Original Article Microvascular invasion as a prognostic indicator in renal cell carcinoma: a systematic review and meta-analysis

Hai Huang^{1*}, Xiu-Wu Pan^{1*}, Yi Huang^{1*}, Dan-Feng Xu², Xin-Gang Cui³, Lin Li¹, Yi Hong¹, Lu Chen¹, Yi Gao¹, Lei Yin¹

¹Department of Urinary Surgery of Changzheng Hospital, Second Military Medical University, 415 Fengyang Road, Shanghai 200003, People's Republic of China; ²Urology Research Center of the Chinese People's Liberation Army, Changzheng Hospital, Second Military Medical University, 415 Fengyang Road, Shanghai 200003, People's Republic of China; ³Department of Urinary Surgery of Eastern Hepatobiliary Surgery Hospital, Second Military Medical University, Shanghai 200438, People's Republic of China. ^{*}Equal contributors.

Received April 13, 2015; Accepted June 10, 2015; Epub July 15, 2015; Published July 30, 2015

Abstract: Microvascular invasion (MVI), an omen of potential hematogenous spread of tumor cells, has been identified as an accepted risk factor for poor prognosis in some solid tumors. But its prognostic value in renal cell carcinoma (RCC) remains disputable. In order to address this question rigorously, we performed a systematical review of the published literature on MVI and RCC prognosis. According to the PRISMA statement, we searched PubMed, Web of science, and Cochrane Library database and identified 33 cohort articles that met the eligibility criteria and involved 14,946 patients (48-2596 per study) in this meta-analysis. Using the random effects model, the association between MVI and four generally recognized end points were estimated, including cancer-specific survival (CSS), recurrence-free survival (RFS), metastasis-free survival (MFS) and overall survival (OS). The presence of MVI was detected in 14.4% of the pathological specimens. A higher incidence of MVI was associated with some acknowledged prognostic risk factors such as higher pathological TNM stages and higher tumor grades. Statistical significance of the combined hazard ratio (HR) was detected for CSS (HR, 2.090; 95% CI, 1.530-2.857), RFS (HR = 2.749; 95% CI, 1.974-3.828), MFS (HR = 1.621; 95% CI, 1.095-2.400). However, the association between MVI and worse overall survival did not address statistical significance (HR = 1.371; 95% CI, 0.978-1.923). These findings suggest that the presence of MVI has a detrimental effect on clinicopathological features of RCC and could serve as a poor prognostic factor for patient with RCC.

Keywords: Microvascular invasion, renal cell carcinoma, meta-analysis, systematic review, prognosis

Introduction

There are estimated 63,920 new cases and 13,860 deaths from renal cancer in the United States in 2014 [1]. Renal cell carcinoma (RCC) is one of the most lethal urologic cancers, more than 80-90% of which are histologically diagnosed as clear cell renal cell carcinoma (ccRCC). Tumor TNM stage and nuclear grade are most frequently used in RCC as prognostic factors. However, even with the resection of the localized tumor, up to a third of patients will go on to develop local recurrence or distant metastasis [2], and the worldwide incidence and mortality rates are raising at a rate of 2-3% per decade [3]. One of the major clinician's concerns is therefore how to identify patients at high risk of

poor outcome. Recently, with the improved knowledge of pathologic parameters such as sarcomatoid/rhabdoid differentiation, tumor necrosis and microvascular invasion, we can make better prognostic evaluations, for a more comprehensive and effective way.

Considering that RCC is one of the most highly vascularized tumors, it is not surprising that vascular invasion is frequently found in these tumors. And macrovascular invasion into the renal vein and/or the vena cava is one of well recognized prognostic factors in RCC, which has been included in TNM staging of RCC. Microvascular invasion (MVI), another type of vascular invasion, refers to the presence of tumor within microscopic venules or veins with



VI: vascular invasion; MVI: microvascular invasion

Figure 1. The flow diagram of the selection process. Flow diagram illustrating the search strategy used according to PRISMA (preferred reporting items for systematic reviews and meta-analyses) statement.

a muscular coat or the lymphatic system, or both. It could be considered as an omen of potential hematogenous spread of tumor cells, which has drawn more and more attention and also been identified as an independent risk factor for poor prognosis in many solid tumors [4-8]. Although numerous studies have been performed to evaluate the impact of MVI in RCC on prognosis of RCC, the results remain disputable. The aim of the present systematical review is to assess the prognostic value of MVI in renal cell carcinoma.

Materials and methods

Search strategy

This meta-analysis was performed and reported following the proposed Preferred Reporting Items for Systematic reviews and Meta-Analyses (PRISMA) [9] statement. We searched electronic databases including PubMed, web of

Cochrane science. and Library for published studies that analyzed the prognostic value of MVI in RCC up to May 31, 2014. The following Medical Subject Headings terms and free text were used: "lymphovascular or lymphatic or microvascular or microvessel or microvenous or microscopic or vascular" AND "invasion or infiltration" AND "renal or kidney" AND "cancer or carcinoma or neoplasm or tumor or mass or tumour" AND "Predict* or prognos* or survival or risk or outcome". The searching strategies and results are shown in Table S1. There was no restriction on population or publication year. Additionally, we conducted a manual search using the bibliographies of all the identified studies, reviews, and editorials to identify references that we may have missed during our primary search.

Selection criteria

Inclusion criteria: (1) studies that included the pathologically confirmed diagnosis of RCC; (2) studies that assessed MVI, and other similar or equivalent concepts such as lymphovascular invasion (LVI), microscopic vascular invasion and microscopic venous invasion; (3) studies in which the primary treatment for RCC patients was limited to surgery with or without adjuvant therapy; (4) studies that analyzed the potential association between the prognosis of RCC patients and MVI; and (5) studies that offered a hazard ratio (HR) and 95% confidence interval (CI) categorically or the data presented were available for calculation of the HR and 95% CI.

Exclusion criteria: (1) studies that were performed on animal models or renal cancer cell lines; (2) letters, review articles, commentaries, clinical guidelines, or case reports; (3) the language of the studies were not English; (4) studies that only analyzed subtypes of RCC and excluded ccRCC; (5) studies with a sample sizes smaller than 30; and (6) if multiple publi-

Study	Staging system	Grading system	MVI+/MVI-	Stage 1-2/ 3-4	Stage 1-2/ Grade1-2/ 3-4 3-4		M0/M1	ccRCC/ non-ccRCC
Belsante [14]	2010 AJCC	Fuhrman	60/359	333/86	288/NA	411/8	419/0	419/0
	2010 AJCC	Fuhrman	21/312	333/0	258/NA	333/0	333/0	333/0
Eisenberg [15]	2010 AJCC	Fuhrman	119/984	713/390	469/634	1106/87	971/132	1103/0
Shindo [16]	2009 AJCC	NA	14/158	172/0	158/14	172/0	172/0	151/21
Drewniak [17]	2010 AJCC	Fuhrman	43/105	37/94	46/73	18/80	13/116	34/39
Steffens [18]	2002 AJCC	Fuhrman	259/1771	1301/729	1738/292	1899/131	1780/250	1721/309
Betsunoh [19]	2009 AJCC	Fuhrman	49/33	50/32	58/24	NA	60/22	82/0
Harada [20]	2009 AJCC	3 grade system	48/74	74/48	99/23	122/0	122/0	104/18
Pichler [21]	2002 AJCC	Fuhrman	99/1655	1064/690	1523/231	1732/32	1754/0	1754/0
Kroeger [22]	2002 AJCC	Fuhrman	475/2121	1496/1100	1614/982	1580/1016	1902/694	2078/518
da Costa [23]	2002 AJCC	Fuhrman	26/116	89/53	91/51	127/15	123/19	99/53
Takayama [24]	2009 AJCC	3 grade system	56/378	451/0	406/15	NA	NA	401/30
Komura [25]	2002 AJCC	Fuhrman	58/112	140/30	140/30	170/0	170/0	131/39
Suzuki [26]	1997 AJCC	3 grade system	32/179	166/45	187/24	211/0	211/0	211/0
Katz [27]	2002 AJCC	Fuhrman	92/749	575/194	589/252	NA	841/0	641/200
Kume [28]	2002 AJCC	Fuhrman	20/128	158/7	156/9	165/0	160/5	151/14
Kim [29]	2002 AJCC	Fuhrman	6/87	93/0	52/41	93/0	93/0	79/14
Rey [30]	2002 AJCC	Fuhrman	23/116	139/30	70/69	139/0	139/0	110/29
May [31]	NA	Fuhrman	70/701	642/129	531/240	758/13	771/0	605/166
Cho [32]	2002 AJCC	Fuhrman	24/275	299/0	253/46	299/0	299/0	299/0
Zubac [33]	2002 AJCC	Fuhrman	7/69	76/0	65/11	76/0	76/0	76/0
Pflanz [34]	2002 AJCC	Thoenes grade	53/554	515/92	432/75	NA	NA	479/128
Horiguchi [35]	2002 AJCC	3 grade system	50/70	93/27	105/15	117/3	103/17	112/8
Dall'Oglio [36]	NA	Fuhrman	59/171	164/86	145/84	216/14	NA	148/82
Klatte [37]	2002 AJCC	Fuhrman	22/497	519/0	414/92	519/0	519/0	409/110
Madbouly [38]	1997 AJCC	Fuhrman	8/40	45/3	41/7	48/0	48/0	43/5
Komai [39]	2002 AJCC	3 grade system	63/38	79/22	96/5	101/0	101/0	97/4
Ito [40]	1997 AJCC	3 grade system	78/100	127/51	162/16	165/13	150/28	140/38
Lee [41]	1997 AJCC	Fuhrman	26/456	382/103	264/221	NA	NA	419/66
Goncalves [42]	NA	Fuhrman	24/71	95/0	63/32	87/8	95/0	56/39
Lang [43]	2002 AJCC	Fuhrman	74/181	172/83	114/141	255/0	255/0	236/19
Ishimura [44]	1998 JUA	3 grade system	70/87	120/37	153/4	157/0	157/0	120/37
Griffiths [45]	NA	Fuhrman	24/152	NA	123/53	176/0	176/0	119/57
VanPonnel [46]	1987 AICC	Fuhrman	51/129	142/38	116/64	180/0	180/0	NA

