

showed clinical resistance to ponatinib. However, 22 cases of compound mutations without T315I patterns of variable effectiveness was demonstrated across the panel of TKIs, which suggests that early detection of compound mutations could be combined with sensitivity studies to shape a therapeutic change in TKIs and thus abort the emergence of resistant clones.

Computer modeling suggested the impact of compound mutations on TKI activity. The proliferation studies noted above found that, for compound mutations not containing T315I, ponatinib and dasatinib had similar activities, except with the Y253H/E55V compound mutation, where dasatinib is considerably more active. The binding sites of ponatinib and dasatinib are known to be different; can modeling explain the difference? Indeed, it can; molecular dynamic simulations showed that Y253H and E255V mutations force a shift in the P loop of the ABL kinase domain, obstructing the ponatinib binding site. Similar simulations sug-

gested poor ponatinib activity in clinically relevant disease evolution, such as the difference in the binding domains of a single T315I and the resistant T315I/E255V mutation. Thus, the authors elegantly followed the interplay of structure, in vitro and in vivo function.

Why is this study important? First, it is a demonstration of how clinical material, wet bench work, and computer modeling can be melded to develop a clear understanding of clinically important biology. Second, it provides a clear roadmap of how future studies can be performed to understand disease resistance. As “targeted therapy” becomes an increasing reality in cancer care, it will become increasingly important to understand and anticipate how Darwinian selection will select for resistance. This manuscript helps prepare us for that future.

REFERENCES

Apperley, J.F. (2007). *Lancet Oncol.* 8, 1018–1029.

Cortes, J.E., Kantarjian, H., Shah, N.P., Bixby, D., Mauro, M.J., Flinn, I., O'Hare, T., Hu, S., Narasimhan, N.I., Rivera, V.M., et al. (2012). *N. Engl. J. Med.* 367, 2075–2088.

Druker, B.J., Guilhot, F., O'Brien, S.G., Gathmann, I., Kantarjian, H., Gattermann, N., Deininger, M.W., Silver, R.T., Goldman, J.M., Stone, R.M., et al.; IRIS Investigators (2006). *N. Engl. J. Med.* 355, 2408–2417.

Khorashad, J.S., Kelley, T.W., Szankasi, P., Mason, C.C., Soverini, S., Adrian, L.T., Eide, C.A., Zabriskie, M.S., Lange, T., Estrada, J.C., et al. (2013). *Blood* 121, 489–498.

O'Hare, T., Shakespeare, W.C., Zhu, X., Eide, C.A., Rivera, V.M., Wang, F., Adrian, L.T., Zhou, T., Huang, W.S., Xu, Q., et al. (2009). *Cancer Cell* 16, 401–412.

Smith, C.C., Lasater, E.A., Zhu, X., Lin, K.C., Stewart, W.K., Damon, L.E., Salerno, S., and Shah, N.P. (2013). *Blood* 121, 3165–3171.

Soverini, S., De Benedittis, C., Machova Polakova, K., Brouckova, A., Horner, D., Iacono, M., Castagnetti, F., Gugliotta, G., Palandri, F., Papayannidis, C., et al. (2013). *Blood* 122, 1634–1648.

Zabriskie, M.S., Eide, C.A., Tantravahi, S.K., Vellore, N.A., Estrada, J., Nicolini, F.E., Khoury, H.J., Larson, R.A., Konopleva, M., Cortes, J.E., et al. (2014). *Cancer Cell*.

DAISY: Picking Synthetic Lethals from Cancer Genomes

Colm J. Ryan,¹ Christopher J. Lord,² and Alan Ashworth^{2,*}

¹Systems Biology Ireland, Conway Institute, University College Dublin, Dublin 4, Ireland

²The Breakthrough Breast Cancer Research Centre, The Institute of Cancer Research, London SW3 6JB, UK

*Correspondence: alana@icr.ac.uk

<http://dx.doi.org/10.1016/j.ccr.2014.08.008>

A better understanding of genetic interactions in cancer might help identify new therapeutic approaches that exploit the concept of synthetic lethality. Ruppin and colleagues have developed a new computational method, DAISY, that predicts such interactions and potentially facilitates the delineation and validation of comprehensive genetic interaction networks.

