Published in final edited form as:

Clin Nucl Med. 2017 December ; 42(12): 912–917. doi:10.1097/RLU.0000000000001820.

⁸⁹Zr-trastuzumab PET/CT for detection of HER2-positive metastases in patients with HER2-negative primary breast cancer

Gary A. Ulaner^{1,2}, David M. Hyman^{3,4}, Serge K. Lyashchenko^{1,2}, Jason S. Lewis^{1,2,5}, and Jorge A. Carrasquillo^{1,2}

¹Department of Radiology, Memorial Sloan Kettering Cancer Center, New York, NY, USA

²Department of Radiology, Weill Cornell Medical College, New York, NY, USA

³Department of Medicine, Memorial Sloan Kettering Cancer Center, New York, NY, USA

⁴Department of Medicine, Weill Cornell Medical College, New York, NY, USA

⁵Molecular Pharmacology Program, Memorial Sloan Kettering Cancer Center, New York, NY, USA

Abstract

Purpose—To determine if imaging with ⁸⁹Zr-trastuzumab, a human epidermal growth factor receptor 2 (HER2)-targeting PET tracer can detect HER2-positive metastases in patients with HER2-negative primary breast cancer.

Methods—As part of an Institutional Review Board (IRB)-approved, prospective clinical trial of ⁸⁹Zr-trastuzumab PET/CT (ClinicalTrials.gov identifier NCT02286843), a second group of 11 patients with HER2-negative primary breast cancer and known metastatic disease were recruited. Patients with confirmed HER2-negative primary breast cancer underwent ⁸⁹Zr-trastuzumab PET/CT to screen for ⁸⁹Zr-trastuzumab-avid lesions suspicious for unsuspected HER2-positive metastases. ⁸⁹Zr-trastuzumab-avid lesions on PET/CT were biopsied and pathologically examined to determine HER2 status.

Results—All 11 patients had confirmed HER2-negative primary breast cancer. Four demonstrated suspicious foci on ⁸⁹Zr-trastuzumab PET/CT. Of these four, one had biopsy-proven HER2-positive metastases. The other three patients with suspicious ⁸⁹Zr-trastuzumab-avid foci had biopsy demonstrating a metastasis that was HER2-negative, and were considered false-positive ⁸⁹Zr-trastuzumab PET foci. Combined with a published report of the first 9 patients, there have been a total of 20 HER2-negative primary breast cancer patients, with 3 patients (15%) having pathologically confirmed HER2-positive distant metastases and 6 (30%) with suspicious ⁸⁹Zr-trastuzumab-avid foci that were HER2-negative on pathology, which were thus considered false-positive ⁸⁹Zr-trastuzumab findings.

Corresponding Author: Gary A. Ulaner, MD, PhD, Memorial Sloan Kettering Cancer Center, 1275 York Ave, Box 77, New York, NY 10065, Phone: (212) 639-3776, Fax: (212) 717-3263. ulanerg@mskcc.org.

Conflict of Interest Disclosure Statement: The authors have no conflicts of interest to declare.

Conclusion—This second group of patients confirms the proof-of-concept that ^{89}Zr -trastuzumab PET/CT detects unsuspected HER2-positive metastases in a subset of patients with HER2-negative primary breast cancer. False-positive ^{89}Zr -trastuzumab-avid foci present a challenge to using this tracer.

INTRODUCTION

Approximately 20% of invasive ductal breast malignancies are classified as HER2-positive due to overexpression of the HER2 protein on the surface of tumor cells or gene amplification [1]. Classification of a breast malignancy as HER2-positive or -negative is an essential component of clinical care, as patients with HER2-positive breast cancer receive specific targeted HER2 therapies that reduce the risk of death, while patients with HER2-negative breast cancer do not [2, 3]. An important question is whether HER2 status could be heterogeneous between sites of malignancy in a single patient. If heterogeneity exists, then classification of HER2 status from one biopsy may inaccurately describe the extent of disease in a patient with the potential to respond to HER2-targeted therapy. Indeed, heterogeneity of tumors both within and across lesions in a single patient is increasingly being recognized [4]. Investigators have identified patients with HER2-positive primary malignancies in which metastases have lost HER2-positivity [5]. Potentially more clinically significant, investigators have identified patients with HER2-negative primary malignancies in which metastases have acquired HER2-positivity. These patients may benefit from HER2-targeted therapies that they are not receiving, as they are not considered to have HER2-positive disease.