 Table 1. Tumor characteristics of the eligible studies

AJCC: American Joint Committee on Cancer; ccRCC: clear cell renal cell carcinoma; non-ccRCC: non clear cell renal cell carcinoma; JUA: Japanese Urological Association.

cations for the same data from the same study group occurred, only the most informative and recent article was recruited into final analysis.

Data extraction

Two investigators (H.H. and P.X.W.) conducted the extraction process independently for the following information: (1) publication and methodology data including first author's surname, publication year, location of the study performed, inclusion and exclusion criteria, study design, period of recruitment, definition of survival, definition of MVI, slice staining methods, NO. of observers, interpretation of MVI, staging system, and nuclear grading system; (2) the baseline data including sample size, gender,

10781

age, follow-up period and treatment, MVI proportion, pathological TNM stage, nuclear grade, and histological subtypes; and (3) statistical data such as HRs and their 95% Cls. We preferred to gather multivariate analysis data. If they were not available, univariate analysis of survival outcomes was extracted instead. Discrepancies between the reviewers were resolved by a consensus meeting with three senior investigators (G.Y., Y.L., and C.X.G.) who made the final decision regarding inclusion or exclusion of the study.

End-points

The outcome measure was the recurrence-free survival (RFS), cancer-specific survival (CSS),

Table 2. Main characteristics of the eligible studies

Study	Year	Country	Recruitment period	No. of patients	Median FU, range (mon)	Study design	Inclusion and exclusion criteria	Definition of survival	Definition of MVI	Staining methods	No. of observers	Interpreta- tion of MVI	Quality scale
Belsante [14]	2014	USA	1997-2010	419	26 (0-150)	Retrospective	yes	yes	yes	NA	NA	NA	5
Eisenberg [15]	2013	USA	2001-2008	1103	78 (0-121)	Retrospective	yes	NA	yes	HE	1	blind	5
Shindo [16]	2013	Japan	1980-2005	172	104.5 (8-308)	Retrospective	yes	yes	NA	EVG	1	NA	4
Drewniak [17]	2013	Poland	2000-2007	148	51 (5-109)	Retrospective	NA	NA	NA	NA	1	NA	5
Steffens [18]	2013	Germany	1990-2011	2030	66 (30-96)	Retrospective	yes	yes	NA	NA	NA	NA	4
Betsunoh [19]	2013	Japan	1999-2012	82	46 (3-112)	Retrospective	yes	yes	NA	NA	2	NA	5
Harada [20]	2012	Japan	1998-2008	122	44 (8-148)	Retrospective	yes	yes	NA	HE/IHC	2	blind	7
Pichler [21]	2012	Austria	1984-2006	1754	82 (0-280)	Retrospective	yes	yes	yes	NA	NA	NA	5
Kroeger [22]	2012	Multination	1981-2009	2596	22.4 (1-212)	Retrospective	NA	yes	yes	NA	NA	NA	4
da Costa [23]	2012	Brazil	1992-2009	142	44	Retrospective	yes	yes	NA	HE/IHC	2	NA	6
Takayama [24]	2011	Japan	1978-2007	431	42.3	Retrospective	yes	yes	NA	NA	NA	NA	4
Komura [25]	2011	Japan	1996-2004	170	50 (28-84)	Retrospective	yes	yes	yes	NA	2	blind	7
Suzuki [26]	2011	Japan	1994-2001	211	81 (4-208)	Retrospective	yes	NA	NA	NA	NA	NA	3
Katz [27]	2011	USA	1989-2004	841	61 (1-209)	Retrospective	yes	yes	yes	HE	1	blind	6
Kume [28]	2010	Japan	1983-2009	165	30.7 (0.4-270.4)	Retrospective	yes	NA	NA	NA	NA	NA	3
Kim [29]	2010	Korea	1995-2004	93	63.6 (10-159)	Retrospective	NA	NA	NA	NA	1	NA	2
Rey [30]	2010	Spain	1993-2005	139	66.2±44.11	Retrospective	NA	NA	yes	NA	NA	NA	3
May [31]	2009	Germany	1992-2006	771	75.7	Retrospective	yes	yes	yes	HE	NA	NA	5
Cho [32]	2009	Japan	1986-2004	502	77.6 (0.4-246.9)	Retrospective	NA	yes	NA	NA	NA	NA	3
Zubac [33]	2008	Norway	1985-1994	76	112.8 (1-232.8)	Retrospective	yes	NA	yes	HE/IHC	2	blind	7
Pflanz [34]	2008	Germany	1992-2007	607	54	Retrospective	NA	yes	NA	HE	NA	NA	3
Horiguchi [35]	2007	Japan	1994-2006	120	24 (2-141)	Retrospective	NA	NA	yes	NA	NA	NA	3
Dall'Oglio [36]	2007	Brazil	1988-2003	230	48 (10-130)	Retrospective	NA	NA	yes	NA	1	NA	3
Klatte [37]	2007	USA	1985-2005	519	49 (1-199)	Retrospective	NA	yes	NA	NA	≥2	NA	4
Madbouly [38]	2007	Saudi Arabia	1990-2004	48	37.7 (12-60)	Retrospective	NA	NA	yes	NA	1	NA	3
Komai [39]	2007	Japan	1986-2004	101	55 (2-187)	Retrospective	NA	NA	NA	NA	NA	NA	2
lto [40]	2006	Japan	1985-2003	178	44.5 (1-232)	Retrospective	NA	yes	NA	NA	NA	NA	3
Lee [41]	2006	Korea	1993-2003	516	50.9 (1-148.6)	Retrospective	yes	yes	NA	NA	NA	NA	4
Goncalves [42]	2004	Brazil	1989-1999	95	45 (14-132)	Retrospective	yes	NA	yes	NA	1	NA	4
Lang [43]	2004	France	1980-1990	255	183	Retrospective	NA	NA	NA	HE/IHC	NA	NA	3
Ishimura [44]	2002	UK	1991-1996	176	44 (25-99)	Retrospective	yes	yes	yes	NA	2	blind	7
Griffiths [45]	2004	Japan	1986-2002	157	45 (6-162)	Retrospective	yes	NA	yes	HE	1	NA	4
VanPoppel [46]	1997	Belgium	1980-1993	180	60 (8-88)	Retrospective	yes	NA	yes	HE/PA/Elastin	NA	NA	4

EVG: Elastica van Gieson; HE: Haematoxylin and eosin; IHC: Immunohistochemistry; MVI: Microvascular invasion; PA: Periodic acid. NA: not available. FU: follow-up.

Study	Year	Country	No. of patients	Median age, range (yr)	Gender (m/f)	Adjuvant therapy (+/-)	RN/PN
Belsante [14]	2014	USA	419	57 (17-85)	247/172	NA	236/183
			333	55.9 (17-85)	194/139	NA	153/180
Eisenberg [15]	2013	USA	1103	62.3 (19-93)	710/393	NA	NA
Shindo [16]	2013	Japan	172	60 (23-82)	133/39	NA	107/65
Drewniak [17]	2013	Poland	148	59.6 (33-79)	102/46	NA	NA
Steffens [18]	2013	Germany	2030	62.3 (20-90)	1316/714	NA	1620/410
Betsunoh [19]	2013	Japan	82	63.1 (39-83)	62/20	22/60	82/0
Harada [20]	2012	Japan	122	65.0 (32-84)	87/35	NA	122/0
Pichler [21]	2012	Austria	1754	62.6 (20-89)	979/775	NA	NA
Kroeger [22]	2012	Multination	2596	61 (19-97)	1685/911	NA	NA
da Costa [23]	2012	Brazil	142	54.7 (23-81)	87/55	NA	100/42
Takayama [24]	2011	Japan	431	60.3 (15-81)	312/119	NA	377/53
Komura [25]	2011	Japan	170	62.4±11.4	114/56	49/121	153/17
Suzuki [26]	2011	Japan	211	59 (16-87)	152/59	90/121	173/38
Katz [27]	2011	USA	841	NA	530/311	NA	622/233
Kume [28]	2010	Japan	165	59 (23-83)	127/38	NA	81/81
Kim [29]	2010	Korea	93	55±11.4	64/29	NA	63/30
Rey [30]	2010	Spain	139	63±11.48	85/54	NA	127/12
May [31]	2009	Germany	771	61.1 (18-84)	488/283	NA	653118
Cho [32]	2009	Korea	299	56 (25-86)	195/104	NA	267/32
Zubac [33]	2008	Norway	76	67 (39-88)	36/40	NA	76/0
Pflanz [34]	2008	Germany	607	61.6 (18-84)	387/220	NA	490/117
Horiguchi [35]	2007	Japan	120	64 (36-81)	83/37	NA	NA
Dall'Oglio [36]	2007	Brazil	230	59 (9-90)	168/62	NA	180/47
Klatte [37]	2007	USA	519	61 (19-88)	320/199	NA	305/214
Madbouly [38]	2007	Saudi Arabia	48	50.7 (20-80)	22/26	NA	48/0
Komai [39]	2007	Japan	101	64 (33-84)	26/75	NA	NA
Ito [40]	2006	Japan	178	59.3±0.9	127/51	NA	NA
Lee [41]	2006	Korea	516	55 (26-81)	360/125	NA	NA
Goncalves [42]	2004	Brazil	95	60 (9-81)	72/23	NA	NA
Lang [43]	2004	France	255	60 (16-87)	169/86	NA	255/0
Ishimura [44]	2004	Japan	157	63.4 (20-84)	99/58	NA	140/17
Griffiths [45]	2002	UK	176	65 (34-88)	120/56	NA	176/0
VanPoppel [46]	1997	Belgium	180	52 (1-180)	107/73	NA	259/6

