

Combined introduction of anti-IL2 receptor antibodies, mycophenolic acid and tacrolimus: effect on malignancies after renal transplantation in a single-centre retrospective cohort study

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Abstract

Background. Several studies suggest that the introduction of tacrolimus (TRL), mycophenolic acid (MPA) and interleukin 2 receptor antibodies (IL2Ra) as single drugs more than a decade ago has not increased the risk of malignancy after renal transplantation. However, only limited data are available on their carcinogenic effects when used in combination as a potent immunosuppressive regimen.

Methods. A retrospective single-centre cohort study on 929 adult renal transplant recipients. Investigation of the effect of two consecutive immunosuppressive regimens [1993–98, $N = 405$, anti-lymphocyte antibodies, cyclosporine and azathioprine (AZA); 1999–2007, $N = 524$, predominantly IL2Ra, TRL and MPA] on the incidence rate of skin cancer, solid tumours and post-transplant lymphoproliferative disease (PTLD).

Results. In total, 365 malignancies developed among 113 patients. As compared to the previous cyclosporine and AZA-based immunosuppression, the introduction of the new immunosuppressive regimen did not increase the incidence rate of skin cancer [rate ratio 0.84; 95% confidence interval (CI) 0.48–1.46], solid tumours (0.89; 95% CI 0.46–1.67) and PTLD (0.82; 95% CI 0.28–2.21). Patients treated with the more recent regimens less frequently developed multiple skin cancers and invasive squamous cell cancer. Skin cancer after transplantation was strongly associated with the development of solid tumours (odds ratio 5.2; $P < 0.0001$). The introduction of the new immunosuppressive drugs reduced the incidence of first year acute rejection from 34.8 to 13.2% ($P < 0.0001$).

Conclusion. Although significantly more efficient in the prevention of acute rejection, the introduction of TRL, MPA and IL2Ra-based immunosuppression after kidney transplantation was not associated with an increased incidence of skin cancer, solid tumours or PTLD.

Keywords: immunosuppression; kidney transplantation; malignancy; mycophenolic acid; tacrolimus

Introduction

Immunosuppression after kidney transplantation is associated with a major increase in the risk of malignancy [1, 2]. Skin cancer is the most commonly encountered malignancy in renal transplant recipients with up to 100-fold increase in risk as compared to an age-adjusted general population [1]. The risk increase is constant over time and long-term cohort studies have documented a 75% incidence of skin cancer and a 33% incidence of non-skin cancer after 30 years of transplantation [3]. Non-Hodgkin lymphoma is the second most common malignancy in kidney transplant recipients with a relative risk of up to 40% as compared to the general population [1]. Post-transplant malignancy is a cause of significant morbidity, can lead to graft loss when immunosuppression is tapered and results in cancer-related death in up to 40–50% of patients with solid tumours or lymphoma [4–6].

During the second half of the 1990s, a new standard regimen, combining induction therapy with interleukin 2 receptor antibodies (IL2Ra) (basiliximab or daclizumab), the calcineurin inhibitor tacrolimus (TRL) and the anti-metabolite agent mycophenolic acid (MPA), progressively replaced the previous combination of induction therapy with anti-lymphocyte antibodies such as thymoglobulin or the anti-CD3 ϵ monoclonal antibody (muromonab-CD3, OKT3®), the calcineurin inhibitor cyclosporine and the anti-metabolite azathioprine (AZA). The introduction of this new regimen was associated with a marked improvement in the immunosuppressive efficacy and reduced the incidence of acute rejection episodes after renal transplantation to <15% in the majority of centres [7, 8]. The increased immunosuppressive

effect of the new regimen has also been shown by the appearance of BK virus nephropathy, a new and previously unreported infectious complication in recipients or organ transplants [9].

The relation between immunosuppression and tumourigenesis is incompletely understood but increasing intensity of immunosuppression has been previously shown to increase the incidence of post-transplant malignancy in a prospective and controlled trial [10]. Several large registry analyses have investigated the effect of the new classes of immunosuppressants on post-transplant malignancy but did not observe an increase in non-Hodgkin lymphoma or solid tumours in transplant recipients treated with IL2Ra or MPA [1, 11–13]. However, these studies had some weaknesses: (i) most had a rather short follow-up time; (ii) outcome measures in large databases were not validated for accuracy; (iii) immunosuppressive drugs were considered individually and not as part of immunosuppressive regimens; (iv) studies did not document whether patients treated with the more recent drugs indeed received a higher immunosuppressive load as indicated by lower rates of acute rejection episodes and graft loss from rejection and (v) only very limited data are available on the impact of the new immunosuppressive drugs on skin cancer.

