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Improving the accuracy of pre-operative survival prediction in renal cell carcinoma with C-reactive protein

SPK Jagdev[1,7], W Gregory[2,7], NS Vasudev[3], P Harnden[4], S Sim[1,3], D Thompson[5], J Cartledge[6], PJ Selby[3] and RE Banks*,[3]

[1]St James's Institute of Oncology, St James's University Hospital, Leeds, UK; [2]Clinical Trials Research Unit, University of Leeds, Leeds, UK; [3]Clinical and Biomedical Proteomics Group, Cancer Research UK Centre, Leeds Institute of Molecular Medicine, St James's University Hospital, Leeds LS9 7TF, UK; [4]Department of Pathology, St James's University Hospital, Leeds, UK; [5]Department of Clinical Biochemistry and Immunology, St James's University Hospital, Leeds, UK; [6]Department of Urology, St James's University Hospital, Leeds, UK

BACKGROUND: Validated objective biomarkers are needed for patients with renal cell carcinoma (RCC) to guide patient management and define high-risk populations for follow-up or for therapeutic purposes.

METHODS: Patients undergoing nephrectomy for RCC ($n = 286$ all stages, 84% with conventional clear cell type) were included with a median duration follow-up of 5 years. The prognostic significance of pre-operative haematological and biochemical variables, including C-reactive protein (CRP) values were examined and whether they added additional information to a recently published pre-operative scoring system was determined.

RESULTS: C-reactive protein was the most significant predictor of overall survival (OS; $\chi^2 = 50.9$, $P < 0.001$). Five-year OS for patients with CRP $\leqslant 15$ mg l^{-1} vs > 15 mg l^{-1} was 72% (95% CI 65–78%) and 33% (95% CI 23–44%), respectively. Similar results were seen for cancer-specific survival (CSS) and disease-free survival. On multivariate analysis, CRP remained highly significant for CSS ($\chi^2 = 17.3$, $P < 0.0001$) and OS ($\chi^2 = 9.8$, $P < 0.002$), in addition to other pre-operative variables including log of neutrophil/lymphocyte ratio, red blood cell count and white cell count. C-reactive protein was significant in addition to the pre-operative nomogram score ($\chi^2 = 12.5$, $P = 0.0004$ for OS, $\chi^2 = 16.2$, $P = 0.0001$ for CSS and $\chi^2 = 8.6$, $P = 0.003$ for DFS) and was still significant when other pre-operative variables were included.

CONCLUSION: C-reactive protein and other haematological and biochemical variables have independent prognostic significance in RCC and may enhance pre-operative scoring systems.

Renal cancer accounts for approximately 3% of adult malignancies with approximately 210 000 cases per year worldwide (GLOBOCAN 2002) (http://www-dep.iarc.fr). Approximately, 70% of patients have clinically localised disease at presentation, but 20–30% of these develop metastatic disease after surgery (Bukowski, 2009). Patients are usually followed up with intermittent imaging tests with a multiplicity of regimens proposed (Rouviere et al, 2006). In the curative setting, therefore, there is a need for prognostic markers that can help to stratify patients according to risk, to enable follow-up schedules to be individualised and to tailor eligibility criteria for trials of neo-adjuvant and adjuvant therapies. In the metastatic setting, such factors may help with patient selection for newer targeted therapies to minimise toxicity and maximise resources.

Many prognostic algorithms or models combine elements of the TNM staging system with tumour grade and combinations of pathological or clinical factors (Galfano et al, 2008; Lane and

Kattan, 2008). However, estimates of risk may be relatively wide for individual patients, not applicable to all subtypes or stages of RCC and some models have not been independently validated. Additionally parameters such as pathological stage and grade are subject to intra- and inter-observer variabilities. More recently, algorithms based on pre-operative variables have been developed. One of these is widely applicable and incorporates age, gender, symptom score, CT-derived size and T stage and presence or absence of metastatic disease (Karakiewicz et al, 2009). Such models have potential value in determining treatment modality, surgical strategy and neo-adjuvant targeting and patient management pre-operatively.

The area of defining new prognostic markers is of active interest (George and Bukowski, 2007; Crispen et al, 2008). Biomarkers in fluids offer the opportunity for more objective and reproducible measurement before operation. Recent examples include the prognostic significance of pre-operative serum sodium and urinary cathepsin D levels (Vasudev et al, 2008, 2009) and CRP (Ito et al, 2006; Karakiewicz et al, 2007; Komai et al, 2007; Kawata et al, 2008; Lamb et al, 2008; Ramsey et al, 2008; Iimura et al, 2009). In this study we have comprehensively analysed the potential pre-operative prognostic significance of a panel of

*Correspondence: Professor RE Banks; E-mail: r.banks@leeds.ac.uk
[7]These authors contributed equally to this work.

haematological and biochemical variables, including CRP in patients with all stages and histological subtypes of RCC undergoing nephrectomy, and examined their potential utility in combination with a recently published pre-operative (Karakiewicz *et al*, 2009) scoring system.

