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Early Prediction of Tumor Response: A Future Strategy for Optimizing Cancer Treatment

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1. Introduction

Positron emission tomography (PET) with ^{18}F-fluorodeoxyglucose (FDG) is successfully capable of imaging glucose metabolism of the tumor cells. Tumor glucose metabolism using FDG-PET has a potential to distinguish viable cancer cells from those in suspension or necrotic components because the degree of tumor FDG uptake is closely associated with its proliferation activity. There is cumulative evidence showing that reduction in FDG uptake value on the early phase after the initiation of chemotherapy more reliably predicts a favorable outcome of patients with breast cancer. Therefore, FDG-PET could help to individualize treatment and to avoid potentially ineffective chemotherapies. In this article, we discuss and illustrate the role and limitations of FDG-PET in the management of neoadjuvant chemotherapy in breast cancer.

2. Tumor metabolic response

FDG-PET has proven useful in the management of various cancers [1]. FDG-PET is known to play an important role in the detection of distant metastasis and recurrence [2, 3]. In addition, it provides quantitative information on tumor glucose metabolic activity, allowing the measurement of metabolic changes and cancer activity shortly after initiation of therapy and before tumor volume reduction [4]. This functional imaging tool may also be useful in predicting tumor response to therapy and optimizing individual treatment [5].

Tumor response to therapy is traditionally assessed by comparison of tumor size before and after treatment, which is determined using anatomical imaging devices such as ultrasonography and computed tomography (CT) on the basis of the Response Evaluation Criteria in Solid

Tumors (RECIST) [6]. Although RECIST has been widely adopted, tumor response must be evaluated for several months until surgery and it does not always reflect the pathological response because of difficulties in distinguishing residual cancer cells from necrotic lesions, fibrosis, or benign masses.

Because of limitations in applying the RECIST criteria using anatomic imaging alone, Wahl et al. proposed a draft framework called the PET Response Criteria in Solid Tumors (PERCIST) [7]. The proposed PERCIST criteria are being used in several current clinical studies. As far as early response assessment with FDG-PET is concerned, clinical studies involving patients with Hodgkin lymphoma and aggressive non-Hodgkin lymphoma have demonstrated that the assessment of changes in FDG uptake on PET imaging after 2 or 3 cycles of chemotherapy was superior for predicting patient prognosis compared with the assessment of morphological changes on computed tomography (CT) [8]. This method was shown to be at least as reliable as definitive response assessment at the end of therapy [8]. In a European multicenter trial monitoring chemotherapy response and survival in 260 patients with lymphoma, an early decrease in FDG uptake on PET after 2 cycles of chemotherapy was significantly correlated with progression-free survival [9]. For other types of cancers, including breast cancer [10], non-small cell lung cancer [11], esophageal cancer [12] [13], gastric cancer [14], and colorectal cancer [15], several clinical studies revealed evidence of the emerging role of FDG-PET in predicting both post-therapeutic clinicopathological response and patient survival.

In our institute, a discrepancy was observed between tumor morphological changes and tumor metabolic activity in a patient treated with neoadjuvant chemotherapy for primary breast cancer. FDG-PET was performed at the onset of chemotherapy, at the midpoint of chemotherapy, and prior to surgery. The primary tumor appeared to grow rapidly after the start of neoadjuvant chemotherapy despite the gradual reduction in glucose accumulation (Figure 1). Postoperative pathological analysis revealed that the lesion was replaced with scar tissue in addition to the presence of massive bleeding and small residual cancer cell nests. In this case, FDG-PET was able to provide more accurate and clinically beneficial information compared with CT.

Clinical studies conducted worldwide have repeatedly revealed the predictive value of FDG-PET in patients with advanced breast cancer treated by chemotherapy. As early as 1993, Wahl et al. studied 11 patients with locally advanced breast cancer before and after 1 cycle of chemotherapy. A significant difference was observed in tumor FDG influx rate (K) from baseline levels between responders and nonresponders (sensitivity: 100%, specificity: 100%) [16]. In 1996, Bassa et al. conducted a retrospective study of 13 patients with breast cancer for whom FDG-PET scans were performed prior to chemotherapy, at the end of the first cycle, at the midpoint of chemotherapy, and before surgery. The mean standardized uptake value (SUV) of the tumor after the first cycle of chemotherapy was significantly lower than the baseline value ($p < 0.01$) [17]. In 2000, reports of clinical studies from two separate institutes showed the usefulness of FDG-PET in the early evaluation of tumor metabolic response to chemotherapy. Schelling et al. demonstrated the ability of FDG-PET to differentiate between responders and nonresponders after the first course of chemotherapy (sensitivity: 100%,
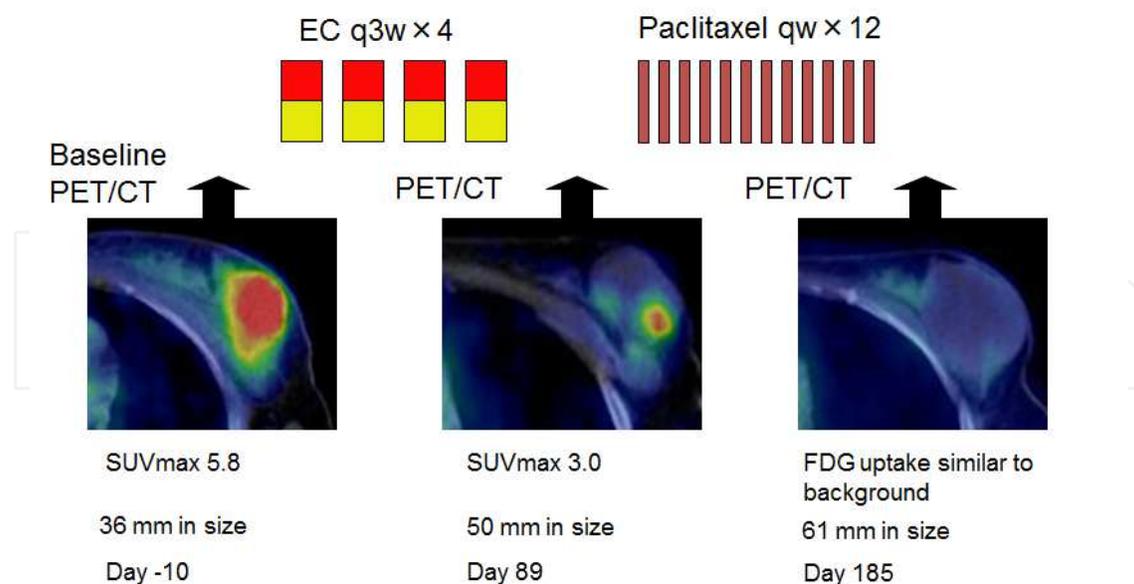
SUVmax decreased from 5.8 to 3.0 at the midpoint of treatment and returned to baseline levels prior to surgery. In contrast, the tumor diameter increased from 36 mm to 50 mm at the midpoint of treatment and to 61 mm prior to surgery.
FDG: fluoro-D-glucose; PET: positron emission tomography; CT: computed tomography
SUVmax: maximal standardized uptake value; EC: epirubicin and cyclophosphamide.

