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PET response assessment in apatinib-treated radioactive iodine-refractory thyroid cancer

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Abstract

This work evaluated the use of the positron emission tomography (PET)/computed tomography (CT) technique to assess the early therapeutic response and predict the prognosis of patients with radioactive iodine-refractory differentiated thyroid cancer (RAIR-DTC) who underwent apatinib therapy. Standardised uptake value (SUV), metabolic tumour volume (MTV) and total lesion glycolysis (TLG), derived from ¹⁸F-FDG PET/CT and SUV from ⁶⁸Ga-NOTA-PRGD2 PET/CT were evaluated. Tumour response was evaluated using the Response Evaluation Criteria in Solid Tumors (RECIST) 1.1. Sixteen of 20 patients achieved partial response (PR) and four of 20 had stable disease (SD) after apatinib therapy. Six progression-free survival (PFS) events occurred. A strong correlation was observed between the best change in the sum of the longest diameters of target lesions (Δ CT%) and ¹⁸F-FDG PET/CT indices after the completion of the first treatment cycle (Δ MTV% ($P=0.0019$), Δ TLG% ($P=0.0021$) and Δ SUVmax% ($P=0.0443$)). A significant difference in PFS was observed between patients with Δ MTV% $<-45\%$ and $\geq-45\%$ ($P=0.0019$) and between patients with Δ TLG% $<-80\%$ and $\geq-80\%$ ($P=0.0065$). Ten of 11 patients presented a decrease in SUVmax on ⁶⁸Ga-NOTA-PRGD2 PET/CT after two cycles of apatinib therapy and showed PR, whereas one patient presenting an increase in SUVmax only showed SD as the best response. When a cut-off value of the target/background ratio at -20% was used, two PFS curves showed a significant difference ($P=0.0016$). Hence, early assessment by ¹⁸F-FDG and ⁶⁸Ga-NOTA-PRGD2 PET/CT was effective in the prediction and evaluation of RAIR-DTC treated with apatinib.

Key Words

- ▶ radioactive iodine-refractory thyroid cancer
- ▶ apatinib
- ▶ PET
- ▶ integrin receptor $\alpha_v\beta_3$ imaging
- ▶ response assessment
- ▶ BRAFV600E
- ▶ TERT promoter mutation

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Introduction

Recent decades have witnessed a rapid increase in the incidence of differentiated thyroid cancer (DTC) worldwide (Chen *et al.* 2016, Siegel *et al.* 2016). Arising from aberrant follicular cells, DTC accounts for nearly

95% of all thyroid neoplasms (Brose *et al.* 2014). Most patients with DTC achieve favourable outcomes with surgery, radioactive iodine (RAI) therapy and thyroxine therapy; however, up to 10% of patients with metastatic

disease are eventually found to have radioactive iodine-refractory DTC (RAIR-DTC) (Schlumberger *et al.* 2015). Because of the low survival rate and limited therapeutic regimen, the treatment of RAIR-DTC has become a hot topic in the recent years (Ho *et al.* 2013, Brose *et al.* 2014, Schlumberger *et al.* 2015, Yang *et al.* 2017). With the development of mechanisms of vascular endothelial growth factor (VEGF) and its receptors (VEGFR), a series of tyrosine kinase inhibitors (TKIs) that target VEGF/VEGFR pathway have been proven effective against RAIR-DTC (Schlumberger *et al.* 2015, Lin *et al.* 2017). Traditionally, patients with RAIR-DTC were evaluated every 2 months after the therapy using the Response Evaluation Criteria in Solid Tumors (RECIST) based on conventional imaging technologies, such as computed tomography (CT). The identification of the biomarkers of an early therapeutic response to TKI therapy is of immense clinical relevance because it may help identify patients who are unlikely to benefit from the therapy. The rapid development of positron emission tomography (PET), PET/CT, new metabolic imaging and receptor-based imaging techniques has led to their use in clinical practice (Wahl *et al.* 2009). Studies have demonstrated the usefulness of ^{18}F -fluorodeoxyglucose (^{18}F -FDG) PET/CT for early response evaluation and prognosis assessment (Carr *et al.* 2010, Takahashi *et al.* 2012, Nakajo *et al.* 2017). Recently, except standardised uptake value (SUV), ^{18}F -FDG PET/CT parameters, such as metabolic tumour volume (MTV) and total lesion glycolysis (TLG), have been shown to be of much clinical relevance in solid tumours such as oesophageal and pancreatic adenocarcinoma (Sakane *et al.* 2017, van Rossum *et al.* 2017). However, little has been reported for DTC, particularly for RAIR-DTC, after TKI therapy.

Among the integrin family, the integrin receptor $\alpha_v\beta_3$ is one of the most important molecules that participate in tumour angiogenesis, invasion and metastasis (Zheng *et al.* 2015, Cheng *et al.* 2016). As a small peptide, arginine–glycine–aspartic acid (RGD) may selectively bind to $\alpha_v\beta_3$. This lock–key relationship allows the use of radionuclide labelled RGD to localise the tumour and for neovascularisation. ^{68}Ga -NOTA-PRGD2 has been used to detect lymph node metastases of lung cancer (Zheng *et al.* 2015). In our previous study, we employed $^{99\text{m}}\text{Tc}$ -3PRGD2 to localise RAIR-DTC lesions (Zhao *et al.* 2012). Furthermore, previous studies have demonstrated a correlation between integrin $\alpha_v\beta_3$ and VEGFR (Ehling *et al.* 2016, Helal-Neto *et al.* 2016). Zhu *et al.* have used RGD to evaluate synovial angiogenesis in patients with rheumatoid arthritis (Zhu *et al.* 2014). On the basis of

previous studies, it would be reasonable to use RGD as a predictor of efficacy and prognosis after TKI therapy targeting the VEGF/VEGFR pathway.

Apatinib, a novel small-molecule TKI that selectively targets VEGFR-2, has been shown to inhibit VEGF-mediated endothelial cell migration and proliferation and decrease tumour microvascular density (Ding *et al.* 2013). Our previous study has shown a quick response and encouraging efficacy of apatinib in RAIR-DTC (Lin *et al.* 2017). Therefore, in this study, we used PET to identify the early response and predict the prognosis of RAIR-DTC treated with apatinib.

Materials and methods

This study was approved by the Institutional Review Board of Peking Medical College Hospital Ethics Committee and was conducted in accordance with the Declaration of Helsinki and Good Clinical Practice guidelines. Written informed consent was obtained from all patients prior to their enrolment. This study was part of the prospective phase II clinical trial registered with ClinicalTrials.gov (number, NCT02731352).

