Published in final edited form as:

Cancer Lett. 2013 May 28; 332(2): 225–228. doi:10.1016/j.canlet.2012.03.005.

TARGETING SURVIVIN IN CANCER

Dario C. Altieri

Prostate Cancer Discovery and Development Program, The Wistar Institute, Philadelphia, PA 19104, U.S.A

Abstract

With almost 4,000 citations in Medline in a little over ten years, survivin has certainly kept scores of investigators busy worldwide. Tangible progress has been made in revealing the multiple functions of survivin, uncovering their wirings as integrated cellular networks, and mapping their exploitation in virtually every human tumor, *in vivo*. Considering the normally long and excruciating timeline of oncology drug discovery, it is clearly a resounding success that a better understanding of survivin biology has led to several clinical trials of survivin-based therapeutics in cancer patients. However, the portfolio of survivin antagonists available in the clinic remains small, pressing the need for a less rigid drug development approach to fully unlock the potential of this unique, albeit unconventional oncology drug target.

Keywords

Survivin; IAP; cancer therapy; signaling networks; small molecule; antisense

1. Overview of survivin

This contribution is not designed to provide a full review on survivin. Comprehensive articles on these topics have been published, which may provide readers with a more in-depth perspective [1,2,3,4,5,6,7]. However, a brief summary of the biology of survivin is presented below to help frame the challenges and opportunities associated with survivin-based therapeutics in cancer.

Survivin is the smallest member of the Inhibitor of Apoptosis (IAP) gene family [8], containing a single Baculovirus IAP Repeat (BIR), which is the hallmark of these molecules. Although a homodimer by X-ray crystallography [8], survivin can function as a monomer for at least some protein-protein interactions, as well as mechanisms of subcellular localization [9]. The control of *survivin* gene expression at the tip of chromosome 17 in humans (17q25) is complex, and involves scores of positive and negative regulators. Several oncogenic transcription factors stimulate expression of the *survivin* gene, whereas multiple tumor suppressors actively repress the *survivin* gene [10]. The dynamic interplay in this transcriptional network controls a unique feature of survivin, namely its sharp differential

© 2012 Elsevier Ireland Ltd. All rights reserved.

Correspondence: Dario C. Altieri, M.D., The Wistar Institute Cancer Center, 3601 Spruce Street, Philadelphia, PA 19104, Tel. (215) 495-6970; FAX (215) 495-6863; daltieri@wistar.org.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

CONFLICT OF INTEREST STATEMENT

The author declares that no conflict of interest exists.

expression in cancer, as opposed to normal tissues, *in vivo* [6,7]. Transcription of the *survivin* gene is strictly cell cycle-regulated, and peaks at mitosis [1]. However, examples of cell cycle-independent *survivin* gene expression have been described, especially in response to growth factor or cytokine stimulation [6]. A survivin protein is extensively post-translationally modified by degradative and non-degradative cycles of ubiquitylation and de-ubiquitylation, as well as phosphorylation [6], which control protein stability, binding to molecular partners and trafficking to various subcellular compartments.

Survivin is a multifunctional protein and is *essential*, in that constitutive or conditional deletion of the *survivin* gene is incompatible with tissue or organism viability [6]. Orthologs of survivin have been found in lower organisms, such as yeast, worms, and flies, suggesting evolutionary conservation of this pathway. In mammalian cells, survivin participates in at least three homeostatic networks: the control of mitosis (1), the regulation of apoptosis (2), and the cellular stress response (3). This classification is not restrictive, as novel functions of surviving are continuously proposed, as well as new roles for known properties. Even within the same network, survivin plays multiple roles. For instance, at mitosis, survivin acts as a passenger protein [11] for proper chromosomal alignment, controls chromatin-associated spindle formation [12], enhances spindle stability via suppression of microtubule dynamics [13], and oversees kinetochore–microtubule attachment [14] in the spindle assembly checkpoint.

As a cytoprotective molecule [6,7], survivin, like all other IAPs except XIAP [8], does not directly inhibit caspases, but interacts with protein partners, most notably XIAP [15]. This complex promotes increased XIAP stability against degradation, activates multiple signaling pathways, including NF κ B, synergistically inhibits caspase-3 and -9, suppresses apoptosis, and accelerates tumor progression, *in vivo*. Other mechanisms of survivin cytoprotection have been proposed, including the ability of mitochondrial-localized pool of survivin to sequester pro-apoptotic Smac away from XIAP [16], or altogether prevent its release from mitochondria [17].