Figure 1. Transversal slices of FDG-PET/CT of a breast lesion before treatment (left), at the completion of the EC regimen (middle), and at the completion of the paclitaxel regimen (right).

specificity: 85%) [18]. Smith et al. also successfully utilized FDG-PET for predicting tumor response after the first cycle of chemotherapy (sensitivity: 90%, specificity: 74%) [19].

Our team investigated the maximum changes in SUV (SUVmax) in 32 primary breast cancer lesions in 30 patients. The patients were treated with neoadjuvant chemotherapy comprising 4 cycles of epirubicine and cyclophosphamide on a triweekly basis and sequential weekly cycles of taxane for 12 weeks [20]. Figure 2 shows representative tumor images on FDG-PET performed at baseline, after one cycle of chemotherapy, after four cycles of chemotherapy, and prior to surgery. The serial images on the upper row show a tumor in which a pathological complete response (pCR) can be observed. The middle row depicts a tumor exhibiting a pathological partial response (pPR), and the lower row illustrates pathological progressive disease (pPD). SUV decreased dramatically in the pCR tumor after one cycle of chemotherapy, after which metabolic activity ceased. SUV change in the pPR tumor was lower than that in the pCR tumor after one cycle of chemotherapy, but SUV gradually diminished during further chemotherapeutic treatment. The pPD tumor showed no significant changes in SUV after treatment. In terms of the optimal threshold of a 40% decrease in SUV, the rate of pathological response in terms of pCR or near pCR was higher (71.4%) in metabolic responders than in nonresponders (12.5%). The sensitivity, specificity, positive predictive value (PPV), and negative predictive value (NPV) were 63%, 92%, 71%, and 88%, respectively.

In 2008, Schwarz–Dose et al. performed the first prospective multicenter trial to evaluate the effectiveness of FDG-PET in predicting early pathological response during chemotherapeutic

A CASE WITH PATHOLOGIC COMPLETE RESPONSE

K.T., 71 y/o, Lt. IDC, Size 46mm, cN1,G3, ER+,PR+,HER2 0,
(Post therapeutic Pathologic Response Grade 3[**])

| SUV max6.13 | SUV max1.95 | SUV max1.59 | SUV max1.14 |
| DAY -14 | DAY 18 | DAY 84 | DAY 179 |
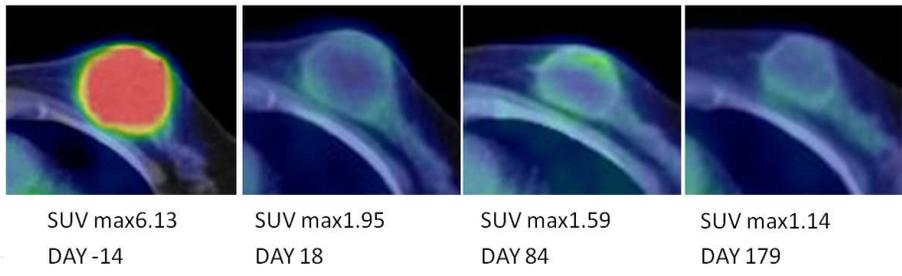
A CASE WITH PATHOLOGIC PARTIAL RESPONSE

A.E., 67 y/o, Lt. IDC, Size 36mm, cN1,G1, ER+,PR+,HER2 1+,
(Post therapeutic Pathologic Response Grade 1a[**])

| SUV max7.01 | SUV max5.23 | SUV max3.86 | SUV max2.59 |
| DAY -7 | DAY 16 | DAY 77 | DAY 158 |
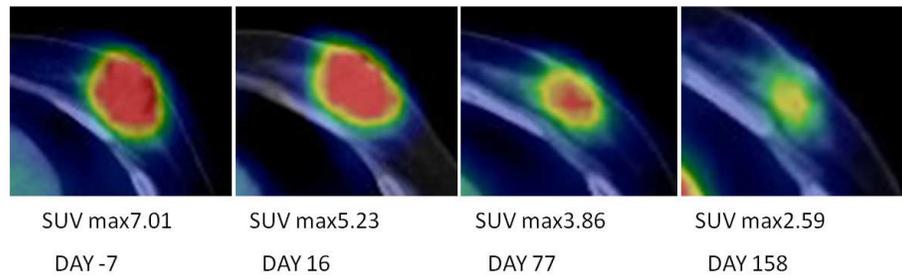
A CASE WITH PROGRESSIVE DISEASE

A.E., 58 y/o, Rt. IDC, Size 40mm, cN0, G3, ER+,PR+,HER2 2+(FISH-),
(Post therapeutic Pathologic Response Grade 0[**])

| SUV max6.21 | SUV max5.11 | SUV max5.09 |
| DAY -11 | DAY19 | DAY 67 |
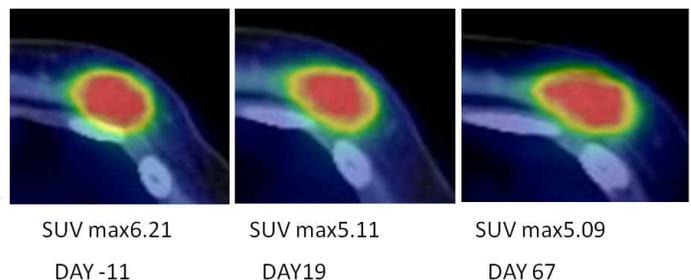
[**]General rules for clinical and pathological recording of breast cancer 2007 (Japan Breast Cancer Society)

pCR: pathological complete response; pPR: pathological partial response; pPD: pathological progressive disease; FDG: fluoro-D-glucose; PET: positron emission tomography; CT: computed tomography; EC: epirubicin and cyclophosphamide

Figure 2. Axial FDG-PET/CT images of a pCR tumor (upper row), a pPR tumor (middle row), and a pPD tumor (lower row). Sequential FDG-PET scans were performed at baseline (left), after 1 cycle of chemotherapy (second from the left), after completion of the EC regimen (third from the left), and at the completion of chemotherapy (right).

treatment in 104 patients with locally advanced breast cancer [10]. In that report, when a 40% decrease in SUV occurred in the first cycle after initiation of chemotherapy compared with baseline values, FDG-PET predicted pCR and pathological macroscopic residual disease at a

high rate, with a sensitivity of 73%, specificity of 63%, PPV of 36%, and NPV of 90% [10]. Other representative studies published in the literature since 2000 are listed in Table 1 [21-25].

In 2011, a meta-analysis in this field summarized 16 articles including a total of 920 patients with breast cancer [26]. To predict histopathological response in primary lesions, the pooled sensitivity, specificity, PPV, NPV, and diagnostic odds ratios were calculated. The results for these parameters were 84% [95% confidence interval (CI), 78%–88%], 66% (95% CI, 62%–70%), 50% (95% CI, 44%–55%), 91% (95% CI, 87%–94%), and 11.90 (95% CI, 6.33%–22.36%), respectively. Although the checkpoints of FDG-PET administration in these trials differed, a subset analysis showed that early response of glucose metabolism after the first or second cycle of chemotherapy provided a significantly better indicator of accuracy compared with a later response (third cycle or later). The subset analysis results showed in Table1. These data indicate that changes in SUV during the first 1–3 cycles of chemotherapy are better indicators of clinical outcome compared with changes during the later cycles.