PATIENTS AND METHODS

Patients

Patients undergoing potentially curative or cytoreductive nephrectomy in Leeds between 1999 and 2006 and who consented to take part were included, excluding patients with familial VHL or polycystic kidney disease. All histological subtypes were included in the analysis (although some separate analyses were also undertaken on patients with clear cell histology only), reflecting the aims of the study in investigating the pre-operative utility of CRP that is, at a time when the histological subtype would be unknown. The study was approved by the Leeds Research Ethics Committee. Clinico-pathological data was largely obtained prospectively with nodal or metastatic disease status determined by CT scan, and all haematological and biochemical variables were measured routinely on pre-operative venous blood samples. C-reactive protein level was measured by a polyethylene glycol (PEG)-enhanced immunoturbidimetric assay on the Siemens Advia 1650 (Siemens Healthcare, Erlangen, Germany).

The pre-operative nomogram score using the variables of gender, age, tumour size and T stage from imaging and a symptom classification score (Karakiewicz *et al*, 2009) were calculated for each patient. Patients with T4 disease on CT staging were treated as T3, as the published nomogram did not include T4.

Statistical methods

The design and analysis of this retrospective study was conducted according to ReMARK guidelines (McShane *et al*, 2005). Survival curves were constructed using the life table method, with significance being determined using the log-rank test. Multivariate analysis of survival data used the proportional hazards model with a forward stepwise approach using a *P*-value for inclusion of 0.01 because of the inclusion of so many variables. Harrell's c-index (Harrell *et al*, 1982) was used to quantify the predictive accuracy of both univariate survival models and multivariate Cox models. This gives a value between 50%, for predictive capacity of the model being no better than random, and 100% for full predictive capacity. The 12 patients with unknown grades were recoded as grade 3, as their survival most closely matched that grade. A similar method was used for the few patients who had unknown values for stage or T stage and for the biochemical/blood variables. This enabled all the patients to be included in the Cox model runs. Additional model runs performed, excluding these patients, demonstrated that this approach was not creating bias. Variables were treated as continuous, or with simple, for example, log transformations in the Cox models, unless analysis using optimum cut-points of the variable gave substantially larger χ^2 values in the Cox model. For CRP, a log transformation was most appropriate. The CRP values reported as <5 were all assumed to be 2.5 for the purposes of the Cox model analyses. An optimum cut-point was one that maximised the χ^2 (or β coefficient in the Cox model) from the log-rank test, comparing survival above and below that cut-point. The Cox models were run with and without the pre-operative model scores to determine whether the accuracy of prediction of survival was improved when CRP and other factors were added to it. Correlations of other factors with the log of CRP values were examined using stepwise linear regression with all variables considered as continuous (detailed results not shown). The r^2 values in the multivariate linear regression indicate how much of the variability in CRP values is explained by the factors included. All *P*-values are two-tailed. Our 1, 2 and 5-year survival figures were compared with those previously published using the pre-operative nomogram (Karakiewicz *et al*, 2009) interpolating between the survival percentages given for different scores in their paper and averaging these over our whole patient population.

Survival was determined based on operation date. DFS was defined from that date until relapse with patients presenting without metastases or with metastases, which were resected being considered as disease free. Patients dying of other causes without relapsing were considered censored for DFS at their date of death.

RESULTS

The demographics and clinico-pathological characteristics of the 286 patients with CRP values available are shown in Table 1. Median follow-up was 5 years. Analyses, which included treatment and its effects, did not alter the findings and so are not reported.

Univariate analysis of CRP and survival

C-reactive protein was a highly significant predictor of OS, cancer-specific survival (CSS) and DFS in all RCC histological subtypes examined (Figures 1A–C). Although a log transformation of CRP was used in the Cox model a cut-point of 15 mg l^{-1} was used when depicting survival and DFS differences, as this incorporated much of the effect. The (univariate) predictive capacity of log of CRP, using Harrell's c-index was 70% for OS, 77% for CSS and 73% for DFS. There was no predictive effect of CRP on non-cancer survival (Figure 1D). The survival effect was similar in clear cell and non-clear cell subtypes (Figure 2).

Multivariate (Cox model) analysis

Univariate and multivariate Cox model results for CSS, OS and DFS are shown in Table 2. C-reactive protein is still highly significantly predictive of survival after inclusion of all the other significant factors for CSS, OS and DFS. Other factors that were independent predictors of CSS on multivariate analysis included tumour stage, grade, WBC, RBC and log of neutrophil/lymphocyte ratio. Log of CRP was the most predictive factor for OS (c-index of 70%) and the second most predictive factor after stage for CSS, increasing the predictive capacity from 81 to 86%. On multivariate analysis of factors for OS, the additional independent predictors included the presence of metastases, necrosis, MVI, RBC and WBC. For DFS, the only independent predictors are CRP, age, stage and MVI. C-reactive protein was the second most predictive factor after stage for DFS, increasing the predictive capacity from 77 to 84%.