The objective of our study was to address these issues in a retrospective cohort study investigating the effect of the current immunosuppressive regimens on the incidence rate of post-transplant malignancies.

Materials and methods

Pre- and post-transplant data from all renal transplantations performed in our centre between January 1993 and December 2007 were extracted from a computer database, which contains routinely collected data from all patients who received a renal transplant at the University of Brussels since 1965. We excluded paediatric transplantations (recipients of <18 years) and combined transplantations from the current analysis. The remaining 929 adult renal transplantations constitute the study population.

Post-transplant tumours

The occurrence of tumours after transplantation was the outcome of interest. The database records the medical history of tumours before transplantation and has a yearly data entry on tumours that develop after transplantation. Data on skin tumours in the database were also cross-checked for completeness with the patient files of the dermatologist who systematically follows all transplant recipients in our centre (V.D.M.). For all tumours identified in the database, one of the investigators (P.B.) conducted a detailed review of the patient's medical records to ascertain the type of malignancy [skin cancer, solid tumour or post-transplant lymphoproliferative disease (PTLD)]; the clinical and pathological tumour classification at diagnosis according to the American Joint Committee on cancer (AJCC) TNM staging system [14]; the type of treatment (surgery, radiotherapy, chemotherapy or other) and the clinical evolution (death from the disease, death from other cause, tumour remission, evolving tumour) in the patient's medical records. Only histologically diagnosed tumours were taken into account. Skin cancers were classified as *in situ* squamous cell carcinomas (SCCs), invasive SCCs, basal cell carcinomas (BCCs), melanomas, Kaposi's sarcomas (skin confinement or visceral involvement) or other. According to the AJCC histopathological grading system, SCCs and BCCs were graded in well differentiated (G1), moderately differentiated (G2) or undifferentiated (G4). PTLDD's were classified according to the World Health Organisation Classification (early lesions, polymorphic, monomorphic, other) [15]; localization (gastro-intestinal, pulmonary, central nervous system, kidney graft, liver or other); B- or T-cell neoplasms and Epstein-Barr virus (EBV) status. Finally, solid tumours were classified as kidney carcinoma, upper tract urothelial carcinoma, bladder carcinoma, colo-rectal carcinoma, breast carcinoma, lung

carcinoma, prostate carcinoma or other. Information on post-transplant malignancies was collected until either death with a functioning graft, returning to dialysis for >1 month or loss to follow-up occurred.

Immunosuppressive therapy

The type of immunosuppression regimen was the exposure of interest in this cohort study. Patients received different types of immunosuppression during the 15-year follow-up. During the first period from 1993 to 1998, the majority of patients received an immunosuppressive regimen based on induction with anti-CD3 monoclonal antibodies (muromonab-CD3, OKT3®), cyclosporine and AZA. During the year 1999, the three components of this regimen were progressively replaced. IL2Ra became the standard induction therapy, whereas thymoglobulin (ATG) use was restricted to patients at high immunological risk. AZA was replaced by MPA and TRL became the standard calcineurin inhibitor ($N = 407$ versus 127 patients treated with the microemulsion formulation of cyclosporine). Transplantations were therefore subdivided into two successive periods of immunosuppression from 1993 to 1998 ('<1999'; $N = 405$) and from 1999 to 2007 ('≥1999'; $N = 524$). All patients received methylprednisolone intravenously at the dose of 250 and 125 mg on the day of transplantation and the first post-operative day, followed by tapered doses as maintenance therapy previously described [8].

Data analysis

Categorical and continuous variables with or without normal distribution were summarized as proportions, means with SD and medians with the interquartile range, respectively. Hypothesis testing of associations was done with the chi-squared test, *t*-test and Wilcoxon rank-sum test.

Immunological graft loss was calculated by the Kaplan–Meier method, censoring graft losses due to causes unrelated to acute or chronic rejection, such as patient death, technical failure or recurrence of primary kidney disease.

