UNIVERSITÉ DE NANTES

_____

FACULTÉ DE MÉDECINE

_____

Année : 2020 N°

THÈSE

pour le

DIPLÔME D'ÉTAT DE DOCTEUR EN MÉDECINE

DES de Néphrologie

par

Thibault LETELLIER

Né le 29/08/1993 à Evry

_____

Présentée et soutenue publiquement le 10 décembre 2020

_____

Association entre l'utilisation d'hydrochlorothiazide et le risque de cancer cutané non-mélanome dans une population de patients transplantés rénaux

_____

Président : Monsieur le Professeur Jacques DANTAL

Membres du jury : Madame le Professeur Brigitte DRENO
Madame le Docteur Caroline VIGNEAU-VICTORRI
Monsieur le Docteur Guillaume LAMIRAULT

Directeur de thèse : Monsieur le Docteur Simon VILLE

UNIVERSITÉ DE NANTES

Remerciements

A Monsieur le Docteur Simon VILLE, directeur de thèse,
Merci de m'avoir proposé ce travail dès le début de mon internat, merci pour ton encadrement, ta réactivité, ta motivation, ta disponibilité et ton aide à toutes les étapes, dans la bonne humeur.

A Monsieur le Professeur Jacques DANTAL, président du jury,
Merci de votre enseignement au cours de mon internat, de votre supervision de ce travail et d'avoir accepté de faire partie de mon jury de thèse ainsi que de le présider.

A Madame le Professeur Brigitte DRENO, à Madame le Docteur Caroline VIGNEAU-VICTORRI, à Monsieur le Docteur Guillaume LAMIRAULT,
Merci de l'honneur que vous me faites en tant que membre du jury de thèse, a fortiori malgré les conditions sanitaires que l'on connaît.

A tous les médecins et professeurs qui m'ont encadré, particulièrement dans le service de Néphrologie, mais également en Cardiologie à la Roche sur Yon, en Médecine interne, en Médecine intensive – réanimation,
Merci de votre enseignement, de la formation que vous continuez à me dispenser, aussi bien sur le plan théorique que pratique.
Merci, et c'est le plus important pour moi, de votre disponibilité, de votre bonne humeur, de l'ambiance, c'est un plaisir d'apprendre et de travailler dans ces conditions.

A mes amis, que je ne citerai pas mais qui se reconnaîtront, aussi bien au lycée à Charleville-Mézières, au cours de mes premières années universitaires à Reims, et à l'internat à Nantes depuis 3 ans,
Merci pour votre soutien, et surtout merci pour tous les moments partagés, passés et futurs, essentiels pour moi.

A ma famille, particulièrement à mes parents et à ma sœur, Margaux,
Merci pour l'environnement de mon enfance, propice à la curiosité, à l'éducation mais aussi au rire et au divertissement.
Merci de m'avoir motivé pour les études, pour la médecine, puis encouragé en toutes circonstances, merci pour ce soutien sans faille, socle de tout le reste.

A Lauriane,
Merci pour ta présence au quotidien, pour ta patience en (presque) toute circonstance,
Merci pour ton soutien inébranlable, pour ta foi en moi démesurée,
Merci pout tout, en fait.

UNIVERSITÉ DE NANTES

Sommaire

I) Prérequis

L'insuffisance rénale chronique est un problème majeur de santé publique, en effet, elle affecte de 8 à 16% de la population mondiale et représente la 12ème cause de mortalité toutes causes confondues en 2017. (1–3)
Près de 3 millions de personnes dans le monde, au stade terminal de cette maladie, sont actuellement traités par une technique de suppléance rénale (hémodialyse, dialyse péritonéale ou transplantation rénale). (4)

Pour les patients éligibles, la transplantation rénale est à privilégier face aux autres techniques de suppléance, en raison d'une amélioration de la survie et de la qualité de vie à long terme, pour un coût inférieur. (5,6)

Cependant, dans l'optique de prévenir le rejet du greffon rénal, des thérapeutiques immunosuppressives sont nécessaires au long cours.
Le schéma thérapeutique de première intention recommandé en 2020 à la phase initiale d'une transplantation rénale est le suivant : (7)

- Traitement d'induction par un agent déplétant lymphocytaire (par exemple sérum antilymphocytaire de lapin) ou par antagoniste du récepteur à l'interleukine 2. (8,9)
- Traitement d'entretien, par l'association d'un inhibiteur de calcineurine (plutôt tacrolimus que ciclosporine) à un agent antiprolifératif (mycophenolate mofetil ou acide mycofenolique), et dans certains cas à une corticothérapie. (10,11)

Ces traitements ont des effets indésirables, dominés par l'augmentation du nombre d'infections, de leur gravité, et l'apparition d'infections opportunistes, qu'elles soient bactériennes, virales, fongiques ou parasitaires. (12)

La deuxième grande catégorie de pathologies favorisées par les immunosuppresseurs sont les cancers.
En effet, après 20 ans de transplantation, l'incidence cumulée de néoplasies solides est estimée à plus de 25%. (13)
De plus, il s'agit de la deuxième cause de mortalité après transplantation d'organe solide, derrière les maladies cardiovasculaires. (14)

Si pour certains cancers, déjà très fréquents, l'incidence semble être peu (cancer colorectal ou pulmonaire) ou pas (cancer du sein ou de la prostate) augmentée dans une population de transplantés, d'autres y sont très largement surreprésentés.
Il s'agit notamment des cancers favorisés par une infection virale comme les sarcomes de Kaposi, les cancers ano-génitaux ou encore les lymphoprolifération post-transplantation. (15)

Les cancers cutanés non mélanomes (CCNM), sont les néoplasies les plus fréquentes à la fois en population générale caucasienne, où leur incidence ne cesse d'augmenter, et parmi les transplantés. (16,17)

Les CCNM englobent différents types de cancers cutanés mais ils sont très majoritairement représentés par les carcinomes basocellulaires (CBC) et épidermoïdes cutanés (CEC).

Historiquement, le risque de CEC et de CBC en transplantation d'organe solide était estimé de 65 à 250 fois et de 10 à 16 fois celui de la population générale, respectivement. (18)
Plus récemment, ce sur-risque semble avoir diminué à 20 fois celui de patients non immunodéprimés, ce qui reste très élevé. (19,20)

Cette diminution depuis les années 1990 s'explique par plusieurs facteurs.
Le premier facteur est probablement le changement dans les thérapies immunosuppressives, et notamment l'arrêt de l'utilisation en première intention de l'azathioprine en transplantation rénale, au profit des dérivés du mycophenolate. (21,22)
Deux autres raisons possibles à cette diminution sont à la fois une meilleure prévention des facteurs de risque, notamment de l'exposition solaire, et un meilleur suivi dermatologique, par rapport à antérieurement, mais également en comparaison avec la population générale. (23)

A l'exception de l'immunosuppression, de nombreux facteurs sont susceptibles d'influencer la survenue des CCNM.
L'âge en est le principal, avec plus de 80% des cas survenant après 60 ans. Par ailleurs, les hommes ont une incidence et une prévalence plus élevées de CCNM. (24)

Un autre facteur de risque important, notamment en transplantation, est l'exposition au papillomavirus humain, détecté plus fréquemment dans les CCNM qu'en peau saine, et ce d'autant plus souvent qu'il s'agit de patients immunodéprimés. (25)

En tête de file des facteurs de risque modifiables se situe l'exposition solaire, et notamment aux rayons ultra-violets (UV). On retrouve en effet une association dose-dépendante entre l'exposition solaire et la survenue de CEC, tandis qu'une exposition aiguë répétée, a fortiori dans l'enfance, est plutôt associée à un risque de CBC. (26–28)
Cette association est d'autant plus marquée lorsqu'il s'y associe un phototype à risque, c'est-à-dire une peau et des yeux clairs, et surtout l'incapacité à bronzer. (29)

Les UVB sont responsables de dommages directs à l'ADN et à l'ARN par la formation de liens covalents entre les bases pyrimidiques, à l'origine de la création de photo-produits mutagènes. (30,31)
De leur côté, les UVA induisent des dégâts indirects à l'ADN via un stress photo-oxydatif et la formation d'espèces réactives de l'oxygène. (32)

En corrélation avec la toxicité cutanée des UV, l'utilisation d'un médicament photosensibilisant favorise également la survenue de CCNM. (33)

Une réaction de photosensibilité est définie par la survenue d'effets indésirables cutanés liés à l'association d'une prise médicamenteuse et de l'exposition au rayonnement solaire. (34)

Parmi ces réactions de photosensibilité, on distingue photo-toxicité directe du médicament, souvent dose-dépendante, et réaction photo-allergique, médiée par le système immunitaire, bien que la plupart des médicaments incriminés puissent induire les deux types de réaction. (35)

Après exposition solaire en présence d'un agent photo-toxique dans l'épiderme ou le derme, on observe la création d'espèces réactives de l'oxygène, pouvant entraîner des dommages cellulaires protéiques et lipidiques voire de l'ADN, responsables des lésions cutanées observées. (36)

Dans le cas d'une réaction photo-allergique, l'agent incriminé est modifié par le rayonnement, et agit ensuite comme un haptène. Cela entraîne une réaction d'hypersensibilité retardée, médiée par les cellules de Langerhans et le recrutement de cellules lymphocytaires T. (37)

Ces deux types de réaction peuvent promouvoir la carcinogénèse, via des dommages directs à l'ADN (mutagénèse photochimique) et via l'inflammation qui s'ensuit. (38,39)

Ces mécanismes sont résumés dans la **Figure 1.** (40)

Des exemples concrets à cette photocarcinogénèse ont été clairement identifiés. Le plus parlant, et le plus ancien, de ces exemples est très certainement celui de la PUVA-thérapie, qui consiste en l'administration d'un médicament photosensibilisant, le méthoxsalène, avant d'exposer la peau à des rayonnements UV-A. Cette thérapeutique a prouvé son efficacité depuis plus de 40 ans dans le traitement du psoriasis. (41)

Peu de temps après, il a été démontré que ce traitement était fortement associé à la survenue de CEC. (42)

Si le cas caricatural du méthoxsalène a permis de mettre en évidence le concept de photocarcinogénèse liée à une photosensibilisation induite par un médicament, de nombreux autres traitements sont aujourd'hui incriminés.

En transplantation, il existe principalement deux exemples concrets.
Le mieux étudié est l'azathioprine. Ce médicament immunosuppresseur, longtemps utilisé en première intention en transplantation d'organe solide, a en effet été clairement identifié comme ayant un effet photosensibilisant, favorisant l'apparition de CCNM. Cet effet est indépendant et synergique avec le sur-risque qu'induit l'immunosuppression elle-même. (43,44)

Plus récemment, c'est en transplantation pulmonaire qu'un autre médicament photosensibilisant a été clairement associé à la survenue de CCNM. Il s'agit du voriconazole, un antifongique largement utilisé après la greffe de poumons pour prévenir l'infection aspergillaire. (45)

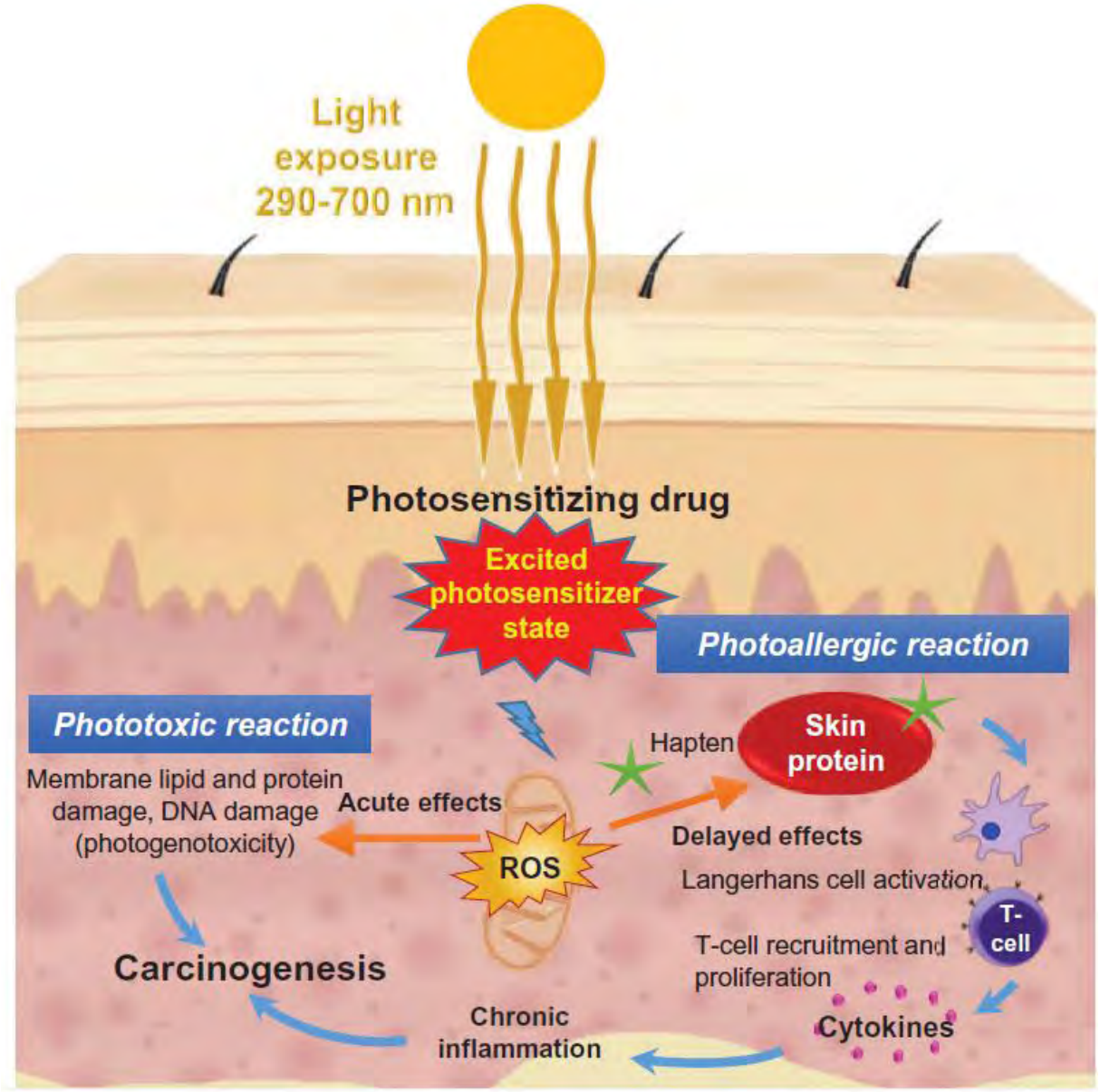

Figure 1. Mécanismes de la photosensibilisation induites par les médicaments. *(© Kreutz R et al. J Hypertens, 2019) (40)*
Un médicament photosensibilisant situé dans le derme ou l'épiderme est excité après absorption de photons de la longueur d'onde appropriée, ce qui génère des espèces réactives de l'oxygène. Celles-ci induisent des dommages cellulaires, causant des effets cytotoxiques directs (= réaction photo-toxique, à gauche). Par ailleurs, un médicament photosensibilisant peut agir comme un haptène qui se lie aux protéines de la peau en présence d'espèces réactives de l'oxygène, entrainant une réponse immunitaire cellulaire retardée (réaction photo-allergique, à droite). Un médicament photosensibilisant facilite donc la carcinogénèse à travers des lésions de l'ADN ou une inflammation chronique.
ROS : espèce réactive de l'oxygène.

En population générale, des études récentes ont mis en évidence une association entre un traitement par hydrochlorothiazide (HCTZ) et la survenue de CCNM.

Inventé en 1958, le chlorothiazide est le premier diurétique à avoir été commercialisé pour réduire la pression artérielle, mais également le premier traitement ayant démontré une amélioration de la morbidité cardiovasculaire lorsque la pression artérielle était effectivement diminuée. (46,47)

Aujourd'hui, les diurétiques thiazidiques, avec en tête de file l'HCTZ, sont toujours utilisés et recommandés parmi les traitements de première intention contre l'hypertension artérielle après plus d'un demi-siècle d'utilisation courante, d'études randomisées et de méta-analyses. (48)

Dans les années qui ont suivi son introduction, l'HCTZ a rapidement été identifié comme étant à l'origine de réactions de photosensibilité. (49,50)
Plus récemment, une augmentation de la toxicité à l'ADN des UV-A a été mise en évidence après la prise d'HCTZ. (51)
La photosensibilisation liée à un médicament est principalement due à la structure chimique de celui-ci, plutôt qu'à son action pharmacologique. Dans le cas de l'HCTZ, c'est le noyau sulfamide qui est le plus probablement en cause. (40)

II) Données cliniques

C'est à partir de 2008 que des études cliniques s'attachent à rechercher une association entre la prise d'HCTZ et la survenue de CCNM.
Les principales d'entre-elles sont présentées ici, par ordre chronologique de parution, ainsi que dans la **Table 1.**

1) Jensen AØ. et al, British Journal of Cancer, 2008 (52)

L'étude se plaçait en population générale dans une région danoise, où les cas de CBC, CEC et de mélanomes multiples étaient appariés à des contrôles sur l'âge, le sexe et la zone de résidence.

L'objectif principal de l'étude était de mettre en évidence l'association entre la prise de diurétiques et la survenue d'un cancer cutané. Les diurétiques étudiés comprenaient les diurétiques de l'anse (furosémide et bumétanide), les diurétiques épargneurs de potassium (amiloride et spironolactone) ainsi que les diurétiques thiazidiques ou apparentés (bendroflumethiazide, HCTZ et indapamide).

Cette étude mettait en évidence une association significative entre la prise d'hydrochlorothiazide et/ou d'amiloride avec la survenue de CEC mais pas de CBC. L'incidence rate ratio (IRR) d'apparition d'un CEC était de 1.58 (intervalle de confiance (CI) à 95% de 1.29 à 1.93) par 10 000 mg d'HCTZ (en dose totale cumulée).
Les autres diurétiques, et notamment les autres thiazidiques, n'étaient pas associés à un sur-risque de CCNM.
Les patients immunodéprimés n'étaient pas exclus, mais cette variable n'était pas incluse dans l'analyse multivariée.

2) Ruiter R. et al, European Journal of Cancer, 2010 (53)

Cette étude hollandaise n'a pas démontré de lien entre l'utilisation de thiazidiques et la survenue de CBC, parmi une cohorte de patients âgés déjà sélectionnée pour une autre étude.

3) De Vries E. et al, British Journal of Dermatology, 2012 (54)

De Vries et al se sont attachés en 2012 à rechercher les facteurs de risques associés à la survenue de CCNM et de mélanomes multiples, en incluant dans 11 centres européens les patients ayant un diagnostic de cancer cutané et en les comparant à une population contrôle.

Les données étaient recueillies prospectivement sous la forme d'auto-questionnaires, notamment concernant l'exposition solaire, les habitudes alimentaires, et la prise de certains médicaments dont des diurétiques thiazidiques (bendroflumethiazide uniquement) pendant au moins 3 mois.
Les patients sous immunosuppresseurs étaient exclus.

Dans cette étude, la prise de bendroflumethiazide (sous la forme d'une variable qualitative binaire) était associée à un sur-risque de survenue de CEC avec un odds ratio (OR) à 1.66 (95% CI de 1.16 à 2.37), sans majorer le risque de CBC.

