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J. Michael Bishop’s Lecture Part 3: Exploiting the Cancer Genome

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Contents

1. Keywords and Terms
2. Lecture Notes
3. Review Questions
4. Answers to Review Questions
5. Discussion Questions
6. Answers to Discussion Questions
7. Explain or Teach These Concepts to a Friend
8. Papers for Journal Club

1. Keywords and Terms

Targeted therapy, chromosomal translocation, acute promyelocytic leukemia (APL), all-trans retinoic acid (ATRA), arsenic trioxide (ATO), mouse model, frontline therapy, combination therapy, bi-modal attack, synthetic lethality, MYC proto-oncogene, CDK-1 cell-cycle kinase, apoptosis, biomarker, chromosomal passenger protein complex, aurora-B kinase, autophagy, genome-wide screens, BRCA genes, poly (ADP-ribose) polymerase (PARP)

2. Lecture Notes

This lecture describes efforts to exploit the genetic paradigm for cancer in the development of new therapeutics for the disease, with focus on a relatively new
approach known as synthetic lethality. The lecture is a formal research seminar and is suitable for advanced undergraduate science students and graduate students.

3. Review Questions

1. What are the two major categories of therapeutic targets in cancer cells?

2. How might these categories be attacked?

3. Describe the current status of each category.

4. What created the therapeutic target in APL and what is the nature of the gene product?

5. Prior to recent work, what was the conventional therapy for the disease? How effective was it?

6. How has that therapy been modified?

7. The current treatment protocol produces a cure rate of ca. 95%. What was the crucial final step in achieving that rate? How was it first authenticated?

8. Why is the current strategy so effective?

9. What is synthetic lethality and how versatile might it be as a therapeutic strategy?

10. Why is MYC a candidate for the synthetic lethal strategy?

11. The lecture describes two therapeutic agents that elicit a synthetic lethal interaction with MYC when the latter is over-expressed in cells. How might that observation be useful?

12. Why was it a surprise that autophagy was responsible for a major part of the synthetic lethal interaction between VX-680 and over-expression of MYC?

13. What synthetic lethal interaction is already in an advanced stage of clinical development?

14. Explain the presumed mechanism of that interaction.
4. Answers to Review Questions

1. Gain and loss of function mutations

2. Gain of function by inhibition. Loss of function by resurrection, replacement or bypass. Both by flank attack with synthetic lethality. The direct approach to loss of function given here is more diversified than what was described in the lecture.

3. Inhibition of gain of function is the most advanced, with a few drugs already in general use and many more under development. No direct approach to loss of function has yet proven practicable. There is one example of synthetic lethality in clinical use, and the potential for other applications has become apparent in preclinical studies.

4. The target was created by a chromosomal translocation that fused a major portion of the alpha-retinoic receptor to the bulk of a previously unidentified protein now know as PML. The result was a dangerous molecular mongrel, PML-APL.

5. Treatment with ATRA in combination with general cytotoxic agents. The cure rate was ca. 80%.

6. By addition of treatment with low doses of ATO. In patients with relatively mild disease, treatment with general cytotoxic agents may be omitted to avoid the severe side effects.

7. Using ATRA and ATO together as frontline therapy. The approach was first authenticated in a mouse model for APL.

8. First, the combination of two agents that attack different domains of the same protein greatly reduces the risk of drug resistance emerging prior to a complete therapeutic response. Second, the therapy kills both the bulk leukemia cells and the stem cells for the leukemia. Third, there is evidence that the therapeutic target may be the only driver for the leukemia - - an unusual circumstance in cancer.

9. The lethal effect of combining two mutations, neither of which is lethal when it stands alone. In principle, it should be useful against both gain and loss of function.

10. First, the gene is vital to cells, so direct inhibition of its function might have severe side effects. Second, the nature of the gene product makes it a difficult target for
current pharmaceuticals. Consequent to these limitations, there is no direct therapy against MYC in view.

11. First, the use of these and related agents against cancers that over-express MYC might be effective without requiring direct inhibition of MYC. Second, over-expression of MYC could represent a biomarker for susceptibility of cancer cells to the two types of therapeutics.

12. Autophagy was traditionally regarded as a survival mechanism for cells. It is now clear, however, that the process can be lethal if forced to extremes, which is apparently what happens in the synthetic lethal interaction between VX-680 and over-expression of MYC.