Table 3. Patient characteristics of the eligible studies

RN: Radical nephrectomy; PN: Partial nephrectomy.

metastasis-free survival (MFS), and overall survival (OS) between patients with or without MVI; the association between microvascular invasion and the clinical outcomes was statistically reflexed by use of hazard ratios (HR).

Quality assessments

Three investigators (G.Y., Y.L., and C.X.G.) evaluation for the enrolled studies according to a predefined form modified on the basis of Graeff's [10], knowing that no generally accepted criteria for the assessment of study quality are available at present. Our quality scale consists of nine criteria with 9 as the maximum score (<u>Table S2</u>). A study with a score \geq 5 was regarded as high quality, and low quality when the score was < 5.

Statistical analysis

Categorical features were gathered and arranged with frequency counts. Continuous data were summarized with medians and rang-

es; Comparisons of quality scales and MVI proportion between eligible studies were evaluated using spearman's rank correlations test by SPSS (Version 19; IBM Corp).

We gathered HRs and their 95% CI of each eligible study to conduct the meta-analysis. In case they were not directly provided, we estimated HRs and their 95% Cl using the available survival data by means of the accepted method [11]. A pooled HR and 95% CI were computed for the risk allele using Stata (Version 12.0; Stata Corp, College station, TX) by a randomeffects model to generate forest plots. If the 95% CI did not overlap with 1 and P < 0.05, the influence of MVI on clinical outcomes was identified as statistically significant. Heterogeneity was quantifiably assessed by use of the Higgins I squared statistic [12] and the Cochran's Q statistic [13]. The I² statistic yielded results ranging between 0 and 100% (0-25%, no heteroge-25-50%, moderate heterogeneity; neity; 50-75%, large heterogeneity; and 75-100%, extreme heterogeneity). P < 0.10 was deemed to stand for notable heterogeneity among studies. Publication bias was evaluated by use of egger's linear regression test and Begg's funnel plot.

In view of the heterogeneity between the studies, we conducted subgroup analyses. First we divided the studies into four groups, Group A with data of patients whose stages, grades, and tumor types were not separated, including all stages and all tumor types, ccRCC and nonccRCC (TanyNanyMany); Group B with data of all RCC types with the emphasis laid on nonmetastatic RCCs (TanyNOMO); Group C with data of all RCC types with the emphasis laid on organ-confined RCCs (T1-2N0M0); and Group D with data of ccRCC only (ccRCC). Then other potential sources of heterogeneity were explored, including publication year, median follow-up, and study location, number of patients, study quality score, and analytical results. When overlapping data appeared, we chose the more informative one.

Results

Study selection and characteristics

Initially, we assembled a total of 1120 articles from the electronic databases, of which 185 duplicate publications were excluded in the first round. Additional 860 articles were excluded after screening the titles and abstracts. Then, we reviewed full texts of the remaining 75 articles, of which 26 were excluded for lacking of sufficient data to estimate the HRs. 4 studies were excluded because they focused on vascular invasion without clear definition. 6 studies were excluded because MVI was assessed for RCC subtypes without including ccRCC, and 6 studies were excluded for the existence of reduplicative data with another study. Finally, 33 studies [14-46] that focused on the association between RCC and MVI were included for meta-analysis, involving a total of 14,946 patients, ranging from 48 to 2,596 per study. A flow diagram of the selection process is showed in Figure 1.

The main features of the 33 eligible studies for aggregation are listed in Tables 1-3. The publishing time of the studies was between 1997 and 2014. The 33 studies originated from Asia (14), Europe (9), the United States (4), multinational research (1) and other regions (5). The median follow-up duration ranged from 24 months to 183 months. Five of these studies included fewer than 100 patients, and 14 studies enrolled more than 200 patients, and 4 studies involved more than 1000 patients. All the included studies were based on the data of retrospective analysis of survival. Other characteristics including tumor features and pathologic outcomes are presented in Table 1. MVI was detected in 14.4% in pathological specimens of the 14,946 patients included in the meta-analysis. And higher frequencies of MVI were found to be associated with higher tumor grades and pathological T stages, distant metastasis and lymph node metastasis in the eligible studies (Table S3). And Eisenberg et al [15] reported a significant correlation between MVI and sarcomatoid differentiation coagulative tumor necrosis, and collecting system invasion. Goncalves et al reported positive association between MVI and perirenal fat invasion [43], in contrast Madbouly et al observed no significant correlation between MVI and perirenal fat invasion [38]. Of the 58 survival analyses, 56 (96.5%) directly provided HRs and their 95% CI for multivariate analysis, and 28 (48.3%) showed no significant correlation between MVI and survival. There was a wide variety of cofactors reported in the multivariate analysis of these studies, among which the most common

Meta-analysis of MVI in RCC



Figure 2. Forest plots of prognosis of microvascular invasion. The horizontal lines correspond to the study-specific hazard ration and 95%. A. CSS for all eligible studies. Cancer-specific survival. B. RFS for all eligible studies. Recurrence-free survival. C. MFS for all eligible studies. Metastasis-free survival. D. OS for all eligible studies. Overall survival.

Table 4. Summarized hazard ratios (including subgroup analysis)

Analysis	Ν	Pooled HR ^a (95% CI)	I ² value (%)	P-value*
CSS				
All studies	20	1.957 (1.498-2.556)	68.5%	0.0001
TanyNanyMany	7	1.435 (1.024-2.011)	74.3%	0.001
TanyN0M0	12	1.757 (1.340-2.304)	47.6%	0.033
T1-2N0M0	5	7.645 (3.647-16.025)	0.0%	0.776
ccRCC	5	1.954 (0.920-4.149)	58.5%	0.047
PY (1997-2009)	9	2.544 (1.720-3.763)	46.7%	0.059
PY (2009-2014)	11	1.594 (1.160-2.191)	65.2%	0.001
Median FU < 60 months	12	2.154 (1.440-3.222)	51.1%	0.021
Median FU \ge 60 months	8	1.821 (1.243-2.668)	81.2%	0.0001
Asian	7	2.326 (1.246-4.342)	37.6%	0.142
Other regions	13	1.886 (1.395-2.549)	76.0%	0.0001
No. of patients < 200	9	3.335 (1.929-5.765)	37.8%	0.117
No. of patients \geq 200	11	1.616 (1.213-2.153)	74.0%	0.0001
Quality scale < 5	12	1.855 (1.295-2.657)	71.2%	0.0001
Quality scale ≥ 5	8	2.196 (1.392-3.463)	67.5%	0.003
Not significant	11	1.195 (1.023-1.395)	0.0%	0.810
Significant	9	3.462 (2.375-5.044)	47.7%	0.054
RFS				
All studie	17	2.749 (1.974-3.828)	60.0%	0.001
TanyN0M0	12	2.278 (1.612-3.218)	58.1%	0.006
T1-2N0M0	6	4.365 (2.540-7.499)	0.0%	0.527
ccRCC	3	2.152 (1.349-3.431)	0.0%	0.469
PY (1997-2009)	9	3.182 (1.668-6.068)	77.6%	0.0001
PY (2009-2014)	8	2.391 (1.844-3.101)	0.0%	0.759
Median FU < 60 months	13	2.754 (1.782-4.257)	65.6%	0.0001
Median FU \ge 60 months	4	2.779 (1.743-4.432)	37.3%	0.188
Asion	8	2.476 (1.311-4.676)	70.6%	0.001
Other regions	9	3.005 (2.174-4.154)	33.7%	0.148
No. of patients < 200	11	3.019 (1.754-5.196)	69.5%	0.0001
No. of patients \geq 200	6	2.613 (1.862-3.666)	27.8%	0.226
Quality scale < 5	11	3.323 (1.905-5.798)	74.0%	0.0001
Quality scale ≥ 5	6	2.292 (1.728-3.042)	0.0%	0.960
Not significant	6	1.554 (0.963-2.510)	52.3%	0.063
Significant	11	3.330 (2.614-4.243)	20.4%	0.249
MFS				
All studies	6	1.621 (1.095-2.400)	75.8%	0.001
TanyNanyMany	2	1.259 (1.026-1.544)	0.0%	0.027
TanyN0M0	2	1.499 (0.683-3.292)	84.2%	0.012
T1-2N0M0	2	10.098 (0.500-203.84)	82.8%	0.016
ccRCC	3	1.409 (0.935-2.124)	79.7%	0.007
OS				
All studies	6	1.371 (0.978-1.923)	44.0%	0.112
TanyNanyMany	3	1.729 (1.248-2.397)	0.0%	0.399
TanyNOMO	3	1.545 (1.139-2.096)	45.1%	0.162

PY: publication year; FU: follow-ups; HR = hazard ratio; 95% CI = 95% confidence interval; a Pooled hazards ratios were obtained from using a DerSimonian-Laird random effects model, applying the inverse of variance as a weighing factor. **P* values obtained from χ^2 -test for heterogeneity. cofactor applied to evaluate the risk of poor survival was the histological grade (<u>Table S4</u>).