It is currently unclear how to best identify patients who have malignancy with heterogeneous HER2 expression. Identification by biopsy would be difficult, as only small samples of tissue from a limited number of lesions are normally sampled. We hypothesized that HER2 heterogeneity may be identified by imaging with a HER2-targeting radiotracer, and designed a prospective clinical trial to evaluate HER2 heterogeneity with ^{89}Zr -trastuzumab PET/CT, which allows for visualization of HER2-positive malignancy [6]. We published the initial results from the first 9 patients and demonstrated the ability of ^{89}Zr -trastuzumab PET/CT to identify HER2-positive metastases in patients with HER2-negative primary breast malignancy [7]. Here we present the results of the second group of patients, comprising 11 patients.

MATERIALS AND METHODS

Patients

A single-center, prospective, IRB-approved protocol (ClinicalTrials.gov identifier NCT02286843) was performed. Patients receiving treatment for metastatic HER2-negative primary breast cancer were identified as potential candidates. Inclusion criteria were: 1) biopsy-proven HER2-negative primary malignancy; 2) biopsy-proven metastatic disease; 3) foci of demonstrable metastases on imaging modalities within six weeks of enrollment; 4) women age > 18 ; and 5) Eastern Cooperative Oncology Group performance score of 0–2. Exclusion criteria were: 1) creatinine $> 2 \times$ upper limit of normal; 2) Aspartate aminotransferase/Alanine aminotransferase $> 2 \times$ upper limit of normal; 3) life expectancy $<$

3 months; 4) pregnancy or lactation; and 5) inability to undergo PET/CT scanning because of weight limits. All patients enrolled in the protocol provided written informed consent.

Confirmation of HER2 Status of the Primary Breast Malignancy

The primary breast cancer tissue sample for each patient was retested for HER2 status to ensure proper classification as HER2-negative. HER2 protein overexpression was evaluated by immunohistochemistry (IHC) using an FDA-approved monoclonal antibody (clone 4B5, Ventana) directed against the internal domain of the c-erbB-2 oncoprotein (HER2). The IHC results were categorized as follows: 0, 1+ = negative result; 2+ = equivocal result; and 3+ = positive result, according to the published American Society of Clinical Oncology (ASCO) guidelines [8] (Table 1). Tissues with 2+ staining (equivocal) were assessed for HER2 amplification with fluorescence in situ hybridization (FISH) as per ASCO guidelines [8] using an FDA-approved probe set (*HER2* IQFISH pharmDx™, Dako), and defined as HER2/CEP17 ratio ≥ 2.0 . Carcinoma with 0 or 1+ IHC staining or 2+ IHC staining and concurrent negative HER2 FISH (HER2/CEP17 ratio < 2.0) were classified as HER2-negative primary breast malignancies.

⁸⁹Zr-Trastuzumab PET/CT

Patients with confirmed HER2-negative primary breast malignancy underwent ⁸⁹Zr-trastuzumab PET/CT. ⁸⁹Zr-trastuzumab is composed of the native trastuzumab, an FDA-approved HER2-targeting monoclonal antibody, conjugated with desferrioxamine and labelled with the positron-emitting metallo-radionuclide zirconium-89 (⁸⁹Zr). ⁸⁹Zr has a half-life of 78 hours, long enough to allow favorable biodistribution of radiolabeled intact antibodies. Our institution has an FDA Investigational New Drug for human ⁸⁹Zr-trastuzumab imaging (MSKCC Investigational New Drug #119907). ⁸⁹Zr-trastuzumab was produced under Good Manufacturing Practice conditions using previously described methodology [9]. The final drug product for human use was manufactured and quality control tested prior to being released for patient administration to ensure that it conformed to the established acceptance specifications for appearance, pH, endotoxin content, sterilizing filter integrity, radiochemical purity, and radionuclidic identity. Sterility and immunoreactivity determinations were performed post-release.