Patients

Patients aged ≥ 18 years with histologically confirmed DTC were recruited. The inclusion criteria were patients who qualified the definition of RAIR-DTC; at least one progressive lesion according to RECIST (version 1.1) and disease progression within the past 14 months according to RECIST (version 1.1). Patients who qualified in at least one of the following criteria were defined as RAIR-DTC: (1) presence of at least one measurable lesion without iodine uptake on radioactive iodine scan; (2) presence of at least one measurable lesion that had progressed within the past 12 months even if it showed positive uptake of radioactive iodine or (3) received cumulative activity of $^{131}\text{I} \geq 600 \text{ mCi}$.

Study design and treatment regimen

Twenty patients were sequentially recruited for apatinib therapy (Jiangsu Hengrui Medicine Co., Ltd, Lianyungang, China). Initially, the first 10 patients were treated with apatinib at an initial dose of 750 mg based on a previous phase I study (Li *et al.* 2010) and a pharmacokinetics study (Ding *et al.* 2013). Despite encouraging efficacy, a high incidence of Grade 3 adverse events (AEs) was observed within 8 weeks of initiation of the treatment;

consequently, the dose was reduced to 500mg for the 10 subsequently enrolled patients. A treatment cycle was defined as 4 weeks (28 days). Treatment interruption due to the toxicity of apatinib was allowed for no more than 2 times or for a maximum duration of 2 weeks (either continuously or cumulatively) in one cycle. Dose reduction by 250mg q.d. each time was accepted, and re-escalation was not accepted. Treatment was continued until disease progression, drug intolerance or the withdrawal of consent for participation in the study. The study diagram is shown in Fig. 1.

The primary endpoints of this phase II clinical trial were the change in serum Tg concentration, disease control rate (DCR) and objective response rate (ORR), and the secondary endpoints were overall survival, progression-free survival (PFS) and duration of response. All patients were required to undergo ^{18}F -fluorodeoxyglucose (^{18}F -FDG) PET/CT scan thrice (at baseline and at the completion of 1st and 2nd treatment cycles). ^{68}Ga -NOTA-PRGD2 PET/CT scan twice (at baseline and after two cycles of treatment) was recommended but not mandatory. The present study aimed to analyse whether PET can be used to assess the early therapeutic response and predict the prognosis of RAIR-DTC treated with apatinib, regardless of the dosage.

Positron emission tomography imaging

^{18}F -FDG PET/CT scan was conducted to assess the response to apatinib. Briefly, patients were instructed to avoid strenuous work or exercise for at least 1 day and fast for >4h before the ^{18}F -FDG (0.15 mCi/kg) injection. After injection, patients rested in a warm, dark room for 40–60 min.

^{68}Ga -NOTA-PRGD2 was synthesised using the method described elsewhere (Zheng *et al.* 2015). The radiochemical purity of the product ^{68}Ga -NOTA-PRGD2 exceeded 95%. After intravenous injection of 111–185 MBq (3–5 mCi) of ^{68}Ga -NOTA-PRGD2, patients were asked to rest for nearly 40 min. The PET acquisitions were performed 1 h after the injection.

The acquisition was performed from skull to mid-thigh (five to six beds position, 2 min per bed) using a Siemens Biograph 64 Truepoint TrueV PET/CT scanner.

The PET/CT data sets were reconstructed using iterative reconstruction (ordered subset expectation maximum, OSEM) and attenuation correction. The results were measured using the software on the PET/CT system's workstation (TrueD, Siemens Medical Systems).

Figure 1
Study design.

Positron emission tomography image interpretation and metabolic parameter measurement

All PET images were interpreted consensually by two nuclear medicine physicians, who were blinded to the clinical data. Semi-quantitative analysis was performed on the PET/CT system's workstation. Target lesions according to RECIST 1.1 were selected at baseline and evaluated during follow-up. The primary tumour was defined as the volume of interest (VOI) and delineated on PET scans using an in-built semi-automatic 3D method.

SUV was derived using the following expression: (decay-corrected administered activity in kilobecquerels per millilitre of tissue volume)/(injected FDG activity in kilobecquerels/body weight in grams). MTV was derived by applying a threshold of 40% of the maximum standardised uptake value (SUVmax) as the lowest limit of the segmentation criteria. The computer-assisted automatically derived contouring margins and regions of interest for both measurements were checked on three sectional images (axial, coronal and sagittal) to ensure the accurate inclusion of all involved nodal sites and the exclusion of adjacent normal structures. TLG was calculated as $SUV_{mean} \times MTV$.

The following quantitative parameters were extracted from VOIs of the ^{18}F -FDG PET/CT scan: maximum and mean SUV (SUVmax and SUVmean), MTV and TLG. For ^{68}Ga -NOTA-PRGD2 PET/CT, only SUVmax and SUVmean were evaluated. Furthermore, the target lesions/background ratios (T/B, SUVmax of target lesions/SUVmean of background) were calculated for further analysis. $\Delta SUV\%$ was calculated using $(SUV - SUV_{baseline})/SUV_{baseline}$, as well as $\Delta MTV\%$ and $\Delta TLG\%$.

^{18}F -FDG positron emission tomography response evaluation

The ^{18}F -FDG response was evaluated using the European Organization for Research and Treatment of Cancer (EORTC) PET study group criteria. Complete metabolic response was achieved when the SUVs of all lesions decreased to the level of uptake equivalent in the background tissue. Partial metabolic response (PMR) was defined as a reduction of a minimum of 25% of the sum of SUVmax of target lesions. Stable metabolic disease (SMD) was classified as an increase in tumour sum of SUVmax less than 25% or decrease less than 25%, and no visible increase in the extent of tumour uptake. Progressive metabolic disease (PMD) was defined as an increase of the

sum of SUVmax >25% or visible increase in the extent of ^{18}F -FDG uptake (>20% in the longest dimension) or when new ^{18}F -FDG uptake appeared in metastatic lesions.

Assessment of biochemical and clinical response

Serum biochemical tests were performed at baseline and every two weeks during the initial two cycles and every 4 weeks after the second cycle. The tests included Tg, thyroglobulin antibody (TgAb) and thyroid-stimulating hormone (TSH).

Tumour diameters were measured at baseline, after one cycle and every two cycles thereafter using CT. The tumour response was evaluated as defined by RECIST (version 1.1). The percentage change in sum of tumour diameters ($\Delta CT\%$) was calculated by $(\text{diameter} - \text{diameter}_{baseline})/\text{diameter}_{baseline}$.

