Risk of infections in renal cell carcinoma (RCC) and non-RCC patients treated with mammalian target of rapamycin inhibitors

M D Kaymakcalan[1], Y Je[2], G Sonpavde[3], M Galsky[4], P L Nguyen[1,5], D Y C Heng[6], C J Richards[7] and T K Choueiri*[,8]

[1]Department of Pharmacy, Dana-Farber Cancer Institute, 450 Brookline Avenue, Boston, MA 02215, USA; [2]Department of Food and Nutrition, Kyung Hee University, 26 Kyungheedae-ro, Dongdaemun-gu, Seoul 130-701, Korea; [3]Department of Medical Oncology, University of Alabama at Birmingham (UAB) Comprehensive Cancer Center, 1824 6th St S, Birmingham, AL 35205, USA; [4]Mount Sinai School of Medicine, Tisch Cancer Institute, 1190 5th Avenue, New York, NY 10029, USA; [5]Department of Radiation Oncology, Brigham and Women's Hospital, 75 Francis Street, Boston, MA 02115, USA; [6]Department of Medical Oncology, Tom Baker Cancer Center and University of Alberta, 1331 29 St NW, Calgary, AB T2N 4N2, Canada; [7]Department of Medicine, Beth Israel Deaconess Medical Center and Harvard Medical School, 330 Brookline Avenue, Boston, MA 02215, USA and [8]Department of Medical Oncology, Dana-Farber Cancer Institute and Harvard Medical School, 450 Brookline Ave, Boston, MA 02215, USA

Background: Mammalian target of rapamycin (mTOR) inhibitors are used in a variety of malignancies. Infections have been reported with these drugs. We performed an up-to-date meta-analysis to further characterise the risk of infections in cancer patients treated with these agents.

Methods: Pubmed and oncology conferences' proceedings were searched for studies from January 1966 to June 2012. Studies were limited to phase II and III randomised controlled trials (RCTs) of everolimus or temsirolimus reporting on cancer patients with adequate safety profiles. Summary incidences, relative risks (RRs), and 95% confidence intervals (CIs) were calculated.

Results: A total of 3180 patients were included. The incidence of all-grade and high-grade infections due to mTOR inhibitors was 33.1% (95% CI, 24.5–43.0%) and 5.6% (95% CI, 3.8–8.3%), respectively. Compared with controls, the RR of all-grade and high-grade infections due to mTOR inhibitors was 2.00 (95% CI, 1.76–2.28, $P < 0.001$) and 2.60 (95% CI, 1.54–4.41, $P < 0.001$), respectively. Subgroup analysis found no difference in incidences or risks between everolimus and temsirolimus or between different tumour types (renal cell carcinoma (RCC) vs non-RCC). Infections included respiratory tract (61.7%), genitourinary (29.4%), skin/soft tissue (4.2%), and others (4.9%).

Conclusion: Treatment with mTOR inhibitors is associated with a significant increase in risk of infections. Close monitoring for any signs of infections is warranted.

Mammalian target of rapamycin (mTOR) inhibitors are targeted agents that have developed into an important therapeutic drug class used in a variety of malignancies. Everolimus (Afinitor, Novartis Pharmaceuticals, East Hanover, NJ, USA) is currently approved by the Food and Drug Administration (FDA) for the treatment of several malignancies including advanced renal cell carcinoma (RCC; Motzer et al, 2008, 2010), progressive pancreatic neuroendocrine tumours (PNET; Yao et al, 2011), subependymal

giant cell astrocytomas associated with tuberous sclerosis (Krueger *et al*, 2010) and most recently in combination with exemestane for advanced hormone receptor-positive, HER-2-negative breast cancer (Baselga *et al*, 2012). Temsirolimus (Torisel, Pfizer, New York, NY, USA) is approved by the FDA for the indication of advanced RCC (Hudes *et al*, 2007). This class of agents inhibits tumour cell proliferation by first binding to an intracellular protein (FKBP-12), which results in a protein–drug complex with mTOR, thereby inhibiting phosphorylation of p70S6 kinase and S6 ribosomal protein in the PI3 kinase/AKT pathway (Nashan, 2002; Abraham and Gibbons, 2007). This process ultimately leads to the downregulation of hypoxia inducible factor genes, including vascular endothelial growth factor, necessary for cancer cell growth, survival, and angiogenesis (Thomas *et al*, 2006).

Mammalian target of rapamycin inhibitors do not usually produce the systemic toxicities that are traditionally seen with chemotherapy such as nausea, vomiting, alopecia, and bone marrow suppression. However, as more targeted agents are developed their different class-specific toxicities are emerging. Trials exploring the safety and efficacy of everolimus and temsirolimus reported some unique side effects including rash, mucositis, infection, pneumonitis, hyperlipidaemia, and hyperglycaemia (Hutson *et al*, 2008; Ravaud, 2011; Elsen *et al*, 2012). Specifically, infections were reported in both everolimus and temsirolimus trials as a common side effect, as a cause of treatment disruption, and in some cases led to fatalities (Galanis *et al*, 2005; Ansell *et al*, 2008; Krueger *et al*, 2010; Motzer *et al*, 2010; Sakaria *et al*, 2010; Tarhini *et al*, 2010; Budde *et al*, 2012; Choueiri *et al*, 2013).

The association of infection with mTOR inhibitors and its possible clinical significance warrants further characterisation. We sought out to assess the magnitude of this problem by looking at the incidence and risk of infections in patients receiving mTOR inhibitors in a large, up-to-date meta-analysis of randomised control trials.

