



Universiteit
Leiden
The Netherlands

RNA splicing in breast cancer progression

Koedoot, E.

Citation

Koedoot, E. (2019, December 17). *RNA splicing in breast cancer progression*. Retrieved from <https://hdl.handle.net/1887/81820>

Version: Publisher's Version

License: [Licence agreement concerning inclusion of doctoral thesis in the Institutional Repository of the University of Leiden](#)

Downloaded from: <https://hdl.handle.net/1887/81820>

Note: To cite this publication please use the final published version (if applicable).

Cover Page



Universiteit Leiden

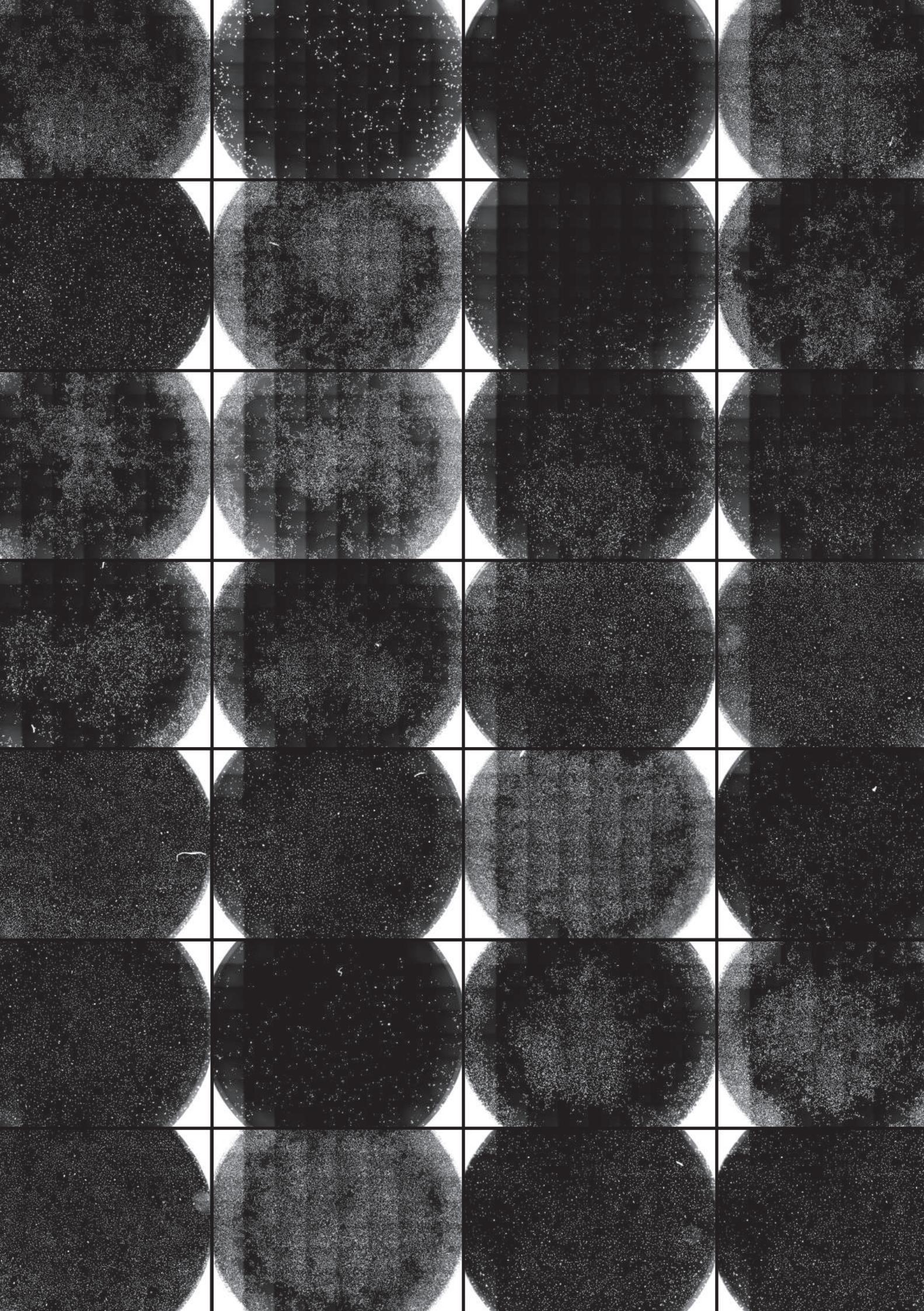


The handle <http://hdl.handle.net/1887/81820> holds various files of this Leiden University dissertation.

Author: Koedoot, E.

Title: RNA splicing in breast cancer progression

Issue Date: 2019-12-17



Appendix

References

Abbreviations

Nederlandse Samenvatting

List of Publications

About the author

◀ IN THE PICTURE

Microscopic view of nuclei of (un)treated breast cancer cells. Nuclei were counted and used as a measure for cell proliferation.

◀ IN BEELD

Microscopisch beeld van celkernen van (on)behandelde borstkankercellen. Door de celkernen te tellen werd het effect van de behandeling op celgroei bepaald.

Appendix

References

1. Ferlay, J. et al. Cancer incidence and mortality patterns in Europe: Estimates for 40 countries and 25 major cancers in 2018. *Eur. J. Cancer* 103, 356–387 (2018).
2. American Cancer Society. Cancer Facts & Figures 2018. *Am. Cancer Soc.* 28–43 (2018). doi:10.1182/blood-2015-12-687814
3. Al, C. et al. *Cancer Incidence in Five Continents, Vol. IX.* (2009).
4. American Cancer Society. Breast Cancer Facts & Figures 2017-2018. (2017).
5. Fouad, T. et al. Overall survival differences between patients with inflammatory and noninflammatory breast cancer presenting with distant metastasis at diagnosis. *Breast Cancer Res Treat* 152, 407–416 (2015).
6. Foulkes, W. D., Smith, I. E. & Reis-Filho, J. S. Triple-Negative Breast Cancer. *N. Engl. J. Med.* 363, 1938–1948 (2010).
7. Perou, C. M. et al. Molecular portraits of human breast tumours. *Nature* 406, 747–752 (2000).
8. Sørlie, T. et al. Gene expression patterns of breast carcinomas distinguish tumor subclasses with clinical implications. *PNAS* 98, 10896–10874 (2001).
9. Gomez, H. L. et al. Efficacy and Safety of Lapatinib As First-Line Therapy for ErbB2-Amplified Locally Advanced or Metastatic Breast Cancer. *J. Clin. Oncol.* 26, 2999–3005 (2008).
10. Vogel, C. et al. Management of ErbB2-positive Breast Cancer: Insights from Preclinical and Clinical Studies with Lapatinib. *Jpn. J. Clin. Oncol.* 40, 999–1013 (2010).
11. Molina, M. A., Codony-servat, J., Albanell, J., Rojo, F. & Baselga, J. Trastuzumab (Herceptin), a Humanized Anti-HER2 Receptor Monoclonal Antibody, Inhibits Basal and Activated HER2 Ectodomain Cleavage in Breast Cancer Cells. *Cancer Res.* 61, 4744–4749 (2001).
12. Pinto, A. C., Ades, F., Azambuja, E. De & Piccart-gehart, M. Trastuzumab for patients with HER2 positive breast cancer: Delivery, duration and combination therapies. *The Breast* 22, S152–S155 (2013).
13. Mohamed, A., Krajewski, K., Cakar, B. & Ma, C. X. Targeted Therapy for Breast Cancer. *Am. J. Pathol.* 183, 1096–1112 (2013).
14. Bauer, K. R., Brown, M., Cress, R. D., Parise, C. A. & Caggiano, V. Descriptive Analysis of Estrogen Receptor (ER)-Negative, Progesterone Receptor (PR)-Negative, and HER2-Negative Invasive Breast Cancer, the So-called Triple-Negative Phenotype. *Cancer* 109, 1721–1728 (2007).
15. Boyle, P. Triple-negative breast cancer: epidemiological considerations and recommendations. *Ann. Oncol.* 23, 8–13 (2012).
16. Sethi, N. & Kang, Y. Unravelling the complexity of metastasis — molecular understanding and targeted therapies. *Nat. Rev. Cancer* 11, 735–748 (2011).
17. Vanharanta, S. & Massagué, J. Origins of metastatic traits. *Cancer Cell* 24, 410–421 (2013).
18. Joyce, J. & Pollard, J. Microenvironmental regulation of metastasis. *Nat. Rev. Cancer* 9, 239–252 (2009).
19. Yates, L. R. et al. Genomic Evolution of Breast Cancer Metastasis and Relapse. *Cancer Cell* 32, 169–184.e7 (2017).
20. Nguyen, D. X., Bos, P. D. & Massagué, J. Metastasis: from dissemination to organ-specific colonization. *Nat. Rev. Cancer* 9, 274–285 (2009).
21. Hanahan, D. & Weinberg, R. A. Hallmarks of cancer: The next generation. *Cell* 144, 646–647 (2011).
22. Hanahan, D. & Weinberg, R. A. The Hallmarks of Cancer. *Cell* 100, 57–70 (2000).
23. Stratton, M. R., Campbell, P. J. & Futreal, P. A. The cancer genome. *Nature* 458, 719–724 (2009).
24. Harney, A. S. et al. Real-time imaging reveals local, transient vascular permeability and tumor cell intravasation stimulated by Tie2hi macrophage-derived VEGFA. *5*, 932–943 (2015).
25. Roussos, E., Condeelis, J. S. & Patsialou, A. Chemotaxis in cancer. *Nat Rev Cancer* 11, 573–587 (2014).
26. Kalluri, R. & Weinberg, R. a. Review series The basics of epithelial-mesenchymal transition. *J. Clin. Invest.* 119, 1420–1428 (2009).
27. Chaffer, C. L. & Weinberg, R. A. A Perspective on Cancer Cell Metastasis. *Science (80-.).* 331, 1559–1564 (2011).
28. Bailly, M. & Condeelis, J. Cell motility: insights from the backstage. *Nat. Cell Biol.* 4, 292–294 (2002).
29. Pollard, T. D. & Borisy, G. G. Cellular Motility Driven by Assembly and Disassembly of Actin Filaments. *Cell* 112, 453–465 (2003).
30. Yamaguchi, H. & Condeelis, J. Regulation of the actin cytoskeleton in cancer cell migration and

