

RESEARCH ARTICLE

Blood Flow and Glucose Metabolism in Stage IV Breast Cancer: Heterogeneity of Response During Chemotherapy

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Abstract

Objective: The purpose of the study was to compare early changes in blood flow (BF) and glucose metabolism (MR_{glu}) in metastatic breast cancer lesions of patients treated with chemotherapy.

Methods: Eleven women with stage IV cancer and lesions in breast, lymph nodes, liver, and bone were scanned before treatment and after the first course of chemotherapy. BF, distribution volume of water (V_d), MR_{glu}/BF ratio, MR_{glu} and its corresponding rate constants K_1 and k_3 were compared per tumor lesion before and during therapy.

Results: At baseline, mean BF and MR_{glu} varied among different tumor lesions, but mean V_d was comparable in all lesions. After one course of chemotherapy, mean MR_{glu} decreased in all lesions. Mean BF decreased in breast and node lesions and increased in bone lesions. V_d decreased in breast and nodes, but did not change in bone lesions. The MR_{glu}/BF ratio decreased in breast and bone lesions and increased in node lesions. In patients with multiple tumor lesions BF and MR_{glu} response could be very heterogeneous, even within similar types of metastases. BF and MR_{glu} increased in lesions of patients who experienced early disease progression or showed no response during clinical follow-up.

Conclusion: BF and MR_{glu} changes separately give unique information on different aspects of tumor response to chemotherapy. Changes in BF and MR_{glu} parameters can be remarkably heterogeneous in patients with multiple lesions.

Key words: Positron emission tomography, Blood flow, Glucose metabolism, Breast cancer, Metastases

Introduction

Stage IV breast cancer is considered incurable. Treatment is directed towards palliation of symptoms and disease stabilization, and with the use of polychemotherapy and taxanes, overall survival can be prolonged [1–2]. Conven-

tionally, clinical response is evaluated after several cycles of chemotherapy by monitoring changes in tumor dimension as determined by physical examination and (anatomical) imaging techniques. There are certain tumor-specific characteristics that play a role in responsiveness to therapy. For example, high glucose metabolism and high degree of (neo) vascularization correlate with poor prognosis, and vascularization and perfusion are important factors in growth and

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metastasizing potential of breast tumors [3–5]. On the other hand, poor tumor perfusion may hamper delivery of intravenous therapy and may lead to hypoxia, which is known to contribute to resistance to standard radiotherapy and chemotherapy and is associated with poor prognosis [6–7]. Overall clinical response rates following chemotherapy, as measured conventionally, vary between 20% and 80%, depending on the (combination of) agent used [2, 8]. This means that many patients are exposed to the morbidity, side effects, and costs of ineffective therapy. There is evidence that measurement of metabolic changes using positron emission tomography (PET) might be a sensitive method to assess early response to treatment [9–13]. The ultimate goal of using PET would be to guide effective treatment to responders, while avoiding unnecessary side effects in nonresponders.

To date, most PET studies in breast cancer assessing response have involved patients with locally advanced breast cancer (LABC) [9–15], and only a few focused on metastatic breast cancer [16–18]. The change emphasizes response assessment. At present, the most commonly used tracer in response monitoring studies is the glucose analogue 2-deoxy-2-[F-18]fluoro-D-glucose (FDG), which measures glucose metabolism. An alternative approach is to measure tumor perfusion *in vivo* using $H_2^{15}O$, a freely diffusible, metabolically inert tracer [18]. Both FDG and $H_2^{15}O$ could provide, either by themselves or combined, better insight into tumor behavior both prior to and during chemotherapy, e.g., by identifying factors that may play a role in responsiveness [13, 14, 19].

Thus, the primary aim of this pilot study was to compare early changes in blood flow (BF) and glucose metabolism (MR_{glu}) in metastatic breast cancer lesions of patients treated with chemotherapy.

Materials and Methods

Patients

Eleven patients with stage IV breast cancer who were scheduled to receive chemotherapy were included in this study. Mean age \pm SD was 55 \pm 9 years (range 42–72 years). All patients had an Eastern Cooperative Oncology Group (ECOG) performance status ranging from 0–2. Metastases were at least 2 cm in diameter and located sufficiently near the heart to make use of an image-derived arterial input function. Metastases were biopsy proven or diagnosed by appropriate image modalities (bone scan, abdominal ultrasound, or abdominal CT). Bone metastases were characterized as lytic, blastic, or mixed based on their appearance on X-ray or CT. Exclusion criteria were pregnancy, diabetes, claustrophobia, and chemotherapy within 6 weeks prior to the study. In addition, bone lesions that had received prior selective radiotherapy were excluded as reference lesions.