MATERIALS AND METHODS

Study selection. An independent review of the Pubmed databases from January 1966 to June 2012 was conducted. Searches were performed, limited to only human studies, with either the key terms 'everolimus,' 'Afinitor,' 'RAD-001' or 'temsirolimus,' 'Torisel,' 'CCI-779.' Citations investigating mTOR inhibitors in a non-oncological setting were excluded. Potentially relevant abstracts were collected and independently coded by four investigators (MDK, GS, CJR, and TKC). The full texts of the selected abstracts were obtained and analysed for appropriate trial design and safety reporting. Only randomised phase II or III studies with a placebo/control arm were used in order to properly calculate relative risk (RR). The Jadad scale was used to assess study quality based on study randomisation, double-blinding practice, and handling of withdrawals (Jadad *et al*, 1996). When more than one publication was drawn from the same clinical trial, the most recent or most complete report was used. The most recent package insert for each agent was assessed for the most current clinical information, and the manufacturer and/or the overall investigator was contacted to obtain clarification on infection information. The process was duplicated using the same search terms and limitations in the American Society of Clinical Oncology online databases of meeting abstracts from 2000 to 2012.

Data extraction. Data extraction was conducted according to the Preferred Reporting Items for Systemic Reviews of Meta-Analyses statement (Moher *et al*, 2009). Any discrepancies between reviewers' classifications of publications were resolved by consensus. The following information was extracted from each study: first author's name, year of publication, trial phase, underlying malignancy, number of enrolled patients, treatment arms, median age and range, median treatment duration and range, median overall survival and 95% confidence interval (CI), number of patients available for analysis, number of all-grade infections attributed to study drug, and number of high-grade infections attributed to study drug. All-grade and high-grade infections were defined according to the National Cancer Institute Common Toxicity Criteria version 3.0 in all trials used for the analysis.

Statistical analysis. We extracted the number of patients with all-grade and high-grade infections and the number of patients who were treated with mTOR inhibitors or control from the selected clinical trials to calculate incidence. The proportion of patients with all-grade infections and 95% CIs were derived from each trial. RRs and 95% CIs were also calculated by comparing the incidence of all-grade infection in patients assigned to mTOR inhibitors with the incidence among controls in the same trial. For studies reporting zero events in a treatment or control arm, we applied a classic half-integer continuity correction to calculate the RR and variance. We then repeated this for high-grade infections.

Statistical heterogeneity among trials included in the meta-analysis was evaluated using Cochrane's Q statistic (Cochran, 1954), and inconsistency was quantified with the I^2 statistic that estimates the percentage of total variation across studies due to heterogeneity rather than chance (Higgins *et al*, 2003). The assumption of homogeneity was considered invalid for P-values < 0.1. To calculate the summary incidence or RRs of all-grade or high-grade infections, we combined trial-specific estimates using random-effects or fixed-effects models depending on the heterogeneity of included trials. When no substantial heterogeneity among trials was found, the summary estimate calculated on the basis of the fixed-effects model was reported by using the inverse variance method. When substantial heterogeneity among trials was observed, the summary estimate calculated on the basis of the random-effects model was reported by using the DerSimonian and Laird (1986) method that considers both within-study and between-study variations. For trials with multiple treatment groups examining varying doses of mTOR inhibitors, we combined the treatment groups for the overall analysis.

To explore the possible reasons for the heterogeneity, we conducted meta-regression analyses to see whether there was a variation in risk estimates by type of drug, underlying malignancy, type of trial, and Jadad score. In addition, we conducted sensitivity analyses by omitting one study at a time to see the influence of each trial on the overall effect estimate. Finally, publication bias was evaluated through funnel plots (that is, plots of study results against precision) and with the Begg's and Egger's test (Begg and Mazumdar, 1994; Egger *et al*, 1997). A two-tailed P-value of < 0.05 was considered statistically significant. All statistical analyses were performed using Stata SE version 12.0 software (Stata Corporation, College Station, TX, USA).

RESULTS

Population characteristics. Our initial search yielded a total of 366 potentially relevant abstracts in Pubmed, with 309 abstracts searching for everolimus studies and 57 abstracts searching for temsirolimus studies. Two hundred and thirty-five everolimus studies were immediately excluded for not being oncological studies. One study from the temsirolimus search was excluded for not relating to an mTOR-inhibiting drug. Subsequently, an additional 118 studies were excluded for one of the following reasons: non-randomised trials, phase I trials, commentaries, review articles, editorials, letters, or only study design reporting. Four everolimus studies were excluded for being duplicates or subgroup-only analyses of previously reported trial data (Motzer
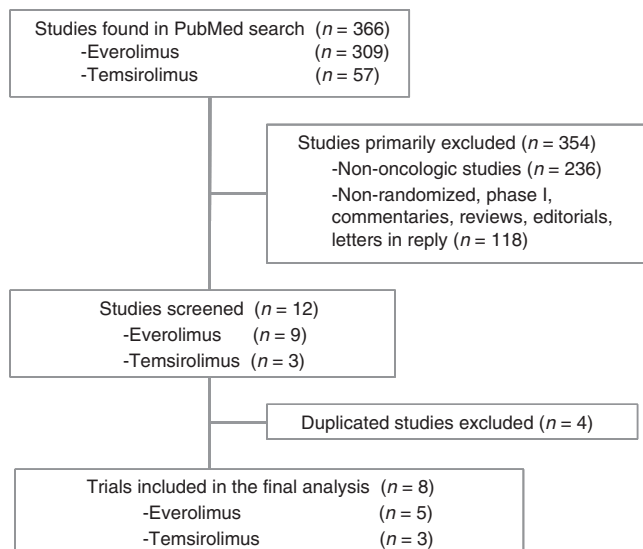
```
┌─────────────────────────────────────────────┐
│ Studies found in PubMed search  (n = 366)   │
│  -Everolimus                    (n = 309)   │
│  -Temsirolimus                  (n = 57)    │
└─────────────────────────────────────────────┘
          │
          │        ┌──────────────────────────────────────────┐
          │        │ Studies primarily excluded (n = 354)     │
          │        │  -Non-oncologic studies (n = 236)        │
          ├────────│  -Non-randomized, phase I,               │
          │        │   commentaries, reviews, editorials,     │
          │        │   letters in reply (n = 118)             │
          │        └──────────────────────────────────────────┘
          │
┌─────────────────────────────────────────────┐
│ Studies screened  (n = 12)                  │
│  -Everolimus               (n = 9)          │
│  -Temsirolimus             (n = 3)          │
└─────────────────────────────────────────────┘
          │
          │        ┌──────────────────────────────────────────┐
          ├────────│ Duplicated studies excluded (n = 4)      │
          │        └──────────────────────────────────────────┘
          │
┌─────────────────────────────────────────────┐
│ Trials included in the final analysis (n = 8)│
│  -Everolimus                    (n = 5)     │
│  -Temsirolimus                  (n = 3)     │
└─────────────────────────────────────────────┘
```

