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Cold Kit PSMA PET Imaging: Phase I study of ^{68}Ga-THP-PSMA PET/CT in patients with prostate cancer

Short running title: **THP-PSMA PET/CT in Prostate Cancer**

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Key Words: prostate specific membrane antigen, PET/CT, prostate cancer, Gallium-68

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Abstract

Objectives: Ga-68 labelled urea-based inhibitors of the prostate-specific membrane antigen (PSMA), such as ^{68}Ga-HBED-PSMA-11, are promising small molecules for targeting prostate cancer. A new radiopharmaceutical ^{68}Ga-THP-PSMA has a simplified design for one-step kit-based radiolabelling. It features the tris(hydroxypyridinone) (THP) ligand, which complexes $^{68}Ga^{3+}$ rapidly at low concentration, room temperature and over a wide pH range, enabling direct elution from a ^{68}Ge/^{68}Ga generator into a lyophilized kit in one-step without manipulation. This Phase I study aimed to assess the safety and biodistribution of ^{68}Ga-THP-PSMA. **Methods:** Cohort A: 8 patients with proven prostate cancer scheduled to undergo prostatectomy underwent PET/CT following administration of ^{68}Ga-THP-PSMA (Gleason score 7-10; PSA mean 7.8, range 5.4-10.6). All patients proceeded to prostatectomy (7 with pelvic nodal dissection). Dosimetry was performed with OLINDA/EXM from multi-time point PET imaging. Cohort B: 6 patients with a positive ^{68}Ga-HBED-PSMA-11 PET/CT underwent comparative ^{68}Ga-THP-PSMA scan. All patients were followed to evaluate for adverse events. **Results:** No adverse events occurred. Cohort A: Six of 8 patients had focal uptake in the prostate (at 2 h, average SUVmax 5.1, range 2.4–9.2) with correlative 3+ staining on PSMA immunohistochemistry on prostatectomy specimens. The two ^{68}Ga-THP-PSMA negative scans had only 1+/2+ staining. The mean effective dose was 2.07E-02 mSv/MBq. Cohort B: ^{68}Ga-THP-PSMA had lower physiologic background uptake compared to ^{68}Ga-HBED-PSMA-11 (parotid: mean SUVmax 3.6 compared to 19.2, liver: 2.7 to 6.3, spleen 2.7 to 10.5, $p<0.001$ for all). In 5 of 6 patients there was concordance in the number of metastases identified with ^{68}Ga-HBED-PSMA-11 and ^{68}Ga-THP-PSMA. 13 of 15 nodal abnormalities were sub-centimetre. In 22 malignant lesions, tumor-to-liver contrast was similar on THP-PSMA compared to HBED-PSMA (4.7 to 5.4, $p=0.15$) despite higher SUVmax with HBED-PSMA (30.3 to 10.7, $p<0.01$). **Conclusion:** ^{68}Ga-THP-PSMA is safe with favourable biodistribution for clinical imaging. Observed focal uptake in the prostate localized to PSMA-expressing malignant tissue on histopathology. Metastatic PSMA-avid foci are also visualized with ^{68}Ga-THP-PSMA PET. Single step production from a GMP cold kit may enable rapid adoption.

Keywords: prostate cancer, positron emission tomography, prostate specific membrane antigen

Prostate specific membrane antigen (PSMA) is a highly overexpressed cell surface glycoprotein in prostate cancer (*1-3*). ^{68}Ga-labeled urea-based small molecules that bind with high affinity to the extra-cellular domain of PSMA, such as ^{68}Ga-HBED-PSMA-11, have rapidly emerged as a disruptive technology for imaging prostate cancer (*4-6*). Positron emission tomography (PET) with ^{68}Ga-HBED-PSMA-11 produces high tumor-to-background contrast and has been rapidly adopted in jurisdictions where the regulatory environment enables on-site, extemporaneously compounded radiotracers to be adopted in the clinic. In other jurisdictions, differing regulatory requirements has limited adoption of PSMA PET. Routine clinical cGMP production of ^{68}Ga-HBED-PSMA11 is limited by synthesis time, and the need for an expensive automated synthesis device and onsite radiochemistry expertise.

^{68}Ga-HBED-PSMA-11 PET/CT has demonstrated clinical utility in both initial staging of newly diagnosed patients and restaging patients in the setting of biochemical recurrence and has rapidly emerged as a practice-changing modality for imaging prostate cancer. More accurate staging of high-risk patients may enable better selection of the most appropriate management strategy and avoid futile locoregional surgery or radiotherapy in patients with metastatic disease. For staging, several studies have demonstrated high sensitivity compared to conventional imaging (*7-10*). In the setting of early biochemical recurrence, PSMA PET/CT has high sensitivity even in patients with PSA below 1.0 ng/ml where conventional imaging is invariably unhelpful (*11*). PSMA PET/CT can have a consequent high management impact in these patients (*12*).

A new radiopharmaceutical, ^{68}Ga-THP-PSMA, for the same diagnostic purpose as ^{68}Ga-HBED-PSMA-11 but designed for greatly simplified one-step kit-based radiolabeling, has been developed (*13*). It features the tris(hydroxypyridinone) (THP) ligand, which complexes $^{68}Ga^{3+}$ rapidly at low concentration, room temperature and over a wide pH range. This enables elution from a ^{68}Ge/^{68}Ga generator, with low germanium-68 breakthrough, into a lyophilized radiopharmaceutical kit in one step. Pre-clinical studies have validated specific binding to PSMA-expressing cells and a biodistribution similar to ^{68}Ga-HBED-PSMA-11(*13*).

This phase I study assesses the safety and biodistribution of ^{68}Ga-THP-PSMA. The trial hypotheses are that (1) ^{68}Ga-THP-PSMA can be safely administered, and (2) its biodistribution will facilitate specific and sensitive identification of PSMA-expressing malignant tissues using PET/CT, with an acceptable absorbed radiation dose.

MATERIALS AND METHODS

This investigator-initiated prospective study was sponsored by the Peter MacCallum Cancer Centre, and ethically approved by the institutional review board. It was prospectively registered (Australian New Zealand Clinical Trials Registry No 12615001324505). All subjects gave informed consent.

Fourteen patients with biopsy proven adenocarcinoma of the prostate were recruited; eight in Group A and six in Group B. Safety and biodistribution of ^{68}Ga-THP-PSMA were assessed in all patients. In Group A, additional aims were to define whole body radiation dose and plasma radiotracer clearance, and correlate uptake with tumor PSMA expression on histopathology. In Group B, the aim was to compare physiologic and pathologic biodistribution in patients with PSMA-avid malignant disease on ^{68}Ga-HBED-PSMA-11 PET/CT.

