Editorial

Par-4: how far will it go?

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The mainstay of cancer treatment typically includes surgery, chemotherapy, and radiation. If cancer is confined to one area, surgery alone can be curative, although it is often combined with adjuvant therapy (1). Radiation is limited to treatment of cells within the scope of the radiation beam, although radio immunotherapy has enhanced radiation therapy particularly in B-cell lymphomas (2). Chemotherapy has a number of forms, including systemically toxic drugs and both drugs and antibodies targeted to specific proteins that are driving the growth of the cancer cells. Chemotherapy can be used to treat systemically both visible and occult cancer cells. However, therapy resistance in tumors is a continuing problem for patients and oncologists alike despite an aggressive treatment regimen. In some cases, early mutations give resistance to tumors, or to a sub-population of tumor cells within a heterogeneous tumor, even prior to treatment. In this case treatment can cause death to sensitive cells allowing the few resistant cells to become the dominant population and lead to tumor recurrence. On the other hand, tumors that initially respond to treatment often become resistant over time as new mutations develop that allow cells to evade death. Such resistance, which may lead to multi-drug resistance, results in tumor recurrence and, ultimately, death (3). In some cases, the mechanism of resistance can be determined, so that a potential new drug can be developed to overcome the resistance, as in the case of imatinib (4,5). However, often no additional drug is suggested by the development of resistance, and other treatments, both targeted and more systemic, are explored with varying degrees of success.

The body has an arsenal of tools to add to the battle against cancer. In addition to the use of therapy to treat existing malignancies, tumor suppressor genes play a major role in the control of cancer and tumorigenesis. Tumor suppressor genes code for regulating proteins that conduct surveillance for and repair of problems arising that lead to oncogenesis and for miRNA molecules that regulate production of target proteins. In many cases, tumor suppressor genes are mutated resulting in the lack of normal, active protein production as is often the case with p53 (6), and in other cases epigenetic changes result in repression of gene expression (7). Knowledge of the mechanism by which tumor suppressor genes are inactivated in cancer offers the potential for a targeted therapy aimed at restoration of tumor suppressor function, whether it be mutational alterations of DNA or epigenetic in origin. In these cases, resistant tumor cells may become sensitive to therapy.

The manuscript entitled "A naturally generated decoy of the prostate apoptosis response-4 protein overcomes therapy resistance in tumors" by Hebbar *et al.* from the laboratory Vivek Rangnekar, addresses the issue of tumor suppressors and therapy resistance in cancer (8). The main focus of this study was to identify factors released from treatment sensitive cancer cells that induce apoptosis in treatment resistant cancer cells. In this manuscript, authors isolated a natural product that can induce apoptosis in drug resistant tumor cells. The natural protein is a caspase 3 cleavage product of the tumor suppressor, Prostate apoptosis response 4, Par-4, that is released from dying cancer cells and causes apoptosis in both therapy sensitive and therapy resistant cancer cells. Notably, this provides a natural targeted therapy for drug resistance tumor cells. Par-4 was first identified in 1994 by Dr. Rangnekar's

group at the University of Kentucky as a protein found in rat prostate cancer cells that were undergoing apoptosis and was absent from rat prostate cancer cells that were not undergoing apoptosis (9). A number of intracellular mechanisms were subsequently described through which Par-4 mediated apoptosis in response to apoptotic stimuli and inhibited survival in cancer cells (10-14). Subsequently, Par-4 was discovered to have a bystander effect on cancer cells located both proximally and distally to cells that expressed Par-4 (15). In this study, the full length Par-4 protein, containing the centrally located SAC domain, identified as the domain responsible for apoptosis in cancer cells, as well as the SAC domain alone, were shown to be secreted from cells expressing active Par-4 and exert the bystander effects through activation of the extrinsic pathway of apoptosis in both nearby and distally located cancer cells.

The natural decoy examined in this study is a 15 kDa fragment from the N terminus of Par-4, called Par-4 Amino-terminal Fragment, or PAF, that enters cancer cells to induce apoptosis, and, as with previous Par-4 events, only affects cancer cells, not normal cells. PAF works by entering cells and stabilizing intracellular Par-4 through binding to Foxo45, an ubiquitin ligase that is responsible for the ubiquitin mediated degradation of Par-4. PAF contains a VASA domain that binds to the SPRY domain of Foxo45 acting as a competitive inhibitor of Foxo45 binding to endogenous Par-4. This activity occurs in both drug sensitive and drug resistant tumor cells.

There are a number of strengths in the study by Hebbar and co-workers. One strength of the study is the use of multiple cell lines. This study examined the paracrine effects of Par-4 in a number of cell types including tumor cells from prostate, breast, melanoma, and lung cancers as well as normal fibroblasts. Also included are mouse embryonic fibroblasts from wild type and Par-4$^{-/-}$ mice. The positive effects of PAF activity on a widely diverse set of cancer cells offers an exciting potential for the future treatment of cancer, and particularly those cancers that have thrived despite aggressive treatment. A second strength is the thoroughness with which the investigators identified the precise portion of the protein that has the effect and how well that was followed by an investigation into the mechanism of action. The study involved the pathways by which PAF exerts its activity and the specificity with which PAF binds to Fbxo45, to inhibit the binding of intracellular Par-4. Furthermore, the authors showed that PAF bound to Fbxo45 itself, competitively inhibiting the binding of endogenous Par-4, rather than binding to Par-4, Akt1, or to some other intermediate, to stabilize Par-4.