A third function of survivin in the cellular stress response is beginning to emerge, and involves the association of survivin with various molecular chaperones, including the aryl hydrocarbon receptor-interacting protein (AIP) [18], Hsp60 [19], and Hsp90 [20]. These interactions may promote adaptation under conditions of cellular stress by maintaining survivin protein stability, folding, and subcellular trafficking, *in vivo*.

2. Targeting survivin in cancer

Because of its nodal properties, and over-expression in virtually every human tumor, survivin has been intensely pursued as a drug target for novel cancer therapeutics [5,6,7]. Consistent with its multifunctional roles, the presence of survivin in cancer has been universally linked to resistance to apoptosis, metastasis, bypass of cell cycle checkpoints, and resistance to therapy. This has been further bolstered by molecular profiling studies and retrospective analyses of patient cohorts, which have consistently identified survivin as a ‘risk factor’ for poor prognosis and disease recurrence [21]. Against this backdrop, putative survivin antagonists would not only function as single-gene inhibitors, but act more broadly as ‘pathway antagonists’ [22], disabling the multiple survivin networks, and thus ideally suited to overcome tumor heterogeneity, *in vivo*.

2.1 - Molecular antagonists

The first described molecular antagonist of survivin is a phosphorothioate antisense oligonucleotide [23], which suppressed survivin mRNA and protein expression, and produced strong anticancer activity in preclinical models. Sponsored by Ely Lilly and Co.,

this agent, designated LY2181308 showed a favorable toxicity profile with evidence of survivin downregulation in a phase I trial in patients with advanced cancers [24]. A locked nucleic acid antisense oligonucleotide to survivin has also been developed by Santaris Pharma, and, more recently, ENZON Pharmaceuticals [25]. A phase I clinical trial with this agent, designated SPC3042 (or ENZ3042), also showed excellent safety [26]. Additional strategies to acutely lower survivin levels in tumor cells involved small interfering RNA (siRNA) or hammerhead ribozymes [27]. In preclinical studies, these agents consistently showed anticancer activity, alone or in combination with chemotherapy, with no detectable systemic toxicity [27]. The delivery of siRNA *in vivo* is challenging, but the apparent success of recent studies [28] suggests that *survivin*-directed gene silencing may at some point be evaluable in cancer patients.

2.2 - Small molecules

Small molecules that directly target survivin have been developed [6], and several clinical trials with these agents have been completed, with more underway. The most extensively studied agent in this class is YM155 [29]. Originally developed by Yamanouchi Pharmaceuticals, and, more recently, Astellas Pharma, YM155 was identified in high-throughput screening for inhibitors of *survivin* promoter activity [29]. This effect was specific for the *survivin* gene, and resulted in potent anticancer activity in preclinical models [29]. Two phase I studies of YM155 in heavily pretreated cancer patients have been published. The trial conducted in the US reported impressive responses, with tumor shrinkage and durable remissions in patients with advanced prostate cancer, large cell non-Hodgkin's lymphoma and non-small cell lung cancer [30]. The Japan phase I trial of YM155 also provided evidence of disease stabilization in nine patients [31]. Importantly, both studies showed a favorable toxicity profile with minimal and rapidly reversible side effects. Two phase II studies of YM155 monotherapy have been recently published. In patients with advanced non small cell lung cancer, YM155 produced two partial responses and disease stabilization in 14 out of 37 evaluable patients, corresponding to a disease control rate of 43% [32]. Consistent with the phase I data, the treatment was well tolerated with the majority of treatment discontinuations not treatment related [32]. The results of a phase II study of YM155 in melanoma were less encouraging. Although well tolerated, YM155 produced only one partial response in patients with stage III and stage IV disease, failing to meet the pre-specified endpoint of two responses in 29 evaluable patients [33].