- invasion. *Biochim Biophys Acta* 1773, 642–652 (2007).
31. Nicholson-dykstra, S., Higgs, H. N. & Harris, E. S. Actin Dynamics: Growth from Dendritic Branches The dendritic nucleation model was devised to. *Curr. Biol.* 15, 346–357 (2005).
32. Sarmiento, C. et al. WASP family members and formin proteins coordinate regulation of cell protrusions in carcinoma cells. *J. Cell Biol.* 180, 1245–1260 (2008).
33. Webb, D. J., Parsons, J. T. & Horwitz, A. F. Adhesion assembly, disassembly and turnover in migrating cells – over and over and over again. *Nat. Cell Biol.* 4, (2002).
34. Broussard, J. A., Webb, D. J. & Kaverina, I. Asymmetric focal adhesion disassembly in motile cells. *Curr. Opin. Cell Biol.* 20, 85–90 (2008).
35. Zaidel-Bar, R., Itzkovitz, S., Ma'ayan, A., Iyengar, R. & Geiger, B. Functional atlas of the integrin adhesome. *Nat. Cell Biol.* 9, 858–867 (2007).
36. Zaidel-bar, R. & Geiger, B. The switchable integrin adhesome. *J. Cell Sci.* 123, 1385–1388 (2009).
37. Lock, J. G. et al. Plasticity in the Macromolecular-Scale Causal Networks of Cell Migration. *PLoS One* 9, (2014).
38. Gupton, S. L. & Waterman-storer, C. M. Spatiotemporal Feedback between Actomyosin and Focal-Adhesion Systems Optimizes Rapid Cell Migration. *Cell* 125, 1361–1374 (2006).
39. Perou, C. M. Molecular Stratification of Triple-Negative Breast Cancers. *Oncologist* 15, 39–48 (2010).
40. Beerling, E. et al. Plasticity between Epithelial and Mesenchymal States Unlinks EMT from Metastasis-Enhancing Stem Cell Capacity. *Cell Rep.* 14, 2281–2288 (2016).
41. Kedrin, D. et al. Intravital imaging of metastatic behavior through a mammary imaging window. *Nat. Methods* 5, 1019–1021 (2008).
42. Riaz, M. et al. MiRNA expression profiling of 51 human breast cancer cell lines reveals subtype and driver mutation-specific miRNAs. *Breast Cancer Res.* 15, (2013).
43. Friedl, P., Sahai, E., Weiss, S. & Yamada, K. M. New dimensions in cell migration. *Nat. Rev. Mol. Cell Biol.* 13, 743–747 (2012).
44. Liu, Y. J. et al. Confinement and low adhesion induce fast amoeboid migration of slow mesenchymal cells. *Cell* 160, 659–672 (2015).
45. Lee, E. Y. et al. Inactivation of the Retinoblastoma Susceptibility Gene in Human Breast Cancers. *Science (80-).* 241, (1988).
46. T'Ang, A., Varley, J. M., Chakraborty, S., Murphree, A. L. & Fung, Y.-K. T. Structural rearrangement of the retinoblastoma gene in human breast carcinoma. *Science (80-).* 242, (1988).
47. Walsh, T. & King, M. Ten Genes for Inherited Breast Cancer. *Cancer Cell* 11, 103–105 (2007).
48. Tan, H., Zhong, Y. & Pan, Z. Autocrine regulation of cell proliferation by estrogen receptor-alpha in estrogen-receptor-alpha-positive breast cancer cell lines. *BMC Cancer* 9, 1–12 (2009).
49. Bhowmick, N., Neilson, E. & Moses, H. Stromal fibroblasts in cancer initiation and progression. *Nature* 432, 332–337 (2004).
50. Jiang, B.-H. & Liu, L.-Z. PI3K/PTEN signaling in angiogenesis and tumorigenesis. *Adv Cancer Res.* 102, 19–65 (2009).
51. Yuan, T. & Cantley, L. PI3K pathway alterations in cancer: variations on a theme. *Oncogene* 27, 5497–5510 (2008).
52. Davies, M. & Samuels, Y. Analysis of the genome to personalize therapy for melanoma. *Oncogene* 29, 5545–5555 (2010).
53. McDonald, E. R. et al. Project DRIVE: A Compendium of Cancer Dependencies and Synthetic Lethal Relationships Uncovered by Large-Scale, Deep RNAi Screening. *Cell* 170, 577–592.e10 (2017).
54. Tsherniak, A. et al. Defining a Cancer Dependency Map. *Cell* 170, 564–576.e16 (2017).
55. Geenen, J. J. J., Linn, S. C., Beijnen, J. H. & Schellens, J. H. M. PARP Inhibitors in the Treatment of Triple-Negative Breast Cancer. *Clin. Pharmacokinet.* 57, 427–437 (2018).
56. Nitulescu, G. M. et al. Akt inhibitors in cancer treatment: The long journey from drug discovery to clinical use (Review). *Int. J. Oncol.* 48, 869–885 (2016).
57. Zhao, S., Chang, S. L., Linderman, J. J., Feng, F. Y. & Luker, G. D. A Comprehensive Analysis of CXCL12 Isoforms in Breast Cancer. *Transl. Oncol.* 7, 429–438 (2014).
58. Duncan, J. S. et al. Dynamic reprogramming of the kinome in response to targeted MEK inhibition in triple negative breast cancer. *Cell* 149, 307–321 (2012).
59. Sieg, D. J. et al. FAK integrates growth-factor and integrin signals to promote cell migration. *Nat. Cell Biol.* 2, 249–257 (2000).
60. Middelbeek, J. et al. TRPM7 is required for breast tumor cell metastasis. *Cancer Res.* 72, 4250–61 (2012).

Appendix

61. Pearlman, A., Rahman, M. T., Upadhyay, K., Loke, J. & Ostrer, H. Ectopic Otoconin 90 expression in triple negative breast cancer cell lines is associated with metastasis functions. *PLoS One* 1–15 (2019).
62. Humphries, B. A. et al. Plasminogen Activator Inhibitor 1 (PAI1) Promotes Actin Cytoskeleton Reorganization and Glycolytic Metabolism in Triple-Negative Breast Cancer. *Mol. Cancer Res.* 17, 1142–1155 (2019).
63. Montagner, M. et al. SHARP1 suppresses breast cancer metastasis by promoting degradation of hypoxia-inducible factors. *Nature* 487, 380–384 (2012).
64. van Roosmalen, W., Le Dévédec, S., Zovko, S., de Bont, H. & van de Water, B. Functional screening with a live cell imaging-based random cell migration assay. *Methods Mol Biol* 769, 435–448 (2011).
65. Fokkelman, M. et al. PhagoKinetic Track Assay: Imaging and Analysis of Single Cell Migration. *Bio-protocol* 6, (2016).
66. Roosmalen, W. Van et al. Tumor cell migration screen identifies SRPK1 as breast cancer metastasis determinant. *J. Clin. Invest.* 125, 1648–1664 (2015).
67. Oltean, S. & Bates, D. O. Hallmarks of alternative splicing in cancer. *Oncogene* 33, 5311–5318 (2014).
68. Black, D. L. Mechanisms of Alternative Pre-Messenger RNA Splicing. *Annu. Rev. Biochem.* 72, 291–336 (2003).
69. Hegele, A. et al. Dynamic Protein-Protein Interaction Wiring of the Human Spliceosome. *Mol. Cell* 45, 567–580 (2012).
70. Ciriello, G. et al. Comprehensive molecular portraits of invasive lobular breast cancer. *Cell* 163, 506–519 (2015).
71. Berger, A., Korkut, A., Kanchi, R., Hegde, A. & Lenoir, W. A comprehensive Pan-Cancer molecular study of gynecologic and breast cancers. *Cancer Cell* 33, 690–705 (2018).
72. Badve, S. et al. Basal-like and triple-negative breast cancers: A critical review with an emphasis on the implications for pathologists and oncologists. *Mod. Pathol.* 24, 157–167 (2011).
73. Fulford, L. G. et al. Basal-like grade III invasive ductal carcinoma of the breast: Patterns of metastasis and long-term survival. *Breast Cancer Res.* 9, 1–11 (2007).
74. Lehmann, B. D. et al. Refinement of Triple-Negative Breast Cancer Molecular Subtypes: Implications for Neoadjuvant Chemotherapy Selection. *PLoS One* 11, e0157368 (2016).
75. Liu, N. Q. et al. Proteomics pipeline for biomarker discovery of laser capture microdissected breast cancer tissue. *J. Mammary Gland Biol. Neoplasia* 17, 155–164 (2012).
76. Nik-Zainal, S. et al. Landscape of somatic mutations in 560 breast cancer whole-genome sequences. *Nature* 534, 47–54 (2016).
77. Morganella, S. et al. The topography of mutational processes in breast cancer genomes. *Nat. Commun.* 7, 1–11 (2016).
78. Stephens, P. J. et al. The landscape of cancer genes and mutational processes in breast cancer. *Nature* 486, 400–4 (2012).
79. Yates, L. R. & Desmedt, C. Translational genomics: Practical applications of the genomic revolution in breast cancer. *Clin. Cancer Res.* 23, 2630–2639 (2017).
80. Liu, N. Q. et al. Comparative proteome analysis revealing an 11-protein signature for aggressive triple-negative breast cancer. *J. Natl. Cancer Inst.* 106, (2014).
81. Roosmalen, 2015, Tumor cell migration identifies SRPK1 as breast cancer metastasis determinant.
82. de Graauw, M. et al. Annexin A2 depletion delays EGFR endocytic trafficking via cofilin activation and enhances EGFR signaling and metastasis formation. *Oncogene* 33, 2610–2619 (2014).
83. Knott, S. et al. Asparagine bioavailability governs metastasis in a model of breast cancer. *Nature* 554, 378–381 (2018).
84. Wagenblast, E. et al. A model of breast cancer heterogeneity reveals vascular mimicry as a driver of metastasis. *Nature* 520, 358–362 (2015).
85. Li, J.-P. et al. The investigational Aurora kinase A inhibitor alisertib (MLN8237) induces cell cycle G2/M arrest, apoptosis, and autophagy via p38 MAPK and Akt/mTOR signaling pathways in human breast cancer cells. *Drug Des. Devel. Ther.* 9, 1627–1652 (2015).
86. Sahai, E., Garcia-Medina, R., Pouysségur, J. & Vial, E. Smurf1 regulates tumor cell plasticity and motility through degradation of RhoA leading to localized inhibition of contractility. *J. Cell Biol.* 176, 35–42 (2007).
87. Nieto, M. A., Huang, R. Y. Y. J., Jackson, R. A. A. & Thiery, J. P. P. EMT: 2016. *Cell* 166, 21–45 (2016).
88. Naidoo, A., Naidoo, K., Yende-zuma, N. & Gengiah, T. N. NIH Public Access. 19, 161–169 (2015).

89. Naffar-Abu-Amara, S. *et al.* Identification of novel pro-migratory, cancer-associated genes using quantitative, microscopy-based screening. *PLoS One* 3, 1–9 (2008).
90. Carpenter, A. E. *et al.* CellProfiler: image analysis software for identifying and quantifying cell phenotypes. *Genome Biol.* 7, R100 (2006).
91. Yan, K., Verbeek, J. & Verbeek, F. in *ISoLA 2012, PARTII, LNCS 7610* 25–41 (2012). doi:10.1007/978-3-319-47166-2_67
92. Love, M. I., Huber, W. & Anders, S. Moderated estimation of fold change and dispersion for RNA-seq data with DESeq2. *Genome Biol.* 15, 1–21 (2014).
93. Subramanian, A. *et al.* Gene set enrichment analysis: A knowledge-based approach for interpreting genome-wide expression profiles. *Proc. Natl. Acad. Sci.* 102, 15545–15550 (2005).
94. Kamburov, A., Wierling, C., Lehrach, H. & Herwig, R. ConsensusPathDB - A database for integrating human functional interaction networks. *Nucleic Acids Res.* 37, D623–D628 (2009).
95. Kim, D., Langmead, B. & Salzberg, S. L. HISAT: A fast spliced aligner with low memory requirements. *Nat. Methods* 12, 357–360 (2015).
96. Anders, S., Reyes, A. & Huber, W. Detecting differential usage of exons from RNA-seq data. *Genome Res.* 22, 2008–2017 (2012).
97. Reyes, A. *et al.* Drift and conservation of differential exon usage across tissues in primate species. *Proc. Natl. Acad. Sci.* 110, 15377–15382 (2013).
98. Shen, S. *et al.* rMATS : Robust and flexible detection of differential alternative splicing from replicate RNA-Seq data. *PNAS* 5593–5601 (2014). doi:10.1073/pnas.1419161111
99. Xia, J., Benner, M. J. & Hancock, R. E. W. NetworkAnalyst -integrative approaches for protein–protein interaction network analysis and visual exploration. *Nucleic Acids Res.* 42, W167–W174 (2014).
100. Wang, Y. *et al.* Gene-expression profiles to predict distant metastasis of lymph-node-negative primary breast cancer. *Lancet* 365, 671–79 (2005).
101. Yu, J. X. *et al.* Pathway analysis of gene signatures predicting metastasis of node-negative primary breast cancer. *BMC Cancer* 7, 182 (2007).
102. Sotiriou, C. *et al.* Gene expression profiling in breast cancer: Understanding the molecular basis of histologic grade to improve prognosis. *J. Natl. Cancer Inst.* 98, 262–272 (2006).
103. Desmedt, C. *et al.* Strong time dependence of the 76-gene prognostic signature for node-negative breast cancer patients in the TRANSBIG multicenter independent validation series. *Clin. Cancer Res.* 13, 3207–3214 (2007).
104. Schmidt, M. *et al.* The humoral immune system has a key prognostic impact in node-negative breast cancer. *Cancer Res.* 68, 5405–5413 (2008).
105. Williams, E. *et al.* The Image Data Resource: A Bioimage Data Integration and Publication Platform. *Nat. Methods* 14, 775–781 (2017).
106. Patsialou, A. *et al.* Selective gene-expression profiling of migratory tumor cells *in vivo* predicts clinical outcome in breast cancer patients. *Breast Cancer Res.* 14, R139 (2012).
107. Minn, A. J. *et al.* Genes that mediate breast cancer metastasis to lung. *Nature* 436, 518–524 (2005).
108. Minn, A. J. *et al.* Lung metastasis genes couple breast tumor size and metastatic spread. *PNAS* 104, 6740–6745 (2007).
109. Wolf, J. *et al.* An *in vivo* RNAi screen identifies SALL1 as a tumor suppressor in human breast cancer with a role in CDH1 regulation. *Oncogene* 33, 4273–4278 (2014).
110. Lee, C. C. *et al.* TCF12 protein functions as transcriptional repressor of E-cadherin, and its overexpression is correlated with metastasis of colorectal cancer. *J. Biol. Chem.* 287, 2798–2809 (2012).
111. Zi, Z., Chapnick, D. A. & Liu, X. Dynamics of TGF- β /Smad Signaling. *FEBS Lett.* 586, 1921–1928 (2012).
112. van 't Veer, L. J. *et al.* Gene expression profiling predicts clinical outcome of breast cancer. *Nature* 415, 530–536 (2002).
113. Schneider, M. *et al.* Human PRP4 kinase is required for stable tri-snRNP association during spliceosomal B complex formation. *Nat. Struct. Mol. Biol.* 17, 216–222 (2010).
114. Hsu, T. Y.-T. *et al.* The spliceosome is a therapeutic vulnerability in MYC-driven cancer. *Nature* 525, 384–388 (2015).
115. Wysocka, J. *et al.* A PHD finger of NURF couples histone H3 lysine 4 trimethylation with chromatin remodelling. *Nature* 442, 86–90 (2006).
116. Frey, W. D. *et al.* BPTF maintains chromatin accessibility and the self-renewal capacity of mammary gland stem cells. *Stem Cell Reports* 9, 23–31 (2017).
117. Wong, J. J. *et al.* Orchestrated Intron Retention Regulates Normal Granulocyte Differentiation.