Figure 1. Selection process for randomised controlled trials included in the meta-analysis.   .

et al, 2008; Beaumont *et al*, 2011; Calvo *et al*, 2011; Tsukamoto *et al*, 2011). The remaining eight trials, including five studies with everolimus (Baselga *et al*, 2009; Motzer *et al*, 2010; Pavel *et al*, 2011; Yao *et al*, 2011; Baselga *et al*, 2012) and three studies with temsirolimus (Hudes *et al*, 2007; Hess *et al*, 2009; Negrier *et al*, 2011), were selected for the meta-analysis. Of the eight selected trials, six were phase III and two were phase II trials as shown in the selection process outlined in Figure 1. The baseline characteristics of each trial are presented in Table 1. There were 2233 patients from the everolimus trials and 947 patients from the temsirolimus trials for a total of 3180 patients available for safety analysis. The selected trials investigated patients with RCC (Hudes *et al*, 2007; Motzer *et al*, 2010; Negrier *et al*, 2011), hormone receptor-positive breast cancer (Baselga *et al*, 2009, 2012), PNET (Yao *et al*, 2011), all types of neuroendocrine tumours (Pavel *et al*, 2011), and mantle cell lymphoma (Hess *et al*, 2009) (Table 1). Inclusion criteria typically required that patients had good performance status and no active baseline infection, as well as adequate bone marrow, hepatic, pulmonary, and renal functions.

Overall incidence and types of infection. All mTOR inhibitor treatment arms were included in the analysis to calculate the overall incidence of infections. All-grade infections due to mTOR inhibitors occurred in 735 patients, representing an incidence of 33.1% (95% CI, 24.5–43.0%), whereas high-grade infections occurred in 100 patients, representing an incidence of 5.6% (95% CI, 3.8–8.3%). When subgrouped by drug type, the incidence of all-grade infections was 27.1% (95% CI, 16.5–41.0%) for everolimus and 41.8% (95% CI, 26.6–58.7%) for temsirolimus. For high-grade infection, the incidence for everolimus was 4.5% (95% CI, 2.3–8.6%) and the incidence for temsirolimus was 7.1% (95% CI, 5.2–9.7%). The incidence for the placebo/control arm was 15.5% (95% CI, 10.2–22.8%) for all-grade infections and was 1.6% (95% CI, 0.8–3.1%) for high-grade infections.

All of the studies used in the meta-analysis documented the rates of both all-grade and high-grade infections, with the exception of one temsirolimus study that did not indicate the incidence of high-grade infections. There were 262 (35.6%) reported infections on mTOR inhibitors in which the type of the infection was not otherwise specified in the studies. Of the six studies that reported on the underlying type of infection (N = 473), the most common were respiratory tract infections, representing 292 patients or 61.7% of all infections due to mTOR inhibitors.

Genitourinary infections (29.4%), skin/soft tissue infections (4.2%), sepsis (1.5%), and gastrointestinal infections (0.2%) were the other infections that were reported in the studies (Table 1). Fungal infections, including candida (1.3%), aspergillosis (0.4%), and others (0.2%) were reported in the studies. Viral infections, including herpes (1.1%) and hepatitis (0.2%), and parasitic infections (0.2%) were also reported (Table 1). Some of these infections were severe and led to the most common cause of fatal adverse events in the trials (Choueiri *et al*, 2013).

RRs of infections and subgroup analysis. All eight trials (five everolimus + three temsirolimus) reported the incidence of all-grade infections and seven trials (five everolimus + two temsirolimus) reported the incidence of high-grade infections, which were used to calculate the RRs of infections associated with mTOR inhibitor use.

The overall RR of developing an all-grade infection for mTOR inhibitors *vs* control was 2.00 (95% CI, 1.76–2.28, P < 0.001). No significant heterogeneity was found among the trials (Q = 5.02, I^2 = 0.0%, P = 0.658; Figure 2). By drug type, everolimus-treated patients (5 trials, 1312 patients) had an increased risk of all-grade infection, with a RR of 2.14 (95% CI, 1.81–2.53, P < 0.001), and temsirolimus-treated patients (3 trials, 612 patients) had an increased risk of all-grade infection, with a RR of 1.81 (CI 95%, 1.48–2.22, P < 0.001). Comparing the RRs of everolimus with temsirolimus demonstrated no significant difference (P = 0.26; Table 2).

When considering only high-grade infection events, the overall RR for mTOR inhibitors *vs* control was 2.60 (CI 95%, 1.54–4.41, P < 0.001). No significant heterogeneity was found among the studies reporting high-grade infections (Q = 7.41, I^2 = 19.1%, P = 0.284) (Figure 2). By drug type, everolimus-treated patients (5 trials, 1,312 patients) had an increased risk of high-grade infection, with a RR of 3.63 (95% CI, 1.66–7.94, P = 0.001). Temsirolimus-treated patients (2 trials, 524 patients) tended to have an increased risk of a high-grade infection, but the RR for temsirolimus *vs* control (RR = 1.97; 95% CI, 0.97–4.03, P = 0.062) did not reach a level of statistical significance because of smaller numbers. No significant differences were observed when comparing the RRs of high-grade infection between everolimus and temsirolimus (P = 0.35; Table 2).