4) Schmidt SAJ. et al, Journal of the European Academy of Dermatology and Venereology, 2015 (55)

Cette étude cas-témoin comparait l'utilisation de médicaments antihypertenseurs chez des cas de mélanomes multiples et de CCNM avec des contrôles appariés sur l'âge, le sexe et la région de résidence, dans le nord du Danemark.
Les patients immunodéprimés étaient exclus.
L'utilisation d'une combinaison de diurétiques associant un thiazidique et un épargneur de potassium était associée à un sur-risque de CEC avec un OR à 2.68 (95% CI de 2.24 à 3.21).
Cette association n'était pas retrouvée avec les thiazidiques seuls, ou pour le CBC.

5) Nardone B. et al, Drug Safety, 2016 (56)

Cette étude américaine sur le sujet concernait la survenue de mélanomes multiples et de CCNM, à la fois sous inhibiteurs de l'enzyme de conversion de l'angiotensine, antagonistes du récepteur de l'angiotensine II ou diurétiques thiazidiques.
L'information disponible était celle de la classe de médicaments, et non celle d'un médicament en lui-même.
Il s'agissait d'une étude de cohorte rétrospective sur un design exposé / non-exposé, concernant uniquement les patients blancs non-hispaniques, et où les transplantés d'organe solide étaient à nouveau exclus.

Après analyse multivariée, la survenue de CBC et CEC était favorisée par un traitement par thiazidiques avec des OR à 2.11 (95% CI de 1.60 à 2.79) et à 4.11 (95% CI de 2.66 à 6.35) respectivement, après ajustement sur l'âge, sexe, la race, le Charlson comorbidities index et la durée de suivi.

6) Su KA. et al, British Journal of Dermatology, 2018 (57)

Il s'agit d'une deuxième étude américaine sur une cohorte de patients hypertendus, dans laquelle les auteurs ont voulu mettre en évidence l'association entre la prise de médicaments antihypertenseurs photosensibilisants et la survenue de CEC.
Sur la base de la littérature et de la décision des auteurs, ces médicaments incluaient les diurétiques (épargneurs de potassium, de l'anse, ou thiazidiques) ainsi que les agonistes des récepteurs α2 adrénergiques.

De la même façon que précédemment, l'étude portait uniquement sur des patients blancs non-hispaniques, et les différentes causes d'immunodépression, telle que la transplantation d'organe solide, étaient des critères d'exclusion.

En comparaison avec les autres patients hypertendus de la cohorte, l'utilisation de médicaments photosensibilisants augmentait le risque de CEC, avec un hazard ratio (HR) ajusté à 1.17 (95% CI de 1.07 à 1.28).
L'analyse multivariée était ajustée sur l'âge, le sexe, le tabagisme, les comorbidités (Charlson comorbidities index), l'antécédent de CEC et l'antécédent d'utilisation d'antihypertenseur photosensibilisant.

Cependant, l'utilisation d'un diurétique thiazidique seul n'était pas associée à la survenue de CEC, mais uniquement l'utilisation d'une combinaison d'antihypertenseurs contenant un thiazidique.

7) Pedersen SA. et al, Journal of the American Academy of Dermatology, 2018 (58)

Ce travail de Pedersen et al est à ce jour l'étude la plus importante publiée sur le sujet. Leur objectif principal était de démontrer l'association entre CCNM et diurétiques thiazidiques.

A l'aide des bases de données nationales exhaustives du Danemark, l'étude portait sur l'ensemble de la population générale de ce pays, à l'exclusion, une fois encore, de toutes les formes d'immunodépression, et notamment des patients transplantés d'organes solides.
Les autres critères d'inclusion étaient d'avoir vécu au moins 10 ans au Danemark avant la date index (date du diagnostic du CCNM), et l'absence d'antécédent de CCNM.

Les auteurs identifièrent 71 533 CBC et 8 629 CEC entre janvier 2004 et décembre 2012. Chaque cas fut apparié à 20 contrôles sur l'âge et le sexe.
L'analyse statistique multivariée était ajustée sur l'âge, le sexe, l'utilisation d'autres médicaments photosensibilisants, certaines pathologies chroniques (diabète, insuffisance rénale chronique, broncho-pneumopathie chronique obstructive et consommation d'alcool abusive), le Charlson comorbidities index et le niveau d'éducation.

L'utilisation d'HCTZ (variable binaire) était associée à la survenue de CBC et de CEC avec des OR à 1.08 (95% CI de 1.05 à 1.10) et à 1.75 (95% CI de 1.66 à 1.85), respectivement.

Une des principales forces de cette étude, du fait du très grand nombre de patient, était l'étude dose-dépendante de cette association.
En effet, pour une dose que les auteurs définissaient comme élevée (dose cumulée > 50 000 mg), les OR étaient de 1.29 (95% CI de 1.23 à 1.35) et 3.98 (95% CI de 3.68 à 4.31) pour carcinomes basocellulaires et épidermoïdes, respectivement.

Cette association dose-dépendante est représentée graphiquement sur la **Figure 2.**

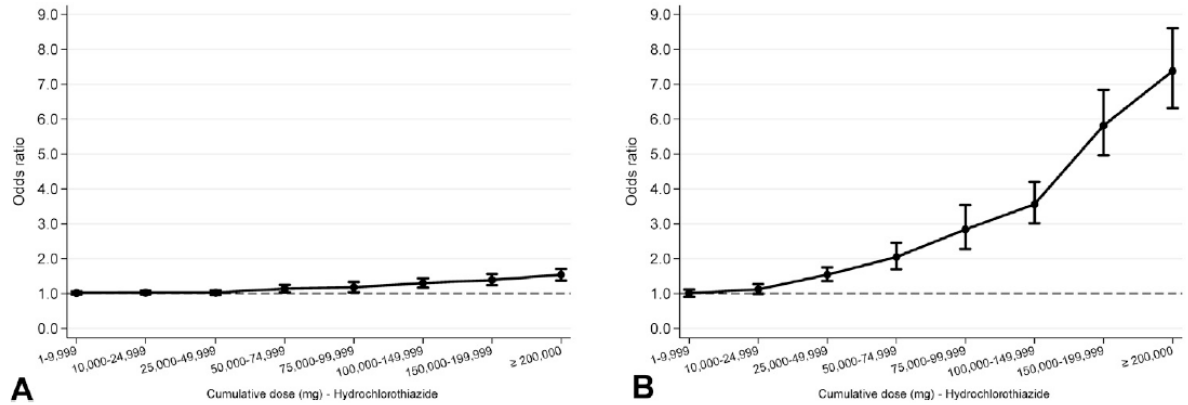

Figure 2. Modèle dose-dépendant entre la dose cumulée d'HCTZ et le risque de carcinome basocellulaire (A) et de carcinome épidermoïde (B). Les barres d'erreur représentent les intervalles de confiance à 95%. (© Pedersen SA. et al, J Am Acad Dermatol, 2018. *(58))*

Les auteurs ont également pratiqué des analyses similaires concernant d'autres diurétiques thiazidiques ou apparentés : le bendroflumethiazide et l'indapamide. Les résultats n'étaient pas significatifs.

S'il existe bel et bien dans cette étude une association dose-dépendante entre prise d'HCTZ et survenue de CBC, on remarque que le sur-risque semble bien supérieur pour les CEC.

Cela peut s'expliquer par la physiopathologie de survenue de ces cancers par rapport à l'exposition solaire. En effet, comme susmentionné l'apparition de CBC semble être plutôt favorisée par une exposition aiguë dans l'enfance, logiquement peu potentialisée par un médicament antihypertenseur, utilisé à l'âge adulte.

8) Pottegård A. et al, British Journal of Cancer, 2019 (59)

Cette étude menée sur la population taïwanaise reprend le même schéma que la précédente, c'est-à-dire les cas de CCNM appariés à des témoins, parmi lesquels on recensait l'exposition à l'HCTZ.

Les critères d'inclusion et d'exclusion étaient les mêmes, ainsi que les variables sur lesquelles l'analyse multivariée était ajustée. Les patients transplantés étaient donc exclus également.
Une différence importante était le regroupement des cas de CBC et de CEC sous une même entité (CCNM).

Les auteurs mettaient en évidence une association significative entre l'exposition à l'HCTZ et la survenue de CCNM, avec un OR ajusté très légèrement augmenté à 1.10 (95% CI de 1.06 à 1.14), et sans dose-dépendance.

Ces résultats sont peut-être partiellement dus au regroupement de CBC et de CEC, mais s'expliquent probablement plutôt par un effet différent des médicaments photosensibilisants dans une population non caucasienne, par conséquent de phototype moins sensible aux UV.
Il s'agit en effet de la première étude à ce jour s'intéressant à une population non européenne ou non blanche non-hispanique américaine.

9) Morales DR. et al, British Journal of Clinical Pharmacology, 2020 : (60)

Le schéma de cette étude anglaise est également le même que celui de l'étude danoise de Pedersen et al, en termes de critères d'inclusion et d'exclusion, de recrutement, de schéma d'étude (cas/témoins) et de variables utilisées en analyse multivariée.
La seule différence était le type de cancers étudiés, qui incluaient CCNM (CBC et CEC séparément), mélanomes multiples, cancers de la lèvre, et de la cavité orale.

L'IRR ajusté de survenue de CBC et CEC sous HCTZ était de 1.10 (95% CI de 1.03 à 1.17) et 1.25 (95% CI de 1.05 à 1.48), respectivement.
Une dose-dépendance était également observée.

***Table** 1. Caractéristiques des études observationnelles recherchant une association entre diurétiques thiazidiques et CCNM.*

Premier auteur	Année	Pays	Type d'étude	Médicament(s)	Carcinome basocellulaire	Carcinome épidermoïde
					Mesure d'association (CI 95%)	Mesure d'association (CI 95%)
Jensen (52)	2008	Danemark	Cas-témoin	Hydrochlorothiazide	IRR : 1.05 (0.95-1.16)	**IRR : 1.58 (1.29-1.93)**
				Bendroflumethiazide	IRR : 0.98 (0.90-1.06)	IRR : 1.03 (0.86-1.22)
				Indapamide	IRR : 0.99 (0.63-1.56)	IRR : 1.20 (0.57-2.54)
Ruiter* (53)	2010	Pays-Bas	Cohorte	Diurétiques thiazidiques	HR : 1.00 (0.95-1.05)	ND
de Vries* (54)	2012	Europe	Cas-témoin	Bendroflumethiazide	OR : 1.27 (0.92-1.75)	**OR : 1.66 (1.16-2.37)**
Schmidt* (55)	2015	Danemark	Cas-témoin	Diurétiques thiazidiques	OR : 1.05 (1.00-1.11)	OR : 1.03 (0.91-1.17)
Nardone* (56)	2016	Etats-Unis	Cohorte	Diurétiques thiazidiques	**OR : 2.11 (1.60-2.79)**	**OR : 4.11 (2.66-6.35)**
Su* (57)	2018	Etats-Unis	Cohorte	Diurétiques thiazidiques	ND	HR : 1.09 (0.99-1.19)
Pedersen* (58)	2018	Danemark	Cas-témoin	Hydrochlorothiazide	**OR : 1.08 (1.05-1.10)**	**OR : 1.75 (1.66-1.85)**
				Bendroflumethiazide	**OR : 1.03 (1.01-1.05)**	OR : 1.02 (0.97-1.08)
				Indapamide	OR : 0.99 (0.92-1.07)	OR : 0.95 (0.79-1.15)
Pottegård* (59)	2019	Taïwan	Cas-témoin	Hydrochlorothiazide	Cancer cutané non mélanome **OR : 1.10 (1.06-1.14)**	
				Indapamide	OR : 0.98 (0.84-1.18)	
Morales* (60)	2020	Angleterre	Cas-témoin	Hydrochlorothiazide	**IRR : 1.10 (1.03-1.17)**	**IRR : 1.25 (1.05-1.48)**

CCNM : cancer cutané non mélanome ; CI : intervalle de confiance ; IRR : incidence rate ratio ; HR : hazard ratio ; OR : odds ratio

*Les patients immunodéprimés étaient exclus.

A la lumière de ces différentes études cliniques, l'agence internationale de recherche sur le cancer (International Agency for Research on Cancer) a identifié l'HCTZ comme étant possiblement carcinogène (grade 2B).

Cela a également mené la plupart des agences sanitaires internationales à publier des alertes de sécurité ou des mises à jour concernant ce sur-risque de CCNM. C'est le cas de l'ANSM (Agence Nationale de Sécurité du Médicament et des produits de santé) en France, de l'EMA (European Medicines Agency) en Europe ou encore de la FDA (Food and Drug Administration) aux Etats-Unis. (61–63)

A ce jour, aucune étude ne s'est intéressée à l'effet de cette exposition à l'HCTZ chez des patients immunodéprimés, transplantés d'organe solide et déjà à risque très élevé de CCNM.

Nous avons donc décidé de conduire notre propre travail sur la cohorte de transplantés rénaux et pancréatiques à Nantes, via DIVAT (cohorte prospective, multicentrique et observationnelle de patients transplantés rénaux).

III) Introduction

In recent decades, kidney transplant outcomes have markedly progressed. However, the use of immunosuppressive therapy, essential for preventing graft rejection, is associated with a significant adverse event profile for patients. Aside from infection, cancer is the main adverse event associated with the use of immunosuppressive agents in solid organ transplant recipients.(13,64)

Keratinocyte cancers, namely squamous-cell carcinomas and basal-cell carcinomas, accounts for more than 90% of skin cancers in transplant recipients. These typically affect more than half of patients over time, occurring 65 to 250 times and 10 to 16 times more frequently than in the general population respectively. (19,22)
In addition, squamous-cell carcinomas typically follow a more aggressive course in transplanted patients, resulting in significant morbidity and mortality.

Aside from immunosuppressive therapy, risk factors for keratinocyte cancer reported in this population include old age, male sex and fair skin type. Whilst cumulative sun exposure is a well-established risk factor for squamous-cell carcinomas, the pattern of sun exposure associated with basal-cell carcinomas is less clear. (17)

For some time, photosensitizing drugs have been suspected to favor the development of squamous-cell carcinomas by potentiating the carcinogenic effect of ultraviolet (UV) radiation.

A recent study conducted on the general population has supported an association between use of hydrochlorothiazide (HCTZ), one of those well-known photosensitizing drugs, and risk of keratinocyte cancer, especially squamous-cell carcinomas. (58)
As the study's focus was on the general population, high-risk groups, including transplant recipients and immunosuppressed patients, were excluded. (58)

We aimed to investigate whether HCTZ use is associated with keratinocyte cancer in transplant recipients on immunosuppressive therapy independently of other known risk factors. We conducted a single-center study using a cohort of kidney, combined kidney pancreas and pancreas transplant recipients.

IV) Méthodes

1) Patient population

We conducted a single-center study on all consecutive adult patients transplanted with a graft that functioned for at least 3 months (kidney, pancreas or combined kidney–pancreas) at Nantes University Hospital between January 1, 2000 and December 31, 2017.

We excluded patients who had been recipients of 3 or more transplantations, as well as those with a prior history of basal-cell carcinoma or squamous-cell carcinoma. We also excluded those on initial maintenance immunosuppressive regimens that included mTOR inhibitors or azathioprine, as these treatments have been implicated as protective and contributing factors to keratinocyte cancers respectively (**Figure 3**).
Transplant patient otherwise received standard medical care

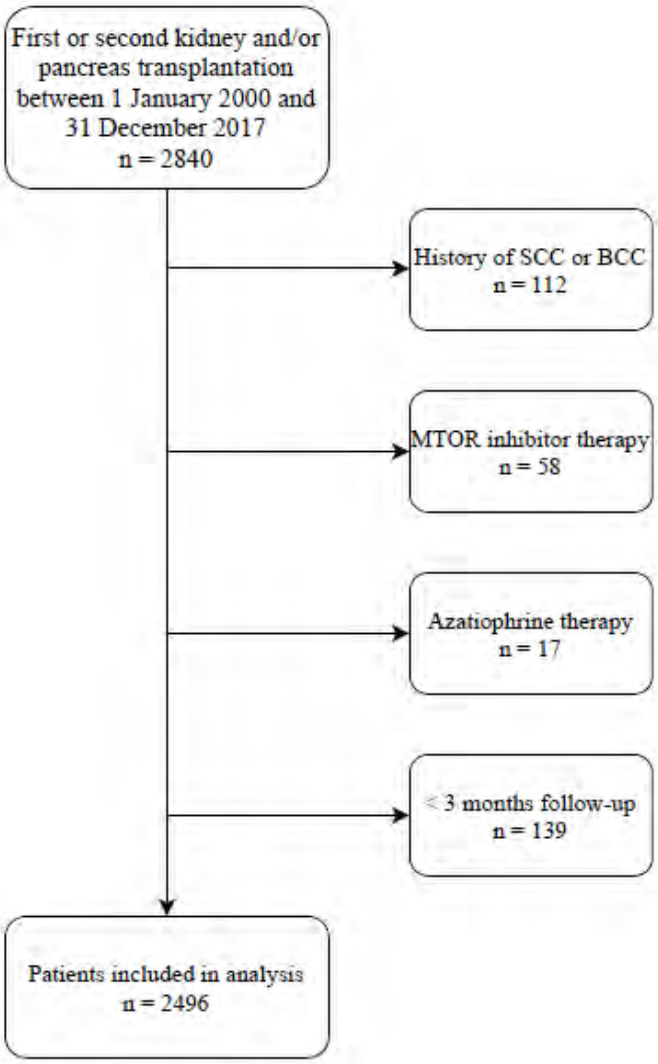

Figure 3. Flowchart of eligibility criterion.

All data were extracted from the French, multi-center, observational and prospective DIVAT cohort of transplanted patients (www.divat.fr).

The clinical and research activities reported are consistent with the Principles of the Declaration of Istanbul as outlined in the 'Declaration of Istanbul on Organ Trafficking and Transplant Tourism'.

More specifically, the DIVAT cohort received CNIL (Commission Nationale de l'Informatique et des Libertés) final agreement (No914184), and the CCTIRS (Advisory Committee on Information Processing in Material Research in the Field of Health) approved the use of its data for scientific research purposes.

2) Immunosuppression

Before 2016 all patients with low-immunological risk received induction immunosuppression with 20 mg of basiliximab at day 0 and day 4 (Simulect, Novartis, Basel, Switzerland) and a 250-mg bolus of methylprednisolone followed by standard post-transplant immunosuppression including calcineurin inhibitor, namely tacrolimus (trough level between 6 and 10 ng/dl) or cyclosporine (trough level between 125 and 200 ng/ml) and mycophenolate mofetil (500–1000 mg/BID) or mycophenolic acid (360–720 mg/BID).

Patients of high-immunological risk (positive reactivity against panel-reactive antibody (PRA) > 75%) and combined kidney–pancreas recipients received induction immunosuppression with rabbit antithymocyte globulin (Thymoglobulin, Genzyme, Cambridge, MA, USA) 6 mg/ kg and a 250-mg bolus of methylprednisolone followed by triple immunosuppression including calcineurin inhibitor, an anti-proliferative drug, and prednisone.

Our standard protocol planned withdrawal of steroids between 1 and 3 months, but some patients remained on triple therapy (rejection and/or high-immunological risk patients) or dual therapy with calcineurin inhibitor and steroids in case of withdrawal of anti-proliferative drug due to poor clinical tolerance and/or infections.