If Par-4 is ubiquitous in cells, selectively kills cancer cells from within via intrinsic pathways, is secreted to kill cancer cells in a bystander effect by binding to the surface of cancer cells and inducing apoptosis by an extrinsic pathway, and is released after cleavage by apoptotic cancer cells to enter and kill other cancer cells, including those resistant to therapy, in a paracrine manner, it is surprising that any cancer is able to grow at all with Par-4 present. However, PAF activity induces apoptosis at about 50–60% frequency in therapy resistant cells. The remaining cells remain resistant. Mechanisms of resistance are numerous and may include overproduction of Akt1, which inactivates Par-4, low expression of Par-4 in resistant cells, or alterations in either apoptosis or survival pathways that function independently of Par-4. The relatively rapid multiplication of cancer cells coupled with the frequency of mutations in tumor suppressor genes enhances the potential for additional mutations an increased heterogeneity of malignant tumors. These issues were well discussed in the manuscript.

The study could have been enhanced by the identification of the mechanism of PAF entry into cancer cells, although that is an ongoing study in the laboratory. The results may also be cautiously optimistic as the half-life of PAF could well be an issue in the efficacy of this peptide as a treatment. In addition, the effects of PAF on solid tumor effects are discussed only within the microenvironment of the tumor and not those resistant tumors existing distally to the sensitive tumors that are releasing PAF as they undergo apoptosis. However, the finding that conditioned media causes apoptosis in cancer cells *in vitro* suggests that PAF has the capability to effect apoptosis in distally located cancer cells as well as those within the same microenvironment as the PAF releasing cells. This is an important issue for the control of distant tumor cells, in particular micrometastatic lesions.

Overall, the study presents interesting and significant data in cancer research. The Rangnekar laboratory has produced a volume of studies on Par-4. Each study is thorough and well done, and this one is no exception. The current manuscript is a welcome addition to the body of literature on Par-4 that has been generated by the Rangnekar laboratory.

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Footnote

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Ethical Statement: The author is accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

References

1. National Cancer Institute. Cancer Treatment. NIH About Cancer. April 6, 2017. Available online: www.cancer.gov/about-cancer/treatment/types
2. Green DJ, Press OW. Whither radioimmunotherapy: to be or not to be? Cancer Res 2017;77:2191-6.
3. Wu Q, Yang Z, Nie Y, et al. Multi-drug resistance in cancer chemotherapeutics: mechanisms and lab approaches. Cancer Lett 2014;347:159-66.
4. Gorre ME, Ellwood-Yen K, Chiosis G, et al. BCR-ABL point mutants isolated from patients with imatinib mesylate-resistant chronic myeloid leukemia remain sensitive to inhibitors of the BCR-ABL chaperone heat shock protein 90. Blood 2002;100:3041-4.
5. Kujawski L, Talpaz M. Strategies for overcoming imatinib resistance in chronic myeloid leukemia. Leuk Lymphoma 2007;48:2310-22.
6. Duffy MJ, Synnott NC, Crown J. Mutant p53 as a target for cancer treatment. Eur J Cancer 2017;83:258-65.
7. Llinàs-Arias P, Esteller M. Epigenetic inactivation of tumour suppressor coding and non-coding genes in human cancer: an update. Open Biol 2017;7. pii: 170152.
8. Hebbar N, Burikhanov R, Shukla N, et al. A Naturally Generated Decoy of the Prostate Apoptosis Response-4 Protein Overcomes Therapy Resistance in Tumors. Cancer Res 2017;77:4039-50.
9. Sells SF, Wood DP Jr, Joshi-Barve SS, et al. Commonality of the gene programs induced by effectors of apoptosis in androgen-dependent and -independent prostate cells. Cell Growth Differ 1994;5:457-66.
10. Goswami A, Qiu S, Dexheimer TS, et al. Par-4 binds to topoisomerase 1 and attenuates its DNA relaxation activity. Cancer Res 2008;68:6190-8.
11. Gurumurthy S, Goswami A, Vasudevan KM, et al. Phosphorylation of Par-4 by protein kinase A is critical for apoptosis. Mol Cell Biol 2005;25:1146-61.
12. Johnstone RW, See RH, Sells SF, et al. A novel repressor, par-4, modulates transcription and growth suppression functions of the Wilms' tumor suppressor WT1. Mol Cell Biol 1996;16:6945-56.
13. Rangnekar VM. Apoptosis mediated by a novel leucine zipper protein Par-4. Apoptosis 1998;3:61-6.
14. Wang BD, Kline CL, Pastor DM, et al. Prostate apoptosis response protein 4 sensitizes human colon cancer cells to chemotherapeutic 5-FU through mediation of an NF kappaB and microRNA network. Mol Cancer 2010;9:98.
15. Burikhanov R, Zhao Y, Goswami A, et al. A novel extrinsic pathway for apoptosis by tumor suppressor Par-4. Cell 2009;138:377-88.

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