar pattern was observed in the proximal region of colons (Fig 2B, Supplemental Fig 3A and 3B) of constitutive as well as induced knockout mice (Supplemental Fig 3C) therefore confirming the observation made in the BrdU-uptake experiment. Thus, the elevated expression levels of Ki-67 detected by immunostaining in cyclin A2-deficient colons concurs with the upregulation of *Mki67* transcripts detected in the RNAseq analysis.

Strikingly, colons deficient for cyclin A2, either constitutive or induced, revealed low- and high-grade dysplasia in around 80% of the mice analyzed that was mostly found in the proximal region (Fig 3A and B). Constitutive knockout mice showed an average occurrence of 4 low-grade dysplasia and nearly 2 high-grade dysplasia per colon, while induced knockout mice displayed a mean of about 3 low-grade and high-grade dysplasia per colon, whereas none were detectable in the control mice (Fig 3C and D). Notably, dysplasia has been already detectable eight days after tamoxifen injection in *VilCreERT2Cna2^{fl/fl}* mice.

Cyclin A2 depletion in colonic epithelial cells leads to abnormal mitoses and DNA damage

We further explored cell cycle features and noticed an increased incidence of mitoses within the crypts when analyzing colons of constitutive and induced knockout mice (at day 8). Colons with constitutive and induced-*Cna2* deletion showed an average of 13 and 24 mitotic figures per mm^2 , respectively, whereas in control animals in average only 2 per mm^2 were found ($n=5$, $p<0.01$ for the constitutive as well as induced knockout mouse strain, Fig 4A and B). Besides, CEC deficient for cyclin A2 harbored nuclear pleomorphism characterized by increased nuclear size compared to controls (Figure 4A and Supplemental Fig 4). Finally, abnormal mitosis, detected by combined staining for α -tubulin to visualize mitotic spindles, γ -

tubulin to label centrosomes and DAPI to identify chromosomes, were prominent in induced cyclin A2-deficient but not control colons (Supplemental Fig 5 A and B).

Defective mitoses can lead to DNA damage (29), and we thus analyzed colons of tamoxifen-treated *VilCreERT2Ccna2fl/fl* mice by IHC using a marker for double-strand breaks (DSB), *i.e.* an anti- γ H2AX antibody, and found an average of more than 70 γ H2AX positive cells/mm² in cyclin A2-deficient CEC compared to 40 in control ones (Fig 4C, n=4, p<0.05). Mre11 is part of the NMR complex involved in the resolution of double-strand DNA (30). We therefore analyzed Mre11 foci formation by confocal microscopy analysis (31) and found that Mre11 foci formation was significantly increased in *VilCreCcna2fl/fl* nuclei (Fig 4D).

Altogether, the alterations found at the protein level correlate with the RNAseq results underpinning the presence of cell cycle perturbations, increased DNA damage and DNA double-strand break repair pathway activation in cyclin A2-deficient CEC.

Activation of regenerative pathways in cyclin A2-deficient colons

Tissue injury induces inflammation and in turn a regenerative response (32). Several pathways have been described that can promote tissue repair in the intestine including YAP/TAZ, NF κ B and WNT/ β -catenin signaling (32, 33). We have previously shown that cyclin A2 inactivation in epithelial cells *in vitro* leads to increased β -catenin stability and activity (18, 19). For this reason we first tested β -catenin expression in the colons of constitutive knockout mice by IHC analysis revealing increased nuclear localization of β -catenin in CEC of cyclin A2 deficient colons by IHC (Fig 5A).

In addition, Western blot analysis of nuclear extracts derived from the constitutive knockout mice displayed elevated levels of both active β -catenin as well as the NF κ B p65 subunit (Fig 5B). We also detected TAZ expression in crypts as well as surrounding stroma in colons of *VilCreCcna2fl/fl* mice (Fig 5 F).

We next examined the activation sequence of the different pathways modulated in cyclin A2-deficient CEC at the protein level. For this, we took advantage of the inducible Cre model (*VilCreERT2Ccna2fl/fl* mice) and examined the altered pathways four days upon *Ccna2* depletion when colons still not display alterations such as changes of crypt architecture or immune cell infiltration (lower panel in Fig 5B and C). Expression levels of active β -catenin and NF κ B were analyzed by Western blot (Fig 5C) and those of γ H2AX and TAZ by IHC (Fig 5E-G). Besides, quantification of mitoses was performed on HE stained colons from induced knockout mice (Fig 5D). Elevated levels of active β -catenin, mitoses as well as γ H2AX were

already detectable at day 4 in cyclin A2-deficient CEC (Fig 5C-E), whereas NFκB and TAZ levels were unaltered at that time point between knockout and control animals (Fig 5C and 5G).

Cyclin A2 deficiency in CEC promotes colon carcinogenesis in mice

To test whether cyclin A2-deficiency modulates colon carcinogenesis, we subjected *VilCreCcna2^{fl/fl}* and *Ccna2^{fl/fl}* mice to chemically induced colon carcinogenesis, an established mouse model resembling the human pathology (39). This model depends on the intraperitoneal injection of the mutagen azoxymethane (AOM) and the subsequent induction of inflammation by adding dextran sodium sulphate (DSS) in the drinking water and is therefore named colitis-associated carcinogenesis (CAC). DSS is toxic to mucosal epithelial cells specifically in the colon, and the destruction of the mucosal barrier leads to chronic inflammation. In the classical protocol mice receive three DSS treatments for 5 days within 60 days. *VilCreCcna2^{fl/fl}* mice displayed an about twofold more important weight loss at days 10 to 15 following the first DSS administration compared to control mice (Supplemental Fig 6B). We therefore decided to not expose the mice to additional DSS administrations (Supplemental Fig 6A). Mice were sacrificed sixty days after the AOM injection and colons isolated and embedded in paraffin. The grade of colitis was evaluated as described in Supplemental Table 2 (40) and was significantly enhanced in cyclin A2-deficient mice (average score of 4.5 for the mutant against 1.5 for the control animals, n=6, p≤0.05), which notably included submucosa as well as transmural disruption by immune cell infiltration that was not detectable in control mice (Fig 6A-B). The colon architecture was strongly affected in cyclin A2-deficient mice (Fig 6C) and incidence of low- and high-grade dysplasia was significantly higher in *VilCreCcna2^{fl/fl}* mice and rather rare in control mice (Fig 6D). Moreover, the surfaces of both low- and high-grade dysplasia were significantly higher in cyclin A2-deficient *versus* control animals (Fig 6E). In addition, adenocarcinomas were only detectable in cyclin A2 mutant mice, but not in control animals (Fig 6D-E). IHC analysis revealed increased expression levels of activated β-catenin, γH2AX as well as IL-6 in cyclin A2-deficient colons by comparison with controls (Fig 7).

Altogether, these observations show that cyclin A2 deficiency in CEC promotes chemically-induced colon carcinogenesis in mice.

Cyclin A2 expression in CRC patient has a prognostic value

Our finding that cyclin A2 deficiency in mice promotes spontaneous development of dysplasia in the colon as well as induced colon carcinogenesis prompted us to perform a meta-analysis of *CCNA2* expression on available public datasets (Supplemental Table 5). The characteristics of the 2,239 patients with CRC profiled are summarized in Table 2. Briefly, the median patients' age was 68 years (range, 19 to 97), and the sex-ratio was balanced with 47% of females. The most frequent location was the right colon, followed by the left colon and sigmoid. The pathological stage included more stages III-IV than I-II. The pathological grade was mainly 2, and the mismatch repair (MMR) status was microsatellite stable (MSS) in most of cases. The CMS classification identified 20% of samples as CMS1 (microsatellite instability immune), 32% as CMS2 (canonical), 17% as CMS3 (metabolic), and 31% as CMS4 (mesenchymal).

CCNA2 expression was heterogeneous across the 2,239 primary CRC tumors analyzed (Fig 9A). Compared to normal colon (NC) tissues, *CCNA2* transcripts are up-regulated in most of the primary tumors. In contrast, expression levels in metastases of CRC (ANOVA, $p=4.16 \times 10^{-54}$) were significantly lower in comparison to those in primary tumors (Fig 8A). In addition, *CCNA2* transcript levels were found to be lower in the stages III/IV *versus* stages I/II (Fig 8B). In parallel, we evaluated cyclin A2 protein expression by IHC on a TMA of CRC biopsies taken from 65 patients from stage I to IV (described in Supplemental Table 3) employing two different cyclin A2 antibodies (Fig 8C and Supplemental Fig 8A; see material and methods). Cyclin A2 expression was quantified as the number of epithelial positive cells per mm^2 of tumor area. The IHC analysis confirmed the transcriptome data by revealing cyclin A2 protein levels to be lower in stages III and IV *versus* early stages (Fig 8C and Supplemental Fig 8A). In addition, we observed lower cyclin A2 levels in stage II MSS compared with MSI tumors (Fig 8C and Supplemental Fig 8A). We next evaluated *CCNA2* levels according to the CMS consensus classification (8) and observed that *CCNA2* expression was the highest in the CMS1 class, the lowest in the CMS4 class, and intermediary in CMS2 and CMS3 classes (Fig 8E).

CCNA2 expression was then measured as discrete value after comparison with the median expression of the 95 normal colon samples; upregulation, thereafter designated “*CCNA2*^{high}” (N=1139), was defined by a CRC/NC ratio ≥ 2 and no upregulation (“*CCNA2*^{non-high}”; N=1100), by a CRC/NC ratio < 2 . We searched for correlations between the *CCNA2*^{high} or *CCNA2*^{non-high} classification and the clinicopathological and molecular variables of the 2239 primary CRC samples (Table 2) and found no correlation between cyclin A2 mRNA levels and patients' age or sex. By contrast, when compared with the *CCNA2*^{non-high}, the *CCNA2*^{high} class was associated (Fisher's exact test) with more frequent right and left tumor locations ($p=2.04 \times 10^{-02}$), pathological

stages I and II ($p=3.19e^{-08}$; Table 2 and Fig 8B), pathological grade 3 ($p=5.99e^{-03}$) as well as MSI status ($p=1.48e^{-05}$) and CMS1 molecular subtype ($p=1.00e^{-06}$).

Relapse-Free-Survival (RFS) data were available for 1,882 out of 2,239 operated pCRC samples. The median follow-up was 42 months (range, 1-212); 496 patients (26%) displayed an event, and the 5-year RFS was 70% (95%CI 68-73; Supplemental Fig 7A). Interestingly, the clinical outcome was different between the two *CCNA2*-based classes, with 281 events (31%) in the *CCNA2*^{non-high} class (N=894) versus 215 events (22%) in the *CCNA2*^{high} class (N=988; $p=2.32e^{-06}$, Fisher's exact test; Table 2). The 5-year relapse-free survival (RFS) was lower (65%) for *CCNA2*^{non-high} patients (95%CI 61-69) versus 75% (95%CI 72-79) for the *CCNA2*^{high} class, respectively ($p=8.86e^{-06}$; Fig 8D). In univariate analysis for RFS (Table 3), the Hazard Ratio (HR) for the event was 0.67 (95%CI 0.56-0.80) in the *CCNA2*^{high} class when compared to the *CCNA2*^{non-high} class ($p=1.01e^{-05}$, Wald test). Other variables associated with relapse free survival (RFS) included the tumor location ($p=1.35e^{-06}$), the pathological stage ($p=2.78e^{-15}$), the MMR status ($p=2.97e^{-04}$) (with the *CCNA2*^{non-high} class being predominantly associated to the MSS status therefore concurring with the results obtained with the TMA) unlike patients' age and sex and pathological grade. In multivariate analysis (Table 3), the *CCNA2*-based classification remained associated with RFS ($p=2.99e^{-03}$, Wald test), as well as tumor location (right and sigmoid colon), pathological stage, and MMR status. Of note, it remained significant ($p=4.13e^{-04}$) in multivariate analysis incorporating the CMS molecular classification (Table 3). As shown in Fig 8F, the RFS of the eight classes defined by both the *CCNA2* expression and the CMS was affected ($p=1.82e^{-06}$, log-rank test): *CCNA2* expression correlated with the clinical outcome of three CMS, with a significantly reduced RFS in *CCNA2*^{non-high} tumor patients by comparison to the *CCNA2*^{high} class for the CMS1 (59% versus 73, $p=1.64e^{-02}$), CMS3 (62% versus 79%, $p=0.05$) and CMS4 consensus subtypes (60% versus 74%, $p=2.08e^{-02}$) (Fig 8E).

To further explore the biological alterations associated with *CCNA2* expression, we compared the whole-genome expression profiles of *CCNA2*^{non-high} (N=222) and *CCNA2*^{high} (N=237) TCGA samples (learning set) (Supplemental Fig 7B and C). We identified 92 genes differentially expressed, including 20 genes overexpressed and 72 genes underexpressed in the *CCNA2*^{high} class (Supplemental Fig 7C; Table 4). A 92-gene GES based on metagene was used to classify samples. As expected, this GES was efficient to class the 459 samples of the learning set ($p=1.08e^{-52}$, t-test). More importantly, its robustness was validated in a completely independent validation set including the 1780 remaining samples ($p=9.22e^{-180}$, t-test; Supplemental Fig 7B). A detailed analysis of the 92 genes revealed that the 20 most significant

altered genes were upregulated involved in cell proliferation and DNA repair (mitotic cell cycle regulation, genomic instability and replicative immortality pathways) (Table 4).

Discussion

Cyclins are well-established drivers of the cell cycle by activating a specific family of kinases, the cyclin-dependent kinases (CDKs). Gene amplification as well as overexpression of cyclins have been frequently detected in cancer tissues, and cyclins have been proposed as biomarkers or therapeutic targets (41). For example, inhibition of cyclin A2 complexes has been shown to impair proliferation of some tumor cell lines (42).

In this report we describe that cyclin A2 deficiency in CECs induces epithelial changes in the mucosa, inflammation, increased cell proliferation and dysplasia in the colon rendering mice more susceptible to chemically-induced colon carcinogenesis. Elevated activation of β -catenin, abnormal mitosis as well as DNA DSBs are already detectable at day 4 after cyclin A2 inactivation and appear therefore to be the initial events. The IHC analysis performed suggests a spatial separation of both events in cyclin A2 deficient colons with nuclear staining of active β -catenin being more frequent in the lower part of colonic crypts, whereas γ H2AX staining localized rather in the upper crypt region (Figs 4,5).

The WNT/ β -catenin pathway is well established to trigger regeneration in certain tissues by controlling the stem cell compartment and has also been shown to cooperate with the YAP/TAZ pathway during colonic tissue repair and regeneration (33, 43). One study showed that cyclin A2 is involved in stabilizing the Axin/GSK3 destruction complex thus promoting β -catenin degradation (44) that could explain β -catenin nuclear accumulation detected in cyclin A2-deficient colons. In line with this is our previous description that cyclin A2 depletion in murine mammary epithelial cells promotes β -catenin activity (18, 19).

A previous study established a role of cyclin A2 in DNA DSB repair by generating mutant mice bearing cyclin A2 knockout and hypomorphic alleles resulting in lower cyclin A2 expression (45). These mice fail to up-regulate the meiotic recombination 11 (Mre11) nuclease that causes impaired resolution of stalled replication forks, reduced repair of double-stranded DNA breaks, aberrant chromosome segregation and increased spontaneous and chemically-induced tumor development (Kanakkanthara et al., 2016). The same study described that cyclin A2 binds to Mre11 mRNA thus promoting its translation. We detected increased Mre11 foci formation in cyclin A2-deficient CEC suggesting that the increased DNA damage detected in the colons of cyclin A2-deficient mice results rather from a defective regulation of DNA replication and/or chromosome segregation. Indeed, phosphorylation of CDC6 by cyclin A/CDK2 has been shown to prevent re-replication during S phase and G2 (46). Moreover,

cyclin A2/CDK activity has been demonstrated to be essential for centrosome duplication and proper sister chromatid segregation (47). Of note, mitoses detected in *VilCreCna2fl/fl* mice, were mostly located in the middle part of colonic crypts whereas γ H2AX positive cells were predominantly identified in the upper part. This is in accordance with the fact that generation of DSB following abnormal mitosis takes place once a cell exits the mitotic process entering the next cell cycle (48) thus during migration toward the top part of the crypts. Notably, high expression levels of γ H2AX have been shown to predict a poor prognosis for CRC patients (49) and a more recent report describes a role for cyclin A2 in preventing chromosomal defects (CIN) in colon cancer cells (50).

It is well established that there is a crosstalk between DNA damage and inflammation and that either event can initiate the other one (51). Colons of cyclin A2-deficient mice display different signs of inflammation such as infiltration of macrophages and IL6 production, which correlated with an increased activation of NF κ B in the colonic epithelial cells. In addition, rectal prolapse was detectable in *VilCreCna2fl/fl* mice, a phenomenon which has been previously described in a mouse model, which develops colitis spontaneously (27). Increased expression and activation of NF κ B has been observed in IBD and CRC patients, especially in mucosal macrophages and epithelial cells, accompanied by production of pro-inflammatory cytokines including IL6 (26). Finally, YAP/TAZ overexpression has been shown to promote tumor formation (52) and to correlate with poor prognosis in CRC (53). We therefore conclude that DNA damage combined with uncontrolled activation of several regenerative pathways promotes the development of dysplasia in cyclin A2-deficient mice.

Most studies evaluating the prognostic value of cyclin A2 in CRC concluded that cyclin A2 overexpression is an indicator for poor prognosis (54–56) whereas one report described a higher survival rate for patient with tumors expressing high levels of cyclin A2 (57). To clarify the prognostic value of cyclin A2 in CRC we performed a meta-analysis of 11 publicly available transcriptome datasets comprising 2239 primary tumor samples. This analysis revealed elevated *CCNA2* transcript levels in primary tumors compared with normal tissues as well as metastases concurring with our previous analysis of biopsies using IHC and Western blot analysis (17). *CCNA2* transcript expression was higher in tumors of stage I and II patients compared with those of stage III and IV. Importantly, this concurs with cyclin A2 protein expression analysis by IHC on our TMA. For the first time, we described that *CCNA2* expression differs among CMS subtypes, with the highest in CMS1 and lowest in CMS4 consensus subtype. A stratification of the transcriptome samples in high and non-high expressers by taking the *CCNA2* transcript expression ratio of CRC *versus* normal tissue ≥ 2 as a cut-off displayed a

higher RFS in the *CCNA2*^{high} class. Importantly, such favorable prognostic value of high *CCNA2* expression persisted in multivariate analysis incorporating the classical prognostic factors or the CMS subtypes, suggesting independent prognostic value. Whole-transcriptome supervised analysis of the two *CCNA2* classes identified that the 20 most significant modulated genes upregulated in the *CCNA2*^{high} versus the *CCNA2*^{non-high} class were all involved in cell proliferation and DNA repair. Though expression data of multicellular tumor tissues cannot be directly correlated with the ones obtained in cell type specific knockout mice, it is remarkable that *CCNA2*^{high} CRC tumors and cyclin A2 deficiency in murine CEC trigger similar mechanisms such as DNA damage. We conclude that the expression pattern of cyclin A2 in CRC tissues mirrors distinct roles during colon carcinogenesis, such as driving cell proliferation in early stages, when highly expressed, but promoting aggressiveness in later stages, when expression levels are lower.

Taken together, our results establish that cyclin A2 deficiency in murine colonic epithelial cells induces DNA damage, inflammation and activation of different regenerative pathways triggering dysplasia. This renders mice more susceptible to chemically-induced colon carcinogenesis by elevated tumor initiation as well as aggressiveness. The observations made in mice are complemented by a meta-analysis of 2239 primary CRC tumors displaying that high *CCNA2* expression is associated with better prognosis in CRC patients. Moreover, we describe here for the first time that *CCNA2* expression differs between the previously described CMS subtypes. Importantly, dissecting the patient cohort into the consensus molecular subtypes identifies *CCNA2*^{non-high} expression as a bad prognosis factor within CMS1, CMS3 and CMS4, but not CMS2. In conclusion, *CCNA2* expression was strongly correlated with several clinicopathological factors of CRC and may represent a favorable prognostic marker in patients with CRC.

Methods

Generation of *cyclin A2* conditional knockout mice in the intestinal epithelium

To generate *cyclin A2* intestinal epithelium-specific knockout mice, *Ccna2* *fl/fl* mice (23) were crossed with transgenic mice carrying either a constitutively active Cre recombinase (*vil-Cre*) (24), which is expressed from the embryonic development (day 9) throughout adulthood in intestinal epithelial cells, or a tamoxifen-inducible Cre under the same promoter (*vil-Cre-ER^{T2}*) (24)), which was induced in adult mice by two intraperitoneal injections of tamoxifen (Sigma, 150 mg/kg). Mice were maintained on a C57BL6/129SV background for the *cyclin A2 vil-Cre* strain and C57BL/6 genetic background for the *cyclin A2 vil-Cre-ER^{T2}* mice. Experimental groups were caged together according to gender. Genomic DNA from mice tails and colonic epithelial cells was used for genotyping using the primers listed in Supplemental Table 1.

