British Journal of Cancer www.nature.com/bjc

ARTICLE

Clinical Studies

Exposure-response relationship of nivolumab and ipilimumab in patients with metastatic renal cell carcinoma from the randomised phase 2 BIONIKK study

Benoit Blanchet[1,2✉], Alicja Puszkiel[1,3], Anne Jouinot[4], Mostefa Bennamoun[5], Denis Maillet[6,7], Delphine Borchiellini[8], Brigitte Laguerre[9], Diane Pannier[10], Marine Gross-Goupil[11], Christine Chevreau[12], Philippe Barthélémy[13], Christophe Tournigand[14,15], Elodie Coquan[16], Gwenaelle Gravis[17], Eve Lepicard[18], Wolf Herman Fridman[19], Catherine Sautes-Fridman[19], Stéphane Oudard[20], Cheng-Ming Sun[19] and Yann-Alexandre Vano[19,21]

BACKGROUND: We aimed to investigate the exposure-response (E/R) relationship for ipilimumab and nivolumab in metastatic clear cell renal cell carcinoma (m-ccRCC) patients from the randomised phase 2 BIONIKK trial (EudraCT 2016-003099-28).
METHODS: This study included patients treated with either single-agent nivolumab (Nivo monotherapy group, $n = 39$) or nivolumab plus ipilimumab (Ipi/Nivo group, $n = 71$). Trough plasma concentrations (C_{min}) were assayed at week 6 after treatment start. Cox proportional hazard and logistic regression models were used to investigate the E/R relationship between C_{min} and clinical outcomes.
RESULTS: Low nivolumab C_{min} (<median) was not identified as an independent risk factor for progression in either the Nivo monotherapy group (HR 2.03, 95% CI [0.93–4.44]; $p = 0.076$) or the Ipi/Nivo group (HR 1.06, 95% CI [0.63–1.79]; $p = 0.83$). Interestingly, low ipilimumab C_{min} (<4.9 µg/mL) was independently associated with worse PFS in the Ipi/Nivo group (HR 1.77, 95% CI [1.03–3.05]; $p = 0.040$). In both groups, neither nivolumab C_{min} nor ipilimumab C_{min} was associated with the risk of death or grade ≥ 3 TRAEs occurrence.
CONCLUSIONS: This study suggests an E-R relationship for the efficacy of ipilimumab in m-ccRCC patients treated in combination with nivolumab. Prospective validation of our efficacy threshold in larger cohorts or phase 3 trials is essential prior to the implementation of a pharmacokinetically guided strategy.

British Journal of Cancer; https://doi.org/10.1038/s41416-026-03340-1

BACKGROUND

The therapeutic landscape for metastatic clear cell renal cell carcinoma (m-ccRCC) has significantly evolved over the past 5 years, particularly in the frontline setting with the use of immune checkpoint inhibitor-based combination regimens [1]. Nivolumab, a programmed cell death-1 (PD-1) inhibitor, associated with ipilimumab, a cytotoxic T-lymphocyte-associated protein 4 (CTLA-4) inhibitor, was among the first regimens to demonstrate a survival benefit over sunitinib in the frontline treatment of m-ccRCC, as shown in the CheckMate 214 trial [2]. This pivotal study led to the European Medicines Agency (EMA) approval of the nivolumab-ipilimumab combination in 2019. However, concerns