PET scans were performed within 5 days before the first and second course of chemotherapy, respectively. Regimens included FAC (5-fluorouracil 500 mg/m², cyclophosphamid 500 mg/m², and doxorubicin 50 mg/m² 3-weekly), Doc (docetaxel 100 mg/m² 3-weekly), AT (doxorubicin 50 mg/m² and docetaxel 100 mg/m² 3-

weekly), CAT (cisplatinum 50 mg/m², doxorubicin 50 mg/m², paclitaxel 100 mg/m² 3-weekly), and Vinorelbine (25 mg/m² weekly). The Medical Ethics Committees of the VU University Medical Centre and Amstelland Hospital approved the study.

Tumor Response and Clinical Follow-Up

Referring physicians were blinded to results of the second PET scan. After the second PET scan, patients returned to regular follow-up with their treating physician. Clinical endpoint was time to progression (TTP). Disease progression was defined by the treating physician, as directed by symptoms, blood tests, and/or physical examination during follow-up using appropriate imaging modalities, i.e., ultrasound or CT for liver lesions and bone scan, CT, or MRI for bone lesions. Progression of lymph nodes and liver metastases was defined by the RECIST criteria [20]. For bone metastases, any increase in size of known metastases or the appearance of new metastases was interpreted as progression.

Acquisition Protocol and Image Processing

All PET scans were performed on an ECAT EXACT HR+ scanner (Siemens/CTI, Knoxville, TN, USA), which consists of 32 rings of bismuth germinate crystal detectors and has a 15.5-cm axial field of view. Patients fasted for at least 6 h prior to scanning. Following a 10-min transmission scan to correct the subsequent emission scans for photon attenuation, 1,100 MBq of $H_2^{15}O$ was injected intravenously, simultaneously starting a 10-min dynamic emission scan (12 \times 5 s, 12 \times 10 s, 6 \times 20 s and 10 \times 30 s frames). Next, a second dynamic emission scan with a total duration of 60.5 min (1 \times 30 s, 6 \times 5 s, 6 \times 10 s, 3 \times 20 s, 5 \times 30 s, 5 \times 60 s, 8 \times 150 s, 6 \times 300 s frames) was started, with an intravenous injection of 370 MBq of FDG at the start of the second frame. The 30 s background frame was used to correct for any residual ^{15}O activity remaining from the $H_2^{15}O$ scan. *All data were acquired in 2D mode.* During the FDG scan venous samples were collected at 35, 45, and 55 min postinjection for measurement of plasma glucose and for quality control of the image-derived input function [21]. All PET data were corrected for decay, dead time, scatter, random coincidences, and measured photon attenuation. All scans were reconstructed as 128 \times 128 matrices using filtered back projection (FBP) with a 0.5 Hanning filter, resulting in a transaxial spatial resolution of \sim 7 mm in the centre of the field of view. For definition of tumor regions of interest (ROIs), summed images of the FDG scan and $H_2^{15}O$ scan were reconstructed using ordered subset expectation maximization (OSEM), which was followed by 5 mm Gaussian smoothing, resulting in the same transaxial spatial resolution as for the FBP images [22].

PET Data Analysis

Arterial input curves were derived from 15 mm ROIs placed over the aorta, left atrium, and left ventricle [23], using the FBP images of the fourth and fifth frames for the FDG scan [24] and the first 12 frames for the $H_2^{15}O$ scan [25].

All tumor ROIs were defined on OSEM reconstructed images of the FDG scan. These ROIs were used to generate time-activity curves from the dynamic frames of both the FDG and $H_2^{15}O$ scans. ROIs were drawn using a semiautomatic region-growing method, including only pixels with a cutoff of 75% of the maximum activity within a lesion [26].

For $H_2^{15}O$, the standard single-tissue compartment model was used [18]. Blood flow (BF) was not calculated for liver lesions because estimates using the single-tissue compartment model may not be reliable for the liver [27]. Fitting was performed both with and without an arterial blood volume component, and the best fit was determined by Akaike and Schwarz criteria, as described previously [24].

For FDG, Patlak graphical analysis [28] with an acquisition interval of 10–60 min p.i. was used to calculate glucose metabolism (MR_{glu}). The two-tissue compartment model with three rate constants [24, 29] was used in addition, in order to acquire more detailed information on delivery (K_1) and phosphorylation (k_3) of FDG.

Finally, the MR_{glu} to BF ratio was calculated, a metabolic parameter introduced by Mankoff et al. as an indirect indicator of glucose use relative to glucose delivery and used as a predictor of macroscopic complete response [13]. This ratio is proportional to $K_1/BF (= \text{glucose extraction fraction}) \times k_3/(k_2+k_3)$.

Statistics

For all lesions, mean \pm standard deviation (SD) was calculated for MR_{glu} , rate constants K_1 and k_3 , BF, V_d , and MR_{glu}/BF before treatment and after one course of chemotherapy. Differences in parameters before and after chemotherapy were tested for significance using the two-tailed Student *t*-test. Pearson correlation coefficients were estimated between BF and MR_{glu} , between BF, K_1 , and k_3 , and between the MR_{glu}/BF ratio, K_1 and k_3 , both before and during treatment. Multilevel analysis was performed to correct for multiple lesions within the same patient. A *p* value < 0.05 was considered statistically significant.