PFS was defined as the time from initial drug administration until disease progression or death, whichever occurred first. DCR was defined as complete response (CR), partial response (PR) or stable disease (SD); ORR was defined as CR and PR according to RECIST (version 1.1).

AEs were recorded and assessed at baseline until 4 weeks after the last drug administration in the study.

Measurement of Tg, TgAb and TSH

Tg and TgAb levels were determined using the electrochemiluminescence immunoassay (Roche Diagnostics GmbH) with functional sensitivity values of 0.100 ng/mL and 10 IU/mL, respectively. TSH was determined using the chemiluminescence immunoassay (Siemens Healthcare Diagnostics), with a measuring range from 0.004 to 150 μ IU/mL. TgAb values >100 IU/mL were considered positive. $\Delta Tg\%$ was calculated as $(Tg - Tg_{baseline})/Tg_{baseline}$.

BRAFV600E and TERT promoter mutation analysis

The primary tumour lesions of thyroid were used for the DNA analysis. Genomic DNA was extracted and analysed using the protocol reported previously (Yang *et al.* 2017).

Statistical analysis

The predictive accuracy of the changes in the four metabolic parameters (SUVmax, SUVmean, MTV and TLG) for tumour response was evaluated using receiver-operating characteristic (ROC) analysis; the area under the

curve (AUC) was used as an index of accuracy. Optimal cut-off values were derived from the ROC curves aiming for the best sensitivity and specificity. PFS curve was estimated using Kaplan–Meier analysis and compared using the log-rank (Mantel–Cox) test; the median and values around the median reduction in the four metabolic parameters were used to find the optimal cut-off values for PFS prediction analysis.

Categorical variables are expressed as frequencies and percentage; continuous variables are expressed as mean $\pm$ standard deviation (s.d.) or median and interquartile range when appropriate. A two-sample *t*-test or Wilcoxon rank-sum test was used to assess between-group differences. Correlation between two variables was analysed using Pearson or Spearman test according to the data. SPSS (version 19.0; SPSS) and Prism 6 (GraphPad Software) were used for statistical analyses. $P < 0.05$ was considered statistically significant.

Results

Patients

A total of 20 patients were enrolled in this study. The mean age of patients was 54.7 ± 13.3 (range, 27–78) years; the male-to-female ratio was 1:1. All patients underwent total or near-total thyroidectomy and lymph node (LN) resection, and the diagnosis of DTC was confirmed on histopathological examination. Fifteen of 20 patients lost the ability of RAI uptake in the metastatic lesions at the first RAI treatment after successful remnant ablation. The baseline characteristics of the included patients are summarised in Table 1. Five patients experienced failure of pre-TKI therapy; of these, four patients were treated with sorafenib and one patient was treated with both sorafenib and vandetanib. All patients underwent ^{18}F -FDG PET/CT scans on three occasions, while 11 patients underwent ^{68}Ga -NOTA-PRGD2 PET/CT on two occasions. Nine patients presented pulmonary metastases only, one patient presented bone metastases only and one patient presented mediastinal LN metastases. The other nine patients presented both pulmonary and bone ($N=5$) or head ($N=2$) or neck ($N=2$) metastases (Table 1).

Efficacy

The RECIST response to apatinib

During apatinib therapy, 16 (80%) patients achieved PR and four (20%) patients achieved SD, according to RECIST 1.1 (Table 1). The DCR and ORR of the study were 100%

(20/20) and 80% (16/20), respectively. After apatinib therapy, the sum of the longest diameters of target lesions decreased from 37.1 ± 15.5 to 20.7 ± 9.0 cm. The best change in the sum of the longest diameters of target lesions from baseline ($\Delta\text{CT}\%$) in all patients was -42.2% (range, -0.9% to -73.2% ; Fig. 2A).

From March 2016 to August 2017, six PFS events occurred, which included five disease progressions and one death. A female patient died of pulmonary infection after 5.53 months of therapy. Until she died, her target lesion response was still PR according to CT. Another patient quit the clinical trial after 6 months and showed SD at the end of the treatment. The estimated median PFS for patients with endpoints ($N=6$) was 6.5 months. The other 13 patients were still under follow-up with a treatment time range from 7.5 to 17.5 months.

Change in serum Tg levels

Tg levels were available for 15 out of the 20 patients. Five patients were excluded from the analyses; among these, three patients presented positive TgAb as previously described, and the other two patients showed quite low levels of Tg (0.43 and 0.36 ng/mL at baseline, respectively).

The mean Tg dropped from 2455.9 (range, 0.36–13,808) ng/mL to 101.2 (range, 396.1–0.1) ng/mL after apatinib therapy (Fig. 2B).

For the 15 Tg-eligible patients, the $\Delta\text{Tg}\%$ at 2-week ($\Delta\text{Tg}_{2w}\%$) and one cycle ($\Delta\text{Tg}_{1\text{cycle}}\%$) and the lowest $\Delta\text{Tg}\%$ were calculated. The $\Delta\text{Tg}_{\text{lowest}}\%$ ranged from -33.2% to -98.5% , $\Delta\text{Tg}_{2w}\%$ from $+47.4\%$ to -91.3% and $\Delta\text{Tg}_{1\text{cycle}}\%$ from $+38.7\%$ to -95.2% . Four patients showed an interim increase in Tg at 2 weeks or one cycle, followed by decrease during follow-up.

PET findings

^{18}F -FDG PET/CT scan

For one-cycle assessment, 16 PMR, three SMD and one PMD were detected according to the EORTC criteria. For two-cycle assessment, one patient was excluded as the target lesion was not amenable to evaluation because of the influence of distinct uptake of the heart. As a result, 14 patients achieved PMR and five had SMD.

At baseline, the initial mean sum of SUVmax of FDG uptake in target lesions was 12.27 (median, 8.38; range, 1.59–42.41). After one cycle of therapy, the mean SUVmax decreased to 5.41 (median, 4.26; range, 1.30–27.73). The mean SUVmax after two cycles of therapy was 4.51 (median, 4.36; range, 0.32–9.67). The other parameters also

Table 1 Characteristics of patients.