[1]Biologie du Médicament-Toxicologie, Hôpital Cochin, Institut du Cancer Paris CARPEM, AP-HP. Centre – Université Paris Cité, Paris, France. [2]CNRS, INSERM, CiTCoM, U1268, Université de Paris, Paris, France. [3]Inserm, UMR-S1144, Université Paris Cité, Paris, France. [4]Université Paris Cité, CNRS, INSERM, Institut Cochin, F-75014 Paris, France. [5]Department of Medical Oncology, Institut Mutualiste Montsouris, Paris, France. [6]Department of Medical Oncology, IMMUCARE, Centre Hospitalier Lyon Sud, Institut de Cancérologie des Hospices de Lyon (IC-HCL), Pierre-Bénite, France. [7]Faculté de médecine Jacques Lisfranc, Saint-Etienne, France. [8]Department of Medical Oncology, Centre Antoine Lacassagne, Université Côte d'Azur, Nice, France. [9]Department of Medical Oncology, Centre Eugene - Marquis, Rennes, France. [10]Department of Medical Oncology, Centre Oscar Lambret, Lille, France. [11]Department of Medical Oncology, Centre Hospitalier Universitaire de Bordeaux - Hôpital Saint-André, Bordeaux, France. [12]Department of Medical Oncology, Institut Universitaire du Cancer -Toulouse- Oncopole, Toulouse, France. [13]Department of Medical Oncology, Hôpitaux universitaires de Strasbourg, Strasbourg, France. [14]Department of Medical Oncology, Hôpital Henri-Mondor, AP-HP Centre - Université de Paris Est, Créteil, France. [15]INSERM, IMRB, Créteil, France. [16]Department of Medical Oncology, Centre de Lutte Contre le Cancer François Baclesse, Caen, France. [17]Department of Medical Oncology, Institut Paoli-Calmettes, Aix Marseille Univ, INSERM, CNRS, CRCM, Immunity and Cancer Team, Marseille, France. [18]ARTIC - Association pour la Recherche de Thérapeutiques Innovantes en Cancérologie (ARTIC), Hôpital Européen Georges Pompidou, AP-HP. Centre – Université Paris Cité, Paris, France. [19]Centre de Recherche des Cordeliers, INSERM, Equipe labellisée Ligue contre le Cancer, Université Paris Cité, Sorbonne Université, Paris, France. [20]Department of Medical Oncology, Hôpital Européen Georges Pompidou, Institut du Cancer Paris CARPEM, AP-HP. Centre – Université Paris Cité, Paris, France. [21]Department of Medical Oncology, Hôpital Foch, Université de Versailles St Quentin, Suresnes, France. ✉email: benoit.blanchet@aphp.fr

Received: 25 June 2025 Revised: 2 December 2025 Accepted: 25 January 2026
Published online: 06 February 2026

were raised by EMA experts regarding the added value of ipilimumab in terms of efficacy, especially given its association with increased risks of adverse events. Consequently, the EMA requested that Bristol Myers Squibb conduct a randomized controlled trial comparing the nivolumab-ipilimumab combination to nivolumab monotherapy in frontline m-ccRCC (CheckMate 8Y8 [NCT03873402]). To date, the results of this trial have not been reported yet.

Since the introduction of immune checkpoint inhibitors (ICIs), substantial efforts have been made to identify predictive biomarkers of efficacy and immune-related toxicities [3]. Among these, treatment dosing has been a key area of investigation. While higher doses of ipilimumab have been linked to better efficacy but with more frequent grade ≥3 adverse events in patients with advanced melanoma [4, 5], these findings have not been consistently replicated in m-ccRCC, particularly in combination with nivolumab. Notably, in patients treated with nivolumab monotherapy, dose range from 0.3 to 10 mg/kg did not appear to influence efficacy or safety outcomes in m-ccRCC [6]. Furthermore, a single study has compared different dosing schedules of nivolumab plus ipilimumab in m-ccRCC (nivolumab 1 mg/kg with ipilimumab 3 mg/kg versus nivolumab 3 mg/kg with ipilimumab 1 mg/kg), demonstrating comparable efficacy but higher toxicity with the higher dose of ipilimumab (3 mg/kg) [7].

Pharmacokinetic (PK) studies have investigated the exposure-response (E-R) of nivolumab and ipilimumab in advanced malignancies. Both of them show a dose-proportional increase in plasma exposure [8], but a time-varying clearance depending on tumour burden and factors related to the cachectic status (albumin, inflammatory syndrome) has been described in multiple PK studies [9–12]. Thus, their elimination increased with both higher tumour burden and protein catabolism in relation to cachectic status. Different studies demonstrated that nivolumab plasma exposure does not drive efficacy at the recommended dose (3 mg/kg) [9, 13, 14]; however, increased early clearance of nivolumab has been identified as a surrogate for disease state [9, 12, 13, 15]. Regarding toxicity, no significant exposure-response relationship has been reported at the recommended daily dose (3 mg/kg) [12–14, 16–18], consistent with the lack of a dose-response relationship observed up to 10 mg/kg during the clinical development of nivolumab [19]. For ipilimumab, different studies reported an E-R relationship for efficacy and toxicity in patients treated for advanced melanoma or hepatocellular carcinoma (HCC) [20–22]. Overall, data on the E-R relationship are largely derived from melanoma or non-small cell lung carcinoma cohorts, with very sparse data available for m-ccRCC patients treated in nivolumab monotherapy [12, 15] and none in combination with ipilimumab.

