1. **Does biomarker testing prevent those who receive a breast cancer diagnosis from getting lymphedema?**

   Development of lymphedema is primarily caused by removal of lymph nodes at the time of breast surgery and the use of radiation to treat lymph nodes. After surgery / radiation, fluid cannot be as easily taken up by the lymphatic system and brought back to circulation, causing swelling. There are certainly factors that increase the risk of lymphedema – such as increasing number of lymph nodes removed and the choice of a mastectomy surgery versus a lumpectomy. However, the decision to remove additional lymph nodes, or perform mastectomy, or perform radiation to the lymph nodes is generally based on the extent of the breast cancer and are needed to prevent cancer recurrence. You might consider 'sentinel lymph node' biopsies a sort of biomarker – with this procedure, a small number of lymph nodes can be removed, with a more extensive lymph node dissection reserved to patients who have significant disease in the lymph nodes.

2. **Is biomarker testing covered by health insurance? Are there different ways to conduct biomarker testing? What are some of the extraordinary cases that benefited from biomarker testing?**

   Most of the biomarkers I discussed during my talk are covered by health insurance, but coverage may vary from plan to plan. Often times even if such testing is not initially covered, it will be covered when your oncologist has further discussions with the insurance company (i.e. prior authorization). Tests that are not yet commonly used such as the use of circulating tumor DNA to detect earlier recurrences may not be covered.

   The term 'biomarker' is a very vague term referring to any biologic measurement that informs us about a disease process. As such, there are a number of different ways biomarker testing can be performed. This can range from staining a cancer tissue specimen and looking at the slide under the microscopic to determine if it stains for certain proteins, to performing sequencing of the tumor's DNA or 'genetic code' to identify mutations that can be targeted.

   One of the most extraordinary new biomarkers from my perspective is HER2-low, which is performed by staining cancer cells for the HER2 protein. This has been performed routinely for years, but only patients with high levels of HER2 expression have previously benefited from HER2-directed therapies such as trastuzumab (Herceptin). However, in the past 6 months, a new study has shown that patients with low levels of HER2 can benefit from the antibody-drug conjugate trastuzumab deruxtecan (Enhertu). This drug acts as a smart bomb, delivering 8 molecules of chemotherapy to cells that express HER2, and can effectively treat cancers even when the HER2 expression is low or heterogeneous. What was extraordinary is that patients treated with this new therapy had their cancers controlled for twice as long when compared to traditional chemotherapy, so it is a really great breakthrough.

3. **How do the tests you speak of compare with the RGCC (Greek) test?**

I have not previously heard of the RGCC / Greek test, but doing some brief research, this appears to be a test based on circulating tumor cells to characterize mutations for a cancer or to detect cancer / cancer recurrence earlier. For characterizing mutations in a cancer with circulating tumor DNA, I typically use FoundationOne Liquid CDx or Guardant360 CDx. These tests are FDA approved to detect tumor mutations that can be targeted in breast, ovarian, prostate, and lung cancer. These approvals are based on the fact that large number of patients have had both their tumor and circulating DNA tested, and confirmed that the blood based tests detected the same mutations that were present on the tumor. As far as I am able to see, similar studies have not been conducted with the RGCC / Greek test, and I would suggest using the tests that have been most well studied and validated, where you can be confident that your results are reproducible and accurate.

4. **My understanding is that my oncologist does not use markers. Can you explain why some do not.**
   I believe this is referring to tumor markers. Tumor markers such as CA 15-3, CA 27-29 and CEA can be elevated in patients with breast cancer, but are neither sensitive or specific – in other words, some patients may have breast cancer and the tests will not be elevated, and some patients may have elevations unrelated to cancer. Because of the inaccuracies in these tests, they are not useful for identifying patients at risk for disease recurrence. In patients with known metastatic cancer, tumor markers can be useful if a cancer is difficult to measure with CT scans or other imaging tests. If your cancer is not measurable then oncologists will sometimes use tumor marker changes to determine if a treatment change is needed. However, if your cancer is measurable on imaging scans (which is most often the case in metastatic disease) then it is better to use the changes on imaging scans to make treatment decisions rather than tumor markers, and thus oncologists may not measure tumor markers.

   Circulating tumor DNA is a more accurate way to potentially identify early cancer recurrence. However, patients who have evidence of circulating tumor DNA at an early timepoint may remain cancer free for 5 years or longer. Ongoing studies are testing whether giving chemotherapy for patients who have positive testing for circulating tumor DNA can prevent recurrences. It might seem like a straightforward topic – if patients with circulating tumor DNA present are at a high risk for recurrence – why not give try some additional treatments? But the challenge is that all treatments have risks, which can at times be debilitating or fatal, and many oncologists do not want to offer treatments where it is unclear whether patients will benefit. There are many trials ongoing in this space and we will hopefully have more data in the near future, until then the ideal setting to have circulating tumor DNA measured would be as part of a clinical trial.

5. **Is biomarker testing the same thing as either genetic testing or Oncotype DX test of the tumor?**
   A ‘biomarker’ is any measurement of a biologic / disease process – so genetic testing and Oncotype DX are both biomarkers, but there are many others.

6. **Does being HER2 low (IHC 2+/FISH negative) impact the treatment someone would receive?**
   Yes – but currently only for patients with metastatic disease. Such patients are eligible for treatment with trastuzumab deruxtecan (Enhertu). There are ongoing studies looking at the use

of this treatment for early stage breast cancer, but currently the treatment is not approved for early stage disease. Also, please note that HER2-low is defined as IHC 1+ or 2+ (and FISH negative) – so patients with either of these results would be eligible.

7. **Interested in the discussion/opinion of PARP inhibitors and PALB2 mutation.**
   Very insightful question – PALB2 encodes a protein that interacts closely with BRCA2, and patients with mutations in PALB2 have similar cancers and those with BRCA mutations. In breast cancer, there are ongoing studies looking at the use of PARP inhibitors in patients with PALB2 germline mutations. Treatment of patients with metastatic breast cancer and a PALB2 mutation could therefore be candidates for clinical trials of PARP inhibitors, or if they have no alternative treatments available for such a patient and there is no trial available in their area, perhaps a PARP inhibitor could be requested through compassionate use. Additionally, there is an unanswered question about the use of PARP inhibitors in patients where they weren't born with a BRCA1/2 mutation, but their tumor developed the BRCA1/2 mutation independently. Studies are also looking at PARP inhibitors in such patients.

8. **What is your opinion about diet after a breast cancer diagnosis? Would you recommend really lowering sugar intake or does it not make any difference? Curious after the PET scan sugar portion**
   Cancer does uptake sugar more heavily than health tissue (aside from a few organs like brain, heart), allowing for detection on an FDG PET scan. However, most studies have shown minimal impact of specific diets on cancer outcomes – as although cancer takes up sugar, your normal healthy cells also need sugar, and there's not a good way to starve the cancer cells without starving the rest of the body. For patients undergoing cancer treatment, it is also important that patients avoid any excessively strict dietary changes, as malnourishment could impact our ability to give effective treatments. For patients in recovery after their cancer diagnosis, especially for hormone receptor positive cancers, there is some evidence that maintaining a healthy body weight may reduce risks of recurrence. But it is unclear that using one type of diet (i.e. low fat vs low carb vs other) to reach a health weight is better than any other.