Another direct small molecule inhibitor of survivin is tetra-O-methyl nordihydroguaiaretic acid (M(4)N), which also acts as a transcriptional repressor of the *survivin* promoter, potentially by antagonizing Sp1-dependent gene expression [34]. This compound, designated Terameprecol (EM-1421) [35] has shown good preclinical activity with an impressive 88% bioavailability, *in vivo* [36]. Terameprecol has been formulated for systemic delivery to cancer patients, and a phase I study in patients with advanced solid malignancies has shown favorable safety and disease stabilization in 8 out of 25 evaluable patients [35]. Another phase I study of Terapremecol in 16 heavily pretreated patients with adult myelogenous leukemia (AML) has also shown favorable safety, one partial response and disease stabilization in 5 patients [37]. In addition, Terameprecol has been formulated as a 1% or 2% vaginal ointment for local application in women with papillomavirus- or herpes simplex virus-associated carcinogenesis. Two phase I studies with Terameprecol ointment have shown excellent safety, no adverse events and no systemic absorption of the agent [38,39].

2.3 - Cancer vaccine/immunotherapy

Because of its differential expression in cancer, as opposed to normal tissues, it has been hypothesized that cancer patients may recognize survivin as a non-self protein, and mount

an immune response to it [40]. This concept has been validated in the clinic, and sera from cancer patients contain antibodies [41], and cytolytic T cells against survivin [42]. This immune recognition has been mapped in detail [43,44], and dendritic cells pulsed with survivin peptides, survivin-containing tumor lysates or transduced/transfected with survivin, elicit cytolytic T cell responses and MHC-restricted anticancer activity *in vitro* [45,46], and in preclinical models [47]. Several phase I/II trials of survivin-directed immunotherapy have been reported [40,48]. In these studies, survivin-based vaccination was invariably safe, devoid of significant side-effects, and associated with antigen-specific immunologic responses. Although no objective responses were noted, two phase I/II trials with infusion of dendritic cells pulsed with survivin showed durable (>6 months) disease stabilization in 24% of melanoma patients [49], and 50% of renal cell carcinoma patients [50]. A phase I/II trial of systemic delivery of protamine-protected survivin mRNA in melanoma was also safe, produced detectable T cell responses, and achieved one complete response out of seven evaluable patients [51]. Anecdotal reports also suggest that survivin-based vaccination may be effective against metastatic disease [52].

3. Survivin-based therapeutics: what's ahead?

Given the herculean task of oncology drug discovery and development [53], moving from discovery to the clinic in ten years is undoubtedly a resounding success for the survivin field. The approaches described above remain promising, and their excellent safety profile bodes well for their continued evaluation in the clinic. There were concerns that the low but detectable level of survivin in normal tissues [54], and its essential role in stem cells [55], could compromise the exploitation of this target, *in vivo*. In reality, this has not occurred, which may reflect a mechanism of 'survivin addiction' [56] specifically by the tumor, or, alternatively, the existence of 'qualitative' differences in the wiring of survivin networks in transformed cells, as opposed to normal tissues [6].

For the next wave of survivin-based therapeutics, combination studies with cytotoxics or radiation appear as obvious choices. Hopefully, these protocols will take advantage of the better understanding of survivin biology accumulated over the past decade. For instance, *sequential* inhibition of survivin in tumor cells pretreated with taxanes, and thus arrested at mitosis with hyperpolarized microtubules, produces a much increased anticancer activity with no toxicity in preclinical models [57], so that a similar *sequential* combination strategy could be envisioned for survivin small molecule or antisense antagonists. Conversely, suppression of survivin has been invariably shown to sensitize disparate tumor cells to ionizing radiation, and this mechanism can be also readily exploited for novel radiotherapy protocols.

There may be also concrete opportunities to rationally combine survivin antagonists with various molecular agents. For instance, survivin is a target gene of various oncogenic signaling circuits, including STAT3, Akt, EGFR [6], Notch [58], and Wnt [59]. Although small molecule inhibitors of these pathways have produced modest gains in the clinic as monotherapy, it is possible that the combined inhibition of one of their critical downstream targets, i.e. surviving may enhance and broaden their efficacy, without compromising safety. Finally, because surviving is a target of both VEGF [60] and Ang-1 [61], and is critically required for endothelial cell viability [62], its inhibitors could be rationally incorporated in current anti-angiogenic regimens, for instance in patients with renal cell cancer or hepatocellular carcinoma receiving multi-kinase inhibitors.