After 2016 patient of low-immunological risk received a low dose (3 mg/Kg) of rabbit antithymocyte globulin as induction therapy instead of basiliximab.

3) Available data

Recipient characteristics were age, gender, transplantation rank (first transplantation or re-transplantation), the initial renal disease (possibly recurrent or not), the renal replacement therapy. Donor feature was the donor type (living or deceased). Baseline transplantation parameters were transplantation type, cold ischemia time, number of HLA A-B-DR incompatibilities, induction therapy, maintenance treatment, use of steroids and occurrence of delayed graft function. Post-transplantation parameters were acute rejection, other malignancies and maintenance treatment exposure.

All patients underwent a dermatological examination undertaken by a specialist prior to the transplant and, in accordance with guidelines regarding outpatient surveillance of kidney transplant recipients, every year after the transplant. Histologically confirmed keratinocyte cancers were entered into the database.
Regarding HCTZ exposure, we extracted the list of patients prescribed HCTZ in the post-transplant period using prescription data collected longitudinally contemporaneously in our electronic system. We reviewed each prescription for accuracy in terms of start and stop dates (considered meaningful from one month of exposure) and dose. The follow-up and the collection of data ceased upon a patient's return to dialysis or at death.

4) Statistical analyses

We considered the transplant as the statistical individual.
Outcomes were defined by time from transplantation to the first occurrence of a squamous-cell carcinoma and basal-cell carcinoma.
Participants who did not have an outcome were censored at graft failure (preemptive re-transplantation, return to dialysis or death), when they were lost to follow-up or at the end of the study, whichever came first.

The cumulative incidence curves of HCTZ, squamous-cell carcinoma and basal-cell carcinoma were obtained using the Aalen-Johansen estimator, using graft failures as competing events. (65)
The median follow-up time was estimated using the reverse Kaplan-Meier. (66)

Multivariable cause-specific time-varying Cox models were used to estimate the relationship between HCTZ and the hazard of squamous-cell carcinoma and basal-cell carcinoma, with HCTZ designated as time-dependent variable as is usually recommended to prevent "immortal time" bias in the analysis of treatment effect in observational studies. (67,68)

Graft failures were right-censored. (69) In our main analysis, we studied the dichotomized variable of cumulative duration since the start of HCTZ. Indeed, construing HCTZ exposure as a time-dependent variable allowed patients, initially all in the untreated group, to move and remain into the treatment group from when HCTZ was commenced.

The hazard proportionality assumption was verified from the Schoenfeld residuals. (70) For baseline continuous covariates, the log-linearity assumption has been checked in unadjusted analysis if the Bayesian Information Criterion was not reduced using natural spline transformation compared to the inclusion of the covariate in its natural scale.

Time-dependent covariates related to immunosuppression, were based on one year-time windows. Patients were categorized as exposed for post-transplantation periods during which they received the treatment, and unexposed during other periods. (71,72)

Based on theoretical considerations and known biological effects, we included a priori the following variables in the multivariable models : (i) baseline parameter : age, sex, re-transplantation, type of transplantation, type of donor, HLA-A -B -DR mismatch ≥ 4, induction therapy, maintenance treatment at transplantation and use of steroids at transplantation ; (ii) time-varying covariates : rejection, maintenance treatment during follow-up (calcineurin inhibitor, mycophenolate derivatives, mTOR inhibitor) and other malignancies.

We also considered covariates that were significantly associated with the outcomes ($p < 0.1$) in unadjusted models. We did not consider interaction term. Patients who had data missing for covariates retained in the multivariable models were excluded.

A series of sensitivity analyses were performed.

Firstly, we estimated the effect of HCTZ use on different subgroups of patients : i) solitary kidney transplant recipients, ii) patients receiving depleting induction therapy, iii) patients receiving non-depleting induction therapy iv) patient receiving cyclosporine at transplantation and v) patients receiving tacrolimus at transplantation.

Secondly, in order to explore a potential dose-response relationship, considering that HCTZ doses remained relatively stable regardless of when it was initiated post transplantation, we initially studied the cumulative duration of HCTZ. However, as the log-linear association between the cumulative duration of use and outcomes could not be demonstrated (likely due to the low number of patients exposed to HCTZ combined with a relatively rare occurrence of events), such analysis was impracticable.

We then categorized exposure to HCTZ according to three categories. In the corresponding multivariable Cox models, the time-dependent variable associated with HCTZ use was ‘no use’, ‘mild-term use’ when the cumulative HCTZ use was < 2 years and ‘long-term use’ when it was ≥ 2 years.

We used R version 3.6.1 and the package ‘base’, ‘dplyr’, ‘survival’, ‘etm’, ‘plotrix’, ‘splines’, ‘lattice’, ‘prodlim’, ‘forestplot’, ‘ReporteRs’ packages for all data analyses.

V) Results

1) Recipient demographic characteristics

We included 2496 recipients of kidney (n=2155), combined kidney/pancreas (n=282) and pancreas transplants (n=59). Patient characteristics subdivided by HCTZ status are displayed in **Table 2.**

Prior to transplantation, no patient was receiving HCTZ, which was consequently started after transplantation in all 279 (11%) participants exposed at least one month to HCTZ during transplantation period.

Probabilities of exposure to HCTZ at 5, 10 and 15 years post-transplantation were **10%** (95% confidence interval (CI) from 8 to 11%), **14%** (95% CI from 12 to 15%) and **16%** (95% CI from 14 to 18%), respectively (**Figure 4**).

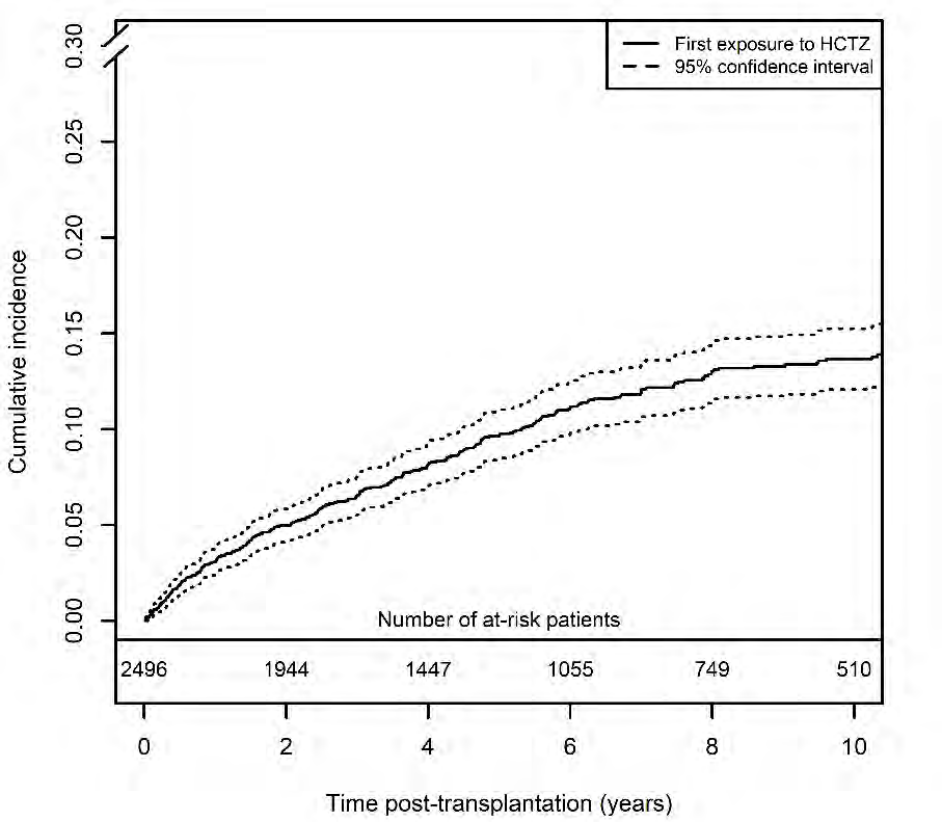

Figure 4. Cumulative incidence curve of Hydrochlorothiazide *(Aalen-Johansen estimator, re-transplantations, returns to dialysis and deaths as competing events).*

Mean duration of exposure and cumulative dose of HCTZ was **2.5 years** (95% CI from 2.1 to 2.8%) and **14436 mg** (95% CI from 12238 to 16634mg) respectively. Median follow-up time was **5.9 years** (range from 0.2 to 18.8 years).

During the follow-up, 343 deaths with a functioning graft, 439 returns to dialysis, 154 basal-cell carcinomas and 132 squamous-cell carcinomas occurred.

UNIVERSITÉ DE NANTES

Table 2. *Characteristics of 2496 transplant recipients included in the analysis according their exposure to Hydrochlorothiazide (HCTZ) during follow-up.*

	Whole sample (n=2496)			no HCTZ (n=2217)			HCTZ (n=279)		
	NA	**M**	**SD**	**NA**	**m**	**SD**	**NA**	**m**	**SD**
Recipient age	0	49	14	0	49	14	0	50	13
	NA	**n**	**%**	**NA**	**n**	**%**	**NA**	**n**	**%**
Male sex	0	1538	62	0	1353	61	0	185	66
Initial nephropathy	0			0			0		
Chronic tubulo-interstitial nephritis/ADPKD/Congenital uropathy		962	39		880	40		82	29
Diabetes		467	19		413	19		54	19
Glomerulopathy		643	26		567	26		76	27
Other		232	9		206	9		26	9
Vascular nephropathy		192	8		151	7		41	15
Kidney replacement therapy	46			41			5		
Hemodialysis		1757	72		1544	71		213	78
Peritoneal dialysis		217	9		200	9		17	6
Preemptive transplantation		476	19		432	20		44	16
Type of transplantation	0			0			0		
Combined kidney-pancreas		282	11		254	11		28	10
Kidney		2155	86		1909	86		246	88
Pancreas		59	2		54	2		5	2
Retransplantation	0	436	17	0	399	18	0	37	13
Deceased donor	0	2200	88	0	1948	88	0	252	90
HLAA B Dr mismatches >= 4	2	1264	51	2	1113	50	0	151	54
Cold ischemia time >= 18 hours	21	1018	41	20	882	40	1	136	49
Delayed graft function	78	811	34	73	705	33	5	106	39
Rejection	0	301	12	0	264	12	0	37	13
Other malignancies	0	193	8	0	163	7	0	30	11
Induction treatment	0			0			0		
Depleting		1219	49		1098	50		121	43
Non-depleting		1213	49		1066	48		147	53
None		64	3		53	2		11	4
Calcineurin inhibitors	0			0			0		
Cyclosporine		361	14		303	14		58	21
Tacrolimus		2108	84		1892	85		216	77
None		27	1		22	1		5	2
Steroid free regimen	0	482	19	0	416	19	0	66	24
mTOR inhibitors during follow-up	0	434	17	0	364	16	0	70	25

No HCTZ corresponds to patients never exposed to HCTZ during their follow-up, HCTZ group corresponds to the other patients.

ADPKD, autosomal dominant polycystic kidney disease; HLA, human leukocyte antigen; mTOR, mechanistic target of rapamycin; m, mean; NA: not available (missing); sd, standard deviation.

2) Basal-cell carcinoma

The cumulative incidence curve for basal-cell carcinoma is presented in **Figure 5.** Cumulative incidence rates at 5, 10 and 15 years post-transplantation for basal-cell carcinoma were **4%** (95% CI from 3 to 5%), **8%** (95% CI from 6 to 9%), and **11%** (95% CI from 9 to 13%) respectively.

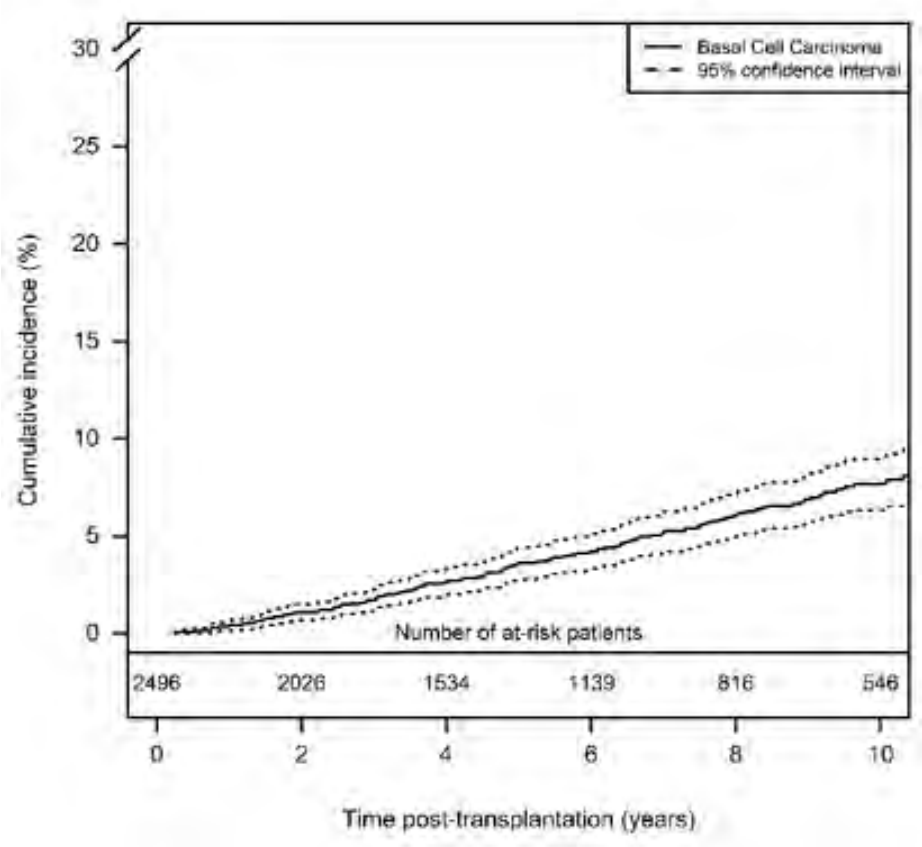

Figure 5. Cumulative incidence curve for Basal Cell Carcinoma *(Aalen-Johansen estimator, re-transplantations, returns to dialysis and deaths as competing events).*

Table 3 presents unadjusted analysis and the final multivariable model from which, amongst 2,496 patients, 23 were excluded due to missing data.

Multivariable Cox models were adjusted for baseline parameters: age, sex, re-transplantation, type of transplantation, type of donor, HLA-A -B -DR mismatch ≥ 4, induction therapy, maintenance treatment at transplantation, use of steroids at transplantation, initial nephropathy, cold ischemia time, and for time-varying covariates: rejection, maintenance treatment during follow-up (calcineurin inhibitor, mycophenolate derivatives, mTOR inhibitor) and other malignancies.

The confounder-adjusted Hazard Ratio (HR) associated with the HCTZ exposure was **0.63 (95% CI from 0.35 to 1.15).**

Table 3. *Results of the unadjusted and multivariable cause-specific time-dependent Cox models studying the risk of basal cell carcinoma (n = 2473).*

	Unadjusted Cox models			Adjusted Cox model		
	HR	**95% CI**	**p-value**	**HR**	**95% CI**	**p-value**
HTCZ exposure	0.79	[0.45 ; 1.40]	0.42	0.63	[0.35 ; 1.15]	0.13
Recipient age	1.05	[1.04 ; 1.07]	<0.001	1.05	[1.04 ; 1.07]	<0.001
Male sex	1.39	[0.99 ; 1.95]	0.06	1.52	[1.04 ; 2.23]	0.03
Initial nephropathy			0.005			0.28
Chronic tubulo-interstitial nephritis/ADPKD/Congenital uropathy (vs. Glomerulopathy)	1.22	[0.83 ; 1.77]		1.25	[0.84 ; 1.86]	
Diabetes (vs. Glomerulopathy)	0.53	[0.29 ; 0.96]		0.72	[0.31 ; 1.63]	
Other (vs. Glomerulopathy)	0.81	[0.42 ; 1.57]		0.65	[0.33 ; 1.28]	
Vascular nephropathy (vs. Glomerulopathy)	1.94	[1.09 ; 3.46]		1.07	[0.57 ; 2.01]	
Kidney replacement therapy			0.26	-	-	-
Hemodialysis (vs. Preemptive transplantation)	0.96	[0.63 ; 1.47]		-	-	-
Peritoneal dialysis (vs. Preemptive transplantation)	1.51	[0.80 ; 2.86]		-	-	-
Kidney transplantation	2.93	[1.44 ; 5.94]	0.003	1.80	[0.60 ; 5.40]	0.29
Retransplantation	1.09	[0.73 ; 1.64]	0.66	1.04	[0.59 ; 1.81]	0.90
Deceased donor	1.03	[0.62 ; 1.71]	0.91	0.97	[0.54 ; 1.72]	0.91
HLAA B Dr mismatches >= 4	0.86	[0.63 ; 1.19]	0.37	0.87	[0.62 ; 1.24]	0.44
Cold ischemia time >= 18 hours	0.74	[0.54 ; 1.02]	0.07	0.56	[0.39 ; 0.80]	0.002
Delayed graft function	0.96	[0.68 ; 1.36]	0.82	-	-	-
Rejection	0.71	[0.36 ; 1.40]	0.33	0.89	[0.45 ; 1.73]	0.72
Other malignancies	0.89	[0.39 ; 2.04]	0.78	0.38	[0.16 ; 0.94]	0.04
Induction treatment			0.83			0.55
Non-depleting (vs. Depleting)	1.11	[0.80 ; 1.53]		0.79	[0.51 ; 1.23]	
None (vs. Depleting)	1.09	[0.45 ; 2.60]		0.99	[0.41 ; 2.43]	
Calcineurin inhibitors			0.663			0.338
Cyclosporine (vs. Tacrolimus)	1.13	[0.79 ; 1.61]		1.31	[0.88 ; 1.93]	
None (vs. Tacrolimus)	0.67	[0.16 ; 2.76]		0.68	[0.14 ; 3.26]	
Calcineurin inhibitor during follow-up	0.46	[0.32 ; 0.67]	<0.001	0.49	[0.28 ; 0.85]	0.01
Steroid free regimen	1.03	[0.71 ; 1.49]	0.88	0.98	[0.66 ; 1.46]	0.92
mTOR during follow-up	1.87	[1.19 ; 2.92]	0.006	1.01	[0.54 ; 1.88]	0.97
Mycophenolate derivatives during follow-up	0.68	[0.46 ; 0.99]	0.04	0.88	[0.58 ; 1.34]	0.54

CI, confidence interval; HR, hazard ratio; ADPKD, autosomal dominant polycystic kidney disease; HCTZ, hydrochlorothiazide; HLA, human leukocyte antigen; mTOR, mechanistic target of rapamycin.

In sensitivity analysis of sub-group of patients, again, no association between HCTZ use and basal-cell carcinoma was found (**Table 5**).

The adjusted HR associated with mild-term (< 2 years) and long-term use (≥ 2 years) of HCTZ were 0.**59 (95% CI from 0.27 to 1.32)** and **0.70 (95% CI from 0.30 to 1.62)** respectively (**Table S1**, see Annexes)

3) Squamous cell carcinoma

The cumulative incidence curve of squamous-cell carcinoma is presented in **Figure 6**. Cumulative incidence rates at 5, 10 and 15 years post-transplantation for squamous-cell carcinoma were **3%** (95% CI from 3 to 4%),**7%** (95% CI from 6 to 8%), and **9%** (95% CI from 8 to 11%), respectively.