Most of the major recent advances in the development of targeted therapies for cancer have originated from the identification and exploitation of genetic dependencies that operate specifically in tumor cells. Many of these dependencies are considered as oncogene addiction effects, where tumor cells become reliant upon the activity of key driver genes such as *BCR-ABL* and *EGFR*; pharma-

cological inhibition of these drivers has therapeutic benefit. Other dependencies include those that arise as a consequence of the “cancer state,” also called nononcogene addictions (Luo et al., 2009). However, in many cases these gene addictions are difficult to target directly with drugs. Moreover, there are many recessive driver mutations in tumor genomes, the so-called tumor suppressors, in which absent

or reduced gene function contributes to tumorigenesis. Synthetic lethality (SL) provides a potential approach to targeting these latter two classes of alterations. The term describes the relationship between two genes whereby individual defects in either gene are compatible with cell viability, but the synthesis or combination of gene defects results in cell death (Ashworth et al., 2011). Recently the

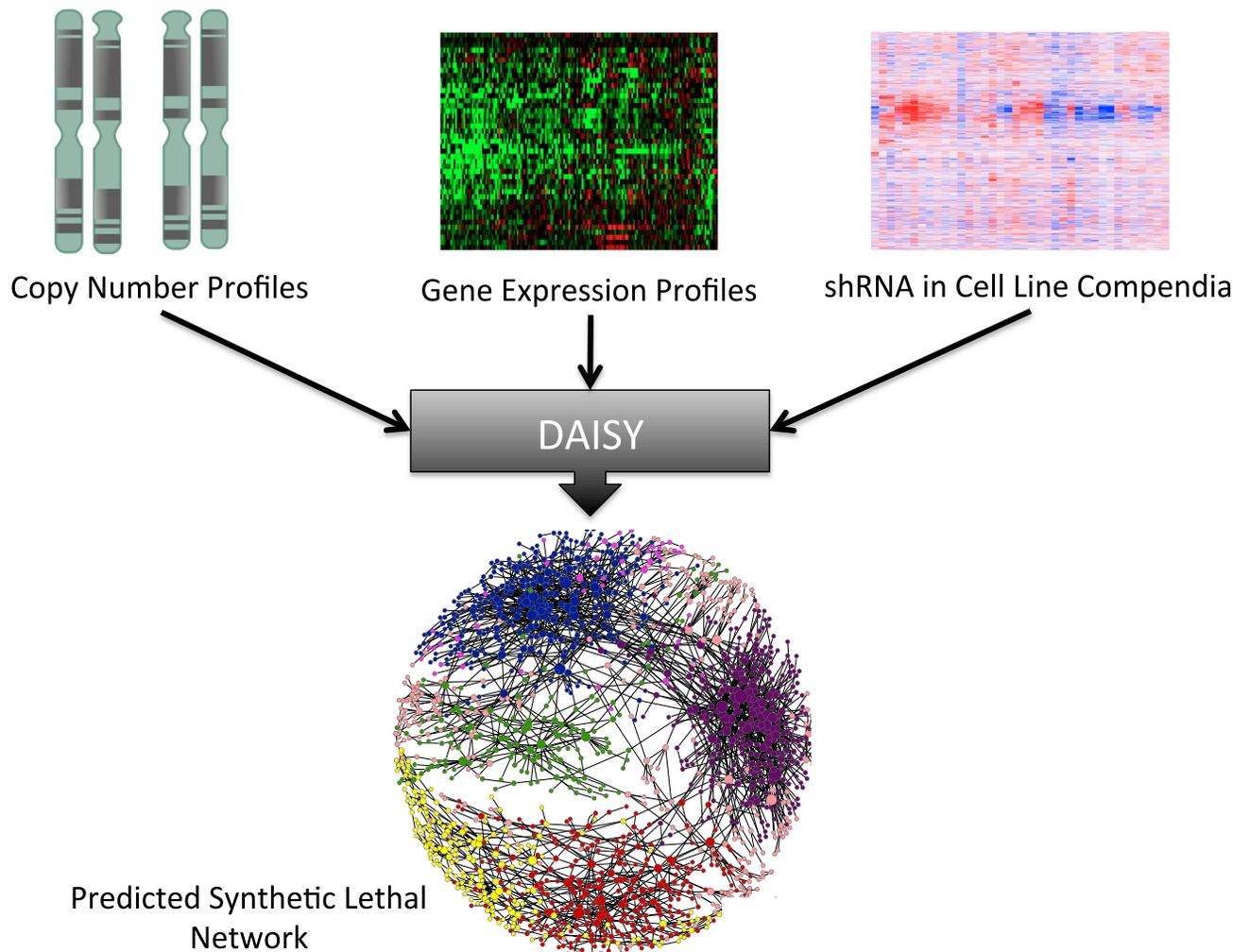


Figure 1. The DAISY Approach to Predicting SL

DAISY integrates information from three data sources—copy number profiles from tumors and tumor cell lines, gene expression profiles, and tumor cell line shRNA screens—to predict SL effects.

exploitation of SL effects involving tumor suppressor genes such as *BRCA1* or *BRCA2* in cancer cells has led to the development of new therapeutic approaches (Lord and Ashworth, 2012).

In yeast and other model organisms, high-throughput reverse genetic screens have enabled the systematic mapping of SL networks on a genome-wide scale (Dixon et al., 2009). In human cancer cell lines, RNA interference (RNAi) and small molecule screens have also been employed to identify SL effects of potential therapeutic value (Sandmann and Boutsos, 2012). Although such screens have proven useful, they have largely focused on the identification of genetic interactions with individual genes or drugs. More systematic approaches have recently been developed (Hart and Moffat, 2013), but

these are not yet as robust as those in more genetically tractable organisms. As a complement and alternative to these relatively elaborate experimental studies, computational methods for predicting SLs would be beneficial. In model systems, many predictive approaches have been proposed, but these primarily focus on extending experimentally derived SL networks rather than de novo prediction of interactions (Wong et al., 2004), limiting their utility for cancer.