In Group A, patients had no prior treatment for prostate carcinoma and were scheduled for prostatectomy. They underwent multiple whole body PET/CT scans as detailed below to determine biodistribution and dosimetry of ^{68}Ga-THP-PSMA. Regions of interest were drawn around standard organs and intensity of uptake measured using SUVmax. Absorbed organ and whole body doses were determined by calculation of time activity values for standard organs using OLINDA/EXM version 1.1 (*14*).

A positive scan was defined by visual analysis if there was uptake in the prostate above background and was localised using a 4-segment prostate model. Intensity of uptake was measured using SUVmax. Nodal or distant metastases were identified by visual interpretation.

All Group A patients proceeded to prostatectomy. Samples were fixed in formalin and handled as per routine practice. 3 µm sections cut from paraffin blocks containing tumor were immunohistochemically stained for PSMA (Dako (Agilent) clone 3E6, 1:100 dilution) using a Ventana Benchmark Ultra automated stainer, with incubation period 32 minutes, high-pH epitope-retrieval buffer using Ventana CC1 solution for 40 minutes, and detection using the Ventana Optiview Detection kit (DAB based). PSMA staining was reviewed by a histopathologist with urologic subspecialty expertise, and scored as absent (0), mild (1+), moderate (2+) or strong (3+) along with the proportion of tumor cells stained. Areas of strong staining for PSMA (3+) were considered positive.

In Group B, inclusion criteria additionally mandated patients with prior clinical ^{68}Ga-HBED-PSMA-11 PET/CT demonstrating at least one unequivocal PSMA-avid focus considered to represent metastatic prostate cancer. Management did not change between the two studies, which were performed within a mean of 12 days (range 2-22). For ^{68}Ga-HBED-PSMA-11, the mean administered activity was 144 MBq (range 63 to 205 MBq), and mean uptake time was 56 min (range 49 to 72 min); patients were imaged on a GE Discovery 690, GE Discovery 710 or Siemen's Biograph 64 PET/CT scanner. ^{68}Ga-THP-PSMA PET/CT acquisition is detailed below. Studies were reviewed blinded to the ^{68}Ga-HBED-PSMA-11 findings and abnormalities were enumerated in prostate bed, regional nodes, non-regional nodes, bone metastases and visceral metastases. Tumor-to-liver and SUVmax of up to 5 lesions per region were measured on both ^{68}Ga-HBED-PSMA-11 and ^{68}Ga-THP-PSMA.

Formulation of ^{68}Ga-THP-PSMA and PET/CT imaging

Group A. ^{68}Ga was eluted from a ^{68}Ge/^{68}Ga generator (iThemba supplied by IDB Holland BV, Netherlands). Plotting the elution profile of this generator showed that the bulk of the Ga-68 eluted with 5 ml of 0.5 N Hydrochloric acid (HCl) was in the second ml. Accordingly, the generator was first eluted with 1 mL 0.5 N HCl into waste. A second syringe containing 1 ml 0.5 N HCl was then connected to the

generator and the outlet was connected via a hypodermic needle directly into an appropriately shielded, vented 40 µg THP-PSMA kit (Theragnostics Ltd, United Kingdom) vial. Radiolabeling was achieved by adding this eluate slowly into the kit and diluting to 5 ml with sterile water for injection. Both needles were then removed and a sample was taken for assessment of pH, Thin Layer Chromatography (TLC), High Pressure Liquid Chromatography (HPLC) and half-life.

Group B. A change in ^{68}Ge/^{68}Ga generator (Galli Eo, IRE-Elit, Fleurus, Belgium) for this substudy involved different radiolabelling method. 30 mL evacuated vial (Gentech, ANSTO Health, Australia) was used to draw the eluate approximately 1 ml directly from the generator into the THP-PSMA vial as per manufacturer's instruction. Another 4 mL of 0.1N HCl was added to the kit and all needles were removed. A sample was taken immediately for quality control.

Quality control studies. Shimadzu HPLC with both UV detector (254 nm) and radiation detection (Ortec 276 Photomultiplier Base with Preamplifier, Amplifier, Bias supply and SCA and a Bicron 1M 11.2 Photomultiplier) was used, with a Phenomenex Kinetex XB-C18 column (150 x 4.6 mm, 5µm) at ambient temperature flow rate 1.0 ml/min and injection volume 100 µl. Approximate retention times under these conditions were 1.5 min for ^{68}Ga, 12.0 min for THP-PSMA and 14.0 min for ^{68}Ga-THP-PSMA. The mobile phase was TFA : Water : Acetonitrile (0.05 : 90 : 10). For TLC, iTLC-SG Varian #SGI0001 strips were used with a mobile phase of 7.7 % w/v ammonium acetate : methanol (50:50) and a spot volume of 5 µl. The approximate Rf was 0.0 for colloidal ^{68}Ga, 0.0 for free unbound ^{68}Ga and 1.0 for ^{68}Ga-THP-PSMA. pH was 6 - 7. The radiochemical yield and purity (activity of ^{68}Ga -THP-PSMA as a percentage of total $^{68}Ga^{3+}$ activity) was >99%. ^{68}Ge breakthrough by half-life measurement was <0.001% for both generators.

Dose and activity administered. The mean administered mass of THP-PSMA was 35 µg $\pm$ 10% (range 32 to 38 µg). The mean administered activity of ^{68}Ga was 238 MBq (range 228 – 270 MBq) for Cohort A, and 232 MBq (range 172 to 275 MBq) for Cohort B.

PET/CT imaging. Images were acquired on a GE Discovery 690 PET/CT device incorporating time-of-flight. Acquisition protocols are summarized in Figure 1. Group A patients underwent repeated imaging for 90 minutes (single low dose CT followed by 7 acquisitions from vertex to upper thighs, 1 minute per bed step). Two further acquisitions were performed at 2 and 3 hours post injection (each with ultra-low dose CT, 1.5 and 2 minutes per bed step, respectively).

In Group B, PET/CT imaging was performed at 15, 60 and 120 minutes from vertex to upper thighs with ultra-low dose CT at 15 and 120 minutes, and low-dose CT at 60 minutes. Patients were asked to void their bladder before the 60 and 120 minutes images.

Plasma sampling. In Cohort A, venous blood samples (5 ml) were taken at 5, 10, 20, 40 60, 90, 120 and 180 minutes. Each was sampled (2 mL) using a pipette for ^{68}Ga counting (Biodex 950 Well Counter, Biodex Medical Systems, New York, USA).

Safety monitoring. All patients were followed for adverse events from the time of ^{68}Ga-THP-PSMA administration for 24 hours. Before injection, baseline electrocardiogram, haematology, biochemistry assay and clotting were measured. Vital signs (blood pressure, heart rate, temperature and oxygen saturation) were recorded every 15 minutes for 3 hours. Blood tests were repeated at 24 hours and the patient was reviewed by a clinician, in person or by telephone, to document any adverse events (AE). Any AE was assessed for severity according to the Common Terminology Criteria for Adverse Events (CTCAE) v4.0 manual.