Appendix

- Cell 154, 583–595 (2013).
118. Braunschweig, U. *et al.* Widespread intron retention in mammals functionally tunes transcriptomes. *Genome Res.* 24, 1774–1786 (2014).
119. Bergeron, D., Pal, G., Beaulieu, Y. B. & Chabot, B. Regulated Intron Retention and Nuclear Pre-mRNA Decay Contribute to PABPN1 Autoregulation. *Mol. Cell. Biol.* 35, 2503–2517 (2015).
120. Simpson, K. J. *et al.* Identification of genes that regulate epithelial cell migration using an siRNA screening approach. *Nat. Cell Biol.* 10, 1027–1038 (2008).
121. van der Weyden, L. *et al.* Genome-wide in vivo screen identifies novel host regulators of metastatic colonization. *Nature* 541, 233–236 (2017).
122. Marabti, E. El & Younis, I. The Cancer Spliceosome: Reprograming of Alternative Splicing in Cancer. *Front. Mol. Biosci.* 5, 1–11 (2018).
123. Marzese, D. M., Manughian, A. O., Javier, P. & Dave, I. J. O. Alternative splicing and cancer metastasis : prognostic and therapeutic applications. *Clin. Exp. Metastasis* 35, 393–402 (2018).
124. Pelisch, F. *et al.* Involvement of hnRNP A1 in the matrix metalloprotease-3-dependent regulation of Rac1 pre-mRNA splicing. *J Cell Biochem* 113, 2319–2329 (2012).
125. He, X. *et al.* Involvement of polypyrimidine tract-binding protein (PTBP1) in maintaining breast cancer cell growth and malignant properties. *Oncogenesis* 3, e84 (2014).
126. Shimoni-Sebag, A., Lebenthal-Loinger, I., Zender, L. & Karni, R. RRM1 domain of the splicing oncoprotein SRSF1 is required for MEK1-MAPK-ERK activation and cellular transformation. *Carcinogenesis* 34, 2498–2504 (2013).
127. Anczuków, O. *et al.* The splicing factor SRSF1 regulates apoptosis and proliferation to promote mammary epithelial cell transformation. *Nat Struct Mol Biol.* 19, 220–228 (2012).
128. Gao, Q. *et al.* Evaluation of cancer dependence and druggability of PRP4 kinase using cellular, biochemical, and structural approaches. *J. Biol. Chem.* 288, 30125–30138 (2013).
129. Petrocca, F. *et al.* A Genome-wide siRNA Screen Identifies Proteasome Addiction as a Vulnerability of Basal-like Triple-Negative Breast Cancer Cells. *Cancer Cell* 24, 182–196 (2013).
130. Black, D. L. Protein Diversity from Alternative Splicing. *Cell* 103, 367–370 (2000).
131. Smith, C. W. J. & Valcárcel, J. Alternative pre-mRNA splicing: the logic of combinatorial control. *Trends Biochem. Sci.* 25, 381–388 (2000).
132. Yang, X. *et al.* Widespread Expansion of Protein Interaction Capabilities by Alternative Splicing. *Cell* 164, 805–817 (2016).
133. David, C. J. & Manley, J. L. Alternative pre-mRNA splicing regulation in cancer: Pathways and programs unhinged. *Genes Dev.* 24, 2343–2364 (2010).
134. Ghigna, C., Valacca, C. & Biamonti, G. Alternative Splicing and Tumor Progression. *Curr. Genomics* 9, 556–570 (2008).
135. Fackenthal, J. D. & Godley, L. A. Aberrant RNA splicing and its functional consequences in cancer cells. *Dis. Model. Mech.* 1, 37–42 (2008).
136. Agafonov, D. E. *et al.* Semiquantitative Proteomic Analysis of the Human Spliceosome via a Novel Two-Dimensional Gel Electrophoresis Method. *Mol. Cell. Biol.* 31, 2667–2682 (2011).
137. Zhu, J., Mayeda, A. & Krainer, A. R. Exon Identity Established through Differential Antagonism between Exonic Splicing Silencer-Bound hnRNP A1 and Enhancer-Bound SR Proteins. *Mol. Cell* 8, 1351–1361 (2001).
138. Tange, T., Damgaard, C. K., Guth, S., Valcárcel, J. & Kjems, J. The hnRNP A1 protein regulates HIV-1 tat splicing via a novel intron silencer element. *EMBO J.* 20, 5748–5758 (2001).
139. House, A. E. & Lynch, K. W. An exonic splicing silencer represses spliceosome assembly after ATP-dependent exon recognition. *Nat. Struct. Mol. Biol.* 13, 937–944 (2006).
140. Ge, H. & Manley, J. L. A Protein Factor, ASF, Controls Cell-Specific Alternative Splicing of SV40 Early Pre-mRNA In Vitro. *Cell* 62, 25–34 (1990).
141. Krainer, A. R., Conway, G. C. & Kozak, D. Purification and characterization of pre-mRNA splicing factor SF2 from HeLa cells. *Genes Dev.* 4, 1158–1171 (1990).
142. Fu, X. & Maniatis, T. The 35-kDa mammalian splicing factor SC35 mediates specific interactions between U1 and U2 small nuclear ribonucleoprotein particles at the 3' splice site. *Proc. Natl. Acad. Sci. USA* 89, 1725–1729 (1992).
143. Jurica, M. S. & Moore, M. J. Pre-mRNA Splicing: Awash in a Sea of Proteins. *Mol. Cell* 12, 5–14 (2003).
144. Wahl, M. C., Will, C. L. & Lührmann, R. The Spliceosome: Design Principles of a Dynamic RNP Machine. *Cell* 136, 701–718 (2009).
145. Faustino, N. & Cooper, T. A. Pre-mRNA splicing and human disease. *Genes Dev.* 17, 419–437 (2003).
146. Stoilov, P. *et al.* Defects in Pre-mRNA Processing as Causes of and Predisposition to Diseases.

- DNA Cell Biol.* 21, 803–818 (2002).
147. Wen, J., Toomer, K. H., Chen, Z. & Cai, X. Genome-wide analysis of alternative transcripts in human breast cancer. *Breast Cancer Res. Treat.* 151, 295–307 (2015).
148. Venables, J. P. et al. Cancer-associated regulation of alternative splicing. *Nat. Struct. Mol. Biol.* 16, 670–676 (2009).
149. Shapiro, I. M. et al. An emt-driven alternative splicing program occurs in human breast cancer and modulates cellular phenotype. *PLoS Genet.* 7, (2011).
150. Valcárcel, J. & Green, M. R. The SR protein family: pleiotropic functions in. *Elsevier Sci. Ltd* S0968-0004, 10039–6 (1996).
151. Hagiwara, M. Alternative splicing: A new drug target of the post-genome era. *Biochim. Biophys. Acta* 1754, 324–331 (2005).
152. Tenenbaum, S. A. & Aguirre-ghiso, J. Dephosphorylation Shows SR Proteins the Way Out. *Mol. Cell* 20, 499–501 (2005).
153. Chen, Y. et al. Mutually exclusive acetylation and ubiquitylation of the splicing factor SRSF5 control tumor growth. *Nat. Commun.* 9, (2018).
154. Huang, Y., Yario, T. A. & Steitz, J. A. A molecular link between SR protein dephosphorylation and mRNA export. *PNAS* 101, 9666–9670 (2004).
155. Sanford, J. R., Ellis, J. D., Cazalla, D. & Cáceres, J. F. Reversible phosphorylation differentially affects nuclear and cytoplasmic functions of splicing factor 2/alternative splicing factor. *PNAS* 102, 15042–15047 (2005).
156. Cáceres, J. F., Screaton, G. R. & Krainer, A. R. A specific subset of SR proteins shuttles continuously between the nucleus and the cytoplasm. *Genes Dev.* 12, 55–66 (1998).
157. Huang, Y., Gattoni, R. & Steitz, J. A. SR Splicing Factors Serve as Adapter Proteins for TAP-Dependent mRNA Export. *Mol. Cell* 11, 837–843 (2003).
158. Buckley, P. T., Khaladkar, M., Kim, J. & Eberwine, J. Cytoplasmic intron retention, function, splicing, and the sentinel RNA hypothesis. *Wiley Interdiscip. Rev. RNA* 5, 223–230 (2014).
159. Archer, S. Y. & Hodin, R. A. Histone acetylation and cancer. *Curr. Opin. Genet. Dev.* 9, 171–174 (1999).
160. Gonzalez, I. et al. A lncRNA regulates alternative splicing via establishment of a splicing-specific chromatin signature. *Nat Struct Mol Biol.* 22, 370–376 (2015).
161. Risso, G. J., Pawellek, A., Ule, J., Lamond, A. I. & Kornblith, A. R. Perturbation of Chromatin Structure Globally Affects Localization and Recruitment of Splicing Factors. *PLoS One* 7, (2012).
162. Sharma, A. et al. Calcium-mediated histone modifications regulate alternative splicing in cardiomyocytes. *PNAS* E4920–E4928 (2014). doi:10.1073/pnas.1408964111
163. Sperling, R. Small non-coding RNA within the endogenous spliceosome and alternative splicing regulation. *Biochim Biophys Acta Gene Regul Mech* S1874-9399, 30537–6 (2019).
164. Yoshida, K. et al. Frequent pathway mutations of splicing machinery in myelodysplasia. *Nature* 478, 64–69 (2011).
165. Yoshida, K. & Ogawa, S. Splicing factor mutations and cancer. *Wiley Interdiscip. Rev. RNA* 5, 445–459 (2014).
166. Ilagan, J. O. et al. U2AF1 mutations alter splice site recognition in hematological malignancies. *Genome Res.* 25, 14–26 (2015).
167. Ellis, M. J. et al. Whole Genome Analysis Informs Breast Cancer Response to Aromatase Inhibition. *Nature* 486, 353–360 (2012).
168. Stephens, P. J. et al. The landscape of cancer genes and mutational processes in breast cancer. *Nature* 486, 400–404 (2012).
169. Nik-Zainal, S. et al. Landscape of somatic mutations in 560 breast cancer whole genome sequences. *Nature* 534, 47–54 (2016).
170. DeBoever, C. et al. Transcriptome Sequencing Reveals Potential Mechanism of Cryptic 3??? Splice Site Selection in SF3B1-mutated Cancers. *PLoS Comput. Biol.* 11, 1–19 (2015).
171. Alsafadi, S. et al. Cancer-associated SF3B1 mutations affect alternative splicing by promoting alternative branchpoint usage. *Nat. Commun.* 7, 1–12 (2016).
172. Shirashi, Y. et al. A comprehensive characterization of cis -acting splicing-associated variants in human cancer. *Genome R* 28, 1111–1125 (2018).
173. Naftelberg, S., Schor, I. E., Ast, G. & Kornblith, A. R. Regulation of Alternative Splicing Through Coupling with Transcription and Chromatin Structure. *Annu. Rev. Biochem.* 84, 165–198 (2015).
174. Wang, Y. et al. A complex network of factors with overlapping affinities represses splicing through intronic elements. *Nat. Struct. Mol. Biol.* 20, (2013).
175. Mazoyer, S. et al. A BRCA1 Nonsense Mutation Causes Exon Skipping. *Am J Hum Genet* 62, 713–715 (1998).