No.	Gender	Age	Pre-TKI therapy	Pathology	T	N	M	Metastases region [‡]	Braf	TERT	RAI dose (mCi)	Best response	Follow-up time (month)	Target lesion assessment
1	M	44	None	PTC	3	1b	1	P, H	+	+	300	PR	17.53	PR
2	M	48	None	PTC	NA	1a	1	P, B	–	+	305	PR	9.27	PD
3	M	78	None	PTC	4	1b	1	P	+	+	300	PR	17.17	PR
4	F	33	None	PTC	4	0	1	P	–	–	200	SD	6.03	SD*
5	M	52	None	PDTC	4	NA	1	P	–	+	350	PR	15.47	PR
6	F	75	None	PTC	4	1b	1	P, B	+	+	230	PR	5.53	PR [#]
7	M	65	Sorafenib	PTC	4	1b	1	P	+	NA	350	PR	15.63	PR
8	F	46	Sorafenib	PTC	3	1b	1	P, B	–	NA	630	PR	3.70	PD
9	F	64	None	PTC	4	1b	1	P	+	+	325	PR	14.87	PR
10	F	50	None	PTC	NA	NA	1	P	+	+	440	PR	14.83	PR
11	F	53	None	PTC, PDTC	3	1b	1	P, H	NA	–	400	PR	9.50	PD
12	F	68	Sorafenib	PTC	4a	1b	1	P	+	NA	300	PR	11.60	PR
13	M	49	None	PTC	NA	1b	1	P, B	+	–	750	PR	10.27	PR
14	F	61	Sorafenib, Vandetanib	PTC	3	NA	1	P, B	–	+	1150	PR	10.33	PR
15	F	27	Sorafenib	PTC	NA	1b	1	P	–	–	350	SD	3.70	PD
16	F	59	None	PTC	4a	1b	1	P	+	–	1100	PR	9.60	PR
17	M	50	None	PTC	1b	1b	1	M	+	+	420	SD	7.50	PD
18	M	38	None	PDTC, FTC	4b	1b	1	N, P	–	NA	320	PR	9.53	PR
19	M	70	None	FTC	1b	NA	1	B	–	–	1100	SD	7.47	SD
20	M	65	None	FTC	4a	1b	1	N, P	NA	NA	150	PR	9.27	PR

[‡]P, pulmonary; H, head; B, bone; M, mediastinum lymph node; N, neck lymph node; *this patient withdrew the consent; [#]this patient died of pulmonary infection. Bold indicates that PFS event occurred in these patients.

FTC, follicular thyroid cancer; NA, not available; PDTC, poorly differentiated thyroid cancer; PTC, papillary thyroid cancer.

exhibited a similar trend (mean SUVmean from 4.37 to 3.42 and 2.87; MTV from 10.87 to 5.73 and 4.78; TLG from 52.68 to 19.73 and 13.02, respectively) (Fig. 2C and D).

Correlation and predictive role of PET finding to morphologic responses

No significant correlation or agreement was observed between RECIST 1.1 and EORTC criteria (Spearman $P=0.217$, Kappa $P=0.197$).

A strong correlation was observed between the best $\Delta CT\%$ and $\Delta MTV\%$ (Spearman r 0.6511, $P=0.0019$), $\Delta TLG\%$ (Spearman r 0.6451, $P=0.0021$) and $\Delta SUV_{max}\%$ (Spearman r 0.4541, $P=0.0443$) at one-cycle treatment. Correlation of $\Delta MTV\%$ (Spearman r 0.7034, $P=0.0008$) and $\Delta TLG\%$ (Spearman r 0.7896, $P<0.0001$) with $\Delta CT\%$ was also significant in two-cycle PET/CT scan (Fig. 3).

Patients were divided into two groups according to the best response (RECIST 1.1): 16 patients in the PR group and three in the SD group (one patient excluded because of the influence of heart uptake as described earlier). The parameters were compared between the two groups. A significant between-group difference was observed with respect to $\Delta MTV\%$ and $\Delta TLG\%$, at the completion of the

first and second treatment cycles (Mann–Whitney U test, $P=0.019$; $P=0.044$, $P=0.019$ and $P=0.014$, respectively).

ROC curve showed that $\Delta MTV\%$ and $\Delta TLG\%$ at one-cycle had a great value in predicting PR response (AUC: 0.859 and 0.797, respectively). The optimal cut-off values of $\Delta MTV\%$ and $\Delta TLG\%$ at one-cycle (-20.78% and -56.34% , respectively) were associated with sensitivity values of 0.75 and 0.75, respectively, and specificity values of 0.94 and 0.81, respectively. In contrast, $\Delta SUV_{max}\%$ and $\Delta SUV_{mean}\%$ did not show any significant predictive value for PR response.

Comparative analyses between biochemistry and imaging responses

All the 15 patients eligible for the Tg analysis showed a significant decrease in the sum of the longest diameters of target lesions. The best $\Delta CT\%$ ranged from -18.6% to -73.2% . For PET response at one cycle, $\Delta SUV_{max}\%$ ranged from 75.6% to -82.7% , $\Delta MTV\%$ from -13.8% to -86.4% and $\Delta TLG\%$ from -19.5% to -95.4% . For the four patients who showed a temporary increase in Tg, the sum of the longest diameters of target lesions all showed shrinkage after apatinib therapy. Further correlation

Figure 2

Clinical response after apatinib therapy. (A) The best change in the sum of the longest diameters of target lesions ($\Delta CT\%$) according to RECIST 1.1. (B) The best change in Tg level after apatinib therapy. (C) Change in SUVmax after one cycle of apatinib therapy. (D) Change in MTV after one cycle of apatinib therapy. PR, partial metabolic response; PR, partial response; SD, stable disease; SMD, stable metabolic disease.

analyses (Spearman test) showed no significant correlation between the $\Delta Tg\%$ (the best, 2 weeks and one cycle) and $\Delta CT\%$ (the best, one cycle and two cycles), $\Delta SUVmax\%$, $\Delta MTV\%$ and $\Delta TLG\%$ at one cycle, respectively. The results indicate no linear relationship between biochemistry and imaging response (for the statistical value and *P* values, [Supplementary Table 1](#), see section on [supplementary data](#) given at the end of this article).

For the 15 patients, only one patient had SD, and the other 14 patients achieved PR as the best response. For the SD patient, a decrease in Tg level was also observed with the best $\Delta Tg\%$ of -98% . The Tg decrease seemed to have no predictive value for the response.