In a recent issue of *Cell*, Jerby-Arnon et al. (2014) describe a new computational approach termed DAISY (*data mining synthetic lethality identification pipeline*) that aims to facilitate the large-scale identification of SLs in cancer. DAISY is elegantly built upon three principles. The first, termed “genomic survival of the fittest”

(gSOF) makes the assumption that SL between two genes can be discovered by identifying pairs of genes whose coinactivation in tumors occurs much less than expected by chance alone; the premise is that cells with inactivation in both partners of a SL pair have a survival disadvantage and thus are rarely observed. Similar approaches based on “mutually-exclusive” mutations have previously been proposed (Ciriello et al., 2012), but large-scale benchmarking and evaluation of results has been lacking. The second component of DAISY exploits published genome-wide short-hairpin RNA (shRNA) screens performed in panels of human tumor cell lines. By combining these data with genomic and transcriptomic profiles of each cell line model, SL pairs were captured by seeking shRNAs that specifically cause

cell inhibition when a particular gene has either reduced expression or reduced copy number. Finally, a third approach exploits the observation that SL pairs frequently engage in functionally related processes and are therefore often coexpressed. Each of these three heuristics is used independently to assign an interaction score for each gene pair using a variety of expression, copy number, and shRNA data sets. These scores are then integrated to predict candidate SLs (Figure 1). In addition to SL, Jerby-Arnon et al. (2014) use the same approach to predict what they term “synthetic dosage lethal” (SDL) interactions, where overexpression of one gene (for example, driven by copy number amplification) renders a second gene essential.

To validate their approach, they first demonstrated that DAISY is capable of predicting dependencies previously identified by RNAi experiments in human cell lines. They evaluated each feature (coexpression, gSOF, and shRNA results) individually and found that a combination of the three proves to be the strongest predictor and that data from shRNA screens in large cell line collections offer little predictive power on their own. To further demonstrate the utility of DAISY, they predicted novel SLs for the tumor suppressor *VHL* and validated a number of these using either siRNAs or drugs. Having shown DAISY capable of predicting known and novel SLs, they applied their algorithm on a genome-wide scale to predict high-confidence SL and SDL networks encompassing ~3,000 SL and ~3,600 SDL interactions. Although these data have considerable utility, a weighted network containing interactions that pass some but not all criteria might also be beneficial to those planning to experimentally test interactions associated with specific genes.