The risk of skin cancer, solid tumour and PTLDD was compared by calculating the Kaplan–Meier estimates of the cumulative incidence. Due to variable follow-up times, the incidence rate of patients who develop the first tumour of interest per 1000 patient-years (py) at risk was calculated. Time at risk was defined as the delay between the date of transplantation and either time of tumour diagnosis, patient death, return to dialysis or loss to follow-up. As many patients developed multiple skin cancers, we also calculated the incidence rate of skin cancer without censoring the patient after the development of the first tumour. The effect of immunosuppression on the risk of post-transplant cancer was assessed by incidence rate ratios with 95% confidence intervals (CIs). The effect of immunosuppression on the occurrence of skin tumours after adjustment for other risk factors was assessed by multivariate negative binomial regression modelling. The effect of risk factors on the occurrence of a first tumour was calculated by using univariate and multivariate competing risk regression (CRR) modelling with the STATA *stcrreg* command. The proportional hazards assumption was tested using Schoenfeld residuals and by graphically displaying the plots of $\ln(-\ln(S))$ against $\ln(t)$. Interaction between variables was tested by creating product terms that were entered into the multivariate models. The Wald test of the corresponding regression coefficient was used for hypothesis testing.

Long-term survival in patients with tumours was estimated by the Kaplan–Meier method. A bilateral *P*-value of <0.05 was used to reject the null hypothesis in all hypothesis testing. All data management and statistical analysis were realized with STATA 11.

Ethical considerations

The procedures of data collection and measures taken to maintain data confidentiality in the database of renal transplant recipients have been reviewed and approved by the ULB Hôpital Erasme Ethics committee. Patients were informed at the moment of listing for renal transplantation that their medical data will be collected in the centre and Eurotransplant databases for medical and research purposes. The protocol of the present study was reviewed and approved by the ULB Hôpital Erasme ethics committee.

Results

Patients transplanted under the more recent immunosuppressive regimen were significantly older and had spent

less time on dialysis before transplantation (Table 1). Total time at risk for the overall cohort was 5896 years with 3577 years for patients transplanted before and 2319 years for patients transplanted from 1999 onwards. About 90% of patients received cyclosporine A (CsA) in combination with AZA in the period before 1999 and a calcineurin inhibitor in combination with MPA thereafter. Mammalian target of rapamycin (mTOR) antagonists were used in only three patients (0.7%) before 1999 but in 11.3% of transplantations thereafter. The use of the new immunosuppressive agents was associated with a highly significant reduction in first year acute rejection episodes from 141/405 (34.8%) to 69/524 (13.2%) [odds ratio (OR) 0.28; 95% CI 0.2–0.4; $P < 0.0001$]. The 5-year Kaplan–Meier estimate of graft loss from rejection was reduced from 13.5 to 8% in the more recent period [hazard ratio (HR) 0.53; 95% CI 0.35–0.79; $P = 0.002$].

Association of the new immunosuppressive regimen with the incidence of cancer after transplantation

In the overall population, 365 malignancies developed among 113 patients (12.2%; 95% CI 10.1–14.3%): 287 skin cancers (78.6%), 58 solid cancers (15.9%) and 20 post-transplant lymphoproliferative disorders or PTLD (5.5%). The development of at least one neoplasia after transplantation was associated with older age (48.2 ± 11.0 versus 42.9 ± 12.5 years in patients without neoplasia; $P < 0.0001$), a history of cancer before transplantation (7.1 versus 2.8%; $P = 0.018$) and Chinese herb nephropathy as primary kidney disease (11.5 versus 4.5%; $P = 0.002$). Treatment of acute rejection and use of mTOR antagonists were not associated with any type of cancer (data not shown).

Kaplan–Meier estimates of the cumulative incidence of skin cancer (Figure 1a), solid tumour (Figure 1b) and PTLD (Figure 1c) did not differ significantly during the two immunosuppressive periods. The Kaplan–Meier method can overestimate the cumulative incidence in the case that a large proportion of patients is censored because of competing outcomes. Univariate CRR with graft loss

and patient death as competing outcomes confirmed that the hazard of malignancy was not increased in the more recent period (CRR HRs of 0.65 for skin cancer, 0.77 for solid tumour and 0.98 for PTLD, $P > 0.05$ for all associations). To take into account the different length of follow-up during the two immunosuppressive periods, rates of cancer per 1000 py at risk and the corresponding rate ratios were calculated (Table 2). Although the incidence rates of a first skin cancer, solid tumour and PTLD did not differ significantly, patients less frequently developed multiple skin cancers during the more recent period (incidence rate ratio 0.66; 95% CI 0.51–0.85; $P = 0.001$).