Despite these hopeful opportunities, the reality is that the current portfolio of surviving antagonists remains small, and unlikely to fully unlock the potential of this target for novel cancer therapeutics. The multiple and likely redundant regulation of the *survivin* promoter in

tumor cells [10] poses formidable challenges for agents like YM155 or Terameprecol to durably shut off *survivin* gene expression in disparate and highly heterogeneous cellular contexts. Antisense molecules have been invariably plagued by issues of specificity, off-target effects or lack of efficacy, and siRNA-based therapeutics is still in its infancy, having to overcome fundamental hurdles in delivery, durable gene silencing, and, ultimately, long-term efficacy in a highly heterogeneous disease.

The lack of a larger portfolio of survivin antagonists likely reflects a parsimonious view that molecules that are not present on the cell surface, or lack inhibitable catalytic activity, are not good targets [22]. For survivin, crystallographic data reveal little in the way of potential druggable sites, typically structural ‘pockets’ of suitable geometry and hydrophobicity. Despite these challenges, the wealth of information collected in the past decade about survivin should open fresh opportunities for drug discovery. For instance, disruption of protein–protein interactions, especially those involving apoptosis regulators, is now feasible, and has generated agents with promising anticancer activity [63]. In this context, proof-of-principle agonists that disrupt the complex between survivin and other network components — for instance, Hsp90 — have been identified [64], and validated for anticancer activity with no toxicity for normal tissues in preclinical studies [64]. Accordingly, it may be possible to envision high-throughput screening strategies to identify small molecule inhibitors of the interaction between survivin and some of its critical partners, for instance Smac [65], XIAP [15], or members of the chromosomal passenger complex [9], which may generate agents with novel specificities and anticancer efficacy. What we have learned over the past decade is that even seemingly small perturbations in survivin protein turnover, post-translational modifications, subcellular localization or binding to molecular partners, has almost invariably catastrophic consequences for tumor cells, resulting in mitotic defects, cell cycle arrest, spontaneous apoptosis, loss of metastatic potential, and inhibition of tumor growth, *in vivo*. As for the survivin antagonists now in the clinic, such novel agents can also be expected to have broad activity in genetically disparate tumor types, a favorable toxicity profile, and synergistic efficacy with conventional or targeted anticancer regimens.

Acknowledgments

This work was supported by National Institutes of Health grants CA78810, CA140043, CA118005 and HL54131. The study sponsors had no role in the study design, in the collection, analysis and interpretation of data; in the writing of the manuscript; and in the decision to submit the manuscript for publication.

REFERENCES

1. Lens SM, Vader G, Medema RH. The case for Survivin as mitotic regulator. *Curr Opin Cell Biol.* 2006; 18:616–622. [PubMed: 16962308]
2. Li F, Brattain MG. Role of the Survivin gene in pathophysiology. *Am J Pathol.* 2006; 169:1–11. [PubMed: 16816356]
3. Kotwaliwale C, Biggins S. Microtubule capture: a concerted effort. *Cell.* 2006; 127:1105–1108. [PubMed: 17174890]
4. Stauber RH, Mann W, Knauer SK. Nuclear and cytoplasmic survivin: molecular mechanism, prognostic, and therapeutic potential. *Cancer Res.* 2007; 67:5999–6002. [PubMed: 17616652]
5. Ryan BM, O'Donovan N, Duffy MJ. Survivin: A new target for anti-cancer therapy. *Cancer Treat Rev.* 2009
6. Altieri DC. Survivin, cancer networks and pathway-directed drug discovery. *Nat Rev Cancer.* 2008; 8:61–70. [PubMed: 18075512]
7. Mita AC, Mita MM, Nawrocki ST, Giles FJ. Survivin: key regulator of mitosis and apoptosis and novel target for cancer therapeutics. *Clin Cancer Res.* 2008; 14:5000–5005. [PubMed: 18698017]
8. Srinivasula SM, Ashwell JD. IAPs: what's in a name? *Mol Cell.* 2008; 30:123–135. [PubMed: 18439892]