Landmark analyses pre- and post-1 year

Cox multivariate analysis of CSS, restricted to deaths within the first year, showed the log of neutrophil/lymphocyte ratio ($\chi^2 = 14.5$, $P = 0.0001$) and WBC ($\chi^2 = 6.1$, $P = 0.01$) to be significant, plus log of CRP ($\chi^2 = 23.4$, $P < 0.0001$), the presence of metastases ($\chi^2 = 12.0$, $P = 0.0005$), N stage ($-$ve *vs* $+$ve, $\chi^2 = 10.6$, $P = 0.001$) and maximum tumour size ($\leqslant 50$ *vs* $51–100$ *vs* >100, $\chi^2 = 9.3$, $P = 0.002$). Analysis restricted to patients who had survived more than 1 year showed stage ($\chi^2 = 25.2$, $P < 0.0001$), grade ($\chi^2 = 9.4$, $P = 0.002$), necrosis ($\chi^2 = 6.4$, $P = 0.01$) and sodium ($\chi^2 = 8.5$, $P = 0.004$) to be the only relevant factors. C-reactive protein was still highly significant univariately in this latter analysis ($\chi^2 = 19.1$, $P < 0.0001$), but was no longer significant multivariately because of its correlations with stage, grade and necrosis. C-reactive protein was significantly correlated with many of the variables analysed (data not shown).

Table 1 Characteristics of the 286 renal cancer patients included in the study

Characteristics	n (%)	Characteristics	Median (range) n (%) for subsets
Age (yrs)			
Median	63	Hb (g per 100 ml)[a]	13.6 (7–20)
Range	29–86		
Gender			
Male	168 (59)	RBC ($\times 10^{12}$ cells per l)	4.6 (2.8–8.0)
Female	118 (41)	≤4.25	70 (24)
		>4.25	215 (75)
		Unknown	1 (0.3)
Histological subtype			
Clear cell	240 (84)	WBC ($\times 10^9$ cells per l)	7.7 (3.2–17.6)
Papillary	25 (9)	≤9.5	229 (80)
Chromophobe	12 (4)	>9.5	56 (20)
		Unknown	1 (0.3)
Collecting duct	3 (1)		
Unclassified	6 (2)		
Grade			
1	7 (2)	Neutrophils ($\times 10^9$ cells per l)	5.2 (2.25–14.79)
2	179 (28)	≤4.9	124 (43)
3	117 (41)	>4.9	160 (56)
4	72 (25)	Unknown	2 (1)
Unknown	12 (4)		
Max tumour size (cm)			
Median	6.1	Lymphocytes ($\times 10^9$ cells per l)	1.7 (0.26–7.35)
Range	0–18	≤1	37 (13)
		>1	248 (87)
		Unknown	1 (0.3)
Necrosis			
Present	94 (33)	Monocytes ($\times 10^9$ cells per l)[a]	0.41 (0.09–1.11)
Absent	183 (64)	Unknown	1 (0.3)
Unknown	9 (3)		
MVI			
Present	118 (41)	Neuts/Lymphs[b]	2.9 (0.75–30.6)
Absent	164 (57)	Unknown	3 (1)
		Mean	3.8
Unknown	4 (1)	Mean log	3.2
pT stage			
1	117 (41)	Platelets ($\times 10^9$ cells per l)	283 (78–884)
2	35 (12)	0–360	215 (75)
3	131 (46)	361–884	68 (24)
4	2 (1)	Unknown	3 (1)
Unknown	1 (0.3)		
N stage			
0	251 (88)	Sodium (mmol l^{-1})	140 (127–147)
1+	35 (12)	127–138	95 (33)
		139–147	190 (66)
		Unknown	1 (0.3)
		Mean	139.5
M stage			
0	228 (80)	Potassium (mmol l^{-1})	4.4 (3.1–6.0)
1	58 (20)	≤4.3	126 (44)
		>4.3	154 (54)
		Unknown	6 (2)
TNM stage			
1	109 (38)	Creatinine (μmol l^{-1})[b]	95 (52–1106)

Table 1 (Continued)

Characteristics	n (%)	Characteristics	Median (range) n (%) for subsets
II	28 (10)		
III	90 (31)		
IV	58 (20)		
Unknown	1 (0.3)	Urea (mmol l^{-1})[a]	5.7 (2–28.8)
CT T stage			
1a	75 (26)	CRP (mg l^{-1})	6.4 (<5–266)
1b	75 (26)	≤15	207 (72)
2	65 (23)	>15	79 (28)
3	43 (15)		
4	11 (4)		
Unknown	12 (6)		
CT tumour size (cm)			
Median	6.0		
Range	2–21		
Unknown	22 (8)		
Symptom score			
1	130 (45)		
2	99 (35)		
3	56 (20)		
Unknown	1 (0.3)		