Authors	Year	Study Type	stage	Population	The timing of PETscans	Early assessment	Endpoint	Cutoff	Se	Sp	PPV	NPV	Accuracy
Martoni, et al.[22]	2010	one center	LABC	34	Baseline,after 2 cycles, 4 cycles, at the end	after 2 cycles	pCR+pMRD	50% in △SUV	100	30	27	100	44
Ueda, et al.[20]	2010	one center	LABC	32	Baseline, after 1 cycycle, after 4 cycles, at the end	after 1 cycle	pCR+less than 3% disappearance of tumor -"-	40% in △SUV	63	92	71	88	84
Kumar, et al.[21]	2009	one center	LABC	23	Baseline, after 2 cycle	after 2 cycles	pCR+pMRD	50% in △SUV	93	75			87
Schwarz-Dose, et al.[10]	2008	multiple centers	LABC	104	baseline, after 1 cycle, after 2 cycle	after 1 cycle	pCR+pMRD	45% in △SUV	73	63	36	90	65
McDemott, et al.[25]	2007	one center	LABC	96	Baseline, after 1cycle, after 2 cycles, at the midpoint, at the end	after 1 cycle	pCR+pMRD	34% in △SUV	100	66			
Berrido-Riedinger, et al.[23]	2007	one center	LABC	50	Baseline, after 2 cycles	after 2 cycles	75% or more absence of tumor	40% in △SUV	77	80			
Rousseau, et al.[24]	2006	one center	LABC	64	Baseline, after 1 cycle, after 2 cycles, after 3 cycles, after 6 cycles	after 2 cycles	50% or more absence of tumor cells	40% in △SUV	89	95		85	
Smith, et al.[19]	2000	one center	LABC	30	Baseline, after 1cycle, after 2 cycles, after 5 cycles, at the end	after 1 cycle	pCRmicro/macro	20% in △DUR	90	74			
Schelling, et al.[18]	2000	one center	LABC	22	Baseline, after 1 cycle, after 2 cycles	after 1 cycle	pCR and pMRD	55% in △SUV	100	85			

| Meta-analysis													
Authors	Year	Study Type	stage	Population	The timing of PET scans	Evaluation of the timing	Endpoint	Cutoff	Se	Sp	PPV	NPV	Accuracy
Wang, et al.[26]	2011	meta-analysis	LABC	920(Total)	Various	Various	Various	Various	84	66	50	91	
				347(subset)	Various	After 1–2 cycles	Various	Various	88	70	61	92	76
				400(subset)	Various	After 3 cycles or later	Various	Various	81	61	34	93	65

LABC, Laegelly advanced breast cancer; pCR, pathological complete response; MRD, macroscope residual disease; Se, sensitivity; Sp, specificity; PPV, positive predictive value; NPV, negative predictive value

Table 1. Optimal timing of FDG PET during chemotherapy

The high sensitivity and high NPV reported above indicate that FDG-PET may be useful for the identification of nonresponders among patients in the early phases of treatment with neoadjuvant chemotherapy. However, in cases where FDG-PET indicates changes in SUV (responders), decision-making regarding continuation of treatment may still be difficult. This problem has been addressed in research on lymphoma patients. Randomized trials have been conducted to determine whether response-guided treatment using early response to therapy as measured by FDG-PET scans is feasible or useful in decreasing the cumulative dose of potentially cytotoxic agents in nonresponders [27], [28]. These trials aim at treatment modification based on PET response by comparing risk-adapted treatment guided by FDG-PET with standard chemotherapy in these patients [29].

An emerging paradigm for a treatment strategy using FDG-PET has been introduced in addition to traditional assessment of tumor response on the basis of staging and tumor subtyping [30]. A specific treatment was chosen from a number of chemotherapeutic drugs, and the usefulness of FDG-PET was analyzed in comparison to assessment based on staging

and subtyping. This new strategy to optimize treatment timing includes staging, subtyping, and response guiding. Staging and subtyping determine favorable treatment options before the initiation of treatment. Functional imaging with FDG-PET offers opportunities to assess tumor response early in the course of treatment. This response-guiding strategy offers the opportunity to revise treatment and improve outcome. FDG-PET not only provides invaluable prognostic information in patients [31] but also supports efforts to switch to more effective treatment options in the early stages of treatment rather than on completion of therapy (Figure 3).
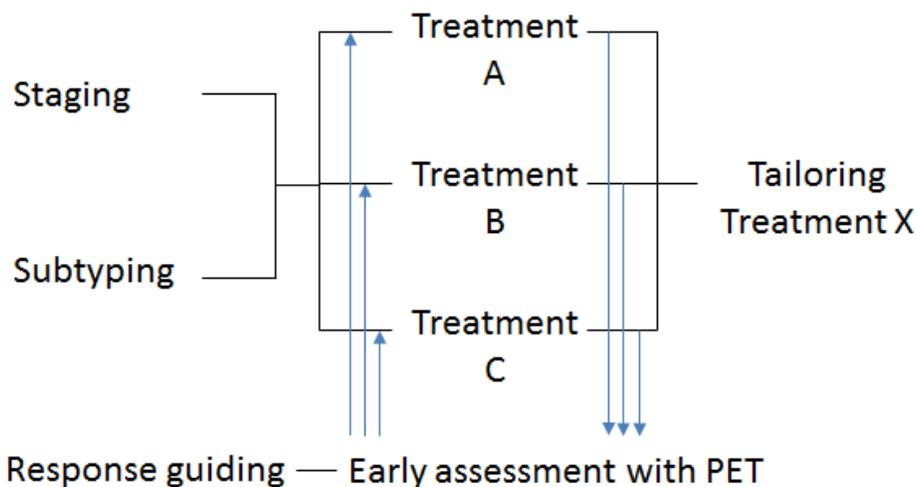
FDG: fluoro-D-glucose; PET: positron emission tomography

Figure 3. Treatment strategy options using FDG-PET in conjunction with staging, subtyping, and response-guiding.

3. Limitations of FDG-PET

Although tumor FDG uptake is an indicator of the viability of cancer cells [31], [32], it is influenced by many biological factors such as stromal cell activity [33], tumor perfusion [34], immune reaction [35], hypoxia [36], [37], and apoptosis. The acute effect of cytotoxic drugs on tumor FDG uptake occurs as a result of high glucose uptake by inflammatory cells and/or energy demand in the process of acute apoptotic death [38], leading to a transient increase in tumor FDG uptake (so-called "flare response") [39], [40]. Therefore, some authors have claimed that the timing of PET scanning soon after the onset of chemotherapy treatment may be crucial. To avoid the flare response, scanning must be delayed for at least 1–2 weeks after the initiation of chemotherapy [41]. PET may be administered immediately before initiation of the second cycle of chemotherapy.

