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MCR RapidIMPACT

Molecular Cancer Research

Mutational Landscape of the Essential Autophagy Gene *BECN1* in Human Cancers

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Abstract

Evidence suggests that the catabolic process of macroautophagy (autophagy hereafter) can either suppress or promote cancer. The essential autophagy gene *ATG6/BECN1* encoding the Beclin1 protein has been implicated as a haploinsufficient tumor suppressor in breast, ovarian, and prostate cancers. The proximity of *BECN1* to the known breast and ovarian tumor suppressor breast cancer 1, early onset, *BRCA1*, on chromosome 17q21, has made this determination equivocal. Here, the mutational status of *BECN1* was assessed in human tumor sequencing data from The Cancer Genome Atlas (TCGA) and other databases. Large deletions encompassing both *BRCA1* and *BECN1*, and deletions of only *BRCA1* but not *BECN1*, were found in breast and ovarian cancers, consistent with *BRCA1* loss being a primary driver mutation in these cancers. Furthermore, there was no evidence for *BECN1* mutation or loss in any other cancer, casting doubt on whether *BECN1* is a tumor suppressor in most human cancers.

Implications: Contrary to previous reports, *BECN1* is not significantly mutated in human cancer and not a tumor-suppressor gene, as originally thought.

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Introduction

Autophagy captures and degrades intracellular proteins and organelles in lysosomes to preserve protein and organelle quality and to recycle building blocks to sustain metabolism and survival in starvation (1, 2). Autophagy promotes the health, function, and survival of cells and tissues, and generally, the loss of autophagy is destructive. In mammals, autophagy deficiency is linked to tissue degeneration, chronic inflammation, susceptibility to metabolic stress, and premature lethality.

There is evidence that autophagy both promotes and suppresses cancer, however, this has not been rigorously addressed in humans (3). Monoallelic disruption of *BECN1* on chromosome 17q21 has been reported in 40% to 75% of human breast, ovarian, and prostate tumors, suggesting that autophagy is a tumor-suppression mechanism (4–6). *BECN1* allelic loss was also found in 9 out of 22 breast cancer cell lines by FISH analysis, although no coding or splice-site mutations were found (4). The small sample sizes and poorly matched comparisons of cell lines and normal tissues and the modest frequencies of loss of heterozygosity used for these investigations is, however, insufficient to support the claim that *BECN1* is a haploinsufficient tumor suppressor.

BECN1 is located on chromosome 17q21 next to *BRCA1*, a known tumor-suppressor gene and whose loss is a driver of breast and ovarian cancer. *BRCA1* is a critical regulator of DNA repair by homologous recombination and its loss causes DNA-repair defects and cancer predisposition (7). The close proximity of *BECN1* and *BRCA1* complicates the determination whether allelic loss of *BECN1* is a driver or passenger mutation in breast and ovarian cancers. Furthermore, the mutational status of *BECN1* in other cancers has not been rigorously assessed.

In support of the concept that autophagy is a tumor-suppression mechanism and that allelic loss of *BECN1* promotes cancer, *Beclin1*$^{+/-}$ mice are prone to mammary hyperplasia, liver and lung carcinomas, and lymphomas (8, 9). However, mosaic whole-body knockout of the essential autophagy gene *Atg5*, or liver-specific knock out of the essential autophagy gene *Atg7*, produces only benign liver hepatomas and no other neoplasms (10). Thus, autophagy defects promote development of benign liver tumors in mice but may also block their progression. Autophagy-