Abbreviations: CRP = C-reactive protein; CT = computed tomography; Hb = haemoglobin; K = potassium; Lymphs = lymphocytes; M = metastasis; monos = monocytes; MVI, microvascular invasion; N = nodal; Na = sodium; Neuts = neutrophils; RBC = red blood cells; T = tumour; WBC = white blood cell count. Reference ranges are: Hb level, female: 11.5–16.0 g per 100 ml, male: 13.5–18.0 g per 100 ml; WBC, 4.0–11.0 $\times 10^9$ cells per l; RBC, 3.8–5.8 $\times 10^{12}$ cells per l; neutrophil count, 2.00–7.5 $\times 10^9$ cells per l; monocyte count, 0.2–0.8 $\times 10^9$ cells per l, platelets count, 150–400 $\times 10^9$ cells per l; sodium level, 135–145 mmol l^{-1}; potassium level, 3.5–5.0 mmol l^{-1}; creatinine level, female: 70–100 μmol l^{-1}, male: 80–115 μmol l^{-1}; urea level, 2.1–8.0 μmol l^{-1}; CRP < 10 mg l^{-1}. [a]Indicates coded as a continuous variable, no obvious cut-points. [b]Indicates coded as a log transformation, again no obvious cut-points.

Pre-operative prognostic variables

Pre-operative nomogram score gave excellent separation of the CSS, OS and DFS curves with similar survival estimates to the original published nomogram estimates (Karakiewicz et al, 2009) (Figure 3A). The predictive accuracy of this nomogram on our data, using Harrell's c-index, was 74% (compared with 86% for the data on which it was validated (Karakiewicz et al, 2007)). The slight early survival advantage in our series appeared to derive predominantly from the patients with higher nomogram scores, and therefore poorer prognosis.

In Cox model analysis, CRP is significant in addition to the nomogram score ($\chi^2 = 12.5$, $P = 0.0004$ for OS, $\chi^2 = 16.2$, $P = 0.0001$ for CSS and $\chi^2 = 8.6$, $P = 0.003$ for DFS), although the increase in predictive accuracy, as measured by Harrell's c-index, was uniformly only about 2%. However, to take OS as an example, predicted survival curves from the Cox model for CRP below and above 15, are still widely separated (Figure 3B), corresponding to the 2% improvement in predictive accuracy after allowing for nomogram score, with an individual having CRP ≤ 15 having an 86% 5-year survival compared with 69% for a patient with CRP > 15 even after allowing for the nomogram score. Including the other significant pre-operative variables from Table 2, CRP was still significant for OS ($\chi^2 = 7.8$, $P = 0.005$), CSS ($\chi^2 = 13.6$, $P = 0.0002$) and DFS ($\chi^2 = 11.2$, $P = 0.0008$). The nomogram score was not as predictive as the other pre-operative factors from Table 2 (CRP, metastases, RBC and WBC) for OS and does not provide extra prognostic information for survival in addition to these variables.
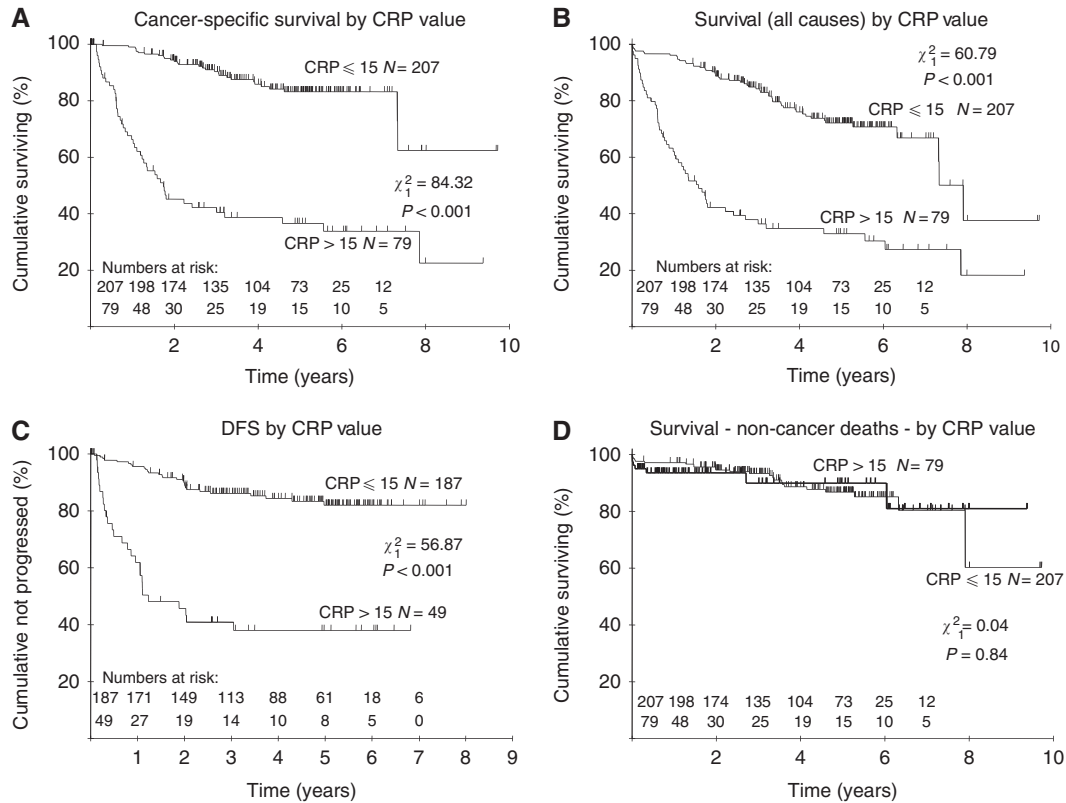
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Figure 1 Kaplan–Meier survival curves demonstrating the effect of CRP in relation to (**A**) CSS, (**B**) OS, (**C**) DFS and (**D**) non-cancer-related survival in patients undergoing nephrectomy for RCC. The 5-year OS was 72% (95% CI 65–78%) for patients with a CRP $\leqslant$ 15 mg l^{-1} and 33% (95% CI 23–44%) for patients with a CRP > 15 mg l^{-1} with corresponding 3-year OS being 84% (95% CI 78–89%) and 38% (95% CI 28–49%). The 5-year CSS was 83% (95% CI 76–88%) for patients with a CRP $\leqslant$ 15 mg l^{-1} and 37% (95% CI 26–49%) for patients with a CRP > 15 mg l^{-1} and corresponding 3-year CSS figures were 90% (95% CI 85–94%) and 42% (95% CI 32–54%). The 5-year DFS was 82% (95% CI 75–87%) for patients with a CRP $\leqslant$ 15 mg l^{-1} and 38% (95% CI 25–53%) for patients with a CRP > 15 mg l^{-1} with corresponding 3-year DFS being 86% (95% CI 80–90%) and 41% (95% CI 28–56%), respectively.
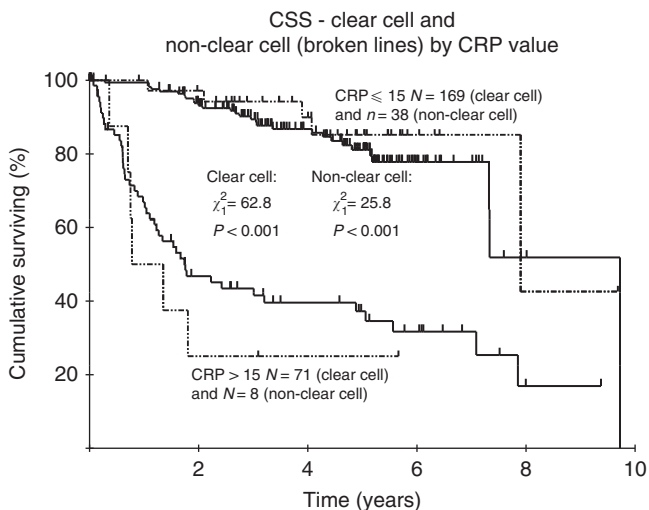