PFS according to FDG PET findings

The parameters of ^{18}F -FDG PET were used in the analyses. For $\Delta SUVmax\%$ and $\Delta SUVmean\%$ at one cycle or two cycles, the use of a cut-off value of -25% (EORTC criteria) failed to show a significant difference in PFS. Fortunately, both $\Delta MTV\%$ and $\Delta TLG\%$ showed promising results. When a cut-off value of -45% was used for $\Delta MTV\%$ at one cycle, a significant difference was observed between

the two PFS curves ($P=0.0019$) ($\geq -45\%$, $N=11$, median PFS 9.3 months (95% CI, 7.0–11.6); $< -45\%$ $N=9$, median PFS not achieved) ([Fig. 4A](#)), as well as $\Delta MTV\%$ at 2-cycle ($P=0.0001$) ($\geq -45\%$, $N=7$, median PFS 7.5 months (95% CI, 4.0–11.0); $< -45\%$ $N=12$, median PFS not achieved). Regarding $\Delta TLG\%$, when a cut-off value of -80% was used, significant differences were also observed at 1-cycle ($P=0.0065$) ($\geq -80\%$, $N=12$, median PFS 9.5 months (95% CI, 6.8–12.2); $< -80\%$ $N=12$, median PFS not achieved; [Fig. 4B](#)) and 2-cycle ($P=0.0029$) ($\geq -80\%$, $N=10$, median PFS 9.3 months (95% CI, 7.0–11.6), $< -80\%$ $N=9$, median PFS not achieved).

^{68}Ga -NOTA-PRGD2 PET/CT scan

Eleven patients underwent ^{68}Ga -NOTA-PRGD2 PET/CT scan. At baseline, the mean sum of SUVmax was 4.42 (range 0.97–8.69). After two treatment cycles, the mean sum of SUVmax decreased to 1.48 (range 0.22–2.18). There was no significant correlation between the reduction in RGD $\Delta SUVmax\%$ and FDG $\Delta SUVmax\%$, $\Delta MTV\%$, $\Delta TLG\%$, the best $\Delta CT\%$ or two-cycle $\Delta CT\%$. A total of 10 patients showed a decrease in SUVmax of ^{68}Ga -NOTA-PRGD2 after apatinib therapy and showed PR as the best

Figure 3

Correlation between MTV (A), TLG (B) change after one cycle of therapy and the best of the sum of the longest diameters of target lesions ($\Delta CT\%$). MTV, metabolic tumour volume; TLG, total lesion glycolysis.

Figure 4

Progression-free survival (PFS) according to different $\Delta MTV\%$ (A) and $\Delta TLG\%$ of one-cycle therapy. MTV, metabolic tumour volume; TLG, total lesion glycolysis.

Figure 5

The PET findings ^{68}Ga -NOTA-PRGD2 PET/CT Patient no. 14 (A) and 15 (B) before (left) and after (right) two cycles of apatinib therapy. (A) After two cycles of therapy, this patient showed a significant decrease in the uptake (SUVmax from 6.532 to 0.74) and presented partial response (PR) after the therapy (still PR after 10.3 months follow-up). (B) Patient showed an increase in the uptake (SUVmax from 0.975 to 1.239) and showed progression disease at the two-cycle (3.7 months) assessment. (C) Progress-free survival according to the ^{68}Ga -NOTA-PRGD2 PET findings.

response (Fig. 5A). One patient who showed an increase in SUVmax of ^{68}Ga -NOTA-PRGD2 after two treatment cycles only had SD as the best response (Fig. 5B). We further used target/background ratio (T/B, SUVmax of target lesions/SUVmean of background) for analysis. The mean T/B ratio decreased from 4.43 (range 1.10–10.04) to 1.86 (range 1.08–3.30). Nine patients presented a notable decrease in T/B ratio ($\Delta\text{T/B}\%$) after therapy (from -39.4% to -75.2%). Furthermore, one patient showed a limited change in T/B after therapy (-0.16%) and the other one showed an increase in T/B after apatinib therapy ($+19.5\%$). These two patients showed PD at four-cycle assessment. When a cut-off value of T/B ratio at -20% was used, the two survival curves showed a significant difference ($P=0.0016$; Fig. 5C). Hence, ^{68}Ga -NOTA-PRGD2 PET/CT may potentially be used not only as a response assessment tool but also as a means to predict post-treatment PFS.

BRAF and TERT promoter mutation

In total, two and five patients were not available for the detection of *BRAF*^{V600E} and *TERT* promoter mutations, respectively. Ten of the 18 patients harboured *BRAF*^{V600E} mutation, whereas eight tested negative. Nine of 15 patients tested positive for *TERT* promoter mutation, whereas six tested negative.

Out of the four patients who showed SD, one harboured *BRAF*^{V600E} mutation. In the PR group, nine of 14 harboured *BRAF* mutation. There was no significant difference between the two groups ($P=0.2745$). Eight of nine patients with *TERT* mutation achieved PR, whereas three of six in the *TERT*-negative group achieved PR; no significant between-group difference was observed with respect to PR rate ($P=0.2352$).

No significant difference was observed with respect to PFS between the *BRAF* mutation-positive and *BRAF*

mutation-negative subgroups ($P=0.3039$) and between the *TERT* mutation-positive and *TERT* mutation-negative groups ($P=0.7481$).

Safety

Most patients experienced treatment-related AEs, ranging from Grade 1–3 AEs; no severe AEs (SAE) related to apatinib were observed till data collection. The most common AEs were hand–foot skin reaction, proteinuria, increased AST levels, fatigue and hypertension. The details are shown in Supplementary Table 1.

Discussion

Recent years have witnessed rapid advances in the field of molecular targeted therapies including TKI therapy for thyroid cancer (Ferrara *et al.* 2003, Xing *et al.* 2013). Thus far, FDA has approved sorafenib and lenvatinib for the treatment of RAI-DTC (Brose *et al.* 2014, Schlumberger *et al.* 2015). Furthermore, a recent study has reported encouraging response rates and efficacy of apatinib (a novel TKI targeting VEGFR-2) in RAI-DTC after 2 months of treatment (Lin *et al.* 2017). In this study, although all patients did not achieve PR with apatinib therapy, the PR rate was still very high (16/20 patients). Out of the five patients who failed to respond to pre-TKI (sorafenib or vandetanib), four showed PR after apatinib therapy and experienced prolonged PFS. These findings are encouraging and suggest the role of apatinib as a salvage treatment after first-line TKI therapy. Nevertheless, not all patients benefited much from apatinib therapy. Therefore, the early prediction of tumour response and prognosis is of great importance for the optimisation of the management of patients on TKI therapy because of the multiple side effects and uncertain efficacy. Therefore, we conducted this study to identify the factors that may

help predict the response to and the outcomes of TKI therapy in patients with RAIR.