Assessment of study quality

The median quality score of the 33 included studies was 4 (mean: 4.13, range: 2-7) (Table 1). The score of 5 or more in methodological assessment indicates high quality, which included 12 (36.4%) studies. 16 of 33 studies present a definition of MVI (Table S5). No significant association was found between quality scores and study size (Spearman's r = 0.091, P = 0.605). Also we did not find statistical difference in quality score in accordance with location of the study performed, median follow-up time and publication year.

Meta-analysis

According to the conceivable heterogeneity between the studies, we used the random effects model to estimate the combined HR of each study. Figure 2 displays a forest plot of the individual HRs and pooled results from the metaanalysis. When we pooled 20 eligible studies into the meta-analysis for cancerspecific survival (CSS), there was a significant correlation between MVI and worse CSS, the pooled HR being 1.957 (95% Cl, 1.498-2.556), while the test of inconsistency (I^2 = 68.5%) failed to eliminate a notable heterogeneity (Figure 2A). The metaanalysis performed on 17

studies that evaluated the correlation between MVI and recurrence-free survival (RFS) showed that the pooled HR was 2.749 (95% CI, 1.974-3.828), despite the large heterogeneity between studies ($I^2 = 60.0\%$) (Figure 2B). Data on MFS was available in six studies, and metaanalysis of MFS suggested that MVI was linked with poor MFS with pooled HR = 1.621 (95% CI, 1.095-2.400). Cochrane Q test (Chi² = 20.63; P = 0.001) and I^2 = 75.8% showed a remarkable heterogeneity (Figure 2C). Six studies with data as regards overall survival (OS), the pooled HR from the meta-analysis suggested that the correlation between MVI and worse OS did not address statistical significance (pooled HR = 1.371; 95% CI, 0.978-1.923). And cochrane Q test (Chi² = 39.96; P = 0.001) with a moderate heterogeneity is shown in the data ($I^2 = 44.0\%$) (Figure 2D).

Assessment of heterogeneity

The meta-analysis of most subgroup again suggested MVI as a prognostic factor despite heterogeneity among some groups (Table 4). It should be noted that the combined HR of CSS and RFS in group C (T1-2NOMO) showed statistical significance (CSS: pooled HR = 7.645, 95% CI, 3.647-16.025, I² = 0.0%; RFS: pooled HR = 4.365, 95% CI, 2.540-7.499, I² = 0.0%) with no heterogeneity. However, the association between MVI and worse CSS in Group D (ccRCC) was statistically insignificant, with a pooled HR = 1.954 (95% CI, 0.920-4.149; P = 0.047 for heterogeneity test; $I^2 = 58.5\%$). Similar results were seen in the MFS where the pooled HR in Group B, C, D did not show statistical significance. And when we pooled the HRs of CSS in the studies with no significance, the pooled HR showed statistical significance instead (pooled HR = 1.195; 95% CI, 1.023-1.395; P = 0.810 for heterogeneity test; $I^2 = 0.0\%$). Moreover, compared with the no statistically significant pooled HR of OS in all eligible studies, The pooled HR of OS in Group A and B showed statistical significance (A: pooled HR = 1.729, 95% Cl, 1.248-2.397, l² = 0.0%; B: pooled HR = 1.545, 95% CI, 1.139-2.096, I² = 0.0%).

Publication bias

We used Begg's funnel plot to examine potential publication bias between the studies (**Figure 3**) and found no exact evidence of funnel plot asymmetry. Begg's test showed no evidence of statistical publication bias (all P > 0.05) between the studies in terms of HR of CSS, RFS, OS and MFS, the *p* values being 0.144, 0.064, 1.000 and 0.452, respectively. Egger's test confirmed the conclusion that no significant publication bias was found in the meta-analysis with respect to RFS, OS and MFS with a *p* value of 0.062, 0.964 and 0.174 respectively, except for CSS with a *p* value of 0.045.

Discussion

Microvascular invasion is defined as the presence of tumor within microscopic veins with a muscular coat, in spite of gross tumor in the renal vein [15], which most probably links with hematogenous spread of tumor cells. Cancer cells intrude into the lymphovascular space, highly proliferate, and then pierce the local vessels or lymphatics to disseminate more extensively [47, 48]. MVI has been identified as a risk factor of lymph node invasion, a recurrence of tumor and distant metastasis in many solid cancers including urothelial tumor [4, 49], lung cancer [5], and hepatocellular carcinoma [7] which has been confirmed in the systematic review studies. And in the liver and testiculars, MVI has been brought into the TNM staging system for improved cancer staging [50, 51]. However, only endometrial/cervical and head and neck cancers consider the presence of MVI as indication for further adjuvant therapy [5]. The prognostic value of MVI has been evaluated in numerous studies, but the results remain equivocal in RCC.

The present meta-analysis consisted of 14,946 RCC patients derived from 33 studies. The individual data were organized according to CSS, DFS, MFS and OS. No statistical difference in quality score was found between the location of the study performed, median follow-up time and publication year. MVI was detected in 14.4% of 14,946 RCC patients. We found a significant correlation between MVI and some acknowledged pathological parameters including pathologic TNM stage and grade.

Due to apparent heterogeneity of the enrolled studies, we used the random-effects model during pooling data. Meta-analysis of the eligible studies addressed a significant association between MVI and CSS, RFS, and MFS, suggesting that MVI is a significant predictor for poor



Figure 3. Begg's Funnel plots for publication bias test. Each point stands for a separate study for the indicated correlation. The horizontal lines represent the mean effects size. A. Beggs Funnel plots for CSS. Cancer-specific survival. B. Beggs Funnel plots for RFS. Recurrence-free survival. C. Beggs Funnel plots for MFS. Metas-tasis-free survival. D. Beggs Funnel plots for OS. Overall survival.

survival regarding cancer related events, but the presence of MVI did not seem to have an unfavorable impact on OS. Most of the subgroup analyses demonstrated similar results. wherein the combined analysis in group C (T1-2N0M0) revealed a significant association between MVI and CSS, RFS with no heterogeneity ($I^2 = 0\%$), which denoted that the presence of MVI predicts poorer prognosis in RCC patients on early pathological stage. However, there was no statistical significance in linking MVI with poor CSS for patients in Group D (ccRCC). When we combined the HRs of CSS in 10 individual studies with negative results, the pooled HR showed statistical significance with no heterogeneity ($I^2 = 0\%$), which further addresses that the prognostic valve of MVI for poor CSS. In addition, compared with the statistically insignificant pooled HR of OS in 6 eligible studies, the pooled HR of OS in Group A (TanyNanyMany) revealed statistical significance with no heterogeneity ($I^2 = 0\%$), suggesting that MVI might be a predictor for high risk of mortality. Notably, heterogeneities of data were detected in most of these subgroup analyses. Thus more studies with larger sample sizes of ccRCC patients or focusing on OS and MFS are needed to further estimate the impact of MVI on prognosis.

The results of the present study should be approached with caution in view of its merits and shortcomings. As a systematic review and meta-analysis, it possesses the power of adequate studies and large numbers of patients to provide more exact evaluation of effects and enable more authentic subgroup analyses. In addition, we found no publication bias using Begg's tests and egger's test for the analysis of association between MVI and RFS, MFS and OS, suggesting that this meta-analysis obtained from these studies approximate the actual results. However, with improved precision, there are several inherent limitations, specifically regarding the potential selection bias that results in heterogeneity between studies. Non-English studies, unpublished studies, and studies that did not provide sufficient data in HRs calculated did not contribute to evaluating of the predictive value of MVI for survival. The first defect is the presence of a slight publication bias of the eligible studies on the summary CSS, indicating the pooled HR may overestimate the true effect size. Another weakness of the present study is heterogeneity in term of different baseline characteristics of patients in each study. Although we take into account the heterogeneity in our meta-analysis by using the random-effects model, the conclusion drawn in this study should be considered prudently. In addition, all the included studies were retrospectively designed, and prospective multicenter trials are needed to seek more exact answers. Finally, although we included 33 studies comprising 14,946 cases for this metaanalysis, relatively few studies were categorized for subgroup analysis and several survival subgroup analyses were lacking in data.