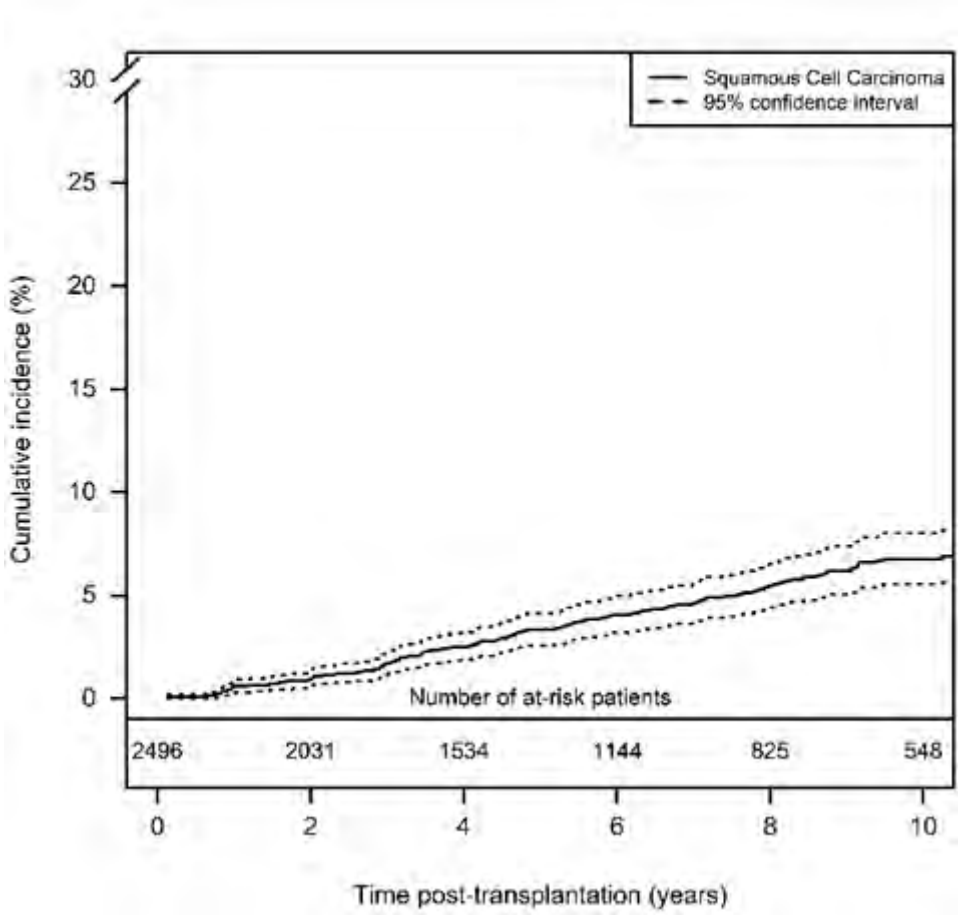

Figure 6. Cumulative incidence curve for Squamous Cell Carcinoma. *(Aalen-Johansen estimator, re-transplantations, returns to dialysis and deaths as competing events).*

Multivariable Cox models were adjusted for baseline parameters: age, sex, re-transplantation, type of transplantation, type of donor, HLA-A -B -DR mismatch ≥ 4, induction therapy, maintenance treatment at transplantation and use of steroids at transplantation, initial nephropathy, cold ischemia time and delayed graft function and for time-varying covariates: rejection, maintenance treatment during follow-up (calcineurin inhibitor, mycophenolate derivatives, mTOR inhibitor) and other malignancies.

Table 4 presents unadjusted analysis and the final multivariable model from which, amongst 2,496 patients, 80 were excluded due to missing data.

The confounder-adjusted HR was **2.04 (95% CI: 1.27 to 3.28), indicating a two-fold higher risk of squamous-cell carcinoma from time of exposure**.

We did not identify a significant dependence of the excess hazard with the time post-transplantation.

Table 4. *Results of the unadjusted and multivariable cause-specific time-dependent Cox models studying the risk of squamous cell carcinoma (n = 2416).*

	Unadjusted Cox models			Adjusted Cox model		
	HR	**95% CI**	**p-value**	**HR**	**95% CI**	**p-value**
HTCZ exposure	2.36	[1.53 ; 3.65]	<0.001	2.04	[1.27 ; 3.28]	0.003
Recipient age	1.07	[1.05 ; 1.08]	<0.001	1.06	[1.04 ; 1.07]	<0.001
Male sex	2.10	[1.40 ; 3.13]	<0.001	2.22	[1.45 ; 3.39]	<0.001
Initial nephropathy			0.006			0.71
Chronic tubulo-interstitial nephritis/ADPKD/Congenital uropathy (vs. Glomerulopathy)	0.75	[0.49 ; 1.16]		0.86	[0.56 ; 1.33]	
Diabetes (vs. Glomerulopathy)	0.53	[0.29 ; 0.97]		0.61	[0.26 ; 1.40]	
Other (vs. Glomerulopathy)	1.43	[0.83 ; 2.46]		1.01	[0.53 ; 1.90]	
Vascular nephropathy (vs. Glomerulopathy)	1.62	[0.88 ; 2.99]		0.73	[0.38 ; 1.42]	
Kidney replacement therapy			0.27	-	-	-
Hemodialysis (vs. Preemptive transplantation)	1.54	[0.91 ; 2.61]		-	-	-
Peritoneal dialysis (vs. Preemptive transplantation)	1.42	[0.63 ; 3.23]		-	-	-
Kidney transplantation	2.49	[1.22 ; 5.06]	0.01	0.68	[0.19 ; 2.36]	0.54
Retransplantation	1.99	[1.36 ; 2.90]	<0.001	2.41	[1.16 ; 4.99]	0.02
Deceased donor	2.79	[1.23 ; 6.34]	0.01	1.65	[0.67 ; 4.05]	0.27
HLAA B Dr mismatches >= 4	0.90	[0.64 ; 1.26]	0.54	0.96	[0.65 ; 1.40]	0.83
Cold ischemia time >= 18 hours	1.60	[1.13 ; 2.29]	0.009	1.32	[0.88 ; 1.98]	0.18
Delayed graft function	1.49	[1.05 ; 2.11]	0.02	0.96	[0.66 ; 1.40]	0.84
Rejection	0.17	[0.04 ; 0.71]	0.02	0.24	[0.06 ; 1.01]	0.06
Other malignancies	0.56	[0.17 ; 1.81]	0.33	0.19	[0.05 ; 0.66]	0.009
Induction treatment			0.83			0.84
Non-depleting (vs. Depleting)	1.04	[0.74 ; 1.48]		1.19	[0.61 ; 2.35]	
None (vs. Depleting)	0.77	[0.28 ; 2.12]		0.97	[0.30 ; 3.18]	
Calcineurin inhibitors			0.01			0.17
Cyclosporine (vs. Tacrolimus)	0.48	[0.29 ; 0.80]		0.67	[0.39 ; 1.16]	
None (vs. Tacrolimus)	0.32	[0.04 ; 2.42]		0.25	[0.03 ; 1.90]	
Calcineurin inhibitor during follow-up	0.39	[0.26 ; 0.59]	<0.001	0.29	[0.16 ; 0.52]	<0.001
Steroid free regimen	0.96	[0.64 ; 1.43]	0.83	1.05	[0.66 ; 1.68]	0.83
mTOR during follow-up	0.93	[0.49 ; 1.76]	0.82	0.30	[0.13 ; 0.68]	0.004
Mycophenolate derivatives during follow-up	0.47	[0.32 ; 0.70]	<0.001	0.64	[0.41 ; 1.01]	0.06

CI, confidence interval; HR, hazard ratio; ADPKD, autosomal dominant polycystic kidney disease; HCTZ, hydrochlorothiazide; HLA, human leukocyte antigen; mTOR, mechanistic target of rapamycin.

As displayed in **Table 5**, sensitivity analysis maintained this association even when analysis was restricted to solitary kidney transplant recipients **(**HR = **2.10**; 95% CI from 1.29 to 3.42).

When we considered patients according to their baseline immunosuppression, the confounder-adjusted HR was **1.94** (95% CI from 0.95 to 3.98) and **1.98** (95% CI from 0.91 to 4.33) for those on non-depleting induction agents and depleting induction agents respectively, and **2.26** (95% CI from 0.49 to 10.48) and **2.18** (95% CI from 1.30 to 3.64) for those on cyclosporine and tacrolimus as calcineurin inhibitor drugs respectively.

Table 5. *Results of the sensitivity analyses.*

	Basal cell carcinoma		Squamous cell carcinoma	
	HR	**95% CI**	**HR**	**95% CI**
Solitary kidney transplantation	0.67	[0.37 ; 1.23]	2.10	[1.29 ; 3.42]
Depleting induction	0.43	[0.17 ; 1.13]	1.98	[0.91 ; 4.33]
Non-depleting induction	0.78	[0.34 ; 1.77]	1.94	[0.95 ; 3.98]
Cyclosporine at transplantation	0.25	[0.04 ; 1.63]	2.26	[0.49 ; 10.48]
Tacrolimus at transplantation	0.87	[0.46 ; 1.64]	2.18	[1.30 ; 3.64]

CI, confidence interval; HR, hazard ratio; HCTZ, hydrochlorothiazide

To further explore a potential dose-response relationship, we performed a sensitivity analysis considering the exposure to HCTZ according to three categories. The adjusted HR associated with a mild-term use (< 2 years) and a long-term use (≥ 2 years) when compared with no use, were **1.99** (95% CI from 1.08 to 3.68) and **2.12** (95% CI from 1.14 to 3.94) respectively (**Table S2**, see Annexes).

VI) Discussion

In our cohort of transplant recipients, we found that, following exposure to HCTZ post-transplantation, patients had a significantly higher risk of developing squamous-cell carcinoma (HR = 2.04 (95% CI: 1.27 to 3.28)), regardless the period of that exposure. By contrast, we have not found an association between HCTZ exposure and the development of basal-cell carcinoma.
The association between HCTZ exposure and the incidence of squamous-cell carcinoma was also found when the analysis was restricted to solitary kidney transplant recipients.

Our findings were made possible due to the availability and use of prospective data on the exposure (i.e. HTCZ) and outcome variable (i.e. keratinocyte cancer) over an extended length of time, combined with a multivariable model that incorporated both time-dependent exposure to HCTZ and confounders (at baseline and some time-dependent) (71) that were not eliminated in the exclusion process (i.e. patients on an mTOR inhibitor or azathioprine at baseline).

More than sixty years ago, HCTZ became the first available oral diuretic and owing to its favorable safety profile, demonstrated a clear benefit in the treatment of hypertension. (73)
However, as soon as it was introduced, severe skin photosensitivity reactions were reported. (49,50)

It is well-established that a number of medications defined as photosensitizing potentiate the erythema reaction to UV light and, through a photocarcinogenic effect, bring about a higher risk of squamous-cell carcinoma. (38)
For instance, azathioprine is well-known to exacerbate skin photosensitivity and is associated with a higher incidence of squamous-cell carcinoma. (22,44)

In 2013, based on limited human evidence,(52,54,74) the International Agency for Research on Cancer highlighted HCTZ as a possible carcinogen (2B).

Since then, nationwide case-control studies in Denmark have unearthed a strong association between HCTZ use and squamous-cell carcinoma. (58,75)
These results were replicated in the United Kingdom, (60) leading to multiple national health agencies publishing safety alerts regarding its use in 2018. (61)

More recently, no association was demonstrated between HCTZ and skin cancer in an Asian population, highlighting the likely importance of baseline photosensitivity to UV as a contributing factor. (59)
A strength of these studies was their ability to demonstrate a dose-response relationship between HCTZ exposure and risk of keratinocyte cancer. In keeping with a lesser role for cumulative UV exposure, (17) only very long periods of HCTZ exposure have been associated with the development of basal-cell carcinoma.

To investigate a potential dose-response relationship in our population, we considered the exposure to HCTZ according to three categories. Results for HR for both mild-term (< 2 years) use and long-term use (≥ 2 years) were comparable.
In other words, we observed an association between HCTZ exposure and squamous-cell carcinoma as soon as a mild-term exposure occurred, and no dose-response relationship.

In the general population, this association has only been demonstrated in those with prolonged exposure to HCTZ (> 3 years) with a demonstrable dose-response relationship. The risk we have demonstrated in our study is comparable to that reported following 6 years of exposure in the original study.
These discrepancies could be related to a potentially additive and interactive effect that HCTZ has on a group of patients intrinsically made vulnerable to squamous-cell carcinoma through their exposure to immunosuppressive agents.
Thus, the absence of a dose-response relationship in our cohort could be, on one hand, superseded by the effect of immunosuppressants and, on the other hand, due to an inadequate follow-up interval compared to the baseline study conducted in the general population. (58)
These are only suppositions, and larger studies in terms of both numbers and length of follow-up are required to investigate them.

Squamous-cell carcinoma in solid organ transplant recipients occurs as a result of complex interactions between immune-mediated mechanisms of tumor promotion (mainly human papilloma virus activation and reduced tumor surveillance) combined with UV-induced carcinogenic effects. (76)
Thus, the emergence of possibly negative synergistic effects with the addition of photosensitizing drugs such as HCTZ, which are suspected to enhance UV-induced DNA damage, is unsurprising. (51)

Similarly, several studies conducted in lung transplant recipients report that voriconazole exposure, used in the prophylaxis and treatment of invasive fungal infection and also implicated as a photosensitizing drug, (77) is an independent risk factor for the development of squamous-cell carcinoma. (45,78,79)

Importantly, we were able to highlight the negative impacts of using HCTZ in a population mainly treated with immunosuppressants belonging to the modern era, namely tacrolimus and mycophenolate mofetil.
Indeed, as stated above, direct UV-induced carcinogenic effects are well-documented with azathioprine - the maintenance therapy historically used in kidney transplantation - but such effects have not been established for cyclosporine, tacrolimus, and mycophenolate mofetil.(44,80)

With improvements in surveillance, this likely explains why the risk of squamous-cell carcinoma in solid organ transplant recipients has drastically decreased in recent decades from 65 to 250 times (18) compared with the general population, to 20 times. (19,20)

In sensitivity analysis, we observed an HR slightly higher with the use of depleting versus non-depleting induction agents, as has been previously reported. (81)

Our observational study suffers from several limitations.

Firstly, it uses data that were not purposefully collected to answer the specific research question. Using data collected from a single center, the number of events detected was also limited.

Furthermore, we lacked information regarding two important risk factors for keratinocyte cancer: UV exposure and skin phenotype (of note, most of the western France population is of fair skin type). However, there is no reason to assume there is a relationship between these risk factors and the use of HTCZ, which may not be considered as confounders.

Secondly, HTCZ use prior to first transplantation would have been an important factor to consider, but this information was not collected in our database.

Thirdly, it would have been interesting to analyze each patient from the point of their first transplantation and then during subsequent dialysis and re-transplantation periods. However, because follow-up data were not available for patients returning to dialysis, we considered the graft as the statistical individual by censoring the return to dialysis without keratinocyte cancers. Among consequences for re-transplantations, prior exposure to HCTZ and other immunosuppressive drugs were not available. To tentatively consider the treatment exposures during previous transplantation or dialysis periods, we included in multivariable models transplantation rank as a proxy.

Finally, other unobserved confounding factors, such as other time-dependent drug exposures, cannot be excluded, making it difficult to establish firm causality.

Our results must be cautiously balanced with the risk related to poorly managed hypertension. Indeed, the overall prevalence of hypertension in kidney transplant recipients is up to 85% and is associated with shortened allograft survival and higher cardiovascular morbidity and mortality. (82)

Thiazide diuretics, including HCTZ, are recommended as a first-line treatment option for hypertension in the general population and appear efficient and safe for kidney transplant recipients. (83,84)

In addition to usual mechanisms, hypertension in kidney transplant recipients results from the use of immunosuppressive medications.

For instance, calcineurin-inhibitor-induced hypertension is related to kidney vasoconstriction, and is efficiently counteracted by the use of calcium-channel blockers. (85)

Moreover, because sodium retention occurs due to activation of the thiazide-sensitive sodium chloride cotransporter by tacrolimus, HCTZ could be especially effective for this group of patients. (86)

Importantly, in the Danish study, other antihypertensive drugs, including the usual alternative to HCTZ, bendroflumethiazide, another thiazide-type diuretic, and indapamide, a thiazide-like diuretic (the other main drug of this class, chlorthalidone, was not studied) were no associated with higher risks of skin cancer and could therefore be considered as an alternative.

In summary, our study suggests an association between HCTZ exposure and the development of squamous-cell carcinoma in kidney transplant recipients with fair skin type. Broader studies will be required to confirm these results. Physicians should carefully evaluate the risks and benefits of HCTZ use, especially in kidney transplant recipients with known non-modifiable risk factors such as fair skin, male sex and old age.

VII) Conclusion

Dans une grande cohorte monocentrique de patients transplantés rénaux et pancréatiques, nous avons pu mettre en évidence un sur-risque de CEC chez les patients traités par hydrochlorothiazide, avec un HR = 2.04 (95% CI : 1.27 à 3.28).
En d'autres termes, dans cette population, les patients sous HCTZ avaient un risque deux fois supérieur de survenue de carcinome épidermoïde cutané par rapport aux autres.

Depuis le début des années 2000, un certain nombre d'études se sont intéressées à cette association, mais il s'agit ici de la première portant spécifiquement sur une population très à risque, les patients transplantés d'organe solide.

A la lumière de ces résultats, et de ceux des études précédentes en population générale, il est important de proposer à la fois une prévention plus importante vis à vis de l'exposition solaire, mais également un suivi dermatologique plus rapproché aux patients transplantés sous HCTZ.

Ce résultat devra être répliqué sur d'autres cohortes, éventuellement prospectives, néanmoins, il semble licite de proposer des alternatives à l'HCTZ chez les patients transplantés d'organe solide exposés aux immunosuppresseurs.
D'un point de vue physiopathologique, il pourrait paraitre illogique de proposer d'autres diurétiques thiazidiques (bendroflumethiazide) ou apparentés (indapamide, chlortalidone), car même si la composante photosensibilisante de ces médicaments n'est pas parfaitement définie, ils possèdent un noyau sulfamide, à l'origine de l'effet photosensibilisant.

Cependant, dans la plupart des études présentées en introduction, ces alternatives semblaient moins favoriser la survenue de CCNM.

Si la situation clinique le permet, il semble judicieux de préférer une autre classe d'antihypertenseur en première intention, chez les patients transplantés, et de privilégier un diurétique de l'anse en cas d'hypertension supposée volo-dépendante.
En effet, la morbidité et la mortalité liées à une hypertension artérielle déséquilibrée sont clairement démontrées.

Ainsi, comme toujours en médecine, c'est l'évaluation personnalisée du rapport bénéfice-risque qui devra finalement guider le choix du praticien.

UNIVERSITÉ DE NANTES

VIII) Bibliographie

1. Jha V, Garcia-Garcia G, Iseki K, Li Z, Naicker S, Plattner B, et al. Chronic kidney disease: global dimension and perspectives. The Lancet. 20 juill 2013;382(9888):260-72.

2. GBD 2017 Causes of Death Collaborators. Global, regional, and national age-sex-specific mortality for 282 causes of death in 195 countries and territories, 1980-2017: a systematic analysis for the Global Burden of Disease Study 2017. The Lancet. oct 2018;392(10159):1736-88.