Patients were intravenously administered 185 MBq $\pm$ 10% of ⁸⁹Zr-trastuzumab over 5 minutes. Radiolabeled ⁸⁹Zr-trastuzumab was brought up to a final mass dose of 50 mg by adding non-radiolabeled trastuzumab at the end of the production, as this has been shown to help optimize tumor targeting [6]. The final mass dose of 50 mg was provided by the Radiochemistry & Molecular Imaging Probe Core at MSKCC to the clinic for patient administration. Five or six days following ⁸⁹Zr-trastuzumab administration, patients underwent PET/CT from the mid-skull to mid-thigh on a dedicated research GE Discovery PET/CT 710 scanner, with an 80mA CT component for attenuation correction and lesion localization. PET/CT images were reconstructed using iterative reconstruction and displayed in multiplanar reconstructions. ⁸⁹Zr-trastuzumab PET/CT scans were interpreted by two different nuclear medicine experts (GAU, JAC), both experienced in the use of PET antibody radiotracers. Both readers were able to compare ⁸⁹Zr-trastuzumab PET/CT results with prior imaging studies. Physiologic ⁸⁹Zr-trastuzumab uptake was expected in the blood pool, liver,

gallbladder, bowel, and kidney. Radiotracer uptake in areas that are not physiologic were graded both qualitatively and semi-quantitatively. For qualitative scoring, lesions were scored as suspicious or not suspicious. Only those foci qualitatively scored as suspicious by both readers were considered suspicious foci. Semi-quantitative analysis of tracer uptake was performed by recording maximum standardized uptake value (SUV) of suspicious lesions. Three-dimensional volumes of interest were placed in these areas and tracer uptake was quantified using SUV, calculated as decay-corrected mean region of interest activity ($\mu\text{Ci}/\text{ml}$) / (injected dose (μCi) / body weight (g)).

Biopsy and Pathologic Evaluation of Suspicious ^{89}Zr -Trastuzumab Foci

Image-guided biopsy was selected in concert with an experienced oncologic interventional radiologist to minimize risks to the patient while obtaining high-quality samples. Biopsy specimens were evaluated by board-certified breast pathology specialists. The IHC results were categorized according to published ASCO guidelines [8], as discussed above and in Table 1. Carcinoma with 3+ IHC staining or 2+ IHC staining and concurrent positive HER2 FISH were classified as HER2-positive metastases.

RESULTS

Patient Characteristics

Between December 2015 and May 2016, a second group of 11 women with HER2-negative primary invasive ductal breast cancer completed the study protocol. These were designated patients number 10 to 20, following the first 9 patients described in the initial publication [7]. Patient characteristics and study results are summarized in Table 2.

Sites of Distant Metastases at Time of Protocol Enrollment

All 11 patients had at least one site of metastatic disease at the time of protocol enrollment. The most common sites of distant metastases were bone (n=7), nodal (n=7), and liver (n=7), followed by lung (n=2), pleural (n=1), and chest wall (n=1). Nine of the eleven patients had multiple organ system metastatic involvement.

Confirmation of HER2 Status of the Primary Breast Malignancy

On IHC retesting of the patients' archived primary breast cancer specimens, all 11 patients had confirmed HER2-negative primary malignancies according to published ASCO guidelines [8]. Three patients had HER2 IHC scores of 0, while seven had HER2 IHC scores of 1+, and one had a HER2 IHC score of 2+ with a FISH of 1.4.

^{89}Zr -Trastuzumab PET/CT

All 11 patients underwent ^{89}Zr -trastuzumab PET/CT. Patients were monitored for 30 minutes after tracer injection and called the following day; no side effects were observed or reported. Vital signs were recorded before and after injection and no changes of clinical significance were observed.

In four of eleven patients (36%), both readers observed foci of ^{89}Zr -trastuzumab avidity considered suspicious for HER2-positive disease. In two patients, the suspicious ^{89}Zr -

trastuzumab foci were in pleura, while one patient exhibited suspicious ^{89}Zr -trastuzumab foci in bone, and one patient exhibited suspicious ^{89}Zr -trastuzumab foci in the liver.

Biopsy and Pathologic Evaluation of Suspicious ^{89}Zr -Trastuzumab Foci

Image-guided biopsy was performed in all four patients with suspicious ^{89}Zr -trastuzumab-avid foci.