Statistical considerations. The number of patients was a pragmatic sample size for a safety study. Data analysis was descriptive. In Cohort B, uptake of ^{68}Ga-THP-PSMA and ^{68}Ga-HBED-PSMA was compared using a paired t-test.

RESULTS

Fourteen patients were screened and all proceeded to study participation and completion. No adverse events attributed to ^{68}Ga-THP-PSMA administration were recorded in any of the patients.

Group A. Baseline cohort characteristics and results are summarized in Table 1. Six of eight patients had increased uptake in the prostate above background. SUVmax of dominant prostate abnormality was 5.1 (range 2.4 – 9.2) at 120 minutes. The earlier time-point acquisitions acquired for purposes of dosimetry calculation were not suitable for determination of prostate uptake, owing to single CT acquisition at 0 minutes and subsequent bladder filling resulting in attenuation correction artefact in the surrounding pelvis.

Physiologic uptake was seen in lacrimal glands, salivary glands, liver, spleen and duodenum. The intensity of uptake in these organs was low (see Table 2). High kidney, ureteric and bladder activity was seen owing to renal excretion.

The mean effective dose was 2.07E-02 mSv/MBq (see Table 3). As patients were asked not to void during the imaging period and bladder accumulation occurs, this represents an overestimate to routine clinical practice. Plasma clearance was measured in 7 patients; technical difficulties obviated measurement in one. Findings were consistent with all patients demonstrating 2-phase exponential clearance with a mean of 5.9 minute (range 2.6 – 8.4) first phase half-time followed by 91.1 minute (range 67.2 – 118.5) second phase clearance (see Figure 2).

All patients proceeded to prostatectomy. 3+ intensity PSMA immunohistochemistry staining was seen in 6 THP-PSMA positive scans, and 1+ / 2+ staining in the 2 THP-PSMA negative scans (Figure 3). Seven of eight underwent pelvic nodal dissection. Pathologic pelvic nodal involvement was identified in 2 patients without corresponding ^{68}Ga-THP-PSMA abnormality, although these were both <1 mm in size, well below imaging resolution. One patient had focal intense uptake in sub-cm pelvic nodes without pathologic abnormality. Follow-up at 6 months confirmed accuracy of initial ^{68}Ga-THP-PSMA PET with

rising PSA, ^{68}Ga-HBED-PSMA11 and contrast-enhanced CT confirming nodal progression (see Figure 4).

Group B

^{68}Ga-THP-PSMA with imaging at 60 minutes had lower background physiologic uptake in salivary glands, liver and spleen compared to ^{68}Ga-HBED-PSMA-11 (parotid: mean SUVmax 3.6 compared to 19.2, $p<0.001$; liver: 2.7 compared to 6.3, $p<0.001$; spleen 2.7 compared to 10.5, $p<0.001$). In all 6 patients, metastatic lesions were identified on both HBED- and THP-PSMA (Table 3, Figure 5-6). For lesional analysis, a total of 22 malignant lesions were measured (15 lymph node, 6 bone. 1 prostate). On correlative CT, 13 of 15 nodes were less than 1cm in short axis diameter; mean of 9.9mm long axis (range 5-17) and 7.2mm short axis (range 3-15). In 5 of 6 patients there was concordance in the number of metastases identified on ^{68}Ga-HBED-PSMA-11 and ^{68}Ga-THP-PSMA. On a lesional analysis, tumor-to-liver contrast ratio was not different between the two radiotracers with a mean 5.4 for ^{68}Ga-HBED-PSMA compared to 4.7 for ^{68}Ga-THP-PSMA ($p=0.15$). Absolute SUVmax was significantly higher on ^{68}Ga-HBED-PSMA with a mean 30.3 compared to 10.7 ($p<0.01$). In one patient, 4 abnormal foci were identified on ^{68}Ga-HBED-PSMA PET compared to 2 on the ^{68}Ga-THP-PSMA; the two lesions only identified on HBED-PSMA were in the prostate and a 3 mm internal iliac node. No further follow-up was available to further validate the ground truth. There was no management change in this patient due to the difference in the two scans.

DISCUSSION

In this study, we provide the first-in-man validation of ^{68}Ga-THP-PSMA, the first clinical tracer to utilize the novel THP chelator, enabling labelling in less than five minutes at room temperature. The DOTA chelator, incorporated in the widely used DOTATATE, requires pH adjustment, heating followed by cooling, and sometimes a purification step, necessitating either significant manual handling or use of an

automated synthesis device. This increases expense and may not be compliant with Good Manufacturing Practice (GMP). Moreover, cartridge-based automated synthesis typically takes more than 30 minutes, which is suboptimal for a short-life radionuclide such as ^{68}Ga. HBED chelation can be performed at room temperature but this produces a mixture of *cis/trans* geometric isomers (*5*). Recently, favourable labelling properties for the DATA chelator for labelling DATATOC in a kit formulation have been described(*15*). THP-PSMA has been developed as a single step kit-formulated GMP radiopharmaceutical, requiring only addition of unprocessed, unfractioned generator eluate to a single vial. Depending on the generator used, however, additional simple steps may be required. The one step kit can also be used with generators with <0.001% ^{68}Ge breakthrough and appropriate elution volume, such as the GalliaPharm Generator from Eckert and Ziegler (7).

^{68}Ga-THP-PSMA imparts low absorbed doses, with effective doses similar to those published for ^{68}Ga-HBED-PSMA-11 (*16*). In this study a mean effective dose of 4.9 mSv was observed. Recently published ^{68}Ga-PSMA-11 guidelines, however, recommend 1.8 – 2.2 MBq per kilogram bodyweight (*17*), which would result in 2.4 - 2.9 mSv exposure from the PET component when using ^{68}Ga-THP-PSMA. This is significantly lower than other imaging tests typically used for imaging prostate cancer such as ^{18}F-fluorocholine, ^{18}F-fluorodeoxyglucose, bone scintigraphy or dedicated contrast-enhanced CT.

^{68}Ga-THP-PSMA had significantly lower background physiologic uptake compared to ^{68}Ga-HBED-PSMA-11. Specifically, salivary and tear glands (lacrimal, parotid and submandibular), liver and small bowel uptake was significantly lower. The mechanism of uptake in salivary glands remains uncertain perhaps reflecting non-specific radiotracer excretion, PSMA expression in these tissues or an unknown target with similar recognition properties. As there is no known expression of PSMA in the liver or spleen, the differential uptake possibly reflects particulate radioactivity with ^{68}Ga known to generate colloidal ^{68}Ga between pH 3 and 7. The observed tumor-to-liver contrast with ^{68}Ga-THP-PSMA-11 was similar to ^{68}Ga-HBED-PSMA despite absolute uptake being lower. Sub-centimetre abnormalities were frequently visualized, reinforcing the high sensitivity of PSMA PET imaging.