9. Jeyapakash AA, Klein UR, Lindner D, Ebert J, Nigg EA, Conti E. Structure of a Survivin-Borealin-INCENP Core Complex Reveals How Chromosomal Passengers Travel Together. *Cell*. 2007; 131:271–285. [PubMed: 17956729]
10. Guha M, Altieri DC. Survivin as a global target of intrinsic tumor suppression networks. *Cell Cycle*. 2009; 8:2708–2710. [PubMed: 19717980]
11. Ruchaud S, Carmena M, Earnshaw WC. Chromosomal passengers: conducting cell division. *Nat Rev Mol Cell Biol*. 2007; 8:798–812. [PubMed: 17848966]
12. Sampath SC, Ohi R, Leismann O, Salic A, Pozniakovski A, Funabiki H. The chromosomal passenger complex is required for chromatin-induced microtubule stabilization and spindle assembly. *Cell*. 2004; 118:187–202. [PubMed: 15260989]
13. Rosa J, Canovas P, Islam A, Altieri DC, Doxsey SJ. Survivin modulates microtubule dynamics and nucleation throughout the cell cycle. *Mol Biol Cell*. 2006; 17:1483–1493. [PubMed: 16407408]
14. Sandall S, Severin F, McLeod IX, Yates JR 3rd, Oegema K, Hyman A, Desai A. A Bir1-Sli15 complex connects centromeres to microtubules and is required to sense kinetochore tension. *Cell*. 2006; 127:1179–1191. [PubMed: 17174893]
15. Dohi T, Xia F, Altieri DC. Compartmentalized phosphorylation of IAP by protein kinase A regulates cytoprotection. *Mol Cell*. 2007; 27:17–28. [PubMed: 17612487]
16. Song Z, Yao X, Wu M. Direct interaction between survivin and Smac/DIABLO is essential for the anti-apoptotic activity of survivin during taxol-induced apoptosis. *J Biol Chem*. 2003; 278:23130–23140. [PubMed: 12660240]
17. Ceballos-Cancino G, Espinosa M, Maldonado V, Melendez-Zajgla J. Regulation of mitochondrial Smac/DIABLO-selective release by survivin. *Oncogene*. 2007; 26:7569–7575. [PubMed: 17546047]
18. Kang BH, Altieri DC. Regulation of survivin stability by the aryl hydrocarbon receptor-interacting protein. *J Biol Chem*. 2006; 281:24721–24727. [PubMed: 16807248]
19. Ghosh JC, Dohi T, Kang BH, Altieri DC. Hsp60 regulation of tumor cell apoptosis. *J Biol Chem*. 2008; 283:5188–5194. [PubMed: 18086682]
20. Fortugno P, Beltrami E, Plescia J, Fontana J, Pradhan D, Marchisio PC, Sessa WC, Altieri DC. Regulation of survivin function by Hsp90. *Proc Natl Acad Sci U S A*. 2003; 100:13791–13796. [PubMed: 14614132]
21. Paik S, Shak S, Tang G, Kim C, Baker J, Cronin M, Baehner FL, Walker MG, Watson D, Park T, Hiller W, Fisher ER, Wickerham DL, Bryant J, Wolmark N. A multigene assay to predict recurrence of tamoxifen-treated, node-negative breast cancer. *N Engl J Med*. 2004; 351:2817–2826. [PubMed: 15591335]
22. van der Greef J, McBurney RN. Innovation: Rescuing drug discovery: in vivo systems pathology and systems pharmacology. *Nat Rev Drug Discov*. 2005; 4:961–967. [PubMed: 16341061]
23. Li F, Ackermann EJ, Bennett CF, Rothmel AL, Plescia J, Tognin S, Villa A, Marchisio PC, Altieri DC. Pleiotropic cell-division defects and apoptosis induced by interference with survivin function. *Nat. Cell Biol*. 1999; 1:461–466. [PubMed: 10587640]
24. Molckovsky A, Siu LL. First-in-class, first-in-human phase I results of targeted agents: Highlights of the 2008 American Society of Clinical Oncology meeting. *J Hematol Oncol*. 2008; 1:20. [PubMed: 18959794]
25. Hansen JB, Fisker N, Westergaard M, Kjaerulff LS, Hansen HF, Thue CA, Rosenbohm C, Wissenbach M, Orum H, Koch T. SPC3042: a proapoptotic survivin inhibitor. *Mol Cancer Ther*. 2008; 7:2736–2745. [PubMed: 18790754]
26. Walker K, Padhiar M. AACR-NCI-EORTC--21st International Symposium. Molecular targets and cancer therapeutics--Part 1. *IDrugs*. 2010; 13:7–9. [PubMed: 20024838]
27. Pennati M, Folini M, Zaffaroni N. Targeting survivin in cancer therapy. *Expert Opin Ther Targets*. 2008; 12:463–476. [PubMed: 18348682]
28. Davis ME, Zuckerman JE, Choi CHJ, Seligson D, Tolcher A, Alabi CA, Yen Y, Heidel JD, Ribas A. Evidence of RNAi in humans from systemically administered siRNA via targeted nanoparticles. *Nature*. 2010; 464:1067–1070. [PubMed: 20305636]
29. Nakahara T, Takeuchi M, Kinoyama I, Minematsu T, Shirasuna K, Matsuhisa A, Kita A, Tominaga F, Yamanaka K, Kudoh M, Sasamata M. YM155, a Novel Small-Molecule Survivin