Multivariate analysis by negative binomial regression modelling identified age at transplantation, male gender and the immunosuppressive period as independent risk factors for post-transplant skin cancer (Table 3). Age acted as a negative confounder because adjustment for age further reduced the incidence rate ratio of skin cancer in patients receiving the new immunosuppressive regimen to 0.22 (95% CI 0.09–0.54; $P = 0.002$). When the negative binomial regression model was restricted to 373 patients who received the TRL–MPA association as compared to 437 patients treated with CsA–AZA, the adjusted incidence rate ratio of skin cancer was 0.12 (95% CI 0.04–0.4; $P = 0.001$). Patient age and Chinese herb nephropathy were the only independent predictors for the development of solid tumours in the multivariate CRR model. Chinese herb nephropathy was associated with a >4-fold increase in the hazard of solid tumours. This was exclusively due to the known increase in urinary tract cancer [16, 17]. Twenty-one of 22 cancers that developed in the 50 patients with Chinese herb nephropathy were urinary tract cancers. The incidence of other solid tumours (2%) was comparable to patients with other nephropathies (3%).

Association of the new immunosuppressive regimens with the stage and clinical evolution of post-transplant tumours

Skin cancer. Most of the skin cancers were SCCs ($n = 178$; 62%), either *in situ* ($n = 130$; 45.3%) or invasive

Table 1. Main characteristics of kidney transplant recipients during two successive immunosuppressive periods

| Characteristics | <1999 (N = 405) | ≥1999 (N = 524) | P |
|---|------------------|------------------|---------|
| Male gender | 253 (62.5%) | 326 (62.2%) | 0.94 |
| Age at transplantation (years) ^a | 39.4 ± 10.9 | 46.8 ± 12.6 | <0.0001 |
| Retransplantation | 75 (18.5%) | 109 (20.8%) | 0.38 |
| Duration of dialysis (months) ^b | 33.5 (19.4–58.0) | 27.9 (13.6–49.6) | 0.002 |
| First-year acute rejection | 141 (34.8%) | 69 (13.2) | <0.0001 |
| History of cancer | 11 (2.7%) | 20 (3.8%) | 0.35 |
| Splenectomy | 15 (3.7%) | 20 (3.8%) | 0.93 |
| Chinese herb nephropathy | 34 (8.4%) | 16 (3.1%) | <0.0001 |
| Follow-up time (years) ^b | 8.8 (5.0–12.5) | 4.4 (2.3–6.5) | <0.0001 |
| Immunosuppressive agents ^c | | | |
| ATG–OKT3/IL2Ra/none (%) | 89.9/0/11.1 | 21.6/57.3/21.2 | <0.0001 |
| AZA/MPA (%) | 88.4/10.6 | 0.4/91.2 | <0.0001 |
| CsA/TRL (%) | 95.1/4.2 | 22.1/72.0 | <0.0001 |
| Sirolimus/everolimus (%) | 0.7/0 | 8.6/2.7 | <0.0001 |

^aMean ± SD.

^bMedian (interquartile range).

^cAccording to intention to treat.