Appendix

176. Kim, E. et al. SRSF2 Mutations Contribute to Myelodysplasia Through Mutant- Specific Effects on Exon Recognition. *Cancer Cell* 27, 617–630 (2015).
177. Fruman, D. A. & Rommel, C. PI3K and Cancer: Lessons, Challenges and Opportunities. *Nat Rev Drug Discov* 13, 140–156 (2014).
178. Dhillon, A. S., Hagan, S., Rath, O. & Kolch, W. MAP kinase signalling pathways in cancer. *Oncogene* 26, 3279–3290 (2007).
179. Gil, E. M. Targeting the PI3K/AKT/mTOR pathway in estrogen receptor-positive breast cancer. *Cancer Treat. Rev.* 40, 862–871 (2014).
180. Santen, R. J. et al. The role of mitogen-activated protein (MAP) kinase in breast cancer. *J. ster* 80, 239–256 (2002).
181. Pace, P., Taylor, J., Suntharalingam, S., Coombes, R. C. & Ali, S. Human Estrogen Receptor beta Binds DNA in a Manner Similar to and Dimerizes with Estrogen Receptor alpha. *J. biol* 272, 25832–25838 (1997).
182. Hah, N. et al. A Rapid, Extensive, and Transient Transcriptional Response to Estrogen Signaling in Breast Cancer Cells. *Cell* 145, 622–634 (2011).
183. Frasor, J. et al. Profiling of Estrogen Up- and Down-Regulated Gene Expression in Human Breast Cancer Cells: Insights into Gene Networks and Pathways Underlying Estrogenic Control of Proliferation and Cell Phenotype. *Endocrinology* 144, 4562–4574 (2003).
184. Honma, N. et al. Clinical importance of estrogen receptor-beta evaluation in breast cancer patients treated with adjuvant tamoxifen therapy. *J Clin Oncol* 26, 3727–3734 (2008).
185. Guo, L. et al. Significance of ERbeta expression in different molecular subtypes of breast cancer. *Diagn. Pathol.* 9, 20 (2014).
186. Chantzi, N. et al. Estrogen receptor beta 2 is associated with poor prognosis in estrogen receptor alpha-negative breast carcinoma. *J Cancer Res Clin Oncol* 139, 1489–1498 (2013).
187. Gökmen-Polar, Y. et al. Expression levels of SF3B3 correlate with prognosis and endocrine resistance in estrogen receptor-positive breast cancer. *Mod. Pathol.* 28, 677–685 (2015).
188. Lahsaee, S., Corkery, D. P., Anthes, L. E., Holly, A. & Dellaire, G. Estrogen receptor alpha (ESR1)-signaling regulates the expression of the taxane-response biomarker PRP4K. *Exp. Cell Res.* 340, 125–131 (2016).
189. Ohe, K. et al. HMGA1a Induces Alternative Splicing of the Estrogen Receptor-alpha Gene by Trapping U1 snRNP to an Upstream Pseudo-5' Splice Site. *Front. Mol. Biosci.* 5, 1–8 (2018).
190. Nassar, G. et al. Comparative analysis of nuclear estrogen receptor alpha and beta interactomes in breast cancer cells. *Mol. Biosyst.* 7, 667–76 (2011).
191. Dago, D. N. et al. Estrogen receptor beta impacts hormone-induced alternative mRNA splicing in breast cancer cells. *BMC Genomics* 16, 367 (2015).
192. Corkery, D. P. et al. Prp4k is a her2-regulated modifier of taxane sensitivity. *Cell Cycle* 14, 1059–1069 (2015).
193. Castiglioni, F. et al. Role of exon-16-deleted HER2 in breast carcinomas. *Endocr. Relat. Cancer* 13, 221–232 (2006).
194. Mitra, D. et al. An oncogenic isoform of HER2 associated with locally disseminated breast cancer and trastuzumab resistance. *Mol Cancer Ther* 8, 2152–2163 (2009).
195. Azios, N. G., Romero, F. J., Denton, M. C., Doherty, J. K. & Clinton, G. M. Expression of herstatin , an autoinhibitor of HER-2/neu , inhibits transactivation of HER-3 by HER-2 and blocks EGF activation of the EGF receptor. *Oncogene* 20, 5199–5209 (2001).
196. Hu, P. et al. Sequestering ErbB2 in endoplasmic reticulum by its autoinhibitor from translocation to cell surface: An autoinhibition mechanism of ErbB2 expression. *Biochem. Biophys. Res. Commun.* 342, 19–27 (2006).
197. Hu, P. et al. In Vivo Identification of the Interaction Site of ErbB2 Extracellular Domain With its Autoinhibitor. *J. Cell. Physiol.* 205, 335–343 (2005).
198. Todorović-Raković, N., Nešković-Konstantinović, Z. & Nikolić-Vukosavljević, D. Cross-talk between ER and HER2 in breast carcinoma. *Arch. Oncol.* 14, 146–150 (2006).
199. Best, A. et al. Expression of Tra2beta in cancer cells as a potential contributory factor to neoplasia and metastasis. *Int. J. Cell Biol.* 2013, (2013).
200. Chang, Y., Hsu, Y., Chen, Y., Wang, Y. & Huang, S.-M. Theophylline exhibits anti-cancer activity via suppressing SRSF3 in cervical and breast cancer cell lines. *Oncotarget* 8, 101461–101474 (2017).
201. Rengasamy, M. et al. The PRMT5/WDR77 complex regulates alternative splicing through ZNF326 in breast cancer. *Nucleic Acids Res.* 45, 11106–11120 (2017).
202. Wu, Y. et al. Function of HNRNPC in breast cancer cells by controlling the dsRNA-induced interferon response. *EMBO J.* 1–19 (2018). doi:10.15252/embj.201899017

203. Cannizzaro, E., Bannister, A. J., Han, N., Alendar, A. & Kouzarides, T. DDX3X RNA helicase affects breast cancer cell cycle progression by regulating expression of KLF4. *FEBS Lett.* 592, 2308–2322 (2018).
204. Hu, Y. *et al.* Splicing factor hnRNPA2B1 contributes to tumorigenic potential of breast cancer cells through STAT3 and ERK1/2 signaling pathway. *Tumor Biol.* 1–11 (2017). doi:10.1177/1010428317694318
205. Koh, C. M. *et al.* MYC regulates the core pre-mRNA splicing machinery as an essential step in lymphomagenesis. *Nature* 523, 96–100 (2015).
206. Anczuków, O. *et al.* SRSF1-Regulated Alternative Splicing in Breast Cancer. *Mol. Cell* 60, 105–117 (2015).
207. Gao, Y. & Koide, K. Chemical perturbation of Mcl-1 pre-mRNA splicing to induce apoptosis in cancer cells. *ACS Chem Biol* 8, 895–900 (2013).
208. Talmadge, J. & Fidler, I. AACR Centennial Series: The Biology of Cancer Metastasis: Historical Perspective. 70, 5649–5669 (2010).
209. Fidler, I. J. The pathogenesis of cancer metastasis: the ‘seed and soil’ hypothesis revisited. *Nat. Rev. Cancer* 3, 1–6 (2003).
210. Gonzalez, D. M. & Medici, D. Signaling mechanisms of the epithelial-mesenchymal transition. *Sci Signal* 7, (2015).
211. Li, J. *et al.* An alternative splicing switch in FLNB promotes the mesenchymal cell state in human breast cancer. *Elife* 7, 1–28 (2018).
212. Fici, P. *et al.* Splicing factor ratio as an index of epithelial-mesenchymal transition and tumor aggressiveness in breast cancer. *Oncotarget* 8, 2423–2436 (2017).
213. Warzecha, C. C., Sato, T. K., Nabet, B., Hogenesch, J. B. & Carstens, R. P. ESRP1 and ESRP2 are epithelial cell type-specific regulators of FGFR2 splicing. *Mol Cell* 33, 591–601 (2009).
214. Martinez-Contreras, R. *et al.* hnRNP proteins and splicing control. *Adv Exp Med Biol* 623, 123–147 (2007).
215. Cammas, A. *et al.* hnRNP A1-mediated translational regulation of the G quadruplex-containing RON receptor tyrosine kinase mRNA linked to tumor progression. *Oncotarget* 7, 16793–16805 (2016).
216. Fiegen, D. *et al.* Alternative Splicing of Rac1 Generates Rac1b, a Self-activating GTPase. *J. Biol. Chem.* 279, 4743–4749 (2004).
217. Matos, P., Collard, J. G. & Jordan, P. Tumor-related Alternatively Spliced Rac1b Is Not Regulated by Rho-GDP Dissociation Inhibitors and Exhibits Selective Downstream Signaling. *J. Biol. Chem.* 278, 50442–50448 (2003).
218. Schnelzer, A. *et al.* Rac1 in human breast cancer: overexpression, mutation analysis, and characterization of a new isoform, Rac1b. *Oncogene* 19, 3013–3020 (2000).
219. Radisky, D. C. *et al.* Rac1b and reactive oxygen species mediate MMP-3-induced EMT and genomic instability. *Nature* 436, 123–127 (2005).
220. Harvey, S. E. *et al.* Coregulation of alternative splicing by hnRNPM and ESRP1 during EMT. *RNA* 24, 1326–1338 (2018).
221. Xu, Y. *et al.* Cell type-restricted activity of hnRNPM promotes breast cancer metastasis via regulating alternative splicing. *Genes Dev.* 28, 1191–1203 (2014).
222. Sun, H. *et al.* HnRNPM and CD44s expression affects tumor aggressiveness and predicts poor prognosis in breast cancer with axillary lymph node metastases. *Genes Chromosom. Cancer* 56, 598–607 (2017).
223. Huot, M.-E., Vogel, G. & Richard, S. Identification of a Sam68 ribonucleoprotein complex regulated by epidermal growth factor. *J. Biol. Chem.* 284, 31903–31913 (2009).
224. Watermann, D. O. *et al.* Splicing factor Tra2-β1 is specifically induced in breast cancer and regulates alternative splicing of the CD44 gene. *Cancer Res.* 66, 4774–4780 (2006).
225. Shimonov-sebag, A., Lebenthal-loinger, I., Zender, L. & Karni, R. RRM1 domain of the splicing oncprotein SRSF1 is required for MEK1-MAPK-ERK activation and cellular transformation. *Carcinogenesis* 34, 2498–2504 (2013).
226. Bonomi, S. *et al.* HnRNP A1 controls a splicing regulatory circuit promoting mesenchymal-to-epithelial transition. *Nucleic Acids Res.* 41, 8665–8679 (2013).
227. Corkery, D. P. *et al.* Loss of PRP4K drives anoikis resistance in part by dysregulation of epidermal growth factor receptor endosomal trafficking. *Oncogene* 37, 174–184 (2018).
228. Bondy-Chorney, E. *et al.* RNA binding protein RALY promotes Protein Arginine Methyltransferase 1 alternatively spliced isoform v2 relative expression and metastatic potential in breast cancer cells. *Int. J. Biochem. Cell Biol.* 91, 124–135 (2017).
229. Tien, J. F. *et al.* CDK12 regulates alternative last exon mRNA splicing and promotes breast cancer