Besides, only a few included studies incorporated immunohistochemical (IHC) analysis in cases negative for MVI by examination of H&E stained sections. The reason is that the use of IHC staining is not common for routine clinical practice. Knowing that this added measurement may increase the detection rate of MVI [27], rigorous morphological criteria should be established to standardize the diagnosis of MVI reproducibly, which is crucial for exerting its predictive value in daily clinical settings.

Conclusions

The results of the present meat-analysis show that estimates of the significance of MVI in RCC patients vary substantially between studies. Our meta-analysis indicates that the presence of MVI has a detrimental effect on survival and clinicopathological features in RCC and therefore could serve as an independent prognostic factor of CSS, RFS, and MFS. It could also be used to predict RCC patients who need further adjuvant therapies.

Acknowledgements

This work was supported by grants from the National Natural Science Foundation of China for Youths (No. 81001136 and 81202020), the National Natural Science Foundation of China (No. 30973006, 81170637), Shanghai Municipal Committee of Science and Technology General Program for Medicine (No. 11JC-1402302), the Key Project of Science and Innovation Foundation of Shanghai Ministry of Education (14zz084), and the Military Fund for Health Care (13BJZ29).

Disclosure of conflict of interest

None.

Address correspondence to: Dr. Dan-Feng Xu, Urology Research Center of The Chinese People's Liberation Army, Changzheng Hospital, Second Military Medical University, 415 Fengyang Road, Shanghai 200003, People's Republic of China. Tel: 86-21-81885732; E-mail: danfengxu_urology@163. com; Dr. Xin-Gang Cui, Department of Urinary Surgery of Changzheng Hospital, Second Military Medical University, 415 Fengyang Road, Shanghai 200003, People's Republic of China. Tel: 86-21-81885732; 86-21-81885736; Fax: 86-21-81885732; E-mail: cuixingang_urology@163.com

References

- [1] Siegel R, Ma J, Zou Z, Jemal A. Cancer statistics, 2014. CA Cancer J Clin 2014; 64: 9-29.
- [2] Janzen NK, Kim HL, Figlin RA, Belldegrun AS. Surveillance after radical or partial nephrectomy for localized renal cell carcinoma and management of recurrent disease. Urol Clin North Am 2003; 30: 843-852.
- [3] Gupta K, Miller JD, Li JZ, Russell MW, Charbonneau C. Epidemiologic and socioeconomic burden of metastatic renal cell carcinoma (mrcc): a literature review. Cancer Treat Rev 2008; 34: 193-205.
- [4] Kim H, Kim M, Kwak C, Kim HH, Ku JH. Prognostic significance of lymphovascular invasion in radical cystectomy on patients with bladder cancer: a systematic review and metaanalysis. PLoS One 2014; 9: e89259.
- [5] Mollberg NM, Bennette C, Howell E, Backhus L, Devine B, Ferguson MK. Lymphovascular invasion as a prognostic indicator in stage i nonsmall cell lung cancer: a systematic review and meta-analysis. Ann Thorac Surg 2014; 97: 965-971.
- [6] Heinzelbecker J, Gross-Weege M, Weiss C, Horner C, Trunk MJ, Erben P, Haecker A, Bolenz C. Microvascular invasion of testicular nonseminomatous germ cell tumors: implications of separate evaluation of lymphatic and blood vessels. J Urol 2014; 192: 593-9.
- [7] Rodriguez-Peralvarez M, Luong TV, Andreana L, Meyer T, Dhillon AP, Burroughs AK. A systematic review of microvascular invasion in hepatocellular carcinoma: diagnostic and prognostic variability. Ann Surg Oncol 2013; 20: 325-339.
- [8] Grimm M. Prognostic value of clinicopathological parameters and outcome in 484 patients with oral squamous cell carcinoma: microvascular invasion (v+) is an independent prognostic factor for oscc. Clin Transl Oncol 2012; 14: 870-880.
- [9] Moher D, Liberati A, Tetzlaff J, Altman DG; PRISMA Group. Preferred reporting items for

systematic reviews and meta-analyses: the PRISMA statement. Int J Surg 2010; 8: 336-41.

- [10] de Graeff P, Crijns AP, de Jong S, Boezen M, Post WJ, de Vries EG, van der Zee AG, de Bock GH. Modest effect of p53, egfr and her-2/neu on prognosis in epithelial ovarian cancer: a meta-analysis. Br J Cancer 2009; 101: 149-159.
- [11] Tierney JF, Stewart LA, Ghersi D, Burdett S, Sydes MR. Practical methods for incorporating summary time-to-event data into meta-analysis. Trials 2007; 8: 16.
- [12] Higgins JP, Thompson SG. Quantifying heterogeneity in a meta-analysis. Stat Med 2002; 21: 1539-1558.
- [13] Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. BMJ 2003; 327: 557-560.
- [14] Belsante M, Darwish O, Youssef R, Bagrodia A, Kapur P, Sagalowsky Al, Lotan Y, Margulis V. Lymphovascular invasion in clear cell renal cell carcinoma--association with disease-free and cancer-specific survival. Urol Oncol 2014; 32: 23-30.
- [15] Eisenberg MS, Cheville JC, Thompson RH, Kaushik D, Lohse CM, Boorjian SA, Costello BA, Leibovich BC. Association of microvascular and capillary-lymphatic invasion with outcome in patients with renal cell carcinoma. J Urol 2013; 190: 37-43.
- [16] Shindo T, Masumori N, Kobayashi K, Fukuta F, Hirobe M, Tonooka A, Hasegawa T, Kitamura H, Tsukamoto T. Long-term outcome of small, organ-confined renal cell carcinoma (rcc) is not always favourable. BJU Int 2013; 111: 941-945.
- [17] Drewniak T, Sandheim M, Jakubowski J, Juszczak K, Stelmach AW. Prognostic factors of overall survival in renal cancer patients-single oncological center study. Cent European J Urol 2013; 66: 283-291.
- [18] Steffens S, Ringe KI, Schroeer K, Lehmann R, Rustemeier J, Wegener G, Schrader M, Hofmann R, Kuczyk MA, Schrader AJ. Does overweight influence the prognosis of renal cell carcinoma? Results of a multicenter study. Int J Urol 2013; 20: 585-592.
- [19] Betsunoh H, Fukuda T, Anzai N, Nishihara D, Mizuno T, Yuki H, Masuda A, Yamaguchi Y, Abe H, Yashi M, Fukabori Y, Yoshida K, Kamai T. Increased expression of system large amino acid transporter (lat)-1 mrna is associated with invasive potential and unfavorable prognosis of human clear cell renal cell carcinoma. BMC Cancer 2013; 13: 509.
- [20] Harada K, Miyake H, Kusuda Y, Fujisawa M. Expression of epithelial-mesenchymal transition markers in renal cell carcinoma: impact on prognostic outcomes in patients undergoing

radical nephrectomy. BJU Int 2012; 110: E1131-E1137.

- [21] Pichler M, Hutterer GC, Chromecki TF, Jesche J, Groselj-Strele A, Kampel-Kettner K, Pummer K, Zigeuner R. Prognostic value of the leibovich prognosis score supplemented by vascular invasion for clear cell renal cell carcinoma. J Urol 2012; 187: 834-839.
- [22] Kroeger N, Rampersaud EN, Patard J, Klatte T, Birkhaeuser FD, Shariat SF, Lang H, Rioux-Leclerq N, Remzi M, Zomorodian N, Kabbinavar FF, Belldegrun AS, Pantuck AJ. Prognostic value of microvascular invasion in predicting the cancer specific survival and risk of metastatic disease in renal cell carcinoma: a multicenter investigation. J Urol 2012; 187: 418-423.
- [23] da Costa WH, Rocha RM, da Cunha IW, da Fonseca FP, Guimaraes GC, Zequi SD. Cd133 immunohistochemical expression predicts progression and cancer-related death in renal cell carcinoma. World J Urol 2012; 30: 553-558.
- [24] Takayama T, Sugiyama T, Kai F, Suzuki T, Nagata M, Imanishi T, Mizuno T, Sato S, Furuse H, Mugiya S, Ozono S. Characteristics of aggressive variants in t1a renal cell carcinoma. J Cancer Res Clin 2011; 137: 1653-1659.
- [25] Komura K, Inamoto T, Black PC, Koyama K, Katsuoka Y, Watsuji T, Azuma H. Prognostic significance of body mass index in asian patients with localized renal cell carcinoma. Nutr Cancer 2011; 63: 908-915.
- [26] Suzuki K, Nishiyama T, Hara N, Akazawa K, Takahashi K. Kattan postoperative nomogram for renal cell carcinoma: predictive accuracy in a japanese population. Int J Urol 2011; 18: 194-199.
- [27] Katz MD, Serrano MF, Humphrey PA, Grubb RLI, Skolarus TA, Gao F, Kibel AS. The role of lymphovascular space invasion in renal cell carcinoma as a prognostic marker of survival after curative resection. Urol Oncol 2011; 29: 738-744.
- [28] Kume H, Suzuki M, Fujimura T, Fukuhara H, Enomoto Y, Nishimatsu H, Homma Y. Distant metastasis of renal cell carcinoma with a diameter of 3 cm or less-which is aggressive cancer? J Urol 2010; 184: 64-68.
- [29] Kim JM, Song PH, Kim HT, Park TC. The prognostic factors for patients with pt1a renal cell carcinoma. Korean J Urol 2010; 51: 233-238.
- [30] Rey Rey J, Leon Ramirez D, Lopez Garcia S, Fernandez Vazquez P, Benavente Delgado J, Ojea Calvo A. Pathological prognostic indicators in renal cell carcinoma. Actas Urol Esp 2010; 34: 71-77.
- [31] May M, Brookman-Amissah S, Kendel F, Knoll N, Roigas J, Hoschke B, Miller K, Gilfrich C, Pflanz S, Gralla O. Validation of a postoperative prognostic model consisting of tumor microvascular invasion, size, and grade to predict

disease-free and cancer-specific survival of patients with surgically resected renal cell carcinoma. Int J Urol 2009; 16: 616-621.