- Suppressant, Induces Regression of Established Human Hormone-Refractory Prostate Tumor Xenografts. *Cancer Res.* 2007; 67:8014–8021. [PubMed: 17804712]
30. Tolcher AW, Mita A, Lewis LD, Garrett CR, Till E, Daud AI, Patnaik A, Papadopoulos K, Takimoto C, Bartels P, Keating A, Antonia S. Phase I and pharmacokinetic study of YM155, a small-molecule inhibitor of survivin. *J Clin Oncol.* 2008; 26:5198–5203. [PubMed: 18824702]
 31. Satoh T, Okamoto I, Miyazaki M, Morinaga R, Tsuya A, Hasegawa Y, Terashima M, Ueda S, Fukuoka M, Ariyoshi Y, Saito T, Masuda N, Watanabe H, Taguchi T, Kakihara T, Aoyama Y, Hashimoto Y, Nakagawa K. Phase I study of YM155, a novel survivin suppressant, in patients with advanced solid tumors. *Clin Cancer Res.* 2009; 15:3872–3880. [PubMed: 19470738]
 32. Giaccone G, Zatloukal P, Roubec J, Floor K, Musil J, Kuta M, van Klaveren RJ, Chaudhary S, Gunther A, Shamsili S. Multicenter phase II trial of YM155, a small-molecule suppressor of survivin, in patients with advanced, refractory, non-small-cell lung cancer. *J Clin Oncol.* 2009; 27:4481–4486. [PubMed: 19687333]
 33. Lewis KD, Samlowski W, Ward J, Catlett J, Cranmer L, Kirkwood J, Lawson D, Whitman E, Gonzalez R. A multi-center phase II evaluation of the small molecule survivin suppressor YM155 in patients with unresectable stage III or IV melanoma. *Invest New Drugs.* 2009 Oct 15. Epub Ahead of print.
 34. Chang CC, Heller JD, Kuo J, Huang RC. Tetra-O-methyl nordihydroguaiaretic acid induces growth arrest and cellular apoptosis by inhibiting Cdc2 and survivin expression. *Proc Natl Acad Sci U S A.* 2004; 101:13239–13244. [PubMed: 15329416]
 35. Smolewski P. Terameprocol, a novel site-specific transcription inhibitor with anticancer activity. *IDrugs.* 2008; 11:204–214. [PubMed: 18311658]
 36. Park R, Chang CC, Liang YC, Chung Y, Henry RA, Lin E, Mold DE, Huang RC. Systemic treatment with tetra-O-methyl nordihydroguaiaretic acid suppresses the growth of human xenograft tumors. *Clin Cancer Res.* 2005; 11:4601–4609. [PubMed: 15958646]
 37. Zhu X, Ma Y, Liu D. Novel agents and regimens for acute myeloid leukemia :2009 ASH annual meeting highlights. *J. Hematol. Oncol.* 2010; 3:17. [PubMed: 20416083]
 38. Khanna N, Dalby R, Connor A, Church A, Stern J, Frazer N. Phase I clinical trial of repeat dose terameprocol vaginal ointment in healthy female volunteers. *Sex. Transm. Dis.* 2008; 35:577–582. [PubMed: 18418294]
 39. Khanna N, Dalby R, Tan M, Arnold S, Stern J, Frazer N. Phase I/II clinical safety studies of terameprocol vaginal ointment. *Gynecol Oncol.* 2007; 107:554–562. [PubMed: 17905420]
 40. Andersen MH, Sorensen RB, Schrama D, Svane IM, Becker JC, Thor Straten P. Cancer treatment: the combination of vaccination with other therapies. *Cancer Immunol Immunother.* 2008
 41. Rohayem J, Diestelkoetter P, Weigle B, Oehmichen A, Schmitz M, Mehlhorn J, Conrad K, Rieber EP. Antibody response to the tumor-associated inhibitor of apoptosis protein survivin in cancer patients. *Cancer Res.* 2000; 60:1815–1817. [PubMed: 10766164]
 42. Schmidt SM, Schag K, Muller MR, Weck MM, Appel S, Kanz L, Grunebach F, Brossart P. Survivin is a shared tumor-associated antigen expressed in a broad variety of malignancies and recognized by specific cytotoxic T cells. *Blood.* 2003; 102:571–576. [PubMed: 12576330]
 43. Casati C, Dalerba P, Rivoltini L, Gallino G, Deho P, Rini F, Belli F, Mezzanzanica D, Costa A, Andreola S, Leo E, Parmiani G, Castelli C. The apoptosis inhibitor protein survivin induces tumor-specific CD8+ and CD4+ T cells in colorectal cancer patients. *Cancer Res.* 2003; 63:4507–4515. [PubMed: 12907624]
 44. Idenoue S, Hirohashi Y, Torigoe T, Sato Y, Tamura Y, Hariu H, Yamamoto M, Kurotaki T, Tsuruma T, Asanuma H, Kanaseki T, Ikeda H, Kashiwagi K, Okazaki M, Sasaki K, Sato T, Ohmura T, Hata F, Yamaguchi K, Hirata K, Sato N. A potent immunogenic general cancer vaccine that targets survivin, an inhibitor of apoptosis proteins. *Clin Cancer Res.* 2005; 11:1474–1482. [PubMed: 15746049]
 45. Schmitz M, Diestelkoetter P, Weigle B, Schmachtenberg F, Stevanovic S, Ockert D, Rammensee HG, Rieber EP. Generation of survivin-specific CD8+ T effector cells by dendritic cells pulsed with protein or selected peptides. *Cancer Res.* 2000; 60:4845–4849. [PubMed: 10987296]

