

We are IntechOpen, the world's leading publisher of Open Access books Built by scientists, for scientists

4,800

Open access books available

122,000

International authors and editors

135M

Downloads

Our authors are among the

154

Countries delivered to

TOP 1%

most cited scientists

12.2%

Contributors from top 500 universities



WEB OF SCIENCE™

Selection of our books indexed in the Book Citation Index
in Web of Science™ Core Collection (BKCI)

Interested in publishing with us?
Contact book.department@intechopen.com

Numbers displayed above are based on latest data collected.

For more information visit www.intechopen.com



Imagen Thecniques in Renal-Cell Carcinoma

L. León, M. Ramos, M. Lázaro, S. Vázquez,
M. C. Areses, O. Fernandez, U. Anido, J. Afonso and
L. A. Aparicio

Additional information is available at the end of the chapter

<http://dx.doi.org/10.5772/54190>

1. Introduction

Over 64000 new renal-cell carcinomas (RCC) are annually detected in the United States, and 13000 people will die from the disease. Most RCC are discovered incidentally on medical imaging and a great percentage of them may be treated by surgery, but one third of patients will present either with locally advanced tumor or with metastases[1]. In addition, another third of patients may develop metastatic disease after initial treatment.

In cancer patients imaging techniques are essential in three aspects. First, at the time of diagnosis and the extension study. Ultrasonography, computed tomography (CT) and magnetic resonance imaging (MRI) are currently available to evaluate renal masses.

Second, since most RCC are now early-stage disease suitable for surgery with curative intent, the patient is candidate to follow-up during years. Early detection of recurrence is vital, because single-organ disease may be trated by metastasectomy. Again, CT and MRI are essential in this setting. Also, these imaging modalities are useful to follow-up people with increased susceptibility for RCC, since we have tools to identify at least a subset of these patients.

And third, imaging techniques are fundamental to evaluate the response to treatment. RECIST criteria, published in 2000 and revised in 2009, has become the most widely accepted guideline for evaluate response [2]. Although RECIST criteria have been proved as a useful tool to asses response in solid tumors, some limitations have been noted. One of these limitations are observed in patients treated with specific targeted therapies [3].

Traditionally, RCC have been remarkably resistant to chemo- and radiotherapy. Over the last decade, there has been an increasing knowledge about pathophysiological processes in RCC

including oncology pathways due to a specific driver mutations: silencing von-Hippel Lindau gene, angiogenesis alterations, evasion of apoptosis or sustained angiogenesis. These features have enabled the emergence of a wide spectrum of novel oncology drugs that are designed to target and interfere with specific aberrant biological pathways. Therefore, morphological criteria may not provide meaningful data in this setting and the incorporation of new imaging techniques (MRI diffusion, perfusion CT, PET scan, etc....) in the diagnosis of extension and assessment of efficacy of this drugs may provide unique physiological data that can be correlated with histopathological changes and may provide functional information.

In this chapter we will review the main techniques of radiological diagnosis and staging, the role of new imaging techniques and we will also discuss the validity of the classical criteria of interpretation of response.

2. Common techniques of characterization of renal lesions

2.1. Ultrasound

Ultrasound (US) is one of the most common techniques used in the initial evaluation of renal lesions. It is a low cost and easy access technique and it also allows avoiding the exposure to ionizing radiation and the use of contrast (Figure 1).



Figure 1. Solid mass in right upper kidney (5 x 3.7 cm).

Although it is an observer-dependent technique, it allows monitoring renal lesions growth and distinguish between cystic and solid lesions. Ultrasonographic features of cystic lesions that allow distinction with malignant lesions or abscesses are:

1. Round morphology, smooth and well-defined walls, separating it from the surrounding parenchyma.
2. There is a strong posterior wall indicating good transmission through the cyst and enhanced transmission beyond the cyst.
3. Absence of internal echoes. The presence of thickened internal septa, calcifications, or mural vascularity indicate malignancy.

One of its limitations is the evaluation and characterization of small lesions. Jamis found that CT detected more renal lesions, especially if they were noncontour deforming. 5% of 2 cm lesions were not detected with CT, an 30% were missed in US. Of lesions under 1 cm, 24% were not detected in TC versus 80% with US [4]. Moreover, given the variability in the echogenicity of malignant kidney, it can be difficult, in the case of isoechoic images, the identification and distinction of these lesions.

In recent years it has become increasingly important the use of contrast-enhanced ultrasound (CEUS). Current CEUS consist of intravenously injected microbubbles that increase the number of reflectors in the vascular space. It has different utilities. It is useful in the differential diagnosis of solid and cystic lesions so as to characterize cystic lesions in benign or malignant [5]. Solid lesions show early arterial enhancement, normally lower than surrounding parenchyma. The delayed enhancement varies and after an arterial phase lesions are isoechoic relative to parenchyma. Often because of intralésional necrosis, there are intralésional areas without contrast enhancement.

It is of particular interest the characterization of complex cystic lesions. Some studies have reported a sensitivity and specificity similar to CT [6] [7]. It can be considered a valid alternative to CT and MRI in monitoring these lesions that need prolonged follow [8]. It may also be useful in detecting small renal masses, improving the accuracy of simple ultrasound, since it allows to observe changes in the thickness of the cortical pyramidal space, not visible in simple US.

2.2. CT scan

Computed tomography (CT) is the modality of choice for the diagnosis and study of extension of renal carcinoma, with a sensitivity greater than 95% (Figure 2) [9]. In addition, the development of multidetector CT has allowed an increase in the rate of detection and diagnosis in early stages [10].

For the evaluation of suspicious lesions, it is advisable to have a specific protocol. This should include a scan without contrast to determine the presence of calcification or fatty tissue within the tumor, and will serve as baseline study to study if these lesions enhance after contrast administration.

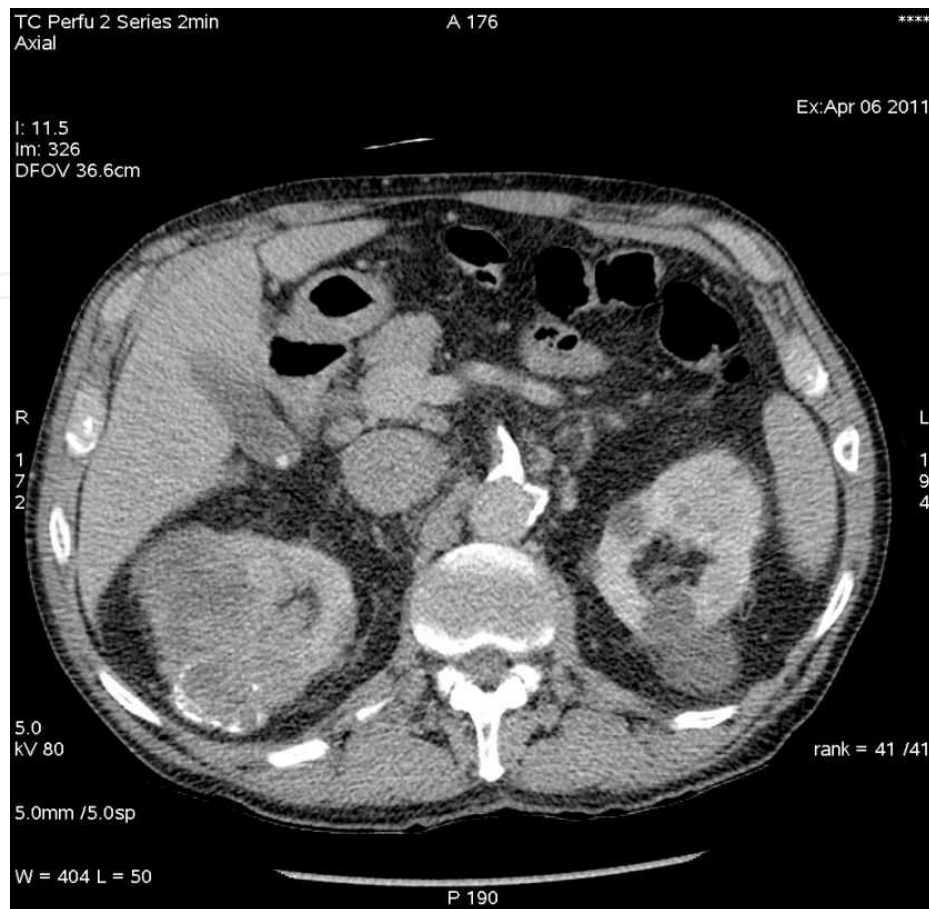


Figure 2. CT scan, right renal cell carcinoma.

The three perfusion renal phases defined in CT diagnosis are: corticomedullary phase, nephrographic phase, and renal elimination phase (or excretory phase) [11].

The images in the corticomedullary phase help to identify the lesion and its vascular supply, being optimal for detecting or excluding tumor invasion of the renal veins [12]. The nephrographic and elimination phase help detecting renal masses, especially those of small size.