To determine whether the observed increases in RRs of all-grade and high-grade infections were influenced by tumour type, we performed a subgroup analysis of RCC, the most commonly occurring malignancy (3 studies and 1197 total patients or 37.6% of all patients), *vs* all other malignancies. The RR of all-grade infection in patients treated with RCC was 1.84 (95% CI, 1.53–2.21; P < 0.001), whereas the RR of all-grade infection in patients with other malignancies (five trials) was 2.18 (95% CI, 1.82–2.60; P < 0.001). No significant difference was observed in RRs of all-grade infection between the patients with RCC or non-RCC (P = 0.25). Similarly, the RR of high-grade infection in patients with RCC treated with mTOR inhibitors (two trials) was 2.76 (95% CI, 1.31–5.81; P = 0.007). For non-RCC patients (five trials), the RR of all-grade infection was 2.46 (95% CI, 1.16–5.19; P = 0.018). No difference was found when comparing the RRs of high-grade infection in patients having RCC with other malignancies (P = 0.63; Table 2).

Study quality. Randomised treatment allocation sequences were generated in all trials used in the analysis. All five everolimus trials were double-blinded, placebo-controlled, and of highest quality achieving the highest Jadad score of 5. All three temsirolimus trials were not double-blinded nor placebo-controlled, but they did all have active treatment controls. All studies were of good methodological quality according to the 5-point Jadad score (all trials ≥ 3). To further assess study quality, we also compared the RRs of all-grade and high-grade infection among phase II *vs* phase III trials. There were no statistically significant differences between

Table 1. Baseline characteristics of the included trials in the meta-analysis

Author, year	Phase	Histology	Patients enrolled	Treatment arms/ dose	Median age (years) (range)	Median treatment duration (months) (Range)	Median OS (months) (range)	Median PFS (months) (range)	Patients for safety analysis	No. of infections as a result of study drug		Reported all-grade infections	Quality
										All grade	High grade		
Everolimus													
Baselga et al, 2012	3	HR(+) BC	724	Everolimus 10mg QD+exemestane 25 mg QD Placebo+exemestane 25 mg QD	62 (34–93) 61 (28–90)	6.0 3.3	Not reached Not reached	11 4.1	482 238	241 59	24 4	Resp, GU, sepsis, candida, hepatitis C	5
Baselga et al, 2009	2	HR(+) BC	270	Everolimus 10mg QD+letrozole 25 mg QD Placebo+letrozole 2.5 mg QD	69 (46–88) 67 (43–84)	Not Reported Not Reported	N/A N/A	N/A N/A	137 132	3 2	1 1	Resp, skin/soft tissue	5
Motzer et al, 2010	3	RCC	416	Everolimus 10mg QD Placebo	61 (27–85) 60 (29–79)	4.7 (0.63–15) 2 (0.7–6.5)	14.8 14.4	4.9 (4.0–5.5) 1.9 (1.8–1.9)	274 137	101 24	27 1	Resp, GU, sepsis, candida aspergillosis	5
Pavel et al, 2011	3	NET	429	Everolimus 10mg QD+octreotide 30mg Q28D Placebo+octreotide 30mg Q28D	60 (22–83) 60 (27–81)	9.2 (0.25–40.7) 9.1 (<0.25–38)	Not reached Not reached	16.4 (13.7–21.2) 11.3 (8.4–14.6)	215 211	42 13	11 1	NOS	5
Yao et al, 2011	3	PNET	410	Everolimus 10mg QD Placebo	58 (23–87) 57 (20–82)	9.2 4	Not reached Not reached	13.7 (11.2–18.8) 5.7 (5.4–8.3)	204 203	83 38	0 1	Resp, GU	5
Temsirolimus													
Hess et al, 2009	3	MCL	162	Temsirolimus 175mg followed by 75 mg QW Temsirolimus 175mg followed by 25 mg QW Investigator's choice of therapy	68 (44–87) 68.5 (43–85) 64.5 (39–88)	3 (0.25–24.4) 3.5 (0.25–43) 5 (0.25–8.7)	12.8 (8.6–19.3) 10 (7.2–14.6) 9.7 (5.8–15.1)	4.8 (3.1–8.1) 3.4 (1.9–5.5) 1.9 (1.6–2.5)	54 54 53	15 11 4	4 2 2	NOS	3
Hudes et al, 2007	3	RCC	626	Temsirolimus 25 mg QW INF-α 3 mIU–18 mIU TIW Temsirolimus 15 mg QW+INF-α 6 mIU TIW	58 (32–81) 60 (23–86) 59 (32–82)	3.8 (3.5–3.9) 1.9 (1.9–2.2) 2.5 (1.9–3.6)	10.9 (8.6–12.7) 7.3 (6.1–8.8) 8.4 (6.6–10.3)	5.5 (3.9–7.0) 3.1 (2.2–3.8) 4.7 (3.9–5.8)	208 200 208	118 50 70	9 7 22	Resp, GU	3
Negrier et al, 2011	2	RCC	171	Temsirolimus 25 mg QW+bevacizumab 10 mg kg^{-1} Q2W Sunitinb 50mg Q6W Q6W days 1–28 INF-α 9mIU TIW+bevacizumab 10 mg kg^{-1} Q2W	62 (33–83) 61.2 (33–83) 61.9 (40–79)	5.1 (0–12) 10.4 (0.5–12) 7.2 (1.0–12)	Not reached Not reached Not reached	8.2 (7.0–9.6) 8.2 (5.5–11.7) 16.8 (6.0–26)	88 42 40	51 16 12	N/A N/A N/A	Resp, GU, skin/soft tissue, GI, sepsis, fungal, Candida, herpes, parasitic	3