One cause of confusion regarding the results of FDG-PET is the heterogenous results concerning the pathological criteria of outcome, which distinguishes responders from nonresponders. The overall prognosis of patients remains unconfirmed. In addition, variations in dose and

different combinations of drugs add to the confusion. The optimal timing of PET scanning may depend on dose intensity or regimen. If the regimen changes during the course of treatment, the results may be affected because the mechanism of drug sensitivity differs according to tumor characteristics.

The optimal timing of FDG-PET scanning after initiation of chemotherapy for the prediction and elimination of progressive disease (PD) has also not been determined till date. In clinical practice, most physicians are more concerned about tumor progression during neoadjuvant chemotherapy than about achieving pCR. Because chemotherapeutic treatment may increase genomic instability in some tumors, and because chemotherapy resistance may develop in severe cases with hypoxia, neoadjuvant chemotherapy may be contraindicated. However, the role of FDG-PET in the early prediction of PD remains to be established.

The optimal threshold of metabolic change on FDG-PET also remains unclear. A recent draft standard of PERCIST has been advocated as response criteria in solid tumors [7]. It recommends a ≥30% decrease in SUV as a cutoff value for partial metabolic response, which is associated with clinical outcome after chemotherapy. However, specific response criteria for breast cancer must be defined in order to increase the accuracy of prognosis. Further prospective research is needed to determine the optimal cut-off value for predicting tumor response.

Thorough evaluation of cost-effectiveness and prognostic impact of early switching from ineffective neoadjuvant chemotherapy to a more effective regimen is essential. In a simulation study, Schegerin et al. found that early prediction of tumor response using functional imaging devices such as FDG-PET facilitated tailoring of treatment options, which had economic benefits [42]. Further clinical trials must be conducted in order to shed further light on this topic.

Finally, the European Organization for Research and Treatment of Cancer PET study group recommended improvements in the quality of tumor imaging with FDG-PET. Interinstitutional bias is another factor in the usefulness of FDG-PET as a prognostic tool. The available data are insufficient to define the optimal time after injection of FDG and the optimal dose of FDG at which SUV should be measured [41].

4. Early changes in metabolism using molecular-targeted drugs and endocrine therapy

Functional imaging devices may be useful in the assessment of the biological activity of molecular-targeted drugs [43]. These agents are predominantly cytostatic in nature, that is, they modulate biological behavior and arrest cell cycling rather than totally killing cancer cells [44]. In cases treated with these agents, the traditional endpoints used to evaluate the effects of cytotoxic drugs, such as RECIST criteria, are insufficient and sometimes inappropriate for the prediction of therapeutic outcome. For example, antagonists of the epidermal growth factor receptor (EGFR), such as trastuzumab and cetuximab, block membranous EGFR in cancer cells, halt cell cycling, and induce apoptosis. These drugs and many others require different evaluation criteria to determine their efficacy. FDG-PET may be useful in establishing these criteria.

Small molecule drugs to block protein kinases, such as gefitinib, have been used for the inhibition of tumor proliferation and neovascularization. A rapid decrease in FDG uptake at 48 hour was seen in lung cancer xenografts treated with gefitinib. Su et al. also reported a very early decrease at 2 hour in gefitinib-sensitive cancer cells and no change in resistant cancer cells [45]. Other reports stated that a reduction in metabolism within 1 week after the commencement of therapy was associated with sensitivity to certain drugs, for example, an epidermal growth factor receptor/human epidermal growth factor receptor 2 (HER2) dual kinase inhibitor (lapatinib) used for the treatment of breast cancer, a c-kit inhibitor (imatinib mesylate) used for the treatment of gastrointestinal stromal tumors (GIST), a mammalian target of rapamycin inhibitor (rapamycin) used for the treatment of GIST, and various other drugs used for the treatment of uterine and neuroendocrine carcinomas and sarcomas [44]. Moreover, in the clinical setting, FDG-PET was reported to be useful for the evaluation of treatment response to sunitinib, a multitarget tyrosine kinase inhibitor, in patients with GIST resistant to treatment with imatinib [46].

Endocrine therapy is one of the most common treatment strategies in patients with estrogen receptor (ER)-positive breast cancer. Successful cytostatic drugs include ER antagonists such as tamoxifen, which induce the deprivation of estrogen production, aromatase inhibitors, ER downregulators, or fulvestrant. In 2011, we reported that changes in SUV at approximately 2 weeks after treatment with letrozole, an aromatase inhibitor, was correlated with a drop in proliferative rate of cancer cells measured by immunohistochemical staining of Ki67 [47]. With a tentative threshold value of a 40% decrease in SUV, Ki67 index values were significantly decreased in metabolic responders. The Immediate Preoperative Anastrozole, Tamoxifen, or Combined with Tamoxifen (IMPACT) randomized trial revealed that 2 weeks of treatment with the aromatase inhibitor anastrozole suppressed the Ki67 index (as compared with a percentage of baseline expression) to a significantly greater extent than did tamoxifen alone or tamoxifen in combination with anastrozole. The affiliated study showed that after 2 weeks of endocrine therapy, Ki67 index values predicted recurrence-free survival in individual patients [48]. A positive correlation between the Ki67 index and tumor SUV has been reported in some studies [32, 49], [50]. Mortazavi-Jehanno et al. investigated the predictive value of metabolic response in patients with metastatic breast cancer after 8 weeks of endocrine therapy, demonstrating that progression-free survival is related to metabolic response [51]. These observations suggest that changes in tumor SUV after endocrine therapy may be associated with favorable prognosis. Therefore, the biological basis of changes in FDG uptake using cytostatic drugs may be associated more with intracellular pathways of metabolism and cell cycling than with cytotoxic agents.

4.1. Tumor metabolic flare

Tumor flare reaction denotes a sudden and temporary worsening of tumor-related symptoms after the initiation of treatment [52]. Several studies reported that radiotherapy and some types of chemotherapeutic agents induce diffusely elevated FDG accumulation because of inflammation. Weber suggested a careful inspection of the degree and pattern of FDG uptake to distinguish between radiation-induced inflammation and residual cancer activity [53]. As

mentioned earlier, the EORTC PET study group recommends that after baseline FDG-PET scanning and before the initiation of chemotherapy, serial scanning using FDG-PET should be performed 1–2 weeks after the first course [41]. Therefore, the consensus till date has been that a waiting period of 1–2 weeks should be observed after initial drug administration or radiotherapy in order to avoid the inflammatory response and accurately evaluate tumor activity. However, metabolic flare does not necessarily indicate treatment failure or cancer progression. Table 2 lists the imaging studies reporting the association between tumor metabolic flare and tumor response to treatment.