Results

Baseline

All patients were scanned twice (at baseline and after one course of therapy) with FDG and $H_2^{15}O$, with the exception of one patient for whom $H_2^{15}O$ was not available on the second scan for technical reasons. The same number of lesions was identified on the FDG and $H_2^{15}O$ scans. Uptake was adequate (moderate to high) in baseline lesions, so no conventional

imaging was necessary to help identify these lesions. On the baseline scans, 45 measurable lesions were found: eight breast tumors, 17 lymph nodes, 15 bone, and five liver metastases. Lymph node metastases included axillary and supraclavicular nodes. The median follow-up period was 13 months. Patient characteristics are summarized in Table 1.

Baseline MR_{glu} in normal breast was $0.01 \pm 0.004 \mu\text{mol}\cdot\text{ml}^{-1}\cdot\text{min}^{-1}$ vs. $0.11 \pm 0.07 \mu\text{mol}\cdot\text{ml}^{-1}\cdot\text{min}^{-1}$ in breast tumors ($p < 0.03$). MR_{glu} was comparable in breast tumors and lymph node metastases, and higher in bone and liver metastases (Table 2). BF and V_d values used in this study were estimated without an arterial blood volume component, as both Akaike and Schwarz criteria indicated that inclusion of arterial blood volume did not significantly improve the quality of the fits. Normal breast had a mean baseline BF of 0.04 ± 0.03 vs. $0.43 \pm 0.23 \text{ ml}\cdot\text{ml}^{-1}\cdot\text{min}^{-1}$ in breast tumors ($p < 0.02$). Mean BF was highest in node metastases ($0.60 \pm 0.27 \text{ ml}\cdot\text{ml}^{-1}\cdot\text{min}^{-1}$) and lowest in bone lesions ($0.41 \pm 0.24 \text{ ml}\cdot\text{ml}^{-1}\cdot\text{min}^{-1}$). V_d was $0.13 \pm 0.06 \text{ ml}\cdot\text{ml}^{-1}$ in normal breast tissue. V_d values in breast lesions were similar to those in nodal and bone lesions. The MR_{glu}/BF ratio was lowest in node lesions (0.23 ± 0.15), followed by breast tumors (0.31 ± 0.19) and bone lesions (0.41 ± 0.29).

Chemotherapy-Induced Changes

The same 45 baseline lesions were identified on the FDG scans after one course of therapy. One patient with bilateral breast lesions and ten node metastases could only be scanned with FDG the second time, however, so 33 lesions were evaluable on the post-therapy $H_2^{15}O$ scans of ten women: six breast tumors, seven lymph nodes, 15 bone, and five liver metastases.

After one cycle of chemotherapy, mean MR_{glu} decreased in all lesions. Compared with baseline, these reductions were statistically significant for breast tumors, nodes, and bone lesions and of borderline significance for liver lesions (see Table 2).

Mean BF decreased in all lesions except bone metastases (Table 2 and Fig. 1a). Mean V_d decreased in breast tumors

Table 1. Patient characteristics

| Patient no. | Age | Histology | Tumor lesions | Therapy | TTP (months) | TTD (months) |
|-------------|-----|-----------|---|---------|--------------|--------------|
| 1 | 72 | Ductal | Bone (lytic) | FAC | 6 | 9 |
| 2 | 42 | Lobular | Liver | Vino | 11 | 14 |
| 3 | 47 | Ductal | Supraclavicular nodes | Doc | 2 | 2 |
| 4 | 42 | Adeno | Liver | Doc | 5 | 20 |
| 5 | 53 | Lobular | Breast ^a , axillary nodes | AT | 4 | 5 |
| 6 | 58 | Adeno | Breast, bone (mixed) | CAT | NER | NER |
| 7 | 65 | Lobular | Breast ^b , supraclavicular nodes | FAC | 5 | 6 |
| 8 | 55 | Lobular | Breast, axillary nodes, bone (mixed) | Vino | 4 | 26 |
| 9 | 59 | Adeno | Breast, cervical nodes, bone (sclerotic) | FAC | 12 | 24 |
| 10 | 49 | Ductal | Breast, axillary nodes, bone (lytic), liver | Doc | 4 | 13 |
| 11 | 58 | Ductal | bone (mixed) | FAC | 9 | 13 |

TTP Time to progression, TTD time till death, NER no evidence of recurrence, ductal ductal carcinoma, lobular lobular carcinoma, adeno adenocarcinoma, FAC 5-fluorouracil, cyclophosphamid, doxorubicin, Doc docetaxel, AT doxorubicin, docetaxel, CAT cisplatinum, doxorubicin, paclitaxel, Vino Vinorelbine

