assays are used to monitor autophagic activity.

Importantly, the study by Anderson et al. (2015) demonstrates that, when tested in mouse xenograft tumor models, orally applied CB-5083 is well tolerated and results in a dose-dependent induction of ER stress and apoptosis in tumor tissues. Accordingly, the tumor volume is significantly reduced in several xenograft-based solid tumor mouse models, whereas, in the same assay, proteasome inhibitors are barely active. Lastly, the authors close by showing that p97 expression levels and the Ras-MAPK pathway activity correlate with the sensitivity to CB-5083.

Overall, the new p97 inhibitor reported here will surely serve as another invaluable tool to advance our understandings on the diverse biological mechanisms regulated by p97. Most significantly, the demonstration that drugs disrupting the cellular proteostasis network at another level can effectively kill cancer cells derived from solid tumors in mouse models is a strong testimony to the proteostasis addition theory, and the findings reported here will potentially open a new path to circumvent obstacles in treatment of solid tumors. In this regard, the study will certainly prompt tremendous interest in developing additional p97 inhibitors as well as inhibitors targeting other proteostasis regulators. Structural validation of the inhibitor-p97 interactions will certainly be launched in order to come up with rational designs to improve p97 inhibitors, which may overcome the inevitable rise of resistance to the current inhibitor. Meanwhile, given the great pharmacological property, low toxicity to normal cells, easy administration route, strong potency, and high specificity, CB-5083 seems to have a good chance to endure through the long and costly clinical testing and bring significant benefit to patients bearing proteostasis-addicted tumors.

REFERENCES

Anderson, D.J., Le Moigne, R., Kjakovic, S., Kumar, B., Rice, J., Wong, S., Wang, J., Yao, B., Valle, E., Kiss von Soly, S., et al. (2015). Cancer Cell *28*, this issue, 653–665.

Chou, T.F., Li, K., Frankowski, K.J., Schoenen, F.J., and Deshaies, R.J. (2013). ChemMedChem *8*, 297–312.

Deshaies, R.J. (2014). BMC Biol. *12*, 94–107.

Ju, J.S., Fuentealba, R.A., Miller, S.E., Jackson, E., Piwnica-Worms, D., Baloh, R.H., and Weihl, C.C. (2009). J. Cell Biol. *187*, 875–888.

Luo, J., Solimini, N.L., and Elledge, S.J. (2009). Cell *136*, 823–837.

Magnaghi, P., D'Alessio, R., Valsasina, B., Avanzi, N., Rizzi, S., Asa, D., Gasparri, F., Cozzi, L., Cucchi, U., Orrenius, C., et al. (2013). Nat. Chem. Biol. *9*, 548–556.

Meyer, H., Bug, M., and Bremer, S. (2012). Nat. Cell Biol. *14*, 117–123.

Navon, A., and Ciechanover, A. (2009). J. Biol. Chem. *284*, 33713–33718.

Wang, Q., Mora-Jensen, H., Weniger, M.A., Perez-Galan, P., Wolford, C., Hai, T., Ron, D., Chen, W., Trenkle, W., Wiestner, A., and Ye, Y. (2009). Proc. Natl. Acad. Sci. USA *106*, 2200–2205.

Wang, Q., Shinkre, B.A., Lee, J.G., Weniger, M.A., Liu, Y., Chen, W., Wiestner, A., Trenkle, W.C., and Ye, Y. (2010). PLoS ONE *5*, e15479.

Tumor-Educated Platelets as Liquid Biopsy in Cancer Patients

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Real-time monitoring of changes in cells or cell products released from malignant lesions into the blood has opened new diagnostic avenues ("liquid biopsy"). In this issue of *Cancer Cell*, Best and colleagues describe that tumor-associated blood platelets provide specific information on the location and molecular composition of the primary tumor.