Appendix

- cell invasion. *Nucleic Acids Res.* 45, 6698–6716 (2017).
230. Lin, J.-C., Lin, C.-Y., Tarn, W.-Y. & Li, F.-Y. Elevated SRPK1 lessens apoptosis in breast cancer cells through RBM4-regulated splicing events. *RNA* 20, 0–11 (2014).
231. Zheng, Y. et al. PHF5A Epigenetically Inhibits Apoptosis to Promote Breast Cancer Progression. *Cancer Res* 78, 3190–3207 (2018).
232. Gaytan-cervantes, J. et al. Sam36 regulates the alternative splicing of survivin DEx3. *JBC* (2017). doi:10.1074/jbc.M117.800318
233. Bielli, P., Bordi, M., Biasio, V. Di & Sette, C. Regulation of BCL-X splicing reveals a role for the polypyrimidine tract binding protein (PTBP1 / hnRNP I) in alternative 5' splice site selection. *Nucleic Acids Res.* 42, 12070–12081 (2014).
234. Koumbadinga, G. A. et al. Increased stability of heterogeneous ribonucleoproteins by a deacetylase inhibitor. *Biochim. Biophys. Acta - Gene Regul. Mech.* 1849, 1095–1103 (2015).
235. Paronetto, M. P., Achsel, T., Massiello, A., Chalfant, C. E. & Sette, C. The RNA-binding protein Sam68 modulates the alternative splicing of Bcl-x. *J. Cell Biol.* 176, 929–939 (2007).
236. Revil, T., Pelletier, J., Toutant, J., Cloutier, A. & Chabot, B. Heterogeneous Nuclear Ribonucleoprotein K Represses the Production of Pro-apoptotic Bcl-xs Splice Isoform. *J. Biol. Chem.* 284, 21458–21467 (2009).
237. Milek, M. et al. DDX54 regulates transcriptome dynamics during DNA damage response. *Genome Res.* 27, 1344–1359 (2017).
238. Sampath, J. et al. Human SPF45, a Splicing Factor, Has Limited Expression in Normal Tissues, Is Overexpressed in Many Tumors, and Can Confer a Multidrug-Resistant Phenotype to Cells. *Am. J. Pathol.* 163, 1781–1790 (2003).
239. Hayes, G. M., Carrigan, P. E. & Miller, L. J. Serine-arginine protein kinase 1 overexpression is associated with tumorigenic imbalance in mitogen-activated protein kinase pathways in breast, colonic, and pancreatic carcinomas. *Cancer Res.* 67, 2072–2080 (2007).
240. Liu, T. et al. TRA2A Promoted Paclitaxel Resistance and Tumor Progression in Triple-Negative Breast Cancers via Regulating Alternative Splicing. *Mol. Cancer Ther.* 16, 1377–1388 (2017).
241. Gabriel, M. et al. Role of the splicing factor SRSF4 in cisplatin-induced modifications of pre-mRNA splicing and apoptosis. *BMC Cancer* 15, 227 (2015).
242. David, C. J., Chen, M., Assanah, M., Canoll, P. & Manley, J. L. HnRNP proteins controlled by c-Myc deregulate pyruvate kinase mRNA splicing in cancer. *Nature* 463, 364–368 (2010).
243. Babic, I. et al. EGFR Mutation-Induced Alternative Splicing of Max Contributes to Growth of Glycolytic Tumors in Brain Cancer. *Cell Metab.* 17, 1000–1008 (2013).
244. Han, J. et al. Hypoxia is a Key Driver of Alternative Splicing in Human Breast Cancer Cells. *Sci. Rep.* 7, 1–17 (2017).
245. Minchenko, O. H., Ogura, T., Opentanova, I. L., Minchenko, D. O. & Esumi, H. Splice isoform of 6-phosphofructo-2-kinase/ fructose-2,6-bisphosphatase-4: Expression and hypoxic regulation. *Mol. Cell. Biochem.* 280, 227–234 (2005).
246. Gasparini, G. Prognostic value of vascular endothelial growth factor in breast cancer. *Oncologist* 5 Suppl 1, 37–44 (2000).
247. Moreira, I., Fernandes, P. & Ramos, M. Vascular endothelial growth factor (VEGF) inhibition - a critical review. *Anti-Cancer Agents* 7, (2007).
248. Harper, S. J. & Bates, D. O. VEGF-A splicing : the key to anti-angiogenic therapeutics? *Nat Rev Cancer* 8, 880–887 (2008).
249. Bates, D. O. et al. VEGF165b, an Inhibitory Splice Variant of Vascular Endothelial Growth Factor, Is Down-Regulated in Renal Cell Carcinoma. *Cancer Res.* 62, 4123–4131 (2002).
250. Woolard, J. et al. VEGF165b, an Inhibitory Vascular Endothelial Growth Factor Splice Variant: Mechanism of Action, In vivo Effect On Angiogenesis and Endogenous Protein Expression. *Cancer Res.* 64, 7822–7835 (2004).
251. Mavrou, A. et al. Serine Arginine Protein Kinase-1 (SRPK1) inhibition as a potential novel targeted therapeutic strategy in prostate cancer. *Oncogene* 34, 4311–4319 (2015).
252. Amin, E. M. et al. WT1 mutants reveal SRPK1 to be a downstream angiogenesis target by altering VEGF splicing. *Cancer Cell* 20, 768–780 (2011).
253. Nowak, D. G. et al. Regulation of Vascular Endothelial Growth Factor (VEGF) splicing from pro-angiogenic to anti-angiogenic isoforms: A novel therapeutic strategy for angiogenesis. *J. Biol. Chem.* 285, 5532–5540 (2010).
254. Finley, S. D. & Popel, A. S. Predicting the effects of anti-angiogenic agents targeting specific VEGF isoforms. *AAPS J.* 14, 500–9 (2012).
255. Giampietro, C. et al. The alternative splicing factor Nova2 regulates vascular development and lumen formation. *Nat. Commun.* 1–15 (2015). doi:10.1038/ncomms9479

256. Macara, I. G. Par Proteins: Partners in Polarization. *Curr. Biol.* 14, 160–162 (2004).
257. Koh, W., Mahan, R. D. & Davis, G. E. Cdc42- and Rac1-mediated endothelial lumen formation requires Pak2, Pak4 and Par3, and PKC- dependent signaling. *J. Cell Sci.* 121, 989–1001 (2008).
258. Iden, S. *et al.* A distinct PAR complex associates physically with VE-cadherin in vertebrate endothelial cells. *EMBO Rep.* 7, 1239–1246 (2006).
259. Ladd, J. J. *et al.* Autoantibody Signatures Involving Glycolysis and Splicesome Proteins Precede a Diagnosis of Breast Cancer among Postmenopausal Women. *Cancer Res.* 73, 1502–1514 (2013).
260. Katayama, H. *et al.* An Autoimmune Response Signature Associated with the Development of Triple-Negative Breast Cancer Reflects Disease Pathogenesis. *Cancer Res.* 75, 3246–3255 (2015).
261. Nakajima, H. *et al.* New antitumor substances, FR901463, FR901464 and FR901465. II. Activities against experimental tumors in mice and mechanism of action. *J Antibiot* 49, 1204–1211 (1996).
262. Mizui, Y. *et al.* Pladienolides, New substances from culture of streptomyces platensis MER-11107 III. *J. Antibiot. (Tokyo)*. 57, 188–196 (2004).
263. Sakai, Y. *et al.* GEX1 compounds, novel antitumor antibiotics related to herboxidiene, produced by Streptomyces sp. II. The effects on cell cycle progression and gene expression. *J. Antibiot. (Tokyo)*. 55, 863–72 (2002).
264. Albert, B. J. *et al.* Meayamycin inhibits pre-mRNA splicing and exhibits picomolar activity against multidrug resistant cells. *Mol. Cancer Ther.* 8, 2308–2318 (2009).
265. Kaida, D. *et al.* Spliceostatin A targets SF3b and inhibits both splicing and nuclear retention of pre-mRNA. *Nat. Chem. Biol.* 3, 576–583 (2007).
266. Kotake, Y. *et al.* Splicing factor SF3b as a target of the antitumor natural product pladienolide. *Nat. Chem. Biol.* 3, 570–575 (2007).
267. Otsuka, K., Yamamoto, Y. & Ochiya, T. Regulatory role of resveratrol, a microRNA-controlling compound, in HNRNPA1 expression, which is associated with poor prognosis in breast cancer. *Oncotarget* 9, 24718–24730 (2018).
268. Iwai, K. *et al.* Anti-tumor efficacy of a novel CLK inhibitor via targeting RNA splicing and MYC-dependent vulnerability. *EMBO Mol. Med.* 10, 1–15 (2018).
269. Zaharieva, E., Chipman, J. K. & Soller, M. Alternative splicing interference by xenobiotics. *Toxicology* 296, 1–12 (2012).
270. Batson, J. *et al.* Development of Potent, Selective SRPK1 Inhibitors as Potential Topical Therapeutics for Neovascular Eye Disease. *ACS Chem. Biol.* 12, 825–832 (2017).
271. Hatcher, J. M. *et al.* SRPKIN-1: A Covalent SRPK1/2 Inhibitor that Potently Converts VEGF from Pro-angiogenic to Anti-angiogenic Isoform. *Cell Chem Biol* 25, 460–470 (2018).
272. Summerton, J. Morpholino antisense oligomers: the case for an RNase H-independent structural type. *Biochim. Biophys. Acta* 1489, 141–158 (1999).
273. Geary, R. S. Antisense oligonucleotide pharmacokinetics and metabolism. *Expert Opin. Metab. Toxicol.* 5, 381–392 (2009).
274. Moulton, H. M. & Moulton, J. D. Morpholinos and their peptide conjugates: Therapeutic promise and challenge for Duchenne muscular dystrophy. *Biochim. Biophys. Acta* 1798, 2296–2303 (2010).
275. Havens, M. A. & Hastings, M. L. Splice-switching antisense oligonucleotides as therapeutic drugs. *Nucleic Acids Res.* 44, 6549–6563 (2016).
276. Denichenko, P. *et al.* Specific inhibition of splicing factor activity by decoy RNA oligonucleotides. *Nat. Commun.* 10, (2019).
277. Smith, I. *et al.* Evaluation of RNAi and CRISPR technologies by large-scale gene expression profiling in the Connectivity Map. *Plos Biol.* 15, 1–23 (2017).
278. Wan, J., Sazani, P. & Kole, R. Modification of HER2 pre-mRNA alternative splicing and its effects on breast cancer cells. *Int J Cancer* 124, 772–777 (2009).
279. Pan, Q., Shai, O., Lee, L. J., Frey, B. J. & Blencowe, B. J. Deep surveying of alternative splicing complexity in the human transcriptome by high-throughput sequencing. *Nat. Genet.* 40, 1413–1416 (2009).
280. Wang, E. T. *et al.* Alternative isoform regulation in human tissue transcriptomes. *Nature* 456, 470–476 (2008).
281. Zhu, Y., Qiu, P. & Ji, Y. TCGA-Assembler: an open-source pipeline for TCGA data downloading, assembling and processing. *Nat Methods* 11, 599–600 (2014).
282. Forbes, S. A. *et al.* COSMIC: exploring the world's knowledge of somatic mutations in human cancer. *Nucleic Acids Res.* 43, 805–811 (2015).
283. Scotti, M. M. & Swanson, M. RNA mis-splicing in disease Marina. *Nat Rev Genet* 17, 19–32 (2015).