Patient 11 was a 40-year-old woman who was diagnosed in October 2013 with an ER-positive / HER2-negative primary invasive ductal carcinoma. At the time of protocol enrollment, an FDG PET/CT demonstrated FDG-avid bone, liver, nodal, and chest wall malignancy. Bone and chest wall lesions had been pathologically confirmed as metastases. ^{89}Zr -trastuzumab PET/CT was performed in March 2016 and demonstrated ^{89}Zr -trastuzumab-avid osseous and nodal lesions. The most avid lesion was a right femoral focus (SUVmax 10.1). Biopsy of the right femoral focus demonstrated HER2 IHC of 2+ and FISH of 1.6. As FISH was < 2.0 , this was considered a false-positive ^{89}Zr -trastuzumab focus.

Patient 14 was a 47-year-old woman diagnosed in June 2011 with an ER-positive / HER2-negative primary invasive ductal carcinoma. At the time of protocol enrollment, an FDG PET/CT demonstrated FDG-avid liver, nodal, and pleural malignancy (Figure 1). Hepatic and pleural lesions had been pathologically confirmed as metastases. ^{89}Zr -trastuzumab PET/CT was performed in May 2016 and demonstrated ^{89}Zr -trastuzumab-avid pleural (SUV 6.9) and liver (SUV 10.9) lesions. The pleural lesion was selected for biopsy as the lesion with the lowest risk for sampling. Biopsy of the pleural lesion demonstrated HER2 IHC of 2+ and FISH of 2.4. As FISH was > 2.0 , this was considered a true-positive ^{89}Zr -trastuzumab focus for a HER2-positive distant metastasis.

Patient 17 was a 51-year-old woman diagnosed in November 2010 with an ER-positive / HER2-negative primary invasive ductal carcinoma. At the time of protocol enrollment, an FDG PET/CT demonstrated FDG-avid bone, nodal, and lung malignancy (Fig. 2). A lung lesion had been pathologically confirmed as metastasis. ^{89}Zr -trastuzumab PET/CT was performed in May 2016 and demonstrated ^{89}Zr -trastuzumab-avid lung (SUV 11.3) lesions. Biopsy of a lung lesion demonstrated HER2 IHC of 1+, which was considered a false-positive ^{89}Zr -trastuzumab focus.

Patient 20 was a 58-year-old woman diagnosed in August 2003 with an ER-positive / HER2-negative primary mixed invasive ductal and lobular carcinoma. At the time of protocol enrollment, an FDG PET/CT demonstrated FDG-avid bone, liver, and nodal malignancy (Fig. 3). A bone and liver lesions had been pathologically confirmed as metastases. ^{89}Zr -trastuzumab PET/CT was performed in June 2016 and demonstrated ^{89}Zr -trastuzumab-avid liver (SUV 15.7) lesions. Biopsy of a liver lesion demonstrated HER2 IHC of 1+, which was considered a false-positive ^{89}Zr -trastuzumab focus.

The remaining seven patients did not have suspicious foci of ^{89}Zr -trastuzumab avidity.

DISCUSSION

This study describes a second group of 11 patients in a prospective clinical trial of HER2-targeted imaging with ^{89}Zr -trastuzumab PET/CT to evaluate for unsuspected HER2-positive metastases in patients with HER2-negative primary breast cancer. It demonstrates that in a small subset of patients with HER2-negative primary breast cancer, HER2-targeted imaging can detect previously unsuspected HER2-positive disease.

Between the current group of 11 patients and a previously published initial group of 9 patients [7], a completed cohort of 20 patients with HER2-negative primary breast cancer have now been evaluated. In total, 3 of 20 patients (15%) were demonstrated to have biopsy-proven unsuspected HER2-positive metastases. In another 6 of 20 patients (30%), suspicious ^{89}Zr -trastuzumab-avid foci resulted in biopsies with HER2-negative pathology, which are being conservatively considered false-positive ^{89}Zr -trastuzumab-avid foci.