CONCLUSION

^{68}Ga-THP-PSMA can be manufactured quickly using a generator and cold-kit vial analogous to procedures widely used for ^{99m}Tc radiotracer production. ^{68}Ga-THP-PSMA can be safely administered, with absorbed radiation doses less than 3mSv from the PET component. Focal uptake in the prostate correlates with histopathologic PSMA expression. Background physiologic uptake was significantly lower with ^{68}Ga-THP-PSMA compared to ^{68}Ga-HBED-PSMA. Sub-centimetre metastases were frequently identified with both radiotracers with similar tumor-to-liver contrast despite lower absolute uptake with ^{68}Ga-THP-PSMA.

DISCLOSURE

The study received funding from Theragnostics Limited (UK). PJB and GEM are named inventors on patents whose claims encompass the THP chelator. JDY is funded by the King's College London and Imperial College London EPSRC Centre for Doctoral Training in Medical Imaging (EP/L015226/1). We acknowledge support from KCL and UCL Comprehensive Cancer Imaging Centre funded by CRUK and EPSRC in association with the MRC and DoH (England), and the NIRH Biomedical Research Centre awarded to Guy's and St Thomas' NHS Foundation Trust in partnership with King's College London and King's College Hospital NHS Foundation Trust.

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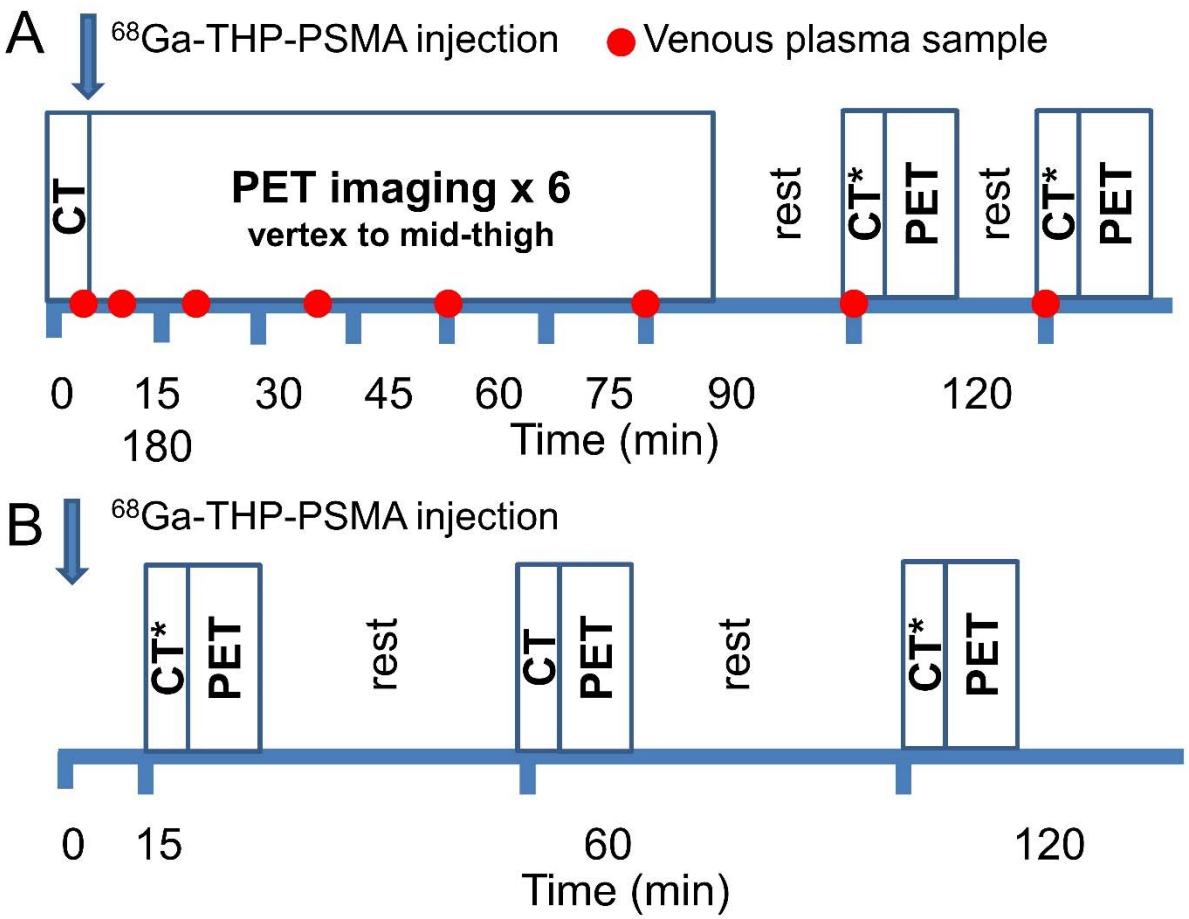

FIGURE 1. Summary of PET/CT acquisition protocol for (A) Cohort A and (B) Cohort B. *ultra-low dose CT for attenuation correction.