^aMultifocal tumor

^bBilateral tumor

Table 2. PET parameters before and after one course of chemotherapy for normal breast and tumor lesions

| Parameter | Baseline mean (\pm SD) | After 1 \times CTh mean (\pm SD) | <i>p</i> |
|---------------------------------------|---------------------------|---------------------------------------|----------|
| MR _{glu} (μ mol/ml/min) | | | |
| Normal breast | 0.01 \pm 0.004 | 0.01 \pm 0.004 | 0.97 |
| Breast tumor | 0.11 \pm 0.07 | 0.07 \pm 0.04 | 0.002 |
| Nodes | 0.11 \pm 0.04 | 0.06 \pm 0.06 | <0.001 |
| Bone metastases | 0.15 \pm 0.10 | 0.12 \pm 0.09 | 0.003 |
| Liver metastases | 0.19 \pm 0.09 | 0.11 \pm 0.04 | 0.06 |
| K_1 (ml/ml/min) | | | |
| Normal breast | 0.02 \pm 0.01 | 0.02 \pm 0.01 | 0.59 |
| Breast tumor | 0.11 \pm 0.05 | 0.13 \pm 0.07 | 0.41 |
| Nodes | 0.18 \pm 0.09 | 0.13 \pm 0.12 | 0.23 |
| Bone metastases | 0.16 \pm 0.08 | 0.24 \pm 0.13 | 0.25 |
| Liver metastases | 0.26 \pm 0.20 | 0.25 \pm 0.10 | 0.26 |
| K_3 (min ⁻¹) | | | |
| Normal breast | 0.02 \pm 0.01 | 0.01 \pm 0.01 | 0.42 |
| Breast tumor | 0.05 \pm 0.03 | 0.03 \pm 0.02 | 0.05 |
| Nodes | 0.07 \pm 0.04 | 0.05 \pm 0.02 | 0.11 |
| Bone metastases | 0.06 \pm 0.04 | 0.05 \pm 0.03 | 0.18 |
| Liver metastases | 0.10 \pm 0.09 | 0.03 \pm 0.02 | 0.11 |
| BF (ml/ml/min) | | | |
| Normal breast | 0.04 \pm 0.03 | 0.04 \pm 0.02 | 0.88 |
| Breast tumor | 0.43 \pm 0.23 | 0.35 \pm 0.15 | 0.09 |
| Nodes | 0.60 \pm 0.27 | 0.21 \pm 0.17 | 0.15 |
| Bone metastases | 0.41 \pm 0.24 | 0.51 \pm 0.29 | 0.26 |
| V_d (ml/ml) | | | |
| Normal breast | 0.13 \pm 0.06 | 0.10 \pm 0.07 | 0.95 |
| Breast tumor | 0.69 \pm 0.17 | 0.63 \pm 0.28 | 0.29 |
| Nodes | 0.73 \pm 0.19 | 0.54 \pm 0.29 | 0.17 |
| Bone metastases | 0.67 \pm 0.21 | 0.67 \pm 0.30 | 0.77 |
| MR _{glu} /BF (μ mol/ml) | | | |
| Normal breast | 0.17 \pm 0.16 | 0.14 \pm 0.08 | 0.75 |
| Breast tumor | 0.31 \pm 0.19 | 0.21 \pm 0.03 | 0.06 |
| Nodes | 0.23 \pm 0.15 | 0.32 \pm 0.21 | 0.79 |
| Bone metastases | 0.41 \pm 0.29 | 0.26 \pm 0.15 | 0.05 |

SD Standard deviation, CTh chemotherapy

and nodes and remained unchanged in bone lesions (Fig. 1b). Compared with baseline, however, changes in BF and V_d were not statistically significant (Table 2).

There was an increase in K_1 in breast tumors and bone lesions and a decrease in nodes and liver lesions, while k_3 decreased in all lesions. Except for the decrease in k_3 in breast lesions ($p < 0.05$), changes in K_1 and k_3 were not statistically significant in other lesions.

Changes in the MR_{glu}/BF ratio were only significant for bone lesions ($p < 0.05$), although they approached significance for breast tumors ($p = 0.06$).

Correlation Between Parameters

Correlation between baseline MR_{glu} and BF was moderate for breast lesions ($r = 0.79$, Fig. 2 and Table 3), and even lower for nodes and bone metastases ($r = 0.64$ and 0.49 , respectively). Excluding the 12 lesions from the patient with bilateral breast lesions and ten node metastases that did not receive the second H₂¹⁵O scan from the baseline, analysis resulted in only moderately lower correlation values ($r = 0.76$ and $r = 0.51$ for breast and nodes, respectively). After therapy, correlation between MR_{glu} and BF improved dramatically for breast

lesions ($r = 0.98$), but remained similar for nodes and bone metastases ($r = 0.62$ and $r = 0.50$, respectively).

At baseline, correlation between BF and K_1 or BF and k_3 was weak to moderate for breast lesions ($r = 0.43$ and $r = 0.58$, respectively), but after chemotherapy, there was a significant improvement in correlation ($r = 0.86$, $p < 0.03$ and $r = 0.94$, $p < 0.005$, respectively). In contrast, in bone lesions, correlation between BF and K_1 was similar before and after therapy ($r = 0.62$ and $r = 0.59$, respectively), while the modest baseline correlation between BF and k_3 ($r = 0.52$) was lost after therapy ($r = 0.15$). Because of the small number of lymph nodes after therapy, changes in correlation between BF and MR_{glu} parameters should be viewed with caution (see Table 3).