Recently, the BIONIKK multicenter randomized phase 2 trial was conducted to test whether molecular profiling could help guide first-line treatment (nivolumab, nivolumab plus ipilimumab or anti-VEGFR tyrosine kinase inhibitor (VEGFR-TKI)) in m-ccRCC patients [23]. In this context, we leveraged the ancillary program of the BIONIKK trial to evaluate the E-R relationship of nivolumab and ipilimumab in this population.

METHODS

Study design and participants

BIONIKK (EudraCT 2016-003099-28) was previously reported as a biomarker-driven, open-label, non-comparative, randomised, phase 2 trial from 15 university hospitals or expert cancer centres in France [23]. The eligibility criteria and trial design were previously described [23].

Treatment randomisation was based on different tumour groups (ccrcc1 to ccrcc1 4), which were determined according to transcriptomic data. Patients in the ccrcc1 and ccrcc4 groups received nivolumab monotherapy or nivolumab in combination with ipilimumab. Those in the ccrcc2 and ccrcc3 groups were treated with nivolumab-ipilimumab or VEGFR-TKI. This ancillary study only included patients treated with ICI (nivolumab monotherapy or ipilimumab plus nivolumab combination) for whom a plasma sample was available at week 6 after the start of treatment. The trial protocol was approved by the French Health authorities and ethics committee. All patients had signed a written informed consent before the beginning of this study.

Procedures

Patients in the Nivo monotherapy group received intravenous nivolumab 240 mg every 2 weeks. In the Nivo/Ipi group, patients were treated with intravenous nivolumab 3 mg/kg plus ipilimumab 1 mg/kg every 3 weeks up to four doses (three or more doses were mandatory to continue study treatment), followed by intravenous nivolumab 240 mg every 2 weeks. In case of severe toxicity, nivolumab and/or ipilimumab administration was delayed as previously reported in the original study [23]. Adverse events were graded until the end of the study (set at the end of treatment or after 18 months, whichever occurred first), according to the National Cancer Institute—Common Toxicity Criteria for Adverse Events (NCI-CTCAE) version 4.0. Disease assessments were performed with computed tomography or magnetic resonance imaging at baseline, 10 weeks after randomization and every 12 weeks thereafter. Progressive disease was confirmed by a consecutive assessment 6 weeks later. If not confirmed, patients were able to continue ICI-based treatment until the next evidence of progression. ICI was administered until progression, unacceptable toxicity or death.

Measurement of ICI plasma concentrations

Blood samples were collected just before the next ICI infusion at week 6 after treatment initiation, corresponding to nivolumab C_{min} after 3 doses in the Nivo monotherapy group and nivolumab and ipilimumab C_{min} after 2 doses in the Ipi/Nivo group, assuming no delay in theoretical administration. Blood was drawn into 5 mL EDTA Vacutainer tubes and then centrifuged. Plasma was transferred to polypropylene tubes and kept at −80 °C until assay. Nivolumab and ipilimumab concentrations were measured using the mAbXmise kit® (Promise, Grenoble, France) for extraction and then high-performance liquid chromatography-tandem mass spectrometry (LC-MS/MS) for quantification. This method was validated according to the EMA guidelines for bioanalytical methods, with intra- and inter-assay coefficients of analytical variability lower than 7.7% and 12.4%, respectively [24]. The lower limit of quantification (LLOQ) was set at 2 µg/mL for nivolumab and ipilimumab. Whatever ICI, plasma concentration below LLOQ was considered as half of LLOQ (i.e. 1 µg/mL) during the statistical analysis. The present LC-MS/MS method has been orthogonally cross-validated with two other techniques in order to ensure its robustness [25].

Antidrug antibodies (ADA) status screening

The presence of ipilimumab ADA (IPI-ADA) and nivolumab ADA (NIVO-ADA) at week 6 after treatment initiation was determined using qualitative anti-nivolumab and anti-ipilimumab ADA ELISA kits (AssayGenie, Dublin, Ireland). All experiments were performed according to the manufacturer's instructions.

Study endpoints

The primary endpoint was the Progression-free survival (PFS), defined as the time from randomization to the first RECIST-defined progression or death. Secondary endpoints included objective response rate (ORR), overall survival (OS) and incidence rate of grade ≥ 3 treatment-related adverse events (TRAE). ORR was defined as the percentage of patients having a partial or complete response according to RECIST, version 1.1 [26]. OS was defined as time between randomization and death (all causes included). Patients with no tumour progression or death at the end of follow-up were censored at the last date of evaluation.