While medical imaging of oncology patients has traditionally been used for the detection of tumors, staging, and evaluation of treatment response, this study suggests that targeted medical imaging may be able to help select patients for targeted systemic therapies. Over 900,000 women are currently living with metastatic breast cancer, with more than 50,000 diagnosed each year [10]. Eighty percent of these women have HER2-negative primary malignancies. If 15% of patients with metastatic HER2-negative primary breast cancer could be found to harbor HER2-positive metastases, this would represent a current population of 135,000 women. Thus, targeted HER2 imaging has the potential to play a clinically valuable role in identifying patients who may benefit from HER2-targeted therapies.

A limitation of this approach is the currently high level of presumably false-positive suspicious ^{89}Zr -trastuzumab-avid foci. This leads to unneeded workup and biopsies. The reason for these false positive foci is unclear. One possibility is pathologic sampling or processing errors. For example, decalcification of osseous lesions may decrease the intensity of HER2 IHC staining, resulting in a false-negative pathologic result [16]. If a lesion exhibits heterogeneity of HER2 expression, it is possible that in any one biopsy, the HER2-positive malignancy was under-sampled. While this could result in erroneous pathologic analysis, given the high number of false positives, it is likely that at least some represent non-specific ^{89}Zr -trastuzumab uptake in HER2-negative lesions. Another possible explanation is free ^{89}Zr being released from its chelator during the long five-day uptake period. When loosely chelated, ^{89}Zr is known to be a bone seeker [11]. Indeed, evidence suggests that when ^{89}Zr is chelated to antibodies with desferrioxamine, radioactivity accumulates in the bone [12]. Thus, non-specific binding of potentially free ^{89}Zr at sites of osseous turnover associated with bone metastases could result in false-positive osseous foci. Another possible explanation for presumed false-positive ^{89}Zr -trastuzumab-avid foci could be related to *in vivo* HER2 internalization rates or affinity differences that would not necessarily be reflected by IHC or FISH [13]. More specific HER2-targeting radiotracers will need to be explored. One potential alternative is ^{89}Zr -pertuzumab, which demonstrated HER2 specificity in animal models [14].

HER2 negative patients have been reported response to trastuzumab therapy. It would be of interest to evaluate if patients with positive Zr-89 trastuzumab scans but negative HER2 status by conventional criteria respond to trastuzumab. This would raise the potential of using Zr-89 trastuzumab as a predictive biomarker [15] and selecting patients for HER2-target therapy based on imaging.

It is interesting to note that the intensity of ⁸⁹Zr-trastuzumab avidity did not discriminate between true-positive and false-positive lesions for HER2 expression. The most ⁸⁹Zr-trastuzumab-avid lesion in the study was a hepatic lesion (patient #20, SUV 15.7), which had HER2-negative pathology. Likewise, a substantially less ⁸⁹Zr-trastuzumab-avid lesion (patient #14, SUV 6.9) had HER2-positive pathology and was a true positive.

Another interesting issue was that even when a ⁸⁹Zr-trastuzumab-avid focus was considered a false positive, there could be noticeable heterogeneity in HER2 expression between the primary malignancy and the avid metastasis. For example, patient #11 had a primary malignancy with an IHC of 0. The ⁸⁹Zr-trastuzumab-avid osseous metastasis that was biopsied had an IHC of 2+ and FISH of 1.6. While IHC 2+ / FISH 1.6 is considered HER2-negative by ASCO guidelines, it still represents a change from the primary malignancy. The value of ⁸⁹Zr-trastuzumab PET/CT for evaluating this type of heterogeneity needs further evaluation.

The strength of this study is its design as a prospective clinical trial, while its weakness is the still small sample size. Ethical standards and logistical obstacles prevent biopsy of all ⁸⁹Zr-trastuzumab foci; thus, pathology is classified on the basis of a limited number of biopsies. Likewise, ethical standards prevent biopsy confirmation of HER2-negative status of metastases in patients without suspicious foci on ⁸⁹Zr-trastuzumab imaging.

In conclusion, ⁸⁹Zr-trastuzumab PET/CT can be used to detect unsuspected HER2-positive metastases in a subset of patients with HER2-negative primary breast cancer. This demonstrates that targeted imaging can identify patients amenable to targeted therapies. Presumed false-positive results currently limit the translation of these techniques, but there is great potential for more specific methodology to have substantial clinical value in the future.