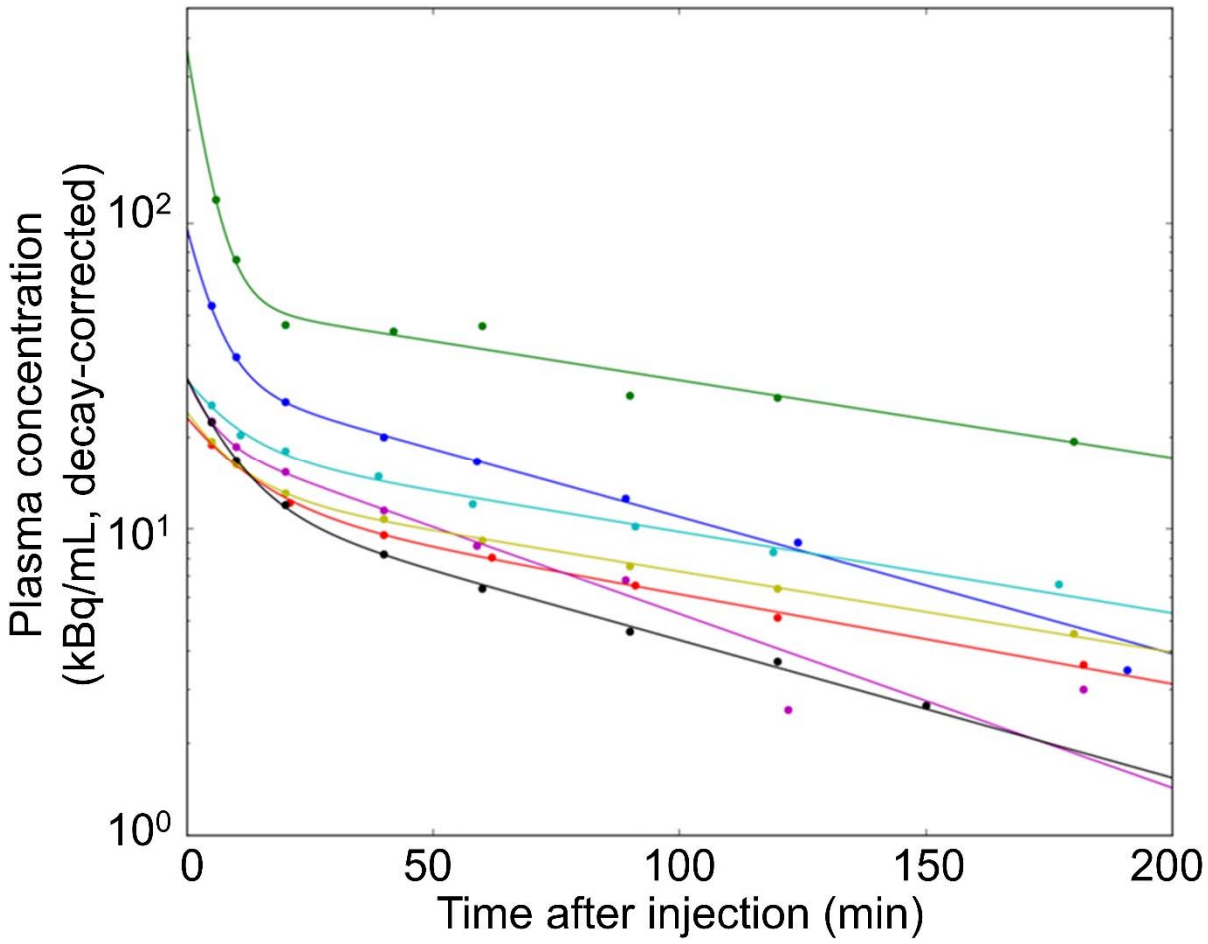

FIGURE 2. Plasma clearance of ^{68}Ga-THP-PSMA with each patient represented in a different color.

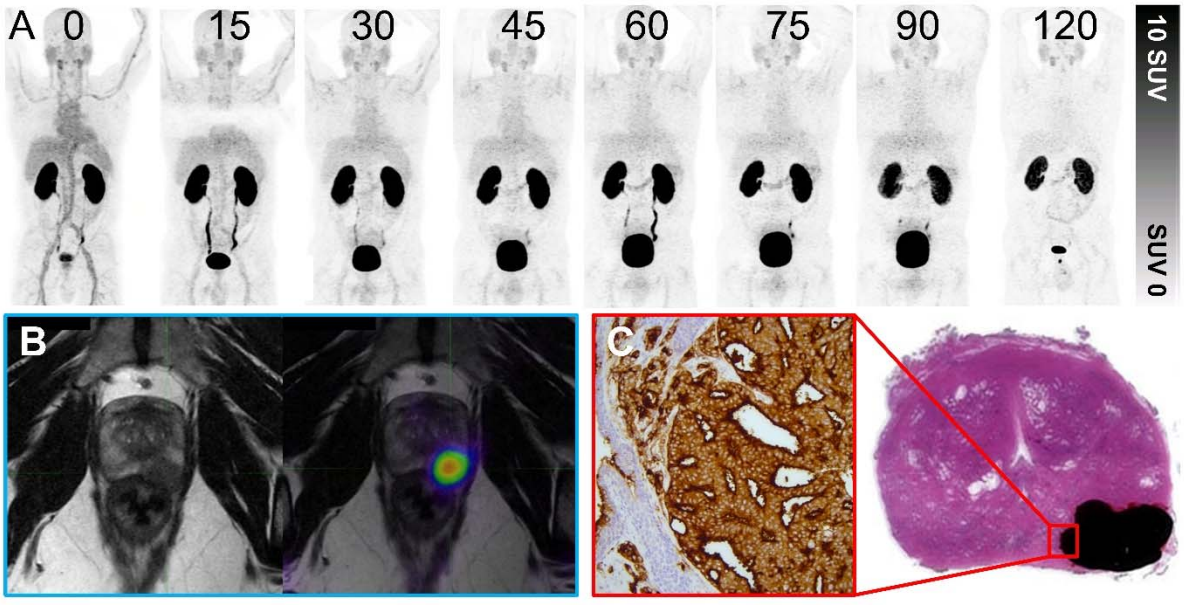

FIGURE 3. Patient with Gleason 4 + 4 = 8 prostate adenocarcinoma, PIRADS-5 on prior multi-parametric MRI. (A) ^{68}Ga-THP-PSMA PET multi-time point PET images from 0 to 120 minutes demonstrating rapid blood pool clearance and low background activity. (B) 120 minute PET image after voiding fused to MRI T2 sequence demonstrating focal uptake in the left posterior mid-zone lesion. (C) Histopathologic correlation following prostatectomy with area of adenocarcinoma shaded in black, and zoomed-in area demonstrating 3+ staining with PSMA immunohistochemistry.