Immunohistochemical and immunofluorescence analyses on paraffin tissue section:

Isolated colons specimens were rolled up lengthwise and embedded in paraffin after fixation in 10% formalin (DiaPATH F0046, MM France) for 24 hours. Immunohistochemistry was performed on 4- μ m sections. Tissue slides were deparaffinized and rehydrated by performing the following washes: xylene (2x5 min), 100 % ethanol (5 min), 96% ethanol (3 min), 70% ethanol (3 min) and dH₂O (5 min). Tissue slides were incubated with 10 mM sodium citrate pH6.0 (T0050, DiaPATH) for 20 min at 100°C or Tris 10mM-EDTA 1mM pH9.0, for antigen retrieval, depending on the antibody. Following two washes in PBS-Tween 0.2%, tissue slides were incubated in PBS-3% hydrogen (#H1009, Sigma) for 5 min at room temperature for peroxidase inactivation. After blocking in 2.5 % blocking serum-5% BSA-5% non-fat milk for 30 min at room temperature, tissue slides were incubated with primary antibody for 1 hour at 4°C. Then corresponding secondary antibody reagents (ImmPRESSTM kit, Vector Laboratories), directed against rabbit, rat or mouse antibodies were used for detection. The following antibodies were used: rabbit anti-Cyclin A2 (Abcam, ab181591, 1 :1000 dilution), rabbit anti-Ki67 (Abcam, ab16667, 1 :200 dilution), rat anti-F4/80 (Hycult biotech, HM1066 1 :800 dilution), rabbit anti-non-phospho (Active) β -catenin (Cell signaling, #8814, 1 :1000 dilution), rabbit anti- γ H2AX (Abcam, ab11174, 1 :500), rat anti-IL6 (Novus Biological, IL6/1270, 1:100), rabbit anti-TAZ (Novus Biological, NBP1-85067, 1:100).

For hematoxylin and eosin staining, tissue slides were incubated in Xylene (3x3 min), 100% ethanol (3x3 min), 95% ethanol (3 min), 80% ethanol (3 min) and 5 min in deionized water. Slides were then incubated in hematoxylin (Vector labs) for 3 min, extensively rinsed in

tap water and dipped 8 to 12 times in acid ethanol (0.03N HCl-70% ethanol). After rinsing in tap water, tissue slides were transferred in deionized water for 2 minutes then stained in Eosin for 30 second (Vector Labs) followed by three incubations of 5 min in 95% ethanol then 100% ethanol. After three incubations in Xylene, coverslips were mounted using Permount media (Fisher Scientific). For the tissue microarray (TMA) analysis, the rabbit anti-Cyclin A2 antibody (Abcam, ab181591, 1:1000 dilution), validated on *Ccna2*-deficient mouse tissues as well as rabbit anti-Cyclin A2 from Novocastra (NCL-CYCLINA), were used.

To carry out immunofluorescence analysis, 4µm thick tissue slides were deparaffinized and rehydrated as described above and were incubated in 1mM EDTA antigen retrieval buffer (pH9.0) for the alpha and gamma-tubulin, or in 10 mM citrate (pH6) for the anti-Mre11 staining. Blocking was performed for 30 minutes in 2,5 % blocking serum-5% BSA-5% non-fat milk at room temperature, and tissue slides were then incubated with primary antibodies (anti-Me11, Novus Biologicals NB-100-142, 1:500 dilution; anti-alpha-tubulin, Novus Biologicals NB-600-506, 1:1000 dilution, anti-gamma tubulin, Sigma, T3320, 1:1000 dilution) for 1 hour at 4°C. Depending on primary antibodies, slides were incubated with goat anti-mouse, rat or rabbit secondary antibodies conjugated to either Alexa-488 or Alexa-555 (ThermoFisher Scientific) in addition with DAPI (0.1 µg/ml) for 1 hour at room temperature. Acquisition of the Mre11 staining was performed on the Leica confocal SP5-SMD and images were analyzed using Imaris software. Quantification of the Mre11 staining was performed using ImageJ. For the alpha/gamma-tubulin co-staining, images acquisition in the proximal colon was performed using Metamorph software on the Upright Zeiss Axioimager Z2 and images were analyzed using ImageJ software.

BrdU incorporation assay

For proliferation studies, mice were intraperitoneally injected with 100µg/g of Bromodeoxyuridine (BrdU, B9285, Sigma) diluted in PBS. Colon specimens were dissected 2 hours after injection, flushed with cold PBS, and embedded in paraffin after fixation in 10% formalin for 24hours. Incorporation of BrdU in proliferating intestinal epithelial cells was detected using an anti-BrdU antibody (Biolegend, 1:100 dilution) after deparaffinization of the tissues, antigen retrieval in citrate buffer, as described above, and DNA denaturation using 2N HCl for 1hour at 37°C followed by an incubation in 0.1M borax buffer pH9. Revelation was performed using the Avidin/Biotin Vectastain System kit (Vectorlab, USA) according to the protocol.

Isolation of colonic epithelial cells (CEC)

Colons were removed from mice and flushed with cold PBS. Fat and adherent connective tissue were taken out, colons were cut longitudinally then placed in 10 ml of cold wash buffer (PBS, 2% FBS). Colons were treated with 10 ml of CEC buffer (PBS, 1% BSA, 1 mM EDTA, 1 mM DTT, 5.6 mM Glucose) at 37°C for 45 minutes under continuous shaking to release CECs. Purity of isolated cells was validated by FACS by using the cell surface markers EpCAM-APC (Biolegend, 118213, 1:50 dilution) and CD45-PE (Biolegend, 103105, 1:100 dilution), CD90.2-FITC (BD, 553013, 1:50 dilution) to identify epithelial and immune cells respectively.

Isolation of CEC cytoplasmic and nuclear fractions

Cells were treated with ice-cold hypotonic buffer (10 mM Hepes pH 7.9, 10 mM KCl, 0.1 mM EDTA, 0.1 mM EGTA, 0.5 mM PMSF, 1 mM DTT and 1×Complete inhibitor tabs, Roche, for 15 min), then treated with 10% NP40 and vortexed. The nuclei were pelleted by centrifugation (14,000 rpm, 15s) while the supernatant was kept as the cytoplasmic fraction. Nuclei were lysed in 20 mM Hepes pH 7.9, 0.4 mM NaCl, 1 mM EDTA, 1 mM EGTA, 0.5 mM PMSF, 1 mM DTT, 1×complete inhibitor tabs (Sigma). The supernatant was collected as the nucleoplasmic fraction after centrifugation (14,000 rpm, 5min).

Western blot analysis

Protein samples were prepared in 4xLaemmli buffer, separated in sodium dodecyl sulfate-polyacrylamide gels and electro blotted onto nitrocellulose membranes (Millipore) using a wet/tank transfer system (Bio-Rad, Hercules, CA). The following antibodies were used in this study: anti-Cyclin A2 (Abcam, ab181591, 1:1000 dilution), anti-NF-κB p65 (Cell Signaling, #8242, 1:1000 dilution) and anti-non-phospho (Active) β-catenin (Cell Signaling, #8814, 1:1000 dilution). All the experiments were performed in triplicate and at least three times. Quantification of at least three immunoblots was carried out using the ImageJ software (National Institute of Health). Experimental values are expressed as a ratio of protein levels over control with the TFIIB (Biolegend, 1:2000 dilution) and Histone H3 (Abcam, Ab1791, 1:1000 dilution) signal for nucleoplasmic fraction or β-actin (Sigma, 1:5000 dilution) for total cell lysates.

RNA sequencing samples preparation and bioinformatic analysis

CEC were isolated as described above from the proximal colon part from 4 weeks-old

Ccna2 fl/fl vil-Cre (KO) and *Ccna2 fl/fl* (control) mice. RNA was extracted using the Roche High Pure RNA isolation kit (Roche Molecular Systems, Inc.) according to the manufacturer protocol and purity as well as integrity controlled using the Agilent RNA 6000 nano kit (Agilent Technologies). RNA sequencing was performed on an Illumina NextSeq500™ platform. The quantification of the gene transcripts was performed using the Kallisto program (58), which is based on pseudo-alignment principle, with default settings. GRCm38 Refseq was used to annotate the transcripts. We used the STAR program (59) to align the reads on the *Mus musculus* reference genome (GRCm38/mm10). For the differential expression analysis, we first filtered the transcripts with too low counts among the samples by using the *filterByExpr* function of edgeR, with default settings. To perform the data normalization and differential expression analysis (DEA) of *Ccna2* deficient *versus* control samples, we ran standard steps of edgeR and DESeq2 in parallel on the filtered data. For both DEA methods, we applied a cutoff threshold of 0.05 on the adjusted p-values and a cutoff threshold of 2 on fold changes. The two resulting lists of significantly differentially expressed genes were intersected. We separated the up-regulated and the down-regulated genes of this unique list. To perform the functional enrichment analysis, these sub-lists were finally submitted to the dedicated function of the gProfileR R package, available on Bioconductor. The R code used is provided in the supplementary data.

Colitis-associated carcinogenesis

For induction of colitis-associated carcinogenesis in the *VilCrecyclinA2fl/fl* mice and their corresponding controls, mice were intraperitoneally injected with 6.25 mg/kg azoxymethane (AOM; Sigma). Mice were treated by one cycle of 5 days with 1.75% (w/v) dextran sodium sulphate (DSS; TdB Consultancy AB) administered in the drinking water at day 5 of the protocol. Weight of the mice was checked every two days for two months, except for the week following the DSS treatment where the weight was measured every day. Mice were then sacrificed, and colon specimens prepared for histological analysis after fixation in 10% neutral buffered formalin. Tissues were embedded in paraffin, and 4-µm thick sections were stained with hematoxylin and eosin or incubated with the following primary antibodies (anti-active β-catenin, anti-γH2AX, and anti-IL6) as above reported. Colitis was scored as outlined in Supplemental Table 2.

Patient samples

Details of the TMA generated at CRB–ICM (agreement number BB-033-00059) in Montpellier (France) are listed in Supplemental Table 3. Some tissue samples were generated as duplicates from the same patients but considered as individual templates, for the statistical analysis, due to tumor heterogeneity.

For the transcriptome meta-analysis, we collected clinicopathological and gene expression data of tissues from normal colon (NC), primary colorectal adenocarcinoma samples (pCRC) and metastases from CRC samples (mCRC) from ten public data sets and one data set generated at the Institut Paoli Calmettes (Marseille, France) comprising at least one probe set representing *CCNA2*. Sets and raw data were collected from the National Center for Biotechnology Information (NCBI)/Genbank GEO, ArrayExpress and TCGA databases (Supplemental Table 5). Samples were profiled using whole-genome DNA microarrays (Affymetrix) or RNA sequencing (Illumina). The analyzed data set contained a total of 2,401 samples, including 95 normal (NC) samples, 2,239 primary tumor (pCRC) samples, and 67 metastases (mCRC) samples included in the present analysis (mCRC, located in the liver (n=47) and lung (n=20)).

Gene expression data analysis

Data analysis required a step of pre-analytic processing. We first normalized each data set separately, by using Robust Multichip Average (RMA) (60) with the non-parametric quantile algorithm for the raw Affymetrix data. Normalization was done in R using Bioconductor and associated packages. We then mapped hybridization probes across the different microarrays represented as previously reported (61). When multiple probes mapped to the same GeneID, we retained the one with the highest variance in each data set. We log2-transformed the available TCGA RNAseq data that were already normalized. Next, we extracted *CCNA2* mRNA expression and corrected the batch effects through the 11 studies using z-score normalization. Briefly, for each expression value in each study separately, *CCNA2* values were transformed by subtracting the mean of the gene in that data set divided by its standard deviation, mean and standard deviation being measured on primary samples. *CCNA2* expression was measured as discrete value after comparison with median expression in the 95 NC samples; upregulation, thereafter designated “*CCNA2*^{high}” was defined by a CRC/NC ratio ≥ 2 and no upregulation (“*CCNA2*^{non-high}”) by a CRC/NC ratio < 2 . The Consensus Molecular Subtype (CMS) classification (8) was based on the tool CMScaller made by Eide et al. (62). Finally, to explore the biological pathways associated with our *CCNA2*-based classification, we applied a supervised analysis using learning and validation sets. The

learning set included the 459 samples of the TCGA data set that included 222 CCNA2^{non-high} and 237 CCNA2^{high} samples. We used a moderated t-test with empirical Bayes statistic included in the limma R packages. False discovery rate (FDR) (63) was applied to correct the multiple testing hypothesis and significant genes were defined by the following thresholds: $p < 5\%$, $q < 25\%$ and fold change (FC) superior to $|2x|$. The resulting gene expression signature (GES) was based on a metagene score defined as the difference between mean expression of genes upregulated and mean expression of genes downregulated in the CCNA2^{high} samples and using a cut-off equal to 0. This score was then applied to both learning and validation sets to test the robustness of the GES (t-test). Ontology analysis of the gene list was based on GO biological processes of the Database for Annotation, Visualization and Integrated Discovery (DAVID; <http://david.abcc.ncifcrf.gov/>).

Statistical analysis

Correlations between the CCNA2 expression-based classes (non-high *versus* high) and the clinicopathological factors were calculated with the Fisher's exact test for the binary variables and the Student's t-test for the continuous variables. Our primary endpoint, relapse-free survival (RFS), was calculated from the date of diagnosis until the date of metastatic relapse or death from CRC. The follow-up was measured from the date of diagnosis to the date of last news for event-free patients. Survival was calculated using the Kaplan-Meier method and curves were compared with the log-rank test. Univariate and multivariate analyses were done using Cox regression analysis (Wald test). The variables tested in univariate analysis included the CCNA2-based classification (non-high *versus* high), patients' age and sex, tumor location, pathological stage and grade, MMR status, and the CMS classification. Multivariate analysis incorporated all variables with a p-value inferior to 5% in univariate analysis. All statistical tests were two-sided at the 5% level of significance. Statistical analysis was done using the survival package (version 2.30) in the R software (version 2.15.2). This article is written in accordance with reporting recommendations for tumor marker prognostic studies criteria (REMARK) (64).

Statistical analysis of quantifications for the mouse study was performed in GraphPad using an unpaired t-test except for the Nuclear size distribution where the Kolmogorov-Smirnov test using R software was performed.

Animal study approval:

Mouse experiments were performed in strict accordance with the guidelines of the

European Community (86/609/EEC), the French National Committee (87/848) for care and use of laboratory animals and were approved by the Regional Ethics committee under the 01556.02 reference number.

Acknowledgments:

We are grateful to the excellent platforms in Montpellier, i.e. RIO imaging platform, the histology and animal experimentation platforms RHEM and RAM, as well as the IGMM mouse facility. Many thanks to Thierry Gostan for his help on the statistical analysis and Ula Hibner for carefully reading the manuscript. We are grateful to Maguy Del Rio for her help on cyclin A2 mRNA expression in patients. This work was supported by INCA (project 2013-111), by the SIRIC Montpellier Cancer Grant INCa_Inserm_DGOS_12553 and La Ligue contre Cancer -comité 34 (BL).

Author's contribution:

YG, MG, CP, VP, RT, HT, JB, CM, QDS, PF, FB-M, CL, SG, RA, EM, BL: conducting experiments, acquiring data and analyzing data.

EM, BL, MH: designing research studies and analyzing data.

YG, FB, JMB, PS, EM, BL, MH: writing manuscript.

References

1. <http://gco.iarc.fr/today/fact-sheets-cancers>
2. Zlobec I, Lugli A. Prognostic and predictive factors in colorectal cancer. *J. Clin. Pathol.* 2008;61(5):561–569.
3. Cuyle P-J, Prenen H. Current and future biomarkers in the treatment of colorectal cancer. *Acta Clin. Belg.* 2017;72(2):103–115.
4. Kannarkatt J, Joseph J, Kurniali PC, Al-Janadi A, Hrinczenko B. Adjuvant Chemotherapy for Stage II Colon Cancer: A Clinical Dilemma. *J. Oncol. Pract.* 2017;13(4):233–241.
5. McCleary NJ, Benson AB, Dienstmann R. Personalizing Adjuvant Therapy for Stage II/III Colorectal Cancer. *Am. Soc. Clin. Oncol. Educ. Book Am. Soc. Clin. Oncol. Annu. Meet.* 2017;37:232–245.
6. Beaugerie L et al. High risk of anal and rectal cancer in patients with anal and/or perianal Crohn's disease. *Clin. Gastroenterol. Hepatol. Off. Clin. Pract. J. Am. Gastroenterol. Assoc.* [published online ahead of print: November 30, 2017]; doi:10.1016/j.cgh.2017.11.041
7. Ma H et al. Pathology and genetics of hereditary colorectal cancer. *Pathology (Phila.)* 2018;50(1):49–59.
8. Guinney J et al. The consensus molecular subtypes of colorectal cancer. *Nat. Med.* 2015;21(11):1350–1356.
9. Yasmeen A, Berdel WE, Serve H, Müller-Tidow C. E- and A-type cyclins as markers for cancer diagnosis and prognosis. *Expert Rev. Mol. Diagn.* 2003;3(5):617–633.

10. Bendris N et al. Cyclin A2: a genuine cell cycle regulator?. *Biomol. Concepts* 2012;3(6):535–543.
11. Blanchard JM. To be or not to be a proliferation marker?. *Oncogene* 2014;33(8):954–955.
12. Jirawatnotai S et al. A function for cyclin D1 in DNA repair uncovered by protein interactome analyses in human cancers. *Nature* 2011;474(7350):230–234.
13. Li Z et al. Cyclin D1 induction of cellular migration requires p27(KIP1). *Cancer Res.* 2006;66(20):9986–9994.
14. Li Z et al. Cyclin D1 regulates cellular migration through the inhibition of thrombospondin 1 and ROCK signaling. *Mol. Cell. Biol.* 2006;26(11):4240–4256.
15. Li Z, Wang C, Prendergast GC, Pestell RG. Cyclin D1 functions in cell migration. *Cell Cycle Georget. Tex* 2006;5(21):2440–2442.
16. McAllister SS, Becker-Hapak M, Pintucci G, Pagano M, Dowdy SF. Novel p27(kip1) C-terminal scatter domain mediates Rac-dependent cell migration independent of cell cycle arrest functions. *Mol. Cell. Biol.* 2003;23(1):216–228.
17. Arsic N et al. A novel function for Cyclin A2: control of cell invasion via RhoA signalling. *J. Cell Biol.* 2011;in press.
18. Bendris N et al. Cyclin A2, a novel regulator of EMT [Internet]. *Cell Mol Life Sci* [published online ahead of print: May 31, 2014];http://www.ncbi.nlm.nih.gov/entrez/query.fcgi?cmd=Retrieve&db=PubMed&dopt=Citation&list_uids=24879294. cited

19. Cheung CT et al. Cyclin A2 modulates EMT via β -catenin and phospholipase C pathways. *Carcinogenesis* [published online ahead of print: May 19, 2015]; doi:10.1093/carcin/bgv069
20. Wang D et al. Prefoldin 1 promotes EMT and lung cancer progression by suppressing cyclin A expression. *Oncogene* 2017;36(7):885–898.
21. Kanakkanthara A et al. Cyclin A2 is an RNA binding protein that controls Mre11 mRNA translation. *Science* 2016;353(6307):1549–1552.
22. Gopinathan L et al. Loss of Cdk2 and cyclin A2 impairs cell proliferation and tumorigenesis. *Cancer Res.* 2014;74(14):3870–3879.
23. Kalaszczyńska I et al. Cyclin A is redundant in fibroblasts but essential in hematopoietic and embryonic stem cells. *Cell* 2009;138(2):352–365.
24. el Marjou F et al. Tissue-specific and inducible Cre-mediated recombination in the gut epithelium. *Genes. N. Y. N 2000* 2004;39(3):186–193.
25. Minoo P, Zlobec I, Peterson M, Terracciano L, Lugli A. Characterization of rectal, proximal and distal colon cancers based on clinicopathological, molecular and protein profiles. *Int. J. Oncol.* 2010;37(3):707–718.
26. Taniguchi K, Karin M. IL-6 and related cytokines as the critical lynchpins between inflammation and cancer. *Semin. Immunol.* 2014;26(1):54–74.
27. Laubitz D et al. Colonic gene expression profile in NHE3-deficient mice: evidence for spontaneous distal colitis. *Am. J. Physiol. Gastrointest. Liver Physiol.* 2008;295(1):G63–G77.
28. Sobecki M et al. Cell-Cycle Regulation Accounts for Variability in Ki-67 Expression Levels. *Cancer Res.* 2017;77(10):2722–2734.

29. Hayashi MT, Karlseder J. DNA damage associated with mitosis and cytokinesis failure. *Oncogene* 2013;32(39):4593–4601.
30. van den Bosch M, Bree RT, Lowndes NF. The MRN complex: coordinating and mediating the response to broken chromosomes. *EMBO Rep.* 2003;4(9):844–849.
31. Rein K, Stracker TH. The MRE11 complex: an important source of stress relief. *Exp. Cell Res.* 2014;329(1):162–169.
32. Karin M, Clevers H. Reparative inflammation takes charge of tissue regeneration. *Nature* 2016;529(7586):307–315.
33. Yui S et al. YAP/TAZ-Dependent Reprogramming of Colonic Epithelium Links ECM Remodeling to Tissue Regeneration. *Cell Stem Cell* 2018;22(1):35-49.e7.
34. De Robertis M et al. The AOM/DSS murine model for the study of colon carcinogenesis: From pathways to diagnosis and therapy studies. *J. Carcinog.* 2011;10:9.
35. Erben U et al. A guide to histomorphological evaluation of intestinal inflammation in mouse models. *Int. J. Clin. Exp. Pathol.* 2014;7(8):4557–4576.
36. Hydbring P, Malumbres M, Sicinski P. Non-canonical functions of cell cycle cyclins and cyclin-dependent kinases. *Nat. Rev. Mol. Cell Biol.* 2016;17(5):280–292.
37. Chen W, Lee J, Cho SY, Fine HA. Proteasome-mediated destruction of the cyclin a/cyclin-dependent kinase 2 complex suppresses tumor cell growth in vitro and in vivo. *Cancer Res.* 2004;64(11):3949–3957.
38. Clevers H, Loh KM, Nusse R. Stem cell signaling. An integral program for tissue renewal and regeneration: Wnt signaling and stem cell control. *Science* 2014;346(6205):1248012.