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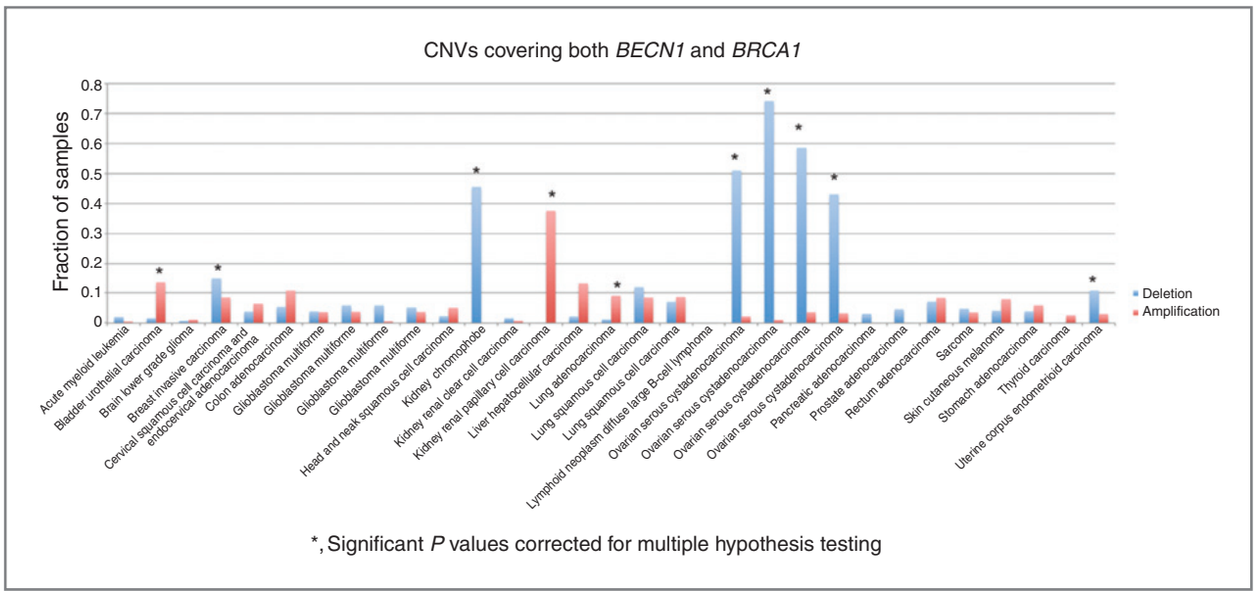

Figure 1. CNVs covering both *BRCA1* and *BECN1*.

independent functions of *Becn1* may also contribute to the suppression of non-liver neoplasms or these events may be limited to genetically manipulated mice and not relevant to human cancers. The vast majority of germline mutations in *BRCA1* are loss-of-function mutations (frameshift, indels, nonsense, or missense mutations), or focal deletions, not gross deletions in the *BRCA1* locus at 17q21 that extend to encompass *BECN1*. Thus *Beclin1*$^{+/-}$ mice do not reflect a human condition.

In contrast, autophagy promotes the survival of tumor cells in hypoxic tumor regions (11) as well as the growth, survival, and malignancy of RAS- and BRAF-driven cancers (3, 12–15). Autophagy promotes tumorigenesis by suppressing p53 activation and maintaining mitochondrial function essential for cellular metabolism and survival (16). Without autophagy, tumors accumulate defective mitochondria, have growth and metabolic defects, and progresses to a more benign fate. This is consistent with a large body of literature indicating that autophagy is required for survival in starvation and stress, functions that are conserved from yeast to mammals that are also important for growth of cancer (2, 3, 17).

Germline mutations in *BRCA1*, *BRCA2*, and *PALB2* predispose to hereditary breast cancer and the three proteins function together to maintain genome stability by promoting faithful repair of double-strand breaks by homologous recombination (18). Mammary epithelial cell–specific knockout of *Palb2* causes mammary tumorigenesis with long latency that is suppressed by allelic loss of *Becn1*, suggesting that autophagy is tumor promoting (19). Deletion of *Trp53* abrogates tumorigenesis impairment upon allelic loss of *Becn1* in *Palb2*-deficient mammary tumors, thus the combination of autophagy defect and loss of a critical DNA-repair mechanism augments the p53 antitumor response (19). Because loss of both Palb2 and autophagy promote DNA damage and p53 activation (18, 20, 21), this explains enhanced p53 activity and why autophagy suppresses the p53 response and mammary tumorigenesis.