Figure 2 Kaplan–Meier survival curves demonstrating the effect of CRP in relation to CSS for clear cell histology (- - -) and non-clear cell histology in patients undergoing nephrectomy for RCC.

Log of CRP was correlated with nomogram score ($r = 0.50$, $P < 0.001$), and this changed only imperceptibly when CRP was analysed as a categorical variable above and below 15 ($r = 0.53$,

$P < 0.001$). C-reactive protein appeared to provide most additional explanatory power in the lower nomogram score ranges, although the numbers became small. For CSS, there was an effect in the <100 range, with 3 out of 104 deaths in the low CRP group and 2 out of 10 in the high CRP group ($\chi^2 = 10.7$, $P = 0.001$). For DFS, CRP again gave additional prognostic information in the good nomogram score categories (<100: low CRP vs high CRP $\chi^2 = 7.3$, $P = 0.007$ and 100–150: $\chi^2 = 5.7$, $P = 0.02$).

DISCUSSION

Previous studies have reported the independent prognostic significance of CRP measured before nephrectomy in patients with RCC in studies ranging from 40 to 313 patients (Jabs *et al*, 2005; Ito *et al*, 2006; Karakiewicz *et al*, 2007; Komai *et al*, 2007; Kawata *et al*, 2008; Lamb *et al*, 2008; Ramsey *et al*, 2008; Iimura *et al*, 2009). We confirm these, but importantly our study involving 286 patients is the first to include all stages, histological subtypes and all of CSS, OS and DFS, hence broadening the applicability. Although the comparison of CRP with scoring systems, such as the Kattan, UISS and SSIGN scores, has been made (Karakiewicz *et al*, 2007; Ramsey *et al*, 2008), we also show for the first time the independence of CRP and several other biochemical and haematological variables compared with a recently proposed pre-operative prognostic algorithm. This may indicate a potential contribution to such models, which may allow the more accurate stratification of individuals for treatment and monitoring. Pre-operatively, this could be of importance in determining patient

Table 2 Cox model predictors of (a) CSS, (b) OS and (c) DFS.