39. Kim SI et al. Cyclin-dependent kinase 2 regulates the interaction of Axin with beta-catenin. *Biochem. Biophys. Res. Commun.* 2004;317(2):478–483.
40. Kanakkanthara A et al. Cyclin A2 is an RNA binding protein that controls Mre11 mRNA translation. *Science* 2016;353(6307):1549–1552.
41. Petersen BO, Lukas J, Sørensen CS, Bartek J, Helin K. Phosphorylation of mammalian CDC6 by cyclin A/CDK2 regulates its subcellular localization. *EMBO J.* 1999;18(2):396–410.
42. Kabeche L, Compton DA. Cyclin A regulates kinetochore microtubules to promote faithful chromosome segregation. *Nature* 2013;502(7469):110–113.
43. Ganem NJ, Pellman D. Linking abnormal mitosis to the acquisition of DNA damage. *J. Cell Biol.* 2012;199(6):871–881.
44. Lee Y-C et al. High expression of phospho-H2AX predicts a poor prognosis in colorectal cancer. *Anticancer Res.* 2015;35(4):2447–2453.
45. Li J-A, Liu B-C, Song Y, Chen X. Cyclin A2 regulates symmetrical mitotic spindle formation and centrosome amplification in human colon cancer cells. *Am. J. Transl. Res.* 2018;10(8):2669–2676.
46. Georgakilas AG, Kotsinas A. Editorial: DNA Damage and Inflammation under Stress. *Front. Genet.* 2017;8:152.
47. Zanconato F, Cordenonsi M, Piccolo S. YAP/TAZ at the Roots of Cancer. *Cancer Cell* 2016;29(6):783–803.

48. Yuen H-F et al. TAZ expression as a prognostic indicator in colorectal cancer. *PloS One* 2013;8(1):e54211.
49. Handa K, Yamakawa M, Takeda H, Kimura S, Takahashi T. Expression of cell cycle markers in colorectal carcinoma: superiority of cyclin A as an indicator of poor prognosis. *Int. J. Cancer* 1999;84(3):225–233.
50. Bahnassy AA et al. Cyclin A and cyclin D1 as significant prognostic markers in colorectal cancer patients. *BMC Gastroenterol.* 2004;4:22.
51. Nozoe T, Inutsuka S, Honda M, Ezaki T, Korenaga D. Clinicopathologic significance of cyclin A expression in colorectal carcinoma. *J. Exp. Clin. Cancer Res. CR* 2004;23(1):127–133.
52. Li J-Q et al. Cyclin A correlates with carcinogenesis and metastasis, and p27(kip1) correlates with lymphatic invasion, in colorectal neoplasms. *Hum. Pathol.* 2002;33(10):1006–1015.
53. Bray NL, Pimentel H, Melsted P, Pachter L. Near-optimal probabilistic RNA-seq quantification. *Nat. Biotechnol.* 2016;34(5):525–527.
54. Dobin A et al. STAR: ultrafast universal RNA-seq aligner. *Bioinforma. Oxf. Engl.* 2013;29(1):15–21.
55. Irizarry RA et al. Exploration, normalization, and summaries of high density oligonucleotide array probe level data. *Biostat. Oxf. Engl.* 2003;4(2):249–264.
56. Bertucci F, Finetti P, Viens P, Birnbaum D. EndoPredict predicts for the response to neoadjuvant chemotherapy in ER-positive, HER2-negative breast cancer. *Cancer Lett.* 2014;355(1):70–75.

57. Eide PW, Bruun J, Lothe RA, Sveen A. CMScaller: an R package for consensus molecular subtyping of colorectal cancer pre-clinical models. *Sci. Rep.* 2017;7(1):16618.
58. Hochberg Y, Benjamini Y. More powerful procedures for multiple significance testing. *Stat. Med.* 1990;9(7):811–818.
59. McShane LM et al. REporting recommendations for tumor MARKer prognostic studies (REMARK). *Nat. Clin. Pract. Urol.* 2005;2(8):416–422.

Figure Legends

Figure 1: Cyclin A2 deletion leads to epithelial changes in the colon mucosa and inflammation. (A). Cyclin A2 depletion at the protein level in colonic epithelial cells (CEC) of constitutive (*VilCreCcna2fl/fl*, left panel) and tamoxifen-induced knockout mice (*VilCreERT2Ccna2fl/fl*, right panel) by comparison to control animals (*Ccna2fl/fl*) displayed by Western Blot analysis. **(B, C, D).** Immunostaining for cyclin A2 **(B)**, F4/80 **(C)** and IL-6 **(D)** of the indicated parts of colon derived from control (*Ccna2fl/fl*), and constitutive (*VilCreCcna2fl/fl*) cyclinA2-deficient mice. Blow Up: 2.5X. Scale bars: 100µm.

Figure 2: Cyclin A2 depletion in colonic epithelial cells induces cell proliferation. (A). Analysis of cell proliferation in colonic crypts of control (*Ccna2fl/fl*) and cyclinA2-deficient (*VilCreCcna2fl/fl*) mice by BrdU incorporation. Left panel: Representative images of immunostainings for BrdU from the distal and proximal parts of the colons taken from control (*Ccna2fl/fl*) and cyclinA2-deficient (*VilCreCcna2fl/fl*) mice 2 hours after intraperitoneal injection of the nucleoside analog. Right panel: Quantification of the numbers of BrdU positive cells per crypt segment. Crypts (depicted in red) were subdivided in 3 parts (dotted lines) for the analysis (bottom, middle and top, n=120 crypts for the distal and n=110 for the proximal part from 3 different mice; mean values $\pm$ SEM are provided, **** p<0.0001; two-tailed unpaired t-test). **(B).** Representative images for Ki67 immunostaining in the proximal part of the colon from control (*Ccna2fl/fl*) animals, induced knockout mice 8 days after tamoxifen treatment (*VilCreERT2Ccna2fl/fl*) and constitutive knockout mice (*VilCreCcna2fl/fl*). Scale bars: 100µm.

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Student t-test test (expressed as mean $\pm$ SD, $p=9.32e^{-09}$). **(C)**. Cyclin A2 protein expression determined by immunostaining using the anti-cyclinA2 antibody from Abcam validated on cyclin A2-deficient colon tissue and expressed as number of positive cells per mm² of tumor analyzed from a TMA of CRC tumor samples derived from stage I (n=23), II (n=48 with 24 MSS tumors and 24 MSI tumors =), III (n=30) and IV (n=26) patients. ($p<0.01$ for the analysis between stage I and II-MSS, $p<0.05$ for comparison between stage I and stage IV, $p=0.01$ for stage II-MSI to IV). **(D)**. Kaplan-Meier RFS Relapse-Free-Survival (RFS) of patients defined as carrier of *CCNA2*^{high} (n=988) and non-high (n=894) tumors. Survivals were calculated using the Kaplan-Meier method and were compared with the log-rank test ($p=8.86e^{-06}$). **(E)**. Analysis of *CCNA2* mRNA levels in the different consensus CRC subtypes, i.e. CMS1 (n=389), CMS2 (n=640), CMS3 (343) and CMS4 (n=604) expressed as log² over normal colon as described in **(A)** ($p=2.57e^{-79}$ by the ANOVA test). **(F)**. Kaplan-Meier RFS curves of all patients with pCRC according to both the *CCNA2*-based classification (*CCNA2*^{high} and *CCNA2*^{non-high} classes with plain curve and dashed curve respectively) and the CMS subtypes ($p=1.64e^{-02}$ for the CMS1 subtype, 0.174 for CMS2, 0.053 for CMS3 and $2.08e^{-02}$ for CMS4).

Supplemental Figure 1: Cyclin A2-deficiency in colonic epithelial cells induces architectural changes in the mucosa and inflammation. (A). HE staining illustrating the subdivision of the colon in 3 parts for the histological analysis, i.e. the distal part close to the rectum, transverse and proximal part, close to the caecum. **(B).** Quantification of the F4/80 staining in control (n=3), constitutive (n=3) and inducible mutant mice (n=4; expressed as mean $\pm$ SEM, *p<0.05; two-tailed unpaired t-test) expressed as number of F4/80 positive cells per mm² of colon tissue. **(C).** Representative HE staining of irregular formed crypts in colons from a constitutive and inducible (day8 following tamoxifen injection) knockout mouse by comparison to colons from control animals. Scale bar: 100 μ m.

Supplemental Figure 2: RNA-seq analysis of altered expression of genes involved in cell cycle regulation and DNA double-strand break repair in cyclin A2 deficient colonic epithelial cells (CEC). (A). Sashimi plot of *Ccna2* reads for each mouse-derived CEC samples analyzed by RNA-seq. Plots show deletion of exons 2 to 7 in transcripts of CEC from *VilCreCcna2^{fl/fl}* mice compared to controls. The coverage for each alignment track is plotted as a bar graph. Arcs representing splice junctions connect exons. Arcs display the number of reads split across the junction (junction depth). Genomic coordinates and the gene annotation track are shown below the junction tracks. **(B).** Gene Ontology over-representation analysis showing the genes up-regulated in *Ccna2^{fl/fl}* mutant mice compared to controls. The top 20 of Gene Ontology terms (Biological Process) are shown here. The p-values have been corrected for multiple testing by the Benjamini-Hochberg method. In this dot plot, the color represents the adjusted p-values and the dot size represents the number of genes for each term. The gene ratio is shown on the x-axis. **(C).** Alterations in the double-strand break KEGG pathway in cyclin A2 deficient CEC. Genes up-regulated in *VilCreCcna2^{fl/fl}* mice samples are in red, whereas downregulated genes are in blue. The scale indicated on the figure represent the log₂ Fold change. In grey are the genes absent from our data. For all analyses, only p-values <0.05 were considered as statistically significant. FC: Fold change; DSB: Double-strand break

Supplemental Figure 3: Increased proliferation of colonic epithelial cells from cyclin A2-deficient mice. (A, B, C). Quantification of Ki67 expression by IHC of the different parts of crypts from the distal and proximal part of the colon from control (n= 66 crypts analyzed from 3 different mice) and constitutive cyclin A2-deficient mice **(A, B)** and proximal part of the colon from induced knockout mice at day 8 following inactivation **(C)** compared to controls (n=120

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Supplemental Figure 4: Increased nuclear size of colonic epithelial cells in cyclin A2-deficient mice. Distribution curve representing the nuclear size of *Ccna2^{fl/fl}* colonic epithelial cells (white, $n=398$ for the left panel and 578 for the right panel from 3 mice), *VilCreCcna2^{fl/fl}* (black, left panel, $n=400$ from 3 mice, $p < 1.10 \times 10^{-16}$) and *VilCreERT2Ccna2^{fl/fl}* (black, right panel, $n=566$, $p < 1.10 \times 10^{-16}$) colonic epithelial cells; p-values were determined using Kolmogorov-Smirnov test.

Supplemental Figure 5: Examples of mitoses in colons of cyclin A2-deficient mice. (A). Immunofluorescence analysis of mitosis (indicated by red circles) using an anti- α -tubulin antibody (green) to stain for the mitotic spindle in combination with γ -tubulin (centrosome in red) and DAPI for DNA in a colon of *VilCreERT2Ccna2^{fl/fl}* mice at day 8 following inactivation (right panel) by comparison to controls (left panel). Scale bar: $100\mu\text{m}$. **(B).** The upper image shows normal mitosis, the lower panel several examples of abnormal mitoses observed in a *Ccna2*-deficient colon. Blow Up: 2.5X

Supplemental Figure 6: Protocol and weight monitoring of the mice during the colitis-associated carcinogenesis. (A). Schematic representation of the modified AOM/DSS protocol applied to *VilCreCcna2^{fl/fl}* and control mice (see Figs 6 and 7). **(B).** Monitoring of the relative weight (expressed as percentage relative to the weight at the beginning of the protocol) of *VilCreCcna2^{fl/fl}* and control mice during the AOM/DSS protocol.

Supplemental Figure 7: Cyclin A2 expression at the mRNA and protein level in CRC patients. (A). Relapse-Free-Survival (RFS) curve of the overall patients analyzed for cyclin A2 mRNA expression levels **(B).** Metagene-based prediction score of outcome (using Student t-test and expressed as mean $\pm$ SD) of the *CCNA2^{high}* samples compared to those of *CCNA2^{non-high}* samples in the learning set (left) and in the independent validation set (right). **(C).** Volcano plot showing the 92 genes differentially expressed in the learning set (TCGA). Genes up-regulated in the *CCNA2^{high}* samples are colored in red and genes down-regulated in green.

Supplemental Figure 8: Cyclin A2 protein expression in CRC tumor samples from different stages. (A). Cyclin A2 expression analyzed on the same TMA shown in Figure 8, but using a different anti-cyclin A2 antibody from Novocastra. (Mean values $\pm$ SEM, $p < 0.05$ for the analysis between stage I and II-MSS, $p < 0.01$ for comparison between stage I and stage III, $p < 0.001$ for stage I to IV, $p < 0.01$ for stage II-MSI to stage IV, two-tailed unpaired t-test). **(B).** Representative immunostaining of stage I, II-MSS, II-MSI, III and IV tumor samples. Scale bar: 100 μ m

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A

B

C

D

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Table1 : List of the 1109 genes differentially expressed between *VilCreCcn2fl/fl* mice and *Ccn2fl/fl* mice samples (fold change > 2, adjusted p-value |FDR| <0.05).
FC: Fold change, CPM: Counts-per-million, FDR: False Discovery Rate

Gene	edgeR log FC	edgeR log CPM	edgeR p-value	edgeR FDR	edgeR Significant differential expression
Ckb	3,401	7,238	8,98E-10	1,43E-05	Up in KO
Lama5	3,536	6,228	2,44E-08	9,62E-05	Up in KO
Pck1	-1,665	6,962	3,32E-08	9,62E-05	Down in KO
Fbln1	3,592	3,066	7,37E-08	9,76E-05	Up in KO
Nid1	4,862	2,592	2,92E-08	9,62E-05	Up in KO
0610040F04Rik	2,954	3,266	4,14E-08	9,62E-05	Up in KO
Gsdmc3	-3,495	6,308	9,91E-08	1,12E-04	Down in KO
LOC108167804	3,666	2,659	1,58E-07	1,48E-04	Up in KO
Cdc7	2,276	3,896	5,14E-08	9,76E-05	Up in KO
Bmp3	-2,567	5,019	7,24E-08	9,76E-05	Down in KO
Slc25a45	2,526	2,539	6,22E-08	9,76E-05	Up in KO
Thpo	2,319	2,996	2,53E-07	1,83E-04	Up in KO
Gm34583	3,145	1,775	1,92E-07	1,61E-04	Up in KO
Hsd17b13	-2,805	3,987	8,40E-08	1,03E-04	Down in KO
Grb10	4,234	1,705	4,24E-08	9,62E-05	Up in KO
Igf2bp1	3,063	3,504	2,41E-07	1,82E-04	Up in KO
Ccdc88a	3,332	1,428	2,51E-08	9,62E-05	Up in KO
Erc2	3,380	1,245	3,15E-07	1,98E-04	Up in KO
Prlr	-3,696	3,615	3,22E-07	1,98E-04	Down in KO
Eomes	4,194	2,904	1,45E-07	1,48E-04	Up in KO
Ptges	3,070	2,040	5,54E-07	2,44E-04	Up in KO
Crip2	2,927	4,087	3,23E-07	1,98E-04	Up in KO
Tgfbr2	1,439	5,664	1,52E-07	1,48E-04	Up in KO
Cacna1e	-5,906	2,036	1,02E-06	3,44E-04	Down in KO
Add3	-1,446	5,984	2,24E-07	1,78E-04	Down in KO
Gm34496	4,532	1,437	1,79E-07	1,58E-04	Up in KO
Adamts15	-2,500	3,342	6,00E-07	2,57E-04	Down in KO
Eda2r	3,558	2,197	3,44E-06	6,77E-04	Up in KO
Skp2	1,891	3,185	6,87E-07	2,79E-04	Up in KO
Slco2b1	-2,211	3,926	3,86E-07	2,13E-04	Down in KO
Rarb	3,239	1,309	7,19E-07	2,79E-04	Up in KO
Atp6v0e2	1,438	5,087	3,58E-07	2,11E-04	Up in KO
Rasgrf2	-2,413	2,836	4,98E-07	2,33E-04	Down in KO
Sult4a1	3,830	1,593	5,23E-07	2,37E-04	Up in KO
Ccn1	-1,663	7,311	4,96E-07	2,33E-04	Down in KO
Rnf186	-1,638	6,794	4,22E-07	2,16E-04	Down in KO
Tmem184c	1,974	2,893	1,36E-06	3,88E-04	Up in KO
Cpm	-1,305	4,115	7,16E-07	2,79E-04	Down in KO
Pear1	1,301	3,892	1,07E-06	3,51E-04	Up in KO
Gja1	3,281	3,297	1,93E-06	4,94E-04	Up in KO
Slc27a2	-2,990	1,336	2,80E-07	1,93E-04	Down in KO
Myo5a	2,997	2,173	1,36E-06	3,88E-04	Up in KO
Tpbp	2,599	3,888	1,13E-06	3,51E-04	Up in KO
Mmp7	4,861	3,968	6,89E-06	1,07E-03	Up in KO

Neto2	2,173	1,660	7,75E-07	2,93E-04	Up in KO
Lce3b	6,735	1,797	9,16E-07	3,31E-04	Up in KO
Chga	-2,393	5,481	1,22E-06	3,65E-04	Down in KO
Sobp	-4,192	1,184	3,95E-07	2,13E-04	Down in KO
Apoc2	3,139	4,090	1,60E-06	4,32E-04	Up in KO
Gch1	1,372	5,684	6,46E-07	2,70E-04	Up in KO
Chgb	-2,066	5,881	1,10E-06	3,51E-04	Down in KO
MyI7	2,746	4,027	2,57E-06	5,82E-04	Up in KO
DII1	-1,659	2,701	2,13E-06	5,37E-04	Down in KO
Atg9b	4,034	2,396	4,87E-06	8,73E-04	Up in KO
Hfm1	5,051	1,082	4,39E-07	2,18E-04	Up in KO
Gnal	2,476	5,000	1,60E-06	4,32E-04	Up in KO
Gsdmc2	-2,613	8,110	2,63E-06	5,87E-04	Down in KO
Spr1a	1,420	6,106	1,15E-06	3,51E-04	Up in KO
Rec8	2,049	4,258	1,52E-06	4,24E-04	Up in KO
Stil	1,108	4,570	1,11E-06	3,51E-04	Up in KO
Hoxd4	-2,948	2,379	2,54E-06	5,82E-04	Down in KO
E2f7	2,917	2,815	4,39E-06	8,20E-04	Up in KO
Matk	-2,085	1,741	3,05E-06	6,39E-04	Down in KO
Aldh3b2	1,583	4,330	1,79E-06	4,75E-04	Up in KO
Fer1l4	1,394	4,829	3,45E-06	6,77E-04	Up in KO
Nr4a1	-1,517	7,679	2,23E-06	5,46E-04	Down in KO
Gm1600	2,490	0,757	9,90E-07	3,42E-04	Up in KO
9330182L06Rik	2,596	1,195	9,71E-06	1,34E-03	Up in KO
Sult1c2	-1,923	4,975	3,06E-06	6,39E-04	Down in KO
Il3ra	2,185	1,678	2,37E-06	5,63E-04	Up in KO
Piezo1	1,644	5,170	2,27E-06	5,46E-04	Up in KO
E130008D07Rik	1,913	1,579	3,84E-06	7,35E-04	Up in KO
Agmo	-2,472	2,476	6,68E-06	1,06E-03	Down in KO
Cacna1a	-2,097	1,893	6,53E-06	1,06E-03	Down in KO
Gm32949	1,278	2,912	4,56E-06	8,34E-04	Up in KO
Polq	1,653	2,547	4,25E-06	8,04E-04	Up in KO
Fer1l4	2,884	2,510	2,27E-06	5,46E-04	Up in KO
Lrrc8b	1,254	5,407	9,61E-07	3,39E-04	Up in KO
Atad2	1,404	4,750	3,31E-06	6,65E-04	Up in KO
Ces1d	-1,778	4,999	3,06E-06	6,39E-04	Down in KO
Myo18b	7,302	1,261	7,05E-08	9,76E-05	Up in KO
5730507C01Rik	1,886	3,329	5,60E-06	9,68E-04	Up in KO
3930402G23Rik	2,013	2,164	1,04E-05	1,39E-03	Up in KO
Gspt2	1,649	2,185	1,26E-05	1,58E-03	Up in KO
Sh3tc2	3,209	2,329	8,17E-06	1,20E-03	Up in KO
Zmat4	-4,692	0,428	4,03E-07	2,13E-04	Down in KO
Ccl5	-3,404	3,267	1,40E-05	1,62E-03	Down in KO
Tmem266	2,632	2,308	7,00E-06	1,08E-03	Up in KO
Gsdmc4	-2,325	8,275	9,70E-06	1,34E-03	Down in KO
Sass6	1,229	3,509	7,91E-06	1,17E-03	Up in KO
Baiap3	-2,271	1,585	3,14E-06	6,39E-04	Down in KO
Mtap	2,200	2,190	6,83E-06	1,07E-03	Up in KO
Slc7a11	3,632	2,450	1,72E-05	1,80E-03	Up in KO
Tpm2	1,104	4,553	5,44E-06	9,60E-04	Up in KO
Tcf24	2,717	1,637	7,88E-06	1,17E-03	Up in KO
Cul9	-2,160	1,234	9,91E-06	1,36E-03	Down in KO