3. Fraser SDS, Roderick PJ. Kidney disease in the Global Burden of Disease Study 2017. Nat Rev Nephrol. 2019;15(4):193-4.

4. Liyanage T, Ninomiya T, Jha V, Neal B, Patrice HM, Okpechi I, et al. Worldwide access to treatment for end-stage kidney disease: a systematic review. The Lancet. 16 mai 2015;385(9981):1975-82.

5. Wolfe RA, Ashby VB, Milford EL, Ojo AO, Ettenger RE, Agodoa LY, et al. Comparison of mortality in all patients on dialysis, patients on dialysis awaiting transplantation, and recipients of a first cadaveric transplant. N Engl J Med. 2 déc 1999;341(23):1725-30.

6. Tonelli M, Wiebe N, Knoll G, Bello A, Browne S, Jadhav D, et al. Systematic Review: Kidney Transplantation Compared With Dialysis in Clinically Relevant Outcomes: Systematic Review of Kidney Transplantation. Am J Transplant. oct 2011;11(10):2093-109.

7. Kidney Disease: Improving Global Outcomes (KDIGO) Transplant Work Group. KDIGO clinical practice guideline for the care of kidney transplant recipients. Am J Transplant. nov 2009;9 Suppl 3:S1-155.

8. Szczech LA, Berlin JA, Aradhye S, Grossman RA, Feldman HI. Effect of anti-lymphocyte induction therapy on renal allograft survival: a meta-analysis. J Am Soc Nephrol. nov 1997;8(11):1771-7.

9. Webster AC, Ruster LP, McGee R, Matheson SL, Higgins GY, Willis NS, et al. Interleukin 2 receptor antagonists for kidney transplant recipients. Cochrane Database Syst Rev. 20 janv 2010;(1):CD003897.

10. Webster AC, Woodroffe RC, Taylor RS, Chapman JR, Craig JC. Tacrolimus versus ciclosporin as primary immunosuppression for kidney transplant recipients: meta-analysis and meta-regression of randomised trial data. BMJ. 8 oct 2005;331(7520):810.

11. Remuzzi G, Lesti M, Gotti E, Ganeva M, Dimitrov BD, Ene-Iordache B, et al. Mycophenolate mofetil versus azathioprine for prevention of acute rejection in renal transplantation (MYSS): a randomised trial. The Lancet. 7 août 2004;364(9433):503-12.

12. Fishman JA. Infection in Organ Transplantation. Am J Transplant. 2017;17(4):856-79.

13. Au E, Wong G, Chapman JR. Cancer in kidney transplant recipients. Nat Rev Nephrol. 2018;14(8):508-20.

14. van de Wetering J, Roodnat JI, Hemke AC, Hoitsma AJ, Weimar W. Patient survival after the diagnosis of cancer in renal transplant recipients: a nested case-control study. Transplantation. 27 déc 2010;90(12):1542-6.

15. Vajdic CM, McDonald SP, McCredie MRE, van Leeuwen MT, Stewart JH, Law M, et al. Cancer incidence before and after kidney transplantation. JAMA. 20 déc 2006;296(23):2823-31.

16. Lomas A, Leonardi-Bee J, Bath-Hextall F. A systematic review of worldwide incidence of nonmelanoma skin cancer. Br J Dermatol. mai 2012;166(5):1069-80.

17. Madan V, Lear JT, Szeimies R-M. Non-melanoma skin cancer. The Lancet. 2010;375:13.

18. Euvrard S, Kanitakis J, Claudy A. Skin Cancers after Organ Transplantation. N Engl J Med. 2003;11.

19. Rizvi SMH, Aagnes B, Holdaas H, Gude E, Boberg KM, Bjørtuft Ø, et al. Long-term Change in the Risk of Skin Cancer After Organ Transplantation: A Population-Based Nationwide Cohort Study. JAMA Dermatol. janv 2017;153(12):1270-7.

20. Menzies S, O'Leary E, Callaghan G, Galligan M, Deady S, Gadallah B, et al. Declining incidence of keratinocyte carcinoma in organ transplant recipients. Br J Dermatol. 2019;181(5):983-91.

21. Coghill AE, Johnson LG, Berg D, Resler AJ, Leca N, Madeleine MM. Immunosuppressive Medications and Squamous Cell Skin Carcinoma: Nested Case-Control Study Within the Skin Cancer after Organ Transplant (SCOT) Cohort. Am J Transplant. févr 2016;16(2):565-73.

22. Vos M, Plasmeijer EI, van Bemmel BC, van der Bij W, Klaver NS, Erasmus ME, et al. Azathioprine to mycophenolate mofetil transition and risk of squamous cell carcinoma after lung transplantation. J Heart Lung Transplant. juill 2018;37(7):853-9.

23. Ismail F, Mitchell L, Casabonne D, Gulati A, Newton R, Proby CM, et al. Specialist dermatology clinics for organ transplant recipients significantly improve compliance with photoprotection and levels of skin cancer awareness. Br J Dermatol. nov 2006;155(5):916-25.

24. Diffey BL, Langtry J a. A. Skin cancer incidence and the ageing population. Br J Dermatol. sept 2005;153(3):679-80.

25. Wang J, Aldabagh B, Yu J, Arron ST. Role of human papillomavirus in cutaneous squamous cell carcinoma: a meta-analysis. J Am Acad Dermatol. avr 2014;70(4):621-9.

26. Kricker A, Armstrong BK, English DR, Heenan PJ. A dose-response curve for sun exposure and basal cell carcinoma. Int J Cancer. 8 févr 1995;60(4):482-8.

27. Rosso S, Zanetti R, Martinez C, Tormo MJ, Schraub S, Sancho-Garnier H, et al. The multicentre south European study « Helios ». II: Different sun exposure patterns in the aetiology of basal cell and squamous cell carcinomas of the skin. Br J Cancer. juin 1996;73(11):1447-54.

28. English DR, Armstrong BK, Kricker A, Fleming C. Sunlight and cancer. Cancer Causes Control. mai 1997;8(3):271-83.

29. Holm-Schou A-SS, Philipsen PA, Wulf HC. Skin cancer phototype: A new classification directly related to skin cancer and based on responses from 2869 individuals. Photodermatol Photoimmunol Photomed. mars 2019;35(2):116-23.

30. Rünger TM. How different wavelengths of the ultraviolet spectrum contribute to skin carcinogenesis: the role of cellular damage responses. J Invest Dermatol. sept 2007;127(9):2103-5.

31. Cadet J, Wagner JR. DNA base damage by reactive oxygen species, oxidizing agents, and UV radiation. Cold Spring Harb Perspect Biol. 1 févr 2013;5(2).

32. Ridley AJ, Whiteside JR, McMillan TJ, Allinson SL. Cellular and sub-cellular responses to UVA in relation to carcinogenesis. Int J Radiat Biol. mars 2009;85(3):177-95.

33. Karagas MR, Stukel TA, Umland V, Tsoukas MM, Mott LA, Sorensen HT, et al. Reported use of photosensitizing medications and basal cell and squamous cell carcinoma of the skin: results of a population-based case-control study. J Invest Dermatol. déc 2007;127(12):2901-3.

34. Monteiro AF, Rato M, Martins C. Drug-induced photosensitivity: Photoallergic and phototoxic reactions. Clin Dermatol. oct 2016;34(5):571-81.

35. Drucker AM, Rosen CF. Drug-Induced Photosensitivity: Culprit Drugs, Management and Prevention. Drug Saf. oct 2011;34(10):821-37.

36. Glatz M, Hofbauer GFL. Phototoxic and photoallergic cutaneous drug reactions. Chem Immunol Allergy. 2012;97:167-79.

37. Tokura Y. Drug photoallergy. J Cutan Immunol Allergy. 2018;1(2):48-57.

38. Stern RS. Photocarcinogenicity of drugs. Toxicol Lett. déc 1998;102-103:389-92.

39. Gocke E. Photochemical mutagenesis: examples and toxicological relevance. J Environ Pathol Toxicol Oncol. 2001;20(4):285-92.

40. Kreutz R, Algharably EAH, Douros A. Reviewing the effects of thiazide and thiazide-like diuretics as photosensitizing drugs on the risk of skin cancer: J Hypertens. juin 2019;1.

41. Parrish JA, Fitzpatrick TB, Tanenbaum L, Pathak MA. Photochemotherapy of psoriasis with oral methoxsalen and longwave ultraviolet light. N Engl J Med. 5 déc 1974;291(23):1207-11.

42. Stern RS, Laird N, Melski J, Parrish JA, Fitzpatrick TB, Bleich HL. Cutaneous squamous-cell carcinoma in patients treated with PUVA. N Engl J Med. 3 mai 1984;310(18):1156-61.

43. O'Donovan P, Perrett CM, Zhang X, Montaner B, Xu Y-Z, Harwood CA, et al. Azathioprine and UVA light generate mutagenic oxidative DNA damage. Science. 16 sept 2005;309(5742):1871-4.

44. Hofbauer GFL, Attard NR, Harwood CA, McGregor JM, Dziunycz P, Iotzova-Weiss G, et al. Reversal of UVA Skin Photosensitivity and DNA Damage in Kidney Transplant Recipients by Replacing Azathioprine: Skin Photosensitivity in Kidney Transplant Recipients. Am J Transplant. janv 2012;12(1):218-25.

45. Feist A, Lee R, Osborne S, Lane J, Yung G. Increased incidence of cutaneous squamous cell carcinoma in lung transplant recipients taking long-term voriconazole. J Heart Lung Transplant. nov 2012;31(11):1177-81.

46. Freis ED, Wanko A, Wilson IM, Parrish AE. Chlorothiazide in hypertensive and normotensive patients. Ann N Y Acad Sci. 3 févr 1958;71(4):450-5.

47. Effects of treatment on morbidity in hypertension. Results in patients with diastolic blood pressures averaging 115 through 129 mm Hg. JAMA. 11 déc 1967;202(11):1028-34.

48. Mancia G, Rosei EA, Azizi M, Burnier M, Clement DL, Coca A, et al. 2018 ESC/ESH Guidelines for the management of arterial hypertension. Eur Heart J. 2018;98.

49. Harber LC, Lashinsky AM, Baer RL. Photosensitivity due to chlorothiazide and hydrochlorothiazide. N Engl J Med. 31 déc 1959;261:1378-81.

50. Harber LC, Lashinsky AM, Baer RL. Skin manifestations of photosensitivity due to chlorothiazide and hydrochlorothiazide. J Invest Dermatol. sept 1959;33:83-4.

51. Kunisada M, Masaki T, Ono R, Morinaga H, Nakano E, Yogianti F, et al. Hydrochlorothiazide Enhances UVA-Induced DNA Damage. Photochem Photobiol. mai 2013;89(3):649-54.

52. Jensen AØ, Thomsen HF, Engebjerg MC, Olesen AB, Sørensen HT, Karagas MR. Use of photosensitising diuretics and risk of skin cancer: a population-based case–control study. Br J Cancer. nov 2008;99(9):1522-8.

53. Ruiter R, Visser LE, Eijgelsheim M, Rodenburg EM, Hofman A, Coebergh J-WW, et al. High-ceiling diuretics are associated with an increased risk of basal cell carcinoma in a population-based follow-up study. Eur J Cancer. sept 2010;46(13):2467-72.

54. de Vries E, Trakatelli M, Kalabalikis D, Ferrandiz L, Ruiz-de-Casas A, Moreno-Ramirez D, et al. Known and potential new risk factors for skin cancer in European populations: a multicentre case-control study. Br J Dermatol. août 2012;167 Suppl 2:1-13.

55. Schmidt S a. J, Schmidt M, Mehnert F, Lemeshow S, Sørensen HT. Use of antihypertensive drugs and risk of skin cancer. J Eur Acad Dermatol Venereol. août 2015;29(8):1545-54.

56. Nardone B, Majewski S, Kim AS, Kiguradze T, Martinez-Escala EM, Friedland R, et al. Melanoma and Non-Melanoma Skin Cancer Associated with Angiotensin-Converting-Enzyme Inhibitors, Angiotensin-Receptor Blockers and Thiazides: A Matched Cohort Study. Drug Saf. 2017;40(3):249-55.

57. Su KA, Habel LA, Achacoso NS, Friedman GD, Asgari MM. Photosensitizing antihypertensive drug use and risk of cutaneous squamous cell carcinoma. Br J Dermatol. 2018;179(5):1088-94.

58. Pedersen SA, Gaist D, Schmidt SAJ, Hölmich LR, Friis S, Pottegård A. Hydrochlorothiazide use and risk of nonmelanoma skin cancer: A nationwide case-control study from Denmark. J Am Acad Dermatol. avr 2018;78(4):673-681.e9.

59. Pottegård A, Pedersen SA, Schmidt SAJ, Lee C-N, Hsu C-K, Liao T-C, et al. Use of hydrochlorothiazide and risk of skin cancer: a nationwide Taiwanese case-control study. Br J Cancer. 2019;121(11):973-8.

60. Morales DR, Pacurariu A, Slattery J, Kurz X. Association between hydrochlorothiazide exposure and different incident skin, lip and oral cavity cancers: A series of population-based nested case-control studies. Br J Clin Pharmacol. juill 2020;86(7):1336-45.

61. Hydrochlorothiazide - Risque de cancer de la peau non-mélanome (carcinome basocellulaire, carcinome épidermoïde) - Lettre aux professionnels de santé - ANSM : Agence nationale de sécurité du médicament et des produits de santé [Internet]. [cité 6 nov 2020]. Disponible sur: https://www.ansm.sante.fr/S-informer/Informations-de-securite-Lettres-aux-professionnels-de-sante/Hydrochlorothiazide-Risque-de-cancer-de-la-peau-non-melanome-carcinome-basocellulaire-carcinome-epidermoide-Lettre-aux-professionnels-de-sante

62. PRAC recommendations on signals adopted at the 3-6 September 2018 PRAC meeting. EMA : European Medicines Agency. [Internet]. 2018. [cité 6 nov 2020]. Disponible sur : https://www.ema.europa.eu/en/documents/prac-recommendation/prac-recommendations-signals-adopted-3-6-september-2018-prac-meeting_en-0.pdf.

63. FDA approves label changes to hydrochlorothiazide to describe small risk of non-melanoma skin cancer. FDA [Internet]. 20 août 2020 [cité 6 nov 2020]; Disponible sur: https://www.fda.gov/drugs/drug-safety-and-availability/fda-approves-label-changes-hydrochlorothiazide-describe-small-risk-non-melanoma-skin-cancer

64. Rosales BM, De La Mata N, Vajdic CM, Kelly PJ, Wyburn K, Webster AC. Cancer mortality in kidney transplant recipients: An Australian and New Zealand population-based cohort study, 1980-2013. Int J Cancer. 2020;146(10):2703-11.

65. Aalen OO, Johansen S. An Empirical Transition Matrix for Non-Homogeneous Markov Chains Based on Censored Observations. Scand J Stat. 1978;5(3):141-50.

66. Schemper M, Smith TL. A note on quantifying follow-up in studies of failure time. Control Clin Trials. août 1996;17(4):343-6.

67. Shariff SZ, Cuerden MS, Jain AK, Garg AX. The secret of immortal time bias in epidemiologic studies. J Am Soc Nephrol. mai 2008;19(5):841-3.

68. Liu J, Weinhandl ED, Gilbertson DT, Collins AJ, St Peter WL. Issues regarding « immortal time » in the analysis of the treatment effects in observational studies. Kidney Int. févr 2012;81(4):341-50.

69. Andersen PK, Geskus RB, de Witte T, Putter H. Competing risks in epidemiology: possibilities and pitfalls. Int J Epidemiol. juin 2012;41(3):861-70.

70. Schoenfeld D. Partial residuals for the proportional hazards regression model. Biometrika. 1 avr 1982;69(1):239-41.

71. Xie D, Yang W, Jepson C, Roy J, Hsu JY, Shou H, et al. Statistical Methods for Modeling Time-Updated Exposures in Cohort Studies of Chronic Kidney Disease. Clin J Am Soc Nephrol. 7 nov 2017;12(11):1892-9.

72. Dekker FW, de Mutsert R, van Dijk PC, Zoccali C, Jager KJ. Survival analysis: time-dependent effects and time-varying risk factors. Kidney Int. oct 2008;74(8):994-7.

73. Moser M, Macaulay AI. Chlorothiazide as an adjunct in the treatment of essential hypertension. Am J Cardiol. févr 1959;3(2):214-9.

74. Robinson SN, Zens MS, Perry AE, Spencer SK, Duell EJ, Karagas MR. Photosensitizing Agents and the Risk of Non-Melanoma Skin Cancer: A Population-Based Case–Control Study. J Invest Dermatol. août 2013;133(8):1950-5.

75. Pottegård A, Hallas J, Olesen M, Svendsen MT, Habel LA, Friedman GD, et al. Hydrochlorothiazide use is strongly associated with risk of lip cancer. J Intern Med. oct 2017;282(4):322-31.

76. Mittal A, Colegio OR. Skin Cancers in Organ Transplant Recipients. Am J Transplant. oct 2017;17(10):2509-30.

77. Haylett AK, Felton S, Denning DW, Rhodes LE. Voriconazole-induced photosensitivity: photobiological assessment of a case series of 12 patients. Br J Dermatol. janv 2013;168(1):179-85.

78. Singer JP, Boker A, Metchnikoff C, Binstock M, Boettger R, Golden JA, et al. High cumulative dose exposure to voriconazole is associated with cutaneous squamous cell carcinoma in lung transplant recipients. J Heart Lung Transplant. juill 2012;31(7):694-9.

79. Vadnerkar A, Nguyen MH, Mitsani D, Crespo M, Pilewski J, Toyoda Y, et al. Voriconazole exposure and geographic location are independent risk factors for squamous cell carcinoma of the skin among lung transplant recipients. J Heart Lung Transplant. nov 2010;29(11):1240-4.

80. Voskamp P, Bodmann CA, Koehl GE, Tensen CP, Bavinck JNB, Willemze R, et al. No acceleration of UV-induced skin carcinogenesis from evenly spread dietary intake of cyclosporine in contrast to oral bolus dosages. Transplantation. 27 nov 2013;96(10):871-6.

81. Bustami RT, Ojo AO, Wolfe RA, Merion RM, Bennett WM, McDiarmid SV, et al. Immunosuppression and the risk of post-transplant malignancy among cadaveric first kidney transplant recipients. Am J Transplant. janv 2004;4(1):87-93.

82. Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, et al. 2018 Practice Guidelines for the management of arterial hypertension of the European Society of Hypertension and the European Society of Cardiology: ESH/ESC Task Force for the Management of Arterial Hypertension. J Hypertens. 2018;36(12):2284-309.

83. Taber DJ, Srinivas TM, Pilch NA, Meadows HB, Fleming JN, McGillicuddy JW, et al. Are thiazide diuretics safe and effective antihypertensive therapy in kidney transplant recipients? Am J Nephrol. 2013;38(4):285-91.

84. Moes AD, Hesselink DA, van den Meiracker AH, Zietse R, Hoorn EJ. Chlorthalidone Versus Amlodipine for Hypertension in Kidney Transplant Recipients Treated With Tacrolimus: A Randomized Crossover Trial. Am J Kidney Dis. juin 2017;69(6):796-804.