Acknowledgments

We acknowledge funding from a Department of Defense Breast Cancer Research Program Breakthrough Award BC132676 (GAU) and a Genentech research grant (GAU). The authors gratefully acknowledge the MSKCC Radiochemistry and Molecular Imaging Probe Core (NIH grant P30 CA08748) for additional support.

Financial Support: Department of Defense Breast Cancer Research Program Breakthrough Award BC132676 (GAU) and Genentech research grant (GAU). The authors gratefully acknowledge the MSKCC Radiochemistry and Molecular Imaging Probe Core (NIH grant P30 CA08748) for additional support.

References

1. Slamon DJ, Clark GM, Wong SG, et al. Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. *Science*. 1987; 235:177–182. [PubMed: 3798106]
2. Slamon DJ, Leyland-Jones B, Shak S, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. *N Engl J Med*. 2001; 344:783–792. [PubMed: 11248153]

3. Romond EH, Perez EA, Bryant J, et al. Trastuzumab plus adjuvant chemotherapy for operable HER2-positive breast cancer. *N Engl J Med*. 2005; 353:1673–1684. [PubMed: 16236738]
4. McGranahan N, Swanton C. Biological and therapeutic impact of intratumor heterogeneity in cancer evolution. *Cancer Cell*. 2015; 27:15–26. [PubMed: 25584892]
5. Niikura N, Liu J, Hayashi N, et al. Loss of human epidermal growth factor receptor 2 (HER2) expression in metastatic sites of HER2-overexpressing primary breast tumors. *J Clin Oncol*. 2012; 30:593–599. [PubMed: 22124109]
6. Dijkers EC, Oude Munnink TH, Kosterink JG, et al. Biodistribution of ⁸⁹Zr-trastuzumab and PET imaging of HER2-positive lesions in patients with metastatic breast cancer. *Clin Pharmacol Ther*. 2010; 87:586–592. [PubMed: 20357763]
7. Ulaner GA, Hyman DM, Ross DS, et al. Detection of HER2-Positive Metastases in Patients with HER2-Negative Primary Breast Cancer Using ⁸⁹Zr-Trastuzumab PET/CT. *J Nucl Med*. 2016; 57:1523–1528. [PubMed: 27151988]
8. Wolff AC, Hammond ME, Hicks DG, et al. Recommendations for Human Epidermal Growth Factor Receptor 2 Testing in Breast Cancer: American Society of Clinical Oncology/College of American Pathologists Clinical Practice Guideline Update. *J Clin Oncol*. 2013
9. Vosjan MJ, Perk LR, Visser GW, et al. Conjugation and radiolabeling of monoclonal antibodies with zirconium-89 for PET imaging using the bifunctional chelate p-isothiocyanatobenzyl-desferrioxamine. *Nat Protoc*. 2010; 5:739–743. [PubMed: 20360768]
10. Johnson RH, Chien FL, Bleyer A. Incidence of breast cancer with distant involvement among women in the United States, 1976 to 2009. *JAMA*. 2013; 309:800–805. [PubMed: 23443443]
11. Abou DS, Ku T, Smith-Jones PM. In vivo biodistribution and accumulation of ⁸⁹Zr in mice. *Nucl Med Biol*. 2011; 38:675–681. [PubMed: 21718943]
12. Deri M, Ponnala S, Kozlowski P, et al. p-SCN-BN-HOPO: A Superior Bifunctional Chelator for Zr ImmunoPET. *Bioconjug Chem*. 2015
13. Rudnick SI, Lou J, Shaller CC, et al. Influence of affinity and antigen internalization on the uptake and penetration of Anti-HER2 antibodies in solid tumors. *Cancer Res*. 2011; 71:2250–2259. [PubMed: 21406401]
14. Marquez BV, Ikotun OF, Zheleznyak A, et al. Evaluation of (⁸⁹)Zr-pertuzumab in Breast cancer xenografts. *Mol Pharm*. 2014; 11:3988–3995. [PubMed: 25058168]
15. Ulaner GA, Riedl CC, Dickler MN, et al. Molecular Imaging of Biomarkers in Breast Cancer. *J Nucl Med*. 2016; 57(Suppl 1):53S–59S. [PubMed: 26834103]

Figure 1. 47-year-old woman with primary ER-positive, HER2-negative invasive ductal breast carcinoma and known metastases in the liver, nodes, and pleura. (A) Axial CT and PET images from a contrast-enhanced FDG PET/CT through the chest demonstrates FDG-avid right pleural masses (SUVmax 6.0, arrows). (B) Axial CT and PET images from a non-contrast-enhanced ⁸⁹Zr-trastuzumab PET/CT at the same level demonstrates ⁸⁹Zr-trastuzumab avidity in the pleural lesions (SUVmax 6.9, arrows).