Appendix

284. Dutertre, M., Vagner, S. & Auboeuf, D. Alternative splicing and breast cancer. *RNA Biol.* 7, 403–411 (2010).
285. Chen, M., Zhang, J. & Manley, J. L. Turning on a fuel switch of cancer - hnRNP proteins regulate alternative splicing of pyruvate kinase mRNA. *Cancer Res.* 70, 8977–8980 (2010).
286. Lee, S. C.-W. & Abdel-Wahab, O. Therapeutic targeting of splicing in cancer. *Nat Med* 22, 976–986 (2016).
287. Smid, M. et al. Breast cancer genome and transcriptome integration implicates specific mutational signatures with immune cell infiltration. *Nat. Commun.* 7, 1–9 (2016).
288. Györffy, B. et al. An online survival analysis tool to rapidly assess the effect of 22,277 genes on breast cancer prognosis using microarray data of 1,809 patients. *Breast Cancer Res. Treat.* 123, 725–731 (2010).
289. Brock, G. clValid : An R Package for Cluster Validation. *J. Stat. Softw.* 25, (2008).
290. Kamburov, A., Stelzl, U., Lehrach, H. & Herwig, R. The ConsensusPathDB interaction database: 2013 Update. *Nucleic Acids Res.* 41, 793–800 (2013).
291. Zambelli, F., Pesole, G. & Pavesi, G. Pscan: Finding over-represented transcription factor binding site motifs in sequences from co-regulated or co-expressed genes. *Nucleic Acids Res.* 37, 247–252 (2009).
292. Nguyen, P. L. et al. Breast cancer subtype approximated by estrogen receptor, progesterone receptor, and HER-2 is associated with local and distant recurrence after breast-conserving therapy. *J. Clin. Oncol.* 26, 2373–2378 (2008).
293. Bentzon, N., Düring, M., Rasmussen, B. B., Mouridsen, H. & Kroman, N. Prognostic effect of estrogen receptor status across age in primary breast cancer. *Int. J. Cancer* 122, 1089–1094 (2008).
294. Zhidkova, N. I., Belkin, A. M. & Mayne, R. Novel isoform of beta 1 integrin expressed in skeletal and cardiac muscle. *Biochemical and Biophysical Research Properties* 214, 279–285 (1995).
295. Van der Flier, A., Kuikman, I., Baudoin, C., Vanderneut, R. & Sonnenberg, A. A novel beta 1 integrin isoform produced by alternative splicing: Unique expression in cardiac and skeletal muscle. *FEBS Lett.* 369, 340–344 (1995).
296. Kim, J. H. et al. MCL-1ES, a novel variant of MCL-1, associates with MCL-1L and induces mitochondrial cell death. *FEBS Lett.* 583, 2758–2764 (2009).
297. Gov, E. & Arga, K. Y. Differential co-expression analysis reveals a novel prognostic gene module in ovarian cancer. *Sci. Rep.* 7, 1–10 (2017).
298. Cai, L. et al. Genomic regression analysis of coordinated expression. *Nat. Commun.* 8, 1–10 (2017).
299. Qin, S., Ma, F. & Chen, L. Gene regulatory networks by transcription factors and microRNAs in breast cancer. *Bioinformatics* 31, 76–83 (2015).
300. Bradner, J. E., Hnisz, D. & Young, R. A. Transcriptional Addiction in Cancer. *Cell* 168, 629–643 (2017).
301. Bleckmann, S. C. et al. Activating Transcription Factor 1 and CREB Are Important for Cell Survival during Early Mouse Development Activating Transcription Factor 1 and CREB Are Important for Cell Survival during Early Mouse Development. *Mol. Cell. Biol.* 22, 1919–1925 (2002).
302. Gyorffy, B., Surowiak, P., Budczies, J. & Lánczky, A. Online survival analysis software to assess the prognostic value of biomarkers using transcriptomic data in non-small-cell lung cancer. *PLoS One* 8, (2013).
303. Gyorffy, B., Lánczky, A. & Szállási, Z. Implementing an online tool for genomewide validation of survival-associated biomarkers in ovarian-cancer using microarray data from 1287 patients. *Endocr. Relat. Cancer* 19, 197–208 (2012).
304. Boroughs, L. K. & Deberardinis, R. J. Metabolic pathways promoting cancer cell survival and growth. *Nat. Cell Biol.* 17, 351–359 (2016).
305. Cairns, R. A. Drivers of the warburg phenotype. *Cancer J. (United States)* 21, 56–61 (2015).
306. LeBlue, V. S. et al. PGC-1alpha mediated mitochondrial biogenesis and oxidative phosphorylation to promote metastasis. *Nat Cell Biol* 16, 992–1015 (2004).
307. Witkiewicz, A. K. et al. Using the ‘reverse Warburg effect’ to identify high-risk breast cancer patients: Stromal MCT4 predicts poor clinical outcome in triple-negative breast cancers. *Cell Cycle* 11, 1108–1117 (2012).
308. Sotgia, F. et al. Mitochondrial metabolism in cancer metastasis Visualizing tumor cell mitochondria and the “ reverse Warburg effect ” in positive lymph node tissue. *Cell Cycle* 11, 1445–1454 (2012).
309. Mertins, P. et al. Proteogenomics connects somatic mutations to signaling in breast cancer. *Nature* 534, 55–62 (2016).
310. Kosti, I., Jain, N., Aran, D., Butte, A. J. & Sirota, M. Cross-tissue Analysis of Gene and Protein

- Expression in Normal and Cancer Tissues. *Sci. Rep.* 6, 1–16 (2016).
311. Yin, H.-L. *et al.* $\beta 1$ Integrin as a Prognostic and Predictive Marker in Triple-Negative Breast Cancer. *Int. J. Mol. Sci.* 17, 1432 (2016).
312. dos Santos, P. B., Zanetti, J. S., Ribeiro-Silva, A. & Beltrão, E. I. C. Beta 1 integrin predicts survival in breast cancer: a clinicopathological and immunohistochemical study. *Diagn. Pathol.* 7, 104 (2012).
313. Belkin, A. M. *et al.* $\beta 1D$ integrin displaces the $\beta 1A$ isoform in striated muscles: Localization at junctional structures and signaling potential in nonmuscle cells. *J. Cell Biol.* 132, 211–226 (1996).
314. Luo, M. & Guan, J.-L. Focal Adhesion Kinase: a Prominent Determinant in Breast Cancer Initiation, Progression and Metastasis. *Cancer Lett.* 289, 127–139 (2011).
315. Reddy, K. B., Nabha, S. M. & Atanaskova, N. Role of MAP kinase in tumor progression and invasion. *Cancer Metastasis Rev.* 22, 395–403 (2003).
316. Mousavi, S. A. *et al.* Assessing the prognostic factors, survival, and recurrence incidence of triple negative breast cancer patients, a single center study in Iran. *PLoS One* 14, 1–17 (2019).
317. James, M., Dixit, A., Robinson, B., Frampton, C. & Davey, V. Outcomes for Patients with Non-metastatic Triple-negative Breast Cancer in New Zealand. *Clin. Oncol.* 31, 17–24 (2019).
318. Jr, H. G. *et al.* Survival Study of Triple-Negative and Non-Triple- Negative Breast Cancer in a Brazilian Cohort. *Clin. Med. Insights* 12, 1–10 (2018).
319. Anders, C. K. & Carey, L. A. Biology, Metastatic Patterns, and Treatment of Patients with Triple-Negative Breast Cancer. *Clin. Breast Cancer* 9, S73–81 (2009).
320. Wolf, K. *et al.* Compensation mechanism in tumor cell migration: Mesenchymal-amoeboïd transition after blocking of pericellular proteolysis. *J. Cell Biol.* 160, 267–277 (2003).
321. Ye, X. & Weinberg, R. A. Epithelial-Mesenchymal Plasticity: A central regulator of cancer progression. *Trends Cell Biol.* 25, 675–686 (2015).
322. Wink, S. *et al.* Quantitative High Content Imaging of Cellular Adaptive Stress Response Pathways in Toxicity for Chemical Safety Assessment. *Chem. Res. Toxicol.* 27, 338–355 (2014).
323. Poser, I. *et al.* BAC TransgeneOmics: a high-throughput method for exploration of protein function in mammals. *Nat. Methods* 5, 409–415 (2008).
324. Hiemstra, S. *et al.* Comprehensive Landscape of Nrf2 and p53 Pathway Activation Dynamics by Oxidative Stress and DNA Damage. *Chem. Res. Toxicol.* 30, 923–933 (2017).
325. Will, C. L. & Lu, R. Spliceosome Structure and Function. *Cold Spring Harb Perspect Biol* 1–23 (2011).
326. Jeromin, S. *et al.* SF3B1 mutations correlated to cytogenetics and mutations in NOTCH1, FBXW7, MYD88, XPO1 and TP53 in 1160 untreated CLL patients. *Leukemia* 28, 108–117 (2014).
327. Furney, S. J. *et al.* SF3B1 mutations are associated with alternative splicing in uveal melanoma. *Cancer Discov.* 3, 1122–1129 (2013).
328. Shepard, P. J. & Hertel, K. J. Protein family review The SR protein family. *Genome Biol.* 10, 1–9 (2009).
329. Rozanov, D. V *et al.* Molecular signature of MT1-MMP: Transactivation of the downstream universal gene network in cancer. *Cancer Res.* 68, 4086–4096 (2008).
330. Saijo, S. *et al.* Serine/arginine-rich splicing factor 7 regulates p21-dependent growth arrest in colon cancer cells. *J. Med. Investigig.* 63, (2016).
331. Fu, Y. U. & Wang, Y. SRSF7 knockdown promotes apoptosis of colon and lung cancer cells. *Oncol. Lett.* 15, 5545–5552 (2018).
332. Olst, E. S. *et al.* A genome-wide siRNA screen for regulators of tumor suppressor p53 activity in human non-small cell lung cancer cells identifies components of the RNA splicing machinery as targets for anticancer treatment. *Mol. Oncol.* 11, 534–551 (2017).
333. Shirai, C. L. *et al.* Mutant U2AF1-expressing cells are sensitive to pharmacological modulation of the spliceosome. *Nat. Commun.* 8, (2017).
334. Jacob, A. G. & Smith, C. W. J. Intron retention as a component of regulated gene expression programs. *Hum. Genet.* 136, 1043–1057 (2017).
335. Ge, Y. & Porse, B. T. The functional consequences of intron retention: Alternative splicing coupled to NMD as a regulator of gene expression. *Bioessays* 36, 236–243 (2013).
336. Dvinge, H. & Bradley, R. K. Widespread intron retention diversifies most cancer transcriptomes. *Genome Med.* 7, 45 (2015).
337. Alphen, R. J. Van, Wiemer, E. A. C., Burger, H. & Eskens, F. The spliceosome as target for anticancer treatment. *Br. J. Cancer* 100, 228–232 (2009).
338. Sundaramoorthy, S., Vázquez-novelle, M. D., Lekomtsev, S., Howell, M. & Petronczki, M. Functional genomics identifies a requirement of pre-mRNA splicing factors for sister chromatid cohesion. *EMBO J.* 33, 2623–2642 (2014).