Animal experiments

	Year	Treatment	Cancer type	Origin of cell lines	Tracer	Modality	Animal type	Time of flare reaction occurred	Comments
Furuta, et al.[54]	1997	radiotherapy	NNE,GLS,KYG	various	FDG	PET	mice	2 hours	A flare was observed in radiosensitive tumors
Aliga, et al.[56]	2007	doxorubicin	MC4L2, MC7L1	breast	FDG	PET	mice	7 days	A flare reaction was observed 7 days after treatment of doxorubicin, methotrexate, letrozole, or placebo
Aide, et al.[55]	2009	cisplatin	NCCIT	testicular	FDG	PET	rats	2 days	A flare was related to a transient cell cycle arrest and apoptosis but did not reveal refractory disease
Bjurberg, et al.[40]	2009	cisplatin	HNxSCC24	head and neck	FDG	PET	mice	1 day	A flare occurred early after cisplatin treatment in responding tumors
Bjurberg, et al.[57]	2010	cisplatin	HNxSCC24	head and neck	2-NBDG	Fluorescene microscope	cell culture	2 days	

Clinical studies

	Year	Treatment	Clinical staging	Origin	Tracer	Modality	Time of flare reaction occurred	Outcome
Schneider JA, et al.[58]	1994	Paclitaxel	metastatic, bone metastasis	breast		Scintigraphy	after 2 cycle (4-6wk)	A flare response of bone metastasis after 2 cycle resulted in improvement on follow-up scan
Mortimer JE, et al.[59]	2001	Tamoxifen	Advanced/metastatic	breast	FDG, FES	PET	7-10 days	Responders had increase in SUV for FDG (28.4±23.3) while non-responders had reduce in SUV (-10.2±16.2) p = 0.0002
Dehdashti, et al.[60]	2009	Estradiol	Advanced	breast	FDG, FES	PET	1 day	Responders had increase in SUV for FDG (20.9±24.2) while non-responders had reduce in SUV (-4.3±11.0) p < 0.0001

Table 2. Tumor metabolic flare on very early phase of treatment

In animal experiments, Furuta et al. reported a flare reaction detected by FDG-PET in nude mice with ependymoblastoma, small cell lung cancer, and glioblastoma at 2 h after irradiation with 10 Gy [54]. They claimed that flare intensity was strongest in the ependymoblastoma, the most radiosensitive of these tumors, whereas the two less radiosensitive tumors showed no increase in FDG uptake during the observation period. In human testicular cancer xenografts in nude rats that received cisplatin, Aide et al. reported that FDG-PET scanning detected a peak FDG uptake on day 2, followed by a marked decrease on day 7 despite the lack of change in tumor volume [55]. They observed a transient S and G2/M cell cycle arrest and a marked increase in apoptosis within this phase. A very early increase in FDG uptake may explain the flare reaction that represents increased tumor metabolism in apoptotic cells as well as in cells that exhibit transient cell cycle arrest.

Aliga et al. reported a similar result mice after the administration of doxorubicin to decrease tumor burden in BALB/c mice with breast cancer [56]. They observed a rapid decrease in tumor FDG uptake 24 hour after chemotherapy and a transient accumulation in FDG uptake on day 7. They suggested that a partial agonistic effect of chemotherapy, apoptosis, cell repair mechanisms, or intratumoral inflammation may be responsible for this flare reaction.

Using the fluorescent glucose analog 2-NBDG, Bjurberg et al. reported a very early increase in metabolism in three squamous cell carcinoma cell lines (LU-HNxSCC-7, LU-HNxSCC-24, and LU-CX-2) after exposure to cisplatin [40] [57]. The flare reaction was observed within 3 days after exposure in these cells, whereas FDG uptake in nonmalignant fibroblastic cells was low.

Contrary to recent animal studies revealing a flare phenomenon-associated tumor response, only a few clinical studies have utilized FDG-PET in detecting tumor flare. As early as 1996, Schneiders et al. reported the use of scintigraphy to detect a flare reaction in bone lesions in patients with metastatic disease; the flare reaction occurred despite a favorable overall outcome [58]. This phenomenon may represent enhanced osteoblastic activity, rapid bone repair, and improved blood flow around the responding lesions. Welch et al. conducted an FDG-PET study in which a paradoxical flare phenomenon was detected within 7 to 10 days after the initiation of tamoxifen in patients with breast cancer [59]. An increase in FDG uptake was observed in responding tumors during week 1 after tamoxifen initiation ; however, this was not observed in nonresponding tumors. The transient increase in FDG uptake could reflect hormone receptor-related changes in tumor metabolism, which was predictive of a favorable outcome. Partial agonist–antagonist activity in selective ER modulators such as tamoxifen is known to differ over the treatment course after tissue exposure. The slow onset of action of tamoxifen and the fact that its effects as an estrogen agonist peak 1–2 weeks after the onset of therapy may contribute to the development of the flare reaction. The same group of investigators conducted a clinical study including an estradiol challenge test to predict hormone sensitivity in women with locally advanced or metastatic ER-positive breast cancer. FDG-PET after 30 mg estradiol induced a metabolic flare, showing greater responsiveness to endocrine therapy and better overall survival in flare patients than in non-flare patients [60].

Results of those animal studies lead to the speculation that tumor metabolic flare is related to the following sequence of events: transient cell cycle arrest, apoptosis, induction of the cancer immune system, and hemodynamic reaction. As for estrogen-positive breast cancer and endocrine therapy, estrogen stimulation may be closely associated with an increase in glucose uptake [61]. At lease, this does not necessarily indicate refractory disease.

Taken together, the underlying presumption in these studies is that changes in FDG uptake in response to chemotherapy occur in three phases (Figure 4). In the first phase, cellular damage followed by inflammation and vascular changes occurs within hours or days, resulting in accumulation of FDG and increased uptake in cancer cells and inflammatory cells such as neutrophils (inflammatory phase). In this phase, pro-inflammatory cytokines may be released because of the tumor's response to chemotherapy, and these cytokines accelerate tumor cell proliferation, activate the immune system, and increase blood flow, resulting in accumulation of tumor FDG. Second, apoptosis, cell cycle arrest, and diminishing inflammation, which occur within days or weeks, lead to a decrease in FDG uptake in cancer cells (apoptotic phase). Decreased blood flow due to vascular damage caused by chemotherapy may deteriorate the

tumor microenvironment and promote apoptosis. In addition, tumor cells may be replaced by fibrotic cells, shrinking the tumor several weeks after the initiation of chemotherapy. FDG uptake in the tumor then decreases because of volume reduction (volume reduction phase). In this last phase, morphological changes can be identified by CT or magnetic resonance imaging. Therefore, metabolic flare precedes decreased uptake of FDG. This is not necessarily a confounding factor; rather, it provides an insight into pharmacodynamics [62]. Further understanding of cancer metabolic flare in the early phase after chemotherapy can aid in strategic planning of successful therapy.

	Inflammatory phase	Apoptotic phase	Volume reduction phase
Time frame	Within hours to days	Within days to weeks	Within weeks to months
FDG uptake	↑↑ (Flare)	→ or ↓	↓
Mechanism	1. Cellular damage due to drug exposure and energy demand against hypoxic stress. 2. Induction of inflammatory cells such as neutrophils 3. Hemodynamic reaction due to inflammation	1. Apoptosis or Cell cycle arrest results in decrease in number of viable cells. 2. Accumulation of inflammatory cells is diminishing. 3. Tumor vessels are regressed.	1. Tumor shrinks because cancer nests replace fibrosis 2. Vessels remodeling and normalization

FDG: fluoro-D-glucose

Figure 4. Tumor FDG uptake in response to chemotherapy hypothetically occurs in three biological phases. In the first phase, FDG accumulation occurs because of cellular damage, inflammation, and vascular reaction. In the second phase, apoptosis results in decreasing cellularity. In the third phase, the tumor decreases in size.