The appearance of renal carcinoma in CT varies depending on the size of the tumor vasculature, the extent of necrosis or intratumoral cystic changes. Enhancement of a renal lesion shows that it is hypervascular; this is the most important finding in the evaluation of renal masses, being a useful parameter in differentiating histological subtypes.

Different groups have shown as clear cell carcinoma has a higher contrast enhancement than other histological subtypes, especially papillary carcinomas [13]. Zhang et al show that 90% of the clear cell renal carcinoma are hypervascular and heterogeneous (with solid hypervascular foci and low attenuation foci by necrotic or cystic changes). Seventy five % of papillary carcinomas were hypovascular and 90% had an uniform pattern or peripheral uptake while chromophobe tumors often show a moderate and homogeneous enhancement [10].

Tumors less than 3 cm sometimes have a smooth contour, they are homogeneous and difficult to distinguish from some benign lesions. Renal cystic carcinomas usually have thickened walls and septa, sometimes with calcification. Three-dimensional CT is important in staging renal cell carcinoma, with the objective of identifying patients having a resectable tumor and to define the best therapeutic option. The value of CT is limited to the study of the perirenal fat. Various criteria have been used to describe the appearance of perirenal fat infiltration. Trabeculation of perirenal fat is not a reliable sign of tumor involvement, and is found in approximately 50% of patients with localized tumors T1 and T2. It can be caused by edema, vascular congestion, or prior inflammation [14]. The presence of a nodule uptake in perirenal fat, is considered the most specific finding of perirenal invasion, with high specificity (98%) but low sensitivity (46%) [15].

Helical CT has also been shown to have high accuracy in the diagnosis of renal vein invasion with a negative predictive value of 97% and a positive predictive value of 92% (Figure 3) [16].

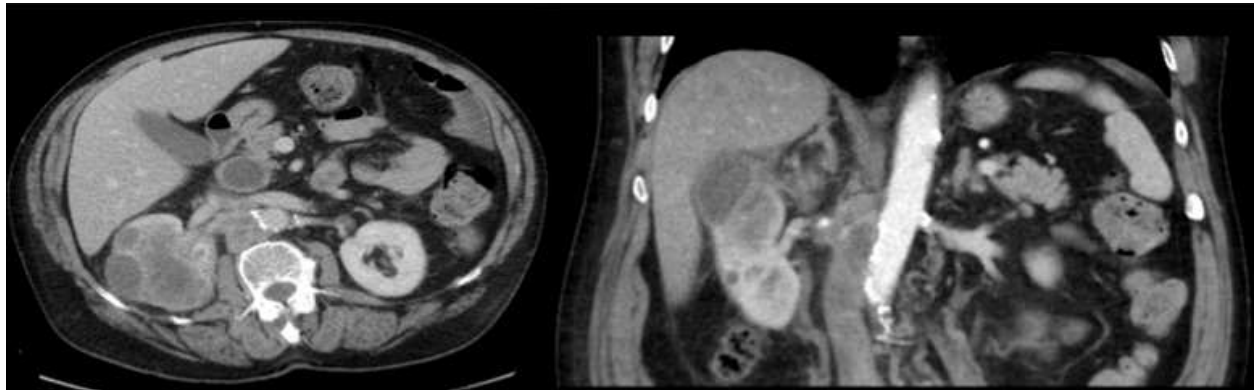


Figure 3. Renal cell carcinoma with thrombosis of the inferior vena cava.

The adrenal evaluation is important because if no abnormalities are detected on CT, adrenalectomy can be avoided. CT has a high negative predictive value in the detection of adrenal involvement by RCC. When the adrenal gland is enlarged, displaced or not displayed an adrenalectomy should be considered [17].

The study of lymph node is based primarily on its size. It is considered that a lymph node could be metastatic when its diameter is greater than 1 cm. However this approach has a limited specificity and sensitivity (between 3 and 43% in different studies) because the size increase may be due to inflammatory changes.

The nodal enhancement pattern helps differentiate between reactive and malignant lymphadenopathy. Metastatic lymph nodes can be enhanced after administration of contrast, especially if the primary tumor is highly vascularized.

Finally, given that CT plays an important role in detecting distant disease, it is necessary to conduct a study of the chest and abdomen in the staging of metastatic disease.

2.3. Magnetic resonance imaging

Magnetic resonance imaging (MRI) is useful when computed tomography cannot be performed, but it has not proved to be superior to CT in the detection or characterization of renal masses. The study should include T1 and T2 sequences and opposed-phase images to detect intratumoral fat. Dynamic study after paramagnetic contrast administration is essential.

Both CT and MRI have high reliability in delineating the extent of intratumoral thrombus, since it could change the surgical approach. However, MRI is more sensitive than CT to differentiate between tumoral and non-tumoral thrombus. The tumoral thrombus is heterogeneous or hyperintense on T2-weighted images, with marked enhancement on the postcontrast images, and, sometimes, it is seen the continuity with the renal tumor. The tumor thrombus is hypointense, not homogeneous and does not enhance after contrast administration [18].

Also, as discussed below, MRI can help us to distinguish between different histological subtypes of renal cell carcinoma (RCC), and between these ones and benign tumors such as oncocytomas and angiomyolipomas.

Clear cell RCC usually shows a signal intensity similar to that of the renal parenchyma on T1-weighted images and it's high intensity on T2-weighted images (Figure 4). Central necrosis is common, and it is typically seen as a homogeneous hypointense area in the center of the mass on T1-weighted images, and hyperintense, rarely hypointense, on T2-weighted images [19]. If intratumoral hemorrhage occurs, the appearance of this will depend on the degree of degradation of its components. A hypointense ring, or pseudocapsule, is sometimes seen on both T1 and T2-weighted images, and is due to compression of the adjacent renal parenchyma by the tumor growth. Breakage of this pseudocapsule correlates with advanced stage and higher nuclear grade [20]. This histological subtype tends to be hypervascular, with heterogeneous enhancement during the arterial phase. You can also appreciate renal vein thrombus in more aggressive and advanced tumors. They can also be predominantly cystic, with only a few areas of solid component [21].

The type I papillary RCC is characterized by a homogeneous hyposignal on T2-weighted images, with homogeneous low-level enhancement after contrast administration [22]. Sometimes they show necrosis and hemorrhage. Type II papillary RCC have a more complex appearance, with hemorrhage and necrosis. It is common to see a hemorrhagic cystic mass with enhancing papillary projections at the periphery. In both types is frequent the presence of a fibrous capsule [23].

Chromophobe RCC may show cystic changes within a solid mass. It is not common the presence of necrotic foci, even in large tumors. Its appearance on MR can be identical to those of clear cell RCC [23].

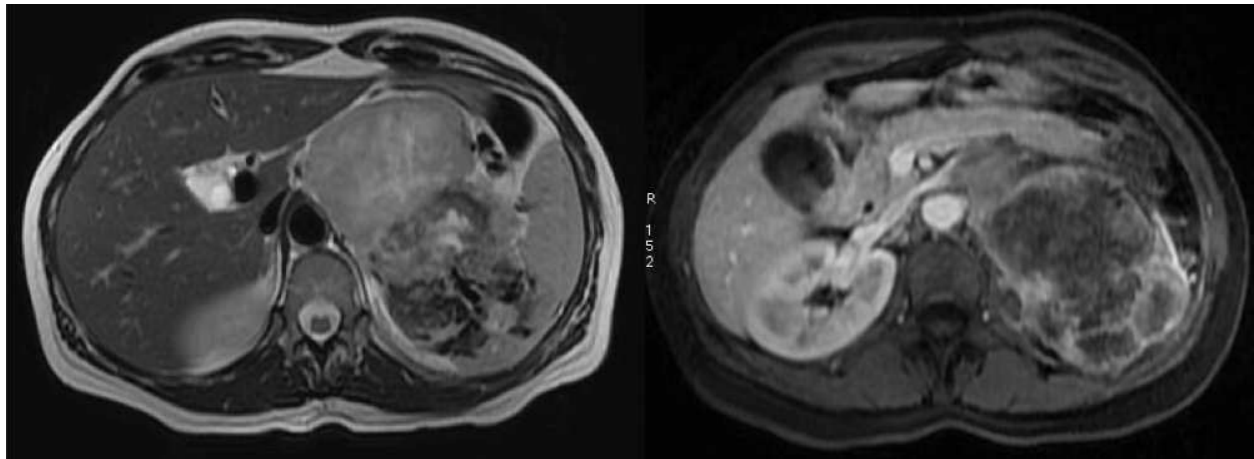


Figure 4. Clear cell RCC. Heterogeneous tumor with focal posterior bleeding. T2 (left side) and T1 sequence (right side) with fast gradient contrast, showing renal vein invasion. Courtesy Dr. Armesto-Pérez.