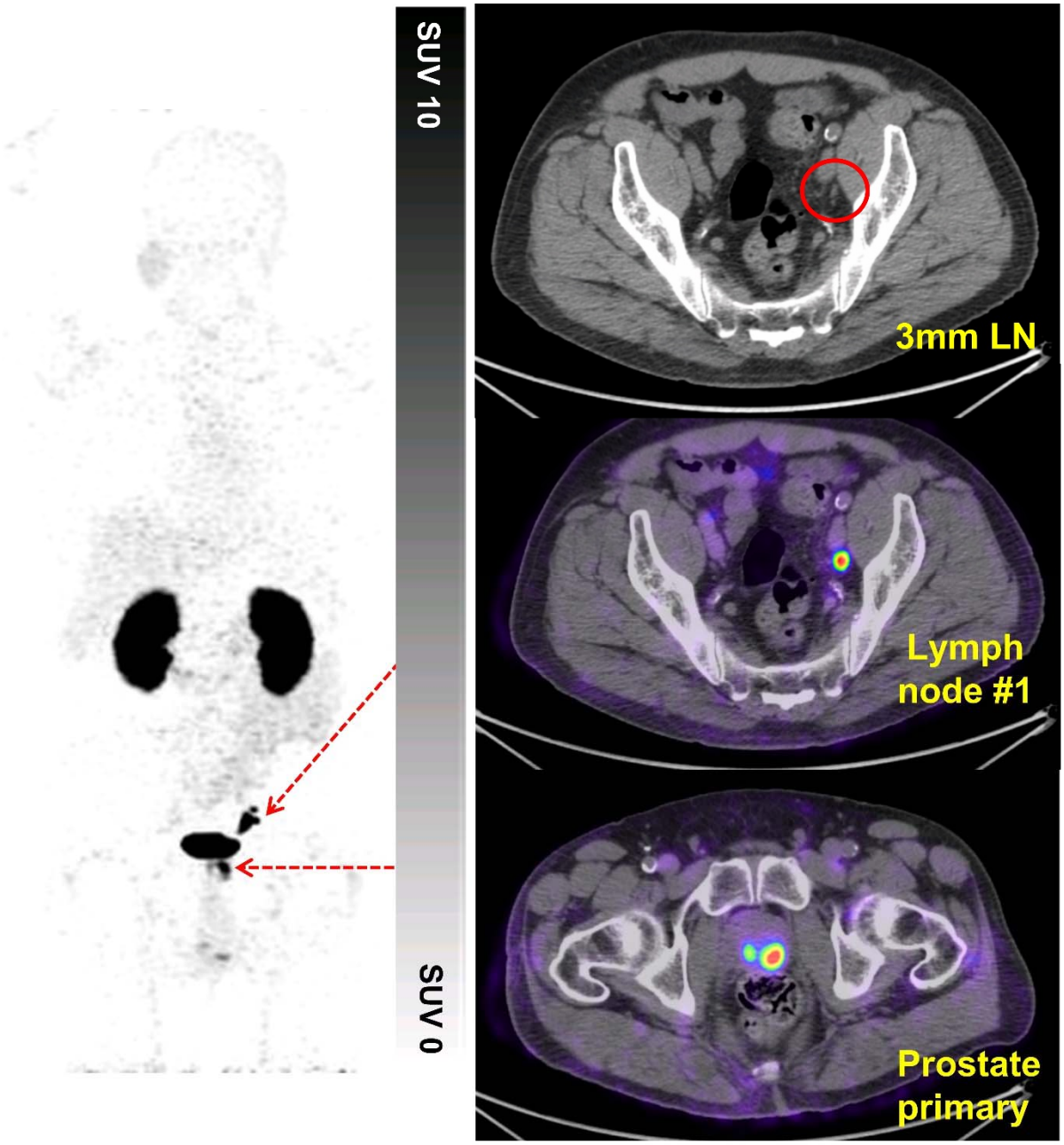

FIGURE 4. Patient with Gleason 4 + 5 = 9 prostate adenocarcinoma with high uptake in the primary prostate tumor and focal uptake in several left external iliac lymph nodes less than 5 mm in size (120 minute image shown). The patient proceeded to prostatectomy and pelvic nodal clearance. Pathologic staging demonstrated no nodal involvement (pN0 stage). Follow-up demonstrated persistently elevated and rising PSA, with repeat imaging at 6 months confirming presence and progression of baseline ^{68}Ga-THP-PSMA PET/CT findings. The pN0 staging is therefore interpreted as false negative due to sampling error.

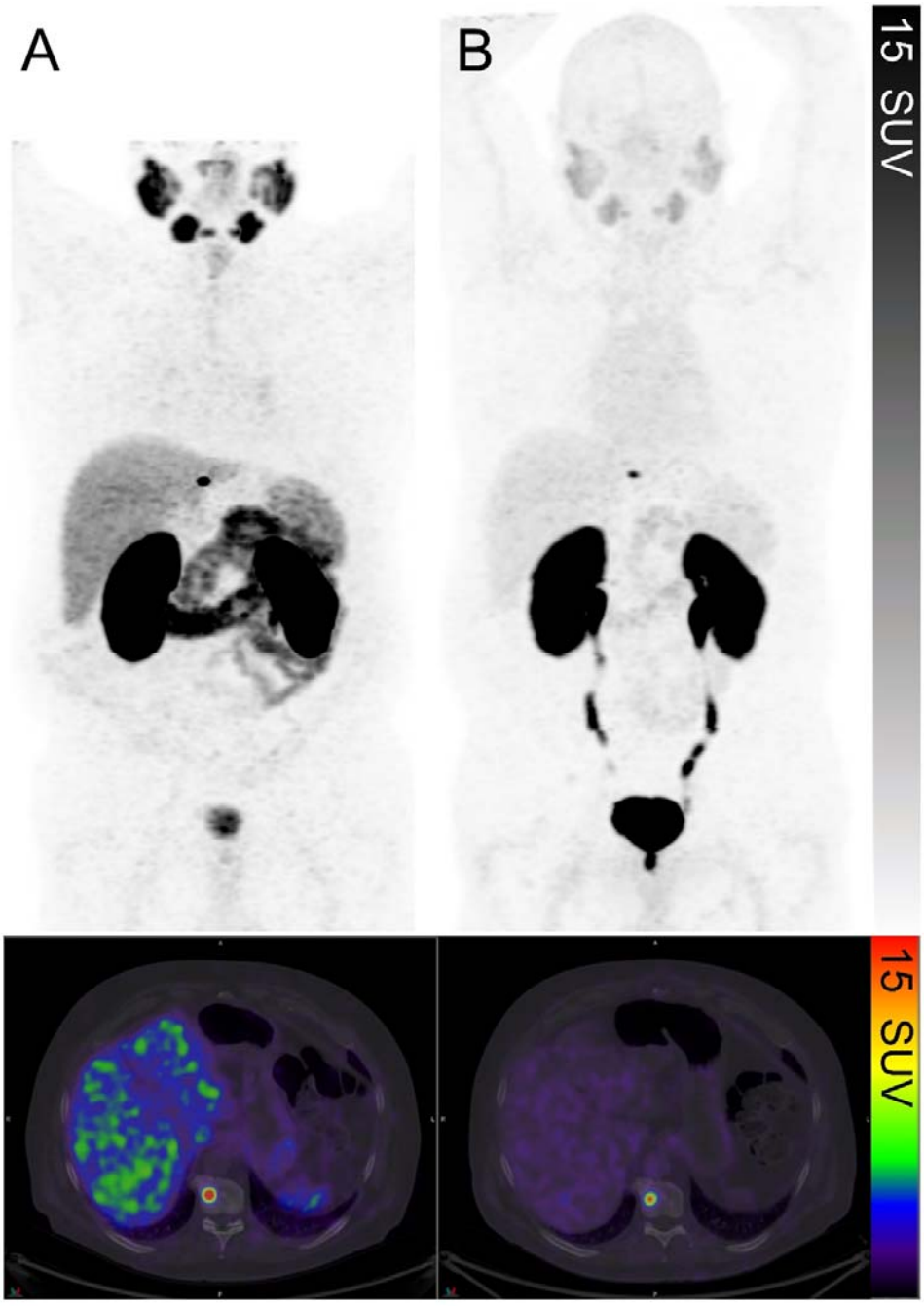

FIGURE 5. Comparison of (A) ^{68}Ga-HBED-PSMA PET/CT (177MBq, 56 minute uptake period) to (B) ^{68}Ga-THP-PSMA PET/CT (232 MBq, 60 minute uptake period) in the same patient. Solitary focal intense abnormality in the T10 vertebral body is well-visualised on both. Note the lower background physiologic uptake on the THP-PSMA study.