Statistical analysis

For descriptive analyses, qualitative variables were expressed as number (%) and quantitative variables as median [interquartile range, IQR] or [95% confidence interval, 95% CI]. Comparisons between 2 groups were performed using Wilcoxon and Chi-square tests. Comparison between ≥3 groups were performed using Kruskal Wallis test. Normal distribution assumption for quantitative variables was evaluated by histograms and Shapiro–Wilk test. Variables deviating from the normal distribution were dichotomized for regression models. In the Ipi/Nivo group, univariate and

multivariate linear regression models were used to identify determinants of interindividual variability in ICI plasma exposure, using dose-normalized C_{min} (100-mg dose for ipilimumab and 240 mg for nivolumab). In the multivariate model, only those determinants with a p-value < 0.05 in the univariate analysis were retained. Regarding exposure–efficacy analysis, survival curves were obtained with Kaplan–Meier estimates and compared with log-rank test. Univariate and multivariate Cox proportional hazard regressions were used to identify risk factors of PFS and OS. Significant variables in univariate analyses ($p < 0.05$), as well as established prognostic factors and potential determinants of plasma exposure were included in the multivariate models. Results were expressed as Hazard Ratio (HR) with 95% CI. Regarding exposure–safety analysis, univariate logistic regression model was used to test the association between grade ≥ 3 TRAE onset and biological and clinical variables in the whole cohort ($n = 110$ patients), regardless of treatment group. Significant variables in univariate analyses as well as potential confounders were included in the multivariate models. Results were expressed as Odds Ratio (OR) with 95% CI. All p-values were two-sided, and the level of significance was set at $p < 0.05$. Statistical analyses were performed with the software R (version 4.1.0, packages *survival*, *survcomp*, *gtsummary* and *ggpubr*) with RStudio (version 1.4.1717).

RESULTS

Study cohort

One hundred and ninety-nine m-ccRCC patients were included in the BIONIKK study. Among them, 58 and 101 patients were enroled in the Nivo monotherapy and Ipi/Nivo groups, respectively, with plasma availability in 39 (67%) and 71 (70%) patients, respectively. Overall, 110 patients were eligible for the E/R analysis (Supplementary Fig. 1). Baseline characteristics of this subgroup of patients were similar to those of the overall BIONNIK population ($n = 199$), except, as expected, for the molecular group that guided the treatment (ICI or VEGFR-TKI) (Table 1). In this ancillary study, patients received immunotherapy only, allocated according to molecular group as follows: in the Nivo monotherapy group, ccrcc1 ($n = 28$) and ccrcc4 ($n = 11$); in the Ipi/Nivo group, ccrcc1 ($n = 28$), ccrcc2 ($n = 28$), ccrcc3 ($n = 1$) and ccrcc4 ($n = 14$). Baseline patient characteristics including albuminemia and CRP level were similar in the Nivo monotherapy and Ipi/Nivo groups (Supplementary Table 1). After a median follow-up of 39.6 [IQR 30.0–48.6] months, 89 patients (81%) had progressed and 49 (45%) had died.