5. Summary

Of late, cancer treatment is frequently optimized on the basis of tumor subtype and stage. Breast cancer is characterized by ER and HER2 status in addition to tumor size and distant metastatic involvement. The advent of commercialized kits containing multiple molecular biomarker assays has shifted the focus in tumor categorization from pathology to molecular analysis. However, rapid development and wide availability of a broad range of new drugs has exceeded the discovery of tumor subtyping methods or the establishment of biomarkers predictive of chemosensitivity. Therefore, a new paradigm that involves response-guided strategies during initial treatment, which can aid in decision-making about the next treatment option, is emerging for cancer treatment. Early response assessment using FDG-PET may eventually be applicable for planning and evaluating future treatment strategies. In future, patients could be allocated to standard or investigational chemotherapy regimens on the basis of metabolic response. Furthermore, in phase 1/2 studies determining the optimal dose of a

new drug, nonresponsive patients can be eliminated on the basis of metabolic response. Glucose metabolism analysis using FDG-PET will be one of several critical factors for evaluating tumor response to chemotherapy. Integration of multiple functional imaging systems may also be useful in predicting early tumor response to chemotherapy.

Finally, considering the phenomenon of tumor metabolic flare, clinical trials must be conducted to determine the best timing for the administration of FDG-PET. This information would be useful to predict tumor response in the very early stages of treatment.

Acknowledgements

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References

[1] Juweid ME, Cheson BD: Positron-emission tomography and assessment of cancer therapy. *The New England journal of medicine* 2006, 354(5):496-507.

[2] Cermik TF, Mavi A, Basu S, Alavi A: Impact of FDG PET on the preoperative staging of newly diagnosed breast cancer. *European journal of nuclear medicine and molecular imaging* 2008, 35(3):475-483.

[3] Niikura N, Ueno NT: The Role of F-FDG-Positron Emission Tomography/Computed Tomography in Staging Primary Breast Cancer. *Journal of Cancer* 2010, 1:51-53.

[4] Zhu A, Lee D, Shim H: Metabolic positron emission tomography imaging in cancer detection and therapy response. *Seminars in oncology* 2011, 38(1):55-69.

[5] Weber WA: Assessing tumor response to therapy. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 2009, 50 Suppl 1:1S-10S.

[6] Therasse P, Arbuck SG, Eisenhauer EA, Wanders J, Kaplan RS, Rubinstein L, Verweij J, Van Glabbeke M, van Oosterom AT, Christian MC *et al*: New guidelines to evaluate

the response to treatment in solid tumors. European Organization for Research and Treatment of Cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. *Journal of the National Cancer Institute* 2000, 92(3):205-216.

[7]   Wahl RL, Jacene H, Kasamon Y, Lodge MA: From RECIST to PERCIST: Evolving Considerations for PET response criteria in solid tumors. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 2009, 50 Suppl 1:122S-150S.

[8]   MacManus MP, Seymour JF, Hicks RJ: Overview of early response assessment in lymphoma with FDG-PET. *Cancer imaging : the official publication of the International Cancer Imaging Society* 2007, 7:10-18.

[9]   Gallamini A, Hutchings M, Rigacci L, Specht L, Merli F, Hansen M, Patti C, Loft A, Di Raimondo F, D'Amore F *et al*: Early interim 2- [18F]fluoro-2-deoxy-D-glucose positron emission tomography is prognostically superior to international prognostic score in advanced-stage Hodgkin's lymphoma: a report from a joint Italian-Danish study. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology* 2007, 25(24):3746-3752.

[10] Schwarz-Dose J, Untch M, Tiling R, Sassen S, Mahner S, Kahlert S, Harbeck N, Lebeau A, Brenner W, Schwaiger M *et al*: Monitoring primary systemic therapy of large and locally advanced breast cancer by using sequential positron emission tomography imaging with [18F]fluorodeoxyglucose. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology* 2009, 27(4):535-541.

[11] Rebollo-Aguirre AC, Ramos-Font C, Villegas Portero R, Cook GJ, Llamas Elvira JM, Romero Tabares A: Is FDG-PET suitable for evaluating neoadjuvant therapy in non-small cell lung cancer? Evidence with systematic review of the literature. *Journal of surgical oncology* 2010, 101(6):486-494.

[12] Lordick F, Ott K, Krause BJ, Weber WA, Becker K, Stein HJ, Lorenzen S, Schuster T, Wieder H, Herrmann K *et al*: PET to assess early metabolic response and to guide treatment of adenocarcinoma of the oesophagogastric junction: the MUNICON phase II trial. *The lancet oncology* 2007, 8(9):797-805.

[13] Kwee RM: Prediction of tumor response to neoadjuvant therapy in patients with esophageal cancer with use of 18F FDG PET: a systematic review. *Radiology* 2010, 254(3):707-717.

[14] Lorenzen S, von Gall C, Stange A, Haag GM, Weitz J, Haberkorn U, Lordick F, Weichert W, Abel U, Debus J *et al*: Sequential FDG-PET and induction chemotherapy in locally advanced adenocarcinoma of the Oesophago-gastric junction (AEG): the Heidelberg Imaging program in Cancer of the oesophago-gastric junction during Neoadjuvant treatment: HICON trial. *BMC cancer* 2011, 11:266.

[15] de Geus-Oei LF, Vriens D, van Laarhoven HW, van der Graaf WT, Oyen WJ: Monitoring and predicting response to therapy with 18F-FDG PET in colorectal cancer: a

systematic review. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 2009, 50 Suppl 1:43S-54S.

[16] Wahl RL, Zasadny K, Helvie M, Hutchins GD, Weber B, Cody R: Metabolic monitoring of breast cancer chemohormonotherapy using positron emission tomography: initial evaluation. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology* 1993, 11(11):2101-2111.

[17] Bassa P, Kim EE, Inoue T, Wong FC, Korkmaz M, Yang DJ, Wong WH, Hicks KW, Buzdar AU, Podoloff DA: Evaluation of preoperative chemotherapy using PET with fluorine-18-fluorodeoxyglucose in breast cancer. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 1996, 37(6):931-938.

[18] Schelling M, Avril N, Nahrig J, Kuhn W, Romer W, Sattler D, Werner M, Dose J, Janicke F, Graeff H *et al*: Positron emission tomography using [(18)F]Fluorodeoxyglucose for monitoring primary chemotherapy in breast cancer. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology* 2000, 18(8): 1689-1695.

[19] Smith IC, Welch AE, Hutcheon AW, Miller ID, Payne S, Chilcott F, Waikar S, Whitaker T, Ah-See AK, Eremin O *et al*: Positron emission tomography using [(18)F]-fluorodeoxy-D-glucose to predict the pathologic response of breast cancer to primary chemotherapy. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology* 2000, 18(8):1676-1688.