Subsequent analyses in the paper highlight the utility of the SL and SDL high-

confidence networks in predicting both patient and cell line phenotypes. The SL network is applied to predict the sensitivity of cell lines to RNAi of specific genes, while the SDL network is used for predicting sensitivity to specific drugs. A drug targeting a specific gene is predicted to impact growth if its SDL partner is overexpressed in the cell line being measured. This effect is shown to be cumulative—the greater the number of SDL partners of a gene are overexpressed, the more likely that a drug targeting a gene is to impact cell growth, suggesting that the predicted interactions may be synthetic sick (causing a growth defect) rather than SL (causing cell death).

In an indirect validation of the predicted networks, the authors suggest that tumors with low expression of two SL partners should have reduced fitness, potentially indicating an improved outcome for patients. Using a large cohort of breast cancer patients, the authors demonstrate that this might indeed be the case; patients with tumors underexpressing two SL partners have a better overall survival. This effect is also cumulative; the more SL pairs a tumor underexpresses, the better the outcome.

In the present study, the authors have focused primarily on SLs and SDLs associated with copy number alterations (loss and amplification respectively). This approach enables uniform application of the DAISY algorithm to all genes in a variety of copy-number data sets. However, many tumor suppressors and the majority of oncogenes are subject to more subtle missense mutations (Vogelstein et al., 2013), and information on these are largely missing from the predicted SL networks. In principle, there is no reason that the DAISY approach cannot be applied in a more fine-grained approach to identify synthetic dependencies associated with specific oncogenic mutations.

Perhaps the most promising aspect of the DAISY approach is that it relies primarily on data from sequencing and gene expression studies. As the number of patient and cell line samples with such data available is increasing exponentially, we can expect significant improvements in the accuracy and coverage of the predicted SL networks. This may facilitate the identification of SL interaction partners for rare driver mutations. For more commonly mutated genes (e.g., *KRAS*, *PTEN*), it may become possible to identify higher-order dependencies (e.g., gene X is essential if *KRAS* and *APC* are both mutated) of therapeutic relevance.

REFERENCES

- Ashworth, A., Lord, C.J., and Reis-Filho, J.S. (2011). *Cell* 145, 30–38.
- Ciriello, G., Cerami, E., Sander, C., and Schultz, N. (2012). *Genome Res.* 22, 398–406.
- Dixon, S.J., Costanzo, M., Baryshnikova, A., Andrews, B., and Boone, C. (2009). *Annu. Rev. Genet.* 43, 601–625.
- Hart, T., and Moffat, J. (2013). *Nat. Methods* 10, 397–399.
- Jerby-Arnon, L., Pfretzer, N., Waldman, Y.Y., McGarry, L., James, D., Shanks, E., Seashore-Ludlow, B., Weinstock, A., Geiger, T., Clemons, P.A., et al. (2014). *Cell* 158, 1199–1209.
- Lord, C.J., and Ashworth, A. (2012). *Nature* 481, 287–294.
- Luo, J., Solimini, N.L., and Elledge, S.J. (2009). *Cell* 136, 823–837.
- Sandmann, T., and Boutros, M. (2012). *Curr. Opin. Genet. Dev.* 22, 36–44.
- Vogelstein, B., Papadopoulos, N., Velculescu, V.E., Zhou, S., Diaz, L.A., Jr., and Kinzler, K.W. (2013). *Science* 339, 1546–1558.
- Wong, S.L., Zhang, L.V., Tong, A.H., Li, Z., Goldberg, D.S., King, O.D., Lesage, G., Vidal, M., Andrews, B., Bussey, H., et al. (2004). *Proc. Natl. Acad. Sci. USA* 101, 15682–15687.