Functional imaging modalities, such as PET, represent an invaluable clinical tool. ^{18}F -FDG PET/CT parameters, such as MTV and TLG, have been shown to predict the therapeutic response in patients with oesophageal cancer (Nakajo *et al.* 2017). However, their role in thyroid cancer (particularly for RAIR-DTC) is not well characterised. To the best of our knowledge, our work may be the first research focusing on the ^{18}F -FDG PET/CT parameters in RAIR-DTC. Both $\Delta\text{MTV}\%$ and $\Delta\text{TLG}\%$ showed encouraging values in predicting response and prognosis compared with $\Delta\text{SUVmax}\%$ and $\Delta\text{SUVmean}\%$. Parameters such as MTV and TLG reflect the metabolic status of the entire tumour and are potentially more accurate for tumour characterisation than is SUVmax, which only reveals a single voxel among the tumour region. In our study, $\Delta\text{MTV}\%$ and $\Delta\text{TLG}\%$ were found valuable in two aspects. First, because of the strong correlation between the best $\Delta\text{CT}\%$ and $\Delta\text{MTV}\%$ (Spearman r 0.6511, $P=0.0019$) and between the best $\Delta\text{CT}\%$ and $\Delta\text{TLG}\%$ (Spearman r 0.6451, $P=0.0021$), clinicians may use $\Delta\text{MTV}\%$ and $\Delta\text{TLG}\%$ at the first cycle to predict the shrinkage of tumour lesions. Furthermore, the extent of $\Delta\text{MTV}\%$ and $\Delta\text{TLG}\%$ also showed a significant difference between the PR and SD groups ($P=0.019$, $P=0.044$, $P=0.019$ and $P=0.014$, respectively). ROC analyses also demonstrated the usefulness of $\Delta\text{MTV}\%$ and $\Delta\text{TLG}\%$ in predicting PR. This provides clinicians a convenient tool for the early prediction and evaluation, which may help avoid unnecessary TKI therapy. Second, we found that both $\Delta\text{MTV}\%$ and $\Delta\text{TLG}\%$ showed a correlation with PFS. As one of the most important parameter for cancer patients, PFS may reflect the patient's benefit and prognosis. The ability to predict disease progression at an early stage is of vital importance because it may facilitate any change in the treatment regimen and optimise patient management strategy. Consequently, both clinicians and radiologists may use ^{18}F -FDG PET/CT to predict the prognosis of patients undergoing TKI therapy during the early stages. This may help avoid the unnecessary treatment of patients who are unlikely to benefit from TKI and to timely identify the PD status.

The best time for early PET is unclear. Takahashi *et al.* conducted PET scan 2 days after gefitinib therapy in patients with lung cancer (Takahashi *et al.* 2012). Wang *et al.* performed PET 14 days after chemotherapy for gastric cancer. Carr *et al.* conducted PET scan 7 days after sunitinib therapy for thyroid cancer but failed to achieve any significant result (Carr *et al.* 2010). It is widely

known that due to the inflammatory reaction induced by chemotherapy for lymphoma, it is recommended that ^{18}F -FDG PET in patients with lymphoma should be performed 3 weeks after the completion of chemotherapy (Barrington *et al.* 2014). Although it is unclear whether TKI can result in the same consequences, we opted for PET scan at one cycle (28 days) after TKI therapy.

In this work, we also used ^{68}Ga -NOTA-PRGD2 PET/CT as an evaluator and predictor, probably the earliest work in the thyroid field. Previous study used $^{99\text{m}}\text{Tc}$ -3PRGD2 to detect and locate the RAIR lesions while simultaneously revealing the uptake of RGD in lesions of RAIR-DTC. We further used ^{68}Ga -labelled RGD and PET imaging for pre- and post-treatment evaluation of uptake at the target lesions. The uptake of RGD may reflect neovascularisation as it selectively binds to $\alpha_v\beta_3$ expressed in neo-vascular areas in tumour tissues. Like all of the anti-angiogenesis drugs, apatinib targets VEGFR-2 and exerts its anti-tumour effect by blocking the VEGF/VEGFR waterfall signal pathway and inhibiting neovascularisation. Till now, there has been no specific tool to predict the prognosis after TKI therapy targeting VEGF/VEGFR pathway. Bao *et al.* used ^{18}F -Alfatide II to monitor the early response in U87MG tumour mice (Bao *et al.* 2016). Hence, it would be a useful tool if RGD imaging could reveal the response and prognosis in patients treated with TKI. In this work, ^{68}Ga -NOTA-PRGD2 was not only effective in evaluation of response, but was also found to be a potential predictor of prognosis after TKI therapy. According to our results, two patients with an insignificant decrease in RGD uptake (revealed by $\Delta\text{T/B}\%$) showed PD after two cycles of apatinib therapy. Despite the small sample size, ^{68}Ga -NOTA-PRGD2 may prove to be a useful tool for response evaluation and prognostic assessment of patients treated with TKI.

In recent years, advances in molecular pathology have played a key role in unravelling the pathogenesis of cancers, such as the role of $\text{BRAF}^{\text{V600E}}$ and TERT promoter in thyroid cancer (Xing 2013, Xing *et al.* 2013, 2014). Xing *et al.* reported the pejorative survival of thyroid patients harbouring $\text{BRAF}^{\text{V600E}}$ or TERT mutation (Xing *et al.* 2013, 2014). Yang *et al.* also found that Chinese patients with TERT mutation were refractory to radioactive iodine therapy (Yang *et al.* 2017). Initial work also showed a relationship between $\text{BRAF}^{\text{V600E}}$ mutation and dedifferentiation of thyroid cancer (Xing 2007). Similarly, patients harbouring both $\text{BRAF}^{\text{V600E}}$ and TERT mutation were refractory to radioactive iodine, which may result in unfavourable prognosis (Yang *et al.* 2017). Previous studies have reported the prognosis of patients

with *BRAF*^{V600E} mutation after TKI therapy (Brose *et al.* 2014, Tahara *et al.* 2017); in both placebo- and sorafenib/lenvatinib-treated groups, patients with the *BRAF*^{V600E} mutation showed a better response than those with wild-type *BRAF*. Moreover, PFS benefit was maintained in patients harbouring the *BRAF*^{V600E} mutation. However, the *BRAF* mutation status was not predictive of TKI benefit according to the biomarker–treatment interaction analysis. Regarding *TERT* promoter mutation, not much evidence is available for patients with RAIR treated with TKI. Our study included 14 patients with *BRAF*^{V600E} or *TERT* promoter mutation. We found no significant association of *BRAF*^{V600E} or *TERT* promoter mutation status with the therapeutic response or PFS, which may suggest that TKI therapy improves the unfavourable prognoses of patients harbouring the *BRAF*^{V600E} or *TERT* promoter mutation under the natural course of the disease.