Pharmacokinetics and ADA status

At week 6 after treatment initiation, the median nivolumab C_{min} (range) was statistically different between the Nivo monotherapy and Ipi/Nivo groups (56.5 (6.6–96.7) vs 30.7 (1.0–56.7) µg/mL, $p < 0.0001$; respectively) due to different dosing regimens. The median ipilimumab C_{min} (range) was 4.9 (1.0–22.5) µg/mL. The incidence of NIVO-ADA was 13% ($n = 5/39$) and 14% ($n = 10/71$) in the Nivo monotherapy and Ipi/Nivo groups, respectively. Three patients (4%) had IPI-ADA in the Ipi/Nivo group. The median dose-normalized nivolumab C_{min} was not significantly different in patients with NIVO-ADA compared to those without in the Nivo monotherapy group (40.0 [IQR 26.1–56.5] vs 57.0 [IQR 42.7–61.6] µg/mL, respectively; $p = 0.32$) while it was statistically lower in the Ipi/Nivo group (17.9 [IQR 13.3–19.8] vs 31.5 [IQR 24.7–31.8] µg/mL, respectively; $p = 0.0017$) (Supplementary Fig. 2). No statistical difference in median dose-normalized ipilimumab C_{min} was observed between patients with and without IPI-ADA (4.6 [IQR 4.1–7.6] vs 6.1 [IQR 3.7–8.5] µg/mL, respectively; $p = 1.0$). No statistical difference in plasma exposure was observed for either nivolumab or ipilimumab according to the molecular group in both groups (Supplementary Fig. 3A–C). In the Nivo monotherapy group, patients with poor IMDC risk had lower C_{min} compared to other patients ($p = 0.035$), whereas no statistical differences in ipilimumab or nivolumab C_{min} were observed in the Nivo/Ipi group (Supplementary Fig. 3D–F). In univariate analyses using only PK data from the Ipi/Nivo group, CRP level, albuminemia, ALP and NIVO-ADA status were significantly associated ($p < 0.05$) with nivolumab C_{min}, while CRP level and albuminemia were significantly associated with ipilimumab C_{min}. Regardless of the nature of ICI, dose-normalized plasma exposure was inversely correlated with both baseline CRP and albumin levels (Supplementary Fig. 4). In multivariate

Table 1. Baseline demographic and disease characteristics of the BIONIKK cohort.

Variable	BIONIKK population with PK samples ($n = 110$)	Overall BIONIKK population treated with ICI ($n = 199$)	p-value[d]
Age (years)	64.1 [37.5–82.6]	64.1 [21.7–83.6]	0.99
Gender, *n* (%)			0.25
Female	21 (19)	51 (26)	
Male	89 (81)	148 (74)	
ECOG performance status, *n* (%)			0.58
0	86 (78)	145 (73)	
1	20 (18)	46 (23)	
2	4 (4)	8 (4)	
Previous nephrectomy, *n* (%)			0.91
No	29 (26)	55 (28)	
Yes	81 (74)	144 (72)	
IMDC risk group, *n* (%)			0.72
Favourable	35 (32)	59 (30)	
Intermediate	49 (44)	98 (49)	
Poor	26 (24)	42 (21)	
Number of metastasis, *n* (%)			1.0
≥2	82 (75)	148 (74)	
1	28 (25)	51 (26)	
Molecular group [a], *n* (%)			0.044
Ccrcc1	56 (51)	83 (42)	
Ccrcc2	28 (25)	73 (37)	
Ccrcc3	1 (1)	9 (5)	
Ccrcc4	25 (23)	34 (17)	
Albumin (g/L)[b]	42.0 [14.9–52.0]	42.0 [14.9–52.0]	0.99
CRP (mg/L)[b]	5.7 [0.5–308.5]	6.2 [0.5–308.5]	0.88
eGFR (mL/min)[b,c]	65.0 [5.1–187.0]	66.0 [5.1–187.0]	0.39
LDH (UI/L)[b]	193 [104–524]	197 [90–895]	0.34
ALT (UI/L)[b]	21 [5–77]	21 [5–90]	0.84
AST (UI/L)[b]	19 [8–66]	20 [8–145]	0.40
ALP (UI/L)[b]	84 [33–637]	86 [33–1010]	0.85

Quantitative results are expressed as median [range].
ALP alkaline phosphatase, *ALT* alanine aminotransferase, *AST* aspartate aminotransferase, *CRP* C–reactive protein, *ECOG* Eastern Cooperative Oncology Group, *ICI* immune checkpoint inhibitor, *IMDC* International Metastatic Renal Cell Carcinoma Database Consortium, *eGFR* estimated glomerular filtration rate, *LDH* lactate dehydrogenase, *PK* pharmacokinetic.
[a]Distinct profiles of clear cell renal cell tumors (ccrcc1 to 4).
[b]Baseline value.
[c]Glomerular filtration rate was estimated according to the Modification of Diet in Renal Disease (MDRD) equation.
[d]p-value : difference between two groups.