The MR Imaging appearance of oncocytomas is variable and nonspecific. They are typically spherical and well-defined masses with hypointense on T1-weighted images and hyperintense on T2-weighted images, in most cases. The central scar, when present, has a stellate appearance with low signal intensity on T1-weighted images and high signal intensity on T2-weighted images, and it may show delayed enhancement after contrast administration. Sometimes are surrounded by a well-defined hypointense capsule [24].

Angiomyolipoma with a predominant fatty component is isointense relative to fat on all MR Imaging sequences and its signal intensity is higher than that of the renal parenchyma on T1-weighted images. Fat-suppression sequences are also useful. Lipid-poor angiomyolipomas are difficult to distinguish from clear cell RCC with current imaging methods, so may occasionally be required histopathological evaluation to establish the correct diagnosis [21].

Diffusion-weighted imaging may be useful in differentiating between RCC and oncocytoma and in the characterization of the different histological subtypes of RCC. Angiomyolipoma, due to the presence of fat, can give false positives, but it is characterized through conventional sequences [25] [26].

The whole-body MRI, at present, is positioning itself as one of the techniques of choice for evaluation of bone marrow in patients with suspected bone metastases with a sensitivity / specificity (> 90%) higher than the radiology conventional CT and bone scans, and similar to PET-CT (Figure 5) [27].

Complement the study with diffusion-weighted imaging, besides allowing a faster interpretation and greater detection of subtle findings could add specificity to the study [28]. This is particularly relevant with the progressive increased use of new anti tumor drugs in which this technique may allow better assessment of tumor response [29].

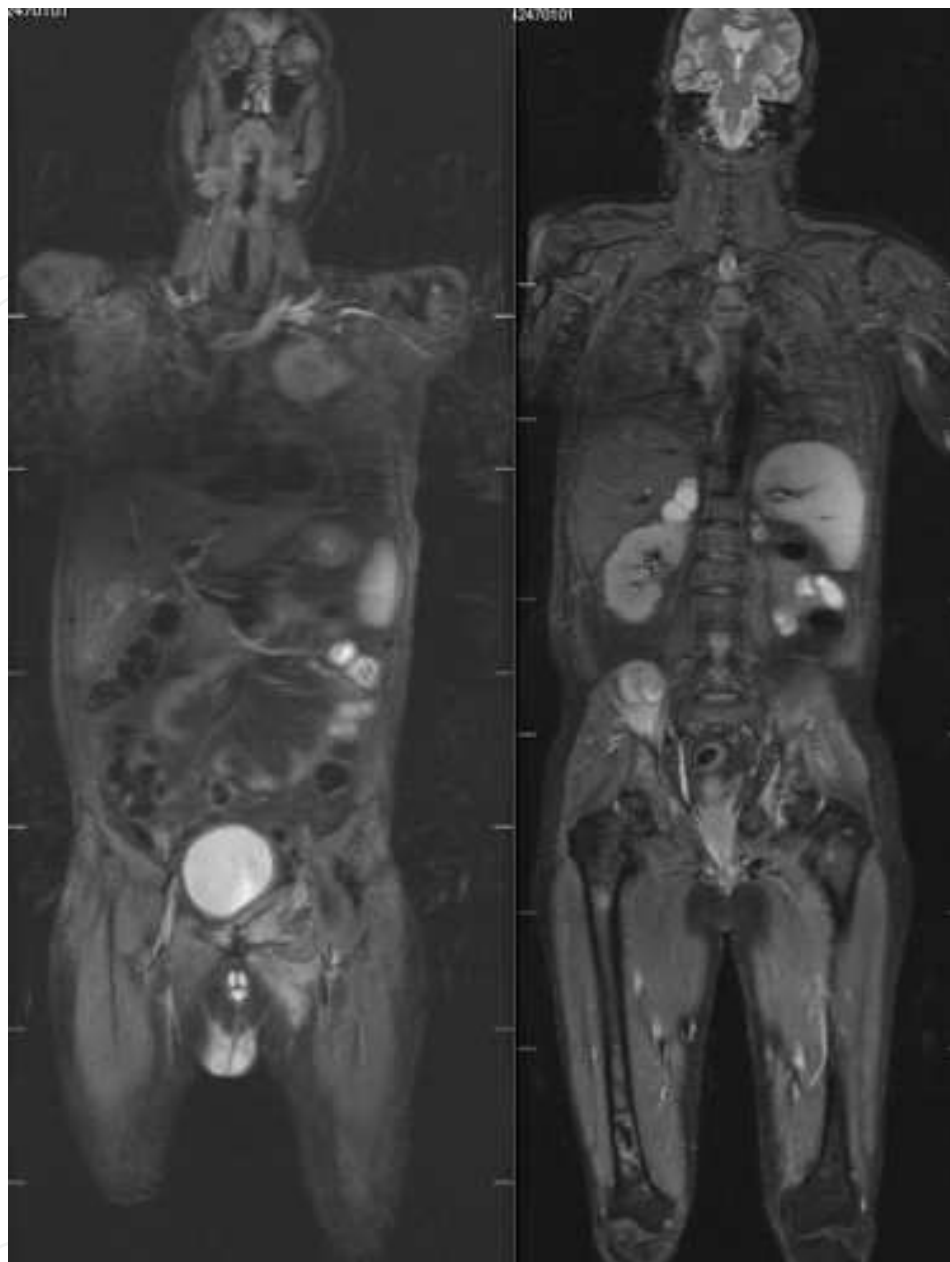


Figure 5. Whole body MRI. Bone, lung and liver metastases. Courtesy Dr. Armesto-Pérez.

2.4. Bone scan

Bone metastases in RCC is reported in 17-37% of patients and its early identification may have prognosis importance because its early intervention leads to significant reduction in patient morbidity. Bone scintigraphy is a very useful tool in diagnosis of bone metastases when those lesions have sufficient osteoblastic reaction (Figure 6). However, bone metastases in RCC usually appear as large expansive lytic lesions, most commonly in the axial skeleton and are poorly visualized in bone scintigraphy [30], showing variable uptake, with a sensitivity between 10-60% in the diagnosis of this metastases in preselected patients with RCC and high probabili-

ty of skeletal involvement with underestimation of the extension of the metastatic involvement, being clearly inferior to other techniques such magnetic resonance imaging or PET scan [31]. Because most bone metastases are symptomatic, most of authors recommend the use of bone scintigraphy only in symptomatic patients with or without raised level of alkaline phosphatase [32,33], although others believe that because its poor sensitivity, the routine use of bone scintigraphy in RCC needs to be questioned [34,35].