	Univariate				Multivariate				
Variable	χ^2	HR	HR CI[a]	*P*-value	χ^2	HR	HR CI[a]	*P*-value	c-Index[d]
					CSS				
(a)									
$\mathrm{Log_{10}(CRP)}$	66.1	4.4	3.1–6.3	<0.001	17.3	2.2	1.5–3.3	<0.0001	86.2
Age (continuous)	0.4	1.01	0.99–1.03	0.52				NS	
Stage	87.7	3.0	2.3–3.9	<0.001	42.7	2.4	1.8–3.3	<0.0001	80.8
Grade	61.5	3.7	2.6–5.3	<0.001	8.9	1.8	1.2–2.7	0.003	86.7
T stage	60.9	3.1	2.2–4.3	<0.001				NS	
N stage (−ve *vs* +ve)	34.0	6.1	3.8–10.0	<0.001				NS	
M stage	66.2	7.3	4.7–11.6	<0.001				NS	
Necrosis (Y/N)	43.5	4.8	3.0–7.7	<0.001				NS	
MVI (Y/N)	63.9	7.2	4.2–12.4	<0.001				NS	
Max tumour size ⩽50, 51–100, >100	40.3	2.8	2.1–3.9	<0.001				NS	
Gender	2.7[b]	1.5	0.9–2.4	0.10				NS	
Histology: clear cell *vs* rest	0.8[c]	1.3	0.69–2.6	0.37				NS	
RBC ⩽4.25 *vs* >4.25	14.7	2.6	1.6–4.1	<0.001	6.7	2.0	1.2–3.2	0.009	87.6
Hb (continuous)	35.6	1.45	1.3–1.6	<0.001				NS	
WBC ⩽9.5 *vs* >9.5	6.8	2.0	1.2–3.4	0.009	7.1	2.2	1.3–3.7	0.008	86.3
Neuts ⩽4.9 *vs* >4.9	16.1	2.7	1.6–4.5	<0.001				NS	
Lymphs ⩽1 *vs* >1	9.4	2.5	1.5–4.3	0.002				NS	
Monos (continuous)	6.8	6.6	1.7–26	0.009				NS	
Platelets ⩽360 *vs* >360	28.7	3.6	2.3–5.7	<0.001				NS	
$\mathrm{Log_{10}(neuts/lymphs)}$	24.1	2.5	1.8–3.5	<0.001	8.3	4.2	1.6–11	0.004	87.5
Na ⩽138 *vs* >138	16.3	2.6	1.6–4.0	<0.001				NS	
K ⩽4.3 *vs* >4.3	10.3	2.2	1.3–3.5	0.001				NS	
Urea (continuous)	0.1	1.01	0.94–1.1	0.80				NS	
Log (creatinine)	0.01	1.03	0.55–1.9	0.92				NS	
(b)					**OS**				
$\mathrm{Log_{10}(CRP)}$	50.9	3.1	2.3–4.1	<0.001	9.8	1.8	1.2–2.5	0.002	70.4
Age (continuous)	2.3	1.01	1.00–1.03	0.13				NS	
Stage	48.2	1.8	1.5–2.2	<0.001				NS	
Grade	34.5	2.2	1.6–2.8	<0.001				NS	
T stage	33.7	1.9	1.5–2.4	<0.001				NS	
N stage (−ve *vs* +ve)	36.2	4.7	3–7.2	<0.001				NS	
M stage	44.0	4.2	2.8–6.3	<0.001	21.7	2.9	1.9–4.5	<0.0001	73.5
Necrosis (Y/N)	33.4	3.2	2.1–4.7	<0.001	7.8	1.9	1.2–2.9	0.005	76.3
MVI (Y/N)	44.0	3.8	2.5–5.6	<0.001	9.5	2.0	1.3–3.2	0.002	78.5
Max tumour size ⩽50, 51–100, >100	26.5	2.0	1.6–2.7	<0.001				NS	
Gender	5.4[b]	1.6	1.1–2.4	0.02				NS	
Histology: clear cell *vs* rest	1.1[c]	1.3	0.77–2.4	0.29				NS	
RBC ⩽4.25 *vs* >4.25	18.2	2.4	1.6–3.6	<0.001	9.6	2.0	1.3–3.0	0.002	77.2
Hb (continuous)	31.2	1.3	1.2–1.5	<0.001				NS	
WBC ⩽9.5 *vs* >9.5	12.8	2.2	1.5–3.4	<0.001	14.1	2.4	1.6–3.7	0.0002	75.4
Neuts. ⩽4.9 *vs* >4.9	17.3	2.3	1.5–3.6	<0.001				NS	
Lymphs. ⩽1 *vs* >1	5.5	1.9	1.1–3.1	0.02				NS	
Monos (continuous)	9.6	6.5	2.1–20.4	0.002				NS	
Platelets ⩽360 *vs* >360	16.9	2.4	1.6–3.5	<0.001				NS	
Log(neuts/lymphs)	20.1	2.1	1.5–2.8	<0.001				NS	
Na ⩽138 *vs* >138	16.9	2.2	1.5–3.3	<0.001				NS	
K ⩽4.3 *vs* >4.3	8.1	1.8	1.2–2.6	0.004				NS	
Urea (continuous)	2.8	1.04	1.0–1.09	0.09				NS	
Log (creatinine)	2.9	1.5	0.98–2.2	0.09				NS	
(c)					**DFS**				
CRP (⩽15 *vs* >15)	38.3	4.0	2.6–5.9	<0.001	23.2	2.9	1.9–4.4	<0.0001	83.9
Age (continuous)	1.4	1.01	0.99–1.04	0.23	6.1	1.03	1.01–1.06	0.01	85.5
Stage	60.1	3.3	2.3–4.7	<0.001	26.6	2.6	1.7–3.8	<0.0001	77.4
Grade	35.2	3.1	2.1–4.5	<0.001				NS	
T stage	50.0	3.2	2.2–4.6	<0.001				NS	
N stage (−ve *vs* +ve)	14.9	5.5	2.7–11.4	<0.001				NS	
M stage	−								
Necrosis (Y/N)	30.1	4.7	2.7–8.0	<0.001				NS	
MVI (Y/N)	39.0	5.6	3.2–9.9	<0.001	7.5	2.3	1.2–4.2	0.006	85.0
Max tumour size ⩽50, 51–100, >100	35.0	3.1	2.1–4.6	<0.001				NS	
Gender	4.9[b]	1.9	1.05–3.3	0.03				NS	