[20] Ueda S, Tsuda H, Saeki T, Osaki A, Shigekawa T, Ishida J, Tamura K, Abe Y, Omata J, Moriya T *et al*: Early reduction in standardized uptake value after one cycle of neoadjuvant chemotherapy measured by sequential FDG PET/CT is an independent predictor of pathological response of primary breast cancer. *The breast journal* 2010, 16(6): 660-662.

[21] Kumar A, Kumar R, Seenu V, Gupta SD, Chawla M, Malhotra A, Mehta SN: The role of 18F-FDG PET/CT in evaluation of early response to neoadjuvant chemotherapy in patients with locally advanced breast cancer. *European radiology* 2009, 19(6):1347-1357.

[22] Martoni AA, Zamagni C, Quercia S, Rosati M, Cacciari N, Bernardi A, Musto A, Fanti S, Santini D, Taffurelli M: Early (18)F-2-fluoro-2-deoxy-d-glucose positron emission tomography may identify a subset of patients with estrogen receptor-positive breast cancer who will not respond optimally to preoperative chemotherapy. *Cancer* 2010, 116(4):805-813.

[23] Berriolo-Riedinger A, Touzery C, Riedinger JM, Toubeau M, Coudert B, Arnould L, Boichot C, Cochet A, Fumoleau P, Brunotte F: [18F]FDG-PET predicts complete pathological response of breast cancer to neoadjuvant chemotherapy. *European journal of nuclear medicine and molecular imaging* 2007, 34(12):1915-1924.

[24] Rousseau C, Devillers A, Sagan C, Ferrer L, Bridji B, Campion L, Ricaud M, Bourbouloux E, Doutriaux I, Clouet M *et al*: Monitoring of early response to neoadjuvant che-

motherapy in stage II and III breast cancer by [18F]fluorodeoxyglucose positron emission tomography. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology* 2006, 24(34):5366-5372.

[25] McDermott GM, Welch A, Staff RT, Gilbert FJ, Schweiger L, Semple SI, Smith TA, Hutcheon AW, Miller ID, Smith IC *et al*: Monitoring primary breast cancer throughout chemotherapy using FDG-PET. *Breast cancer research and treatment* 2007, 102(1): 75-84.

[26] Wang Y, Zhang C, Liu J, Huang G: Is 18F-FDG PET accurate to predict neoadjuvant therapy response in breast cancer? A meta-analysis. *Breast cancer research and treatment* 2012, 131(2):357-369.

[27] Plathow C, Weber WA: Tumor cell metabolism imaging. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 2008, 49 Suppl 2:43S-63S.

[28] Dann EJ, Bar-Shalom R, Tamir A, Haim N, Ben-Shachar M, Avivi I, Zuckerman T, Kirschbaum M, Goor O, Libster D *et al*: Risk-adapted BEACOPP regimen can reduce the cumulative dose of chemotherapy for standard and high-risk Hodgkin lymphoma with no impairment of outcome. *Blood* 2007, 109(3):905-909.

[29] Aridgides P, Bogart J, Shapiro A, Gajra A: PET Response-Guided Treatment of Hodgkin's Lymphoma: A Review of the Evidence and Active Clinical Trials. *Advances in hematology* 2011, 2011:309237.

[30] Fleming IN, Gilbert FJ, Miles KA, Cameron D: Opportunities for PET to deliver clinical benefit in cancer: breast cancer as a paradigm. *Cancer imaging : the official publication of the International Cancer Imaging Society* 2010, 10:144-152.

[31] Ueda S, Tsuda H, Asakawa H, Shigekawa T, Fukatsu K, Kondo N, Yamamoto M, Hama Y, Tamura K, Ishida J *et al*: Clinicopathological and prognostic relevance of uptake level using 18F-fluorodeoxyglucose positron emission tomography/computed tomography fusion imaging (18F-FDG PET/CT) in primary breast cancer. *Japanese journal of clinical oncology* 2008, 38(4):250-258.

[32] Ueda S, Kondoh N, Tsuda H, Yamamoto S, Asakawa H, Fukatsu K, Kobayashi T, Yamamoto J, Tamura K, Ishida J *et al*: Expression of centromere protein F (CENP-F) associated with higher FDG uptake on PET/CT, detected by cDNA microarray, predicts high-risk patients with primary breast cancer. *BMC cancer* 2008, 8:384.

[33] Martinez-Outschoorn UE, Pestell RG, Howell A, Tykocinski ML, Nagajyothi F, Machado FS, Tanowitz HB, Sotgia F, Lisanti MP: Energy transfer in "parasitic" cancer metabolism: mitochondria are the powerhouse and Achilles' heel of tumor cells. *Cell Cycle* 2011, 10(24):4208-4216.

[34] Miles KA, Griffiths MR, Keith CJ: Blood flow-metabolic relationships are dependent on tumour size in non-small cell lung cancer: a study using quantitative contrast-en-

hanced computer tomography and positron emission tomography. *European journal of nuclear medicine and molecular imaging* 2006, 33(1):22-28.

[35] Spaepen K, Stroobants S, Dupont P, Bormans G, Balzarini J, Verhoef G, Mortelmans L, Vandenberghe P, De Wolf-Peeters C: [(18)F]FDG PET monitoring of tumour response to chemotherapy: does [(18)F]FDG uptake correlate with the viable tumour cell fraction? *European journal of nuclear medicine and molecular imaging* 2003, 30(5): 682-688.

[36] Goh V, Engledow A, Rodriguez-Justo M, Shastry M, Peck J, Blackman G, Endozo R, Taylor S, Halligan S, Ell P *et al*: The flow-metabolic phenotype of primary colorectal cancer: assessment by integrated 18F-FDG PET/perfusion CT with histopathologic correlation. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 2012, 53(5):687-692.

[37] Dooms C, van Baardwijk A, Verbeken E, van Suylen RJ, Stroobants S, De Ruysscher D, Vansteenkiste J: Association between 18F-fluoro-2-deoxy-D-glucose uptake values and tumor vitality: prognostic value of positron emission tomography in early-stage non-small cell lung cancer. *Journal of thoracic oncology : official publication of the International Association for the Study of Lung Cancer* 2009, 4(7):822-828.

[38] Jin S, DiPaola RS, Mathew R, White E: Metabolic catastrophe as a means to cancer cell death. *Journal of cell science* 2007, 120(Pt 3):379-383.

[39] Archer CD, Parton M, Smith IE, Ellis PA, Salter J, Ashley S, Gui G, Sacks N, Ebbs SR, Allum W *et al*: Early changes in apoptosis and proliferation following primary chemotherapy for breast cancer. *British journal of cancer* 2003, 89(6):1035-1041.

[40] Bjurberg M, Henriksson E, Brun E, Ekblad L, Ohlsson T, Brun A, Kjellen E: Early changes in 2-deoxy-2- [18F]fluoro-D-glucose metabolism in squamous-cell carcinoma during chemotherapy in vivo and in vitro. *Cancer biotherapy & radiopharmaceuticals* 2009, 24(3):327-332.