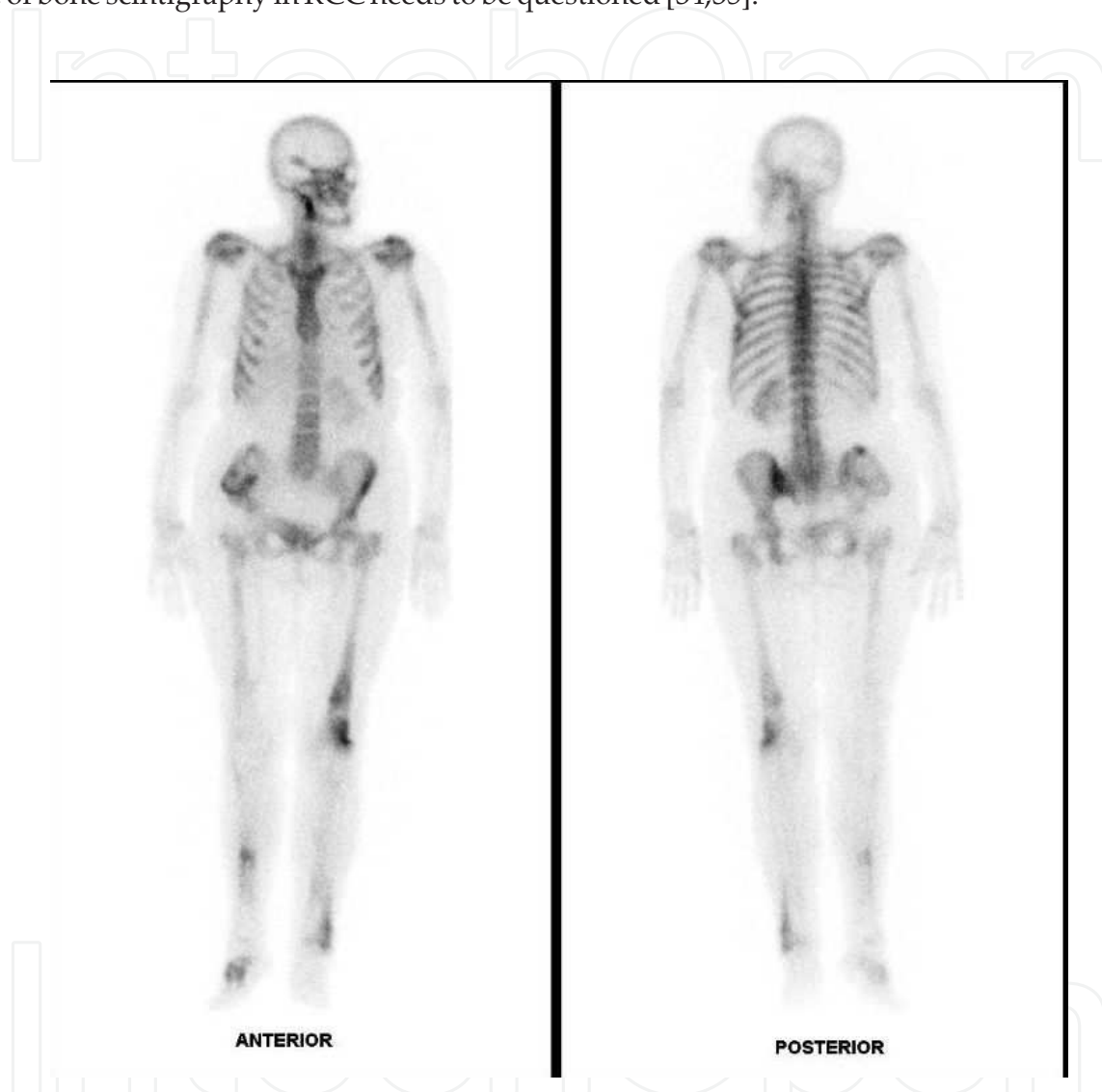


Figure 6. Bone scan of a patient with renal cancer showing metastases in the right tibia, left femur, pelvis and rib cage.

2.5. PET

We can study the PET role of in RCC from three points of view: localized disease, extensive disease and monitoring treatment response.

Localized disease: Most of publications in this patient subset have been made in a retrospective way and many of them studied patients from the PET archive and not from the popula-

tion of patients with a suspicious kidney mass. More recently we have known the result of a prospective study that examined 18 patients with renal lesions suspicious for malignancy diagnosed on CT, MRI or ultrasound [36]. In all patients, a FDG-PET/CT was made and diagnosis of malignancy was suspected when intensity on PET was greater than intensity in the renal parenchyma and it was different from the physiological excretion in the collecting system. Patients underwent nephrectomy or surgical resection of the renal mass with the respective histological analysis. PET showed a sensitivity of 46,6% and a specificity of 66,6%. The median diameter and Furhman grade of FDG positive malignant lesions were significantly higher than in FDG-negative malignant lesions ($p < 0,05$). It is difficult to draw conclusions with a study involving a sample of patients so small, but we can see that about half of the patients could not be diagnosed by PET, so probably we will have to expect better results with this diagnostic technique before introducing it as part of a routine preoperative diagnosis of RCC. A modification of the technique is the immunological PET, using ^{124}I -cG250 (chimeric girentuximab labeled with ^{124}I) because cG250 functions as an epitope of CAIX, a transmembrane enzyme that is almost universally expressed in clear cells RCC cells. With this modality has been observed a 94% sensitivity and 100% specificity, with positive and negative predictive values of 100% and 90% respectively, in a population of 26 patients with renal masses suspicious for malignancy [37].

Extensive disease: Although in the metastatic RCC PET has better sensitivity (63-100%) than in localized disease, some authors believe that FDG-PET currently appears to be too unreliable to recommend its routine use in the staging of RCC, because it is less sensitive than radiological imaging for retroperitoneal lymphadenopathy and bone or lung metastases [38]. However, this technique may have a place detecting recurrence and probably an associated prognostic value (Figure 7). In a recent study, the authors found a sensitivity and specificity of 81% and 71% respectively, for FDG-PET in the diagnosis of recurrence, with correct diagnosis in all cases of intra-abdominal (lymph nodes, local recurrence and adrenal glands) and bone recurrence, with a clear trend for better 5-year survival in PET-negative patients compared with PET-positive patients: 83% versus 46% respectively [39].

Monitoring treatment response: Systemic treatment in metastatic RCC is represented for multikinase inhibitors like sorafenib and sunitinib. These drugs are active because of their capacity of inhibition on the tyrosine kinase receptor VEGF and the platelet-derived growth factor receptor, in the endothelial cells and pericytes. Because expression of Glut (a downstream product of HIF transcriptional activity), it is conceivable that intensity of FDG uptake may be reflective of the magnitude of the entire pathway [40]. In other words, the variable intensity of FDG-PET in RCC may reflect variable strength of the HIF signaling pathway. Kayani et al. studied prospectively 44 treatment naive metastatic RCC. A basal (pretreatment) FDG-PET was made and then repeated it at 4 and 16 weeks of treatment. The most intense lesion of each patient ($\text{SUV} > 2.5$) was used as the index lesion and they defined metabolic response as a decrease of $> 20\%$ in SUV and metabolic disease progression as an increase of $> 20\%$ or development of new metastatic lesions. In the first comparison (after 4 treatment weeks) they found a metabolic response in 24 (57%) patients but without correlation with the PFS or overall survival. In the second comparison (16 treatment weeks), 12 (28%) pa-

tients had metabolic disease progression, which correlated with decreased OS and PFS 8HR: 5.96 [95% CI: 2.43-19.02] and 12.13 [95% CI: 3.72-46.51]), respectively [41]. With these results, we can conclude that the FDG-PET probably may be more useful in diagnosing tumor progression than treatment response. Another point that deserves to be examined is whether the cutoff of 20% is appropriate to differentiate responders from those who do not.

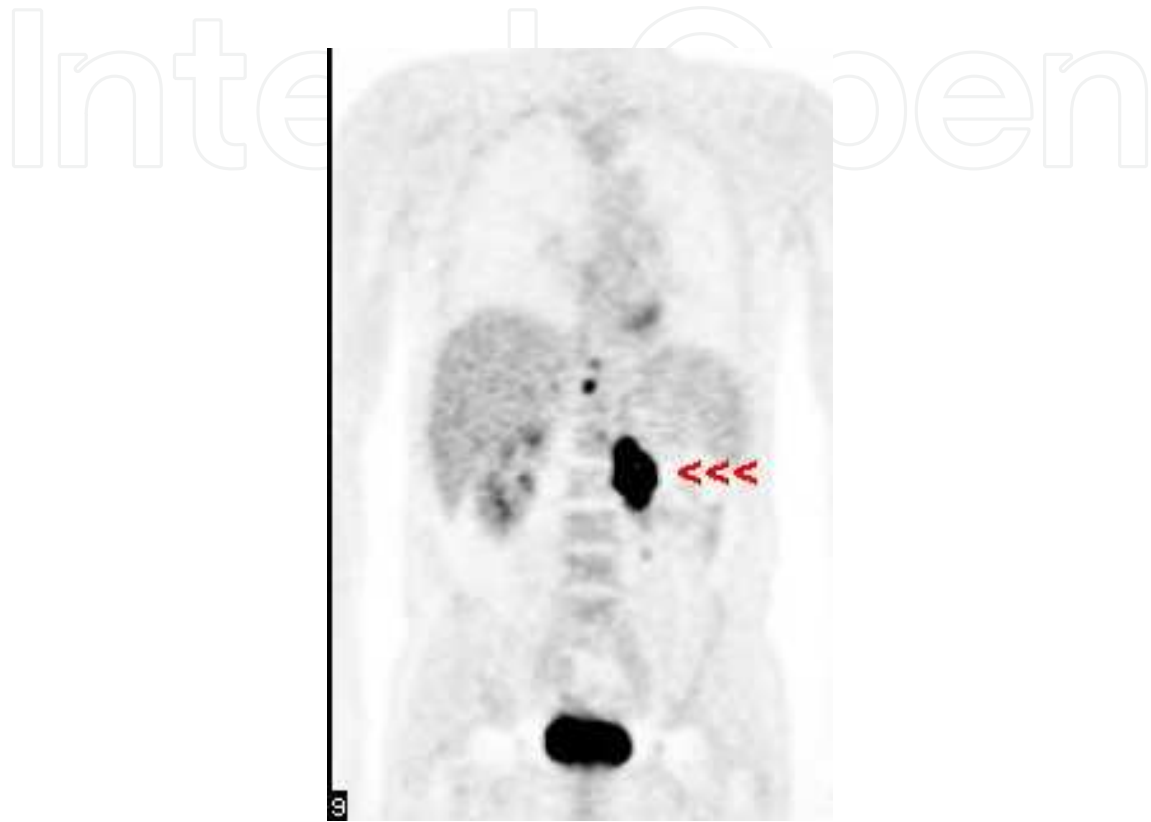


Figure 7. PET scan showing loco-regional recurrence (red arrows), in a patient with a previous left nephrectomy.