Figure 2.

51-year-old woman with primary ER-positive, HER2-negative invasive ductal breast carcinoma. (A) FDG PET MIP demonstrates multiple FDG-avid metastases, which correspond to lesions in the lung (arrow), nodes (arrowhead), and bone (curved arrow). (B) Axial CT and PET images from a contrast-enhanced FDG PET/CT through the chest, displayed in lung CT windows, demonstrate an FDG-avid left lung mass (SUVmax 7.3, arrow). (C) ^{89}Zr -trastuzumab PET MIP demonstrates physiologic tracer in the blood pool, liver, and bowel, as well as a suspicious ^{89}Zr -trastuzumab-avid focus in the upper chest (arrow). (D) Axial CT and PET images from a non-contrast-enhanced ^{89}Zr -trastuzumab PET/CT demonstrate that the suspicious ^{89}Zr -trastuzumab-avid focus corresponds to the left lung mass (SUVmax 11.3, arrowhead).

Figure 3.

58-year-old woman with primary ER-positive, HER2-negative invasive ductal breast carcinoma and known metastases in the bone, liver, and nodes. (A) Axial CT and PET images from a non-contrast-enhanced FDG PET/CT through the abdomen demonstrates an FDG-avid liver metastasis (SUVmax 5.6, arrows). (B) Axial CT and PET images from a non-contrast-enhanced ⁸⁹Zr-trastuzumab PET/CT at the same level demonstrates ⁸⁹Zr-trastuzumab avidity in the liver metastasis (SUVmax 15.7, arrows).

Table 1

Algorithm for defining HER2 expression *

First, HER2 testing is performed with IHC. IHC results:	
0 or 1+	HER2-negative
2+	Equivocal
3+	HER2-positive
If IHC is 2+ (equivocal), then dual-probe HER2 FISH is performed. FISH results:	
HER2/CEP17 ratio $\geq$ 2.0	HER2-positive
HER2/CEP17 ratio < 2.0	HER2-negative

CEP17 = chromosome enumeration probe 17, FISH = fluorescence in situ hybridization, HER2 = Human epidermal growth factor receptor 2, IHC = immunohistochemistry.

*
as per ASCO guidelines [8]

Table 2

Patient demographics and study results

Patient	Age	Sites of distant metastases at enrollment	Confirmatory IHC of primary breast cancer	Suspicious foci on ⁸⁹ Zr- trastuzumab PET/CT?	Image-guided biopsy results
10	54	Bone, Nodes	HER2 1+	None	
11	40	Bone, Liver, Nodes, Chest Wall	HER2 0	Bone (SUV 10.1) and Nodal (SUV 5.1)	HER2 IHC 2+ FISH 1.6
12	56	Liver	HER2 1+	None	
13	43	Nodes, Lung	HER2 1+	None	
14	47	Liver, Nodes, Pleura	HER2 2+ FISH 1.4	Liver (SUV 10.9) and Pleura (SUV 6.9)	HER2 IHC 2+ FISH 2.4
15	50	Bone, Liver	HER2 1+	None	
16	49	Bone, Liver	HER2 0	None	
17	51	Bone, Nodes, Pleura	HER2 0	Lung (SUV 11.3)	HER2 1+
18	48	Bone, Liver, Nodes	HER2 1+	None	
19	60	Liver	HER2 1+	None	
20	58	Bone, Liver, Nodes	HER2 1+	Liver (SUV 15.7)	HER2 1+

Patients 11–20, all with primary invasive ductal breast cancer, are reported here. The first 9 patients were reported in the initial results of the trial [7] HER2 = Human epidermal growth factor receptor 2, IHC = Immunohistochemistry, FISH = Fluorescence In Situ Hybridization.