The important unanswered question here is whether mutations in essential autophagy genes are found in human cancers using current genomic information, and if they are found, are they loss- or gain-of-function mutations? Note that recent assessment of oncogenes and tumor-suppressor genes assembled from the current human tumor sequencing data does not include any autophagy genes (22), but this was not examined specifically. To begin to resolve the potential conflicting role of autophagy in human cancer, we examined the publicly available human tumor sequencing and gene expression databases (including The Cancer Genome Atlas, TCGA) to determine the mutational and expression status of *BECN1* in a broad array of human cancers. We first assessed *BECN1* for single-nucleotide variations (SNV) and copy-number variations (CNV) in human breast, ovarian, and prostate cancer genome sequences. Because *BECN1* is adjacent to *BRCA1*, we specifically looked for deletions of *BECN1* that do not encompass *BRCA1*. We found enrichment for truncating mutations of *BRCA1*, deletion of the chromosomal region that included *BRCA1* only, and deletions affecting both *BRCA1* and *BECN1*, but not truncating mutations of *BECN1* or deletions of only *BECN1*. Analysis of all other cancers that lack *BRCA1* deletion indicated no significant recurrence of SNVs or CNVs in *BECN1*. Thus, *BECN1* is not mutated or specifically deleted in human cancer, indicating that it is not a tumor-suppressor gene.

Materials and Methods

CNVs

To study the copy-number status of *BECN1* and *BRCA1* in different cancers, we downloaded more than 10,000 processed copy-number data from the TCGA portal

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(https://tcga-data.nci.nih.gov/tcga/). The TCGA Consortium collected tumor and matched normal samples from 24 different cancers from which to perform single-nucleotide polymorphism (SNP) and comparitive genomic hybridization (CGH) microarray analysis on genomic DNA to find CNVs are listed in Supplementary Table S1. The cancers for which we obtained CNV data include acute myeloid leukemia, bladder urothelial carcinoma, brain lower grade glioma, breast invasive carcinoma, cervical squamous cell carcinoma and endocervical adenocarcinoma, colon adenocarcinoma, glioblastoma multiforme, head and neck squamous cell carcinoma, kidney chromophobe, kidney renal clear cell carcinoma, kidney renal papillary cell carcinoma, liver hepatocellular carcinoma, lung adenocarcinoma, lung squamous cell carcinoma, lymphoid neoplasm diffuse large B-cell lymphoma, ovarian serous cyst adenocarcinoma, pancreatic adenocarcinoma, prostate adenocarcinoma, rectum adenocarcinoma, sarcoma, skin cutaneous melanoma, stomach adenocarcinoma, thyroid carcinoma, and uterine corpus endometrioid carcinoma. Tumor samples represent primary as well as metastatic tumors.

The TCGA consortium performed CNV calling and provided level 3 data for CNVs including segment mean values and number of markers for all CNVs detected. Briefly, the segment mean is the average of the $\log_2$ ratio of probes in the segment [$\log_2$ (observed intensity/reference intensity)] and represents the extent of copy-number changes for that particular genomic segment, whereas the number of markers is the number of probes present in that segmental region. To extract a set of high confidence CNVs, we used a threshold of 0.2 in segment mean value for amplifications and −0.2 for deletions. We derived these thresholds by examining the distribution of segment mean values from tumor and normal samples. In addition, we require the number of markers spanning a CNV to be at least 10 to decrease false positives in calling CNVs. We tested the sensitivity of our method by finding the previously reported CNVs (amplifications in *PIK3CA*, *EGFR*, *FOXA1*, and *HER2; deletions in MLL3*, *PTEN*, *RB1*, and *MAP2K4*) in breast invasive cancer (23). We used the CNVs that pass these criteria for further analysis and identify all CNVs that overlap *BECN1* or *BRCA1*.

We used the matched tumor and normal samples to determine the somatic CNVs. We identify CNVs as germline in the tumor if there was an overlapping CNV in the matched normal. The ratio of deletions to amplifications of somatic CNVs found across the genome provide a background ratio for comparison with the ratio found at a particular locus. If the ratio of deletion to amplifications is different at a locus than the genome average, then there may be selection for deletions or amplifications at that locus. We used the two-tailed Fisher exact test for determining statistical significance using the average number of deletions and duplications per sample for the background and the number of samples with deletion and amplifications for the locus.

Somatic mutations

The TCGA provides somatic mutations detected from whole genome and whole-exome sequencing from matched tumor and normal samples as level 2 data (Supplementary Table S2). We extracted the somatic mutations for *BECN1* and *BRCA1* and indicated their type as missense, nonsense, silent, splice site, and insertion or deletion resulting in frame shift or in-frame (Supplementary Tables S3 and S4).