3. New techniques in imaging of renal tumors

The introduction of functional imaging techniques have allowed us to study in vivo physiological processes of tissues and tumors. Techniques such as computed tomography (CT) or magnetic resonance (MR) allow us to study tumor perfusion (angiogenesis). Positron emission tomography (PET) scan or spectroscopy RM is useful in the evaluation of tumor metabolism while diffusion RM allows the study of the diffusion of water molecules through the diffusion sequences (cellularity) to assess hypoxia phenomena or changes in the lymph nodes function. All these techniques can obtain information on the tumor microenvironment, including levels of oxygenation, tumor cell proliferation or vascularization and open a different dimension in the study of patients: diagnosis, staging, treatment planning, evaluation of response or follow-up [42] [43].

For example, dynamic techniques (MRI or CT) seem most appropriate for assessing anti-vascular drug response or acting in the VEGF/ PDGFR pathway, such as bevacizumab, whose mechanism of action appears to focus on normalization of tumor vascularization [Jain 2005], while the PET appears to do better in the case of drugs such as cetuximab, acting in the EGFR pathway [44] [45] [46].

3.1. Perfusion-CT

Perfusion CT is based on the temporal change of the attenuation of tissues after intravenous administration of iodinated contrast. This study consists of two phases. The first phase lasts between 40 and 60 seconds in which the enhancement is mainly due to the contrast distribution in the intravascular space and its rapid passage to the extracellular space. This phase requires high temporal resolution (one acquisition per second). In the second phase the contrast enhancement depends on its distribution between intra- and extravascular compartments. In this period the acquisition is more spaced and lasts between 2 and 5 minutes [47] [48] [49,50].

This functional technique can be used to measure a number of parameters including vascular blood flow, blood volume, mean transit time, peak enhancement, time to peak enhancement and capillary permeability. Several studies have validated functional CT data as a biomarker of angiogenesis [47] [51]. There is growing interest on the use of CT perfusion in oncology with multiple applications that may be helpful: differential diagnosis between benign and malignant neoplasms, identifying tumors of unknown origin (with impaired liver perfusion with occult metastatic disease), definition of prognosis (with best response in tumors with more perfusion), monitoring response to treatment and development of new drugs (Figures 8 and 9) [50]. The technique is being applied in multiple tumor types: head and neck, lung, liver, pancreas, colorectal cancer, lymphoma and prostate.

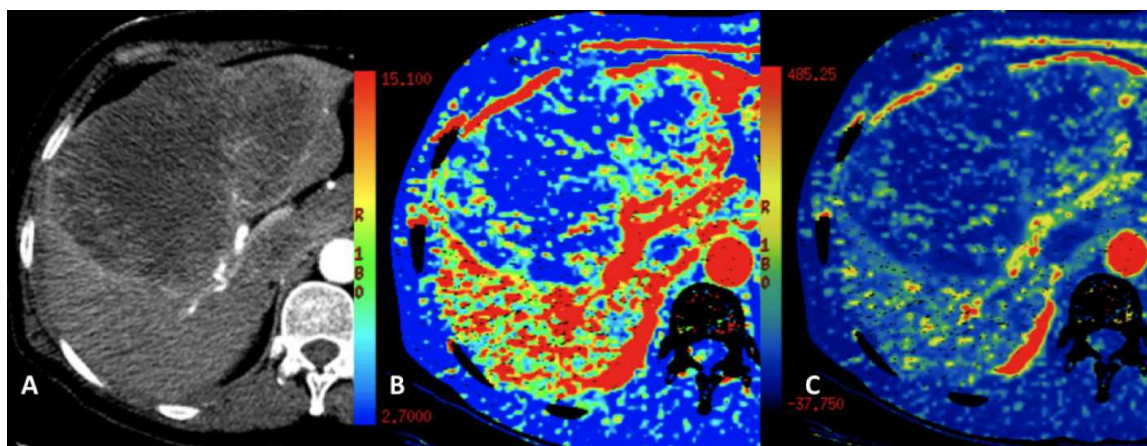


Figure 8. Renal Cancer. Liver metastasis treated with temsirolimus. Axial CT image (A) and blood volume (B) and blood flow (C) parametric maps show low perfusion parameters in metastasis. Courtesy Dr. García Figueiras.

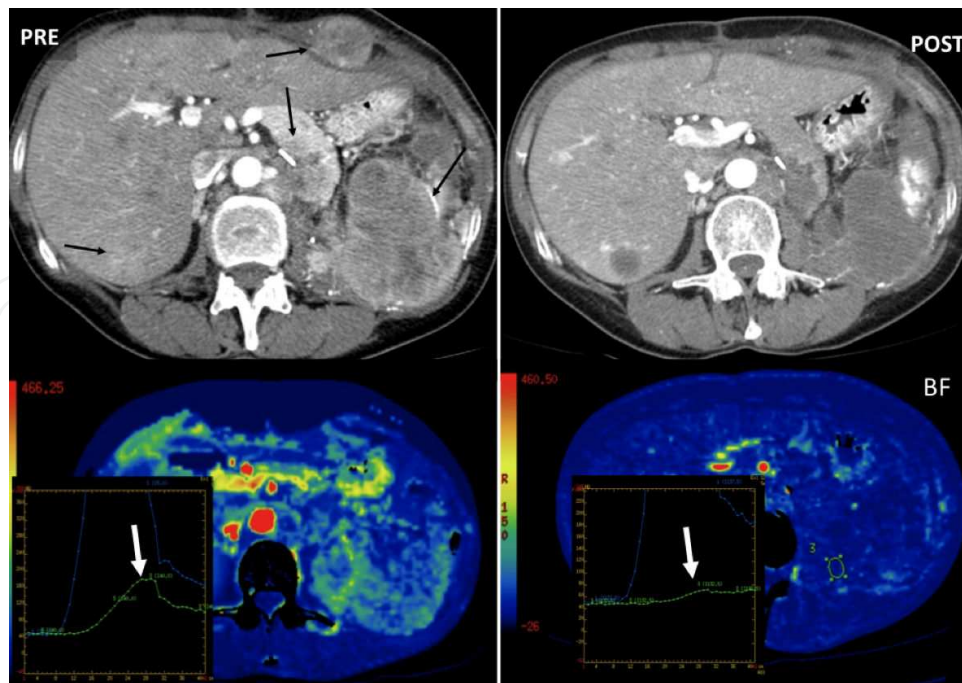


Figure 9. Renal Cancer with diffuse metastatic disease (black arrows) in a 58 year-old female patient. Pre-therapy (left column) and 10 days post-sunitinib (right column). Axial CT images, blood flow (BF) parametric maps, and curves time-density show a partial response with disappearance of some metastatic foci, necrotic changes in many of them, a change in enhancement curve (white arrows) and a BF decrease by 95% in tumor. Courtesy Dr. García Figueiras.

3.2. DCE-MRI

Other functional imaging techniques not specifically focused on the study of angiogenesis, such as diffusion MRI, enabling the study of tumor cellularity and having quantitative parameters such as the apparent diffusion coefficient (ADC). Thus, tumors with high cellularity show low ADC [52] [53]. Moreover, since tumor response is associated with destruction of tumor cells, it is generally associated with increased ADC tumor lesions. The diffusion thus evaluate the apoptotic and necrotic effect but not angiogenesis, main target of new drugs.

Preliminary studies have shown significant changes very early in the flow, blood volume and perfusion with tumor therapy. There is a relationship between changes in K_{trans} , K_{ep} and the area under the curve and the response in different tumors, showing a very marked functional changes in the vascular supply to the tumor [54] [55]. Therefore these techniques could be worth to select those patients who will respond to drugs with an early evaluation of the response using functional imaging.

In a subgroup of patients enrolled in the phase II study discontinuation of sorafenib, DCE-MRI was performed before and after initiation of treatment. Radiological response by RECIST criteria was observed in 4/17 patients (ORR 24%), and time to progression was 12.9 months. K_{trans} decreased significantly during treatment with sorafenib (60.3% decrease,

95% CI 46.1 to 74.6%). The percentage decrease in K_{trans} and change in tumor size was significantly associated with progression-free survival ($p = 0.01$ and 0.05 , respectively).

3.3. PET

Finally, molecular techniques such as PET show a limited role in the study of metastatic renal cancer, since this tumor usually has a low activity of glucose metabolism (pathway assessed by ^{18}F -fluorodeoxyglucose, the most widely used radiotracer). Only in cases where the tumor shows an increased metabolism of glucose, PET may be useful in the assessment of the disease and its response to therapy. Other radiotracers that allow the study of important characteristics such as tumor hypoxia, cell proliferation or angiogenesis itself, are still under evaluation and implementation in clinical practice [56]. In an experimental way it is evaluating the introduction of functional imaging techniques in clinical studies, to develop translational research in oncology imaging applications. In a NCI trial, Dr. Hoffman (University of Utah) is using DCE-MRI and various types of PET ($H2150$ -PET, FDG-PET, FDL-PET) in monitoring response to multi-targeted treatment in renal cancer patients.