Abbreviations: GI = gastrointestinal; GU = genitourinary; HR(+) BC = hormone receptor-positive breast cancer; INF-α = interferon alpha; MCL = mantle cell lymphoma; mIU = million international units; NET = neuroendocrine tumour; No. = number; NOS = not otherwise specified; OS = overall survival; PFS = progression-free survival; PNET = pancreatic neuroendocrine tumour; QD = once a day; QW = once a week; Q28D = every 28 days; Q2W = twice weekly; Q6W = every 6 weeks; QoW = once a month; RCC = renal cell carcinoma; Resp = respiratory; TIW = three times a week.

Relative risks of all-grade infections:

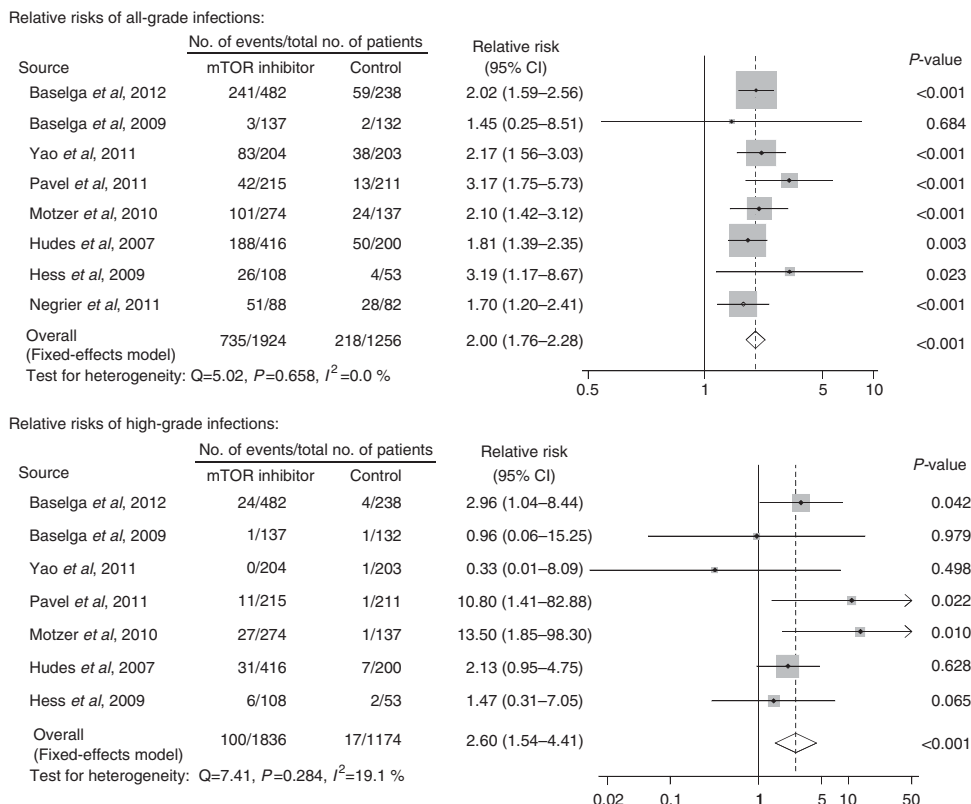
Relative risks of high-grade infections:

Figure 2. Relative risks of all-grade and high-grade infections associated with mTOR inhibitors.

Table 2. Incidences and relative risks of all-grade and high-grade infections associated with mTOR inhibitors						
	No. of infection/No. of subjects		Incidence			
	Number of studies	mTOR inhibitors	Placebo	mTOR inhibitors	Placebo	Relative risk (95% CI)	Test for heterogeneity
All grade						
Overall	8	735/1924	218/1256	33.1% (24.5–43.0)	15.5% (10.2–22.8)	2.00 (1.76–2.28); $P<0.001$	$Q=5.02$, $P=0.658$, $I^2=0.0\%$
Everolimus	5	470/1312	136/921	27.1% (16.5–41.0)	12.2% (6.8–21.1)	2.14 (1.81–2.53); $P<0.001$	$Q=2.13$, $P=0.711$, $I^2=0.0\%$
Temsirolimus	3	265/612	82/335	41.8% (26.6–58.7)	22.2% (12.4–36.5)	1.81 (1.48–2.22); $P<0.001$	$Q=1.36$, $P=0.506$, $I^2=0.0\%$
High grade						
Overall	7	100/1836	17/1174	5.6% (3.8–8.3)	1.6% (0.8–3.1)	2.60 (1.54–4.41); $P<0.001$	$Q=7.41$, $P=0.284$, $I^2=19.1\%$
Everolimus	5	63/1312	8/921	4.5% (2.3–8.6)	1.0% (0.5–2.0)	3.63 (1.66–7.94); $P=0.001$	$Q=5.97$, $P=0.202$, $I^2=32.9\%$
Temsirolimus	2	37/524	9/253	7.1% (5.2–9.7)	3.6% (1.9–6.7)	1.97 (0.97–4.03); $P=0.062$	$Q=0.17$, $P=0.681$, $I^2=0.0\%$
Abbreviations: CI = confidence interval; mTOR = mammalian target of rapamycin; No. = number.						

the phase subgroups for either grade (all-grade $P=0.33$; high-grade $P=0.57$). To assess the influence of each study on the overall RRs of all-grade and high-grade infection, we performed a sensitivity analysis by omitting one study at a time and found that there were no studies that had a significant impact on the overall RR.