Capg	1,515	6,616	5,54E-06	9,68E-04	Up in KO
Apoc1	4,382	0,172	8,03E-07	2,97E-04	Up in KO
Ntrk2	-1,624	2,806	2,06E-05	1,88E-03	Down in KO
Rab31	1,736	3,879	6,67E-06	1,06E-03	Up in KO
Defa24	10,107	1,246	7,86E-05	4,04E-03	Up in KO
Rrm2	2,852	5,377	1,89E-05	1,85E-03	Up in KO
Syt8	2,491	2,007	9,65E-06	1,34E-03	Up in KO
Stom	1,629	6,475	5,91E-06	9,87E-04	Up in KO
Ulbpl	2,587	1,065	3,11E-06	6,39E-04	Up in KO
Akr1b8	1,389	7,708	7,66E-06	1,16E-03	Up in KO
1700017B05Rik	1,222	4,551	5,69E-06	9,72E-04	Up in KO
1700007E05Rik	2,413	1,347	1,42E-05	1,62E-03	Up in KO
Me1	2,220	6,645	1,29E-05	1,58E-03	Up in KO
Spag16	2,970	1,366	1,20E-05	1,51E-03	Up in KO
Ddit4l	3,741	0,596	1,53E-05	1,71E-03	Up in KO
Gbp8	-4,429	2,608	4,80E-05	2,99E-03	Down in KO
Maf	1,418	3,640	1,06E-05	1,40E-03	Up in KO
Pvt1	1,998	1,573	8,42E-06	1,23E-03	Up in KO
Cit	1,244	4,163	1,84E-05	1,85E-03	Up in KO
Sh3bp1	1,311	3,848	7,07E-06	1,08E-03	Up in KO
Spr2a3	1,419	9,911	1,47E-05	1,66E-03	Up in KO
Edn2	-1,979	4,829	1,05E-05	1,39E-03	Down in KO
Plb1	3,222	0,704	8,03E-05	4,10E-03	Up in KO
Sis	2,551	4,360	1,93E-05	1,85E-03	Up in KO
Zfp677	-1,966	1,770	1,41E-05	1,62E-03	Down in KO
Ms4a10	2,343	1,620	1,88E-05	1,85E-03	Up in KO
Mybl1	1,666	2,331	1,91E-05	1,85E-03	Up in KO
Slc16a3	2,615	4,105	2,03E-05	1,88E-03	Up in KO
Scgb2b15	3,725	1,443	2,35E-05	1,97E-03	Up in KO
Scgb2b17	3,725	1,443	2,35E-05	1,97E-03	Up in KO
9130227L01Rik	1,518	1,851	1,38E-05	1,62E-03	Up in KO
Macrod1	-1,305	3,458	1,20E-05	1,51E-03	Down in KO
Cotl1	1,270	6,433	1,17E-05	1,50E-03	Up in KO
Pcsk6	3,771	1,252	3,31E-05	2,43E-03	Up in KO
1700016C15Rik	2,754	0,162	1,37E-06	3,88E-04	Up in KO
Ddias	1,779	2,562	1,92E-05	1,85E-03	Up in KO
Gm31423	2,135	3,296	2,42E-05	2,00E-03	Up in KO
Tph1	-2,188	3,917	2,15E-05	1,92E-03	Down in KO
Acta1	-2,504	7,231	2,64E-05	2,08E-03	Down in KO
Cyp4b1	-2,024	4,851	1,64E-05	1,75E-03	Down in KO
Kcnh8	-3,517	0,474	1,03E-05	1,39E-03	Down in KO
Vav1	-1,463	1,967	1,59E-05	1,71E-03	Down in KO
Bmp8b	1,635	2,405	2,18E-05	1,93E-03	Up in KO
Igfbp4	3,283	2,937	4,73E-05	2,96E-03	Up in KO
Ttc7b	1,351	2,494	2,79E-05	2,19E-03	Up in KO
Steap4	2,025	2,626	1,91E-05	1,85E-03	Up in KO
Pcsk1n	-2,889	1,231	4,98E-05	3,05E-03	Down in KO
Rad54b	1,075	2,778	7,30E-05	3,84E-03	Up in KO
Trpm5	-2,748	1,553	1,10E-05	1,43E-03	Down in KO
Mki67	1,555	7,569	1,75E-05	1,82E-03	Up in KO
Fat2	2,168	0,787	1,29E-05	1,58E-03	Up in KO
Suox	2,065	5,819	2,53E-05	2,04E-03	Up in KO

Tnni1	1,844	5,309	3,05E-05	2,32E-03	Up in KO
Rtn4rl1	-1,227	2,216	3,75E-05	2,63E-03	Down in KO
LOC108168178	3,396	2,345	6,25E-05	3,51E-03	Up in KO
Gipc3	2,408	1,351	4,66E-05	2,94E-03	Up in KO
Brca1	2,186	3,929	2,87E-06	6,24E-04	Up in KO
Cyp27a1	-1,371	3,655	1,78E-05	1,84E-03	Down in KO
Il1rn	1,508	6,015	2,24E-05	1,95E-03	Up in KO
Slc28a2	-3,076	1,659	6,44E-05	3,56E-03	Down in KO
Slc7a9	1,902	3,298	3,13E-05	2,34E-03	Up in KO
Prss16	4,095	0,549	4,31E-05	2,76E-03	Up in KO
Bmp1	-1,960	3,137	2,58E-05	2,07E-03	Down in KO
Scgb2b10	3,267	1,995	6,13E-05	3,50E-03	Up in KO
Grik5	-1,302	3,119	2,50E-05	2,04E-03	Down in KO
Rcn1	-2,959	1,360	3,41E-05	2,48E-03	Down in KO
Gm32555	1,437	1,907	3,56E-05	2,56E-03	Up in KO
Tcf7l1	1,771	4,710	3,65E-05	2,58E-03	Up in KO
Tnni2	3,374	-0,117	5,78E-06	9,76E-04	Up in KO
Gm40595	2,148	1,649	6,66E-05	3,61E-03	Up in KO
Gprin1	1,690	1,007	1,91E-05	1,85E-03	Up in KO
Frmd3	2,672	0,074	0,00014307	5,63E-03	Up in KO
Atf7ip2	2,462	1,226	0,00010795	4,83E-03	Up in KO
Prss27	2,669	1,843	5,51E-05	3,28E-03	Up in KO
Phlda3	2,986	2,258	8,12E-05	4,12E-03	Up in KO
Fam212b	2,564	0,584	1,50E-05	1,69E-03	Up in KO
Gm41896	3,314	2,087	0,0001096	4,88E-03	Up in KO
Nsl1	1,730	2,923	0,00011348	4,97E-03	Up in KO
Zfp618	-1,464	3,723	3,17E-05	2,36E-03	Down in KO
Ap1s2	1,524	1,800	1,93E-05	1,85E-03	Up in KO
Asic1	1,432	1,474	4,16E-05	2,74E-03	Up in KO
Jag2	1,619	2,978	3,70E-05	2,60E-03	Up in KO
Tcp1l12	-1,063	2,858	3,40E-05	2,47E-03	Down in KO
Ces1e	-1,043	3,782	2,84E-05	2,21E-03	Down in KO
Smc2	1,483	4,865	3,00E-05	2,30E-03	Up in KO
Adamts18	-7,563	0,324	1,54E-05	1,71E-03	Down in KO
Jph1	3,635	-0,046	3,54E-05	2,55E-03	Up in KO
Idh1	1,059	7,856	3,35E-05	2,45E-03	Up in KO
Prodh	-1,418	3,734	3,96E-05	2,70E-03	Down in KO
Agmat	3,508	0,029	2,03E-05	1,88E-03	Up in KO
Ccp110	1,355	3,166	3,66E-05	2,58E-03	Up in KO
Ticrr	1,546	3,360	3,90E-05	2,68E-03	Up in KO
Fads3	1,503	5,044	3,87E-05	2,68E-03	Up in KO
Cd3g	-2,844	1,892	6,94E-05	3,70E-03	Down in KO
2810417H13Rik	2,600	5,507	0,00010218	4,72E-03	Up in KO
Npnt	3,520	-0,169	2,06E-05	1,88E-03	Up in KO
Slc25a48	2,335	0,426	3,88E-05	2,68E-03	Up in KO
LY6d	2,171	3,992	6,53E-05	3,59E-03	Up in KO
Krt27	4,641	-0,383	4,89E-06	8,73E-04	Up in KO
Cpe	1,643	3,242	5,13E-05	3,08E-03	Up in KO
Rasd1	-1,168	5,337	2,99E-05	2,30E-03	Down in KO
Zcchc3	3,046	-0,211	6,08E-06	1,01E-03	Up in KO
Diaph3	1,434	3,237	8,57E-05	4,25E-03	Up in KO
Bglap3	1,343	4,299	4,24E-05	2,74E-03	Up in KO

Pla2g7	2,194	2,236	6,20E-05	3,50E-03	Up in KO
LOC101055907	1,655	0,825	5,98E-05	3,50E-03	Up in KO
Casp12	-3,578	1,080	4,87E-05	3,00E-03	Down in KO
Cadm1	2,090	1,064	3,65E-05	2,58E-03	Up in KO
Zwilch	1,579	3,599	5,69E-05	3,37E-03	Up in KO
Ly6c1	3,318	1,884	0,00013489	5,48E-03	Up in KO
Elavl2	7,881	-0,839	1,05E-05	1,39E-03	Up in KO
Scara5	-2,933	-0,394	1,93E-06	4,94E-04	Down in KO
Pdx1	3,696	-0,562	6,15E-06	1,01E-03	Up in KO
Brca2	1,716	2,663	9,04E-05	4,34E-03	Up in KO
Fanca	1,857	4,120	6,16E-05	3,50E-03	Up in KO
Ntn4	-1,651	2,274	8,37E-05	4,18E-03	Down in KO
Rtn4r	-1,290	3,390	4,69E-05	2,95E-03	Down in KO
Nr2e3	-2,997	-0,038	1,56E-05	1,71E-03	Down in KO
Fndc5	-2,902	2,180	0,00012079	5,09E-03	Down in KO
Tdrp	2,477	1,771	0,00011869	5,08E-03	Up in KO
Casc5	1,195	3,552	3,80E-05	2,65E-03	Up in KO
Btbd19	1,387	3,467	5,06E-05	3,08E-03	Up in KO
Cmah	1,701	6,998	6,70E-05	3,61E-03	Up in KO
C77370	1,268	2,860	0,00010455	4,76E-03	Up in KO
Hoxd3	-2,936	0,502	5,51E-05	3,28E-03	Down in KO
Proc	1,622	1,985	0,00010789	4,83E-03	Up in KO
Apbb1ip	-1,638	1,371	5,99E-05	3,50E-03	Down in KO
Lgr5	-2,425	1,218	8,62E-05	4,26E-03	Down in KO
Cd3e	-2,907	1,303	9,75E-05	4,56E-03	Down in KO
Cdkn2a	5,251	-0,312	1,34E-05	1,60E-03	Up in KO
Nebi	2,117	1,396	0,00019362	6,55E-03	Up in KO
Trim29	1,799	2,910	6,72E-05	3,61E-03	Up in KO
Gm38563	1,741	1,357	5,02E-05	3,07E-03	Up in KO
Card11	-2,200	1,481	7,51E-05	3,91E-03	Down in KO
Limch1	-3,790	-0,399	4,18E-05	2,74E-03	Down in KO
Hpgds	-1,326	1,626	0,00015104	5,78E-03	Down in KO
Krt23	-1,958	2,665	8,71E-05	4,27E-03	Down in KO
Tlcd2	1,312	4,310	4,81E-05	2,99E-03	Up in KO
Psat1	2,060	3,425	0,00011085	4,90E-03	Up in KO
Wnk2	-1,234	3,656	4,09E-05	2,74E-03	Down in KO
Tiam1	1,692	3,820	1,37E-05	1,61E-03	Up in KO
H2-T3	-1,836	2,683	7,06E-05	3,75E-03	Down in KO
Ppp4r4	2,211	0,878	3,95E-05	2,70E-03	Up in KO
Pde1c	-2,698	0,104	0,0003991	9,64E-03	Down in KO
Gm33198	3,080	3,415	0,00022703	7,07E-03	Up in KO
Gm30069	3,343	1,198	0,00017353	6,27E-03	Up in KO
Cd74	-5,375	7,263	0,00015249	5,82E-03	Down in KO
Dclk3	-2,542	1,715	0,00013131	5,40E-03	Down in KO
Gm28207	6,370	-0,121	3,61E-06	7,00E-04	Up in KO
Amigo2	-3,506	-0,060	2,20E-05	1,93E-03	Down in KO
Cpne5	1,945	0,393	3,27E-05	2,42E-03	Up in KO
Vav2	-1,856	4,232	9,34E-05	4,42E-03	Down in KO
Pcdhga7	-3,265	-0,291	9,73E-05	4,56E-03	Down in KO
Fmnl3	1,476	2,154	8,12E-05	4,12E-03	Up in KO
Plcb2	-1,922	0,622	6,57E-05	3,60E-03	Down in KO
Gm39143	2,960	2,502	0,00023242	7,16E-03	Up in KO

Prkcdp	1,382	1,679	0,00013813	5,54E-03	Up in KO
Ccne1	2,112	3,704	0,0001451	5,65E-03	Up in KO
6030426L16Rik	1,314	2,195	1,59E-05	1,71E-03	Up in KO
Rdh9	1,641	5,140	9,35E-05	4,42E-03	Up in KO
Slc6a20a	-2,264	0,094	2,52E-05	2,04E-03	Down in KO
Npc111	1,954	2,434	0,00012135	5,09E-03	Up in KO
Ranbp17	1,832	2,053	0,00019673	6,55E-03	Up in KO
Nav1	-2,366	1,212	7,35E-05	3,85E-03	Down in KO
Ly6d	6,468	4,217	2,28E-05	1,95E-03	Up in KO
Scgb2b20	5,120	-0,432	4,57E-06	8,34E-04	Up in KO
Gimap4	-3,060	2,783	0,00025267	7,43E-03	Down in KO
Mybl2	1,786	3,949	0,0001377	5,54E-03	Up in KO
Map9	1,099	3,523	8,65E-05	4,26E-03	Up in KO
Dyrk3	2,091	1,243	0,00019827	6,56E-03	Up in KO
Hdac11	-1,021	4,915	7,47E-05	3,90E-03	Down in KO
Dnah2	1,077	1,551	0,00011531	5,00E-03	Up in KO
Cyp2d9	1,921	6,037	0,00014406	5,64E-03	Up in KO
Mylip	-1,019	3,791	7,83E-05	4,04E-03	Down in KO
Gdf15	1,672	6,425	9,68E-05	4,55E-03	Up in KO
Gm33617	-2,436	0,193	4,47E-05	2,83E-03	Down in KO
sept-06	-1,225	1,870	6,69E-05	3,61E-03	Down in KO
Sh2d6	-1,323	2,593	0,00014154	5,62E-03	Down in KO
Tfpi	-3,037	-0,187	2,10E-05	1,88E-03	Down in KO
Gm41143	3,238	1,866	0,00026603	7,73E-03	Up in KO
Rgs1	-2,629	3,914	0,00024198	7,30E-03	Down in KO
Slit2	1,348	2,092	0,00016011	6,04E-03	Up in KO
S100a11	1,392	7,024	0,00021938	6,96E-03	Up in KO
Crif2	1,336	2,436	8,24E-05	4,13E-03	Up in KO
Ldhd	1,120	3,598	0,00012091	5,09E-03	Up in KO
Top2a	1,653	6,880	0,00014695	5,68E-03	Up in KO
Ldlrad4	-1,923	1,729	0,00031166	8,42E-03	Down in KO
Fancd2	1,855	2,999	0,00016887	6,15E-03	Up in KO
LOC102638993	1,039	2,997	0,00010004	4,65E-03	Up in KO
Ephx1	1,595	4,553	0,00014415	5,64E-03	Up in KO
Apol7e	-2,697	2,075	0,00023785	7,26E-03	Down in KO
Epdr1	2,906	1,817	0,00022256	6,99E-03	Up in KO
Pls3	1,028	4,263	8,99E-05	4,34E-03	Up in KO
Ercc6l	2,018	3,258	0,00019667	6,55E-03	Up in KO
Gzma	-3,321	3,071	0,00044965	1,04E-02	Down in KO
Adgrg5	-3,405	1,450	0,00028397	7,96E-03	Down in KO
Tbx21	-3,136	0,759	0,00018317	6,41E-03	Down in KO
Ndc1	1,263	4,189	0,00027088	7,84E-03	Up in KO
Hmcn2	4,107	-0,836	2,08E-05	1,88E-03	Up in KO
H2-DMA	-2,005	3,245	0,00132893	1,97E-02	Down in KO
Rbp7	1,916	1,078	0,00024836	7,35E-03	Up in KO
Tulp3	1,585	1,490	0,00020566	6,63E-03	Up in KO
Mogat2	1,440	3,726	0,00010571	4,80E-03	Up in KO
Peg3	-2,243	-0,053	4,02E-05	2,71E-03	Down in KO
Hck	-1,999	0,854	0,00013494	5,48E-03	Down in KO
BC055324	1,474	2,613	0,00019426	6,55E-03	Up in KO
LOC108168882	2,717	1,652	0,00024768	7,34E-03	Up in KO
Tcf23	1,228	2,819	1,55E-05	1,71E-03	Up in KO

Gm40902	-3,917	0,146	0,00023808	7,26E-03	Down in KO
Thbs1	-1,819	9,472	0,00020567	6,63E-03	Down in KO
Maats1	1,748	0,369	4,39E-05	2,79E-03	Up in KO
Rhobtb3	-1,480	1,976	0,00020774	6,67E-03	Down in KO
Gsta3	2,008	4,334	0,00023994	7,27E-03	Up in KO
4931428L18Rik	2,749	-0,268	4,85E-05	3,00E-03	Up in KO
Rad18	1,406	2,535	0,00021542	6,86E-03	Up in KO
Pltp	1,497	1,440	0,00017613	6,28E-03	Up in KO
Resp18	-2,134	0,748	0,00012934	5,35E-03	Down in KO
Tgtp2	-3,208	1,836	0,00041424	9,81E-03	Down in KO
Pclo	-2,378	0,273	0,00014989	5,77E-03	Down in KO
Krt79	3,984	-0,937	1,72E-05	1,80E-03	Up in KO
Ttc21a	2,186	-0,281	1,89E-05	1,85E-03	Up in KO
Lipo3	-1,285	3,322	0,00010436	4,76E-03	Down in KO
Grin3a	2,292	4,457	0,00029925	8,18E-03	Up in KO
Tagap	-1,660	0,905	0,00010277	4,72E-03	Down in KO
LY6E	2,064	7,369	0,00029836	8,17E-03	Up in KO
Gm38946	2,782	3,268	0,00060339	1,24E-02	Up in KO
Aspg	-1,201	5,882	0,00014036	5,60E-03	Down in KO
Gm35630	2,735	-0,529	2,28E-05	1,95E-03	Up in KO
Gm37416	1,592	2,943	0,00020159	6,60E-03	Up in KO
Kif24	1,198	2,366	0,0002229	6,99E-03	Up in KO
Slc25a4	1,477	4,740	0,00019386	6,55E-03	Up in KO
H2-DMb1	-5,015	2,964	0,000194	6,55E-03	Down in KO
Inpp5d	-2,207	1,519	0,00018372	6,41E-03	Down in KO
Adgrg2	1,504	0,944	0,00014264	5,63E-03	Up in KO
Arnt2	1,398	2,329	0,00018393	6,41E-03	Up in KO
Adck3	-1,575	4,274	0,00017734	6,28E-03	Down in KO
Ugt8a	1,893	3,466	0,00018512	6,43E-03	Up in KO
Sgol2a	1,088	3,790	9,35E-05	4,42E-03	Up in KO
Klrk1	-2,446	0,750	0,00032127	8,58E-03	Down in KO
Sned1	-1,662	4,529	6,32E-05	3,52E-03	Down in KO
Map2	1,860	2,199	0,00030738	8,33E-03	Up in KO
Slc7a15	2,596	1,938	0,00038362	9,43E-03	Up in KO
Adcy4	3,051	0,524	0,00052691	1,13E-02	Up in KO
Dusp5	-1,313	5,696	0,00011462	5,00E-03	Down in KO
Fbxo48	2,148	0,024	4,11E-05	2,74E-03	Up in KO
Gm41101	2,759	2,533	0,00046092	1,05E-02	Up in KO
Trip13	1,596	3,116	0,00024556	7,32E-03	Up in KO
Pbk	2,182	4,688	0,0004331	1,02E-02	Up in KO
Klrd1	-2,763	1,300	0,0002314	7,15E-03	Down in KO
Sh2d2a	-2,136	1,914	0,00063308	1,28E-02	Down in KO
Fam111a	1,913	4,024	0,00028949	8,01E-03	Up in KO
Figl1	1,882	3,125	0,00024609	7,32E-03	Up in KO
Cav2	1,939	3,023	0,00027635	7,89E-03	Up in KO
Foxm1	1,695	4,508	0,00024328	7,30E-03	Up in KO
Ccdc3	2,329	0,536	0,00020863	6,68E-03	Up in KO
Rnf39	-1,170	5,264	9,93E-05	4,62E-03	Down in KO
Cyp3a44	-3,958	0,185	0,00017986	6,31E-03	Down in KO
Cdsn	2,464	0,830	0,00031814	8,51E-03	Up in KO
H2-T10	-1,536	2,482	0,00019496	6,55E-03	Down in KO
Arhgap9	1,710	3,681	0,00064795	1,30E-02	Up in KO