Gene expression

TCGA RNA-seq level 3 data for all cancers, tumor and normal, were processed and normalized and we used the RSEM normalized values for gene expression. Fold change in *BECN1* gene expression between tumor and normal tissue was calculated using median expression of tumors and normal. Significance of differential gene expression change in *BECN1* is calculated using a two-tailed Wilcoxon test and Bonferroni corrected for multiple hypothesis testing.

Results

CNVs in *BECN1*

CNVs in *BECN1* were assessed in the databases indicated in Table S1 from approximately 10,000 normal/tumor pairs. CNVs were classified into three groups defined by whether the CNV overlapped with *BECN1* but not *BRCA1*, overlapped with *BRCA1* but not *BECN1*, or overlapped with both *BECN1* and *BRCA1* (Table 1). Most of the CNVs detected are large and overlap both *BECN1* and *BRCA1*. Each CNV was further identified as a deletion, amplification, or interrupting amplification if only a part of a gene was amplified (not included in the count for amplifications). As expected, breast and ovarian tumors were significantly enriched for having deletions in the locus containing both *BECN1* and *BRCA1* (Fig. 1 and Table 1).

Other tumor types that exhibited significant enrichment for deletions in both *BECN1* and *BRCA1* include kidney chromophobe and uterine corpus endometrioid carcinoma (Fig. 1 and Table 1). Tumor types found having enrichment for amplifications include bladder urothelial carcinoma, kidney renal papillary cell carcinoma, and lung adenocarcinoma. Closer examination found that the CNVs in kidney chromophobe and kidney renal papillary cell carcinoma are whole-chromosome deletions and amplifications, respectively, which are consistent with known loss and gain of chromosome 17 for these two types of tumors (24).

CNVs that overlap *BRCA1* but not *BECN1* were enriched for deletions in breast and ovarian tumors, whereas CNVs that overlap *BECN1* but not *BRCA1* were not enriched for deletions in any tumor (Table 1). These results are consistent with the loss of *BRCA1* being the driver mutation in breast and ovarian tumors. No significant CNVs in *BECN1* were detected in any other cancers (Table 1). Loss of chromosome 17q21 and *BRCA1* has been reported in prostate cancer only very infrequently (0.45%; ref. 25). For prostate adenocarcinoma, we found 9 deletions (covering both *BECN1* and *BRCA1*) and no amplifications (Table 1). The *P* value for enrichment of deletions is 0.024, however, after correcting

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Table 1. Enrichment for deletions and amplifications for *BECN1* and *BRCA1* in cancers

Cancer	Number of tumor/normal pairs	Average CNA per tumor *BECN1* alone		*BRCA1* alone					Both *BECN1* and *BRCA1*		
		Deletion	Amplification	Deletion	Amplification	*P* value	Deletion	Amplification	*P* value	Deletion	Amplification	*P* value
Acute myeloid leukemia	196	146	146	1	1		0	0		4	1	
Bladder urothelial carcinoma	184	99	106	0	0		0	0		3	25	0.014415
Brain lower grade glioma	269	34	32	1	0		0	0		2	3	
Breast invasive carcinoma	999	85	111	3	7		24	4	2.42E−03	149	86	4.00E−03
Cervical squamous cell carcinoma and endocervical adenocarcinoma	155	52	61	1	0		2	0		6	10	
Colon adenocarcinoma	460	70	71	3	2		7	1		25	50	
Glioblastoma multiforme 1	434	29	25	1	0		1	1		17	16	
Glioblastoma multiforme 2	237	30	27	2	1		1	2		14	9	
Glioblastoma multiforme 3	170	35	29	0	0		1	0		10	1	
Glioblastoma multiforme 4	534	106	109	0	1		3	1		28	20	
Head and neck squamous cell carcinoma	390	49	55	0	0		1	1		9	20	
Kidney chromophobe	66	97	75	1	0		1	0		30	0	0.0002139
Kidney renal clear cell carcinoma	550	35	44	0	1		2	0		9	4	
Kidney renal papillary cell carcinoma	168	30	49	0	0		0	0		0	63	8.36E−07
Liver hepatocellular carcinoma	136	52	88	0	2		1	2		3	18	
Lung adenocarcinoma	505	50	66	0	2		0	2		6	46	0.0069843
Lung squamous cell carcinoma 1	117	51	50	1	0		0	0		14	10	
Lung squamous cell carcinoma 2	505	100	109	2	3		7	0		36	44	
Lymphoid neoplasm diffuse large B-cell lymphoma	18	49	48	0	0		0	0		0	0	
Ovarian serous cystadenocarcinoma 1	587	105	109	7	2		10	3		299	13	2.03E−35
Ovarian serous cystadenocarcinoma 2	97	76	92	2	0		3	0		72	1	1.17E−13
Ovarian serous cystadenocarcinoma 3	355	93	121	2	1		3	2		208	13	4.05E−31
Ovarian serous cystadenocarcinoma 4	586	253	363	16	4		47	3	2.42E−12	252	19	8.09E−47
Pancreatic adenocarcinoma	66	29	30	0	0		0	0		2	0	
Prostate adenocarcinoma	195	46	29	1	2		1	0		9	0	
Rectum adenocarcinoma	167	81	83	0	1		0	0		12	14	
Sarcoma	84	71	122	0	1		0	1		4	3	
Skin cutaneous melanoma	338	54	78	0	0		2	2		14	27	
Stomach adenocarcinoma	306	44	59	1	1		6	2		12	18	
Thyroid carcinoma	505	15	18	0	0		1	0		0	13	
Uterine corpus endometrioid carcinoma	505	62	91	3	3		12	2		55	15	1.15E−05