Finally, we attempted to assess the possible association between risk of infection and increased exposure to mTOR inhibitors. In convention with other studies, we calculated the mean of the median durations of therapy as a cut-off, which was found to be 5.8 months. When comparing the RRs of all-grade and high-grade infections due to mTOR inhibitors amongst long-coursed trials

and short-coursed trials, we did not observe a statistically significant difference (all-grade $P = 0.33$; high-grade $P = 0.80$).

Publication bias. The Begg's test and the Egger's test were conducted on the incidence and RR of all-grade infection, and both showed no evidence of publication bias (for RR of all-grade infection, Begg's $P = 0.54$ and Egger's $P = 0.31$). Similarly, the Begg's and Egger's tests were performed on the incidence and RR of high-grade infection, and no evidence of publication bias was found (for RR of high-grade infection, Begg's $P = 0.55$ and Egger's $P = 0.97$).

DISCUSSION

Our meta-analysis combined eight RCTs investigating mTOR inhibitors for the treatment of cancers. To our knowledge, this is the first large study of RCTs demonstrating a significant increase in the risk of infection with the use of mTOR inhibitors in cancer patients. Our analysis found that the risk of developing an infection of any grade was two-fold higher in patients treated with mTOR inhibitors. And, more importantly, there is a 2.6-fold increase in the risk of high-grade infection associated with the use of mTOR inhibitors. The observed infection risk can possibly be explained by the potential immunosuppressant effects of mTOR inhibitors. Everolimus and sirolimus, the principal active metabolite of temsirolimus, are known to inhibit interleukin (IL)-2-, IL-7-, and IL-15-driven proliferation of activated T cells and B cells (Lai and Tan, 1994; Schuler et al, 1997; Shegal, 2003; Chapman and Perry, 2004).

Although there were no statistically significant differences between temsirolimus and everolimus when comparing their RRs, there was a heavier contribution from everolimus to the RR of infection, likely because of the larger number of everolimus patients and studies used in the analysis rather than a distinctive quality of the drug.

With an increased RR of treatment-related infections, it is clear that proper monitoring, immediate intervention, and effective management is crucial to achieve the maximal therapeutic benefit of mTOR inhibitors. After reporting the incidence of infection in everolimus-treated RCC patients, which was similar to the incidence in our analysis (37% vs 33.1%; Motzer et al, 2010), the RECORD-1 Study Group subsequently published recommendations for the management of infections and other adverse events according to the grade of the event (Porta et al, 2011; Ravaud, 2011). These recommendations can be used by clinicians to effectively manage treatment-related infections. Fungal infections such as Candida and Aspergillosis, mycobacterial infections such as tuberculosis, and viral infections such as hepatitis and herpes occurred in the studies used in our analysis and were reported in the prescribing information (Novartis, 2012; Pfizer, 2012). Patients must be appropriately screened for viral, mycobacterial and fungal infections in the right clinical context. Clinicians must fully treat patients with any active infection before the initiation of mTOR inhibitors and must monitor patients during the course of treatment (Porta et al, 2011). Typically, patients with active or recently active infections are excluded from clinical trials; therefore, the true incidence of these infections could be widely underreported. More trials and reporting on these patients must be done in order to gain more insight into the management of this subgroup of patients.

A randomised, double-blinded multicenter trial evaluated the pharmacokinetics of temsirolimus and suggested that there may indeed be a correlation between the cumulative exposure of temsirolimus and certain adverse effects including infection (Boni et al, 2005). In our meta-analysis, patients in the studies with longer treatment durations did not have more risk to develop infections than patients on studies with shorter treatment durations ($P > 0.05$ for all-grade and high-grade). The findings do not support the association of infection risk and cumulative exposure; however, information on the time of occurrence of infection and individual data points on treatment duration may be needed to properly investigate the association.

Despite the size of this meta-analysis, our study has several limitations. First, we only had access to the available data published in the clinical trials, so there were patient variables that were not known, such as co-morbidities, previous treatment exposure, concomitant medications, and dose interruptions. Second, patients in trials have adequate organ and haematological function, which may not be the case in common oncology practice. It is conceivable that the true incidence and risk of treatment-related adverse effects is higher in actual practice. Third, not all of the included studies were double-blinded, but blinding is not always possible with parenteral administration. Although some of the included studies were not blinded, they were all of good methodological quality. Lastly, and despite our attempts, the reported safety data did not disclose the specific aetiologies of all the infections that occurred.

In conclusion, the mTOR inhibitors everolimus and temsirolimus are associated with an increased risk of all-grade and high-grade infections. These targeted agents are of great clinical benefit in various malignancies and the benefits outweigh the risks in the vast majority of cases, and thus their FDA approval. However, the immediate detection and effective management of the potential bacterial, viral, and fungal infections that can occur with these agents is crucial for optimal patient outcomes.

ACKNOWLEDGEMENTS

DISCLAIMER

GS: Speaker's bureau for Novartis, GSK; Advisory board for Novartis, Pfizer. DYCH: Advisory board/consultancy, Pfizer, Novartis, Bayer/Onyx. TKC: Advisory Board, Pfizer, GSK, Novartis, Aveo, Genentech, Bayer/Onyx. The remaining authors declare no conflict of interest.

REFERENCES

Abraham RT, Gibbons JJ (2007) The mammalian target of rapamycin signaling pathway: twists and turns in the road to cancer therapy. *Clin Cancer Res* **13**: 3109–3114.

Ansell SM, Inwards DJ, Rowland Jr KM, Flynn PJ, Morton RF, Moore Jr DF, Kaufmann SH, Ghobrial I, Kurtin PJ, Maurer M, Allmer C, Witzig TE (2008) Low- dose, single temsirolimus for relapsed mantle cell lymphoma. *Cancer* **113**(3): 508–514.