85. Kuypers DRJ, Neumayer HH, Fritsche L, Budde K, Rodicio JL, Vanrenterghem Y, et al. Calcium channel blockade and preservation of renal graft function in cyclosporine-treated recipients: a prospective randomized placebo-controlled 2-year study. Transplantation. 27 oct 2004;78(8):1204-11.

86. Hoorn EJ, Walsh SB, McCormick JA, Fürstenberg A, Yang C-L, Roeschel T, et al. The calcineurin inhibitor tacrolimus activates the renal sodium chloride cotransporter to cause hypertension. Nat Med. 2 oct 2011;17(10):1304-9.

IX) Annexes

1) Liste des abréviations

CCNM	Cancer cutané non-mélanome
CEC	Carcinome épidermoïde cutané
CBC	Carcinome basocellulaire
UV	Ultraviolet
ROS	Espèce réactive de l'oxygène
HCTZ	Hydrochlorothiazide
IRR	Incidence rate ratio
CI	Intervalle de confiance
OR	Odds ratio
HR	Hazard ratio
mTOR	Cible de la rapamycine chez les mammifères
HLA	Antigène leucocytaire humain
APKD	Polykystose rénale autosomique dominante

UNIVERSITÉ DE NANTES

2) Données supplémentaires

Table S1. *Results of the unadjusted and multivariable cause-specific time-dependent Cox models studying the risk of basal cell carcinoma with the exposure to HCTZ in 3 categories (n = 2473).*

	HR	95% CI	p-value
HTCZ exposure			0.327
< 2 years (vs. no use)	0.59	[0.27 ; 1.32]	
>= 2 years (vs. no use)	0.70	[0.30 ; 1.62]	
Recipient age	1.05	[1.04 ; 1.07]	<0.001
Male sex	1.53	[1.04 ; 2.23]	0.029
Initial nephropathy			0.277
Chronic tubulo-interstitial nephritis/ADPKD/Congenital uropathy (vs. Glomerulopathy)	1.25	[0.84 ; 1.86]	
Diabetes (vs. Glomerulopathy)	0.72	[0.31 ; 1.63]	
Other (vs. Glomerulopathy)	0.65	[0.33 ; 1.28]	
Vascular nephropathy (vs. Glomerulopathy)	1.07	[0.57 ; 2.01]	
Kidney transplantation	1.80	[0.60 ; 5.41]	0.291
Retransplantation	1.04	[0.59 ; 1.81]	0.903
Deceased donor	0.96	[0.54 ; 1.72]	0.903
HLAA B Dr mismatches >= 4	0.87	[0.62 ; 1.24]	0.443
Cold ischemia time >= 18 hours	0.56	[0.39 ; 0.80]	0.002
Rejection	0.89	[0.45 ; 1.73]	0.722
Other malignancies	0.38	[0.16 ; 0.93]	0.035
Induction treatment			0.549
Non-depleting (vs. Depleting)	0.79	[0.51 ; 1.23]	
None (vs. Depleting)	0.99	[0.41 ; 2.41]	
Calcineurin inhibitors			0.332
Cyclosporine (vs. Tacrolimus)	1.31	[0.89 ; 1.94]	
None (vs. Tacrolimus)	0.68	[0.14 ; 3.26]	
Steroid free regimen	0.98	[0.66 ; 1.46]	0.920
Calcineurin inhibitor during follow-up	0.49	[0.28 ; 0.85]	0.011
mTOR during follow-up	1.01	[0.54 ; 1.89]	0.969
Mycophenolate derivatives during follow-up	0.88	[0.58 ; 1.34]	0.548

CI, confidence interval; HR, hazard ratio; ADPKD, autosomal dominant polycystic kidney disease; HCTZ, hydrochlorothiazide; HLA, human leukocyte antigen; mTOR, mechanistic target of rapamycin.

UNIVERSITÉ DE NANTES

Table S2. *Results of the unadjusted and multivariable cause-specific time-dependent Cox models studying the risk of squamous cell carcinoma with the exposure to HCTZ in 3 categories (n= 2416). CI, confidence interval; HR, hazard ratio; ADPKD, autosomal dominant polycystic kidney disease; HCTZ, hydrochlorothiazide; HLA, human leukocyte antigen; mTOR, mechanistic target of rapamycin.*

	HR	95% CI	p-value
HTCZ exposure			0.010
< 2 years (vs. no use)	1.99	[1.08 ; 3.68]	
>= 2 years (vs. no use)	2.12	[1.14 ; 3.94]	
Recipient age	1.06	[1.04 ; 1.07]	<0.001
Male sex	2.22	[1.45 ; 3.38]	<0.001
Initial nephropathy			0.704
Chronic tubulo-interstitial nephritis/ADPKD/Congenital uropathy (vs. Glomerulopathy)	0.86	[0.56 ; 1.33]	
Diabetes (vs. Glomerulopathy)	0.61	[0.26 ; 1.40]	
Other (vs. Glomerulopathy)	1.01	[0.53 ; 1.92]	
Vascular nephropathy (vs. Glomerulopathy)	0.73	[0.38 ; 1.42]	
Kidney transplantation	0.68	[0.19 ; 2.36]	0.539
Retransplantation	2.41	[1.16 ; 4.98]	0.018
Deceased donor	1.65	[0.67 ; 4.05]	0.273
HLAA B Dr mismatches >= 4	0.96	[0.65 ; 1.40]	0.830
Cold ischemia time >= 18 hours	1.31	[0.87 ; 1.98]	0.190
Delayed graft function	0.96	[0.66 ; 1.41]	0.846
Rejection	0.24	[0.06 ; 1.01]	0.051
Other malignancies	0.19	[0.05 ; 0.66]	0.009
Induction treatment			0.837
Non-depleting (vs. Depleting)	1.19	[0.61 ; 2.34]	
None (vs. Depleting)	0.97	[0.29 ; 3.18]	
Calcineurin inhibitors			0.169
Cyclosporine (vs. Tacrolimus)	0.67	[0.39 ; 1.16]	
None (vs. Tacrolimus)	0.25	[0.03 ; 1.90]	
Steroid free regimen	1.05	[0.66 ; 1.68]	0.832
Calcineurin inhibitor during follow-up	0.29	[0.16 ; 0.52]	<0.001
mTOR during follow-up	0.29	[0.13 ; 0.67]	0.004
Mycophenolate derivatives during follow-up	0.65	[0.41 ; 1.02]	0.058

CI, confidence interval; HR, hazard ratio; ADPKD, autosomal dominant polycystic kidney disease; HCTZ, hydrochlorothiazide; HLA, human leukocyte antigen; mTOR, mechanistic target of rapamycin.

UNIVERSITÉ DE NANTES

3) Manuscrit

Article

Association between Use of Hydrochlorothiazide and Risk of Keratinocyte Cancers in Kidney Transplant Recipients

Thibault Letellier,[1] *Florent Leborgne*,[2,3] *Clarisse Kerleau*,[1] *Aurélie Gaultier*,[4] *Jacques Dantal*,[1,5] *and Simon Ville*,[1,5] *on behalf of Divat Consortium*[*]

Abstract

Background and objectives Keratinocyte cancers, which primarily comprise squamous cell carcinomas and basal cell carcinomas, represent a major concern and potential risk for kidney transplant recipients. Hydrochlorothiazide, a diuretic widely used to treat hypertension, has been implicated in skin photosensitivity reaction. Recent studies conducted in the general population have found that hydrochlorothiazide use is associated with a higher risk of keratinocyte cancer, especially squamous cell carcinomas. High-risk groups, however, including transplant recipients were excluded from these. Our aim was to investigate whether hydrochlorothiazide use was associated with keratinocyte cancer in kidney transplant recipients on immunosuppressive therapy.

Design, setting, participants, & measurements In a single-center cohort of kidney (n=2155), combined kidney-pancreas (n=282), and pancreas (n=59) transplant recipients from the DIVAT database transplanted between 2000 and 2017 in Nantes (France), we evaluated the association between hydrochlorothiazide exposure and keratinocyte cancers. Multivariable cause-specific, time-varying Cox models were used to estimate the relationship between hydrochlorothiazide exposure and the hazard of squamous cell carcinoma and basal cell carcinoma, with hydrochlorothiazide designated as the time-dependent variable.

Results Among the participants, 279 of 2496 (11%) were exposed to hydrochlorothiazide after the transplantation. Cumulative incidence rates of keratinocyte cancer by 10 and 15 years were of 7% and 9% for squamous cell carcinomas, respectively, and 8% and 11% for basal cell carcinomas, respectively. We found a relationship between exposure to hydrochlorothiazide and the risk of squamous cell carcinomas (hazard ratio, 2.04; 95% confidence interval, 1.27 to 3.28). In contrast, we found no association between hydrochlorothiazide exposure and basal cell carcinomas (hazard ratio, 0.63; 95% confidence interval, 0.35 to 1.15).

Conclusions In a single-center cohort of kidney, combined kidney-pancreas, and pancreas transplant recipients, exposure to hydrochlorothiazide was associated with a two-fold higher risk of squamous cell carcinoma and no higher risk of basal cell carcinoma.

CJASN 15: ●●●–●●●, 2020. doi: https://doi.org/10.2215/CJN.02560220

[1]Institut de Transplantation Urologie Néphrologie, CHU Nantes, Nantes, France
[2]INSERM UMR 1246–SPHERE, Nantes University, Tours University, Nantes, France
[3]IDBC, Pacé, France
[4]CHU de Nantes, Direction de la recherche, Plateforme de Méthodologie et Biostatistique, Nantes, France
[5]Centre de Recherche en Transplantation et Immunologie UMR1064, INSERM, Université de Nantes, Nantes, France

Correspondence: Dr. Simon Ville, Department of Nephrology and Transplantation, CHU de Nantes, 30 Boulevard Jean Monnet, 44000 Nantes, France. Email: simon.ville@chu-nantes.fr

Introduction

In recent decades, kidney transplant outcomes have markedly progressed. However, the use of immunosuppressive therapy, essential for preventing graft rejection, is associated with a significant adverse event profile for patients. Aside from infection, cancer is the main adverse event associated with the use of immunosuppressive agents in solid organ transplant recipients (1,2). Keratinocyte cancers, namely squamous cell carcinomas and basal cell carcinomas, accounts for >90% of skin cancers in transplant recipients. These typically affect more than half of patients over time, occurring 65–250 and 10–16 times, respectively, more frequently than in the general population (3,4). In addition, squamous cell carcinomas typically follow a more aggressive course in patients with transplants, resulting in significant morbidity and mortality. Aside from immunosuppressive therapy, risk factors for keratinocyte cancer reported in this population include old age, men, and fair skin type (3,4). Although cumulative sun exposure is a well-established risk factor for squamous cell carcinomas, the pattern of sun exposure associated with basal cell carcinomas is less clear (5). For some time, photosensitizing drugs have been suspected to favor the development of squamous cell carcinomas by potentiating the carcinogenic effect of ultraviolet (UV) radiation. A recent study conducted in the general population has supported an association between use of hydrochlorothiazide (HCTZ), one of those well-known photosensitizing drugs, and risk of keratinocyte cancer, especially squamous cell carcinomas (6). As the study's focus was on the general population, high-risk groups,

including transplant recipients and immunosuppressed patients, were excluded (6).

Q:5 Q:6 Q:7 Q:8 We aimed to investigate whether HCTZ use is associated with keratinocyte cancer in transplant recipients on immunosuppressive therapy independently of other known risk factors. We conducted a single-center study using a cohort of kidney, combined kidney-pancreas, and pancreas transplant recipients.

Materials and Methods

Patient Population

We conducted a single-center study on all consecutive adult patients transplanted with a graft that functioned for at least 3 months (kidney, pancreas, or combined kidney-pancreas) at Nantes University Hospital between January 1, 2000 and December 31, 2017. We excluded patients who had been recipients of three or more transplantations, as well as those with a prior history of basal cell carcinoma or squamous cell carcinoma. We also excluded those on initial maintenance immunosuppressive regimens that included mammalian target of rapamycin (mTOR) inhibitors or azathioprine, as these treatments have been implicated as protective and contributing factors to keratinocyte cancers, respectively (Figure 1). Patients with transplants otherwise received standard medical care (Supplemental Material).

Q:9 All data were extracted from the French multicenter, observational, and prospective DIVAT cohort of patients with transplants (www.divat.fr). The clinical and research activities reported are consistent with the Principles of the Declaration of Istanbul as outlined in the "Declaration of Istanbul on Organ Trafficking and Transplant Tourism." More specifically, the DIVAT cohort received Commission Nationale de l'Informatique et des Libertés Final Agreement 914184, and the Advisory Committee on Information Processing in Material Research in the Field of Health approved the use of its data for scientific research purposes.

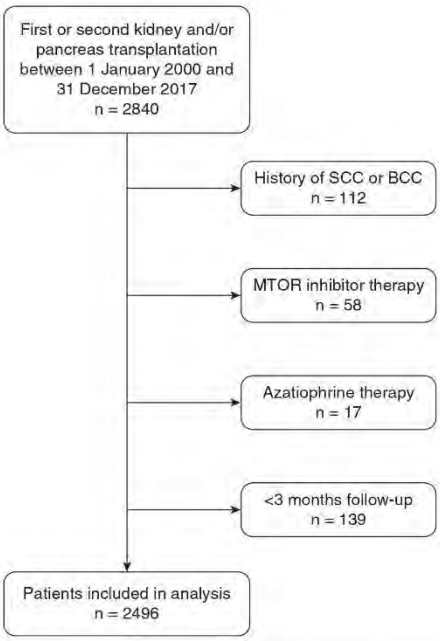

Figure 1. | **Flowchart of eligibility criterion.** BCC, basal cell carcinoma; mTOR, mammalian target of rapamycin; SCC, squamous cell carcinoma.

Available Data

Complete available data are found in Supplemental Material. All patients underwent a dermatologic examination undertaken by a specialist prior to the transplant and in accordance with guidelines regarding outpatient surveillance of kidney transplant recipients, every year after the transplant. Histologically confirmed keratinocyte cancers were entered into the database.

Regarding HCTZ exposure, we extracted the list of patients prescribed HCTZ in the post-transplant period using prescription data collected longitudinally and contemporaneously in our electronic system. We reviewed each prescription for accuracy in terms of start and stop dates (considered meaningful from 1 month of exposure) and dose. The follow-up and the collection of data ceased upon a patient's return to dialysis or at death.

Statistical Analyses

We considered the transplant as the statistical individual. Outcomes were defined by time from transplantation to the first occurrence of a squamous cell carcinoma or basal cell carcinoma. Participants who did not have an outcome were censored at graft failure (preemptive retransplantation, return to dialysis, or death), when they were lost to follow-up, or at the end of the study, whichever came first. The cumulative incidence curves of HCTZ, squamous cell carcinoma, and basal cell carcinoma were obtained using the Aalen–Johansen estimator, using graft failures as competing events (7). The median follow-up time was estimated using the reverse Kaplan–Meier (8). Multivariable cause-specific, time-varying Cox models were used to estimate the relationship between HCTZ and the hazard of squamous cell carcinoma and basal cell carcinoma, with HCTZ designated as time-dependent variable as is usually recommended to prevent "immortal time" bias in the analysis of treatment effect in observational studies (9,10). Graft failures were right censored (11). In our main analysis, we studied the dichotomized variable of cumulative duration since the start of HCTZ. Indeed, construing HCTZ exposure as a time-dependent variable allowed patients, initially all in the untreated group, to move and remain into the treatment group from when HCTZ was commenced. The hazard proportionality assumption was verified from the Schoenfeld residuals (12). For baseline continuous covariates, the log-linearity assumption has been checked in unadjusted analysis if the Bayesian Information Criterion was not reduced using natural spline transformation compared with the inclusion of the covariate in its natural scale. Time-dependent covariates related to immunosuppression were on the basis of 1-year time windows. Patients were categorized as exposed for post-transplantation

Q:17

Table 1. Characteristics of 2496 transplant recipients included in the analysis according their exposure to hydrochlorothiazide during follow-up

COLUMN HEADING	Whole Sample, $n=2496$			No Hydrochlorothiazide, $n=2217$			Hydrochlorothiazide, $n=279$		
	Not Available (Missing)	Mean	SD	Not Available (Missing)	Mean	SD	Not Available (Missing)	Mean	SD
Recipient age, yr	0	49	14	0	49	14	0	50	13
	Not Available (Missing)	*n*	%	Not Available (Missing)	*n*	%	Not Available (Missing)	*n*	%
Men	0	1538	62	0	1353	61	0	185	66
Initial nephropathy	0			0			0		
Chronic tubulointerstitial nephritis/ ADPKD/congenital uropathy		962	39		880	40		82	29
Diabetes		467	19		413	19		54	19
Glomerulopathy		643	26		567	26		76	27
Other		232	9		206	9		26	9
Vascular nephropathy		192	8		151	7		41	15
KRT	46			41			5		
Hemodialysis		1757	72		1544	71		213	78
Peritoneal dialysis		217	9		200	9		17	6
Preemptive transplantation		476	19		432	20		44	16
Type of transplantation	0			0			0		
Combined kidney-pancreas		282	11		254	11		28	10
Kidney		2155	86		1909	86		246	88
Pancreas		59	2		54	2		5	2
Retransplantation	0	436	17	0	399	18	0	37	13
Deceased donor	0	2200	88	0	1948	88	0	252	90
HLA-A -B -Dr mismatches ≥4	2	1264	51	2	1113	50	0	151	54
Cold ischemia time ≥18 h	21	1018	41	20	882	40	1	136	49
Delayed graft function	78	811	34	73	705	33	5	106	39
Rejection	0	301	12	0	264	12	0	37	13
Other malignancies	0	193	8	0	163	7	0	30	11
Induction treatment	0			0			0		
Depleting		1219	49		1098	50		121	43
Nondepleting		1213	49		1066	48		147	53
None		64	3		53	2		11	4
Calcineurin inhibitors	0			0			0		
Cyclosporin		361	14		303	14		58	21
Tacrolimus		2108	84		1892	85		216	77
None		27	1		22	1		5	2
Steroid-free regimen	0	482	19	0	416	19	0	66	24
mTOR inhibitors during follow-up	0	434	17	0	364	16	0	70	25

No hydrochlorothiazide (HCTZ) corresponds to patients never exposed to HCTZ during their follow-up. The HCTZ group corresponds to the other patients. ADPKD, autosomal dominant polycystic kidney disease; KRT, kidney transplant recipient; mTOR, mammalian target of rapamycin.

periods during which they received the treatment and unexposed during other periods (13,14). On the basis of theoretical considerations and known biologic effects, we included *a priori* the following variables in the multivariable models: (*1*) baseline parameters: age, sex, retransplantation, type of transplantation, type of donor, HLA-A -B -DR mismatch greater than or equal to four, induction therapy, maintenance treatment at transplantation, and use of steroids at transplantation; and (*2*) time-varying covariates: rejection, maintenance treatment during follow-up (calcineurin inhibitor, mycophenolate derivatives, or mTOR inhibitor), and other malignancies. We also considered covariates that were significantly associated with the outcomes (P=0.10) in unadjusted models. We did not consider interaction term. Patients who had data missing for covariates retained in the multivariable models were excluded.