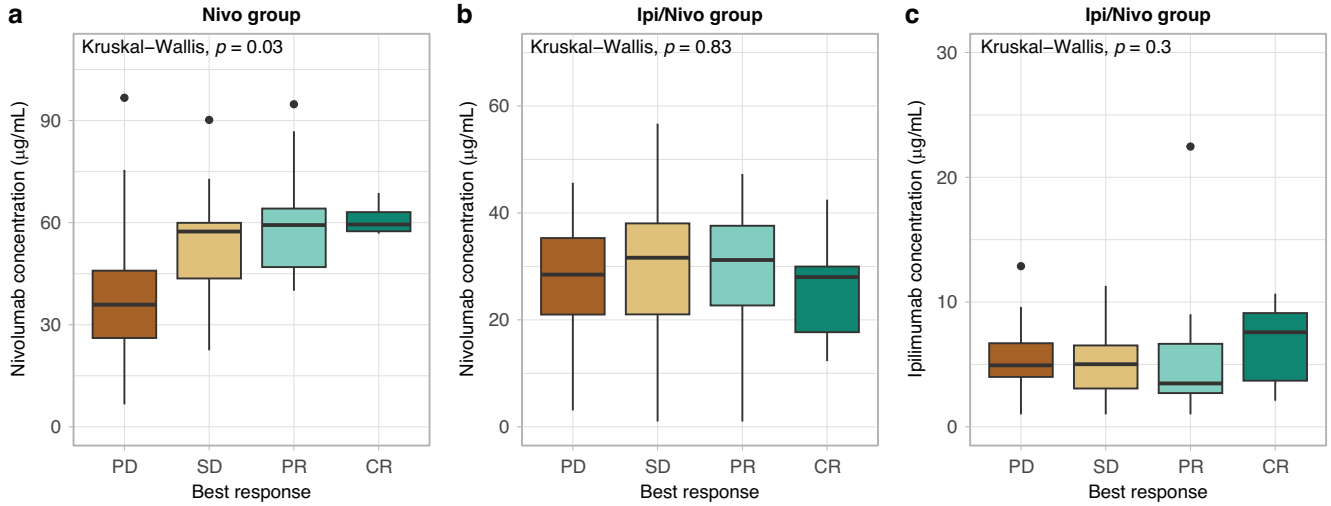

Fig. 1 Best objective response according to trough concentrations of nivolumab and ipilimumab in the Nivo monotherapy group and the Ipi/Nivo group. Panel **a** shows nivolumab trough concentrations in the monotherapy group. Panels **b** and **c** show nivolumab and ipilimumab trough concentrations, respectively, in the combination group. CR complete response, PR partial response, SD stable disease, PD progressive disease.

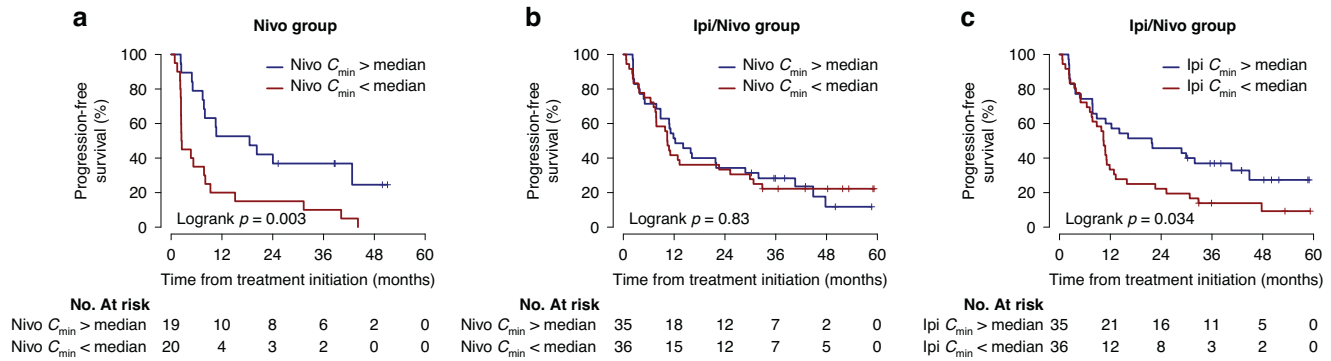

Fig. 2 Kaplan–Meier curves for progression-free survival (PFS) according to trough concentrations (C_{min}) of nivolumab and ipilimumab in the Nivo monotherapy group and the Ipi/Nivo group. Panel **a** shows PFS according to nivolumab C_{min} in the monotherapy group. Panels **b** and **c** show PFS according to nivolumab and ipilimumab C_{min}, respectively, in the combination group.

analyses, albuminemia and NIVO-ADA status were identified as independent determinants of nivolumab C_{min}, in the Ipi/Nivo group while no independent determinant of ipilimumab C_{min} was identified (Supplementary Table 2). In the Nivo monotherapy group, albuminemia was confirmed as an independent determinant of nivolumab C_{min} (data not shown).