4. Response evaluation

4.1. Evaluation of response: Antiangiogenics and mTOR inhibitors

We must consider several issues when assessing the therapeutic response of tumors. The morphological assessment with quantification of changes in size used in the RECIST criteria ("Criteria in Solid Tumors Response") has been our main concern when assessing tumor response [2]. This approach seems true for the use of cytotoxic drugs. However, this assessment is limited, since the macroscopic changes take time to become evident, often are not specific and do not provide information on the physiological and molecular component of tumors [42].

Advances in the field of oncology have led to the development of new drugs in renal cancer as sunitinib, sorafenib, pazopanib, tivoaznib, axitinib, temsirolimus, everolimus and bevacizumab [46]. These drugs (mainly cytostatic) cause little change in lesion size. Therefore, RECIST criteria are not entirely suitable for assessing tumor response, and proper techniques will vary according to the mechanism of action of the drug.

The recent emergence of techniques for the functional study of angiogenesis, such as perfusion CT or dynamic MRI allow obtaining quantitative parameters (blood volume, blood flow mean transit time, k_{trans} V_e , etc.) and would open an interesting field for assessing tumor response in a more objective [57] [58] [59]. This could open the door to the development of a strategy based on the image for the selection of patients to be treated with antiangiogenic therapies. However, each of these techniques has advantages and disadvantages. Thus, CT perfusion shows the drawback of radiation necessary for conducting the studies, whereas in the case of dynamic MRI the analysis of the results is much more complex.

The functional and molecular imaging techniques could offer clear opportunities in the study of renal tumors, but nevertheless, we must not forget that, for validation as biomarkers, would require completing a qualification and validation process, which would pass through standardization in the collection and analysis of the images and the correlation of the parameters obtained with patient outcomes. Once this is achieved, functional-molecular techniques, especially perfusion CT, could become promising tools in the selection of patients for targeted drug therapy and the assessment of the response [57] [58].

5. Criterios RECIST/MASS/CHOI

Classically, oncology response evaluation is based on comparison of pre and post-treatment tumor volume by studying changes in the diameter of the tumors. RECIST criteria in its original version and its 2009 Update 1.1 are applied routinely in oncology practice [2]. However, it is recognized that the response evaluation focused exclusively on size changes have important limitations, including the importance of excluding changes in tumor metabolism or not considering the appearance of necrosis or fibrosis as a factor which may be related to response to treatment. Furthermore, the introduction of new drugs creates the need for a different evaluation of the tumor and treatment response [46].

The limitations of traditional approaches, as the criteria of the World Health Organization (WHO) or Response Evaluation Criteria in Solid Tumors (**RECIST**) in the evaluation of targeted therapies have been widely documented [64] [65] [57]. Therapies that act on tumor vascularity may have underestimated clinical benefit by tumor size change since their mechanism of action (more cytostatic than cytotoxic), produces more stabilization than tumoral responses.

Without abandoning the use of size criteria as a key element in the assessment of patients with metastatic renal cancer, some authors have attempted to obtain early information (**EP-TIC**, Early English Post-therapy Imaging Changes) [66] on the prognosis of patients treated with therapy acting at the VEGF pathway. In this regard, it was demonstrated that a 10% decrease in the sum of the largest diameters of the lesions in the first control, provides information on the subsequent course of patients. Using only tumor size as endpoint criterion would leave aside the use of IV contrast.

Subsequently it was observed a relationship between the degree of tumor enhancement before therapy and the likelihood of response (being higher in those tumors with greater pre-treatment enhancement). Many of these new drugs induce tumor necrosis, causing a dramatic drop in the enhancement of metastatic lesions in the post-therapy evaluation [67]. Based on these observations and on previous experience with gastrointestinal stromal tumors treated with imatinib, a set of tumor response criteria based on changes in size and / or density tumor was established: Choi criteria, modified Choi criteria, MASS criteria and SACT criteria (Table 1) [56].

Criteria	Targeted lesions	Complete response	Partial response	Stable disease	Progressive disease
RECIST version 1.1 (2)	Tumor size 10 mm by CT scan	Disappearance of all lesions	Decrease in size 30%	Does not meet criteria for PR or PD	Increase in tumor size 20% (the sum must also demonstrate an absolute increase of at least 5 mm)
	Tumor size 15 mm by chest X-ray	No new lesions	No new lesions		New lesions
	Maximum of 5 target lesions		No PD of non-target lesions		
EPTIC (Early Posttherapy Imaging Changes) (60)	Establishing the prognosis depending on the % size decrease after antiangiogenic therapy Decreased size $\geq 10\%$ = good prognosis.				
Choi Criteria (61) (3)	Tumor size 15 mm	Disappearance of all lesions	A decrease in size 10% or a decrease in tumor attenuation (HU) 15% on CT	Does not meet criteria for CR, PR or PD	Increase in tumor size 10% and does not meet criteria of PR by tumor attenuation
	Maximum of 10 target lesions	No new lesions	No new lesions, no obvious progression of non-measurable disease	No symptomatic deterioration attributed to PD	New lesions
Modified Choi Criteria (62)	Tumor size 15 mm	Disappearance of all lesions	A decrease in size 10% and a decrease in tumor attenuation (HU) 15% on CT	Does not meet criteria for CR, PR or PD	Increase in tumor size 10% and does not meet criteria of PR by tumor attenuation
	Maximum of 10 target lesions	No new lesions	No new lesions, no obvious progression of non-measurable disease	No symptomatic deterioration attributed to PD	New lesions

Criteria	Targeted lesions	Complete response	Partial response	Stable disease	Progressive disease
SCAT Criteria (29)	Tumor size 10 mm		Decrease in tumor size 20%	Does not meet criteria for PR or PD	Increase in tumor size 20%
	Maximum of 10 target lesions		Decrease in tumor size 10% and half of the non-lung target lesions with 20 HU decreased mean attenuation		New metastases, marked central fill-in of a target lesion or new enhancement in a homogeneously hypoattenuating nonenhancing mass
MASS criteria (63)		Favorable response. No new lesions and any of the following: 1. Decrease in tumor size 20 % 2. One or more predominantly solid enhancing lesions with marked central necrosis or marked decreased attenuation (40 UH)		Does not meet criteria for favorable or unfavorable response	Any of the following: 1. Increase in tumor size of 20 % in the absence of marked central necrosis or marked decreased attenuation 2. New metastases, marked central fill-in, or new enhancement of a previously homogeneously hypoattenuating nonenhancing mass
Functional and molecular image.	No response criteria defined				
CR: complete response. PR: partial response. NC: no changes. PD: progressive disease. RECIST: Response Evaluation Criteria in Solid Tumors. CT: computed tomography. SACT: criteria size and attenuation CT (SACT) criteria. HU: Hounsfield Unit.					

CR: complete response. PR: partial response. NC: no changes. PD: progressive disease. RECIST: Response Evaluation Criteria in Solid Tumors. CT: computed tomography. SACT: criteria size and attenuation CT (SACT) criteria. HU: Hounsfield Unit.

Table 1. CT-based criteria for response evaluation of targeted therapies in renal cell carcinoma.

Each of these criteria has a number of advantages compared to RECIST, but some limitations. So **Choi criteria**, based on the change in size or tumor density on CT (% change in the measured attenuation value at UH), show little advantage over RECIST when establishing the possibility of a long-term response. Furthermore, these show a different utility criteria

depending on the type of drug tested, being most useful in the case of sorafenib (which tends to cause more degree of necrosis in lesions) than for sunitinib.

Modified Choi criteria evaluate existing changes in both size and tumor density after treatment. These criteria could differentiate those patients at risk of disease progression, but shows a tendency to classify patients as responders.

SACT criteria (Size and Attenuation CT) differ from the modified Choi criteria that establish an absolute value of change in tumor density (> 20 UH) rather than a % of change. These criteria are more reliable in the case of low attenuation pre-therapy lesions, in which it is easier to obtain a percentage decrease in density.

Finally, the **MASS criteria** (Morphology, Attenuation, Size, and Structure) include morphological and structural elements regardless of the size and density of lesions. These criteria are intended to take into account the extensive necrotic changes frequently associated with tumor response to these drugs [63].

However, both SACT as MASS criteria are complicated and basically useful in differentiating patients with a long progression-free survival (> 250 days) of those showing a rapid progression (< 250 days). Overall, we consider that in all these criteria contrast enhancement of lesions plays a major role, so that both imaging protocols (volume of contrast acquisition phase, etc.) or factors such as cardiac function patient can significantly influence the results.

6. Summary

The era of molecular biology have created great expectations on our ability to translate these discoveries into effective treatments for patients. Over the last decade, there has been an increasing knowledge about pathophysiological processes that are common to most tumors including: independence from growth signals, insensitivity to growth-inhibitory signals, evasion of apoptosis, limitless potential for replication, sustained angiogenesis, and tissue invasion and metastasis. These major pathways deregulated in cancer have a key role in tumor development and microenvironment. These features have enabled the emergence of a wide spectrum of novel oncologic drugs that are designed to target and interfere with specific aberrant biological pathways. In general, these agents use different strategies to interfere with specific biological targets, such as blocking growth factors, receptors, or tyrosine kinase (TK) action.