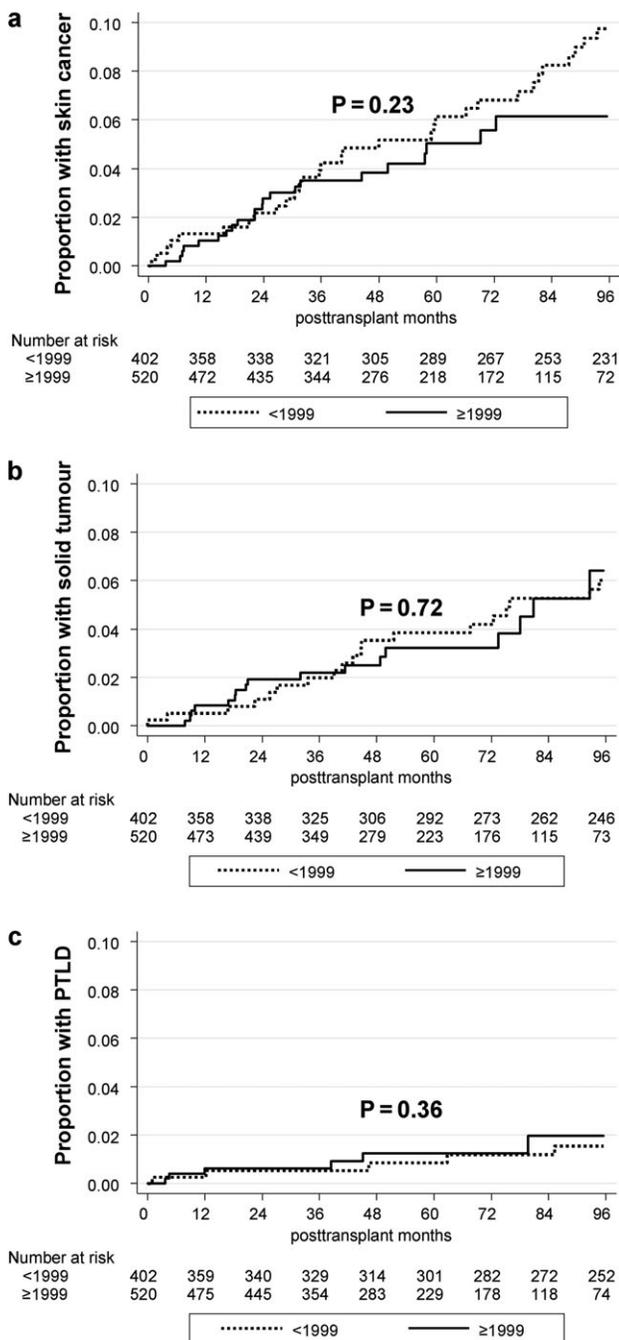


Fig. 1. Kaplan–Meier estimates of the cumulative incidence of cancer by immunosuppressive period up to 96 months after transplantation. Cumulative incidence of skin cancer (a), solid tumours (b) and post-transplant lymphoproliferative disease (c). Population at risk during the follow-up is tabulated below the graphs. Hypothesis testing by the log-rank test with $P > 0.05$ for all comparisons.

($n = 48$; 16.7%); ~80% ($n = 144$; 80.9%) were well differentiated on histological grading, whereas 19.1% ($n = 34$) were moderately differentiated or undifferentiated. BCCs were less common ($n = 80$; 27.9%) with a ratio of SCCs to BCCs of 2.2:1. The median number of tumours in patients with skin cancer per patient was 2 (range 1–43). The distribution of squamous cell skin cancers according to the immunosuppressive regimen is shown in Table 4. A higher

proportion of SCCs that developed in patients treated with CsA–AZA were invasive (31.5 versus 14.6%; $P = 0.02$). Among the 17 patients who developed one or several invasive SCCs, 3 died of a metastatic tumour infiltration (17.6%). In total, 18% (11/61) of patients with skin cancer also developed a solid cancer compared to only 4% (33/868) of patients without skin cancer (OR 5.2; 95% CI 2.5–10.9; $P < 0.0001$).

A total of 14 patients developed Kaposi's sarcoma and 4 patients melanoma without significant differences in stage at diagnosis and evolution between the two immunosuppressive periods.

Solid cancer. The most common solid cancers after kidney transplantation ($n = 58$) were bladder urothelial carcinoma ($n = 18$; 31%), upper-tract urothelial carcinoma ($n = 8$; 13.8%), colo-rectal cancer ($n = 4$; 6.9%), breast cancer ($n = 4$; 6.9%), kidney cancer ($n = 3$; 5.2%) and lung cancer ($n = 3$; 5.2%) (Supplementary table 1). Most solid tumours were diagnosed at a Stages I–II AJCC ($n = 37$; 63.8%), especially bladder urothelial carcinoma ($n = 15$; 83.3%) and upper-tract urothelial carcinoma ($n = 7$; 87.5%) which might be related to systematic screening cystoscopies and bilateral ureteronephrectomy in kidney transplant recipients with Chinese herb nephropathy [16]. Around 36% ($n = 21$; 36.2%) were detected at a late Stages III–IV, mostly colo-rectal cancer ($n = 3$; 75%), lung cancer ($n = 2$; 66.7%) and stomach cancer ($n = 2$; 100%). Overall, 24.6% of all solid tumours were diagnosed at a metastatic stage (Stage IV).

There were no significant differences between the number of solid cancer cases diagnosed at a late Stages III–IV AJCC TNM staging comparing the previous (35.1%) and more recent immunosuppressive periods (38.9%) ($P = 0.786$).