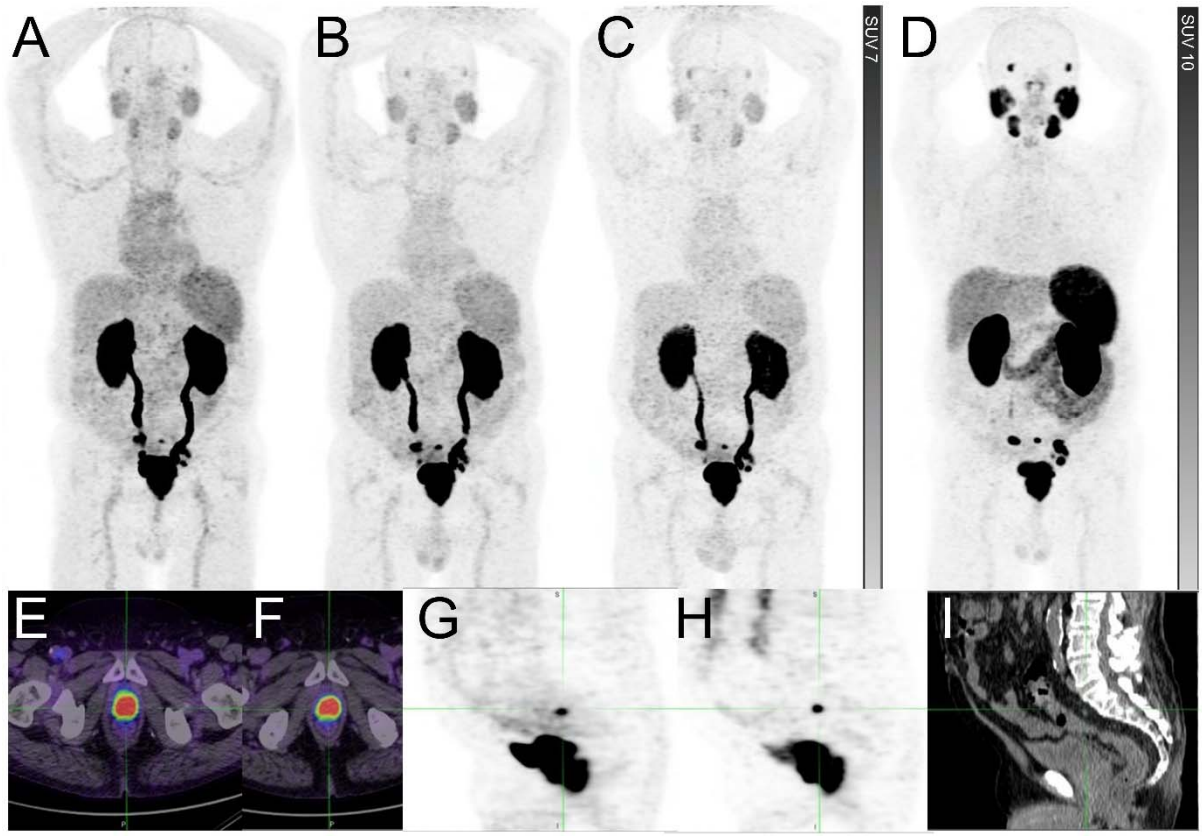

FIGURE 6. Primary staging of patient with Gleason 5+4=9 prostate adenocarcinoma and normal conventional imaging. ^{68}Ga-THP-PSMA MIP images at (A) 15 min, (B) 60 min and (C) 120 min, and (D) ^{68}Ga-HBED-PSMA 60 min MIP image comparison. Focal intense uptake is seen in the primary prostate tumor on (E) ^{68}Ga-THP-PSMA and (F) ^{68}Ga-HBED-PSMA PET/CT. Multiple sub-centimetre pelvic nodal metastases were visualized. (G) ^{68}Ga-THP-PSMA and (F) ^{68}Ga-PSMA-PSMA demonstrate focal uptake corresponding to (I) <5mm pre-sacral node on CT.

TABLE 1: Cohort A: baseline characteristics and results

		pre-op				THP-PSMA		post-op		
No	Age	Gleason Score (GS)	PSA	Clinical T	MRI	prostate +/- SUVmax	nodal +/- SUVmax	GS	Pathologic stage	PSMA IHC
1	71	3 + 5 = 8	10.3	T3	PIRADS4	+ 5.6	-	4 +3 = 7	T3b N1*	3+
2	64	3 + 4 = 7	6.2	T2a/b	PIRADS5	-	-	3 + 4 = 7	T2c Nx	2+
3	55	4 + 3 = 7	9.6	T1c	PIRADS2	+ 2.5	-	4 +3 = 7	T3a N0	3+
4	65	3 + 4 = 7	5.4	T1	PIRADS4	+ 4.0	-	3 + 4 = 7	T2c Nx	3+
5	71	3 + 4 = 7	3.2	T2b	PIRADS5	+ 2.4	-	4 +3 = 7	T3a Nx	3+
6	54	5 + 5 = 10	9	T2b	PIRADS5	-	-	4 + 5 = 9	T3a N1†	1+
7	46	4 + 4 = 8	10.6	T3a	PIRADS5	+ 7.1	-	4 + 5 = 9	T3a N0	3+
8	58	4 + 4 = 8	8	T2c	not performed	+ 9.2	+ 18.4‡	4 + 5 = 9	T3b N0	3+

* 0.6mm in 1 LN. † 0.1mm in 1 LN. ‡confirmed as true positive on follow-up with sampling error resulting in pathologic N0 staging

Abbreviations. PIRADS: Prostate imaging reporting and data system v2. LN: lymph node

TABLE 2: Physiologic uptake (SUVmax) of ^{68}Ga-THP-PSMA compared to ^{68}Ga-HBED-PSMA for Cohort B