The use of new drugs in the treatment of advanced or metastatic kidney cancer, with different mechanisms of action compared to conventional chemotherapy raises new questions. One of the biggest problems with new drugs are produced in the evaluation of the response, and the incorporation of new imaging techniques (MRI diffusion, perfusion CT, nuclear medicine, etc...) in the diagnosis of extension and assessment of efficacy.

In this chapter we have reviewed the main techniques of radiological diagnosis and staging, the value of new imaging modalities, and discuss the validity of the classical criteria of interpretation of response.

Author details

L. León¹, M. Ramos², M. Lázaro³, S. Vázquez⁴, M. C. Areses⁵, O. Fernandez⁵, U. Anido¹, J. Afonso⁶ and L. A. Aparicio⁷

*Address all correspondence to: Aparicio@sergas.es

1 Complejo Hospitalario Universitario de Santiago, Santiago de Compostela, Spain

2 Centro Oncológico de Galica, A Coruña, Spain

3 Complejo Hospitalario Universitario, Vigo, Spain

4 Hospital Universitario Lucus Augusti, Lugo, Spain

5 Complejo Hospitalario Universitario de Ourense, Ourense, Spain

6 Complejo Hospitalario Arquitecto Marcide, Ferrol, Spain

7 Complejo Hospitalario Universitario, A Coruña, Spain

References

- [1] Cohen HT, McGovern FJ. Renal-cell carcinoma. *N Engl J Med* 2005, Dec 8;353(23):2477-90.
- [2] Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). *Eur J Cancer* 2009, Jan;45(2):228-47.
- [3] Choi H, Charnsangavej C, Faria SC, Macapinlac HA, Burgess MA, Patel SR, et al. Correlation of computed tomography and positron emission tomography in patients with metastatic gastrointestinal stromal tumor treated at a single institution with imatinib mesylate: Proposal of new computed tomography response criteria. *J Clin Oncol* 2007, May 1;25(13):1753-9.
- [4] Jamis-Dow CA, Choyke PL, Jennings SB, Linehan WM, Thakore KN, Walther MM. Small (< or = 3-cm) renal masses: Detection with CT versus US and pathologic correlation. *Radiology* 1996, Mar;198(3):785-8.
- [5] Siracusano S, Bertolotto M, Ciciliato S, Valentino M, Liguori G, Visalli F. The current role of contrast-enhanced ultrasound (CEUS) imaging in the evaluation of renal pathology. *World J Urol* 2011, Oct;29(5):633-8.

- [6] Ascenti G, Mazziotti S, Zimbaro G, Settineri N, Magno C, Melloni D, et al. Complex cystic renal masses: Characterization with contrast-enhanced US. *Radiology* 2007, Apr;243(1):158-65.
- [7] Quaia E, Bertolotto M, Cioffi V, Rossi A, Baratella E, Pizzolato R, Cov MA. Comparison of contrast-enhanced sonography with unenhanced sonography and contrast-enhanced CT in the diagnosis of malignancy in complex cystic renal masses. *AJR Am J Roentgenol* 2008, Oct;191(4):1239-49.
- [8] Park BK, Kim B, Kim SH, Ko K, Lee HM, Choi HY. Assessment of cystic renal masses based on bosniak classification: Comparison of CT and contrast-enhanced US. *Eur J Radiol* 2007, Feb;61(2):310-4.
- [9] Maldazys JD, deKernion JB. Prognostic factors in metastatic renal carcinoma. *J Urol* 1986, Aug;136(2):376-9.
- [10] Zhang J, Lefkowitz RA, Ishill NM, Wang L, Moskowitz CS, Russo P, et al. Solid renal cortical tumors: Differentiation with CT. *Radiology* 2007, Aug;244(2):494-504.
- [11] Kopka L, Fischer U, Zoeller G, Schmidt C, Ringert RH, Grabbe E. Dual-phase helical CT of the kidney: Value of the corticomedullary and nephrographic phase for evaluation of renal lesions and preoperative staging of renal cell carcinoma. *AJR Am J Roentgenol* 1997, Dec;169(6):1573-8.
- [12] Sheth S, Scatarige JC, Horton KM, Corl FM, Fishman EK. Current concepts in the diagnosis and management of renal cell carcinoma: Role of multidetector ct and three-dimensional CT. *Radiographics* 2001, Oct;21 Spec No:S237-54.
- [13] Ruppert-Kohlmayr AJ, Uggowitzer M, Meissnitzer T, Ruppert G. Differentiation of renal clear cell carcinoma and renal papillary carcinoma using quantitative CT enhancement parameters. *AJR Am J Roentgenol* 2004, Nov;183(5):1387-91.
- [14] Zagoria RJ, Bechtold RE, Dyer RB. Staging of renal adenocarcinoma: Role of various imaging procedures. *AJR Am J Roentgenol* 1995, Feb;164(2):363-70.
- [15] Johnson CD, Dunnick NR, Cohan RH, Illescas FF. Renal adenocarcinoma: CT staging of 100 tumors. *AJR Am J Roentgenol* 1987, Jan;148(1):59-63.
- [16] Welch TJ, LeRoy AJ. Helical and electron beam CT scanning in the evaluation of renal vein involvement in patients with renal cell carcinoma. *J Comput Assist Tomogr* 1997;21(3):467-71.
- [17] Gill IS, McClennan BL, Kerbl K, Carbone JM, Wick M, Clayman RV. Adrenal involvement from renal cell carcinoma: Predictive value of computerized tomography. *J Urol* 1994, Oct;152(4):1082-5.
- [18] Trinidad C, Martinez C, Delgado C. Tumores urológicos. Imagen en oncología.. In: Madrid: Ed Médica PAnamericana; 2009. p. 67-78.

- [19] Eilenberg SS, Lee JK, Brown J, Mirowitz SA, Tartar VM. Renal masses: Evaluation with gradient-echo gd-dtpa-enhanced dynamic MR imaging. *Radiology* 1990, Aug; 176(2):333-8.
- [20] Yamashita Y, Watanabe O, Miyazaki T, Yamamoto H, Harada M, Takahashi M. Cystic renal cell carcinoma. Imaging findings with pathologic correlation. *Acta Radiol* 1994, Jan;35(1):19-24.
- [21] Pedrosa I, Sun MR, Spencer M, Genega EM, Olumi AF, Dewolf WC, Rofsky NM. MR imaging of renal masses: Correlation with findings at surgery and pathologic analysis. *Radiographics* 2008;28(4):985-1003.
- [22] Roy C, Sauer B, Lindner V, Lang H, Saussine C, Jacqmin D. MR imaging of papillary renal neoplasms: Potential application for characterization of small renal masses. *Eur Radiol* 2007, Jan;17(1):193-200.
- [23] Pedrosa I, Sun MR, Spencer M, Genega EM, Olumi AF, Dewolf WC, Rofsky NM. MR imaging of renal masses: Correlation with findings at surgery and pathologic analysis. *Radiographics* 2008;28(4):985-1003.
- [24] Harmon WJ, King BF, Lieber MM. Renal oncocytoma: Magnetic resonance imaging characteristics. *J Urol* 1996, Mar;155(3):863-7.
- [25] Taouli B, Thakur RK, Mannelli L, Babb JS, Kim S, Hecht EM, et al. Renal lesions: Characterization with diffusion-weighted imaging versus contrast-enhanced MR imaging. *Radiology* 2009, May;251(2):398-407.
- [26] Sandrasegaran K, Sundaram CP, Ramaswamy R, Akisik FM, Rydberg MP, Lin C, Aisen AM. Usefulness of diffusion-weighted imaging in the evaluation of renal masses. *AJR Am J Roentgenol* 2010, Feb;194(2):438-45.
- [27] Sohaib SA, Cook G, Allen SD, Hughes M, Eisen T, Gore M. Comparison of whole-body MRI and bone scintigraphy in the detection of bone metastases in renal cancer. *Br J Radiol* 2009, Aug;82(980):632-9.
- [28] Ohno Y, Koyama H, Onishi Y, Takenaka D, Nogami M, Yoshikawa T, et al. Non-small cell lung cancer: Whole-body MR examination for m-stage assessment--utility for whole-body diffusion-weighted imaging compared with integrated FDG PET/CT. *Radiology* 2008, Aug;248(2):643-54.
- [29] Smith AD, Lieber ML, Shah SN. Assessing tumor response and detecting recurrence in metastatic renal cell carcinoma on targeted therapy: Importance of size and attenuation on contrast-enhanced CT. *AJR Am J Roentgenol* 2010, Jan;194(1):157-65.
- [30] Adiga GU, Dutcher JP, Larkin M, Garl S, Koo J. Characterization of bone metastases in patients with renal cell cancer. *BJU Int* 2004, Jun;93(9):1237-40.
- [31] Staudenherz A, Steiner B, Puig S, Kainberger F, Leitha T. Is there a diagnostic role for bone scanning of patients with a high pretest probability for metastatic renal cell carcinoma? *Cancer* 1999, Jan 1;85(1):153-5.