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Table 2 *(Continued)*

Variable	Univariate				Multivariate				
	χ^2	HR	HR CI[a]	*P*-value	χ^2	HR	HR CI[a]	*P*-value	c-Index[d]
Histology:									
Clear cell *vs* rest	0.52[c]	1.3	0.6–2.8	0.47				NS	
RBC $\leqslant$ 4.25 *vs* >4.25	3.1	1.7	0.97–3.1	0.08				NS	
Hb (continuous)	8.7	1.25	1.1–1.4	0.003				NS	
WBC $\leqslant$ 9.5 *vs* >9.5	0.66	1.3	0.7–2.6	0.42				NS	
Neuts. $\leqslant$ 4.9 *vs* >4.9	4.1	1.75	1.0–3.0	0.04				NS	
Lymphs. $\leqslant$ 1 *vs* >1	0.3	1.3	0.47–3.6	0.59				NS	
Monos (continuous)	5.0	6.7	1.4–33.1	0.025				NS	
Platelets $\leqslant$ 360 *vs* >360	23.4	4.1	2.4–7.0	<0.001				NS	
Log(neuts/lymphs)	3.3	1.5	0.98–2.4	0.07				NS	
Na $\leqslant$ 138 *vs* >138	2.8	1.6	0.93–2.8	0.10				NS	
K $\leqslant$ 4.3 *vs* >4.3	2.4	1.5	0.89–2.6	0.12				NS	
Urea (continuous)	1.13	1.05	0.97–1.1	0.29				NS	
Log (creatinine)	0.32	1.25	0.6–2.6	0.57				NS	

Abbreviations: CI = confidence interval; CSS = cancer-specific survival; DFS = disease-free survival; HR = hazard ratio; NS = not significant; OS, overall survival. Units in table 2 are similar to those shown in table 1. [a]95% confidence interval. [b]Worse survival in male patients. [c]Clear cell with worse survival. [d]% at inclusion of variable.
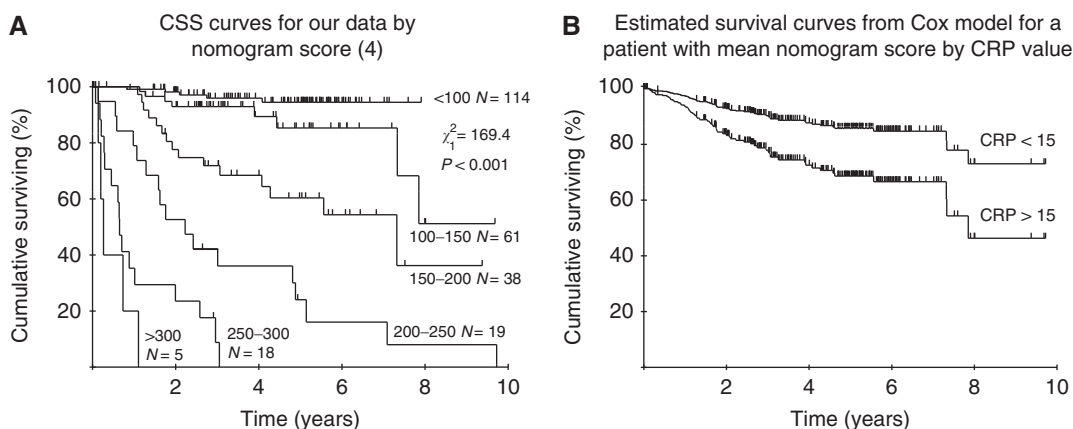

Figure 3 (**A**) Survival (CSS) curves for our data by nomogram score (Karakiewicz *et al*, 2009). The 1-, 2- and 5-year estimates for our data (published nomogram data) (Karakiewicz *et al*, 2009) are 91.9 (87.5%), 83.0 (80.2%) and 72.4% (73.9%), respectively. (**B**) Cox model predicted survival curves for a patient having the mean nomogram score and CRP below and above 15, demonstrating the additional effect of CRP in predicting survival beyond the nomogram score effect in patients undergoing nephrectomy for RCC.

treatment, such as selection of higher-risk patients, given the advent of neo-adjuvant therapies, or in the case of advanced disease those who most warrant cytoreductive nephrectomy with its inherent risks. Conversely, in a post-operative setting, higher risk patients may be identified for adjuvant therapies and/or monitored more intensively, all of which have implications for health economics and patient quality of life.