13. The interaction between inhibition of the PARP enzyme and a deficiency in either of the BRCA genes.

14. A deficiency in either BRCA gene compromises one form of DNA repair, inhibition of PARP compromises another. In combination, the two allow an intolerable amount of DNA damage to persist, leading to cellular suicide.

5. Discussion Questions

1. What might be the most efficient way to identify additional synthetic lethal interactions with gain or loss of function in cancer cells?

2. How might synthetic lethality be used to reduce the risk of acquired drug resistance in cancer cells.

3. What lessons about drug development might the ATO story hold?

4. How might you rebut skepticism about whether autophagy was really the lethal agency in the interaction between VX-680 and MYC?

5. Pharmacological agents are often not as specific as they are designed to be, leading to “off-target” effects. How could you test whether inhibition of the aurora-B kinase was actually responsible for the synthetic lethal interaction with MYC?

6. Given what is presently known about the synthetic lethal interaction between inhibition of PARP and deficiencies in the BRCA genes, what other circumstances might combine with PARP inhibitors to give a synthetic lethal interaction?
6. Answers to Discussion Questions

1. Perform screens that utilize genome-wide inhibitions of individual genes, with either RNAi or pharmacological agents. This can be done blindly with established lines of cancer cells, or with single known loss or gain of functions. For example, such screens have already been reported for an oncogenic mutant of the RAS proto-oncogen and for MYC. Numerous candidates for synthetic lethal interaction with these two genes have been identified. In addition, similar screens identified a synthetic lethal interaction between PARP inhibitors and deficiencies in the BRCA genes.

2. For gain of function, combine a synthetic lethal interaction with one oncogene with direct inhibition of another, or combine two synthetic lethal interactions, particularly two that kill by different means (eg., apoptosis and autophagy). For loss of function, only a combination of two synthetic lethal interactions is presently feasible; direct mitigation of loss of function is not yet practicable.

3. First, the search for active ingredients in traditional Chinese (and other) medications may reveal new candidates as therapeutics for cancer. It would be best, however, to focus on those medications for which there is some sound evidence of efficacy against one or more forms of cancer. Anecdotal folk lore can be very misleading. Second, given two independently effective but not curative therapeutics, there should be no delay in testing their efficacy in combination.

4. Disable autophagy by either genetic or pharmacological means and see whether that impedes the synthetic lethal effect. As mentioned in the lecture, experiments have shown that it does.

5. Determine whether the effect of VX-680 can be replicated by a different inhibitor of the kinase or by depletion of the kinase by RNAi. These experiments have been done. The results sustain the conclusion that the synthetic lethal interaction is indeed due to inhibition of the aurora-B kinase. Depletion of other components of the chromosomal passenger protein complex also create a synthetic lethal interaction with MYC, so it is disablement of the complex that is responsible for the lethal interaction, not some other effect of inhibiting aurora-B kinase.

6. First, any deficiency in DNA repair that is distinctive from that created by inhibition of PARP. Second, treatments that act by damaging DNA and might well overwhelm
the repair mechanisms when they are already compromised by inhibition of PARP. Preliminary results indicate that both of these may be viable options.

7. Explain or Teach These Concepts to a Friend

1. What is meant by targeted therapy, and why is it only now that such therapy is under development for cancer?

2. Explain the potential approaches to targeted therapies for both gain and loss of function, as well as the potential limitations on each.


4. The combination of ATRA and ATO is presently the only truly curative form of targeted therapy. (Although the targeting was serendipitous, it is highly specific.) Explain this exceptional efficacy.

5. Explain the difference between apoptosis and autophagy.

6. Research the literature on your own

7. How were the individual therapeutic efficacies of ATRA and ATO against APL first discovered?

8. What is known about the therapeutic mechanisms of ATRA and ATO?

9. What is the status of the cell-cycle kinases as therapeutic targets in cancer?

10. How was synthetic lethality discovered and in what context was it first suggested as a therapeutic strategy for cancer?

11. What potential therapeutics for MYC have been described in the literature? What is known about their potential efficacy?

12. Investigate the process and functions of autophagy, and explore the difficulties in authenticating its activity.

8. Papers for Journal Club

These papers introduce synthetic lethality as a therapeutic modality for cancer, provide early preclinical proofs of principle, and illustrate the use of genome-wide screens to
obtain a comprehensive inventory of synthetic lethal interactions with individual oncogenes.