Inter-lesion Heterogeneity

Changes in BF and MR_{glu} after the first course of chemotherapy were often remarkably heterogeneous within one patient, not only between different types of lesions, but even within similar lesions. Fig. 3 shows examples of typical heterogeneous response in two patients with multiple lesions. Similarly, heterogeneous response was observed in patient nos. 6, 8, and 10.

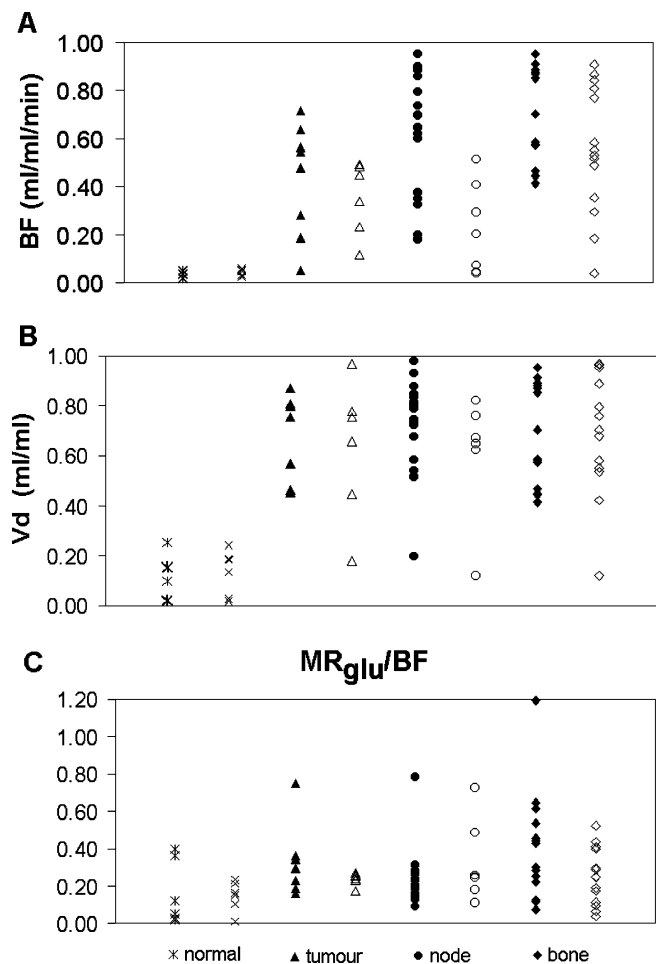


Fig. 1. BF (a), V_d (b), and MR_{glu}/BF (c) at baseline (bold symbols) and after one course of chemotherapy (open symbols) for normal breast and (metastatic) breast cancer lesions.

PET Parameters vs. Clinical Outcome

Because ten of 11 patients showed disease progression during clinical follow-up, patients were subdivided further into those that showed early disease progression (TTP \geq 6 months) vs. those with late (TTP > 6 months) or no progression. Seven patients showed early disease progression. In three patients, TTP was longer than 6 months, and one patient was still in clinical remission at the end of follow-up.

Mean MR_{glu}/BF was lower in patients with late or no progression than in patients with early progression (0.24 ± 0.11 vs. 0.34 ± 0.21), but this difference was not statistically significant ($p=0.21$). Similarly, differences between baseline and post-therapy values of BF, MR_{glu}, and V_d in patients with early progression vs. patients with late or no progression were not statistically significant (data not shown).

When assessed on a lesion basis (Fig. 4), however, MR_{glu} and BF increased only in lesions of patients who progressed early. In contrast, no increase in MR_{glu} and BF was seen in

any lesion of patients with TTP > 6 months. In these patients, MR_{glu} and BF decreased $-32\% \pm 21\%$ (range -8% to -69%) and $-22\% \pm 19\%$ (range -6% to -60%), respectively. No predictive pattern as to the occurrence of progression could be found in changes in V_d or MR_{glu}/BF (Fig. 4).

Discussion

The primary aim of this pilot study was to compare early chemotherapy-induced changes in blood flow and in glucose metabolism in metastatic breast cancer lesions.

The present study is, to our knowledge, the first to report BF and V_d values for regional and distant breast cancer metastases. Baseline MR_{glu}, BF, and V_d were different among different tumor lesions. MR_{glu} and the associated microparameters K_1 and k_3 were highest in liver metastases. MR_{glu} was lowest (and similar) in breast and node lesions. Baseline BF values were comparable in breast and bone lesions, and on average higher in nodes. In contrast, V_d was comparable among all three lesions.