Baselga J, Campone M, Piccart M, Burris HA, Rugo HS, Sahmound T, Noguchi S, Gnant M, Pritchard KI, Lebrun F, Beck JT, Ito Y, Yardley D, Deleu I, Perez A, Bachelot T, Vittori L, Xu Z, Mukhopadhyay P, Lebwohl D, Hortobagyi GN (2012) Everolimus in postmenopausal hormone-receptro-positive advanced breast cancer. *N Engl J Med* **366**: 520–529.

Baselga J, Semiglazov V, van Dam P, Manikhas A, Bellet M, Mayordomo J, Campone M, Kubista E, Greil R, Bianchi G, Steinseifer J, Molloy B, Tokaji E, Gardner H, Phillips P, Stumm M, Lane HA, Dixon JM, Jonat W, Rugo HS (2009) Phase II randomized study of neoadjuvant everolimus plus letrozole compared with placebo letrozole in patients with estrogen receptor-positive breast cancer. *J Clin Oncol* **27**: 2630–2637.

Beaumont J, Butt Z, Baladi J, Motzer RJ, Haas T, Hollaender N, Kay A, Cella D (2011) Patient-reported outcomes in a phase III study of everolimus versus placebo in patients with metastatic carcinoma of the kidney that has

progressed on vascular endothelial growth factor receptor tyrosine kinase inhibitor therapy. *Oncologist* **16**: 632–640.

Begg CB, Mazumdar M (1994) Operating characteristics of a rank correlation test for publication bias. *Biometrics* **50**: 1088–1101.

Boni JP, Leister C, Bender G, Fitzpatrick V, Twine N, Stover J, Dorner A, Immermann F, Burczynski ME (2005) Population pharmacokinetics of CCI-779: correlation to safety and pharmacogenomic responses in patients with advanced renal cell. *Clin Pharmacol Ther* **77**(1): 76–89.

Budde K, Kingswood C, Zonnenberg B, Frost M, Belousova E, Sauter M, Fischerder M, Nonomura N, ebin M, Pei Y, Brakemeier S, Sahmoud T, Shah G, Lincy J, Bissler J (2012) *Everolimus in the treatment of angiolipoma in patients with tuberous sclerosis complex or sporadic lymphangioleimyomatosis: results from EXIST-2*. Presented at 27th Annual European Association of Urology Congress, 24–28 February (2012) Paris, France.

Calvo E, Escudier B, Motzer R, Oudard S, Hutson TE, Porta C, Bracarda S, Grünwald V, Thompson JA, Ravaud A, Kim D, Panneerselvam A, Anak O, Figlin RA (2011) Everolimus in metastatic renal cell carcinoma: subgroup analysis of patients with 1 or 2 previous vascular endothelial growth factor receptor-tyrosine kinase inhibitor therapies enrolled in the phase III RECORD-1 study. *Eur J Cancer* **48**: 333–339.

Chapman TM, Perry CM (2004) Everolimus. *Drugs* **64**(8): 861–872.

Choueiri TK, Je Y, Sonpavde G, Richards CJ, Galsky MD, Nguyen PL, Schutz F, Heng DY, Kaymakcalan M (2013) Incidence and risk of treatment related mortality in cancer patients treated with mammalian target of rapamycin inhibitor. *Ann Oncol*; e-pub ahead of print 8 May 2013; doi:10.1093/annonc/mdt155.

Cochran WG (1954) The combination of estimates from different experiments. *Biometrics* **10**: 101–129.

DerSimonian R, Laird N (1986) Meta-analysis in clinical trials. *Control Clin Trials* **7**: 177–188.

Egger M, Davey Smith G, Schneider M, Minder C (1997) Bias in meta-analysis detected by a simple, graphical test. *BMJ* **315**: 629–634.

Elsen T, Sternberg CN, Robert C, Mulders P, Zbinden S, Izzedine H, Escudier B (2012) Targeted therapies for renal cell carcinoma: review of adverse event management strategies. *J Natl Cancer Inst* **104**: 93–113.

Galanis E, Buckner JC, Maurer MJ, Kreisberg JI, Ballman K, Boni J, Peralba JM, Jenkins RB, Dakhil SR, Morton RF, Jaeckle KA, Scheithauer BW, Dancey J, Hidalgo M, Walsh DJ (2005) Phase II trial of temsirolimus (CCI-779) in recurrent glioblastoma multiforme: a north central cancer treatment group study. *J Clin Oncol* **23**: 5294–5304.

Hess G, Herbrecht R, Romaguera J, Verhoef G, Crump M, Gisselbrecht C, Laurell A, Offner F, Strahs A, Berkenblit A, Hanushevsky O, Clancy J, Hewes B, Moore L, Coiffier B (2009) Phase III study to evaluate temsirolimus compared with investigator's choice therapy for the treatment of relapsed or refractory mantle cell lymphoma. *J Clin Oncol* **27**: 3822–3829.

Higgins JP, Thompson SG, Deeks JJ, Altman DG (2003) Measuring inconsistency in meta-analyses. *BMJ* **327**: 557–560.

Hudes G, Carducci M, Tomczak P, Dutcher J, Figlin R, Kapoor A, Staroslawska E, Sosman J, McDermott D, Bodrogi I, Kovacevic Z, Lesovoy V, Schmidt-Wolf IG, Barbarash O, Gokmen E, O'Toole T, Lustgarten S, Moore L, Motzer RJ (2007) Temsirolimus, interferon alfa, or both for advanced renal-cell carcinoma. *N Engl J Med* **356**: 2271–2281.

Hutson TE, Figlin RA, Kuhn JG, Motzer RJ (2008) Targeted therapies for metastatic renal cell carcinoma: an overview of toxicity and dosing strategies. *Oncologist* **13**: 1084–1096.