C-reactive protein is also associated with prognosis of other tumour types and recently in a study involving 10 408 individuals observed over 16 years as part of the Copenhagen City Heart Study; baseline levels of CRP were associated with increased risk of developing cancer (Allin *et al*, 2009). Importantly, this study used a high-sensitivity assay for CRP, allowing examination of values at a higher level of resolution within the 'normal' interval. Further analysis of CRP in two pooled prospective cohorts from the British Women's Heart and Health study and the Caerphilly cohort and a meta-analysis also found an association with cancer risk (Heikkila *et al*, 2009).

A criticism made in using CRP as a prognostic indicator is its perceived low specificity owing to its being an acute-phase reactant produced by the liver in a variety of inflammatory or infectious diseases. However, highly significant discrimination

between RCC patients with poor and good outcome was possible, and encouragingly CRP was not associated with non-cancer death in our study. The use of repeated baseline measurements (Ledue and Rifai, 2003; Pearson *et al*, 2003) may allow the impact of subclinical or clinical acute inflammatory illnesses to be determined. However, there is evidence in RCC that CRP is produced by normal and malignant renal epithelial cells (Jabs *et al*, 2003, 2005) with a significant correlation between pre-operative serum CRP and tissue CRP mRNA levels.

On multivariate analysis CRP clearly provides additional information to the pre-operative model (Karakiewicz *et al*, 2009). The small increase in predictive accuracy of about 2%, using Harrell's c-index, is not unexpected in such multivariate analyses. Schemper and Henderson, (2000) state that 'the likely amount of predictive accuracy is often low even if there are highly significant and relatively strong prognostic factors'. They recommend graphical comparisons of survival curves and clearly the CRP curves corresponding to the 2% improvement in predictive accuracy are still widely separated even after allowing for the nomogram score. This additional separation is interesting given that one component of the nomogram is symptom scores, which CRP may be expected to partly reflect. Our data shows that CRP is

highly correlated with many factors but in the multivariate linear regression the overall r^2 value for log of CRP only reached 49% and so 51% of the variability in CRP values is unexplained by factors examined. This variable is, therefore, also measuring an independent effect of some nature, and therefore has the potential to predict effects of a previously unknown nature. Its objective measurement and correlation with the other factors such as stage, MVI, necrosis and TNM augurs well for its use as a monitoring variable, as it is obviously related to other factors that are clearly relevant, although not completely assessable before treatment.

In addition to CRP, several other routinely measured biochemical and haematological parameters were shown to have independent prognostic significance, including log of neutrophil/lymphocyte ratio, serum sodium and WBC. Importantly these provided additional information to the pre-operative score (Karakiewicz et al, 2009) in this study. This is in contrast to a previous smaller study (Ramsey et al, 2008) in which on multivariate analysis apart from clinico-pathological variables and the SSIGN, UISS and Kattan scores, only CRP but not other haematological variables was significant for CSS or RFS. This difference is most likely because of the more heterogeneous cohort in our study, as when the patients with metastatic disease were excluded, the neutrophil/lymphocyte ratio was then no longer independently significant. Our model therefore has identified other parameters that could be of value in an integrated scoring system. This is the first report of the neutrophil/lymphocyte ratio as an independent predictive factor of survival in RCC patients, although previously described in other cancers such as ovarian cancer (Cho et al, 2009).

The concept of using additional measurable biomarkers together with clinico-pathological scoring systems is so far restricted to tissue with promising markers, including p53, vimentin and CAIX (Kim et al, 2004) or Ki-67, p53, endothelial/epithelial VEGFR1 and epithelial VEGF-D (Klatte et al, 2009). C-reactive protein is even more attractive being a routinely, relatively inexpensive and objectively measured analyte available pre-operatively. The additional value of this and other routinely measured biochemical and haematological variables with pre-operative nomograms such as that proposed by Karakiewicz et al (2009) should now be explored further prospectively in a larger series of patients. Such studies should include consideration of any known potential confounding factors such as BMI, smoking or HRT (Ledue and Rifai, 2003; Heikkila et al, 2007) and the role of co-morbidities and environmental factors in influencing an individual's baseline value as exemplified in cardiovascular disease (Ruckerl et al, 2009) in which high-sensitivity CRP measurements are currently used in risk assessment (Pearson et al, 2003).

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