As the foremost biomarker of DTC, Tg revealed tumour burden and treatment response. Yang *et al.* reported the practical utility of Tg to predict post-RAI therapy prognosis using the response system of DTC (Yang *et al.* 2016). As Tg is produced both by normal thyroid tissues and DTC cells, it may reflect the tumour burden in patients with DTC, whereas Tg level in patients with RAIR-DTC varied from 0.36 to 13,808 ng/mL in our study. The two patients with low Tg levels (0.36 and 0.43 ng/mL) harboured a large tumour burden. We believe that in this circumstance, since the tumour was undergoing dedifferentiation and was refractory to iodine, Tg might not be a sensitive marker of tumour burden or response as compared to that in patients with well-differentiated DTC. Despite the fact that Tg levels in all patients showed a decreasing trend after apatinib therapy, the Δ Tg% level showed no correlation with imaging findings. Furthermore, four patients showed temporary increase in Tg after apatinib therapy at two or four weeks, followed by a decrease during follow-up. This temporary increase in Tg level might be due to treatment-induced injury and death of tumour cells. Thus, we think that Tg had nothing to do with prognosis but may be of value in predicting response.

In summary, early assessment by ¹⁸F-FDG and ⁶⁸Ga-NOTA-PRGD2 PET/CT may help in prognostic assessment of patients with RAIR-DTC treated with apatinib. Reductions in MTV and TLG were both useful for the prediction of the response and PFS. ⁶⁸Ga-NOTA-PRGD2 might be a potential tool for the evaluation of RAIR-DTC treated with TKI. Both *BRAF*^{V600E} and *TERT* promoter mutations failed to show an obvious influence on the response and PFS after apatinib therapy. Considering the small sample size in this study, these encouraging results

will be verified in a phase III clinical trial (ClinicalTrials.gov, NCT03048877).

Supplementary data

This is linked to the online version of the paper at <https://doi.org/10.1530/ERC-18-0007>.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

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References

- Bao X, Wang MW, Luo JM, Wang SY, Zhang YP & Zhang YJ 2016 Optimization of early response monitoring and prediction of cancer antiangiogenesis therapy via noninvasive PET molecular imaging strategies of multifactorial bioparameters. *Theranostics* **6** 2084–2098. (<https://doi.org/10.7150/thno.13917>)
- Barrington SF, Mikhaeel NG, Kostakoglu L, Meignan M, Hutchings M, Mueller SP, Schwartz LH, Zucca E, Fisher RI, Trotman J, *et al.* 2014 Role of imaging in the staging and response assessment of lymphoma: consensus of the International Conference on Malignant Lymphomas Imaging Working Group. *Journal of Clinical Oncology* **32** 3048–3058. (<https://doi.org/10.1200/JCO.2013.53.5229>)
- Brose MS, Nutting CM, Jarzab B, Elisei R, Siena S, Bastholt L, de la Fouchardiere C, Pacini F, Paschke R, Shong YK, *et al.* 2014 Sorafenib in radioactive iodine-refractory, locally advanced or metastatic differentiated thyroid cancer: a randomised, double-blind, phase 3 trial. *Lancet* **384** 319–328. ([https://doi.org/10.1016/S0140-6736\(14\)60421-9](https://doi.org/10.1016/S0140-6736(14)60421-9))
- Carr LL, Mankoff DA, Goulart BH, Eaton KD, Capell PT, Kell EM, Bauman JE & Martins RG 2010 Phase II study of daily sunitinib in FDG-PET-positive, iodine-refractory differentiated thyroid cancer and metastatic medullary carcinoma of the thyroid with functional imaging correlation. *Clinical Cancer Research* **16** 5260–5268. (<https://doi.org/10.1158/1078-0432.CCR-10-0994>)
- Chen W, Zheng R, Baade PD, Zhang S, Zeng H, Bray F, Jemal A, Yu XQ & He J 2016 Cancer statistics in China, 2015. *CA: A Cancer Journal for Clinicians* **66** 115–132. (<https://doi.org/10.3322/caac.21338>)
- Cheng W, Feng F, Ma C & Wang H 2016 The effect of antagonizing RGD-binding integrin activity in papillary thyroid cancer cell lines. *OncoTargets and Therapy* **9** 1415–1423. (<https://doi.org/10.2147/OTT.S99166>)