NOTE: Only statistically significant enrichments have their Bonferroni corrected *P* values shown. CNA, copy number alteration.

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for multiple hypothesis testing, it is not significant. Prostate adenocarcinoma is a heterogeneous disease and the fraction of this disease where loss of 17q21 is a driver mutation is small compared with breast or ovarian cancer. It is clear, however, that in contrast with previous reports, *BECN1* deletions do not significantly occur in the absence of *BRCA1* deletion.

Somatic mutations

There are 169 and 32 (ratio of 5.28) mutations found in *BRCA1* and *BECN1*, respectively, across all tumor samples (6,632) and the numbers are 137 and 31 (ratio of 4.42) if we exclude breast and ovarian tumors in which *BRCA1* is known to be a tumor suppressor (Supplementary Table S2). The difference in mutation number is mostly explained by the size of the coding region of the two genes (ratio of protein coding length of *BRCA1* to *BECN1* is 4.14).

None of the mutations found in *BECN1* were nonsense or splice-site mutations (Supplementary Table S3) with the potential to alter function and that are frequently found tumor-suppressor mutations. If we restrict analysis to breast and ovarian cancer, there is only one mutation found in *BECN1* and it is a missense mutation in an ovarian tumor. In contrast, there are 32 mutations in *BRCA1* of which 23 are nonsense, splice site, or frameshift mutations, all of which lead to truncation of *BRCA1* (Supplementary Table S4).

Across all cancer data from TCGA, there are 30 missense, 0 nonsense, 0 splice site, and 11 silent mutations for *BECN1* and 135 missense, 20 nonsense, 12 splice site, and 39 silent mutations for *BRCA1*. To find statistical enrichment of missense, nonsense, or splice-site mutations compared with silent mutations, we use as null model the aggregate of mutations across all samples in breast cancer (778 tumors) yielding 31,861 missense, 2,339 nonsense, 1,075 splice site, and 11,677 silent mutations. Because the vast majority of mutations detected in tumors are passenger mutations with little or no selective advantage to the tumors, the ratio of missense to silent mutations (2.73), nonsense to silent mutations (0.20), and splice site to silent mutations (0.09) are good approximations for little or no selection of missense, nonsense, or splice site over silent mutations. Indeed, these ratios are very similar when looking at other cancer types from TCGA. There is statistically significant increase in ratio of nonsense to silent and splice site to silent mutations for *BRCA1* (2.6 and 3.4 fold increase with P value of 0.0008 and 0.0003 using two-tailed χ^2 test with Yate's correction). There is no significant increase in missense over silent mutations for *BRCA1* and *BECN1*, and no increase in nonsense and splice site over silent mutations in *BECN1*. The proportion of missense, nonsense, and splice-site mutations for *BECN1* is statistically consistent with the occurrence of passenger mutations.