- [32] Cho HJ, Kim SJ, Ha US, Hong SH, Kim JC, Choi YJ, Hwang TK. Prognostic value of capsular invasion for localized clear-cell renal cell carcinoma. Eur Urol 2009; 56: 1006-1012.
- [33] Zubac DP, Bostad L, Kihl B, Eide J, Wentzel-Larsen T, Haukaas A. Organ-confined clear cell renal cell carcinoma: the prognostic impact of microvascular invasion, nuclear grade and tumour size. Apmis 2008; 116: 1027-1033.
- [34] Horiguchi A, Asano T, Asano T, Ito K, Sumitomo M, Hayakawa M. Fatty acid synthase over expression is an indicator of tumor aggressiveness and poor prognosis in renal cell carcinoma. J Urol 2008; 180: 1137-1140.
- [35] Pflanz S, Brookman-Amissah S, Roigas J, Kendel F, Hoschke B, May M. Impact of macroscopic tumour necrosis to predict survival of patients with surgically resected renal cell carcinoma. Scand J Urol Nephrol 2008; 42: 507-513.
- [36] Dall'Oglio MF, Antunes AA, Sarkis AS, Crippa A, Leite KR, Lucon AM, Srougi M. Microvascular tumour invasion in renal cell carcinoma: the most important prognostic factor. BJU Int 2007; 100: 552-555.
- [37] Klatte T, Chung J, Leppert JT, Lam JS, Pantuck AJ, Figlin RA, Belldegrun AS. Prognostic relevance of capsular involvement and collecting system invasion in stage I and II renal cell carcinoma. BJU Int 2007; 99: 821-824.
- [38] Madbouly K, Al-Qahtani SM, Ghazwani Y, Al-Shaibani S, Mansi MK. Microvascular tumor invasion: prognostic significance in low-stage renal cell carcinoma. Urology 2007; 69: 670-674.
- [39] Komai Y, Saito K, Sakai K, Morimoto S. Increased preoperative serum c-reactive protein level predicts a poor prognosis in patients with localized renal cell carcinoma. BJU Int 2007; 99: 77-80.
- [40] Ito K, Asano T, Yoshii H, Satoh A, Sumitomo M, Hayakawa M. Impact of thrombocytosis and creactive protein elevation on the prognosis for patients with renal cell carcinoma. Int J Urol 2006; 13: 1365-1370.
- [41] Lee SE, Byun S, Oh JK, Lee SC, Chang IH, Choe G, Hong SK. Significance of macroscopic tumor necrosis as a prognostic indicator for renal cell carcinoma. J Urology 2006; 176: 1332-1337.
- [42] Lang H, Lindner V, Letourneux H, Martin M, Saussine C, Jacqmin D. Prognostic value of microscopic venous invasion in renal cell carcinoma: long-term follow-up. Eur Urol 2004; 46: 331-335.
- [43] Goncalves PD, Srougi M, Dall'Oglio MF, Leite K, Ortiz V, Hering F. Low clinical stage renal cell carcinoma: relevance of microvascular tumor

invasion as a prognostic parameter. J Urol 2004; 172: 470-474.

- [44] Ishimura T, Sakai I, Hara I, Eto H, Miyake H. Microscopic venous invasion in renal cell carcinoma as a predictor of recurrence after radical surgery. Int J Urol 2004; 11: 264-268.
- [45] Griffiths D, Verghese A, Golash A, Kynaston HG, Matthews PN, Hart A, Court JB. Contribution of grade, vascular invasion and age to outcome in clinically localized renal cell carcinoma. BJU Int 2002; 90: 26-31.
- [46] VanPoppel H, Vandendriessche H, Boel K, Mertens V, Goethuys H, Haustermans K, VanDamme B, Baert L. Microscopic vascular invasion is the most relevant prognosticator after radical nephrectomy for clinically nonmetastatic renal cell carcinoma. J Urol 1997; 158: 45-49.
- [47] Alitalo K, Mohla S, Ruoslahti E. Lymphangiogenesis and cancer: meeting report. Cancer Res 2004; 64: 9225-9229.
- [48] Padera TP, Kadambi A, di Tomaso E, Carreira CM, Brown EB, Boucher Y, Choi NC, Mathisen D, Wain J, Mark EJ, Munn LL, Jain RK. Lymphatic metastasis in the absence of functional intratumor lymphatics. Science 2002; 296: 1883-1886.

- [49] Ku JH, Byun SS, Jeong H, Kwak C, Kim HH, Lee SE. Lymphovascular invasion as a prognostic factor in the upper urinary tract urothelial carcinoma: a systematic review and meta-analysis. Eur J Cancer 2013; 49: 2665-2680.
- [50] Albers P, Siener R, Kliesch S, Weissbach L, Krege S, Sparwasser C, Schulze H, Heidenreich A, de Riese W, Loy V, Bierhoff E, Wittekind C, Fimmers R, Hartmann M. Risk factors for relapse in clinical stage i nonseminomatous testicular germ cell tumors: results of the german testicular cancer study group trial. J Clin Oncol 2003; 21: 1505-1512.
- [51] Vauthey JN, Lauwers GY, Esnaola NF, Do KA, Belghiti J, Mirza N, Curley SA, Ellis LM, Regimbeau JM, Rashid A, Cleary KR, Nagorney DM. Simplified staging for hepatocellular carcinoma. J Clin Oncol 2002; 20: 1527-1536.

Table S1. Searching strategies and results

Database	Date	Search strategy	Results
Pubmed	Up to May 2014	#1 renal or kidney [all fields]	846
		#2 cancer or carcinoma or neoplasm or tumor or mass or tumour [all fields]	
		#3 Predict* or prognos* or survival or risk or outcome [all fields]	
		#4 lymphovascular or Lymphatic or microvascular or microvessel or microvenous or microscopic or vascular [all fields]	
		#5 invasion or infiltration [all fields]	
		#1 AND #2 AND #3 AND #4 AND #5	
Web of science	Up to May 2014	#1 renal or kidney [topic]	881
		#2 cancer or carcinoma or neoplasm or tumor or mass or tumour [topic]	
		#3 Predict* or prognos* or survival or risk or outcome [topic]	
		#4 lymphovascular or Lymphatic or microvascular or microvessel or microvenous or microscopic or vascular [topic]	
		#5 invasion or infiltration [topic]	
		#1 AND #2 AND #3 AND #4 AND #5	
Cochrane library	Up to May 2014	#1 renal or kidney [title, abstract, key words]	6
		#2 cancer or carcinoma or neoplasm or tumor or mass or tumour [title, abstract, key words]	
		#3 Predict* or prognos* or survival or risk or outcome [title, abstract, key words]	
		#4 lymphovascular or Lymphatic or microvascular or microvessel or microvenous or microscopic or vascular [title, abstract, key words]	
		#5 invasion or infiltration [title, abstract, key words]	
		#1 AND #2 AND #3 AND #4 AND #5	

Table S2. Criteria for quality assessment

Criterium	Points
1. Is the population under study defined with in- and exclusion criteria?	1
2. Were patient data prospectively collected?	1
3. Are the main prognostic patient and tumour characteristics presented? ^a	1
4. Were the kinds of staining method > 1?	1
5. was the definition of MVI available in the studies?	1
6. Were stainings evaluated by > 1 observer?	1
7. Were the staining results interpreted by pathologists who were blind to the clinicopathological data?	1
8. Is the study endpoint defined?	1
9. Is the time of follow up specified?	1
Max	9

^a: At least four of the following characteristics: age at diagnosis, tumour stage, tumour grade, tumour type.