- Ding J, Chen X, Gao Z, Dai X, Li L, Xie C, Jiang H, Zhang L & Zhong D 2013 Metabolism and pharmacokinetics of novel selective vascular endothelial growth factor receptor-2 inhibitor apatinib in humans. *Drug Metabolism and Disposition* **41** 1195–1210. (<https://doi.org/10.1124/dmd.112.050310>)
- Ehling J, Misiewicz M, von Stillfried S, Mockel D, Bzyl J, Pochon S, Lederle W, Knuechel R, Lammers T, Palmowski M, *et al.* 2016 In situ validation of VEGFR-2 and alpha v ss 3 integrin as targets for breast lesion characterization. *Angiogenesis* **19** 245–254. (<https://doi.org/10.1007/s10456-016-9499-4>)
- Ferrara N, Gerber HP & LeCouter J 2003 The biology of VEGF and its receptors. *Nature Medicine* **9** 669–676. (<https://doi.org/10.1038/nm0603-669>)
- Helal-Neto E, Brandao-Costa RM, Saldanha-Gama R, Ribeiro-Pereira C, Midlej V, Benchimol M, Morandi V & Barja-Fidalgo C 2016 Priming endothelial cells with a melanoma-derived extracellular matrix triggers the activation of alphavbeta3/VEGFR2 axis. *Journal of Cellular Physiology* **231** 2464–2473. (<https://doi.org/10.1002/jcp.25358>)
- Ho AL, Grewal RK, Leboeuf R, Sherman EJ, Pfister DG, Deandreis D, Pentlow KS, Zanzonico PB, Haque S, Gavane S, *et al.* 2013 Selumetinib-enhanced radioiodine uptake in advanced thyroid cancer. *New England Journal of Medicine* **368** 623–632. (<https://doi.org/10.1056/NEJMoa1209288>)
- Li J, Zhao X, Chen L, Guo H, Lv F, Jia K, Yv K, Wang F, Li C, Qian J, *et al.* 2010 Safety and pharmacokinetics of novel selective vascular endothelial growth factor receptor-2 inhibitor YN968D1 in patients with advanced malignancies. *BMC Cancer* **10** 529. (<https://doi.org/10.1186/1471-2407-10-529>)
- Lin Y, Wang C, Gao W, Cui R & Liang J 2017 Overwhelming rapid metabolic and structural response to apatinib in radioiodine refractory differentiated thyroid cancer. *Oncotarget* **8** 42252–42261. (<https://doi.org/10.18632/oncotarget.15036>)
- Nakajo M, Jinguji M, Nakabeppu Y, Nakajo M, Higashi R, Fukukura Y, Sasaki K, Uchikado Y, Natsugoe S & Yoshiura T 2017 Texture analysis of (18)F-FDG PET/CT to predict tumour response and prognosis of patients with esophageal cancer treated by chemoradiotherapy. *European Journal of Nuclear Medicine and Molecular Imaging* **44** 206–214. (<https://doi.org/10.1007/s00259-016-3506-2>)
- Sakane M, Tatsumi M, Hori M, Onishi H, Tsuboyama T, Nakamoto A, Ota T, Eguchi H, Wakasa K, Hatazawa J, *et al.* 2017 Volumetric parameters of 2-deoxy-2-[18F]fluoro-d-glucose positron emission tomography/computed tomography can predict histopathologic treatment response after neoadjuvant chemoradiotherapy in pancreatic adenocarcinoma. *European Journal of Radiology* **94** 64–69. (<https://doi.org/10.1016/j.ejrad.2017.05.021>)
- Schlumberger M, Tahara M, Wirth LJ, Robinson B, Brose MS, Elisei R, Habra MA, Newbold K, Shah MH, Hoff AO, *et al.* 2015 Lenvatinib versus placebo in radioiodine-refractory thyroid cancer. *New England Journal of Medicine* **372** 621–630. (<https://doi.org/10.1056/NEJMoa1406470>)
- Siegel RL, Miller KD & Jemal A 2016 Cancer statistics, 2016. *CA: A Cancer Journal for Clinicians* **66** 7–30. (<https://doi.org/10.3322/caac.21332>)
- Tahara M, Schlumberger M, Elisei R, Habra MA, Kiyota N, Paschke R, Dutcus CE, Hihara T, McGrath S, Matijevic M, *et al.* 2017 Exploratory analysis of biomarkers associated with clinical outcomes from the study of lenvatinib in differentiated cancer of the thyroid. *European Journal of Cancer* **75** 213–221. (<https://doi.org/10.1016/j.ejca.2017.01.013>)
- Takahashi R, Hirata H, Tachibana I, Shimosegawa E, Inoue A, Nagatomo I, Takeda Y, Kida H, Goya S, Kijima T, *et al.* 2012 Early [18F]fluorodeoxyglucose positron emission tomography at two days of gefitinib treatment predicts clinical outcome in patients with adenocarcinoma of the lung. *Clinical Cancer Research* **18** 220–228. (<https://doi.org/10.1158/1078-0432.CCR-11-0868>)
- van Rossum PS, Fried DV, Zhang L, Hofstetter WL, Ho L, Meijer GJ, Carter BW, Court LE & Lin SH 2017 The value of (18)F-FDG PET before and after induction chemotherapy for the early prediction of a poor pathologic response to subsequent preoperative chemoradiotherapy in oesophageal adenocarcinoma. *European Journal of Nuclear Medicine and Molecular Imaging* **44** 71–80. (<https://doi.org/10.1007/s00259-016-3478-2>)
- Wahl RL, Jacene H, Kasamon Y & Lodge MA 2009 From RECIST to PERCIST: Evolving Considerations for PET response criteria in solid tumors. *Journal of Nuclear Medicine* **50** (Supplement 1) 122S–150S. (<https://doi.org/10.2967/jnumed.108.057307>)
- Xing M 2007 BRAF mutation in papillary thyroid cancer: pathogenic role, molecular bases, and clinical implications. *Endocrine Reviews* **28** 742–762. (<https://doi.org/10.1210/er.2007-0007>)
- Xing M 2013 Molecular pathogenesis and mechanisms of thyroid cancer. *Nature Reviews Cancer* **13** 184–199. (<https://doi.org/10.1038/nrc3431>)
- Xing M, Alzahrani AS, Carson KA, Viola D, Elisei R, Bendlova B, Yip L, Mian C, Vianello F, Tuttle RM, *et al.* 2013 Association between BRAF V600E mutation and mortality in patients with papillary thyroid cancer. *JAMA* **309** 1493–1501. (<https://doi.org/10.1001/jama.2013.3190>)
- Xing M, Liu R, Liu X, Murugan AK, Zhu G, Zeiger MA, Pai S & Bishop J 2014 BRAF V600E and TERT promoter mutations cooperatively identify the most aggressive papillary thyroid cancer with highest recurrence. *Journal of Clinical Oncology* **32** 2718–2726. (<https://doi.org/10.1200/JCO.2014.55.5094>)
- Yang X, Liang J, Li T, Zhao T & Lin Y 2016 Preablative stimulated thyroglobulin correlates to new therapy response system in differentiated thyroid cancer. *Journal of Clinical Endocrinology and Metabolism* **101** 1307–1313. (<https://doi.org/10.1210/jc.2015-4016>)
- Yang X, Li J, Li X, Liang Z, Gao W, Liang J, Cheng S & Lin Y 2017 TERT promoter mutation predicts radioiodine-refractory character in distant metastatic differentiated thyroid cancer. *Journal of Nuclear Medicine* **58** 258–265. (<https://doi.org/10.2967/jnumed.116.180240>)
- Zhao D, Jin X, Li F, Liang J & Lin Y 2012 Integrin alphavbeta3 imaging of radioactive iodine-refractory thyroid cancer using 99 mTc-3PRGD2. *Journal of Nuclear Medicine* **53** 1872–1877. (<https://doi.org/10.2967/jnumed.112.107821>)
- Zheng K, Liang N, Zhang J, Lang L, Zhang W, Li S, Zhao J, Niu G, Li F, Zhu Z, *et al.* 2015 68Ga-NOTA-PRGD2 PET/CT for integrin imaging in patients with lung cancer. *Journal of Nuclear Medicine* **56** 1823–1827. (<https://doi.org/10.2967/jnumed.115.160648>)
- Zhu Z, Yin Y, Zheng K, Li F, Chen X, Zhang F & Zhang X 2014. Evaluation of synovial angiogenesis in patients with rheumatoid arthritis using ⁶⁸Ga-PRGD2 PET/CT: a prospective proof-of-concept cohort study. *Annals of the Rheumatic Diseases* **73** 1269–1272. (<https://doi.org/10.1136/annrheumdis-2013-204820>)

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