Gene-expression changes

The differential expression of *BECN1* between tumor and normal tissue for 17 cancer types from TCGA show no significant fold-change greater than 2 (Supplementary Table S5). The greatest decrease in expression of *BECN1* occurs in kidney chromophobe in which the fold-change of tumor to normal is 0.65, which is consistent with loss of chromosome 17 being common in this cancer.

Discussion

Using the genomic information collected on a broad array of human cancers, we assessed the mutational status of the essential autophagy gene *BECN1*. Despite reports indicating allelic loss of *BECN1* in some human cancers, this seems to be explained solely by the proximity of *BECN1* to *BRCA1*. We find no evidence of mutation or focal loss of *BECN1* from the analysis of currently available cancer genomic information. Monoallelic loss of the chromosome 17q21 region that encompasses both *BECN1* and *BRCA1* is found in both breast and ovarian cancer. However, as the region is large, this finding does not support a role for *BECN1* as the driver. Furthermore, there is no finding of nonsense or splice-site mutations in *BECN1* in any other cancers.

Germline missense mutations in *BRCA1* followed by somatic deletion of the remaining allele in tumors are responsible for inherited cancers. In these cancers, the majority of the deletions are large and take out both *BRCA1* and *BECN1* and a hundred others. While focal deletions and somatic, predicted loss-of-function mutations (missense, nonsense, frameshift, and splice-site mutations) are found in *BRCA1*; they are not found in *BECN1*. Furthermore, there are no significant germline mutation or allelic loss of *BECN1* in patients with breast and ovarian cancer, nor are there inactivating mutations in the absence of *BRCA1* mutation or loss. This is in agreement with *BRCA1* deficiency being a driver mutation in breast and ovarian cancer. Indeed, allelic loss of *Becn1* suppresses, rather than promotes, mammary tumorigenesis mediated by *Palb2* deficiency (19). As PALB2 is a regulator of BRCA1 and BRCA2 and a known tumor suppressor (18), this suggests that *Becn1* suppresses tumorigenesis of homologous recombination–deficient cancers rather than promoting it.

One interesting tumor type where autophagy may promote tumor progression not included in the analysis here is hepatomas. Mice with allelic loss of *Becn1*, or biallelic deletion of *Atg5* or *Atg7* in liver are prone to liver tumors. Autophagy deficiency may promote initiation of benign liver tumors by inducing chronic tissue damage, but also autophagy may be needed for progression to more aggressive disease. Indeed, deletion of *Atg7* diverts progression of lung adenocarcinomas to benign oncocytomas (13, 14). It will be of interest to examine the mutational status of autophagy genes in human hepatomas and oncocytomas once the sequencing data become available. This will test if autophagy defects both promote the genesis of hepatomas while they limit tumor progression to benign disease (hepatomas and oncocytomas).

Disclosure of Potential Conflicts of Interest

E. White is a consultant/advisory board member of Forma Therapeutics. No potential conflicts of interest were disclosed by the other authors.

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Development of methodology: S. V Laddha, C.S. Chan
Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.): S. V Laddha, C.S. Chan
Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis): S. V Laddha, C.S. Chan
Writing, review, and/or revision of the manuscript: S. V Laddha, S. Ganesan, C.S. Chan, E. White
Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases): S. V Laddha
Study supervision: C.S. Chan