Study	Tumor feature	MVI (+/-)		Stage		Fuhrman grade			Regional lymph node involvement		Distant metastases			Histologic subtype					PFI		
			pT1-2	pT3-4	Р	1-2	3-4	Р	pNx/N0	pN1	Р	MO	M1	Р	CC	р	С	cd	S	u	
Belsante [14]	ccRCCpTan- yNanyM0	60/359	21/ 312	39/47	< 0.001	20/ 268	40/91	< 0.001	NA	NA		NA	NA		NA	NA	NA	NA	NA	NA	NA
Eisenberg [15]	ccRCCpTan- yNanyMany	119/954	19/ 694	100/ 289	< 0.001	11/ 458	108/ 526	< 0.001	94/ 922	62/25	< 0.001	95/ 876	24/ 108	0.004	119/ 984	5/ 214	1/85	1/45	NA	3/ 17	NA
Kroeger [22]	pTanyNanyMany	475/2121	97/ 1399	378/ 722	< 0.001	170/ 1444	305/ 677	< 0.001	387/ 1193	42/58	< 0.001	192/ 1710	283/ 411	< 0.001	412/ 1666	47/ 309	14/ 135	2/11	NA	NA	NA
Katz [27]	pTanyNanyMO	91/750	23/ 552	68/ 126	< 0.001	274/ 562	65/ 187	0.036	NA	NA		NA	NA		67/ 574	6/ 127	2/26	NA	NA	17/ 20	NA
Rey [30]	pTanyNOMO	23/153	11/ 135	12/18	< 0.001	4/66	19/50	< 0.001	NA	NA		NA	NA		NA	NA	NA	NA	NA	NA	NA
Dall'Oglio [37]	pTanyNanyMany	59/171	26/ 138	33/33	< 0.001	12/ 134	47/37	< 0.001	NA	NA		NA	NA		39/ 109	8/37	2/21	NA	9/4	NA	NA
Madbouly [39]	pT1-2NOMO	8/40	6/39	2/1	< 0.001	6/35	2/5	< 0.001	NA	NA		NA	NA		8/35	0/1	0/4	NA	NA	NA	2/1
Goncalves [43]	pT1-2NanyM0	24/71	24/71	NA		3/60	21/11	< 0.001	16/71	8/0	< 0.001	NA	NA		10/46	NA	2/8	NA	7/1	NA	14/4
Ishimura [45]	pTanyNOMO	70/87	36/84	34/3	< 0.001	65/83	2/2	< 0.001	NA	NA		NA	NA		59/61	4/4	5/10	0/1	NA	NA	NA
Lang [44]	pTanyN0M0	74/181	NA	NA		13/101	61/80	< 0.001	NA	NA		NA	NA		NA	NA	NA	NA	NA	NA	NA
VanPoppel [47]	pTanyNOMO	51/129	31/ 111	20/39	< 0.001	25/90	26/38	< 0.001	NA	NA		NA	NA		NA	NA	NA	NA	NA	NA	NA

Table S3. MVI proportion in the eligible studies

cc: clear cell; p: papillary; c: chromophobe; cd: collecting duct; s: Sarcomatous; u: unclassified; PFI: perirenal fat invasion; NA: not available.

Table S4. Estimation of the hazard ratio

Otherstein	Tumor	Survivol		95% CI					
Study	charateristics	Survival	HK	lower	upper				
Belsante [14]	ccRCCTanyNanyM0	CSS	2.7	0.9	8.4	MVI, tumor stage, grade	not		
Belsante [14]	ccRCCTanyNanyM0	DFS	1.70	0.77	3.60	MVI, tumor stage, grade	not		
Belsante [14]	ccRCCpT1-2N0M0	CSS	12.7	1.7	92.7	MVI, tumor stage, grade	Significant		
Belsante [14]	ccRCCpT1-2N0M0	DFS	4.00	1.20	13.7	MVI, tumor stage, grade	Significant		
Eisenberg [15]	ccRCCpTanyNanyMany	MFS	1.20	0.82	1.60	primary tumor, regional lymph node, distant metastases classifications, tumor size, nuclear grade, coagulative tumor necrosis, MVI	not		
Eisenberg [15]	ccRCCpTanyNanyMany	CSS	1.30	0.95	1.80	primary tumor, regional lymph node, and distant metastases classifications, tumor size, nuclear grade, coagulative tumor necrosis, MVI	not		
Shindo [16]	pT1aN0M0	CSS	8.191	1.808	37.098	interferon, histologic subtype, grade, tumor size, MVI	Significant		
Drewniak [17]	pTanyNanyMany	OS	1.628	1.049	2.525	performance status, smoking history, hemoglobin concentration, AJCC anatomical staging, tumor grade, and presence of microvascular invasion	Significant		
Steffens [18]	pTanyNanyMany	CSS	1.08	0.84	1.38	age and sex, tumor stage, differentiation grade, histopathological subtype, lymphogenous/visceral metastasis, and MVI	not		
Betsunoh [19]	ccRCCpTanyNanyMany	OS	3.222	1.231	8.434	histological grade, pT stage, microscopic vascular invasion, metastasis, and LAT1 mRNA expression	Significant		
Harada [20]	pTanyNOMO	RFS	2.680	1.200	5.987	Mode of presentation, Karnofsky performance status, C-reactive protein level, Pathological stage, Grade, MVI, Histological subtype, E-cadherin, N-cadherin, β -catenin, γ -catenin, Clusterin, Slug, Snail, Twist, Vimentin, ZEB1, ZEB2	Significant		
Pichler [21]	ccRCCpTanyN0M0	10y MFS	2.19	1.54	3.12	T stage, N stage, grade, tumor size, tumor necrosis, MVI	Significant		
Kroeger [22]	pTanyNanyMany	MFS	1.295	1.000	1.677	gender, ECOG PS, Fuhrman grade, size, pT stage, N stage, M stage, MAVI, MVI	Significant		
Kroeger [22]	ccRCCpTanyNanyMany	MFS	1.097	0.832	1.446	gender, ECOG PS, Fuhrman grade, size, pT stage, N stage, M stage, MAVI, MVI	not		
Kroeger [22]	pTanyNanyM0	CSS	1.061	0.686	1.642	gender, ECOG PS, Fuhrman grade, size, pT stage, N stage, M stage, MAVI, MVI	not		
Kroeger [22]	pTanyNanyMany	CSS	0.901	0.730	1.113	gender, ECOG PS, Fuhrman grade, size, pT stage, N stage, M stage, MAVI, MVI	not		
Kroeger [22]	ccRCCpTanyNanyM0	CSS	0.548	0.043	6.980	gender, ECOG PS, Fuhrman grade, size, pT stage, N stage, M stage, MAVI, MVI	not		
da Costa [23]	pTanyNOMO	5y PFS	2.387	1.023	5.570	T stage, tumor size, grade, MVI, necrosis, metastasis, ECOG PS, CD133	Significant		
da Costa [23]	pTanyNOMO	5y CSS	2.382	1.219	4.653	T stage, tumor size, grade, MVI, necrosis, metastasis, ECOG PS, CD133	Significant		
Takayama [24]	pT1aNanyMany	OS	2.058	0.244	1.244	symptom, CRP, size, histological grade, sarcomatoid component, MVI	not		
Takayama [24]	pT1aNanyMany	MFS	2.636	1.111	6.253	age, sex, side, symptom, CRP, size, histological grade, nephrectomy, histology, sarcomatoid component, MVI	Significant		
Komura [25]	pTanyN0M0	OS	1.248	0.363	4.292	mode of presentation, BMI, ECOG performance status, serum level of C-reactive protein (CRP), microvascular invasion, pathological stage, histologic subtype, nuclear grade, adjuvant cytokine therapy, MVI	not		
Komura [25]	pTanyN0M0	CSS	13.845	1.264	151.63	mode of presentation, BMI, ECOG performance status, serum level of C-reactive protein (CRP), microvascular invasion, pathological stage, histologic subtype, nuclear grade, adjuvant cytokine therapy, MVI	Significant		
Komura [25]	pTanyN0M0	RFS	2.978	1.007	8.809	mode of presentation, BMI, ECOG performance status, serum level of C-reactive protein (CRP), microvascular invasion, pathological stage, histologic subtype, nuclear grade, adjuvant cytokine therapy, MVI	Significant		
Suzuki [26]	ccRCCpTanyN0M0	5y RFS	1.913	0.879	4.163	age, sex, symptoms, T stage, tumor size, grade, MVI	not		
Katz [27]	pTanyNanyMO	MFS	0.98	0.58	1.63	LVI, Fuhrman grade, stage, subtype, tumor, size	not		
Katz [27]	pTanyNanyMO	DSS	0.96	0.54	1.69	LVI, Fuhrman grade, stage, subtype, tumor, size	not		
Katz [27]	pTanyNanyMO	OS	1.24	0.87	1.77	LVI, Fuhrman grade, stage, subtype, tumor, size	not		
Kume [28]	pT1aN0M0	MFS	58.121	5.47	617.27	age, sex, side, symptom, size, histology, grade, treatment, sarcomatoid component, MVI	Significant		
Kim [29]	pT1aN0M0	RFS	17.947	1.261	255.376	Fuhrman's nuclear grade, MVI, necrosis in tumor	Significant		