Among the 46 patients who developed 58 solid tumours, in total, 20 patients died of the disease (43.5%), 19 patients were in tumour remission (41.3%), 5 patients had an evolving tumour (10.9%) and 2 patients died of another cause (4.3%). Patient survival with solid cancer was 50.2% (95% CI 33–65%) at 5 years and 40.2% (95% CI 22–57%) at 10 years. Survival after development of a solid tumour tended to be lower in the more recent period [5-year survival of 48.1% (17.4–73.6) versus 73.5% (46.8–88.2); $P = 0.09$].

PTLD. The PTLD ($n = 20$) occurred in the gastro-intestinal tract in 45%, the central nervous system in 20% and the renal graft in 15% of the patients. Active replication of EBV was detected by polymerase chain reaction in 69.2% of 13 tested patients. B-cell lymphomas accounted for the great majority of PTLD in the renal transplant population (93.8%). Monomorphic (53.3%) and polymorphic (46.7%) PTLD were about equally frequent. Eight patients were treated successfully (40%), eight patients died of the disease (40%), three patients had an evolving tumour (15%) and one patient died of another cause (5%). Patient survival with PTLD was 41.6% (95% CI 14–67%) at 5 years. There were no significant differences in the incidence rate of monomorphic or polymorphic PTLD, detection of EBV and clinical outcomes between the two periods of immunosuppression.

Table 2. Incidence rates of malignancies after kidney transplantation during two successive immunosuppressive periods

| Incidence rate ^a | <1999 | ≥1999 | IRR ^b (95% CI) | P |
|---|-------|-------|---------------------------|-------|
| Skin cancer ^c | 11.5 | 9.7 | 0.84 (0.48–1.46) | 0.53 |
| Total number of skin cancers ^d | 56.2 | 37.1 | 0.66 (0.51–0.85) | 0.001 |
| Solid tumour ^c | 8.4 | 7.4 | 0.89 (0.46–1.67) | 0.71 |
| PTLD ^c | 3.7 | 3.0 | 0.82 (0.28–2.21) | 0.94 |

^an/1000 patient-years at risk.^bIncidence rate ratio.^cRate of patients developing first tumour. Time at risk censored after occurrence of first tumour of interest.^dTaking into account multiple tumours developing in the same patient.**Table 3.** Multivariate analysis of risk factors for skin cancer and solid cancers

| Risk factors | Skin cancers ^a | | Solid tumour ^b | |
|--------------------------------|---------------------------|---------|---------------------------|---------|
| | IRR (95% CI) | P | HR (95% CI) | P |
| Age at transplant (per year) | 1.11 (1.06–1.16) | <0.0001 | 1.03 (1.00–1.06) | 0.016 |
| Male gender | 2.81 (1.09–7.24) | 0.001 | 1.08 (0.54–2.20) | 0.83 |
| History of cancer | 0.53 (0.04–7.35) | 0.64 | 2.06 (0.76–5.58) | 0.154 |
| Chinese herb nephropathy | 1.11 (0.20–6.10) | 0.424 | 4.8 (2.14–10.75) | <0.0001 |
| Immunosuppressive period ≥1999 | 0.22 (0.09–0.54) | 0.002 | 0.71 (0.38–1.37) | 0.29 |

^aEffect of risk factors on incidence rate of skin cancers modelled by negative binominal regression; IRR, incidence rate ratio.^bEffect of risk factors on the time to occurrence of first tumour modelled by competing risk regression.**Table 4.** SCC type and grade of differentiation

| SCC type | <1999 | ≥1999 | P |
|--|-------------|------------|-------|
| SCC <i>in situ</i> | 89 (68.5%) | 41 (85.4%) | 0.024 |
| SCC invasive | 41 (31.5%) | 7 (14.6%) | |
| Grade of differentiation | | | 0.052 |
| Well differentiated (G1) | 110 (84.6%) | 34 (70.8%) | |
| Moderately differentiated or undifferentiated (G2 or G4) | 20 (15.4%) | 14 (29.2%) | |
| Total | 130 | 48 | |

Discussion

The present study shows that introduction of the current immunosuppressive regimens based on calcineurin inhibitors, MPA and induction therapy with IL2Ra antibodies did not increase the incidence rate of skin cancer, solid tumours or PTLD. Our observations are in accordance with several previous reports which documented no increase in the risk of PTLD and solid tumours by IL2Ra or MPA in recipients of solid organ transplants [1, 12, 13]. The present report complements these papers by focussing not on individual immunosuppressive agents but the effect of their combined use as what has become the current gold standard of immunosuppression [7]. By confirming in the present series that the newer drugs effectively provide more efficient immunosuppression in terms of acute rejection episodes and graft loss from rejection, our data challenge the commonly held belief that the risk of cancer is directly related to the overall immunosuppressive load.