The peripheral blood of a cancer patient is a pool of cells and/or cell products derived from the primary tumor and different metastatic sites, including circulating tumor cells (CTCs) and stromal cells of the tumor microenvironment (e.g., macrophages) as well as tumor-derived DNA, RNA, and proteins. The analysis of these blood components can, therefore, provide a comprehensive real-time picture of the tumor-associated changes in an individual cancer patient (Figure 1). This information can be used for screening and early detection of cancer, estimation of the risk for metastatic relapse or progression (prognostic information), stratification and real-time monitoring of treatment response, identification of therapeutic targets and resistance mechanisms (biological therapies), and a better understanding of the biology of metastatic development.

In contrast to tissue biopsies, blood samples can be obtained easily and repeatedly from cancer patients. In the last decade, many reports have focused on CTCs and circulating nucleic acids (in particular, tumor-derived DNA fragments and various

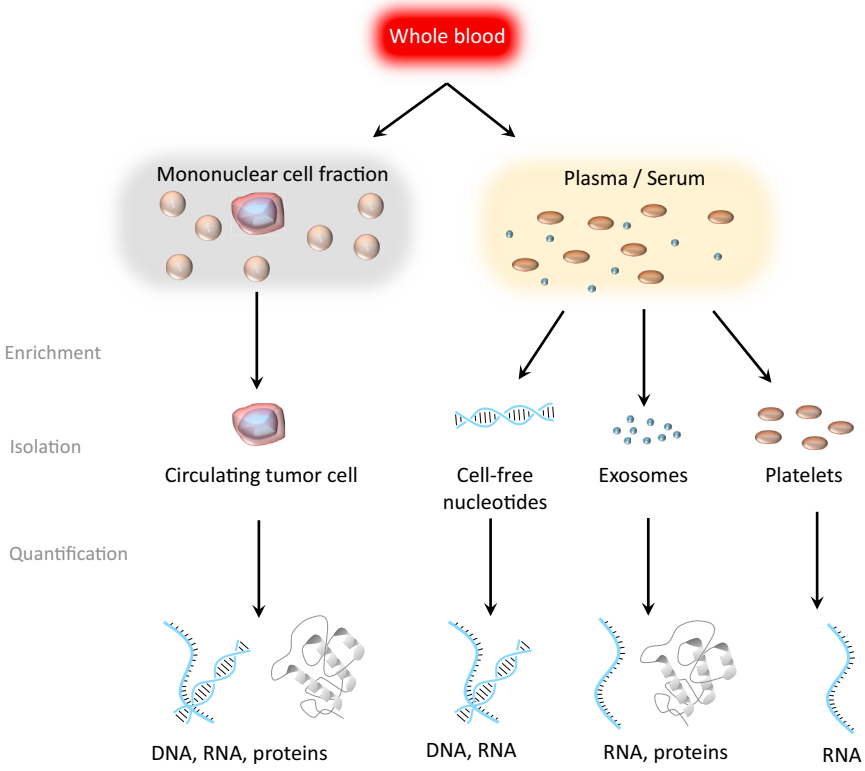

Figure 1. Sources of Liquid Biopsy
Blood can be separated into different fractions in order to enrich for tumor-associated biomarkers. From the mononuclear cell fraction, circulating tumor cells (CTCs) may provide genomic, transcriptomic, and proteomic information on the tumor. From plasma or serum, cell-free nucleotides and exosomes can be further used to interrogate cancer-secreted bioparticles. Tumor educated platelets (TEPs) carry additional information on the location of the tumor in their mRNA.

miRNA species) (Alix-Panabières and Pantel, 2014). More recently, the molecular analysis of tumor-derived microvesicles, in particular, exosomes (Melo et al., 2015), has broadened the spectrum of liquid biopsy applications. Regarding the clinical relevance, molecular analysis of CTCs and circulating tumor DNA (ctDNA) provides important information on therapeutic targets and drug resistance mechanisms in carcinoma patients. Although the ctDNA field is restricted to DNA analyses, investigation of CTCs allows comprehensive studies at the DNA, RNA, and protein level as well as functional studies including the establishment of cell lines and xenografts (Cayrefourcq et al., 2015).