46. Pisarev V, Yu B, Salup R, Sherman S, Altieri DC, Gabrilovich DI. Full-Length Dominant-Negative Survivin for Cancer Immunotherapy. *Clin Cancer Res.* 2003; 9:6523–6533. [PubMed: 14695157]
47. Siegel S, Wagner A, Schmitz N, Zeis M. Induction of antitumor immunity using survivin peptide-pulsed dendritic cells in a murine lymphoma model. *Br J Haematol.* 2003; 122:911–914. [PubMed: 12956760]
48. Friedrichs B, Siegel S, Andersen MH, Schmitz N, Zeis M. Survivin-derived peptide epitopes and their role for induction of antitumor immunity in hematological malignancies. *Leuk Lymphoma.* 2006; 47:978–985. [PubMed: 16840186]
48. Trepiaakas R, Berntsen A, Hadrup SR, Bjorn J, Geertsen PF, Straten PT, Andersen MH, Pedersen AE, Soleimani A, Lorentzen T, Johansen JS, Svane IM. Vaccination with autologous dendritic cells pulsed with multiple tumor antigens for treatment of patients with malignant melanoma: results from a phase I/II trial. *Cytotherapy.* 2010 Apr 29. Epub ahead of print.
50. Berntsen A, Trepiaakas R, Wenandy L, Geertsen PF, thor Straten P, Andersen MH, Pedersen AE, Claesson MH, Lorentzen T, Johansen JS, Svane IM. Therapeutic dendritic cell vaccination of patients with metastatic renal cell carcinoma: a clinical phase 1/2 trial. *J Immunother.* 2008; 31:771–780. [PubMed: 18779742]
51. Weide B, Pascolo S, Scheel B, Derhovanessian E, Pflugfelder A, Eigentler TK, Pawelec G, Hoerr I, Rammensee HG, Garbe C. Direct injection of protamine-protected mRNA: results of a phase 1/2 vaccination trial in metastatic melanoma patients. *J Immunother.* 2009; 32:498–507. [PubMed: 19609242]
52. Wobser M, Keikavoussi P, Kunzmann V, Weininger M, Andersen MH, Becker JC. Complete remission of liver metastasis of pancreatic cancer under vaccination with a HLA-A2 restricted peptide derived from the universal tumor antigen survivin. *Cancer Immunol Immunother.* 2006; 55:1294–1298. [PubMed: 16315030]
53. Kamb A, Wee S, Lengauer C. Why is cancer drug discovery so difficult? *Nat Rev Drug Discov.* 2007; 6:115–120. [PubMed: 17159925]
54. Fukuda S, Pelus LM. Survivin, a cancer target with an emerging role in normal adult tissues. *Mol Cancer Ther.* 2006; 5:1087–1098. [PubMed: 16731740]