Appendix

339. Zanini, I. M. Y., Soneson, C., Lorenzi, L. E. & Azzalin, C. M. Human cactin interacts with DHX8 and SRRM2 to assure efficient pre-mRNA splicing and sister chromatid cohesion. *J. Cell Sci.* 130, 767–778 (2017).
340. Wink, S., Hiemstra, S., Herpers, B. & Water, B. Van De. High- content imaging- based BAC- GFP toxicity pathway reporters to assess chemical adversity liabilities. *Arch. Toxicol.* 91, 1367–1383 (2017).
341. Tyanova, S., Temu, T. & Cox, J. The MaxQuant computational platform for mass spectrometry – based shotgun proteomics. *Nat. Protoc.* 11, 2301–2319 (2016).
342. Perez-Riverol, Y. *et al.* The PRIDE database and related tools and resources in 2019: improving support for quantification data. *Nucleic Acids Res.* 47, 442–450 (2019).
343. Vichai, V. & Kirtikara, K. Sulforhodamine B colorimetric assay for cytotoxicity screening. *Nat. Protoc.* 1, 1112–1116 (2006).
344. Landau, D. A. *et al.* Evolution and impact of subclonal mutations in chronic lymphocytic leukemia. *Cell* 152, 714–726 (2013).
345. Sakaue-sawano, A. *et al.* Genetically Encoded Tools for Optical Dissection of the Mammalian Cell Cycle. *Mol. Cell* 68, 626–639.e5 (2017).
346. Lange, J. De *et al.* Defective sister chromatid cohesion is synthetically lethal with impaired APC/C function. *Nat. Commun.* 6, 1–12 (2015).
347. Peters, J. & Nishiyama, T. Sister Chromatid Cohesion. *Cold spring Harb. Perspect.* 1–18 (2012).
348. Leung, A. K. L. & Lamond, A. I. In vivo analysis of NHPX reveals a novel nucleolar localization pathway involving a transient accumulation in splicing speckles. *J. Cell Biol.* 157, 615–629 (2002).
349. Thakran, P. *et al.* Sde 2 is an intron-specific pre-mRNA splicing regulator activated by ubiquitin-like processing. *EMBO J.* 37, 89–101 (2018).
350. Sivaramakrishnan, M. *et al.* Binding to SMN2 pre-mRNA-protein complex elicits specificity for small molecule splicing modifiers. *Nat. Commun.* 8, (2017).
351. Shpargel, K. B. & Matera, A. G. Gemin proteins are required for efficient assembly of Sm-class ribonucleoproteins. *PNAS* 102, 17372–17377 (2005).
352. Lombardi, M. L. *et al.* The Interaction between Nesprins and Sun Proteins at the Nuclear Envelope Is Critical for Force Transmission between the Nucleus and Cytoskeleton. *J. Biol. Chem.* 286, 26743–26753 (2011).
353. May, C. K. & Carroll, C. W. Differential incorporation of SUN-domain proteins into LINC complexes is coupled to gene expression. *PLoS One* 84, 1–14 (2018).
354. Wang, M. H., Yao, H. P. & Zhou, Y. Q. Oncogenesis of RON receptor tyrosine kinase: A molecular target for malignant epithelial cancers. *Acta Pharmacol. Sin.* 27, 641–650 (2006).
355. Varas, J. *et al.* Absence of SUN1 and SUN2 proteins in Arabidopsis thaliana leads to a delay in meiotic progression and defects in synapsis and recombination. *Plant J.* 81, 329–346 (2015).
356. Turgay, Y. *et al.* SUN proteins facilitate the removal of membranes from chromatin during nuclear envelope breakdown. *J. Cell Biol.* 204, 1099–1109 (2014).
357. Watrin, E., Demidova, M., Watrin, T., Hu, Z. & Prigent, C. Sororin pre-mRNA splicing is required for proper sister chromatid cohesion in human cells. *EMBO Rep.* 15, 948–955 (2014).
358. Lelij, P. Van Der *et al.* SNW1 enables sister chromatid cohesion by mediating the splicing of sororin and APC2 pre-mRNAs. *EMBO J.* 33, 2643–2658 (2014).
359. Oka, Y. *et al.* UBL 5 is essential for pre-mRNA splicing and sister chromatid cohesion in human cells. *EMBO Rep.* 15, 956–964 (2014).
360. Marcotte, R. *et al.* Essential Gene Profiles in Breast, Pancreatic, and Ovarian Cancer Cells. *Cancer Discov.* 173–189 (2012). doi:10.1158/2159-8290.CD-11-0224
361. Kim, J. *et al.* Cohesin interacts with a panoply of splicing factors required for cell cycle progression and genomic organization. (2018).
362. Matsumoto, A. *et al.* Global loss of a nuclear lamina component, lamin A/C, and LINC complex components SUN1, SUN2, and nesprin- 2 in breast cancer. *Cancer Med.* 1547–1557 (2015). doi:10.1002/cam4.495
363. Matsumoto, A. *et al.* Loss of the integral nuclear envelope protein SUN1 induces alteration of nucleoli. *Nucleus* 7, 68–83 (2016).
364. Aouida, M., Eid, A. & Mahfouz, M. M. CRISPR/Cas9-mediated target validation of the splicing inhibitor Pladienolide B. *Biochim. Open* 3, 72–75 (2016).
365. Kashyap, M. K. *et al.* Targeting the spliceosome in chronic lymphocytic leukemia with the macrolides FD-895 and pladienolide-B. *Chronic Lymphocytic Leuk.* 100, 945–954 (2015).
366. Sato, M. *et al.* High antitumor activity of pladienolide B and its derivative in gastric cancer. *Cancer Sci.* 105, 110–116 (2014).
367. American Cancer Society. *Cancer Facts and Figures*. (2017).

368. Anders, C. & Carey, L. Understanding and Treating Triple-Negative Breast Cancer. *Oncology* 22, 1233–1243 (2008).
369. Yao, H. et al. Triple-negative breast cancer: is there a treatment on the horizon? *Oncotarget* 8, 1913–1924 (2017).
370. Di, Z. et al. Ultra High Content Image Analysis and Phenotype Profiling of 3D Cultured Micro-Tissues. *PLoS One* 9, 1–10 (2014).
371. Barretina, J. et al. The Cancer Cell Line Encyclopedia enables predictive modeling of anticancer drug sensitivity. *Nature* 483, 603–607 (2012).
372. Gale, M. et al. Screen-identified selective inhibitor of lysine demethylase 5A blocks cancer cell growth and drug resistance. *Oncotarget* 7, (2016).
373. Garnett, M. J. et al. Systematic identification of genomic markers of drug sensitivity in cancer cells. *Nature* 483, 570–575 (2012).
374. Jansen, V. M. et al. Kinome-Wide RNA Interference Screen Reveals a Role for PDK1 in Acquired Resistance to CDK4/6 Inhibition in ER-Positive Breast Cancer. *Cancer Res.* 77, 2488–2499 (2017).
375. Thrane, S. et al. A kinase inhibitor screen identifies Mcl-1 and Aurora kinase A as novel treatment targets in antiestrogen-resistant breast cancer cells. *Oncogene* 34, 4199–4210 (2015).
376. Vora, S. R. et al. CDK 4/6 Inhibitors Sensitize PIK3CA Mutant Breast Cancer to PI3K Inhibitors. *Cancer Cell* 26, 136–149 (2014).
377. Sachs, N. et al. A Living Biobank of Breast Cancer Organoids Captures Disease Heterogeneity. *Cell* 172, 373–382 (2018).
378. Jabs, J. et al. Screening drug effects in patient-derived cancer cells links organoid responses to genome alterations. *Mol Syst Biol* 13, 1–16 (2017).
379. Wong, A. H. et al. Drug screening of cancer cell lines and human primary tumors using droplet microfluidics. *Sci. Rep.* 7, 1–15 (2017).
380. Bruna, A. et al. A Biobank of Breast Cancer Explants with Preserved Intra-tumor Heterogeneity to Screen Anticancer Compounds. *Cell* 167, 260–274.e22 (2016).
381. Fang, Y. & Eglen, R. M. Three-Dimensional Cell Cultures in Drug Discovery and Development. *SLAS Discov.* 22, 456–472 (2017).
382. Gao, H. et al. High-throughput screening using patient-derived tumor xenografts to predict clinical trial drug response. *Nat. Med.* 21, (2015).
383. Hidalgo, M. et al. A Pilot Clinical Study of Treatment Guided by Personalized Tumorgrafts in Patients with Advanced Cancer. *Mol. Cancer Ther.* 10, 1311–1317 (2011).
384. Katt, M., Placone, A., Wong, A., Xu, Z. & Searson, P. In Vitro Tumor Models: Advantages , Disadvantages , variables , and Selecting the Right Platform. *Front. Bioeng. Biotechnol.* 4, 1–14 (2016).
385. Zanoni, M. et al. 3D tumor spheroid models for in vitro therapeutic screening: a systematic approach to enhance the biological relevance of data obtained. *Sci. Rep.* 6, 1–11 (2016).
386. Drost, J. & Clevers, H. Organoids in cancer research. *Nat. Rev. Cancer* 18, 407–418 (2018).
387. Wetering, M. Van De et al. Prospective Derivation of a Living Organoid Biobank of Colorectal Cancer Patients. *Cell* 161, 933–945 (2015).
388. Zhang, Y. et al. Elevated insulin-like growth factor 1 receptor signaling induces antiestrogen resistance through the MAPK/ERK and PI3K/Akt signaling routes. *Breast Cancer Res.* 13, 1–16 (2011).
389. Neve, R. M. et al. A collection of breast cancer cell lines for the study of functionally distinct cancer subtypes. *Cancer Cell* 10, 515–527 (2006).
390. Prat, A. et al. Characterization of cell lines derived from breast cancers and normal mammary tissues for the study of the intrinsic molecular subtypes. *Breast Cancer Res Treat* 142, 237–255 (2013).
391. Hollestelle, A. et al. Distinct gene mutation profiles among luminal-type and basal-type breast cancer cell lines. *Breast Cancer Res Treat* 121, 53–64 (2010).
392. Kao, J. et al. Molecular Profiling of Breast Cancer Cell Lines Defines Relevant Tumor Models and Provides a Resource for Cancer Gene Discovery. *PLoS One* 4, 1–16 (2009).
393. Charafe-Jauffret, E. et al. Gene expression profiling of breast cell lines identifies potential new basal markers . *Oncogene* 25, 2273–2284 (2006).
394. Qi, Y. & Xu, R. Roles of PLODs in Collagen Synthesis and Cancer Progression. *Front. cell Dev. Biol.* 6, 1–8 (2018).
395. Mayer, E. L. Targeting Breast Cancer with CDK Inhibitors. *Curr Oncol Rep* 17, 15–19 (2015).
396. Tang, A. et al. Aurora kinases: novel therapy targets in cancers. *Oncotarget* 8, 23937–23954 (2017).