Q : 10

A series of sensitivity analyses was performed. First, we estimated the effect of HCTZ use on different subgroups of patients: (*1*) solitary kidney transplant recipients, (*2*) patients receiving depleting induction therapy, (*3*) patients receiving nondepleting induction therapy, (*4*) patient receiving cyclosporin at transplantation, and (*5*) patients receiving tacrolimus at transplantation. Second, in order to explore a potential dose-response relationship, considering that HCTZ doses remained relatively stable regardless of when it was initiated post-transplantation, we initially studied the cumulative duration of HCTZ. However, as the log-linear association between the cumulative duration of use and outcomes could not be demonstrated (likely due to the low number of patients exposed to HCTZ combined with a relatively rare occurrence of events), such analysis was impracticable. We then categorized exposure to HCTZ according to three categories. In the corresponding multivariable Cox models, the time-dependent variables associated with HCTZ use were "no use," "mild-term use" when the cumulative HCTZ use was <2 years, and "long-term use" when it was ≥2 years.

We used R version 3.6.1 and the packages "base," "dplyr," "survival," "etm," "plotrix," "splines," "lattice," "prodlim," "forestplot," and "ReporteRs" for all data analyses.

Results

Recipient Demographic Characteristics

We included 2496 recipients of kidney (n=2155), combined kidney-pancreas (n=282), and pancreas (n=59) transplants. Patient characteristics subdivided by HCTZ status are displayed in Table 1.

Prior to transplantation, no patient was receiving HCTZ, which was consequently started after transplantation in all 279 (11%) participants exposed at least 1 month to HCTZ during the transplantation period. Probabilities of exposure to HCTZ at 5, 10, and 15 years post-transplantation were 10% (95% confidence interval [95% CI], 8% to 11%), 14% (95% CI, 12% to 15%), and 16% (95% CI, 14% to 18%), respectively (Figure 2). Mean duration of exposure and cumulative dose of HCTZ were 2.5 years (95% CI, 2.1 to 2.8 years) and 14,436 mg (95% CI, 12,238 to 16,634 mg), respectively. Median follow-up time was 5.9 years (range from 0.2 to 18.8 years). During the follow-up, 343 deaths with a functioning graft, 439 returns to dialysis, 154 basal cell carcinomas, and 132 squamous cell carcinomas occurred.

Basal Cell Carcinoma

The cumulative incidence curve for basal cell carcinoma is presented in Figure 3A. Cumulative incidence rates at 5, 10, and 15 years post-transplantation for basal cell carcinoma were 4% (95% CI, 3% to 5%), 8% (95% CI, 6% to 9%), and 11% (95% CI, 9% to 13%), respectively. Table 2 presents unadjusted analysis and the final multivariable model from which, among 2496 patients, 23 were excluded due to missing data. Multivariable Cox models were adjusted for baseline parameters (age, sex, retransplantation, type of transplantation, type of donor, HLA-A -B -DR mismatch greater than or equal to four, induction therapy, maintenance treatment at transplantation, use of steroids at transplantation, initial nephropathy, and cold ischemia time) and for time-varying covariates (rejection, maintenance treatment during follow-up [calcineurin inhibitor, mycophenolate derivatives, or mTOR inhibitor], and other malignancies). The confounder-adjusted hazard ratio (HR) associated with the HCTZ exposure was 0.63 (95% CI, 0.35 to 1.15). In sensitivity analysis of the subgroup of patients, again no association between HCTZ use and basal cell carcinoma was found (Table 3). The adjusted HRs associated with mild-term (<2 years) and long-term use (≥2 years) of HCTZ were 0.59 (95% CI, 0.27 to 1.32) and 0.70 (95% CI, 0.30 to 1.62), respectively.

Squamous Cell Carcinoma

The cumulative incidence curve of squamous cell carcinoma is presented in Figure 3B. Cumulative incidence rates at 5, 10, and 15 years post-transplantation for squamous cell carcinoma were 3% (95% CI, 3% to 4%), 7% (95% CI, 6% to 8%), and 9% (95% CI, 8% to 11%), respectively. Table 4 presents unadjusted analysis and the final multivariable model from which, among 2496 patients, 80 were excluded

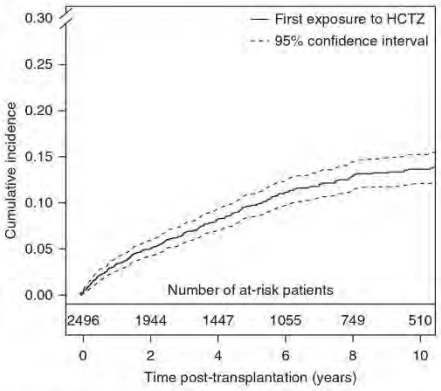

Figure 2. | Cumulative incidence curve of hydrochlorothiazide (HCTZ). Aalen–Johansen estimator, retransplantations, returns to dialysis, and deaths are competing events.

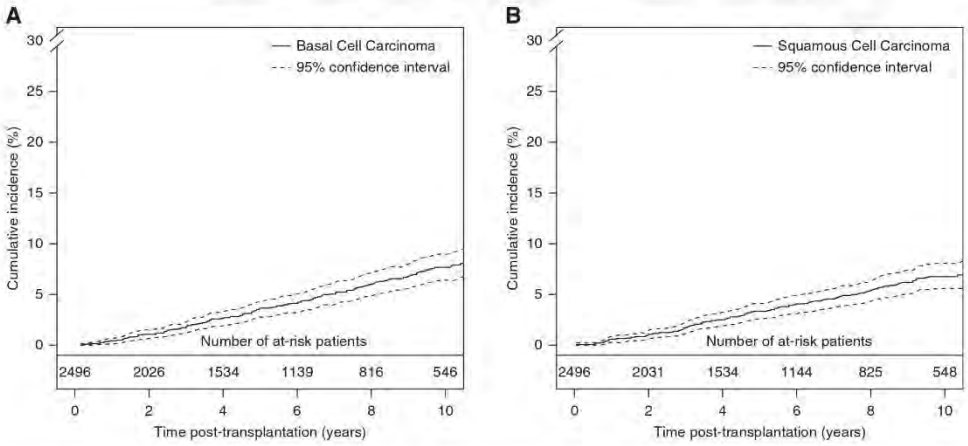

Figure 3. | Cumulative incidence curves for (A) basal cell carcinoma and (B) squamous cell carcinoma. Aalen–Johansen estimator, re-
Q:15 transplantations, returns to dialysis, and deaths are competing events.
Q:16

due to missing data. Multivariable Cox models were adjusted for baseline parameters (age, sex, retransplantation, type of transplantation, type of donor, HLA-A -B -DR mismatch greater than or equal to four, induction therapy, maintenance treatment at transplantation, use of steroids at transplantation, initial nephropathy, cold ischemia time, and delayed graft function) and for time-varying covariates (rejection, maintenance treatment during follow-up [calcineurin inhibitor, mycophenolate derivatives, or mTOR inhibitor], and other malignancies). The confounder-adjusted HR was 2.04 (95% CI, 1.27 to 3.28), indicating a two-fold higher risk of squamous cell carcinoma from time of exposure. We did not identify a significant dependence of the excess hazard with the time post-transplantation. As displayed in Table 3, sensitivity analysis maintained this association even when analysis was restricted to solitary kidney transplant recipients (HR, 2.10; 95% CI, 1.29 to 3.42). When we considered patients according to their baseline immunosuppression, the confounder-adjusted HRs were 1.94 (95% CI, 0.95 to 3.98) and 1.98 (95% CI, 0.91 to 4.33) for those on nondepleting induction agents and depleting induction agents, respectively, and the confounder-adjusted HRs were 2.26 (95% CI, 0.49 to 10.48) and 2.18 (95% CI, 1.30 to 3.64) for those on cyclosporin and tacrolimus, respectively, as calcineurin inhibitor drugs. To further explore a potential dose-response relationship, we performed a sensitivity analysis considering the exposure to HCTZ according to three categories. The adjusted HRs associated with a mild-term use (<2 years) and a long-term use (≥2 years) when compared with no use were 1.99 (95% CI, 1.08 to 3.68) and 2.12 (95% CI, 1.14 to 3.94), respectively.

Discussion

In our cohort of transplant recipients, we found that, following exposure to HCTZ post-transplantation, patients had a significantly higher risk of developing squamous cell carcinoma (HR, 2.04; 95% CI, 1.27 to 3.28), regardless the period of that exposure. By contrast, we have not found an association between HCTZ exposure and the development of basal cell carcinoma.

The association between HCTZ exposure and the incidence of squamous cell carcinoma was also found when the analysis was restricted to solitary kidney transplant recipients.

Our findings were made possible due to the availability and use of prospective data on the exposure (*i.e.*, HTCZ) and outcome variable (*i.e.*, keratinocyte cancer) over an extended length of time, combined with a multivariable model that incorporated both time-dependent exposure to HCTZ and confounders (at baseline and some time dependent [13]) that were not eliminated in the exclusion process (*i.e.*, patients on an mTOR inhibitor or azathioprine at baseline).

More than 60 years ago, HCTZ became the first available oral diuretic, and owing to its favorable safety profile, it demonstrated a clear benefit in the treatment of hypertension (15). However, as soon as it was introduced, severe skin photosensitivity reactions were reported (16,17). It is well established that a number of medications defined as photosensitizing potentiate the erythema reaction to UV light and through a photocarcinogenic effect, bring about a higher risk of squamous cell carcinoma (18). For instance, azathioprine is well known to exacerbate skin photosensitivity and is associated with a higher incidence of squamous cell carcinoma (19,20).

In 2013, on the basis of limited human evidence (21–23), the International Agency for Research on Cancer highlighted HCTZ as a possible carcinogen (2B). Since then, nationwide patient-control studies in Denmark have unearthed a strong association between HCTZ use and squamous cell carcinoma (6,24). These results were replicated in the United Kingdom (25), leading to multiple

Table 2. Results of the unadjusted and multivariable cause-specific, time-dependent Cox models studying the risk of basal cell carcinoma (*n*=2473)

COLUMN HEADING	Adjusted Cox Model			
	Hazard Ratio	95% Confidence Interval	Hazard Ratio	95% Confidence Interval
HTCZ exposure	0.79	[0.45 to 1.40]	0.63	[0.35 to 1.15]
Recipient age	1.05	[1.04 to 1.07]	1.05	[1.04 to 1.07]
Men	1.39	[0.99 to 1.95]	1.52	[1.04 to 2.23]
Initial nephropathy				
Chronic tubulointerstitial nephritis/ ADPKD/congenital uropathy (versus glomerulopathy)	1.22	[0.83 to 1.77]	1.25	[0.84 to 1.86]
Diabetes (versus glomerulopathy)	0.53	[0.29 to 0.96]	0.72	[0.31 to 1.63]
Other (versus glomerulopathy)	0.81	[0.42 to 1.57]	0.65	[0.33 to 1.28]
Vascular nephropathy (versus glomerulopathy)	1.94	[1.09 to 3.46]	1.07	[0.57 to 2.01]
KRT				
Hemodialysis (versus preemptive transplantation)	0.96	[0.63 to 1.47]	—	—
Peritoneal dialysis (versus preemptive transplantation)	1.51	[0.80 to 2.86]	—	—
Kidney transplantation	2.93	[1.44 to 5.94]	1.80	[0.60 to 5.40]
Retransplantation	1.09	[0.73 to 1.64]	1.04	[0.59 to 1.81]
Deceased donor	1.03	[0.62 to 1.71]	0.97	[0.54 to 1.72]
HLA-A -B -Dr mismatches ≥4	0.86	[0.63 to 1.19]	0.87	[0.62 to 1.24]
Cold ischemia time ≥18 h	0.74	[0.54 to 1.02]	0.56	[0.39 to 0.80]
Delayed graft function	0.96	[0.68 to 1.36]	—	—
Rejection	0.71	[0.36 to 1.40]	0.89	[0.45 to 1.73]
Other malignancies	0.89	[0.39 to 2.04]	0.38	[0.16 to 0.94]
Induction treatment				
Nondepleting (versus depleting)	1.11	[0.80 to 1.53]	0.79	[0.51 to 1.23]
None (versus depleting)	1.09	[0.45 to 2.60]	0.99	[0.41 to 2.43]
Calcineurin inhibitors				
Cyclosporin (versus tacrolimus)	1.13	[0.79 to 1.61]	1.31	[0.88 to 1.93]
None (versus tacrolimus)	0.67	[0.16 to 2.76]	0.68	[0.14 to 3.26]
Steroid-free regimen	1.03	[0.71 to 1.49]	0.98	[0.66 to 1.46]
Calcineurin inhibitors during follow-up	0.46	[0.32 to 0.67]	0.49	[0.28 to 0.85]
mTOR inhibitors during follow-up	1.87	[1.19 to 2.92]	1.01	[0.54 to 1.88]
Mycophenolate derivatives during follow-up	0.68	[0.46 to 0.99]	0.88	[0.58 to 1.34]

HCTZ, hydrochlorothiazide; ADPKD, autosomal dominant polycystic kidney disease; KRT, kidney transplant recipient; —, XXX; mTOR, mammalian target of rapamycin.

Q: 18

Q:19 **Table 3. Results of the sensitivity analyses**

COLUMN HEADING	Squamous Cell Carcinoma			
	Hazard Ratio	95% Confidence Interval	Hazard Ratio	95% Confidence Interval
Solitary kidney transplantation	0.67	[0.37 to 1.23]	2.10	[1.29 to 3.42]
Depleting induction	0.43	[0.17 to 1.13]	1.98	[0.91 to 4.33]
Nondepleting induction	0.78	[0.34 to 1.77]	1.94	[0.95 to 3.98]
Cyclosporin at transplantation	0.25	[0.04 to 1.63]	2.26	[0.49 to 10.48]
Tacrolimus at transplantation	0.87	[0.46 to 1.64]	2.18	[1.30 to 3.64]

national health agencies publishing safety alerts regarding its use in 2018 (26,27). More recently, no association was demonstrated between HCTZ and skin cancer in an Asian population, highlighting the likely importance of baseline photosensitivity to UV as a contributing factor (28). A strength of these studies was their ability to demonstrate a dose-response relationship between HCTZ exposure and risk of keratinocyte cancer. In keeping with a lesser role for cumulative UV exposure (5), only very long periods of HCTZ exposure have been associated with the development of basal cell carcinoma.

To investigate a potential dose-response relationship in our population, we considered the exposure to HCTZ according to three categories. Results for HRs for both mild-term (<2 years) use and long-term use (≥2 years) were comparable. In other words, we observed an association between HCTZ exposure and squamous cell carcinoma as soon as a mild-term exposure occurred and no dose-response relationship. In the general population, this association has only been demonstrated in those with prolonged exposure to HCTZ (>3 years) with a demonstrable dose-response relationship. The risk we have demonstrated in our study is comparable with that reported following 6 years of exposure in the original study. These discrepancies could be related to a potentially additive and interactive effect that HCTZ has on a group of patients intrinsically made vulnerable to squamous cell carcinoma through their exposure to immunosuppressive agents. Thus, the absence of a dose-response relationship in our cohort could be, on one hand, superseded by the effect of immunosuppressants and on the other hand, due to an inadequate follow-up interval compared with the baseline study conducted in the general population (6). These are only suppositions, and larger studies in terms of both numbers and length of follow-up are required to investigate them.

Squamous cell carcinoma in solid organ transplant recipients occurs as a result of complex interactions between immune-mediated mechanisms of tumor promotion (mainly human papilloma virus activation and reduced tumor surveillance) combined with UV-induced carcinogenic effects (4). Thus, the emergence of possibly negative synergistic effects with the addition of photosensitizing drugs such as HCTZ, which are suspected to enhance UV-induced DNA damage, is unsurprising (29). Similarly, several studies conducted in lung transplant recipients report that voriconazole exposure, used in the prophylaxis and treatment of invasive fungal infection and also implicated as a photosensitizing drug (30), is an independent risk factor for the development of squamous cell carcinoma (31–33).

Importantly, we were able to highlight the negative effects of using HCTZ in a population mainly treated with immunosuppressants belonging to the modern era, namely tacrolimus and mycophenolate mofetil. Indeed, as stated above, direct UV-induced carcinogenic effects are well documented with azathioprine, the maintenance therapy historically used in kidney transplantation, but such effects have not been established for cyclosporin, tacrolimus, and mycophenolate mofetil (20,34). With improvements in surveillance, this likely explains why the risk of squamous cell carcinoma in solid organ transplant recipients has drastically decreased in recent decades from 65 to 250 times (3) compared with the general population to 20 times (35,36). In sensitivity analysis, we observed an HR slightly higher with the use of depleting versus nondepleting induction agents, as has been previously reported (37).

Our observational study suffers from several limitations. First, it uses data that were not purposefully collected to answer the specific research question. Using data collected from a single center, the number of events detected was also limited. Furthermore, we lacked information regarding two important risk factors for keratinocyte cancer: UV exposure and skin phenotype (of note, most of the western France population is of fair skin type). However, there is no reason to assume there is a relationship between these risk factors and the use of HTCZ, which may not be considered as confounders. Second, HTCZ use prior to first transplantation would have been an important factor to consider, but this information was not collected in our database. Third, it would have been interesting to analyze each patient from the point of their first transplantation and then during subsequent dialysis and retransplantation periods. However, because follow-up data were not available for patients returning to dialysis, we considered the graft as the statistical individual by censoring the return to dialysis without keratinocyte cancers. Among consequences for retransplantations, prior exposure to HCTZ and other immunosuppressive drugs was not available. To tentatively consider the treatment exposures during previous transplantation or dialysis periods, we included in multivariable models the transplantation rank as a proxy. Finally, other unobserved confounding factors, such as other time-dependent drug exposures, cannot be excluded, making it difficult to establish firm causality.