- [32] Hafez KS, Novick AC, Campbell SC. Patterns of tumor recurrence and guidelines for followup after nephron sparing surgery for sporadic renal cell carcinoma. *J Urol* 1997, Jun;157(6):2067-70.
- [33] Levy DA, Slaton JW, Swanson DA, Dinney CP. Stage specific guidelines for surveillance after radical nephrectomy for local renal cell carcinoma. *J Urol* 1998, Apr;159(4):1163-7.
- [34] Koga S, Tsuda S, Nishikido M, Ogawa Y, Hayashi K, Hayashi T, Kanetake H. The diagnostic value of bone scan in patients with renal cell carcinoma. *J Urol* 2001, Dec;166(6):2126-8.
- [35] Blacher E, Johnson DE, Haynie TP. Value of routine radionuclide bone scans in renal cell carcinoma. *Urology* 1985, Nov;26(5):432-4.
- [36] Ozülker T, Ozülker F, Ozbek E, Ozpaçacı T. A prospective diagnostic accuracy study of F-18 fluorodeoxyglucose-positron emission tomography/computed tomography in the evaluation of indeterminate renal masses. *Nucl Med Commun* 2011, Apr;32(4):265-72.
- [37] Divgi CR, Pandit-Taskar N, Jungbluth AA, Reuter VE, Gönen M, Ruan S, et al. Pre-operative characterisation of clear-cell renal carcinoma using iodine-124-labelled antibody chimeric G250 (124i-cg250) and PET in patients with renal masses: A phase I trial. *Lancet Oncol* 2007, Apr;8(4):304-10.
- [38] Mueller-Lisse UG, Mueller-Lisse UL. Imaging of advanced renal cell carcinoma. *World J Urol* 2010, Jun;28(3):253-61.
- [39] Nakatani K, Nakamoto Y, Saga T, Higashi T, Togashi K. The potential clinical value of FDG-PET for recurrent renal cell carcinoma. *Eur J Radiol* 2011, Jul;79(1):29-35.
- [40] Khandani AH, Rathmell WK. Positron emission tomography in renal cell carcinoma: An imaging biomarker in development. *Semin Nucl Med* 2012, Jul;42(4):221-30.
- [41] Kayani I, Avril N, Bomanji J, Chowdhury S, Rockall A, Sahdev A, et al. Sequential FDG-PET/CT as a biomarker of response to sunitinib in metastatic clear cell renal cancer. *Clin Cancer Res* 2011, Sep 15;17(18):6021-8.
- [42] Torigian DA, Huang SS, Houseni M, Alavi A. Functional imaging of cancer with emphasis on molecular techniques. *CA Cancer J Clin* 2007;57(4):206-24.
- [43] Atri M. New technologies and directed agents for applications of cancer imaging. *J Clin Oncol* 2006, Jul;24(20):3299-308.
- [44] Galbraith SM, Maxwell RJ, Lodge MA, Tozer GM, Wilson J, Taylor NJ, et al. Combretastatin A4 phosphate has tumor antivascular activity in rat and man as demonstrated by dynamic magnetic resonance imaging. *J Clin Oncol* 2003, Aug;21(15):2831-42.
- [45] Jain RK. Normalization of tumor vasculature: An emerging concept in antiangiogenic therapy. *Science* 2005, Jan;307(5706):58-62.

- [46] Desar IME, van Herpen CML, van Laarhoven HWM, Barentsz JO, Oyen WJG, van der Graaf WTA. Beyond RECIST: Molecular and functional imaging techniques for evaluation of response to targeted therapy. *Cancer Treat Rev* 2009, Jun;35(4):309-21.
- [47] Jeswani T, Padhani AR. Imaging tumour angiogenesis. *Cancer Imaging* 2005;5:131-8.
- [48] Cuenod CA, Fournier L, Balvay D, Guinebretière J-MM. Tumor angiogenesis: Pathophysiology and implications for contrast-enhanced MRI and CT assessment. *Abdom Imaging* 2006;31(2):188-93.
- [49] Goh V, Padhani AR. Imaging tumor angiogenesis: Functional assessment using MDCT or MRI? *Abdom Imaging* 2006;31(2):194-9.
- [50] Kambadakone AR, Sahani DV. Body perfusion CT: Technique, clinical applications, and advances. *Radiol Clin North Am* 2009, Jan;47(1):161-78.
- [51] Ma S-HH, Le H-BB, Jia B-HH, Wang Z-XX, Xiao Z-WW, Cheng X-LL, et al. Peripheral pulmonary nodules: Relationship between multi-slice spiral CT perfusion imaging and tumor angiogenesis and VEGF expression. *BMC Cancer* 2008;8:186.
- [52] Padhani AR. Diffusion magnetic resonance imaging in cancer patient management. *Semin Radiat Oncol* 2011, Apr;21(2):119-40.
- [53] Li SP, Padhani AR. Tumor response assessments with diffusion and perfusion MRI. *J Magn Reson Imaging* 2012, Apr;35(4):745-63.
- [54] Turkbey B, Kobayashi H, Ogawa M, Bernardo M, Choyke PL. Imaging of tumor angiogenesis: Functional or targeted? *AJR Am J Roentgenol* 2009, Aug;193(2):304-13.
- [55] Gwyther SJ, Schwartz LH. How to assess anti-tumour efficacy by imaging techniques. *Eur J Cancer* 2008, Jan;44(1):39-45.
- [56] van der Veldt AA, Meijerink MR, van den Eertwegh AJ, Boven E. Targeted therapies in renal cell cancer: Recent developments in imaging. *Target Oncol* 2010, Jun;5(2):95-112.
- [57] Figueiras RG, Padhani AR, Goh VJ, Vilanova JC, González SB, Martín CV, et al. Novel oncologic drugs: What they do and how they affect images. *Radiographics* 2011;31(7):2059-91.
- [58] Garcia Figueiras R. CT perfusion techniques in oncologic imaging: A useful tool? *AJR Am J Roentgenol* 2013, Jan 1.
- [59] Flaherty KT, Rosen MA, Heitjan DF, Gallagher ML, Schwartz B, Schnall MD, O'Dwyer PJ. Pilot study of DCE-MRI to predict progression-free survival with sorafenib therapy in renal cell carcinoma. *Cancer Biol Ther* 2008, Apr;7(4):496-501.
- [60] Thiam R, Fournier LS, Trinquart L, Medioni J, Chatellier G, Balvay D, et al. Optimizing the size variation threshold for the CT evaluation of response in metastatic renal cell carcinoma treated with sunitinib. *Ann Oncol* 2010, May;21(5):936-41.

- [61] van der Veldt AA, Meijerink MR, van den Eertwegh AJ, Haanen JB, Boven E. Choi response criteria for early prediction of clinical outcome in patients with metastatic renal cell cancer treated with sunitinib. *Br J Cancer* 2010, Mar 2;102(5):803-9.
- [62] Nathan PD, Vinayan A, Stott D, Juttla J, Goh V. CT response assessment combining reduction in both size and arterial phase density correlates with time to progression in metastatic renal cancer patients treated with targeted therapies. *Cancer Biol Ther* 2010, Jan;9(1):15-9.
- [63] Smith AD, Shah SN, Rini BI, Lieber ML, Remer EM. Morphology, attenuation, size, and structure (MASS) criteria: Assessing response and predicting clinical outcome in metastatic renal cell carcinoma on antiangiogenic targeted therapy. *AJR Am J Roentgenol* 2010, Jun;194(6):1470-8.
- [64] Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). *Eur J Cancer* 2009, Jan;45(2):228-47.
- [65] van Cruijsen H, van der Veldt A, Hoekman K. Tyrosine kinase inhibitors of VEGF receptors: Clinical issues and remaining questions. *Front Biosci* 2009;14:2248-68.
- [66] Krajewski KM, Guo M, Van den Abbeele AD, Yap J, Ramaiya N, Jagannathan J, et al. Comparison of four early posttherapy imaging changes (EPTIC; RECIST 1.0, tumor shrinkage, computed tomography tumor density, choi criteria) in assessing outcome to vascular endothelial growth factor-targeted therapy in patients with advanced renal cell carcinoma. *Eur Urol* 2011, May;59(5):856-62.
- [67] Nishino M, Jagannathan JP, Krajewski KM, O'Regan K, Hatabu H, Shapiro G, Ramaiya NH. Personalized tumor response assessment in the era of molecular medicine: Cancer-specific and therapy-specific response criteria to complement pitfalls of RECIST. *AJR Am J Roentgenol* 2012, Apr;198(4):737-45.