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References

1. Mizushima N, Komatsu M. Autophagy: renovation of cells and tissues. Cell 2011;147:728–41.
2. Rabinowitz JD, White E. Autophagy and metabolism. Science 2010;330:1344–8.
3. White E. Deconvoluting the context-dependent role for autophagy in cancer. Nat Rev Cancer 2012;12:401–10.
4. Aita VM, Liang XH, Murty VV, Pincus DL, Yu W, Cayanis E, et al. Cloning and genomic organization of beclin 1, a candidate tumor suppressor gene on chromosome 17q21. Genomics 1999;59:59–65.
5. Choi AM, Ryter SW, Levine B. Autophagy in human health and disease. N Engl J Med 2013;368:651–62.
6. Liang XH, Jackson S, Seaman M, Brown K, Kempkes B, Hibshoosh H, et al. Induction of autophagy and inhibition of tumorigenesis by beclin 1. Nature 1999;402:672–6.
7. Drost R, Jonkers J. Opportunities and hurdles in the treatment of BRCA1-related breast cancer. Oncogene 2013 Aug 19. [Epub ahead of print].
8. Qu X, Yu J, Bhagat G, Furuya N, Hibshoosh H, Troxel A, et al. Promotion of tumorigenesis by heterozygous disruption of the beclin 1 autophagy gene. J Clin Invest 2003;112:1809–20.
9. Yue Z, Jin S, Yang C, Levine AJ, Heintz N. Beclin 1, an autophagy gene essential for early embryonic development, is a haploinsufficient tumor suppressor. Proc Natl Acad Sci U S A 2003;100:15077–82.
10. Takamura A, Komatsu M, Hara T, Sakamoto A, Kishi C, Waguri S, et al. Autophagy-deficient mice develop multiple liver tumors. Genes Dev 2011;25:795–800.
11. Degenhardt K, Mathew R, Beaudoin B, Bray K, Anderson D, Chen G, et al. Autophagy promotes tumor cell survival and restricts necrosis, inflammation, and tumorigenesis. Cancer Cell 2006;10:51–64.
12. Guo JY, Chen HY, Mathew R, Fan J, Strohecker AM, Karsli-Uzunbas G, et al. Activated Ras requires autophagy to maintain oxidative metabolism and tumorigenesis. Genes Dev 2011;25:460–70.
13. Guo JY, Karsli-Uzunbas G, Mathew R, Aisner SC, Kamphorst JJ, Strohecker AM, et al. Autophagy suppresses progression of K-ras-induced lung tumors to oncocytomas and maintains lipid homeostasis. Genes Dev 2013;27:1447–61.
14. Strohecker AM, Guo JY, Karsli-Uzunbas G, Price SM, Chen GJ, Mathew R, et al. Autophagy sustains mitochondrial glutamine metabolism and growth of BRAFV600E-driven lung tumors. Cancer Discov 2013;3:1272–85.
15. Yang S, Wang X, Contino G, Liesa M, Sahin E, Ying H, et al. Pancreatic cancers require autophagy for tumor growth. Genes Dev 2011;25:717–29.
16. Guo JY, Xia B, White E. Autophagy-mediated tumor promotion. Cell 2013;155:1216–9.
17. Suzuki SW, Onodera J, Ohsumi Y. Starvation induced cell death in autophagy-defective yeast mutants is caused by mitochondria dysfunction. PLoS ONE 2011;6:e17412.
18. Moynahan ME, Jasin M. Mitotic homologous recombination maintains genomic stability and suppresses tumorigenesis. Nat Rev Mol Cell Biol 2010;11:196–207.
19. Huo Y, Cai H, Teplova I, Bowman-Colin C, Chen G, Price S, et al. Autophagy opposes p53-mediated tumor barrier to facilitate tumorigenesis in a model of PALB2-associated hereditary breast cancer. Cancer Discov 2013;3:894–907.
20. Karantza-Wadsworth V, Patel S, Kravchuk O, Chen G, Mathew R, Jin S, et al. Autophagy mitigates metabolic stress and genome damage in mammary tumorigenesis. Genes Dev 2007;21:1621–35.
21. Mathew R, Kongara S, Beaudoin B, Karp CM, Bray K, Degenhardt K, et al. Autophagy suppresses tumor progression by limiting chromosomal instability. Genes Dev 2007;21:1367–81.
22. Vogelstein B, Papadopoulos N, Velculescu VE, Zhou S, Diaz LA Jr, Kinzler KW. Cancer genome landscapes. Science 2013;339:1546–58.
23. Network TCGA. Comprehensive molecular portraits of human breast tumours. Nature 2012;490:61–70.
24. Yusenko MV. Molecular pathology of renal oncocytoma: a review. Int J Urol 2010;17:602–12.
25. Leongamornlert D, Mahmud N, Tymrakiewicz M, Saunders E, Dadaev T, Castro E, et al. Germline BRCA1 mutations increase prostate cancer risk. Br J Cancer 2012;106:1697–701.

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