		^{68}Ga-THP-PSMA				^{68}Ga-HBED-PSMA			
	Pt	Parotid	Liver	Spleen	Blood pool	Parotid	Liver	Spleen	Blood pool
Group A	1	3.0	2.7	2.6	2.6	n/a			
	2	3.4	2.3	2.1	2.6				
	3	2.4	2.7	1.8	2.3				
	4	4.1	3.3	2.7	3.0				
	5	2.9	2.4	1.7	3.1				
	6	3.0	2.9	2.6	2.6				
	7	4.4	3.0	3.9	4.5				
	8	3.5	3.0	2.4	2.5				
Group B	9	5.0	2.6	2.7	1.8	20.0	8.2	9.3	1.8
	10	3.5	4.8	4.8	1.6	18.3	7.2	9.8	1.2
	11	4.4	2.1	2.1	1.7	16.7	4.8	8.8	1.4
	12	3.9	2.2	3.2	1.9	18.7	6.3	15.9	1.7
	13	2.9	2.4	3.2	2.3	14.2	6.2	14.5	1.9
	14	4.2	1.9	1.6	1.9	27.0	5.1	4.4	0.6
	Mean	3.6	2.7	2.7	2.5	19.2	6.3	10.5	1.4

TABLE 3: Absorbed organ and whole body doses of ^{68}Ga-THP-PSMA

RGAN	PT 1	PT 2	PT 3	PT 4	PT 5	PT 6	PT 7	PT 8	MEAN	MEDIAN
	Pt 1	Pt 2	Pt 3	Pt 4	Pt 5	Pt 6	Pt 7	Pt 8	**Mean**	**Median**
Adrenals	7.00E-03	6.01E-03	7.36E-03	5.02E-03	4.85E-03	5.88E-03	5.57E-03	5.33E-03	**5.88E-03**	**5.72E-03**
Brain	1.14E-03	1.31E-03	2.15E-03	1.15E-03	1.20E-03	1.29E-03	9.35E-04	1.47E-03	**1.33E-03**	**1.24E-03**
Gallbladder	9.89E-03	7.54E-03	1.11E-02	7.24E-03	9.00E-03	8.42E-03	1.33E-02	7.03E-03	**9.20E-03**	**8.71E-03**
Lower large intestine	1.92E-02	8.42E-03	1.32E-02	6.63E-03	1.00E-02	9.39E-03	8.78E-03	1.01E-02	**1.07E-02**	**9.71E-03**
Small intestine	1.71E-02	1.44E-02	1.52E-02	2.25E-02	1.53E-02	1.53E-02	1.25E-02	1.36E-02	**1.57E-02**	**1.53E-02**
Stomach	9.89E-03	7.85E-03	1.17E-02	7.49E-03	7.84E-03	8.03E-03	7.70E-03	6.86E-03	**8.42E-03**	**7.84E-03**
Upper large intestine	8.04E-03	7.46E-03	9.96E-03	6.34E-03	7.36E-03	7.24E-03	6.48E-03	7.16E-03	**7.50E-03**	**7.30E-03**
Heart	1.04E-02	1.55E-02	2.80E-02	1.42E-02	1.55E-02	1.62E-02	1.50E-02	1.18E-02	**1.58E-02**	**1.53E-02**
Kidneys	7.26E-02	6.05E-02	9.07E-02	8.89E-02	9.13E-02	9.91E-02	5.87E-02	7.60E-02	**7.97E-02**	**8.24E-02**
Liver	1.83E-02	1.08E-02	1.79E-02	1.10E-02	1.52E-02	1.11E-02	1.05E-02	1.02E-02	**1.31E-02**	**1.10E-02**
Lungs	1.69E-02	1.18E-02	1.85E-02	1.53E-02	1.53E-02	1.56E-02	1.33E-02	1.09E-02	**1.47E-02**	**1.53E-02**
Muscle	5.15E-03	4.23E-03	5.69E-03	3.44E-03	4.50E-03	4.43E-03	4.25E-03	4.04E-03	**4.47E-03**	**4.34E-03**
Pancreas	9.41E-03	7.19E-03	1.46E-02	9.92E-03	1.30E-02	8.82E-03	8.26E-03	8.60E-03	**9.98E-03**	**9.11E-03**
Red marrow	9.86E-03	8.20E-03	1.05E-02	6.87E-03	7.58E-03	7.89E-03	7.57E-03	7.69E-03	**8.27E-03**	**7.79E-03**
Osteogenic cells	1.24E-02	1.07E-02	1.35E-02	8.52E-03	8.61E-03	1.00E-02	1.03E-02	9.52E-03	**1.04E-02**	**1.02E-02**
Skin	4.56E-03	4.13E-03	4.55E-03	2.93E-03	2.79E-03	3.65E-03	3.70E-03	3.56E-03	**3.73E-03**	**3.68E-03**
Spleen	1.56E-02	1.14E-02	1.63E-02	1.47E-02	1.38E-02	1.32E-02	1.05E-02	9.65E-03	**1.31E-02**	**1.35E-02**
Testes	1.12E-02	9.65E-03	1.08E-02	1.05E-02	1.28E-02	1.12E-02	1.03E-02	9.91E-03	**1.08E-02**	**1.06E-02**
Thymus	5.48E-03	4.91E-03	5.73E-03	3.56E-03	3.10E-03	4.31E-03	4.48E-03	4.02E-03	**4.45E-03**	**4.39E-03**
Thyroid	8.34E-03	8.07E-03	1.12E-02	6.05E-03	6.93E-03	6.27E-03	7.57E-03	7.38E-03	**7.72E-03**	**7.47E-03**
Urinary bladder	1.80E-01	1.78E-01	1.22E-01	1.51E-01	4.50E-01	2.67E-01	1.36E-01	3.36E-01	**2.27E-01**	**1.79E-01**
Remainder	7.97E-03	6.80E-03	8.33E-03	5.64E-03	6.28E-03	6.80E-03	6.35E-03	6.38E-03	**6.82E-03**	**6.59E-03**
Effective dose (mSv/MBq)	2.05E-02	1.71E-02	1.75E-02	1.60E-02	3.21E-02	2.25E-02	1.52E-02	2.49E-02	**2.07E-02**	**1.90E-02**

TABLE 4: Number of PSMA-avid metastatic lesions identified

	Gleason Score	PSA (ug/L)	Prostate bed (T)		Regional nodes (N)		Non-regional nodes (M1a)		Bone metastases (M1b)		Visceral metastases (M1c)	
Pt No			THP	HBED	THP	HBED	THP	HBED	THP	HBED	THP	HBED
9	3+4=7	2.1	0	0	0	0	0	0	1	1	0	0
10	3+3=6	14.9	0	0	2	2	1	1	0	0	0	0
11	4+4=8	50.7	0	0	0	0	10	10	>20	>20	0	0
12	4+4=8	3.2	0	0	1	1	0	0	0	0	0	0
13	5+4=9	3	1	1	6	6	0	0	0	0	0	0
14	3+3=6	8	0	1	2	3	0	0	0	0	0	0