In agreement with previous reports, the present study confirms that renal transplant recipients frequently develop multiple and invasive non-melanoma skin cancers [18–21] and that invasive SCC is complicated by tumour general-

ization and death in a significant proportion of patients [21, 22]. Furthermore, the development of skin cancer could be a marker for an increased risk to develop a solid tumour, as patients with skin cancers had a five times increased risk of developing a solid tumour. Interestingly, the incidence rate of multiple and invasive non-melanoma skin cancers was significantly lower during the more recent immunosuppressive period. The use of mTOR antagonists in a small proportion of patients during the more recent period is unlikely to explain the effect as it was also observed when the analysis was restricted to patients treated with TRL and MPA. Withdrawal of AZA from the current regimen could be instrumental because the drug generates mutagenic oxidative DNA lesions by enhancing the effect of ultraviolet A light [23]. A protective effect of MPA has also been suggested by a lower incidence of skin cancer in patients taking 3 g instead of 2 g of MPA per day [24] and by an inhibitory effect of MPA on tumour cell growth and angiogenesis *in vitro* [25].

mTOR antagonists inhibit tumour cell proliferation and angiogenesis and are associated with a lower incidence of cancer after organ transplantation [26, 27]. Conversion to mTOR antagonists has reduced the incidence of non-melanoma skin cancer in high-risk patients [28] but this approach might be unsafe in patients with suboptimal graft function or proteinuria [29]. Our data suggest that conversion from AZA to MPA might be an alternative option in this category of patients.

Our cohort has a high incidence of urinary tract cancer due to the consumption of Chinese herb preparations containing *Aristolochia* species that dramatically increase the risk of urothelial carcinoma [16, 17]. The present cohort with a long-term follow-up confirmed that the carcinogenic effect of aristolochic acid is limited to urinary tract tumours and does not affect the incidence of other solid tumours or skin cancer.

Twenty of the 929 patients in our cohort developed PTLD after transplantation. Previous large registry studies had reported a reduced incidence of PTLD with the use of IL2Ra instead of ATG [12, 30], MPA [12] and cyclosporine as compared to TRL [11]. We were unable to detect an effect of immunosuppression on the incidence of PTLD. This could reflect a lack of association as previously reported [13, 31] or be related to the relatively low incidence rate and insufficient statistical power to detect an association in our cohort.

Our study has several limitations. Firstly, potential risk factors for cancer, such as smoking, sun exposure, and a positive family history could not be assessed in the present retrospective dataset. It is, however, unlikely that these risk factors have changed substantially during the two consecutive periods. Secondly, data were analysed on an intention-to-treat basis that does not take into account subsequent changes in immunosuppressive agents. Differences in follow-up between patients treated with successive immunosuppressive regimens might interfere with the estimation of rates of cancer. We addressed this issue by calculating incidence rates taking into account time at risk and by using competing risk analysis because of censoring for competing outcomes. In spite of these precautions, we cannot completely rule out that the higher incidence of multiple and recurring skin cancers under the previous regimen is caused by more prolonged exposure to immunosuppression. These observations therefore have to be confirmed after a longer follow-up of patients treated with the more recent regimen. Finally, we identified age as a positive confounder during the more recent period but cannot rule out that measures of effect remain confounded by unidentified risk factors.

In conclusion, this study shows that the current immunosuppressive regimen based on TRL, MPA and IL2Ra induction therapy is not associated with an increased incidence rate of malignancy as compared to the previous regimen in spite of improved prevention of graft rejection. Furthermore, the current regimen is associated with a marked reduction in the incidence rate of skin cancer. This suggests that immunosuppressive regimens can differentially affect immunosuppressive mechanisms involved in graft rejection and tumour surveillance.

Supplementary data

Supplementary data are available online at <http://ndt.oxfordjournals.org>.

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Conflict of interest statement. None declared.

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