Mean BF values in breast tumors were higher than previously reported in patients with (locally advanced) breast cancer, namely, 0.43 ± 0.23 ml·ml⁻¹·min⁻¹ in the present study vs. 0.32 ml·ml⁻¹·min⁻¹ [13] and 0.30 ml·ml⁻¹·min⁻¹ [18] in two earlier studies. Interestingly, Wilson et al. [18] did report a mean tumor BF of 0.42 ml·ml⁻¹·min⁻¹ in a subset of patients with Mx or M1 disease, comparable with values in the present study, and higher than values in their patients with M0 disease (0.24 ml·ml⁻¹·min⁻¹). It is known that tumor growth, progression, and metastagenicity require angiogenesis [30, 31]. In fact, microvessel density has been shown to be significantly higher in breast tumors of patients with metastases, than in those without [32]. The reason for the higher BF values in breast tumors of patients with metastatic disease could be due to this phenomenon, signifying a change in tumor biology (higher vascularization) associated with increased metastatic potential. With a mean value of 0.41 ± 0.24 ml·ml⁻¹·min⁻¹, BF in bone lesions was much higher than that reported for normal bone marrow, namely, 0.11 – 0.18 ml·ml⁻¹·min⁻¹ [33].

After chemotherapy, correlation between BF and MR_{glu} increased dramatically for breast lesions, while in bone lesions, correlation was similar before and after therapy. In breast lesions, both k_3 and K_1 contributed to this improvement in correlation, since both showed a significantly higher correlation with BF after therapy, although this was more pronounced for k_3 ($r=0.94$ vs. $r=0.83$, Table 3). This is partly confirmed by the study by Tseng et al. [34] involving chemotherapy-treated LABC patients, who suggested that the phosphorylation step (k_3) was responsible for the improved correlation they observed after chemotherapy.

BF and K_1 did not correlate well before therapy (Table 3), in contrast to findings of Zasadny et al. [19] and Tseng et al. [34]. Both proposed that BF could possibly be replaced by K_1 , at least for some aspects of metabolic analyses. Although correlation between BF and K_1 improved after chemotherapy

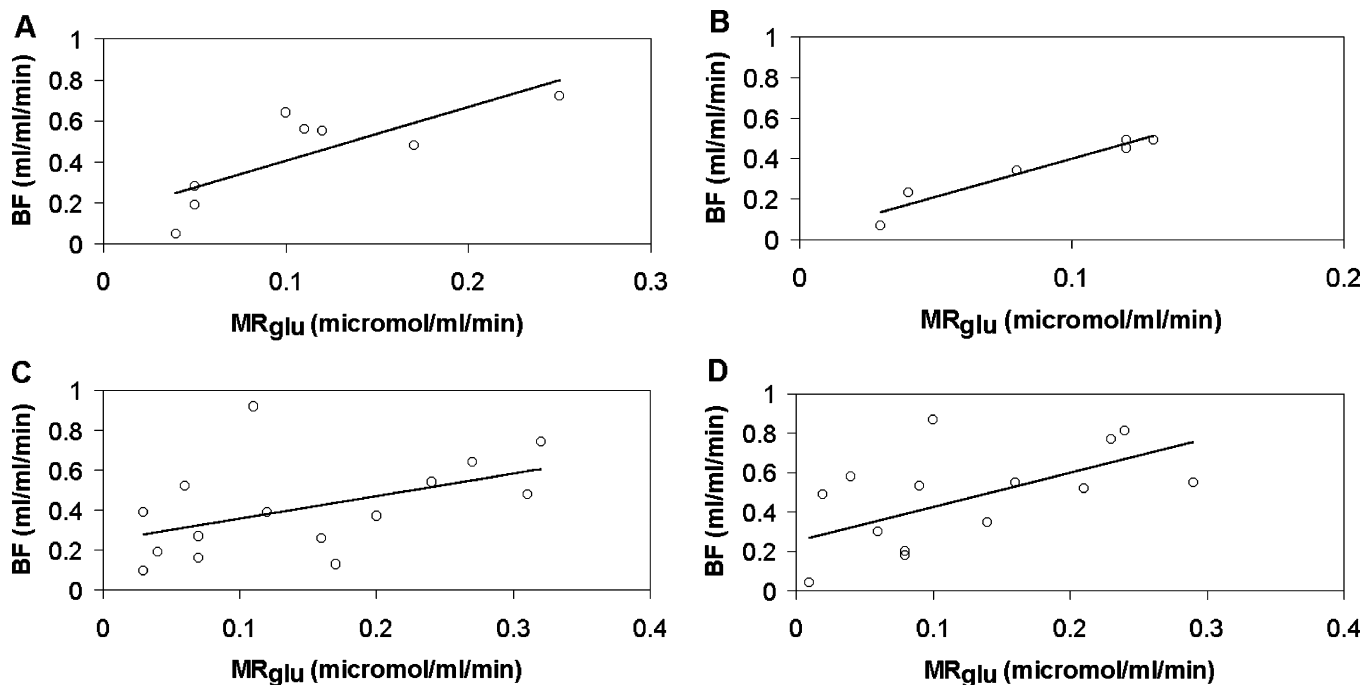


Fig. 2. Correlation between MRglu and BF for breast (a and b) and bone (c and d) lesions before (a and c) and after (b and d) one course of chemotherapy.

in the present study (Table 3), as in Tseng et al. [34], it seems that with the unexplained variance between BF and K_1 found in both studies, BF cannot be reliably replaced by K_1 .