[41] Young H, Baum R, Cremerius U, Herholz K, Hoekstra O, Lammertsma AA, Pruim J, Price P: Measurement of clinical and subclinical tumour response using [18F]-fluorodeoxyglucose and positron emission tomography: review and 1999 EORTC recommendations. European Organization for Research and Treatment of Cancer (EORTC) PET Study Group. *Eur J Cancer* 1999, 35(13):1773-1782.

[42] Schegerin M, Tosteson AN, Kaufman PA, Paulsen KD, Pogue BW: Prognostic imaging in neoadjuvant chemotherapy of locally-advanced breast cancer should be cost-effective. *Breast cancer research and treatment* 2009, 114(3):537-547.

[43] Weber WA, Czernin J, Phelps ME, Herschman HR: Technology Insight: novel imaging of molecular targets is an emerging area crucial to the development of targeted drugs. *Nature clinical practice Oncology* 2008, 5(1):44-54.

[44] Contractor KB, Aboagye EO: Monitoring predominantly cytostatic treatment response with 18F-FDG PET. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 2009, 50 Suppl 1:97S-105S.

[45] Su H, Bodenstein C, Dumont RA, Seimbille Y, Dubinett S, Phelps ME, Herschman H, Czernin J, Weber W: Monitoring tumor glucose utilization by positron emission tomography for the prediction of treatment response to epidermal growth factor receptor kinase inhibitors. *Clinical cancer research : an official journal of the American Association for Cancer Research* 2006, 12(19):5659-5667.

[46] Prior JO, Montemurro M, Orcurto MV, Michielin O, Luthi F, Benhattar J, Guillou L, Elsig V, Stupp R, Delaloye AB *et al*: Early prediction of response to sunitinib after imatinib failure by 18F-fluorodeoxyglucose positron emission tomography in patients with gastrointestinal stromal tumor. *Journal of clinical oncology : official journal of the American Society of Clinical Oncology* 2009, 27(3):439-445.

[47] Ueda S, Tsuda H, Saeki T, Omata J, Osaki A, Shigekawa T, Ishida J, Tamura K, Abe Y, Moriya T *et al*: Early metabolic response to neoadjuvant letrozole, measured by FDG PET/CT, is correlated with a decrease in the Ki67 labeling index in patients with hormone receptor-positive primary breast cancer: a pilot study. *Breast Cancer* 2011, 18(4):299-308.

[48] Dowsett M, Smith IE, Ebbs SR, Dixon JM, Skene A, A'Hern R, Salter J, Detre S, Hills M, Walsh G: Prognostic value of Ki67 expression after short-term presurgical endocrine therapy for primary breast cancer. *Journal of the National Cancer Institute* 2007, 99(2):167-170.

[49] Buck A, Schirrmeister H, Kuhn T, Shen C, Kalker T, Kotzerke J, Dankerl A, Glatting G, Reske S, Mattfeldt T: FDG uptake in breast cancer: correlation with biological and clinical prognostic parameters. *European journal of nuclear medicine and molecular imaging* 2002, 29(10):1317-1323.

[50] Shimoda W, Hayashi M, Murakami K, Oyama T, Sunagawa M: The relationship between FDG uptake in PET scans and biological behavior in breast cancer. *Breast Cancer* 2007, 14(3):260-268.

[51] Mortazavi-Jehanno N, Giraudet AL, Champion L, Lerebours F, Le Stanc E, Edeline V, Madar O, Bellet D, Pecking AP, Alberini JL: Assessment of response to endocrine therapy using FDG PET/CT in metastatic breast cancer: a pilot study. *European journal of nuclear medicine and molecular imaging* 2012, 39(3):450-460.

[52] Basu S, Alavi A: Defining co-related parameters between 'metabolic' flare and 'clinical', 'biochemical', and 'osteoblastic' flare and establishing guidelines for assessing response to treatment in cancer. *European journal of nuclear medicine and molecular imaging* 2007, 34(4):441-443.

[53] Weber WA: Use of PET for monitoring cancer therapy and for predicting outcome. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 2005, 46(6): 983-995.

[54] Furuta M, Hasegawa M, Hayakawa K, Yamakawa M, Ishikawa H, Nonaka T, Mitsu-hashi N, Niibe H: Rapid rise in FDG uptake in an irradiated human tumour xeno-graft. *European journal of nuclear medicine* 1997, 24(4):435-438.

[55] Aide N, Poulain L, Briand M, Dutoit S, Allouche S, Labiche A, Ngo-Van Do A, Nataf V, Batalla A, Gauduchon P *et al*: Early evaluation of the effects of chemotherapy with longitudinal FDG small-animal PET in human testicular cancer xenografts: early flare response does not reflect refractory disease. *European journal of nuclear medicine and molecular imaging* 2009, 36(3):396-405.

[56] Aliaga A, Rousseau JA, Cadorette J, Croteau E, van Lier JE, Lecomte R, Benard F: A small animal positron emission tomography study of the effect of chemotherapy and hormonal therapy on the uptake of 2-deoxy-2- [F-18]fluoro-D-glucose in murine models of breast cancer. *Molecular imaging and biology : MIB : the official publication of the Academy of Molecular Imaging* 2007, 9(3):144-150.

[57] Bjurberg M, Abedinpour P, Brun E, Baldetorp B, Borgstrom P, Wennerberg J, Kjellen E: Early Metabolic Flare in Squamous Cell Carcinoma after Chemotherapy is a Mark-er of Treatment Sensitivity In Vitro. *Nuclear medicine and molecular imaging* 2010, 44(3): 165-169.

[58] Schneider JA, Divgi CR, Scott AM, Macapinlac HA, Seidman AD, Goldsmith SJ, Lar-son SM: Flare on bone scintigraphy following Taxol chemotherapy for metastatic breast cancer. *Journal of nuclear medicine : official publication, Society of Nuclear Medicine* 1994, 35(11):1748-1752.

[59] Mortimer JE, Dehdashti F, Siegel BA, Trinkaus K, Katzenellenbogen JA, Welch MJ: Metabolic flare: indicator of hormone responsiveness in advanced breast cancer. *Jour-nal of clinical oncology : official journal of the American Society of Clinical Oncology* 2001, 19(11):2797-2803.

[60] Dehdashti F, Mortimer JE, Trinkaus K, Naughton MJ, Ellis M, Katzenellenbogen JA, Welch MJ, Siegel BA: PET-based estradiol challenge as a predictive biomarker of re-sponse to endocrine therapy in women with estrogen-receptor-positive breast cancer. *Breast cancer research and treatment* 2009, 113(3):509-517.

[61] Fischer WH, Keiwan A, Schmitt E, Stopper H: Increased formation of micronuclei af-ter hormonal stimulation of cell proliferation in human breast cancer cells. *Mutagene-sis* 2001, 16(3):209-212.

[62] Trigonis I, Jackson A: Imaging pharmacodynamics in oncology: the potential signifi-cance of "flares". *Annals of nuclear medicine* 2010, 24(3):137-147.