55. Blum B, Bar-Nur O, Golan-Lev T, Benvenisty N. The anti-apoptotic gene survivin contributes to teratoma formation by human embryonic stem cells. *Nat Biotechnol.* 2009; 27:281–287. [PubMed: 19252483]
56. Weinstein IB, Joe AK. Mechanisms of disease: Oncogene addiction--a rationale for molecular targeting in cancer therapy. *Nat Clin Pract Oncol.* 2006; 3:448–457. [PubMed: 16894390]
57. O'Connor DS, Wall NR, Porter AC, Altieri DC. A p34(cdc2) survival checkpoint in cancer. *Cancer Cell.* 2002; 2:43–54. [PubMed: 12150824]
58. Lee CW, Raskett CM, Prudovsky I, Altieri DC. Molecular Dependence of Estrogen Receptor-Negative Breast Cancer on a Notch-Survivin Signaling Axis. *Cancer Res.* 2008; 68:5273–5281. [PubMed: 18593928]
59. Kim PJ, Plescia J, Clevers H, Fearon ER, Altieri DC. Survivin and molecular pathogenesis of colorectal cancer. *Lancet.* 2003; 362:205–209. [PubMed: 12885482]
60. O'Connor DS, Schechner JS, Adida C, Mesri M, Rothermel AL, Li F, Nath AK, Pober JS, Altieri DC. Control of apoptosis during angiogenesis by survivin expression in endothelial cells. *Am. J. Pathol.* 2000; 156:393–398. [PubMed: 10666367]
61. Daly C, Wong V, Burova E, Wei Y, Zabski S, Griffiths J, Lai KM, Lin HC, Ioffe E, Yancopoulos GD, Rudge JS. Angiopoietin-1 modulates endothelial cell function and gene expression via the transcription factor FKHR (FOXO1). *Genes Dev.* 2004; 18:1060–1071. [PubMed: 15132996]
62. Tran J, Master Z, Yu JL, Rak J, Dumont DJ, Kerbel RS. A role for survivin in chemoresistance of endothelial cells mediated by VEGF. *Proc Natl Acad Sci U S A.* 2002; 99:4349–4354. [PubMed: 11917134]
63. Lessene G, Czabotar PE, Colman PM. BCL-2 family antagonists for cancer therapy. *Nat Rev Drug Discov.* 2008; 7:989–1000. [PubMed: 19043450]

64. Plescia J, Salz W, Xia F, Pennati M, Zaffaroni N, Daidone MG, Meli M, Dohi T, Fortugno P, Nefedova Y, Gabrilovich DI, Colombo G, Altieri DC. Rational design of shepherdin, a novel anticancer agent. *Cancer Cell*. 2005; 7:457–468. [PubMed: 15894266]
65. Sun C, Nettesheim D, Liu Z, Olejniczak ET. Solution structure of human survivin and its binding interface with Smac/Diablo. *Biochemistry*. 2005; 44:11–17. [PubMed: 15628841]