Besides tumor cells and their products, normal cells present in the tumor microenvironment are also released into the blood stream, and these cells can harbor important information. In this issue of *Cancer Cell*, Best et al. (2015) describe an important potential role of tumor-educated blood platelets (TEPs). The biology behind this new diagnostic role of TEPs is the well-known interaction between blood platelets and tumor cells that is known to affect tumor growth and dissemination (Kuznetsov et al., 2012). This interaction affects not only the expression of relevant genes in tumor cells, but also alters the RNA profile of blood platelets (Best et al., 2015). In total, Best et al. (2015) performed mRNA sequencing of TEPs from 283 platelet samples and distinguished 228 patients with localized and metastasized tumors from 55 healthy individuals with 96% accuracy. Across six different tumor types (non-small cell lung carcinoma, colorectal cancer, glioblastoma, pancreatic cancer, hepatobiliary cancer, and breast cancer), the location of the primary tumor was correctly identified with 71% accuracy. Moreover, MET or *ERBB2*-positive and mutant *KRAS*, *EGFR*, or *PIK3CA* tumors were accurately distinguished using surrogate TEP mRNA profiles.

Interestingly, the tumor-specific educational programs in TEPs were predominantly influenced by tumor type and, to a lesser extent, by tumor progression and metastases (i.e., no significant differences between non-metastasized and metastasized tumors were detected). This is somewhat surprising, because experimental work has shown an influence of blood platelets on tumor cell dissemination (Labelle et al., 2011) and metastatic outgrowth (Kuznetsov et al., 2012). A possible explanation might be the low number of patients analyzed for each tumor type, which may not allow a statistically meaningful stratification in localized and metastatic tumors. Moreover, the set of tumors analyzed was very heterogeneous with regard to the metastatic capacities, e.g., glioblastomas can release tumor cells into the blood circulation but almost never produce overt metastases, while pancreatic and lung cancer are frequently metastasized at primary diagnosis. Thus, larger and more homogeneous datasets are required to address the important question of whether TEPs sequencing can also reveal information on the risk of metastatic relapse, which is the dominant cause of cancer-related deaths in most malignancies.

Although the findings of Best et al. (2015) are very exciting, the requirements for introducing a new cancer screening test into practice are very high. The presented case-control study is a good start, but much larger cohorts of individuals at risk need to be analyzed and followed up prospectively over many years. For example, cancer screening studies on prostate-specific antigen (PSA) in prostate cancer or mammography in breast cancer comprised ten thousands of individuals at risk and follow-up time of 10 or more years. Focusing on study populations at very high risk (e.g., people with a predisposition for cancer) might reduce this effort to some extent. Moreover, many aging people have benign tumors (e.g., skin tumors) or leukocytes carrying cancer-associated mutations (Genovese et al., 2014), and this may cause false-positive findings. Finally, despite all diagnostic efforts, early detection of cancer might not always translate into reduced mortality because tumor cell dissemination can occur very early during tumorigenesis and surgical removal of the primary lesion is no guarantee for cure.

In contrast, the second key finding of Best et al. (2015) that TEPs mRNA profiles could distinguish mutant *KRAS*, *EGFR*, or *PIK3CA* tumors could be readily validated

in future prospective clinical trials in which cancer patients will receive targeted therapies based on these profiles. This is a very important application in modern oncology. Besides, novel chemotherapy drugs (e.g., antibodies or small inhibitors) that target specific receptors or molecular pathways on cancer cells have been developed over the past ten years (Wan et al., 2013). Although these drugs are administered to target metastatic cells, current stratification of therapy is based largely on the analysis of the resected primary tumor. However, metastases occur in many patients years after primary tumor diagnosis, and they can harbor unique genomic alterations different from the bulk of the original primary tumor cells (Kang and Pantel, 2013). Thus, the direct analysis of metastatic cells will reveal important information for a systemic cancer therapy targeting metastatic disease. However, a biopsy of overt metastases is an invasive procedure limited to certain locations and not easily acceptable in the clinic. Moreover, recent work has shown that different metastatic sites harbor different genomic aberrations, and biopsies of one or two accessible metastases may not be representative. Thus, the assessment of the TEP mRNA profile might provide real-time information on the actual status of metastatic lesions. In this regard, future comparative studies will show whether the TEP approach is superior or complementary to other means of liquid biopsy (e.g., ctDNA or CTC analyses) in stratifying individual patients to the appropriate therapy.