Slfn2	-2,086	0,547	0,00010998	4,88E-03	Down in KO
Pcdh17	-2,140	2,945	0,00034049	8,92E-03	Down in KO
Tacc1	-1,520	1,616	0,0002421	7,30E-03	Down in KO
Asf1b	2,117	3,802	0,00039494	9,58E-03	Up in KO
Gzmb	-3,041	3,353	0,00067642	1,34E-02	Down in KO
Esco2	2,042	3,622	0,00036304	9,14E-03	Up in KO
Dpyd	-1,439	5,176	0,00016689	6,14E-03	Down in KO
Sprp2h	3,776	-0,766	0,00010206	4,72E-03	Up in KO
C87977	6,108	-0,342	5,11E-05	3,08E-03	Up in KO
Ptpcr	-2,626	3,892	0,00053854	1,14E-02	Down in KO
Pdgfc	1,329	3,633	0,00032511	8,65E-03	Up in KO
Cd69	-2,418	0,113	8,89E-05	4,32E-03	Down in KO
Eci3	-1,065	3,129	0,00019018	6,55E-03	Down in KO
Ces2b	-1,377	3,007	0,00018802	6,50E-03	Down in KO
Fasl	-3,375	0,300	0,00029067	8,03E-03	Down in KO
Prpt1	-3,184	0,341	0,00020425	6,62E-03	Down in KO
Gpr171	-2,922	0,727	0,0003137	8,46E-03	Down in KO
Plekhh2	1,411	2,877	6,07E-05	3,50E-03	Up in KO
Cd7	-2,629	2,713	0,00050499	1,10E-02	Down in KO
Mns1	1,490	1,896	0,00031673	8,48E-03	Up in KO
Bnip3	1,109	3,642	0,00020752	6,67E-03	Up in KO
E2f8	1,255	4,355	0,00014664	5,68E-03	Up in KO
Gm40034	1,244	1,491	0,00028076	7,95E-03	Up in KO
Limd2	-1,037	1,155	0,00078046	1,46E-02	Down in KO
Pde8b	-1,058	3,653	0,00017477	6,28E-03	Down in KO
ANXA9	1,541	0,787	0,0001737	6,27E-03	Up in KO
Nfam1	2,424	0,101	4,24E-05	2,74E-03	Up in KO
Papln	1,685	1,607	0,0003554	9,09E-03	Up in KO
Farp2	-1,056	5,223	0,00013586	5,51E-03	Down in KO
Rad51	2,117	3,842	0,00046007	1,05E-02	Up in KO
Pgap1	1,068	5,520	0,0001594	6,03E-03	Up in KO
Csrp2	-1,114	6,726	0,00022475	7,03E-03	Down in KO
Slain1	-1,175	1,663	0,00030013	8,19E-03	Down in KO
Fzd3	-1,438	2,213	0,00026632	7,73E-03	Down in KO
Dusp22	1,921	2,742	0,00023086	7,15E-03	Up in KO
P2ry10	-2,851	2,505	0,00066367	1,32E-02	Down in KO
Snhg7os	1,935	0,401	2,90E-05	2,25E-03	Up in KO
Gm36595	3,501	-0,869	0,00010266	4,72E-03	Up in KO
Snx32	1,943	0,443	0,00011226	4,94E-03	Up in KO
E2f2	1,176	5,698	0,00017202	6,24E-03	Up in KO
Ppp2r2c	-2,878	-0,234	0,00012931	5,35E-03	Down in KO
Grasp	-1,404	3,131	0,00027182	7,84E-03	Down in KO
Plat	2,045	0,919	0,00025118	7,40E-03	Up in KO
Arhgef17	1,791	0,334	0,00018817	6,50E-03	Up in KO
Dmrta1	3,492	-0,772	0,00011925	5,09E-03	Up in KO
Nrn1	-5,007	-0,882	2,62E-05	2,08E-03	Down in KO
Fyb	-1,868	1,662	0,00022519	7,03E-03	Down in KO
Clspn	1,970	3,947	0,00047768	1,07E-02	Up in KO
Gm29917	-2,197	-0,286	6,70E-05	3,61E-03	Down in KO
Zap70	-2,516	1,339	0,0004552	1,04E-02	Down in KO
Bmp4	2,883	-0,363	5,73E-05	3,38E-03	Up in KO
Klf8	1,233	1,088	0,00013824	5,54E-03	Up in KO

Lrmp	-1,912	1,983	0,0001612	6,04E-03	Down in KO
Nespas	5,341	-0,998	2,41E-06	5,64E-04	Up in KO
Abcc4	1,377	3,468	0,0002865	8,00E-03	Up in KO
Traf3ip3	-2,769	-0,314	0,00017139	6,23E-03	Down in KO
Gal3st2	-2,489	1,149	0,00043189	1,02E-02	Down in KO
D630023F18Rik	2,872	-0,382	7,97E-05	4,08E-03	Up in KO
Proser2	-1,028	3,768	0,00023064	7,15E-03	Down in KO
Igsf23	1,804	4,634	0,00027807	7,90E-03	Up in KO
Ccnf	1,989	3,831	0,00052194	1,13E-02	Up in KO
Ptpn22	-2,818	2,307	0,00112434	1,78E-02	Down in KO
Gcnt1	1,524	5,372	0,00031613	8,48E-03	Up in KO
Chst12	-2,095	0,436	0,00014991	5,77E-03	Down in KO
Blm	1,408	2,664	0,00047863	1,07E-02	Up in KO
Bfsp1	1,767	3,648	0,00048192	1,07E-02	Up in KO
Treh	2,110	1,682	0,00050212	1,10E-02	Up in KO
Zfp385a	1,692	0,477	0,00023478	7,20E-03	Up in KO
Maml2	-2,458	-0,528	0,00013698	5,54E-03	Down in KO
Xcl1	-1,745	0,536	0,00013202	5,40E-03	Down in KO
Map10	-1,710	0,369	0,00016718	6,14E-03	Down in KO
Rac2	-2,044	2,487	0,00046107	1,05E-02	Down in KO
Sulf2	1,017	5,105	0,00017786	6,28E-03	Up in KO
Nkg7	-2,637	1,551	0,00058856	1,22E-02	Down in KO
Hmgcll1	1,774	0,698	0,00040493	9,65E-03	Up in KO
9130019O22Rik	-1,135	2,454	0,00033361	8,79E-03	Down in KO
Ambp	1,304	1,720	0,00034212	8,94E-03	Up in KO
Cd247	-2,114	0,380	0,00035895	9,12E-03	Down in KO
Ncapg2	1,715	4,266	0,00040333	9,65E-03	Up in KO
Wdhd1	1,125	4,229	0,00035466	9,09E-03	Up in KO
Thop1	1,073	5,676	0,00028402	7,96E-03	Up in KO
2310069B03Rik	2,466	0,931	0,0004787	1,07E-02	Up in KO
Gm28225	4,018	-0,644	0,00028323	7,96E-03	Up in KO
Pik3r5	-1,659	1,069	0,00035203	9,05E-03	Down in KO
Gm8979	-1,286	8,042	0,00036376	9,14E-03	Down in KO
Itk	-3,138	2,047	0,00082063	1,50E-02	Down in KO
Cd200r1	-2,995	0,318	0,00045246	1,04E-02	Down in KO
Slc6a19	1,928	2,035	0,00057332	1,20E-02	Up in KO
Neurod1	-1,859	0,237	0,00022083	6,99E-03	Down in KO
Gm42151	-1,346	1,224	0,00057838	1,21E-02	Down in KO
Gpr18	-3,511	-0,610	0,00017747	6,28E-03	Down in KO
Sox21	-1,331	2,604	0,0004434	1,03E-02	Down in KO
Tgtp1	-3,029	1,778	0,00112874	1,78E-02	Down in KO
Prss22	3,505	0,622	0,0007713	1,45E-02	Up in KO
Arntl2	1,492	1,046	0,00017837	6,28E-03	Up in KO
Acap1	-2,403	2,056	0,00076068	1,45E-02	Down in KO
Gm39847	2,100	-0,608	2,38E-05	1,98E-03	Up in KO
Slamf7	-2,060	0,175	0,00093021	1,60E-02	Down in KO
Tbc1d10c	-2,509	1,386	0,00071363	1,39E-02	Down in KO
Hmgn3	-2,269	-0,457	0,00020112	6,60E-03	Down in KO
Aadac	-1,649	2,704	0,00053154	1,13E-02	Down in KO
Gimap6	-2,511	1,859	0,00078408	1,46E-02	Down in KO
Cyp3a25	-2,621	-0,050	0,00025475	7,47E-03	Down in KO
Cenpi	1,682	2,555	0,00076224	1,45E-02	Up in KO

Grhl1	1,250	1,660	0,0006067	1,25E-02	Up in KO
Mms22l	1,115	3,339	0,00041004	9,74E-03	Up in KO
Per2	-1,276	5,172	0,00038849	9,48E-03	Down in KO
Gm15286	2,523	-0,486	0,00011703	5,05E-03	Up in KO
Gm20472	5,605	-1,410	1,73E-05	1,80E-03	Up in KO
BC048559	1,940	0,541	0,00025976	7,58E-03	Up in KO
Espl1	1,452	3,850	0,00048919	1,08E-02	Up in KO
Dtx1	-3,095	0,363	0,00060777	1,25E-02	Down in KO
Rnd2	1,248	1,475	0,00037306	9,32E-03	Up in KO
Ccl4	-3,316	0,106	0,00044905	1,04E-02	Down in KO
Sult1a1	-1,031	7,910	0,00037979	9,40E-03	Down in KO
Gngt2	-1,105	1,092	0,00055175	1,16E-02	Down in KO
Itgae	-2,328	2,830	0,00093394	1,60E-02	Down in KO
Gm29758	1,183	1,973	0,0005185	1,12E-02	Up in KO
Sectm1a	1,061	4,133	0,00027454	7,86E-03	Up in KO
Selplg	-2,302	2,105	0,00138746	2,02E-02	Down in KO
Smoc2	-2,640	2,430	0,00111427	1,77E-02	Down in KO
Itln1	2,485	0,673	0,0005452	1,15E-02	Up in KO
Des	2,015	-0,057	0,00027983	7,94E-03	Up in KO
Fbp1	-2,067	-0,070	0,00014287	5,63E-03	Down in KO
Arap3	-2,820	0,820	0,0007171	1,40E-02	Down in KO
Trim35	1,312	3,954	0,00053061	1,13E-02	Up in KO
1500009L16Rik	1,060	3,257	0,00034305	8,94E-03	Up in KO
Nav3	5,365	-1,602	1,29E-05	1,58E-03	Up in KO
Gm36840	3,442	-1,205	2,53E-05	2,04E-03	Up in KO
Hvcn1	-2,143	0,720	0,00050702	1,11E-02	Down in KO
Arhgdig	1,791	0,527	0,00064826	1,30E-02	Up in KO
Peg13	-1,119	3,521	0,00035581	9,09E-03	Down in KO
Hoxd8	-3,021	-0,143	0,00082302	1,50E-02	Down in KO
Cd160	-1,748	0,699	0,0002616	7,62E-03	Down in KO
Gm41090	2,834	0,877	0,00089282	1,57E-02	Up in KO
Rasa3	-2,235	1,497	0,00098349	1,65E-02	Down in KO
Ctsn	-2,515	2,357	0,00092757	1,60E-02	Down in KO
LOC108168395	-1,094	4,166	0,00034444	8,95E-03	Down in KO
Mdm1	1,052	2,478	0,00058116	1,21E-02	Up in KO
Akna	-1,175	3,116	0,00043418	1,02E-02	Down in KO
H2-Ab1	-3,759	5,223	0,00052409	1,13E-02	Down in KO
Lgsn	3,242	-0,598	0,00047325	1,06E-02	Up in KO
Rasa1	2,114	0,058	0,0001967	6,55E-03	Up in KO
LOC108168067	1,462	6,302	0,00064785	1,30E-02	Up in KO
Zfp37	1,122	4,152	0,00037627	9,35E-03	Up in KO
Cd96	-2,739	0,740	0,00082467	1,50E-02	Down in KO
Gbp9	-2,333	1,306	0,00101083	1,68E-02	Down in KO
Egr3	-1,500	1,133	0,00047748	1,07E-02	Down in KO
Slc37a2	-3,493	4,708	0,00046958	1,06E-02	Down in KO
2410018L13Rik	3,815	-1,284	4,21E-05	2,74E-03	Up in KO
Abcg1	-1,162	1,469	0,00052335	1,13E-02	Down in KO
Zgrf1	1,572	2,354	0,0001935	6,55E-03	Up in KO
Ptpn	-1,547	1,066	0,00049473	1,09E-02	Down in KO
Rad54l	1,726	3,228	0,0016963	2,29E-02	Up in KO
Bard1	1,657	2,419	0,00086894	1,54E-02	Up in KO
AI504432	-2,648	-0,173	0,00033191	8,78E-03	Down in KO

Gm32298	1,100	3,134	0,00039133	9,52E-03	Up in KO
H2-BI	-1,455	4,772	0,00053458	1,14E-02	Down in KO
Mvb12b	-1,249	2,033	0,00121508	1,87E-02	Down in KO
Rbms3	-1,394	1,834	0,00079133	1,47E-02	Down in KO
Fabp5	1,894	3,386	0,0010978	1,75E-02	Up in KO
Spn	-2,558	1,499	0,00099167	1,66E-02	Down in KO
Phf1	-1,101	5,380	0,00037431	9,32E-03	Down in KO
Slc5a4b	1,493	0,262	0,0002045	6,62E-03	Up in KO
Gen1	1,214	2,549	0,000177	6,28E-03	Up in KO
Sema3d	2,604	-0,627	0,00013998	5,60E-03	Up in KO
Gm13056	1,438	1,321	0,00048886	1,08E-02	Up in KO
Adgrb1	2,340	1,579	0,00123554	1,89E-02	Up in KO
Gbp4	-3,316	1,583	0,00037173	9,31E-03	Down in KO
Ereg	1,821	6,352	0,00091078	1,58E-02	Up in KO
Ackr3	-2,496	0,004	0,00038059	9,40E-03	Down in KO
Sez6l2	-1,538	0,264	0,0002006	6,60E-03	Down in KO
Ptgr1	1,652	4,884	0,00082604	1,50E-02	Up in KO
Sh3rf2	-1,357	3,791	0,00027287	7,84E-03	Down in KO
Slc4a11	1,966	0,161	0,00140527	2,04E-02	Up in KO
Dact1	2,181	0,050	0,00035332	9,07E-03	Up in KO
Gpr34	-3,065	0,549	0,00103438	1,70E-02	Down in KO
C630043F03Rik	1,053	1,441	0,00066492	1,32E-02	Up in KO
Gpr132	-2,437	1,582	0,00110239	1,76E-02	Down in KO
Hist1h3d	2,111	0,622	0,00059078	1,22E-02	Up in KO
Lax1	-2,635	-0,444	0,00018747	6,50E-03	Down in KO
Cdk14	-2,654	-1,063	2,31E-05	1,96E-03	Down in KO
Magi2	2,349	-0,583	0,00016477	6,12E-03	Up in KO
Gimap3	-2,318	2,776	0,0012921	1,94E-02	Down in KO
Ltbp3	1,433	0,174	0,00028873	8,01E-03	Up in KO
Eva1c	-1,933	0,372	0,0001224	5,09E-03	Down in KO
Edn1	-1,341	7,181	0,00071481	1,39E-02	Down in KO
Alox5	-2,095	-0,470	0,00015866	6,01E-03	Down in KO
Il21r	-2,745	2,231	0,00248991	2,91E-02	Down in KO
H2-Eb1	-3,879	4,479	0,00075311	1,44E-02	Down in KO
Anln	1,200	4,368	0,00014664	5,68E-03	Up in KO
Prrx1	2,981	-1,300	1,87E-05	1,85E-03	Up in KO
Per1	-1,320	6,234	0,00063234	1,28E-02	Down in KO
Trib3	1,652	-0,348	4,12E-05	2,74E-03	Up in KO
Gusb	1,121	5,225	0,00034909	9,00E-03	Up in KO
Dsn1	1,579	2,965	0,00098108	1,65E-02	Up in KO
Tle6	1,496	2,492	0,00082007	1,50E-02	Up in KO
Hacd4	1,449	1,198	0,00403165	3,88E-02	Up in KO
Mcm10	1,012	4,344	0,00041552	9,82E-03	Up in KO
Il33	2,056	1,332	0,00087297	1,54E-02	Up in KO
Cdkl1	-2,963	-0,183	0,00036063	9,12E-03	Down in KO
Cxxc4	1,235	0,706	0,00093031	1,60E-02	Up in KO
Kifc5b	1,154	2,615	0,0009351	1,60E-02	Up in KO
Krtap13	3,408	-0,928	0,00033313	8,79E-03	Up in KO
Gm34572	2,323	0,273	0,00084386	1,51E-02	Up in KO
Sla2	-1,960	0,684	0,00073164	1,41E-02	Down in KO
Il2rg	-2,248	1,315	0,00111736	1,77E-02	Down in KO
A930004D18Rik	3,053	-1,025	0,00024058	7,28E-03	Up in KO

LOC108167858	1,990	1,974	0,00165975	2,26E-02	Up in KO
Kif15	1,333	4,258	0,0004485	1,04E-02	Up in KO
Ms4a4b	-3,025	-0,368	0,00063765	1,29E-02	Down in KO
Rtp3	4,166	-1,498	2,36E-05	1,97E-03	Up in KO
Nuf2	1,784	3,525	0,00120281	1,85E-02	Up in KO
Sp140	-1,533	1,058	0,00077628	1,46E-02	Down in KO
Gm30311	1,885	3,748	0,00016428	6,12E-03	Up in KO
Dock2	-2,454	1,305	0,00105386	1,72E-02	Down in KO
Nr4a3	-1,783	2,244	7,62E-05	3,94E-03	Down in KO
Pde4b	-1,743	1,930	0,00104633	1,72E-02	Down in KO
Gm38978	-1,811	-0,257	0,00012251	5,09E-03	Down in KO
Aqp3	4,015	-1,391	4,09E-05	2,74E-03	Up in KO
Gtse1	1,713	3,639	0,0012719	1,92E-02	Up in KO
Gm30438	1,239	0,860	0,00040543	9,65E-03	Up in KO
Coro1a	-1,960	2,502	0,00115885	1,81E-02	Down in KO
Rrm1	1,599	5,706	0,00113276	1,78E-02	Up in KO
H2-Aa	-4,311	4,937	0,00102971	1,70E-02	Down in KO
Gm8995	-1,397	4,066	0,00077434	1,46E-02	Down in KO
Rab37	-1,160	2,253	0,00075906	1,44E-02	Down in KO
Gsdma	-1,669	-0,033	0,00023988	7,27E-03	Down in KO
Dcbld2	1,581	2,845	0,00119513	1,84E-02	Up in KO
Thy1	-2,317	1,130	0,00113314	1,78E-02	Down in KO
Serpinb9	-1,393	2,058	0,00057888	1,21E-02	Down in KO
Cers3	1,936	-0,005	0,00036055	9,12E-03	Up in KO
Glpr2	-1,111	2,099	0,00068699	1,35E-02	Down in KO
sept-01	-1,964	1,576	0,00058776	1,22E-02	Down in KO
Rcsd1	-2,269	0,889	0,00140079	2,04E-02	Down in KO
Enpep	2,226	5,967	0,00171897	2,31E-02	Up in KO
Fut9	-4,068	-0,250	0,00161494	2,22E-02	Down in KO
Notch3	2,253	-0,241	0,00067424	1,34E-02	Up in KO
Kif11	1,242	5,034	0,00084398	1,51E-02	Up in KO
Fli1	-2,238	-0,038	0,00061764	1,26E-02	Down in KO
Cd84	1,221	4,837	0,00072809	1,41E-02	Up in KO
Ptptr	-1,458	1,125	0,00077201	1,45E-02	Down in KO
Nup133	1,021	4,292	0,00071904	1,40E-02	Up in KO
Gm9292	2,713	-1,023	9,40E-05	4,43E-03	Up in KO
Laptm5	-2,201	3,188	0,0016517	2,25E-02	Down in KO
E130012A19Rik	1,338	4,843	0,00080254	1,48E-02	Up in KO
Mndal	-2,065	1,039	0,00230631	2,77E-02	Down in KO
Dna2	1,458	2,924	0,00113832	1,79E-02	Up in KO
A330049N07Rik	1,040	0,893	0,00060597	1,25E-02	Up in KO
BC021614	1,762	2,194	0,00108652	1,75E-02	Up in KO
Gm40405	2,563	1,279	0,0024652	2,88E-02	Up in KO
Klhl13	-2,293	0,083	0,00080823	1,48E-02	Down in KO
Rdh1	1,113	3,620	0,00030731	8,33E-03	Up in KO
Chaf1a	1,471	4,197	0,00113062	1,78E-02	Up in KO
Ccne2	1,289	2,730	0,00096213	1,63E-02	Up in KO
Stat4	-2,912	0,300	0,00127892	1,93E-02	Down in KO
Pde7b	1,503	0,325	0,00071279	1,39E-02	Up in KO
Nptx2	1,846	0,710	0,00097239	1,64E-02	Up in KO
BC025446	1,026	4,401	0,00071202	1,39E-02	Up in KO
Mst1	2,143	-0,437	0,00028165	7,95E-03	Up in KO