Jadad A, Moore R, Carroll D, Jenkinson C, Reynolds DJ, Gavaghan DJ, McQuay HJ (1996) Assessing the quality of reports of randomized clinical trials: Is blinding necessary? *Control Clin Trials* **17**: 1–12.

Krueger DA, Care MM, Holland K, Agricola K, Tudor C, Mangeshkar P, Wilson KA, Byars A, Sahmoud T, Franz DN (2010) Everolimus for subependymal giant-cell astrocytomas in tuberous sclerosis. *N Engl J Med* **363**: 1801–1811.

Lai JH, Tan TH (1994) CD28 signaling causes a sustained down-regulation of I kappa B alpha which can be prevented by the immunosuppressant rapamycin. *J Biol Chem* **269**(48): 30077–30080.

Moher D, Liberati A, Tetzlaff J, Altman DG (2009) Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. *J Clin Epidemiol* **62**: 1006–1012.

Motzer R, Escudier B, Oudard S, Hutson TE, Porta C, Bracarda S, Grunwald V, Thompson JA, Figlin RA, Hollaender N, Kay A, Ravaud A (2010) Phase 3 trial of everolimus for metastatic renal cell carcinoma. *Cancer* **116**: 4256–4465.

Motzer R, Escudier B, Oudard S, Hutson TE, Porta C, Bracarda S, Grunwald V, Thompson JA, Figlin RA, Hollaender N, Urbanowitz G, Berg WJ, Kay A, Lebwohl D, Ravaud A (2008) Efficacy of everolimus in advanced renal cell carcinoma: a double-blind, randomized, placebo-controlled phase III trial. *Lancet* **372**: 449–456.

Nashan B (2002) Review of the proliferation inhibitor everolimus. *Expert Opin Investig Drugs* **11**(12): 1845–1857.

Negrier S, Gravis G, Perol D, Chevreau C, Delva R, Bay JO, Blanc E, Geoffrois L, Rolland F, Legouffe E, Sevin E, Laguerre B, Escudier B (2011) Temsirolimus and bevacizumab, or sunitinib, or interferon alfa and bevacizumab for patients with advanced renal cell carcinoma (TORAVA): a randomized phase 2 trial. *Lancet Oncol* **12**(7): 673–680.

Novartis (2012) *Afinitor (everolimus) Package Insert*. Novartis Pharma Stein AG: Stein, Switzerland.

Pavel ME, Hainsworth JD, Baudin E, Peeters M, Horsch D, Winkler RE, Klimovsky J, Lebwohl D, Jehl V, Wolin EM, Oberg K, Van Custem E, Yao JC (2011) Everolimus plus octreotide long-active repeatable for the treatment of advanced neuroendocrine tumours associated with carcinoid syndrome (RADIANT-2): a randomised, placebo-controlled, phase 3 study. *Lancet* **378**: 2005–2012.

Pfizer Inc (2012) *Torisel (temsirolimus) Package Insert*. Pfizer Inc: Philadelphia, PA.

Porta C, Osanto S, Ravaud A, Climent MA, Vaishampayan U, White DA, Creel P, Dickow B, Fischer P, Gornell SS, Meloni F, Motzer RJ (2011) Management of adverse events associated with the use of everolimus in patients with advanced renal cell carcinoma. *Eur J Cancer* **47**(9): 1287–1298.

Ravaud A (2011) Treatment-associated adverse event management in the advanced renal cell carcinoma patient treated with targeted therapies. *Oncologist* **16**(suppl 2): 32–44.

Sakaria JN, Galanis E, Wu W, Dietz AB, Kaufmann TJ, Gustafson MP, Brown PD, Uhm JH, Rao RD, Doyle L, Giannini C, Jaeckle KA, Buckner JC (2010) Combination of temsirolimus (CCI-779) with chemoradiation in newly diagnosed glioblastoma multiforme (GBM) (NCCTG trial N027D) is associated with increased infectious risks. *Clin Cancer Res* **16**: 5573–5580.

Schuler W, Sedrani R, Cottens S, Häberlin B, Schulz M, Schuurman HJ, Zenke G, Zerwes HG, Schreier MH (1997) SDZ RAD, a new rapamycin derivative: pharmacological properties in vitro and in vivo. *Transplantation* **64**: 36–42.

Shegal SN (2003) Sirolimus: its discovery, biological properties, and mechanism of action. *Transplant Proc* **35**(suppl 3): 7S–14S.

Tarhini A, Kotsakis A, Gooding W, Shuai Y, Petro D, Friedland D, Belani CP, Argiris A (2010) Phase II study of everolimus (RAD001) in previously treated small cell lung cancer. *Clin Cancer Res* **16**(23): 5900–5907.

Thomas GV, Tran C, Mellinghoff IK, Welsbie DS, Chan E, Fueger B, Czernin J, Sawyers CL (2006) Hypoxia-inducible factor determines sensitivity to inhibitors of mTOR in kidney cancer. *Nat Med* **12**: 122–127.

Tsukamoto T, Shinohara N, Tsuchiya N, Hamamoto Y, Maruoka M, Fujimoto H, Niwakawa M, Uemura H, Usami M, Terai A, Kanayama HO, Sumiyoshi Y, Eto M, Akaza H (2011) Phase III trial of everolimus in metastatic renal cell carcinoma: subgroup analysis of japanese patients from RECORD-1. *J Clin Oncol* **41**: 17–24.

Yao JC, Shah MH, Ito T, Bohas CL, Wolin EM, Van Cutsem E, Hobday TJ, Okusaka T, Capdevila J, de Vries EG, Tomassetti P, Pavel ME, Hoosen S, Haas T, Lincy J, Lebwohl D, Oberg K (2011) Everolimus for advanced pancreatic neuroendocrine tumors. *N Engl J Med* **346**: 514–523.