Appendix

397. Bhullar, K. S. *et al.* Kinase-targeted cancer therapies: progress , challenges and future directions. *Mol. Cancer* 17, 1–20 (2018).
398. Cicenas, J. The Aurora kinase inhibitors in cancer research and therapy. *J. Cancer Res. Clin. Oncol.* 142, 1995–2012 (2016).
399. Deshaies, R. J. Proteotoxic crisis , the ubiquitin-proteasome system , and cancer therapy. *BMC Biol.* 12, 1–14 (2014).
400. Adams, J. THE PROTEASOME: A SUITABLE ANTINEOPLASTIC TARGET. *Nat. Rev. Cancer* 4, 349–360 (2004).
401. Sun, Y. *et al.* Effects of an Indolocarbazole-Derived CDK4 Inhibitor on Breast Cancer Cells. *J. Cancer* 2, 36–51 (2011).
402. Choi, J. E. *et al.* Combined treatment with ABT-737 and VX-680 induces apoptosis in Bcl-2- and c-FLIP-overexpressing breast carcinoma cells. *Oncol. Rep.* 33, 1395–1401 (2015).
403. Lamb, R. *et al.* Cell cycle regulators cyclin D1 and CDK4/6 have estrogen receptor-dependent divergent functions in breast cancer migration and stem cell-like activity. *Cell Cycle* 12, 2384–2394 (2013).
404. Maiello, M. R. *et al.* EGFR and MEK Blockade in Triple Negative Breast Cancer Cells. *J. Cell. Biochem.* 2778–2785 (2015). doi:10.1002/jcb.25220
405. Zhang, M. *et al.* Prognostic value of survivin and EGFR protein expression in triple-negative breast cancer (TNBC) patients. *Targ Oncol* 9, 349–357 (2014).
406. Nakai, K., Hung, M. & Yamaguchi, H. A perspective on anti-EGFR therapies targeting triple-negative breast cancer. *Am J Cancer Res* 6, 1609–1623 (2016).
407. Savage, P., Blanchet-cohen, A., Kleinman, C. L., Park, M. & Rogoussis, J. A Targetable EGFR-Dependent Tumor-Initiating Program in Breast Cancer. *Cell Rep.* 21, 1140–1149 (2017).
408. Wang, X. *et al.* EGFR signaling promotes inflammation and cancer stem-like activity in inflammatory breast cancer. *Oncotarget* 8, 67904–67917 (2017).
409. Zheng, Z. *et al.* Correlation between epidermal growth factor receptor and tumor stem cell markers CD44/CD24 and their relationship with prognosis in breast invasive ductal carcinoma. *Med. Oncol.* 32, 1–11 (2015).
410. Ramaiahgari, S. C. *et al.* A 3D in vitro model of differentiated HepG2 cell spheroids with improved liver- like properties for repeated dose high- throughput toxicity studies. *Arch. Toxicol.* 88, 1083–1095 (2014).
411. Ashton, T. M., Mckenna, W. G., Kunz-schughart, L. A. & Higgins, G. S. Oxidative Phosphorylation as an Emerging Target in Cancer Therapy. *Clin. Cancer Res.* 24, 2482–2491 (2018).
412. Riaz, N. *et al.* Mitochondrial DNA copy number variation across human cancers. *Elife* 5, 1–20 (2016).
413. Weerts, M. J. A. *et al.* Low Tumor Mitochondrial DNA Content Is Associated with Better Outcome in Breast Cancer Patients Receiving Anthracycline-Based Chemotherapy. *Clin. Cancer Res.* 23, 4735–4744 (2017).
414. Weerts, M. J. A. *et al.* Mitochondrial DNA content in breast cancer: Impact on in vitro and in vivo phenotype and patient prognosis. *Oncotarget* 7, (2016).
415. Stone, H. R. & Morris, J. R. DNA damage emergency: cellular garbage disposal to the rescue ? *Oncogene* 33, 805–813 (2014).
416. Chen, S. *et al.* Genome-Wide siRNA Screen for Modulators of Cell Death Induced by Proteasome Inhibitor Bortezomib. *Cancer Res.* 70, 4318–4327 (2010).
417. Lovitt, C. J., Shelper, T. B. & Avery, V. M. Doxorubicin resistance in breast cancer cells is mediated by extracellular matrix proteins. *BMC Cancer* 18, 1–11 (2018).
418. Breslin, S. & Driscoll, L. O. The relevance of using 3D cell cultures , in addition to 2D monolayer cultures , when evaluating breast cancer drug sensitivity and resistance. *Oncotarget* 7, (2016).
419. Gangadhara, S., Smith, C., Barrett-lee, P. & Hiscox, S. 3D culture of Her2+ breast cancer cells promotes AKT to MAPK switching and a loss of therapeutic response. *BMC Cancer* 16, 1–12 (2016).
420. Whittle, J. R., Lewis, M. T., Lindeman, G. J. & Visvader, J. E. Patient-derived xenograft models of breast cancer and their predictive power. *Breast Cancer Res.* 17, (2015).
421. Papapetrou, E. P. Patient-derived induced pluripotent stem cells in cancer research and precision oncology. *Nat Med* 22, 1392–1401 (2016).
422. Tsai, H., Trubelja, A., Shen, A. Q. & Bao, G. Tumour-on-a-chip: microfluidic models of tumour morphology , growth and microenvironment. *R. Soc. Publ.* (2017).
423. Gounaris, I. Survival of patients with metastatic breast cancer with or without locoregional therapy. *Lancet Oncol.* 16, e585–e586 (2015).
424. Tzeng, Y. T. *et al.* Kinome-Wide siRNA Screening Identifies Src-Enhanced Resistance of

- Chemotherapeutic Drugs in Triple-Negative Breast Cancer Cells. *Front. Pharmacol.* 9, 1–11 (2018).
425. Jackson, A. L. *et al.* Expressing profiling reveals off-target gene regulation by RNAi. *Nat. Biotechnol.* 21, 635–638 (2003).
426. Doudna, J. A. & Charpentier, E. The new frontier of genome engineering with CRISPR-Cas9. *Science (80-.)*, 346, (2014).
427. Jinek, M. *et al.* A programmable dual RNA-guided DNA endonuclease in adaptive bacterial immunity. *Science (80-.)*, 337, 816–821 (2012).
428. Duda, K. *et al.* High-efficiency genome editing via 2A-coupled co-expression of fluorescent proteins and zinc finger nucleases or CRISPR/Cas9 nickase pairs. *Nucleic Acids Res.* 42, 1–16 (2014).
429. Liang, X. *et al.* Rapid and highly efficient mammalian cell engineering via Cas9 protein transfection. *J. Biotechnol.* 208, 44–53 (2015).
430. Swiech, L. *et al.* In vivo interrogation of gene function in the mammalian brain using CRISPR-Cas9. *Nat. Biotechnol.* 33, 102–106 (2015).
431. Wang, T., Wei, J. J., Sabatini, D. M. & Lander, E. S. Genetic screens in human cells using the CRISPR/Cas9 system. *Science (80-.)*, 343, 80–84 (2014).
432. Koike-yusa, H., Li, Y., Tan, E., Velasco-herrera, M. D. C. & Yusa, K. Genome-wide recessive genetic screening in mammalian cells with a lentiviral CRISPR-guide RNA library. *Nat. Biotechnol.* 32, 267–276 (2014).
433. Shalem, O. *et al.* Genome-Scale CRISPR-Cas9 Knockout Screening in Human Cells. *Science (80-.)*, 343, 84–87 (2014).
434. Gilbert, L. A. *et al.* Genome-Scale CRISPR-Mediated Control of Gene Repression and Activation. *Cell* 159, 647–661 (2014).
435. Sharma, S. & Petsalaki. Application of CRISPR-Cas9 Based Genome-Wide Screening Approaches to Study Cellular Signalling Mechanisms. *Int. J. Mol. Sci.* 19, 1–14 (2018).
436. Unniyampurath, U., Pilankatta, R. & Krishnan, M. N. RNA Interference in the Age of CRISPR: Will CRISPR Interfere with RNAi? *Int. J. Mol. Sci.* 17, (2016).
437. Tajadura-ortega, V. *et al.* An RNAi screen of Rho signalling networks identifies RhoH as a regulator of Rac1 in prostate cancer cell migration. *BMC Biol.* 16, 1–20 (2018).
438. Smolen, G. A. *et al.* A genome-wide RNAi screen identifies multiple RSK-dependent regulators of cell migration. *Genes Dev.* 24, 2654–2665 (2010).
439. Williams, S. P. *et al.* Data Descriptor: Systematic screens of endothelial cell migration and morphology. *Sci. Data* 4, 1–11 (2017).
440. De Pril, R., Perera, T. & Lekkerkerker, A. A high-content screen for inhibitors of cell migration in cancer metastasis using adenoviral knock-down. *Biotech Int* 21, (2009).
441. Edmondson, R., Broglie, J. J., Adcock, A. F. & Yang, L. Three-Dimensional Cell Culture Systems and Their Applications in Drug Discovery and Cell-Based Biosensors. *Assay Drug Dev. Technol.* 12, 207–218 (2014).
442. Duval, K. *et al.* Modeling Physiological Events in 2D vs. 3D Cell Culture. *Physiology* 32, 266–277 (2017).
443. Mseka, T., Bamburg, J. R. & Cramer, L. P. ADF/cofilin family proteins control formation of oriented actin-filament bundles in the cell body to trigger fibroblast polarization. *J. Cell Sci.* 120, 4332–4344 (2007).
444. Kapałczyńska, M. *et al.* 2D and 3D cell cultures – a comparison of different types of cancer cell cultures. *Arch Med Sci* 14, 910–919 (2018).
445. Friedl, P., Bröcker, E. & Zänker, K. S. Integrins, Cell Matrix Interactions and Cell Migration Strategies: Fundamental Differences in Leukocytes and Tumor Cells. *Cell Adhes. Commun.* 6, 225–236 (1998).
446. Lindström, S. *et al.* Genome-wide association study identifies multiple loci associated with both mammographic density and breast cancer risk. *Nat. Commun.* 5, (2015).
447. Michailidou, K., Lindström, S., Dennis, J., Beesley, J. & Hui, S. Association analysis identifies 65 new breast cancer risk loci. *Nature* 551, (2017).
448. Kozlovski, I., Siegfried, Z., Amar, A. & Rotem, S. The role of RNA alternative splicing in regulating cancer metabolism. *Hum. Genet.* 136, 1113–1127 (2017).
449. Narayanan, S. P., Singh, S. & Shukla, S. A saga of cancer epigenetics: linking epigenetics to alternative splicing. *Biochem. J.* 474, 885–896 (2017).
450. Luco, R., Allo, M., Schor, I., Kornblith, A. & Misteli, T. Epigenetics in alternative pre-mRNA splicing. *Cell* 144, 16–26 (2011).
451. Boutz, P. L., Bhutkar, A. & Sharp, P. A. Detained introns are a novel , widespread class of post-

Appendix

- transcriptionally spliced introns. *Genes Dev.* 29, 63–80 (2015).
452. Braun, C. J. *et al.* Coordinated Splicing of Regulatory Detained Introns within Oncogenic Transcripts Creates an Exploitable Vulnerability in Malignant Glioma Article Coordinated Splicing of Regulatory Detained Introns within Oncogenic Transcripts Creates an Exploitable Vulne. *Cancer Cell* 32, 411–426 (2017).
453. Fu, X. Exploiting the Hidden Treasure of Detained Introns. *Cancer Cell* 32, 393–395 (2017).

Abbreviations

2D	Two-dimensional
3D	Three-dimensional
A3SS	Alternative 3' splice site
A5SS	Alternative 5' splice site
AR	Androgen receptor
AS	Alternative splicing
ATF1	Activating transcription factor 1
BAC	Bacterial artificial chromosome
BC	Breast cancer
BSA	Bovine serum albumin
CDK	Cyclin dependent kinase
CLL	Chronic lymphocytic leukemia
CREB1	CAMP responsive element binding protein 1
CREM	Cyclic AMP response element modulatory protein
DDR	DNA damage response
DEG	Differentially expressed gene
DMFS	Distant metastasis-free survival
ECM	Extracellular matrix
EGFR	Epidermal growth factor receptor
EMT	Epithelial-to-mesenchymal transition
ER	Estrogen receptor
FACS	Fluorescence-activated cell sorting
FAK	Focal adhesion kinase
FC	Fold change
FDR	False discovery rate
GFP	Green fluorescent protein
GLM	Generalized linear model
GSEA	Gene set enrichment analysis
HER2	Human epidermal growth factor receptor 2
HIS	Human invasion signature
HMEC	Human mammary epithelial cell
hnRNPs	Heterogeneous nuclear ribonucleoproteins
HR	Hormone receptor
HR	Hazard ratio
HRP	Horseradish peroxidase
IDR	Image data resource
IR	Intron retention
ITGB1	Integrin β 1
KM	Kaplan Meier
KP	Kinasepool
LINC	Linkers of nucleoskeleton and cytoskeleton
LMS	Lung metastasis signature
MAPK	Mitogen activated protein kinase
MDS	Myelodysplastic syndrome
MET	Mesenchymal-to-epithelial transition
MFS	Metastasis-free survival
MXE	Mutually exclusive exon

A

Appendix

NGS	Next generation sequencing
NHEJ	Non-homologous end-joining
Par	Partitioning-defective
PB	Pladienolide B
PC	Pearson correlation coefficient
PCA	Principal component analysis
PDX	Patient-derived xenograft
PFA	Paraformaldehyde
PI3K	Phosphoinositide 3 kinase
PKM1	Pyruvate kinase M1
PKM2	Pyruvate kinase M2
PKT	Phagokinetic track
PPI	Protein-protein interaction
PR	Progesteron receptor
pre-mRNA	pre-mature messenger RNA
PXN	Paxillin
RBM4	RNA-binding motif protein 4
RCM	Random cell migration
RRM	RNA recognition motif
RT	Room temperature
SCC	Sister chromatid cohesion
SDS	Sodium dodecyl sulfate
SE	Exon skipping
SF	Splicing factor
sgRNA	single guide RNA
siRNA	Small interference RNA
snRNA	small nuclear RNA
snRNP	small nuclear ribonucleoproteins
SR	Serine/arginine-rich
SRB	Sulforhodamine B
SRPK	SR-rich protein-specific kinase
SRPK1	SRSF protein kinase 1
SRSF1	Serine and arginine splicing factor 1
STR	Short tandem repeat
TCGA	The cancer genome atlas
TNBC	Triple-negative breast cancer
VEGF	Vascular endothelial growth factor