Srgn	-1,852	1,679	0,00138214	2,02E-02	Down in KO
Il2rb	-2,344	3,237	0,0027074	3,06E-02	Down in KO
Ncapg	1,315	4,390	0,00127388	1,92E-02	Up in KO
Acot12	1,828	-0,234	0,0001218	5,09E-03	Up in KO
Ass1	1,618	4,271	0,00141611	2,05E-02	Up in KO
Sema3f	1,105	3,070	0,00074942	1,43E-02	Up in KO
Arhgap25	-2,605	0,313	0,00091417	1,58E-02	Down in KO
Slc26a7	4,538	-1,657	1,34E-05	1,60E-03	Up in KO
Slc16a2	-1,777	0,470	0,00091949	1,59E-02	Down in KO
Rnf122	-1,138	1,199	0,00076332	1,45E-02	Down in KO
Gm12216	-1,542	0,492	0,00049248	1,08E-02	Down in KO
Gm15441	1,005	1,267	0,00083742	1,51E-02	Up in KO
Ppp3r2	2,372	-0,139	0,00077883	1,46E-02	Up in KO
RASSF5-8	1,483	0,169	0,00267778	3,04E-02	Up in KO
Evi2b	-2,560	0,161	0,00093816	1,61E-02	Down in KO
Sla	-2,499	0,988	0,00286465	3,16E-02	Down in KO
Irx3	2,331	-0,836	0,00035097	9,04E-03	Up in KO
Skap1	-2,199	-0,125	0,00073749	1,41E-02	Down in KO
Vsir	-1,783	0,754	0,00083635	1,51E-02	Down in KO
Ciita	-3,727	2,846	0,00094266	1,61E-02	Down in KO
Prkg2	-1,390	5,844	0,00079704	1,48E-02	Down in KO
Incenp	1,043	5,288	0,00086284	1,54E-02	Up in KO
9030619P08Rik	1,764	0,897	0,00123834	1,89E-02	Up in KO
4930427A07Rik	1,041	4,072	0,00095102	1,62E-02	Up in KO
Sox12	1,415	0,644	0,00090043	1,57E-02	Up in KO
Rhoh	-1,941	0,826	0,00116022	1,81E-02	Down in KO
Cd53	-2,444	1,068	0,00154813	2,16E-02	Down in KO
Phgdh	2,057	4,085	0,00212819	2,63E-02	Up in KO
C330027C09Rik	1,116	4,452	0,00059408	1,23E-02	Up in KO
Gm10354	3,802	-1,816	0,00011972	5,09E-03	Up in KO
Dlgap5	1,360	3,877	0,00128164	1,93E-02	Up in KO
Sash3	-2,333	0,811	0,00135057	1,99E-02	Down in KO
Gprc5b	2,135	-0,533	0,00016307	6,09E-03	Up in KO
Gpr55	-2,909	0,874	0,00200586	2,54E-02	Down in KO
Cdc6	1,065	3,037	0,00143632	2,06E-02	Up in KO
Lck	-1,234	3,014	0,00076412	1,45E-02	Down in KO
Lrp8	1,781	2,381	0,00150065	2,12E-02	Up in KO
Lgals1	1,355	1,000	0,00142939	2,06E-02	Up in KO
Zfp184	1,155	0,956	0,00164003	2,25E-02	Up in KO
Cxcr6	-2,670	0,523	0,00153243	2,15E-02	Down in KO
Bub1	1,231	3,646	0,00117634	1,82E-02	Up in KO
Suv39h2	1,056	1,850	0,00132817	1,97E-02	Up in KO
Neil3	1,668	3,378	0,00184415	2,41E-02	Up in KO
Hsf2bp	1,559	1,159	0,00059494	1,23E-02	Up in KO
Mdfi	1,950	-0,489	0,00012169	5,09E-03	Up in KO
Fosb	-1,088	9,850	0,00125668	1,90E-02	Down in KO
Dmkn	2,754	0,178	0,0001118	4,93E-03	Up in KO
Ska1	1,660	1,681	0,00162236	2,23E-02	Up in KO
AW551984	2,227	0,156	0,00115103	1,80E-02	Up in KO
Mastl	1,139	3,013	0,00090399	1,58E-02	Up in KO
Pou6f1	-1,313	1,675	0,00077777	1,46E-02	Down in KO
Gm42368	1,321	2,831	0,00107493	1,74E-02	Up in KO

Pik3cd	-1,891	2,450	0,00212176	2,62E-02	Down in KO
Lrrc8c	-2,451	0,622	0,00101877	1,69E-02	Down in KO
Tlr2	-1,114	4,337	0,00087864	1,55E-02	Down in KO
Pxdc1	1,435	2,753	0,00135778	1,99E-02	Up in KO
Zfp658	-1,091	1,250	0,00103313	1,70E-02	Down in KO
Bco1	1,018	1,625	0,00164215	2,25E-02	Up in KO
H2-T-ps	-1,313	2,513	0,00086557	1,54E-02	Down in KO
Cd48	-2,437	0,376	0,00130213	1,95E-02	Down in KO
Apob	1,074	8,121	0,00109122	1,75E-02	Up in KO
Nab2	1,053	1,344	0,00188234	2,44E-02	Up in KO
Pigz	2,637	0,228	0,00118072	1,83E-02	Up in KO
Dusp14	1,179	0,420	0,00048971	1,08E-02	Up in KO
D7Ert128e	2,982	-1,528	2,09E-05	1,88E-03	Up in KO
Trp53cor1	3,022	-1,307	0,0003812	9,40E-03	Up in KO
Gm32677	1,159	1,579	0,00107163	1,74E-02	Up in KO
Cyp2r1	-2,136	-0,568	8,92E-05	4,32E-03	Down in KO
Rnf212	-1,170	2,062	0,00103089	1,70E-02	Down in KO
Melk	1,777	4,466	0,00200669	2,54E-02	Up in KO
Pitpnm2	1,062	2,472	0,00147413	2,10E-02	Up in KO
BC030867	1,685	1,857	0,00080399	1,48E-02	Up in KO
Fbn1	-1,698	1,090	0,00123063	1,88E-02	Down in KO
Gm9994	-1,806	0,689	0,00103234	1,70E-02	Down in KO
Dock10	-2,457	2,108	0,00288812	3,17E-02	Down in KO
Arhgap30	-2,238	1,542	0,00209056	2,61E-02	Down in KO
BC018473	1,674	-0,398	0,00038503	9,44E-03	Up in KO
Il7r	-2,368	-0,645	0,00043693	1,02E-02	Down in KO
Cd200r4	-2,827	0,921	0,00217729	2,66E-02	Down in KO
Ptgs1	-1,633	-0,108	0,00058956	1,22E-02	Down in KO
Txk	-2,099	0,345	0,0012203	1,87E-02	Down in KO
Zfp454	-2,330	-1,337	0,00020082	6,60E-03	Down in KO
Pcdh19	3,816	-1,532	0,00016827	6,14E-03	Up in KO
Slc10a2	1,334	7,970	0,00155329	2,16E-02	Up in KO
Tchh	1,241	0,786	0,00105163	1,72E-02	Up in KO
LOC108168870	2,721	1,087	0,00340341	3,52E-02	Up in KO
Ces2a	-1,077	8,142	0,00118673	1,84E-02	Down in KO
Emb	-2,364	1,252	0,0022251	2,70E-02	Down in KO
Dhrs9	1,375	4,954	0,0014565	2,08E-02	Up in KO
Ccl25	1,014	2,032	0,00120081	1,85E-02	Up in KO
Cd2	-2,929	-1,454	6,16E-05	3,50E-03	Down in KO
Scg5	-2,325	-1,070	0,00013268	5,42E-03	Down in KO
Xlr4a	-3,170	1,030	0,00354146	3,60E-02	Down in KO
Rnase1	1,992	-0,899	8,81E-05	4,29E-03	Up in KO
Fam84a	1,185	2,277	0,00161773	2,23E-02	Up in KO
Fyn	-2,026	1,802	0,00202851	2,56E-02	Down in KO
Apol7b	-2,087	0,308	0,00109274	1,75E-02	Down in KO
Mtfr2	1,334	1,488	0,00167937	2,27E-02	Up in KO
Rnf24	1,079	4,004	0,00117138	1,82E-02	Up in KO
Nim1k	2,254	-0,976	0,00029108	8,03E-03	Up in KO
LOC108169043	1,092	4,202	0,00144987	2,08E-02	Up in KO
Ikzf3	-2,315	1,923	0,00401494	3,87E-02	Down in KO
Dmxl2	1,583	0,035	0,00047171	1,06E-02	Up in KO
Nr4a2	-1,315	3,567	0,00065744	1,32E-02	Down in KO

Cyfp2	-2,672	1,425	0,00294777	3,20E-02	Down in KO
P3h2	3,127	-1,444	0,00029403	8,09E-03	Up in KO
Gata4	2,172	1,930	0,0027564	3,10E-02	Up in KO
Spc25	1,288	3,904	0,00170547	2,30E-02	Up in KO
Mcm8	1,067	2,382	0,00282458	3,14E-02	Up in KO
Ak4	1,788	5,407	0,00238826	2,82E-02	Up in KO
Khdc1a	6,083	1,618	2,26E-05	1,95E-03	Up in KO
LOC108168394	-1,909	1,023	0,00154407	2,16E-02	Down in KO
Arhgdib	-1,793	1,119	0,00113428	1,78E-02	Down in KO
Gm33595	-1,455	2,683	0,00159461	2,20E-02	Down in KO
Gsta1	1,071	6,081	0,00133604	1,98E-02	Up in KO
Ppp1r14a	-1,939	-0,704	0,00023325	7,17E-03	Down in KO
Itgb7	-2,170	1,829	0,00255071	2,94E-02	Down in KO
Cdca2	1,034	3,871	0,00045208	1,04E-02	Up in KO
Unc119	1,261	4,731	0,00126293	1,91E-02	Up in KO
Col25a1	-2,167	-0,919	4,17E-05	2,74E-03	Down in KO
St6gal1	-1,444	4,397	0,0019478	2,50E-02	Down in KO
Hcls1	-1,618	0,787	0,0012538	1,90E-02	Down in KO
Cdca5	1,342	3,363	0,00202008	2,55E-02	Up in KO
Rnf183	1,443	-0,171	0,00036027	9,12E-03	Up in KO
Rnf157	1,700	0,219	0,000977	1,65E-02	Up in KO
Lcp2	-2,573	1,352	0,00294786	3,20E-02	Down in KO
Clca4a	1,068	10,143	0,00157117	2,18E-02	Up in KO
Fxyd5	-2,039	0,960	0,00201226	2,55E-02	Down in KO
Cyth4	-2,396	1,542	0,00283807	3,15E-02	Down in KO
Itgb2	-2,086	1,139	0,00252821	2,92E-02	Down in KO
Tmed6	1,844	1,580	0,00270599	3,06E-02	Up in KO
C2	1,269	2,631	0,00179916	2,37E-02	Up in KO
AA414768	1,292	0,382	0,0006395	1,29E-02	Up in KO
Nt5dc2	2,199	1,903	0,00231041	2,77E-02	Up in KO
Dennd4a	-1,061	3,615	0,00110784	1,77E-02	Down in KO
Cnnm1	-1,180	1,730	0,00130997	1,96E-02	Down in KO
Cacna1h	-1,212	4,111	0,0015638	2,17E-02	Down in KO
Skida1	1,564	-0,258	0,00121459	1,87E-02	Up in KO
Ilgp1	-3,884	3,659	0,00219801	2,68E-02	Down in KO
Lpxn	-1,302	1,448	0,00148938	2,11E-02	Down in KO
Cxcl5	2,096	1,069	0,00250601	2,92E-02	Up in KO
Reg4	1,422	5,261	0,00221985	2,70E-02	Up in KO
Trim6	1,137	3,954	0,0008866	1,56E-02	Up in KO
Ifit3b	1,647	2,974	0,00225586	2,73E-02	Up in KO
Traip	1,681	2,817	0,00342784	3,53E-02	Up in KO
Cst6	1,135	1,472	0,00182229	2,39E-02	Up in KO
Nfkbie	-1,044	1,367	0,00124494	1,89E-02	Down in KO
Gm30235	-1,913	0,891	0,00210744	2,62E-02	Down in KO
Agap2	-1,816	0,983	0,00097516	1,65E-02	Down in KO
Grm4	-1,381	1,860	0,00016473	6,12E-03	Down in KO
Cadm4	1,457	-0,069	0,0004695	1,06E-02	Up in KO
Zfp365	2,780	-1,538	4,27E-05	2,74E-03	Up in KO
BC021767	-1,053	2,069	0,00180996	2,38E-02	Down in KO
Ano3	4,854	-1,403	8,14E-05	4,12E-03	Up in KO
Prss41	1,947	0,045	0,00053881	1,14E-02	Up in KO
Malrd1	-1,244	4,700	0,0016996	2,29E-02	Down in KO

Pla2g2a	3,849	3,309	0,00116304	1,81E-02	Up in KO
Pak6	1,029	1,856	0,00195536	2,50E-02	Up in KO
Pygl	-1,165	0,807	0,00101953	1,69E-02	Down in KO
Fmn1l	-2,130	1,642	0,00285589	3,16E-02	Down in KO
Ankle1	1,552	3,303	0,00288601	3,17E-02	Up in KO
Dsel	-1,590	0,628	0,00172537	2,31E-02	Down in KO
Ang4	1,867	4,176	0,00410212	3,91E-02	Up in KO
A930001C03Rik	1,438	0,547	0,00146848	2,09E-02	Up in KO
Glp1r	-1,928	-0,683	0,00051071	1,11E-02	Down in KO
Myo1f	-2,212	0,775	0,00239981	2,83E-02	Down in KO
LOC102639958	1,661	-0,357	0,00056628	1,19E-02	Up in KO
Kntc1	1,586	3,418	0,00037443	9,32E-03	Up in KO
Gm36704	2,121	1,938	0,00358203	3,62E-02	Up in KO
Gm39275	1,314	1,518	0,00198811	2,53E-02	Up in KO
Evx1os	1,888	0,260	0,0021603	2,66E-02	Up in KO
Lpcat4	1,156	6,628	0,00173655	2,32E-02	Up in KO
Gm2115	-1,071	1,827	0,00155288	2,16E-02	Down in KO
Nanos1	1,288	0,793	0,00134964	1,99E-02	Up in KO
Efr3b	1,728	-0,072	0,00123082	1,88E-02	Up in KO
Ncapd2	1,192	5,857	0,00264074	3,01E-02	Up in KO
Mttp	1,433	1,594	0,00136093	2,00E-02	Up in KO
Ifi203	-2,195	0,921	0,00186195	2,42E-02	Down in KO
Xrcc2	1,122	1,664	0,00256017	2,95E-02	Up in KO
Lat	-2,074	0,130	0,00128773	1,94E-02	Down in KO
Cst7	-2,140	-0,698	0,00077552	1,46E-02	Down in KO
Trat1	-3,137	-0,461	0,00051969	1,12E-02	Down in KO
Arhgef6	-2,141	0,006	0,00193523	2,49E-02	Down in KO
Vsig2	1,028	4,574	0,00216662	2,66E-02	Up in KO
Dnah10	1,712	0,508	0,00238498	2,82E-02	Up in KO
Slfn9	1,226	4,176	0,00209848	2,61E-02	Up in KO
Gm34154	2,407	-1,359	9,08E-05	4,34E-03	Up in KO
LOC101056001	1,638	2,274	0,00231015	2,77E-02	Up in KO
C1qtnf6	1,278	5,415	0,00265814	3,03E-02	Up in KO
Fzd2	-1,833	-0,043	0,00129745	1,95E-02	Down in KO
Gpc1	1,085	0,705	0,00094527	1,61E-02	Up in KO
Ecm1	1,206	1,757	0,00296649	3,22E-02	Up in KO
Kctd15	-1,183	0,215	0,00062398	1,27E-02	Down in KO
Akr1c18	1,288	2,589	0,00234465	2,79E-02	Up in KO
Aaas	1,043	3,911	0,00217532	2,66E-02	Up in KO
Acox2	1,020	3,219	0,00172459	2,31E-02	Up in KO
Sema7a	1,523	3,553	0,00181911	2,39E-02	Up in KO
Chtf18	1,428	3,099	0,00281942	3,14E-02	Up in KO
Arhgap22	1,446	-0,498	0,00039924	9,64E-03	Up in KO
9030612E09Rik	-3,220	-1,740	0,00019758	6,55E-03	Down in KO
Hnmt	-1,927	0,151	0,00086534	1,54E-02	Down in KO
Csta1	1,950	0,684	0,00281394	3,14E-02	Up in KO
Gdpd2	-1,199	3,238	0,00211535	2,62E-02	Down in KO
Mad2l1	1,374	4,600	0,00290845	3,19E-02	Up in KO
Megf9	1,007	2,803	0,0021889	2,67E-02	Up in KO
Bag2	1,125	1,795	0,00287442	3,17E-02	Up in KO
Ciart	-1,559	2,709	0,00277656	3,11E-02	Down in KO
Pydc3	-2,108	-0,729	0,0007661	1,45E-02	Down in KO

Gm40892	-3,124	0,030	0,00251563	2,92E-02	Down in KO
Pole	1,477	3,651	0,00307228	3,29E-02	Up in KO
Nlrc3	-2,017	1,278	0,00331264	3,45E-02	Down in KO
Kif4	1,358	4,498	0,00077532	1,46E-02	Up in KO
Evx1	1,709	2,332	0,0016611	2,26E-02	Up in KO
Hmha1	-1,802	3,060	0,00238475	2,82E-02	Down in KO
Mal	1,238	6,364	0,00233132	2,78E-02	Up in KO
Gm33333	1,282	0,967	0,00188467	2,44E-02	Up in KO
LOC102632156	1,153	0,136	0,00107994	1,75E-02	Up in KO
Slc34a2	1,326	4,835	0,00270176	3,06E-02	Up in KO
Myom3	-1,250	2,630	0,00145181	2,08E-02	Down in KO
Spc24	1,323	3,463	0,00304692	3,27E-02	Up in KO
Pappa	2,283	-0,648	0,0029866	3,23E-02	Up in KO
Prox1	-2,270	-1,118	0,00016049	6,04E-03	Down in KO
Mthfd2	1,229	3,596	0,00298253	3,23E-02	Up in KO
Gm41029	2,285	-0,969	0,00211018	2,62E-02	Up in KO
Dnajb4	-1,033	4,872	0,00186179	2,42E-02	Down in KO
Hic1	-2,235	-0,388	0,00118905	1,84E-02	Down in KO
Adcy7	-2,050	1,697	0,00368368	3,68E-02	Down in KO
Fbxo5	1,293	3,192	0,00297396	3,22E-02	Up in KO
Galnt18	1,939	0,340	0,00282735	3,14E-02	Up in KO
Gm40491	-1,585	0,446	0,00172118	2,31E-02	Down in KO
Lat2	-1,392	1,041	0,00365462	3,66E-02	Down in KO
Hirip3	1,167	4,120	0,00412621	3,92E-02	Up in KO
Gimap5	-1,772	-0,034	0,00188281	2,44E-02	Down in KO
Gm40833	2,915	-1,437	0,00101829	1,69E-02	Up in KO
Gm3776	1,074	5,340	0,00230314	2,77E-02	Up in KO
Sp110	-1,227	2,871	0,00235354	2,80E-02	Down in KO
Oip5	1,281	1,705	0,00321797	3,39E-02	Up in KO
Wdr90	1,024	3,704	0,00252429	2,92E-02	Up in KO
Grap2	-1,677	1,741	0,00315604	3,35E-02	Down in KO
9430085M18Rik	1,742	-0,741	0,00106183	1,73E-02	Up in KO
Tnip3	1,509	3,314	0,00355821	3,61E-02	Up in KO
Ndr4	2,566	-1,564	0,00012006	5,09E-03	Up in KO
Iqgap3	1,378	4,605	0,00251105	2,92E-02	Up in KO
LOC108167332	-2,453	0,211	0,00289759	3,18E-02	Down in KO
Gemin6	1,400	2,178	0,00357961	3,62E-02	Up in KO
Ncaph	1,470	4,325	0,00365846	3,66E-02	Up in KO
LOC108167862	1,584	-0,503	0,00078735	1,47E-02	Up in KO
Gpr146	-1,695	-0,415	0,00066681	1,32E-02	Down in KO
Fanci	1,516	2,761	0,00367239	3,67E-02	Up in KO
Sipa1	-1,091	2,487	0,0020446	2,57E-02	Down in KO
Cd300lf	-1,836	-0,075	0,00041987	9,91E-03	Down in KO
Gm6093	-1,676	1,020	0,00288868	3,17E-02	Down in KO
Gm39942	1,034	5,138	0,00237745	2,81E-02	Up in KO
Sstr2	-1,387	0,890	0,00144519	2,07E-02	Down in KO
LOC108167999	2,534	-0,684	0,00369191	3,68E-02	Up in KO
LOC108169123	2,534	-0,684	0,00369191	3,68E-02	Up in KO
Ccr1	1,390	0,580	0,00163129	2,24E-02	Up in KO
Nek5	-1,496	-0,448	0,00053688	1,14E-02	Down in KO
Id4	1,281	1,923	0,00334691	3,48E-02	Up in KO
Was	-2,069	-0,215	0,00173257	2,32E-02	Down in KO