V_d reflects the amount of tumor tissue that rapidly exchanges water with blood within the time of the $H_2^{15}O$ scan [18], and it has been suggested that this represents the proportion of viable tissue within a tumor [35]. If chemotherapy leads to hypo- or hyperperfusion, it will cause a decrease or increase, respectively, in perfusion, but no change in blood volume and V_d . If therapy causes vascular shutdown, this would lead to a reduction in all three parameters [35]. In breast tumors and nodes, both BF and V_d decreased after therapy, although to different degrees (Table 2). This could mean that chemotherapy results in

some degree of vascular shutdown in these lesions. V_d is also very low in normal breast tissue, however, (see Fig. 1b) so an alternative explanation for the lower V_d in breast tumors after chemotherapy could be that values (partially) return to normal. In contrast, mean BF increased in bone lesions, while V_d remained unchanged. Therefore, chemotherapy apparently caused some hyperperfusion in bone metastases.

Mankoff et al. [13] first introduced the MR_{glu}/BF ratio as a parameter that could predict complete response in LABC patients and suggested that a high ratio indicates high glucose extraction. In fact, this ratio is more complex, as it is proportional to glucose extraction fraction times $k_3/(k_2+k_3)$. In theory, tumors can increase glucose extraction as a survival mechanism under conditions of hypoxia or hypoperfusion [36]. In the present study, the MR_{glu}/BF ratio of patients with late (TTP>6 months) or no progressive disease was indeed lower than in those with early progression, but differences were not statistically significant.

The heterogeneity of response observed in this patient group is of particular interest (see Fig. 3). Obviously, BF and MR_{glu} represent two separate physiological processes that are not necessarily linked. The heterogeneity in some (responding) bone lesions might partly be explained by the so-called ‘osteoblastic flare’ phenomenon [37], a paradoxical increase in tracer uptake after successful response of osteolytic metastases due to greater radioisotope uptake by the healing bone. However, the heterogeneous response was also seen in patients without bone metastases. More importantly, an increase in either BF or MR_{glu} in any lesion appeared to be an adverse prognostic sign, regardless of other, well-responding lesions in the same patient. Mankoff

Table 3. Correlation coefficients between BF, MR_{glu}/BF and micro-parameters before and after one course of chemotherapy

| Parameters | Before CTh (<i>r</i>) | <i>p</i> | After CTh (<i>r</i>) | <i>p</i> |
|---------------------|-------------------------|----------|------------------------|----------|
| Breast tumor | | | | |
| BF and MR_{glu} | 0.79 | 0.02 | 0.98 | 0.001 |
| BF and K_1 | 0.43 | 0.29 | 0.86 | 0.03 |
| BF and k_3 | 0.23 | 0.58 | 0.94 | 0.005 |
| Node | | | | |
| BF and MR_{glu} | 0.64 | 0.005 | 0.62 ^a | 0.14 |
| BF and K_1 | 0.28 | 0.26 | 0.63 ^a | 0.13 |
| BF and k_3 | 0.61 | 0.01 | 0.14 ^a | 0.77 |
| Bone | | | | |
| BF and MR_{glu} | 0.49 | 0.06 | 0.50 | 0.06 |
| BF and K_1 | 0.62 | 0.03 | 0.59 | 0.02 |
| BF and k_3 | 0.52 | 0.04 | 0.15 | 0.59 |

^aOnly seven nodes could be compared on both ^{18}F FDG and $H_2^{15}O$ scans after chemotherapy; see text.