Meta-analysis of MVI in RCC

Rey [30]	pTanyNOMO	CSS	1.225	0.32	4.691	stage, fuhrman's grade, tumor size, necrosis, MVI, sinus invasion	not
May [31]	pTanyNanyMO	DFS	2.33	1.56	3.47	grade, MVI, tumor size	Significant
May [31]	pTanyNanyMO	CSS	2.74	1.75	4.29	grade, MVI, tumor size	Significant
Cho [32]	ccRCCpTanyN0M0	RFS	3.444	1.411	8.409	univariate analysis	Significant
Cho [32]	ccRCCpTanyNOMO	CSS	1.091	0.254	4.673	univariate analysis	not
Zubac [33]	ccRCCpT1-2N0M0	CSS	7.17	2.25	22.78	tumor size, nuclear grade, MVI, age	Significant
Pflanz [34]	pTanyNanyMany	CSS	1.762	0.867	3.578	T stage, N stage, M stage, thoenes grade, tumor size, tumor necrosis, MVI, platelet count	not
Pflanz [34]	pTanyNanyMany	OS	1.541	0.875	2.714	T stage, N stage, M stage, thoenes grade, tumor size, tumor necrosis, MVI, platelet count	not
Horiguchi [35]	pTanyNanyMany	CSS	5.415	1.002	29.259	stage, grade, regional lymph node metastasis, distant metastasis, MVI, FAS expression	Significant
Dall'Oglio [36]	pTanyNanyMany	CSS	5.169	2.275	11.75	Presentation, tumor size, MVI	Significant
Dall'Oglio [36]	pTanyNanyMany	DFS	5.733	2.753	11.94	Presentation, grade, MVI	Significant
Klatte [37]	pT1-2NOMO	RFS	2.37	0.93	6.05	tumour size and the categorical variables capsular involvement, collecting-system invasion, microvascular invasion, ECOG PS, Fuhrman grade, and histological type	not
Madbouly [38]	pT1-2N0M0	DFS	10.34	1.891	56.54	MVI, stage, grade, cell type, perirenal fat invasion, tumor size, and patient age	Significant
Komai [39]	pTanyNOMO	DSS	1.47	0.64	3.45	CRP, anaemia, pT stage, grade, histological cell type, MVI	not
Komai [39]	pTanyNOMO	RFS	0.62	0.33	1.11	CRP, anaemia, pT stage, grade, histological cell type, MVI	not
lto [40]	pTanyNOMO	DFS	7.353	2.227	24.39	age, sex, tumor side, T stage, tumor size, grade, MVI, THC (thrombocytosis), CRP (C-reactive protein)	Significant
Lee [41]	pTanyNanyMany	CSS	1.28	0.57	2.86	presence of metastasis, T stage, grade, MVI, tumor size, sarcomatoid differentiation, tumor necrosis	not
Lee [41]	pTanyNanyMO	CSS	1.67	0.91	3.05	T stage, grade, MVI, tumor size, sarcomatoid differentiation, tumor necrosis	not
Goncalves [42]	pT1-2NanyMO	RFS	6.78	1.80	25.53	MVI, perirenal fat infiltration, nuclear grade, tumor diameter, macrovascular involvement, lymph node metastasis, sarcomatous elements	Significant
Goncalves [42]	pT1-2NanyM0	CSS	15.72	1.57	157.68	MVI, perirenal fat infiltration, nuclear grade, tumor diameter, macrovascular involvement, lymph node metastasis, sarcomatous elements	Significant
Lang [43]	pTanyNOMO	CSS	2.16	1.69	2.71	tumor size, nuclear grade, MVI, age, sex	Significant
Lang [43]	pTanyNOM0	OS	1.82	1.5	2.22	tumor size, nuclear grade, MVI, age, sex	Significant
Ishimura [44]	pTanyNOMO	RFS	1.965	0.632	6.116	age, sex, tumor size, pathological stage, grade, histological type, MVI	not
Ishimura [44]	pTanyNOMO	CSS	2.432	0.528	11.201	age, sex, tumor size, pathological stage, grade, histological type, MVI	not
Ishimura [44]	pT1-2N0M0	RFS	4.41	1.115	16.918	age, sex, tumor size, pathological stage, grade, histological type, MVI	Significant
Ishimura [44]	pT1-2N0M0	CSS	2.284	0.248	21.056	age, sex, tumor size, pathological stage, grade, histological type, MVI	not
Griffiths [45]	pTanyNOMO	DFS	2.15	0.98	4.72	MVI, inferior vena cava invasion, renal vein invasion, grade, age, tumor size, histological type	not
VanPoppel [46]	pTanyNOM0	DFS	4.19	2.11	8.30	age, stage, grade, size, MVI	Significant

DSS: disease special survival; CSS: cancer special survival; RFS: recurrence free survival; DFS: disease free survival; PFS: progression free survival; MFS: metastasis free survival; OSS: overall survival; MVI: microvascular invasion/microscopic vascular invasion; MAVI: macrovascular invasion; LVI: lymphovascular invasion; ECOG PS: Eastern Cooperative Oncology Group performance status.

Meta-analysis of MVI in RCC

Table S5. The definition of MVI in the eligible studies

Study	Year	Research object	Definition
Belsante [14]	2014	Lymphovascular invasion	LVI was defined as the presence of the invasion of cancer cells into blood vessels or the lymphatic system (excluding the renal vein and its muscle containing segmental branches and the inferior vena cava).
Eisenberg [15]	2013	Microvascular invasion/Capil- lary-Lymphatic Invasion	MVI was defined as the presence of tumor within microscopic veins or venules with a muscular coat, regardless of gross tumor in the renal vein. The term CLI was specifically used to define the presence of tumor within a microscopic capillary or lymphatic channel, vessels lacking a muscular coat.
Shindo [16]	2013	Microvascular invasion	NA
Drewniak [17]	2013	Microvascular invasion	NA
Steffens [18]	2013	Microscopic vascular invasion	NA
Betsunoh [19]	2013	Microscopic vascular invasion	NA
Harada [20]	2012	Microvascular invasion	NA
Pichler [21]	2012	Microscopic vascular inva- sion/Macroscopic vascular invasion	MVI was defined as microscopic detection of neoplastic cells invading the vessel wall or neoplastic emboli in the vessel lumen.
Kroeger [22]	2012	Microvascular invasion	MVI was defined as invasion of neoplastic cells in microscopic vessels or tumor emboli in the intratumor microscopic vessels.
da Costa [23]	2012	Microvascular invasion	NA
Takayama [24]	2011	Microvascular invasion	NA
Komura [25]	2011	Microvascular invasion	MVI was defined as tumor cells in an endothelium-lined space by routine light microscopy in whole-mounted RCC specimens.
Suzuki [26]	2011	Microvascular invasion	NA
Katz [27]	2011	Lymphovascular invasion	LVI was considered present if any tumor cells were seen with in the luminal space lined by endothelial cells by visual inspection on H and E stained slides from each tumor.
Kume [28]	2010	Microvascular invasion	NA
Kim [29]	2010	Microvascular invasion	NA
Rey [30]	2010	Microscopic vascular invasion	MVI was defined as the presence of tumor cells in the vascular lumen of the analyzed specimens, which also includes lymphatic vessel invasion.
May [31]	2009	Microvascular invasion	MVI was defined as the unequivocal presence of tumor cells in an endothelium lined space without underlying muscular walls.
Cho [32]	2009	Microvascular invasion	NA
Zubac [33]	2008	Microvascular invasion	MVI was diagnosed only when tumour cell aggregates were seen within lumina covered with CD31-positive cells or when tumour cells penetrated a ves- sel wall. A group of tumour cells invaginated a vessel wall without real invasion. Both sinusoidal and muscular vessels within and close to the tumour were assessed.
Pflanz [34]	2008	Microvascular invasion	NA
Horiguchi [35]	2007	Microvascular invasion	MVI was considered to have occurred when routine pathological examination revealed neoplastic cells in at least 1 endothelium lined space.
Dall'Oglio [36]	2007	Microvascular invasion	The presence of MVI was evaluated and defined as positive when there were neoplastic cells in an endothelium-lined space and/or in the intratumoral microcirculation of the tunica media.
Klatte [37]	2007	Microvascular invasion	NA
Madbouly [38]	2007	Microvascular tumor invasion	MVI was defined as the microscopic detection of neoplastic cells invading the vessel wall or neoplastic emboli in the vessel lumen.
Komai [39]	2007	Microvascular invasion	NA
Ito [40]	2006	Microvascular invasion	NA
Lee [41]	2006	Microvascular invasion	NA
Goncalves [42]	2004	Microvascular tumor invasion	MVI was indicated by neoplastic cells invading the vessel walls or by neoplastic emboli in the vessel lumen.
Lang [43]	2004	Microscopic venous invasion	NA
Ishimura [44]	2004	Microscopic venous invasion	MVI was defined by the presence of a cancer cell in blood vessels based on the examination of hematoxylin-eosin stained specimens.
Griffiths [45]	2002	Inferior vena cava invasion/ Renal vein invasion/Micro- scopic vascular invasion	Vascular invasion was classified into three categories, i.e. (i) inferior vena cava invasion(IVCI), (ii) invasion into the major hilar vessels, designated RVI and (iii) vascular invasion seen microscopically but not in the IVC or major hilar vessels, designated MVI.
VanPoppel [46]	1997	Microvascular invasion	MVI was considered present when tumor was seen in a vessel that is at least 1 or more endothelial cells or the tunica media of the vessel were recog- nized to surround a neoplastic cell group.