Angpt2	-2,404	-0,970	0,00100263	1,67E-02	Down in KO
Pax4	-2,430	-1,450	8,66E-05	4,26E-03	Down in KO
Cd8a	-1,874	2,607	0,0042327	3,98E-02	Down in KO
Nckap1l	-2,115	1,122	0,0032501	3,41E-02	Down in KO
Gbp6	-1,858	3,220	0,00482735	4,33E-02	Down in KO
Gm32796	1,590	-0,559	0,00065055	1,30E-02	Up in KO
Rgs17	-1,111	3,556	0,00269469	3,05E-02	Down in KO
Riad1	-1,033	0,555	0,00105587	1,72E-02	Down in KO
Eme1	1,465	1,955	0,00211991	2,62E-02	Up in KO
Klre1	-1,738	-0,268	0,00167751	2,27E-02	Down in KO
Gm1966	-1,264	2,846	0,00278761	3,12E-02	Down in KO
Ptgis	1,851	-0,004	0,00210069	2,61E-02	Up in KO
Gpx7	1,102	0,482	0,00211976	2,62E-02	Up in KO
Exo1	1,799	2,565	0,00415539	3,93E-02	Up in KO
Per3	-1,091	4,350	0,00357556	3,62E-02	Down in KO
Itgax	-2,161	0,540	0,00383771	3,77E-02	Down in KO
Mfge8	1,457	3,741	0,0044542	4,09E-02	Up in KO
LOC105242408	1,743	0,278	0,00209475	2,61E-02	Up in KO
Ltb4r1	1,093	0,217	0,00214952	2,65E-02	Up in KO
Slc2a12	2,463	-1,412	0,00021187	6,77E-03	Up in KO
Gm2808	-2,788	0,693	0,00537045	4,62E-02	Down in KO
Lysmd2	1,217	1,205	0,00381264	3,75E-02	Up in KO
Lrr1	1,716	0,780	0,00455688	4,15E-02	Up in KO
Cd3d	-1,583	1,185	0,00503479	4,43E-02	Down in KO
Wwtr1	1,003	0,526	0,00114733	1,80E-02	Up in KO
Rad51ap1	1,849	2,775	0,00582729	4,87E-02	Up in KO
Plin4	1,856	0,877	0,00300738	3,24E-02	Up in KO
Gm42245	1,409	-0,466	0,00050762	1,11E-02	Up in KO
St8sia4	-2,672	0,338	0,00276806	3,11E-02	Down in KO
Oprd1	-1,378	-0,455	0,00056625	1,19E-02	Down in KO
Ubd	-4,335	0,870	0,00403909	3,88E-02	Down in KO
Dock3	-1,357	0,106	0,00145582	2,08E-02	Down in KO
Nek2	1,149	4,690	0,00349497	3,57E-02	Up in KO
4930503L19Rik	-2,082	0,443	0,00293957	3,20E-02	Down in KO
Mfap3l	-1,078	5,471	0,00338426	3,50E-02	Down in KO
Lmcd1	1,695	-0,415	0,00117815	1,83E-02	Up in KO
Saxo1	2,712	-1,448	0,00116462	1,81E-02	Up in KO
Soga1	-1,160	3,060	0,00269528	3,05E-02	Down in KO
Rps6ka6	1,034	1,234	0,00355829	3,61E-02	Up in KO
Dpysl3	1,111	3,225	0,00336972	3,49E-02	Up in KO
Gm30762	1,169	0,722	0,00359169	3,62E-02	Up in KO
Psmb8	-1,300	5,397	0,00371619	3,69E-02	Down in KO
E330020D12Rik	1,204	0,074	0,00105118	1,72E-02	Up in KO
Gimap7	-3,027	0,262	0,00500873	4,42E-02	Down in KO
Plekho2	-1,013	1,108	0,00231228	2,77E-02	Down in KO
Tnfsf10	-1,328	3,679	0,00363779	3,65E-02	Down in KO
Ikzf1	-2,243	1,185	0,00488953	4,36E-02	Down in KO
Ric3	-2,181	-1,409	0,00040333	9,65E-03	Down in KO
Cdc45	1,408	3,267	0,0045869	4,17E-02	Up in KO
Atp8b2	-1,309	1,575	0,00288106	3,17E-02	Down in KO
Agt	-1,456	1,196	0,00351218	3,59E-02	Down in KO
Vwa7	1,066	0,698	0,00255446	2,94E-02	Up in KO

Nup62cl	4,365	-1,779	2,83E-05	2,21E-03	Up in KO
Usp2	-1,116	5,106	0,00287021	3,17E-02	Down in KO
B9d1	1,183	1,575	0,00526947	4,56E-02	Up in KO
Kcnab2	-2,111	-0,147	0,00324235	3,40E-02	Down in KO
Bmf	-1,142	2,794	0,00334108	3,47E-02	Down in KO
Ipcef1	-2,164	0,807	0,00309071	3,30E-02	Down in KO
Bub1b	1,066	4,631	0,00369597	3,68E-02	Up in KO
Aldh3b3	2,189	-0,872	0,00142285	2,06E-02	Up in KO
Adgrf1	1,175	3,234	0,00483835	4,33E-02	Up in KO
Gm11735	1,382	1,961	0,00458529	4,17E-02	Up in KO
Myo1g	-2,132	1,378	0,00602351	4,97E-02	Down in KO
Gm36767	-2,117	-1,152	0,00068655	1,35E-02	Down in KO
Ror1	-1,551	0,733	0,00357727	3,62E-02	Down in KO
Gm42286	1,940	-0,178	0,00230177	2,77E-02	Up in KO
Sh2d7	-2,057	-0,017	0,00503451	4,43E-02	Down in KO
Klra7	-2,334	-0,358	0,00324102	3,40E-02	Down in KO
Defb37	-1,309	1,594	0,00251184	2,92E-02	Down in KO
Spaca7	-2,494	-1,411	0,00068735	1,35E-02	Down in KO
Tnfrsf11b	1,377	-0,288	0,00109751	1,75E-02	Up in KO
Iffo1	-1,802	-0,704	0,00183729	2,40E-02	Down in KO
Penk	2,588	-1,228	0,00201792	2,55E-02	Up in KO
Myh10	1,114	0,295	0,0016474	2,25E-02	Up in KO
Gm40498	1,485	0,354	0,00294685	3,20E-02	Up in KO
Pola1	1,232	3,515	0,00464662	4,21E-02	Up in KO
Ndc80	1,433	3,177	0,00547814	4,67E-02	Up in KO
Myt1	-1,321	-0,603	0,00024689	7,33E-03	Down in KO
Snn	1,509	-0,433	0,00112736	1,78E-02	Up in KO
Peli3	-1,230	0,524	0,00137298	2,01E-02	Down in KO
Tmem86a	-1,109	0,642	0,0034141	3,52E-02	Down in KO
4930526L06Rik	1,211	0,123	0,00199884	2,54E-02	Up in KO
Ms4a6b	-2,380	-0,512	0,00308598	3,30E-02	Down in KO
Ptrf	1,025	2,353	0,00442206	4,08E-02	Up in KO
Cd244	-2,227	1,056	0,00559078	4,74E-02	Down in KO
Kcnk7	1,358	-0,007	0,0028547	3,16E-02	Up in KO
Gm15283	1,729	0,336	0,00421957	3,97E-02	Up in KO
Asb2	-2,103	-0,082	0,00356001	3,61E-02	Down in KO
Pex5l	-1,181	1,221	0,00481907	4,32E-02	Down in KO
Map1b	-1,570	-0,651	0,00083233	1,50E-02	Down in KO
Mfng	-2,280	-0,556	0,00110859	1,77E-02	Down in KO
Fcer1g	-1,781	0,807	0,00441485	4,08E-02	Down in KO
Krt12	1,357	2,596	0,00407128	3,89E-02	Up in KO
Adarb2	1,333	0,676	0,00352081	3,59E-02	Up in KO
Tmc3	1,421	-0,013	0,00426629	4,00E-02	Up in KO
Rgs13	-2,421	-0,411	0,00305771	3,28E-02	Down in KO
Smpdl3b	1,375	2,623	0,00586181	4,89E-02	Up in KO
Col18a1	1,292	1,220	0,00401089	3,87E-02	Up in KO
Syn2	-2,024	0,415	0,00513755	4,48E-02	Down in KO
Rbm11	1,624	0,705	0,00132814	1,97E-02	Up in KO
Cd68	1,131	1,631	0,00279074	3,12E-02	Up in KO
Hcst	-1,317	0,082	0,00166313	2,26E-02	Down in KO
Gm38875	-1,159	0,277	0,00246641	2,88E-02	Down in KO
LOC108169069	1,222	0,434	0,00292857	3,19E-02	Up in KO

Smoc1	-2,180	-0,908	0,00194557	2,50E-02	Down in KO
Gm35437	-1,235	0,259	0,00316927	3,36E-02	Down in KO
Slc22a17	-1,479	0,325	0,00353694	3,60E-02	Down in KO
Gm15401	1,106	4,813	0,00540418	4,64E-02	Up in KO
Snora70	-1,289	0,412	0,00254351	2,93E-02	Down in KO
Pnliprp2	1,305	0,829	0,00478028	4,29E-02	Up in KO
Zfp189	-1,109	0,828	0,00384782	3,78E-02	Down in KO
D16Ert472e	-1,519	1,742	0,00490032	4,36E-02	Down in KO
Drc1	1,094	0,572	0,00229024	2,76E-02	Up in KO
Gm42226	1,744	0,129	0,00536451	4,62E-02	Up in KO
Gimap1	-1,072	1,707	0,00578403	4,84E-02	Down in KO
Tbc1d16	-1,079	3,085	0,00494531	4,38E-02	Down in KO
Sgol1	1,149	3,108	0,00556653	4,73E-02	Up in KO
Myrip	-1,221	1,583	0,0044539	4,09E-02	Down in KO
Grhl3	1,620	0,592	0,00440938	4,08E-02	Up in KO
Retnlb	2,515	6,172	0,00355561	3,61E-02	Up in KO
Cps1	1,536	1,201	0,00548605	4,67E-02	Up in KO
Ltb	-1,515	-0,456	0,001354	1,99E-02	Down in KO
Gm14149	1,028	3,532	0,00456121	4,15E-02	Up in KO
Fam110b	-1,292	-0,685	0,00266992	3,04E-02	Down in KO
Pbp2	1,302	1,071	0,00501203	4,42E-02	Up in KO
Capn3	-1,474	-0,024	0,00237893	2,81E-02	Down in KO
Paqr3	1,081	0,143	0,00314244	3,34E-02	Up in KO
Gm15731	1,738	-1,090	0,00062081	1,27E-02	Up in KO
Gpc3	1,689	0,440	0,00492501	4,38E-02	Up in KO
Kif18b	1,029	3,150	0,00559595	4,74E-02	Up in KO
Adamts16	-2,561	-1,370	0,00196829	2,51E-02	Down in KO
Ccdc96	2,005	-1,362	0,00061828	1,26E-02	Up in KO
Olfm1	-1,375	-0,432	0,00258343	2,96E-02	Down in KO
Slc15a1	-1,054	4,518	0,00525143	4,56E-02	Down in KO
Fev	-2,338	-1,524	0,00203973	2,57E-02	Down in KO
Gm12865	1,384	-0,019	0,00388616	3,80E-02	Up in KO
Slc24a3	-1,676	0,333	0,00531138	4,59E-02	Down in KO
2210011K15Rik	1,256	-0,413	0,00111109	1,77E-02	Up in KO
Flrt2	-1,853	-0,400	0,00310965	3,32E-02	Down in KO
Plin5	1,110	-0,072	0,0023164	2,77E-02	Up in KO
Kif1a	-1,729	-0,557	0,00253279	2,93E-02	Down in KO
Pdgfd	-2,177	-0,910	0,00017575	6,28E-03	Down in KO
Slc38a3	1,440	0,297	0,00483105	4,33E-02	Up in KO
Spag5	1,187	4,244	0,00426185	4,00E-02	Up in KO
Hpca	-1,304	0,765	0,00439722	4,08E-02	Down in KO
Klra5	-2,241	-0,140	0,00503401	4,43E-02	Down in KO
Syp	-1,339	-0,205	0,00297213	3,22E-02	Down in KO
Shank3	1,394	-0,432	0,00244118	2,86E-02	Up in KO
Scn3a	-1,541	-0,557	0,00267406	3,04E-02	Down in KO
Ly9	-2,155	-1,122	0,0013158	1,96E-02	Down in KO
Gm20752	1,389	-0,248	0,00385688	3,78E-02	Up in KO
Slc2a6	-1,007	2,152	0,00513141	4,48E-02	Down in KO
Gm29891	-2,175	-0,670	0,00541829	4,64E-02	Down in KO
Npb	1,084	-0,946	0,00314403	3,34E-02	Up in KO
LOC108168988	1,029	0,718	0,00520613	4,53E-02	Up in KO
Slc11a2	1,310	6,113	0,0032312	3,40E-02	Up in KO

Cdh26	-1,934	-1,518	0,00036328	9,14E-03	Down in KO
Pcdhgc5	1,288	0,057	0,00436191	4,05E-02	Up in KO
Homer3	1,124	0,483	0,00595657	4,94E-02	Up in KO
Gm830	-1,784	-0,311	0,00453656	4,14E-02	Down in KO
C130083M11Rik	1,241	-0,284	0,00173645	2,32E-02	Up in KO
4933439K11Rik	-1,131	-0,079	0,00200667	2,54E-02	Down in KO
Tmem173	-1,340	1,124	0,00599396	4,96E-02	Down in KO
Gm41866	-1,379	-0,484	0,00187441	2,43E-02	Down in KO
Mrc1	1,104	0,569	0,00530918	4,59E-02	Up in KO
Fancb	1,361	1,576	0,00534088	4,61E-02	Up in KO
Ttll11	-1,021	0,304	0,00337062	3,49E-02	Down in KO
Zbtb20	-1,289	3,592	0,00251164	2,92E-02	Down in KO
Gm35039	3,206	-0,706	0,00585559	4,89E-02	Up in KO
Gm30806	1,040	0,407	0,00507868	4,45E-02	Up in KO
Clec2i	-2,342	-0,066	0,00580039	4,85E-02	Down in KO
Nudt11	-1,308	-0,643	0,00143302	2,06E-02	Down in KO
Gm9199	-2,035	-1,359	0,00181244	2,39E-02	Down in KO
Mlf1	1,926	-0,757	0,00460405	4,17E-02	Up in KO
Pstpip1	-1,133	0,829	0,00449647	4,12E-02	Down in KO
Adgrf5	1,075	-0,030	0,0024231	2,85E-02	Up in KO
Gpsm3	-1,734	-0,477	0,00495862	4,39E-02	Down in KO
Btbd11	1,122	0,509	0,00262985	3,00E-02	Up in KO
H2afy2	-1,732	-0,621	0,00374077	3,70E-02	Down in KO
Cd27	-2,062	-0,576	0,00414105	3,92E-02	Down in KO
Zfp773	-1,333	-0,646	0,00140651	2,04E-02	Down in KO
Azin2	1,441	0,224	0,00190949	2,46E-02	Up in KO
Pdpx	1,243	-0,182	0,00446192	4,10E-02	Up in KO
Hesx1	1,944	-1,268	0,00230163	2,77E-02	Up in KO
Chrm4	1,298	-0,614	0,00329729	3,44E-02	Up in KO
BC049715	1,427	0,605	0,00415333	3,93E-02	Up in KO
Calcb	4,076	-0,036	0,0017946	2,37E-02	Up in KO
Cd72	-1,674	-0,146	0,00575876	4,82E-02	Down in KO
St18	-1,676	-1,130	0,00125045	1,90E-02	Down in KO
Vmn2r98	1,161	-0,562	0,00218312	2,67E-02	Up in KO
Siglecfl	-1,707	-0,619	0,00508847	4,46E-02	Down in KO
Tspyl3	-1,205	-0,140	0,0049253	4,38E-02	Down in KO
Acsbg1	-1,913	-0,427	0,00544187	4,66E-02	Down in KO
Pfn2	1,014	-0,157	0,00284561	3,15E-02	Up in KO
LOC108167368	1,076	-0,238	0,00182094	2,39E-02	Up in KO
Gm31619	1,527	-0,610	0,0036502	3,66E-02	Up in KO
Gm35543	-1,534	-1,236	0,00094396	1,61E-02	Down in KO
Gm41662	1,087	-0,073	0,00491378	4,37E-02	Up in KO
Pcdhga6	-1,681	-0,882	0,00602192	4,97E-02	Down in KO
Aqp12	-1,528	-1,347	0,00048354	1,07E-02	Down in KO
Gm34389	-1,025	-0,078	0,00374758	3,70E-02	Down in KO
Tet1	-1,771	-0,698	0,00179335	2,37E-02	Down in KO
Faxc	1,423	-0,798	0,00420068	3,96E-02	Up in KO
Rbpms2	1,180	-0,294	0,0044922	4,12E-02	Up in KO
Gm32652	-1,423	-0,869	0,00402076	3,87E-02	Down in KO
Gm16984	-1,256	-0,625	0,00237043	2,81E-02	Down in KO
Klrb1	1,147	-0,125	0,00563657	4,76E-02	Up in KO
Gm8493	1,593	-1,055	0,00311186	3,32E-02	Up in KO

Fbxl16	-1,199	-0,649	0,00250215	2,92E-02	Down in KO
C9	-1,505	-0,784	0,00498674	4,41E-02	Down in KO
Aldh1a3	1,312	-0,548	0,00541827	4,64E-02	Up in KO
Gm30120	1,204	-0,237	0,00588685	4,90E-02	Up in KO
Mum1l1	1,549	-1,332	0,00118804	1,84E-02	Up in KO
Jam3	1,469	-0,966	0,00403924	3,88E-02	Up in KO
AY512931	1,376	-0,497	0,00497673	4,40E-02	Up in KO
Gm42235	1,050	-0,320	0,00521896	4,54E-02	Up in KO
Gm40417	1,591	-1,405	0,00176233	2,34E-02	Up in KO
Zdhhc19	1,023	-1,005	0,00318355	3,36E-02	Up in KO
Duoxa1	1,277	-0,731	0,00320935	3,38E-02	Up in KO
Rarres1	1,467	-1,144	0,00201318	2,55E-02	Up in KO
Kbtbd6	1,055	-0,390	0,00405499	3,88E-02	Up in KO
Fam105a	-1,583	-0,438	0,00308418	3,30E-02	Down in KO
Sall2	-1,257	-1,072	0,00084131	1,51E-02	Down in KO
Rbp1	1,364	-1,022	0,00386579	3,78E-02	Up in KO
Dennd1c	-1,620	0,336	0,00586437	4,89E-02	Down in KO
Ackr2	1,053	-0,641	0,00476788	4,29E-02	Up in KO
LOC108168298	-1,758	-1,714	0,00211104	2,62E-02	Down in KO
Gm30877	1,070	-0,574	0,00358161	3,62E-02	Up in KO
Gm40559	1,563	-1,155	0,00538343	4,63E-02	Up in KO
Gm30247	1,380	-0,963	0,00544597	4,66E-02	Up in KO
Adamts9	1,990	-1,214	0,00458122	4,17E-02	Up in KO
Rab3c	-1,390	-0,607	0,00234698	2,79E-02	Down in KO
Nckap5l	-1,171	-0,858	0,00489819	4,36E-02	Down in KO

Table 2 : Clinicopathological characteristics and correlations with the CCNA2-based classification.

MMR: Mismatch repair, MSI: Microsatellite instable, MSS: Microsatellite stable,
CMS: Consensus Molecular Subtypes, RFS : Relapse Free Survival

Characteristics		All	CCNA2 classes		p-value
			Non-high	High	
Patient's age					0,484
	≤50 years	228 (12%)	105 (11%)	123 (12%)	
	>50 years	1702 (88%)	830 (89%)	872 (88%)	
Patients' sex					0,417
	female	954 (47%)	453 (46%)	501 (48%)	
	male	1069 (53%)	528 (54%)	541 (52%)	
Tumor colon location					2,04E-02
	left	549 (32%)	253 (31%)	296 (34%)	
	right	777 (46%)	362 (44%)	415 (47%)	
	sigmoid	291 (17%)	164 (20%)	127 (14%)	
	transversum	77 (5%)	35 (4%)	42 (5%)	
Pathological stage					3,19E-08
	I-II	834 (51%)	347 (44%)	487 (58%)	
	III-IV	785 (49%)	435 (56%)	350 (42%)	
Pathological grade					5,99E-03
	1	26 (6%)	12 (5%)	14 (6%)	
	2	353 (77%)	190 (83%)	163 (71%)	
	3	77 (17%)	26 (11%)	51 (22%)	
MMR status					1,48E-05
	MSI	128 (19%)	40 (12%)	88 (25%)	
	MSS	563 (81%)	298 (88%)	265 (75%)	
CMS subtypes					1,00E-06
	CMS1	389 (20%)	80 (8%)	309 (31%)	
	CMS2	640 (32%)	298 (30%)	342 (34%)	
	CMS3	343 (17%)	162 (17%)	181 (18%)	
	CMS4	604 (31%)	439 (45%)	165 (17%)	
Follow-up median, months (min-max)		42 (1-212)	40 (1-212)	42 (1-203)	5,00E-03
Events, number (%)		496 (26%)	281 (31%)	215 (22%)	2,32E-06
5-Year RFS [95%CI]		70% [68-73]	65% [61-69]	75% [72-79]	8,86E-06

Table 3: Univariate and multivariate prognostic analyses for RFS.

