Bispecific T-Cell Engaging Antibodies for Cancer Therapy
Patrick A. Baeuerle and Carsten Reinhardt

Background facts:
- CD3 is the signaling portion of the TCR
- CD28 is found on T-cells. It binds b7, a costimulatory molecule found on the surface of profession APC.

1. Briefly describe the protein structure of a BiTE antibody? What two antigens does it recognize?

2. Describe how you might construct a gene that would express a BiTE antibody. What genetic material would you have to begin with, and how would you manipulate that material?

3. Although TCTL may be effective at killing cancer cells early on in the disease, what escape mechanisms ultimately allow the tumor to avoid killing by T-cells.

4. In what way do BiTE antibodies circumvent one viral escape mechanism?

5. Which protein is required at a lower dose for effective treatment, BiTE antibodies or conventional IgG?

6. Why is it necessary to infuse the BiTE antibodies into the patient?

7. Why do BiTE antibodies only appear to stimulate memory Tc cells and not naïve Tc cells.

8. Why do BiTE antibodies only activate Tc cells in close proximity/contact to their target, as opposed to all memory Tc cells, even though all Tc cells would have CD3.

9. Why would BiTE antibodies be more effective at fighting solid tumors than treatment with IgG?

10. When BiTE antibodies were first envisioned, it was thought to be better to link together two Fab fragments, one recognizing CD3 and the other the tumor antigen.
   i) The Fab-based BiTE antibodies are generally more stable than the Fv based ones. Why?
   ii) The origin of the anti-CD3 and anti-tumor antigen antibodies that were used to generate the BiTE-Fab and BiTE-Fv molecules were murine (mouse) hybridomas. Both the bispecific Fab and Fv molecules, caused some patients to develop antibodies against these proteins. How could you modify the protein to reduce this side-effect?