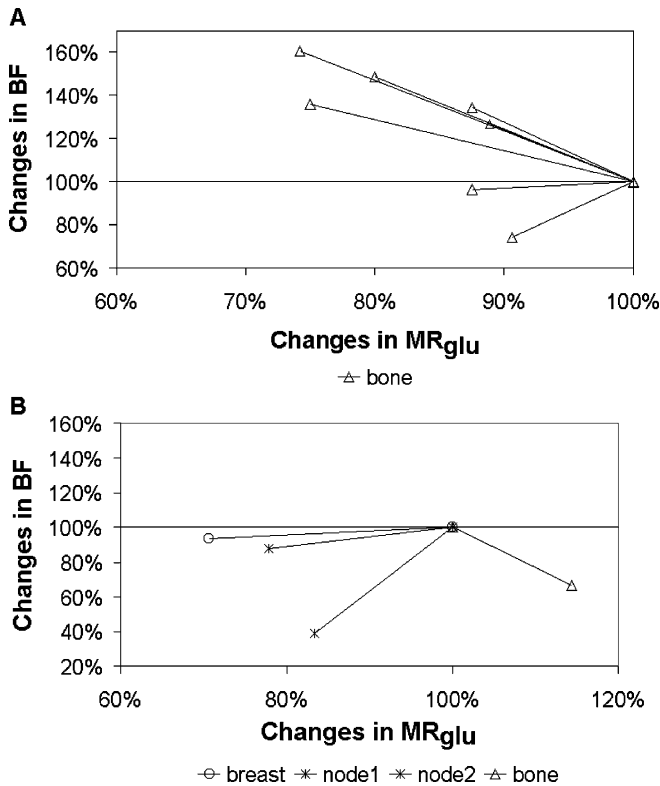


Fig. 3. Two examples of heterogeneous response in *patient no. 1* with bone lesions only (a) and in *patient no. 9* with breast tumor, nodes, and bone metastases (b). MR_{glu} and BF post-chemotherapy are expressed relative to a baseline value of 100%.

et al. [14] also found an average rise in BF after 2 months of chemotherapy in LABC patients who were classified as clinical or pathological nonresponders. This could mean that an early increase in either BF or MR_{glu} during chemotherapy could be used to identify patients who will not respond to therapy or are at risk for early disease progression. Thirdly, there are potential implications for the design of future PET-response monitoring studies involving stage IV breast cancer patients. Conventionally, according to the RECIST classification [20] often used to measure tumor response, only a limited number of ‘measurable’ target lesions are selected at baseline (five per organ, ten in total) and used for subsequent response evaluation. In the light of possible heterogeneous metabolic response, however, it may be more prudent to include and follow-up as many target lesions as possible (within a selected field of view) in order to get a representative view of the overall tumor response in a patient. Bone metastases are considered ‘nonmeasurable’ by the RECIST criteria [20], but both the present study and that of Stafford et al. [17] suggest that serial quantification of response in bone metastases is possible. Moreover, there is some evidence that ¹⁸F¹⁸FDG PET can differentiate between ‘active’ and clinically stable bone metastases [38], that PET is superior (to bone scintigraphy) in detecting osteolytic

bone lesions [39, 40] and that survival in patients with osteolytic lesions is shorter [40]. As such, uniquely, FDG PET avidity may be an indirect sign of clinical aggressiveness of bone metastases.

This study has a number of limitations. Firstly, the number of patients was small, and there was variability in tumor load and treatment. For example, docetaxel and paclitaxel have known anti-angiogenic effects and would obviously have a different effect on blood flow compared to non-anti-angiogenic agents. This could partly explain the inter-patient response heterogeneity as discussed above, but does not explain the heterogeneous response observed within several individual patients.

The purpose of the study was not to investigate the effects of a specific chemotherapeutic agent on blood flow and glucose metabolism, however, but to investigate whether blood flow and glucose metabolism behave differently under influence of chemotherapy. This was the reason for including therapies with different mechanisms of action. Secondly, selection of metastases was restricted by the axial field of view of the scanner and limited to lesions in the vicinity of the heart in order to make use of image-derived arterial input functions. Thus, in some cases, total tumor load was larger than could be evaluated with this dynamic scanning protocol.

Conclusion

Simultaneous measurement of changes in tumor perfusion and glucose metabolism in metastatic breast cancer lesions during chemotherapy offers the possibility to study different biological aspects of tumor response to treatment. Chemotherapy-induced changes in blood flow and glucose metabolism can be markedly heterogeneous even between similar lesions in the same patient.

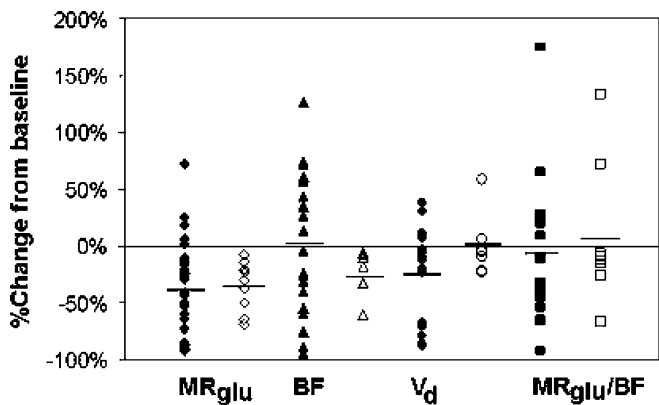


Fig. 4. Percentual changes in MR_{glu}, BF, V_d, and MR_{glu}/BF ratio for all lesions compared to baseline. Patients with early disease progression (TTP ≥ 6 months, *closed symbols*) vs. late or no progression (*open symbols*).

Acknowledgement. The authors would like to thank Dr. Johan Nuyts from the Department of Nuclear Medicine, University Hospital Gasthuisberg, Leuven, Belgium, for providing the semiautomatic program that was used to draw the three-dimensional regions of interest.

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