MMR: Mismatch repair, MSI: Microsatellite instable, MSS: Microsatellite stable,
CMS: Consensus Molecular Subtypes, RFS: Relapse Free Survival, HR: Hazard Ratio

Variables		Univariate			Multivariate			Multivariate		
		N	HR [95CI]	p-value	N	HR [95CI]	p-value	N	HR [95CI]	p-value
Patient's age	>50 vs. ≤50 years	1630	1.02 [0.76-1.39]	0,882						
Patients' sex	male vs. female	1722	1.11 [0.92-1.34]	0,254						
Tumor colon location	right vs. left	1440	0.82 [0.67-1.01]	5,09E-08	489	0.97 [0.69-1.37]	0,880			
	sigmoid vs. left		0.33 [0.22-0.50]		489	0.37 [0.17-0.81]	1,28E-02			
	transversum vs. left		0.20 [0.07-0.53]		489	0.21 [0.03-1.53]	0,124			
Pathological stage	III-IV vs. I-II	1374	2.65 [2.08-3.38]	2,78E-15	489	2.43 [1.72-3.43]	5,03E-07			
Pathological grade	2 vs. 1	408	1.40 [0.51-3.84]	0,463						
	3 vs. 1		1.87 [0.61-5.71]							
MMR status	MSS vs. MSI	606	2.59 [1.55-4.32]	2,97E-04	489	2.65 [1.47-4.76]	1,18E-03			
CMS subtypes	CMS2 vs. CMS1	1657	0.81 [0.61-1.07]	1,44E-04				1700	0.73 [0.56-0.97]	2,90E-02
	CMS3 vs. CMS1		0.99 [0.73-1.36]					1700	0.95 [0.70-1.29]	0,743
	CMS4 vs. CMS1		1.38 [1.06-1.81]					1700	1.23 [0.93-1.62]	0,151
CCNA2 classes	high vs. non-high	1882	0.67 [0.56-0.8]	1,01E-05	489	0.61 [0.44-0.85]	2,99E-03	1700	0.70 [0.58-0.85]	4,13E-04

Table 4 : List of 92 genes differentially expressed between the CCNA2^{High} and CCNA2^{Non-high} classes

Gene Symbol	Entrez Gene ID	CCNA2 ^{Non-high} (log2, average)	CCNA2 ^{High} (log2, average)	CCNA2, high vs. non-high (log2-ratio)	p-value	q-value	Associated to	Associated pathways	Notes
CCNA2	890	9,46	10,57	1,11	4,94E-91	1,90E-87	up CCNA2 ^{High} and CCNA2 ^{Non-high}	TP53, BRCA1, CDKN1A, KRAS	The protein encoded by this gene belongs to the highly conserved cyclin family, whose members function as regulators of the cell cycle. This protein binds and activates cyclin-dependent kinase 2 and thus promotes transition through G1/S and G2/M. MAD2L1 is a component of the mitotic spindle assembly checkpoint that prevents the onset of anaphase until all chromosomes are properly aligned at the metaphase plate. MAD2L1 is related to the MAD2L2 gene located on chromosome 1.
MAD2L1	4085	9,16	10,23	1,06	3,82E-56	4,90E-53	up CCNA2 ^{High} and CCNA2 ^{Non-high}	BRCA1, TERT, MLH1, APC	TRAP1 is relevant in the control of key cell cycle regulators in tumor cells. TRAP1/TBP7 quality control of CDK1 and MAD2 contributes mechanistically to the regulation of mitotic entry and transit. In addition to its role in checkpoint signaling, MAD2 ensures chromosome stability through the regulation of AURORA B. catalytic activation of the spindle assembly checkpoint depends on regulated protein-protein interactions that accelerate the spontaneous but rate-limiting conversion of MAD2 required for mitotic checkpoint complex assembly
NEIL3	55247	5,82	6,82	1,01	7,76E-40	4,26E-37	up CCNA2 ^{High} and CCNA2 ^{Non-high}	TP53, EGFR, VEGF, BRAC1	NEIL3 belongs to a class of DNA glycosylases homologous to the bacterial Fpg/Nei family. These glycosylases initiate the first step in base excision repair by cleaving bases damaged by reactive oxygen species and introducing a DNA strand break via the associated lyase reaction NEIL3 Repairs Telomere Damage during S Phase to Secure Chromosome Segregation at Mitosis. NEIL3 protects genome stability through targeted repair of oxidative damage in telomeres during S/G2 phase.
DEPDC1	55635	7,63	8,72	1,09	7,07E-34	9,38E-32	up CCNA2 ^{High} and CCNA2 ^{Non-high}		DEPDC1 is a novel cell cycle related gene that regulates mitotic progression. DEPDC1 mRNA and protein expression levels were dramatically increased in prostate cancer tissues and cell lines. Overexpression of DEPDC1 promoted, but depletion of DEPDC1 inhibited cell proliferation by regulating the G1-S phase cell cycle transition. DEP domain containing 1 is a novel diagnostic marker and prognostic predictor for hepatocellular carcinoma.
PBK	55872	8,10	9,26	1,16	6,70E-31	4,60E-29	up CCNA2 ^{High} and CCNA2 ^{Non-high}	TGFB, TP53, CDKN1A, TERT	This gene encodes a serine/threonine protein kinase related to the dual specific mitogen-activated protein kinase kinase (MAPKK) family. Evidence suggests that mitotic phosphorylation is required for its catalytic activity. The encoded protein may be involved in the activation of lymphoid cells and support testicular functions, with a suggested role in the process of spermatogenesis. Overexpression of this gene has been implicated in tumorigenesis.
MND1	84057	6,25	7,28	1,03	9,33E-31	6,19E-29	up CCNA2 ^{High} and CCNA2 ^{Non-high}		The T-LAK Cell-originated Protein Kinase Signal Pathway Promotes Colorectal Cancer Metastasis. The product of the MND1 gene associates with HOP2 (MIM 608665) to form a stable heterodimeric complex that binds DNA and stimulates the recombinase activity of RAD51 (MIM 179617) and DMC1 (MIM 602721) (Chi et al., 2007 [PubMed 17639080]). Both the MND1 and HOP2 genes are indispensable for meiotic recombination Data suggest that the human TBPIP/Hop2-Mnd1 complex may ensure proper pairing between homologous chromosomes through its stimulation of strand exchange during meiosis.
MLF1IP	79682	8,29	9,29	1,00	5,86E-30	3,52E-28	up CCNA2 ^{High} and CCNA2 ^{Non-high}		The centromere is a specialized chromatin domain, present throughout the cell cycle, that acts as a platform on which the transient assembly of the kinetochore occurs during mitosis. All active centromeres are characterized by the presence of long arrays of nucleosomes in which CENPA (MIM 117139) replaces histone H3 (see MIM 601128). MLF1IP, or CENPU, is an additional factor required for centromere assembly MLF1IP is correlated with progression and prognosis in luminal breast cancer. MLF1IP significantly promotes prostate cancer cell proliferation and colony formation and significantly inhibits apoptosis without affecting cell cycle phase arrest in prostate cancer cell lines
ESCO2	157570	7,19	8,30	1,12	1,89E-27	8,17E-26	up CCNA2 ^{High} and CCNA2 ^{Non-high}	mitosis	This gene encodes a protein that may have acetyltransferase activity and may be required for the establishment of sister chromatid cohesion during the S phase of mitosis. ESCO2 has an S-phase specific role in the maintenance of genome stability The protein encoded by this gene is involved in cell motility. It is expressed in breast tissue and together with other proteins, it forms a complex with BRCA1 and BRCA2, thus is potentially associated with higher risk of breast cancer.
HMMR	3161	8,43	9,53	1,10	5,60E-27	2,27E-25	up CCNA2 ^{High} and CCNA2 ^{Non-high}	BRCA1	RHAMM expression identifies an aggressive subpopulation of tumor budding cells and is an independent adverse prognostic factor for colorectal cancer patients.
CHAC2	494143	5,73	6,75	1,02	3,05E-26	1,17E-24	up CCNA2 ^{High} and CCNA2 ^{Non-high}		The protein encoded by this gene is a gamma-glutamyl cyclotransferase that catalyzes the conversion of glutathione to 5-oxoproline and cysteinylglycine. It is thought that this gene is upregulated in response to endoplasmic reticulum stress and that the glutathione depletion enhances apoptosis Results showed for the first time that CHAC2 was degraded by the ubiquitin-proteasome pathway and CHAC2 expression inhibited tumor cell growth, proliferation, migration in vitro and in vivo. Mechanistic study showed that CHAC2 induced mitochondrial apoptosis and autophagy through unfolded protein response. CHAC2, downregulated in gastric and colorectal cancers, acted as a tumor suppressor inducing apoptosis and autophagy through unfolded protein response.
HSPA4L	22824	5,95	7,18	1,23	9,47E-14	6,52E-13	up CCNA2 ^{High} and CCNA2 ^{Non-high}	TP53, EGFR, VEGF, BRAC1	

PLA2G3	50487	2,85	4,04	1,19	3,44E-06	3,99E-06	up CCNA2 ^{High} and CCNA2 ^{Non-high}	TGFB	<p>This gene encodes a protein that belongs to the secreted phospholipase A2 family, whose members include the bee venom enzyme. The encoded enzyme functions in lipid metabolism and catalyzes the calcium-dependent hydrolysis of the sn-2 acyl bond of phospholipids to release arachidonic acid and lysophospholipids. This enzyme acts as a negative regulator of ciliogenesis, and may play a role in cancer development by stimulating tumor cell growth and angiogenesis.</p> <p>Phospholipase A2 Group III and Group X Have Opposing Associations with Prognosis in Colorectal Cancer.</p> <p>The protein encoded by this gene is primarily found in the nucleolus, where it can bind to the 5' UTR of the insulin-like growth factor II leader 3 mRNA and may repress translation of insulin-like growth factor II during late development. The encoded protein contains several KH domains, which are important in RNA binding and are known to be involved in RNA synthesis and metabolism.</p> <p>Our results strengthen the association of IMP3 with the regulation of epithelial to mesenchymal transition in breast cancers.</p> <p>Post-transcriptional regulation of cyclins D1, D3 and G1 and proliferation of human cancer cells depend on IMP-3 nuclear localization.</p> <p>Data show that insulin-like growth factor-II mRNA-binding protein 2 and 3 (IMP2/3)-miR-200a-progesterone receptor axis represents a double-negative feedback loop and serves as a new potential therapeutic target for the treatment of Triple-negative breast cancer (TNBC).</p> <p>The membrane-associated protein encoded by this gene is a member of the superfamily of ATP-binding cassette (ABC) transporters. ABC proteins transport various molecules across extra- and intracellular membranes. ABC genes are divided into seven distinct subfamilies (ABC1, MDR/TAP, MRP, ALD, OABP, GCN20, and White). This encoded protein is a member of the ABC1 subfamily, which is the only major ABC subfamily found exclusively in multicellular eukaryotes.</p>
IGF2BP3	10643	4,78	5,87	1,10	4,96E-06	5,52E-06	up CCNA2 ^{High} and CCNA2 ^{Non-high}	TP53, BRCA1, Myc	<p>This gene encodes a member of the ZIC family of C2H2-type zinc finger proteins. The encoded protein may act as a transcriptional repressor. Studies in mouse and Xenopus support a role for this gene in neural crest development. Elevated expression of this gene has been observed in various human cancers and may contribute to cancer progression.</p> <p>Identification of zinc finger protein of the cerebellum 5 as a survival factor of prostate and colorectal cancer cells.</p> <p>ZIC5 is highly upregulated in non-small cell lung cancer tumor tissues and may act as an oncogene by influencing CCNB1 and CDK1 complex expression.</p> <p>basolateral surface of epithelial cells</p> <p>COLORECTAL Polymeric Immunoglobulin Receptor Expression is Correlated with Hepatic Metastasis and Poor Prognosis in Colon Carcinoma Patients with Hepatic Metastasis.</p> <p>This gene encodes a member of the vasopressin/oxytocin subfamily of G protein-coupled receptors. The encoded membrane protein acts as a receptor for neuropeptide S and affects a variety of cellular processes through its signaling. Increased expression of this gene in ciliated cells of the respiratory epithelium and in bronchial smooth muscle cells is associated with asthma.</p>
ABCA12	26154	2,23	3,28	1,04	2,16E-05	2,05E-05	up CCNA2 ^{High} and CCNA2 ^{Non-high}	VEGFA	
ZIC5	85416	2,72	3,82	1,10	1,16E-04	9,21E-05	up CCNA2 ^{High} and CCNA2 ^{Non-high}	CDK1, CCNB1	
PIGR	5284	13,83	14,94	1,11	1,80E-04	1,37E-04	up CCNA2 ^{High} and CCNA2 ^{Non-high}	TGFb, Cdk6	
NPSR1	387129	2,50	3,62	1,12	2,38E-04	1,76E-04	up CCNA2 ^{High} and CCNA2 ^{Non-high}		
REG4	83998	9,87	11,19	1,32	2,39E-04	1,77E-04	up CCNA2 ^{High} and CCNA2 ^{Non-high}		
CALB1	793	2,16	3,18	1,03	3,50E-04	2,49E-04	up CCNA2 ^{High} and CCNA2 ^{Non-high}		<p>The protein encoded by this gene is a member of the calcium-binding protein superfamily that includes calmodulin and troponin C. Originally described as a 27 kDa protein, it is now known to be a 28 kDa protein. It contains four active calcium-binding domains, and has two modified domains that are thought to have lost their calcium binding capability. This protein is thought to buffer entry of calcium upon stimulation of glutamate receptors.</p>
REG1A	5967	8,26	9,63	1,37	4,95E-04	3,39E-04	up CCNA2 ^{High} and CCNA2 ^{Non-high}		
CYP2W1	54905	8,10	7,09	-1,01	5,27E-04	3,59E-04	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
GNB4	2786	8,55	7,51	-1,05	1,20E-04	9,46E-05	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
SFRP2	6423	8,79	7,69	-1,09	7,63E-05	6,31E-05	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
LY6G6D	58530	6,93	5,48	-1,45	2,98E-05	2,73E-05	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
CEL	1056	8,56	7,31	-1,25	2,80E-05	2,58E-05	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
KRT23	25984	9,03	7,79	-1,24	1,92E-05	1,84E-05	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
NPTX2	4885	7,13	6,10	-1,03	1,21E-05	1,21E-05	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
NTRK2	4915	6,16	5,15	-1,01	8,35E-06	8,79E-06	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
TNNC2	7125	6,55	5,48	-1,07	2,27E-06	2,77E-06	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
PRSS33	260429	5,53	4,44	-1,09	2,27E-06	2,77E-06	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
CST1	1469	8,71	7,64	-1,07	1,70E-06	2,14E-06	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
F7	2155	4,00	2,91	-1,09	5,46E-07	7,65E-07	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
MUC12	10071	10,57	9,41	-1,16	3,74E-07	5,49E-07	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
OGN	4969	4,27	3,10	-1,17	2,79E-07	4,22E-07	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
DES	1674	9,78	8,47	-1,32	2,44E-07	3,75E-07	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
CHRD12	25884	6,04	4,76	-1,28	2,18E-07	3,38E-07	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
SLC1A7	6512	6,03	4,87	-1,16	1,94E-07	3,05E-07	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
C7	730	6,30	5,22	-1,08	1,63E-07	2,61E-07	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
RHCG	51458	4,73	3,64	-1,08	5,37E-08	9,62E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
PALM3	342979	5,52	4,49	-1,03	2,57E-08	5,02E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
CASQ2	845	3,43	2,25	-1,18	1,21E-08	2,59E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
ITGBL1	9358	6,75	5,64	-1,11	9,64E-09	2,10E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}		
HSPB6	126393	7,27	6,20	-1,07	9,48E-09	2,08E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}		

C6orf15	29113	3,90	2,48	-1,42	8,61E-09	1,91E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}
LMO3	55885	4,51	3,43	-1,08	6,44E-09	1,47E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}
PKP1	5317	7,30	6,22	-1,08	6,18E-09	1,42E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}
DRD2	1813	4,96	3,55	-1,41	4,78E-09	1,13E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}
SLC7A4	6545	6,37	5,29	-1,08	4,35E-09	1,03E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}
L1CAM	3897	6,66	5,39	-1,27	4,26E-09	1,02E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}
CHRD1	91851	6,12	4,80	-1,32	4,18E-09	1,00E-08	down CCNA2 ^{High} and CCNA2 ^{Non-high}
LONRF2	164832	2,74	1,71	-1,03	3,14E-09	7,79E-09	down CCNA2 ^{High} and CCNA2 ^{Non-high}
ADCY5	111	4,99	3,97	-1,02	2,91E-09	7,29E-09	down CCNA2 ^{High} and CCNA2 ^{Non-high}
HAND2	9464	4,68	3,55	-1,13	1,99E-09	5,15E-09	down CCNA2 ^{High} and CCNA2 ^{Non-high}
SYNM	23336	8,15	7,12	-1,03	1,24E-09	3,36E-09	down CCNA2 ^{High} and CCNA2 ^{Non-high}
PLXNB3	5365	5,59	4,45	-1,14	7,56E-10	2,16E-09	down CCNA2 ^{High} and CCNA2 ^{Non-high}
THBS4	7060	5,60	3,95	-1,65	6,96E-10	2,01E-09	down CCNA2 ^{High} and CCNA2 ^{Non-high}
CRTAC1	55118	2,67	1,67	-1,00	6,72E-10	1,94E-09	down CCNA2 ^{High} and CCNA2 ^{Non-high}
CILP	8483	5,46	4,13	-1,33	4,88E-10	1,46E-09	down CCNA2 ^{High} and CCNA2 ^{Non-high}
LOC100127888	100127888	6,76	5,61	-1,15	2,51E-10	7,94E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
APOD	347	7,58	6,42	-1,16	2,46E-10	7,82E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
SLIT2	9353	5,29	4,08	-1,21	1,49E-10	4,97E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
ADAM33	80332	5,36	4,35	-1,01	1,20E-10	4,08E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
SFRP4	6424	9,04	7,57	-1,47	9,83E-11	3,41E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
C5orf23	79614	4,29	3,12	-1,17	8,32E-11	2,93E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
MYH11	4629	11,21	9,96	-1,24	5,47E-11	2,02E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
GAP43	2596	3,16	2,10	-1,06	3,27E-11	1,26E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
NGFR	4804	5,26	4,16	-1,10	2,80E-11	1,10E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
PSD	5662	5,64	4,59	-1,05	2,71E-11	1,07E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
GPR133	283383	5,49	4,48	-1,01	2,66E-11	1,06E-10	down CCNA2 ^{High} and CCNA2 ^{Non-high}
PTGIS	5740	6,98	5,76	-1,21	1,63E-11	6,73E-11	down CCNA2 ^{High} and CCNA2 ^{Non-high}
CSDC2	27254	4,64	3,56	-1,08	1,39E-11	5,82E-11	down CCNA2 ^{High} and CCNA2 ^{Non-high}
ATP1A2	477	3,87	2,58	-1,29	1,21E-11	5,19E-11	down CCNA2 ^{High} and CCNA2 ^{Non-high}
NALCN	259232	3,09	2,00	-1,08	9,93E-12	4,33E-11	down CCNA2 ^{High} and CCNA2 ^{Non-high}
RGMA	56963	6,89	5,82	-1,07	7,70E-12	3,44E-11	down CCNA2 ^{High} and CCNA2 ^{Non-high}
PLIN4	729359	5,65	4,16	-1,49	4,34E-12	2,06E-11	down CCNA2 ^{High} and CCNA2 ^{Non-high}
SGCA	6442	4,02	2,84	-1,18	2,66E-12	1,31E-11	down CCNA2 ^{High} and CCNA2 ^{Non-high}
TNXB	7148	7,77	6,68	-1,09	2,33E-12	1,16E-11	down CCNA2 ^{High} and CCNA2 ^{Non-high}
CAPS	828	8,03	7,02	-1,01	1,67E-12	8,58E-12	down CCNA2 ^{High} and CCNA2 ^{Non-high}
COMP (= THSB5)	1311	7,59	5,73	-1,86	1,17E-12	6,27E-12	down CCNA2 ^{High} and CCNA2 ^{Non-high}
HSPB7	27129	6,08	4,82	-1,26	1,16E-12	6,26E-12	down CCNA2 ^{High} and CCNA2 ^{Non-high}
VIPR2	7434	4,14	3,08	-1,05	1,06E-12	5,79E-12	down CCNA2 ^{High} and CCNA2 ^{Non-high}
C1orf175	374977	5,79	4,74	-1,05	9,91E-13	5,45E-12	down CCNA2 ^{High} and CCNA2 ^{Non-high}
LRRN2	10446	5,69	4,62	-1,07	4,33E-13	2,57E-12	down CCNA2 ^{High} and CCNA2 ^{Non-high}
SCUBE2	57758	6,06	5,06	-1,00	3,52E-13	2,13E-12	down CCNA2 ^{High} and CCNA2 ^{Non-high}
ELN	2006	9,83	8,81	-1,02	1,51E-13	9,91E-13	down CCNA2 ^{High} and CCNA2 ^{Non-high}
IGFN1	91156	3,13	1,86	-1,28	8,81E-14	6,12E-13	down CCNA2 ^{High} and CCNA2 ^{Non-high}
SPEG	10290	6,13	4,79	-1,33	6,97E-14	4,94E-13	down CCNA2 ^{High} and CCNA2 ^{Non-high}
PDZD4	57595	4,21	3,20	-1,01	2,21E-14	1,69E-13	down CCNA2 ^{High} and CCNA2 ^{Non-high}
ISLR2	57611	4,83	3,74	-1,09	4,48E-14	3,89E-14	down CCNA2 ^{High} and CCNA2 ^{Non-high}
PALM	5064	6,90	5,89	-1,01	4,09E-15	3,58E-14	down CCNA2 ^{High} and CCNA2 ^{Non-high}
GGT7	2686	9,14	8,06	-1,08	1,20E-15	1,14E-14	down CCNA2 ^{High} and CCNA2 ^{Non-high}
PRELP	5549	7,74	6,23	-1,51	7,20E-16	7,12E-15	down CCNA2 ^{High} and CCNA2 ^{Non-high}