Before these clinical trials can be conducted, the robustness of the assay and factors confounding the blood platelet mRNA profile need to be investigated. The authors recommend isolation of the platelet fraction within 48 hr after blood withdrawal; however, research on many cell types has indicated that mRNA profiles will change during this time period. In addition, systemic factors such as chronic or transient inflammatory diseases or cardiovascular events and other non-cancerous diseases may also influence the platelet mRNA profile.

Taken together, the ability of TEPs to pinpoint the location of the primary tumor might advance the use of liquid biopsies for cancer diagnostics. Further validation is warranted to determine the clinical utility of TEP profiles for blood-based liquid biopsy. The validation of liquid biopsy assays is an important task of the new European consortium CANCER-ID that comprises more than 30 institutions from academia and industry (http://www.cancer-id.eu).

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REFERENCES

Alix-Panabières, C., and Pantel, K. (2014). Nat. Rev. Cancer *14*, 623–631.

Cayrefourcq, L., Mazard, T., Joosse, S., Solassol, J., Ramos, J., Assenat, E., Schumacher, U., Costes, V., Maudelonde, T., Pantel, K., and Alix-Panabières, C. (2015). Cancer Res. *75*, 892–901.

Genovese, G., Kähler, A.K., Handsaker, R.E., Lindberg, J., Rose, S.A., Bakhoum, S.F., Chambert, K., Mick, E., Neale, B.M., Fromer, M., et al. (2014). N. Engl. J. Med. *371*, 2477–2487.

Kang, Y., and Pantel, K. (2013). Cancer Cell *23*, 573–581.

Kuznetsov, H.S., Marsh, T., Markens, B.A., Castaño, Z., Greene-Colozzi, A., Hay, S.A., Brown, V.E., Richardson, A.L., Signoretti, S., Battinelli, E.M., and McAllister, S.S. (2012). Cancer Discov. *2*, 1150–1165.

Labelle, M., Begum, S., and Hynes, R.O. (2011). Cancer Cell *20*, 576–590.

Melo, S.A., Luecke, L.B., Kahlert, C., Fernandez, A.F., Gammon, S.T., Kaye, J., LeBleu, V.S., Mittendorf, E.A., Weitz, J., Rahbari, N., et al. (2015). Nature *523*, 177–182.

Wan, L., Pantel, K., and Kang, Y. (2013). Nat. Med. *19*, 1450–1464.

Best, M.G., Sol, N., Kooi, I., Tannous, J., Westerman, B.A., Rustenburg, F., Schellen, P., Verschueren, H., Post, E., Koster, J., et al. (2015). Cancer Cell *28*, this issue, 666–676.

Welcoming Treat: Astrocyte-Derived Exosomes Induce PTEN Suppression to Foster Brain Metastasis

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Metastasis to distant organs depends on pathological crosstalk between tumor cells and various tissue-specific stromal components. Zhang and colleagues recently demonstrated that astrocyte-derived exosomal miR-19a reversibly downregulated PTEN expression in cancer cells, thereby increasing their CCL2 secretion and recruitment of myeloid cell to promote brain metastasis.

Stephen Paget's visionary "seed and soil" hypothesis underscored the importance of mutual compatibility between tumor cells and host organs in the formation of metastatic lesions. Such compatibility may stem from the intrinsic characteristics of cancer cells and the host microenvironments, but could also be developed though their co-evolution during adaptive