Our results must be cautiously balanced with the risk related to poorly managed hypertension. Indeed, the overall prevalence of hypertension in kidney transplant recipients is up to 85% and is associated with shortened allograft

Table 4. Results of the unadjusted and multivariable cause-specific, time-dependent Cox models studying the risk of squamous cell carcinoma (*n*=2416)

COLUMN HEADING	Adjusted Cox Model			
	Hazard Ratio	95% Confidence Interval	Hazard Ratio	95% Confidence Interval
HTCZ exposure	2.36	[1.53 to 3.65]	2.04	[1.27 to 3.28]
Recipient age	1.07	[1.05 to 1.08]	1.06	[1.04 to 1.07]
Men	2.10	[1.40 to 3.13]	2.22	[1.45 to 3.39]
Initial nephropathy				
Chronic tubulointerstitial nephritis/ ADPKD/congenital uropathy (versus glomerulopathy)	0.75	[0.49 to 1.16]	0.86	[0.56 to 1.33]
Diabetes (versus glomerulopathy)	0.53	[0.29 to 0.97]	0.61	[0.26 to 1.40]
Other (versus glomerulopathy)	1.43	[0.83 to 2.46]	1.01	[0.53 to 1.90]
Vascular nephropathy (versus glomerulopathy)	1.62	[0.88 to 2.99]	0.73	[0.38 to 1.42]
KRT			—	—
Hemodialysis (versus preemptive transplantation)	1.54	[0.91 to 2.61]	—	—
Peritoneal dialysis (versus preemptive transplantation)	1.42	[0.63 to 3.23]	—	—
Kidney transplantation	2.49	[1.22 to 5.06]	0.68	[0.19 to 2.36]
Retransplantation	1.99	[1.36 to 2.90]	2.41	[1.16 to 4.99]
Deceased donor	2.79	[1.23 to 6.34]	1.65	[0.67 to 4.05]
HLA-A -B -Dr mismatches ≥4	0.90	[0.64 to 1.26]	0.96	[0.65 to 1.40]
Cold ischemia time ≥18 h	1.60	[1.13 to 2.29]	1.32	[0.88 to 1.98]
Delayed graft function	1.49	[1.05 to 2.11]	0.96	[0.66 to 1.40]
Rejection	0.17	[0.04 to 0.71]	0.24	[0.06 to 1.01]
Other malignancies	0.56	[0.17 to 1.81]	0.19	[0.05 to 0.66]
Induction treatment				
Nondepleting (versus depleting)	1.04	[0.74 to 1.48]	1.19	[0.61 to 2.35]
None (versus depleting)	0.77	[0.28 to 2.12]	0.97	[0.30 to 3.18]
Calcineurin inhibitors				
Cyclosporin (versus tacrolimus)	0.48	[0.29 to 0.80]	0.67	[0.39 to 1.16]
None (versus tacrolimus)	0.32	[0.04 to 2.42]	0.25	[0.03 to 1.90]
Steroid-free regimen	0.96	[0.64 to 1.43]	1.05	[0.66 to 1.68]
Calcineurin inhibitors during follow-up	0.39	[0.26 to 0.59]	0.29	[0.16 to 0.52]
mTOR inhibitors during follow-up	0.93	[0.49 to 1.76]	0.30	[0.13 to 0.68]
Mycophenolate derivatives during follow-up	0.47	[0.32 to 0.70]	0.64	[0.41 to 1.01]

HCTZ, hydrochlorothiazide; ADPKD, autosomal dominant polycystic kidney disease; KRT, kidney transplant recipient; —, XXX; mTOR, mammalian target of rapamycin.

survival and higher cardiovascular morbidity and mortality (38). Thiazide diuretics, including HCTZ, are recommended as a first-line treatment option for hypertension in the general population (39), and they seem efficient and safe for kidney transplant recipients (40,41). In addition to usual mechanisms, hypertension in kidney transplant recipients results from the use of immunosuppressive medications. For instance, calcineurin inhibitor–induced hypertension is related to kidney vasoconstriction, and it is efficiently counteracted by the use of calcium-channel blockers (42). Moreover, because sodium retention occurs due to activation of the thiazide-sensitive sodium chloride cotransporter by tacrolimus (43), HCTZ could be especially effective for this group of patients. Importantly, in the Q:11 Danish study, other antihypertensive drugs, including the usual alternative to HCTZ, bendroflumethiazide, another thiazide-type diuretic, and indapamide, a thiazide-like diuretic (the other main drug of this class, chlorthalidone, was not studied), were no associated with higher risks of skin cancer and could therefore be considered as an alternative.

In summary, our study suggests an association between HCTZ exposure and the development of squamous cell carcinoma in kidney transplant recipients with fair skin type. Broader studies will be required to confirm these results. Physicians should carefully evaluate the risks and benefits of HCTZ use, especially in kidney transplant recipients with known nonmodifiable risk factors, such as fair skin, men, and old age.

Disclosures

Q:12 J. Dantal reports receiving research funding from Novartis. All remaining authors have nothing to disclose.

Funding

Q:13 None.

Acknowledgments

Q:14 J. Dantal, F. Leborgne, T. Letellier, and S. Ville conceived and/or designed the study; A. Gaultier, C. Kerleau, and T. Letellier acquired data; J. Dantal, A. Gaultier, F. Leborgne, T. Letellier, and S. Ville played an important role in interpreting the results; J. Dantal, F. Leborgne, T. Letellier, and S. Ville drafted or revised the manuscript; and J. Dantal, F. Leborgne, T. Letellier, S. Ville, and members of the Divat Consortium approved the final version.

Supplemental Material

This article contains the following supplemental material online at http://cjasn.asnjournals.org/lookup/suppl/doi:10.2215/CJN.02560220/-/DCSupplemental.

Supplemental Material. Methods.

References

1. Au E, Wong G, Chapman JR: Cancer in kidney transplant recipients. *Nat Rev Nephrol* 14: 508–520, 2018
2. Rosales BM, De La Mata N, Vajdic CM, Kelly PJ, Wyburn K, Webster AC: Cancer mortality in kidney transplant recipients: An Australian and New Zealand population-based cohort study, 1980-2013. *Int J Cancer* 146: 2703–2711, 2020
3. Euvrard S, Kanitakis J, Claudy A: Skin cancers after organ transplantation. *N Engl J Med* 348: 1681–1691, 2003
4. Mittal A, Colegio OR: Skin cancers in organ transplant recipients. *Am J Transplant* 17: 2509–2530, 2017
5. Madan V, Lear JT, Szeimies R-M: Non-melanoma skin cancer. *Lancet* 375: 673–685, 2010
6. Pedersen SA, Gaist D, Schmidt SAJ, Hölmich LR, Friis S, Pottegård A: Hydrochlorothiazide use and risk of nonmelanoma skin cancer: A nationwide case-control study from Denmark. *J Am Acad Dermatol* 78: 673–681.e9, 2018
7. Aalen OO, Johansen S: An empirical transition matrix for non-homogeneous Markov chains based on censored observations. *Scand J Stat* 5: 141–150, 1978
8. Schemper M, Smith TL: A note on quantifying follow-up in studies of failure time. *Control Clin Trials* 17: 343–346, 1996
9. Shariff SZ, Cuerden MS, Jain AK, Garg AX: The secret of immortal time bias in epidemiologic studies. *J Am Soc Nephrol* 19: 841–843, 2008
10. Liu J, Weinhandl ED, Gilbertson DT, Collins AJ, St Peter WL: Issues regarding 'immortal time' in the analysis of the treatment effects in observational studies. *Kidney Int* 81: 341–350, 2012
11. Andersen PK, Geskus RB, de Witte T, Putter H: Competing risks in epidemiology: Possibilities and pitfalls. *Int J Epidemiol* 41: 861–870, 2012
12. Schoenfeld D: Partial residuals for the proportional hazards regression model. *Biometrika* 69: 239–241, 1982
13. Xie D, Yang W, Jepson C, Roy J, Hsu JY, Shou H, Anderson AH, Landis JR, Feldman HI; Chronic Renal Insufficiency Cohort (CRIC) Study Investigators: Statistical methods for modeling time-updated exposures in cohort studies of chronic kidney disease. *Clin J Am Soc Nephrol* 12: 1892–1899, 2017
14. Dekker FW, de Mutsert R, van Dijk PC, Zoccali C, Jager KJ: Survival analysis: Time-dependent effects and time-varying risk factors. *Kidney Int* 74: 994–997, 2008
15. Moser M, Feig PU: Fifty years of thiazide diuretic therapy for hypertension. *Arch Intern Med* 169: 1851–1856, 2009
16. Harber LC, Lashinsky AM, Baer RL: Photosensitivity due to chlorothiazide and hydrochlorothiazide. *N Engl J Med* 261: 1378–1381, 1959
17. Harber LC, Lashinsky AM, Baer RL: Skin manifestations of photosensitivity due to chlorothiazide and hydrochlorothiazide. *J Invest Dermatol* 33: 83–84, 1959
18. Stern RS: Photocarcinogenicity of drugs. *Toxicol Lett* 102-103: 389–392, 1998
19. Vos M, Plasmeijer EI, van Bemmel BC, van der Bij W, Klaver NS, Erasmus ME, de Bock GH, Verschuuren EAM, Rácz E: Azathioprine to mycophenolate mofetil transition and risk of squamous cell carcinoma after lung transplantation. *J Heart Lung Transplant* 37: 853–859, 2018
20. Hofbauer GFL, Attard NR, Harwood CA, McGregor JM, Dziunycz P, Iotzova-Weiss G, Straub G, Meyer R, Kamenisch Y, Berneburg M, French LE, Wüthrich RP, Karran P, Serra AL: Reversal of UVA skin photosensitivity and DNA damage in kidney transplant recipients by replacing azathioprine. *Am J Transplant* 12: 218–225, 2012
21. Jensen AØ, Thomsen HF, Engebjerg MC, Olesen AB, Sørensen HT, Karagas MR: Use of photosensitising diuretics and risk of skin cancer: A population-based case-control study. *Br J Cancer* 99: 1522–1528, 2008
22. de Vries E, Trakatelli M, Kalabalikis D, Ferrandiz L, Ruiz-de-Casas A, Moreno-Ramirez D, Sotiriadis D, Ioannides D, Aquilina S, Apap C, Micallef R, Scerri L, Ulrich M, Pitkänen S, Saksela O, Altsitsiadis E, Hinrichs B, Magnoni C, Fiorentini C, Majewski S, Ranki A, Stockfleth E, Proby C; EPIDERM Group: Known and potential new risk factors for skin cancer in European populations: A multicentre case-control study. *Br J Dermatol* 167[Suppl 2]: 1–13, 2012
23. Robinson SN, Zens MS, Perry AE, Spencer SK, Duell EJ, Karagas MR: Photosensitizing agents and the risk of non-melanoma skin cancer: A population-based case-control study. *J Invest Dermatol* 133: 1950–1955, 2013
24. Pottegård A, Hallas J, Olesen M, Svendsen MT, Habel LA, Friedman GD, Friis S: Hydrochlorothiazide use is strongly associated with risk of lip cancer. *J Intern Med* 282: 322–331, 2017
25. Morales DR, Pacurariu A, Slattery J, Kurz X: Association between hydrochlorothiazide exposure and different incident skin, lip and oral cavity cancers: A series of population-based nested case-control studies. *Br J Clin Pharmacol* 86: 1336–1345, 2020

26. Agence nationale de sécurité du médicament et des produits de santé: Hydrochlorothiazide - Risque de cancer de la peau non-mélanome (carcinome basocellulaire, carcinome épidermoïde) - Lettre aux professionnels de santé, 2018. Available at: https://www.ansm.sante.fr/S-informer/Informations-de-securite-Lettres-aux-professionnels-de-sante/Hydrochlorothiazide-Risque-de-cancer-de-la-peau-non-melanome-carcinome-basocellulaire-carcinome-epidermoide-Lettre-aux-professionnels-de-sante. Accessed May 20, 2020
27. United Kingdom Government: Hydrochlorothiazide: Risk of non-melanoma skin cancer, particularly in long-term use, 2018. Available at: https://www.gov.uk/drug-safety-update/hydrochlorothiazide-risk-of-non-melanoma-skin-cancer-particularly-in-long-term-use. Accessed May 20, 2020
28. Pottegård A, Pedersen SA, Schmidt SAJ, Lee C-N, Hsu C-K, Liao T-C, Shao S-C, Lai EC-C: Use of hydrochlorothiazide and risk of skin cancer: A nationwide Taiwanese case-control study. *Br J Cancer* 121: 973–978, 2019
29. Kunisada M, Masaki T, Ono R, Morinaga H, Nakano E, Yogianti F, Okunishi K, Sugiyama H, Nishigori C: Hydrochlorothiazide enhances UVA-induced DNA damage. *Photochem Photobiol* 89: 649–654, 2013
30. Haylett AK, Felton S, Denning DW, Rhodes LE: Voriconazole-induced photosensitivity: Photobiological assessment of a case series of 12 patients. *Br J Dermatol* 168: 179–185, 2013
31. Feist A, Lee R, Osborne S, Lane J, Yung G: Increased incidence of cutaneous squamous cell carcinoma in lung transplant recipients taking long-term voriconazole. *J Heart Lung Transplant* 31: 1177–1181, 2012
32. Singer JP, Boker A, Metchnikoff C, Binstock M, Boettger R, Golden JA, Glidden DV, Arron ST: High cumulative dose exposure to voriconazole is associated with cutaneous squamous cell carcinoma in lung transplant recipients. *J Heart Lung Transplant* 31: 694–699, 2012
33. Vadnerkar A, Nguyen MH, Mitsani D, Crespo M, Pilewski J, Toyoda Y, Bermudez C, Kwak EJ, Silveira FP, Clancy CJ: Voriconazole exposure and geographic location are independent risk factors for squamous cell carcinoma of the skin among lung transplant recipients. *J Heart Lung Transplant* 29: 1240–1244, 2010
34. Voskamp P, Bodmann CA, Koehl GE, Tensen CP, Bavinck JNB, Willemze R, Geissler EK, de Gruijl FR: No acceleration of UV-induced skin carcinogenesis from evenly spread dietary intake of cyclosporine in contrast to oral bolus dosages. *Transplantation* 96: 871–876, 2013
35. Rizvi SMH, Aagnes B, Holdaas H, Gude E, Boberg KM, Bjørtuft Ø, Helsing P, Leivestad T, Møller B, Gjersvik P: Long-term change in the risk of skin cancer after organ transplantation: A population-based nationwide cohort study. *JAMA Dermatol* 153: 1270–1277, 2017
36. Menzies S, O'Leary E, Callaghan G, Galligan M, Deady S, Gadallah B, Lenane P, Lally A, Houlihan DD, Morris PG, Sexton DJ, McCormick PA, Egan JJ, O'Neill JP, Conlon PJ, Moloney FJ: Declining incidence of keratinocyte carcinoma in organ transplant recipients. *Br J Dermatol* 181: 983–991, 2019
37. Bustami RT, Ojo AO, Wolfe RA, Merion RM, Bennett WM, McDiarmid SV, Leichtman AB, Held PJ, Port FK: Immunosuppression and the risk of post-transplant malignancy among cadaveric first kidney transplant recipients. *Am J Transplant* 4: 87–93, 2004
38. Weir MR, Burgess ED, Cooper JE, Fenves AZ, Goldsmith D, McKay D, Mehrotra A, Mitsnefes MM, Sica DA, Taler SJ: Assessment and management of hypertension in transplant patients. *J Am Soc Nephrol* 26: 1248–1260, 2015
39. Williams B, Mancia G, Spiering W, Agabiti Rosei E, Azizi M, Burnier M, Clement D, Coca A, De Simone G, Dominiczak A, Kahan T, Mahfoud F, Redon J, Ruilope L, Zanchetti A, Kerins M, Kjeldsen S, Kreutz R, Laurent S, Lip GYH, McManus R, Narkiewicz K, Ruschitzka F, Schmieder R, Shlyakhto E, Tsioufis K, Aboyans V, Desormais I; List of authors/Task Force members: 2018 practice guidelines for the management of arterial hypertension of the European society of hypertension and the European society of cardiology: ESH/ESC task force for the management of arterial hypertension [published correction appears in *J Hypertens* 37: 456, 2019 10.1097/HJH.0000000000002026. *J Hypertens* 36: 2284–2309, 2018
40. Taber DJ, Srinivas TM, Pilch NA, Meadows HB, Fleming JN, McGillicuddy JW, Bratton CF, Thomas B, Chavin KD, Baliga PK, Egede LE: Are thiazide diuretics safe and effective antihypertensive therapy in kidney transplant recipients? *Am J Nephrol* 38: 285–291, 2013
41. Moes AD, Hesselink DA, van den Meiracker AH, Zietse R, Hoorn EJ: Chlorthalidone versus amlodipine for hypertension in kidney transplant recipients treated with tacrolimus: A randomized crossover trial. *Am J Kidney Dis* 69: 796–804, 2017
42. Kuypers DRJ, Neumayer HH, Fritsche L, Budde K, Rodicio JL, Vanrenterghem Y; Lacidipine Study Group: Calcium channel blockade and preservation of renal graft function in cyclosporine-treated recipients: A prospective randomized placebo-controlled 2-year study. *Transplantation* 78: 1204–1211, 2004
43. Hoorn EJ, Walsh SB, McCormick JA, Fürstenberg A, Yang C-L, Roeschel T, Paliege A, Howie AJ, Conley J, Bachmann S, Unwin RJ, Ellison DH: The calcineurin inhibitor tacrolimus activates the renal sodium chloride cotransporter to cause hypertension. *Nat Med* 17: 1304–1309, 2011

Received: February 28, 2020 **Accepted:** September 28, 2020

*The Divat Consortium includes Gilles Blancho, Julien Branchereau, Diego Cantarovich, Agnès Chapelet, Clément Deltombe, Fadi Fakhouri, Lucile Figueres, Claire Garandeau, Magali Giral, Caroline Gourraud-Vercel, Maryvonne Hourmant, Lola Jacquemont, Sabine Le Bot, and Aurélie Meurette.

Published online ahead of print. Publication date available at www.cjasn.org.

UNIVERSITÉ DE NANTES

Vu, le Président du Jury,
(tampon et signature)

Professeur Jacques DANTAL

Vu, le Directeur de Thèse,
(tampon et signature)

Docteur Simon VILLE

Vu, le Doyen de la Faculté,

Professeur Pascale JOLLIET

UNIVERSITÉ DE NANTES

NOM : LETELLIER PRENOM : Thibault

Titre de Thèse : Association entre l'utilisation d'hydrochlorothiazide et le risque de cancer cutané non mélanome dans une population de patients transplantés rénaux

Association between use of hydrochlorothiazide and risk of keratinocyte cancers in kidney transplant recipients

---

ABSTRACT

Keratinocyte cancers represent a major concern for kidney transplant recipients.
Hydrochlorothiazide seems to be associated with a higher risk of keratinocyte cancer in general population. Our aim was to investigate whether hydrochlorothiazide use was associated with keratinocyte cancer in kidney transplant recipients.
In a single center of kidney (n = 2155), combined kidney-pancreas (n = 282), and pancreas (n = 59) transplant recipients, transplanted between 2000 and 2017 in Nantes (France), we evaluated the association between hydrochlorothiazide exposure and keratinocyte cancers.
Among participants, 279 of 2496 (11%) were exposed to hydrochlorothiazide after the transplantation. We found a relationship between exposure to hydrochlorothiazide and the risk of squamous-cell carcinomas (HR = 2.04, 95% CI: 1.27 to 3.28). In contrast, we found no association between hydrochlorothiazide exposure and basal-cell carcinomas (HR = 0.63, 95% CI: 0.35 to 1.15).

RESUME

Les cancers cutanés non-mélanome (principalement carcinome basocellulaire et carcinome épidermoïde cutané) sont un problème majeur pour les patients transplantés rénaux.
L'hydrochlorothiazide semble favoriser la survenue de cancers cutanés non-mélanome en population générale. Notre objectif était d'établir si l'utilisation d'hydrochlorothiazide en était un facteur de risque chez des patients transplantés rénaux.
Dans une cohorte monocentrique de patients transplantés à Nantes entre 2000 et 2017 de rein (n = 2155), rein-pancréas (n = 282) et pancréas (n = 59), nous avons évalué l'association entre l'exposition à l'hydrochlorothiazide et les cancers cutanés non-mélanome.
Parmi les participants, 279 sur 2496 (11%) étaient exposés à l'hydrochlorothiazide après la transplantation. Nous avons mis une évidence une relation entre l'exposition à l'hydrochlorothiazide et le risque de carcinome épidermoïde cutané (HR = 2.04, 95% CI : 1.27 to 3.28). A contrario, nous n'avons pas relevé d'association entre hydrochlorothiazide et carcinome basocellulaire (HR = 0.63, 95% CI : 0.35 to 1.15).

KEY-WORDS

KIDNEY TRANSPLANTATION; KERATINOCYTE CANCER; SQUAMOUS-CELL CARCINOMA; THIAZIDE DIURETICS

MOTS-CLES

TRANSPLANTATION RENALE ; CANCER CUTANE NON-MELANOME ; CARCINOME EPIDERMOÏDE CUTANE ; DIURETIQUES THIAZIDIQUES