Exposure-efficacy relationship

In the studied cohort ($n = 110$), the ORR was 54% (95% CI, 41–65) in the Nivo/Ipi group ($n = 38/71$) versus 38% (95% CI, 23 to 55) in the Nivo monotherapy group ($n = 15/39$), with complete responses in 9 patients (13%) versus 4 patients (10%). The DCR was 82% (95% CI, 71 to 90) in the Nivo/Ipi group ($n = 58/71$) versus 67% (95% CI, 50 to 81) in the Nivo monotherapy group ($n = 26/39$). At week 6, the median nivolumab C_{min} was significantly different among RECIST response groups (PD, SD, PR and CR) in the Nivo monotherapy group ($p = 0.03$) (Fig. 1a). In contrast, neither nivolumab C_{min} nor ipilimumab C_{min} was statistically different according to RECIST response groups in the Ipi/Nivo group (Fig. 1b, c). Similarly, patients with an objective response had higher median nivolumab C_{min} compared to others (59.3 [IQR 49.2–64.1] vs 43.6 [IQR 30.0–59.2] µg/mL, respectively; $p = 0.023$) in the Nivo monotherapy group, as well as in those with disease control (58.4 [IQR 46.7–62.9] vs 35.9 [IQR 26.1–45.9] µg/mL, respectively; $p = 0.0044$) (Supplementary Fig. 5). Conversely, neither nivolumab nor ipilimumab C_{min} was statistically different according to ORR ($p = 0.76$ and $p = 0.75$, respectively) and DCR ($p = 0.91$ and $p = 0.66$, respectively) in the Ipi/Nivo group (Supplementary Fig. 5).

The median PFS was 7.8 [95% CI, 4.9 to 18.5] months and 11.1 [95% CI, 9.9 to 21.8] months in the Nivo monotherapy and Ipi/Nivo groups, respectively. In the Nivo monotherapy group, median PFS was statistically shorter in patients with a nivolumab C_{min} below the median (i.e. <56.5 µg/mL) versus those with C_{min} above the median (2.5 [95% CI, 2.3 to 9.3] vs 18.5 [95% CI, 8.0 to not reached, NR months, respectively; log rank-test $p = 0.003$), while no difference was observed with the median cut of 30.7 µg/mL in the Ipi/Nivo group (10.4 [95% CI, 7.7 to 25.2] vs 12.3 [95% CI, 8.8 to 31.9] months, respectively; log rank-test $p = 0.83$) (Fig. 2a, b). Interestingly, patients with an ipilimumab C_{min} below the median (i.e. <4.9 µg/mL) had shorter median PFS than those with C_{min} above the median (10.4 [95% CI, 7.7 to 12.6] vs 21.8 [95% CI, 8.8 to NR] months, respectively; log rank-test $p = 0.034$) (Fig. 2c). The median PFS was not significantly associated with NIVO-ADA status in the Nivo monotherapy group (2.3 [95% CI, 2.2 to NR] vs 7.8 [95% CI, 4.9 to 18.5] months in patients with NIVO-ADA vs others, respectively; log rank-test $p = 0.80$) (Supplementary Fig. 6A), and in the Ipi/Nivo group (21.3 [95% CI, 9.9 to NR] vs 10.9 [95% CI, 7.8 to 21.8] months in patients with NIVO-ADA vs others, respectively; log rank-test $p = 0.15$) (Supplementary Fig. 6B). No statistical difference in median

Table 2. Univariate and multivariate Cox proportional hazard analysis of risk factors for progression in the two treatment groups.

	Nivo monotherapy group ($n = 39$)		Ipi/Nivo group ($n = 71$)	
Variable	**Hazard ratio (95% CI)**	***p*-value**	**Hazard ratio (95% CI)**	***p*-value**
Univariate analysis				
Age (per 1 year increase)	0.96 (0.93–1.00)	0.053	0.96 (0.94–0.99)	0.013
Sex				
Male vs female	0.87 (0.39–1.91)	0.73	0.76 (0.37–1.56)	0.46
Nephrectomy				
Yes vs No	0.54 (0.26–1.11)	0.09	0.68 (0.37–1.25)	0.21
IMDC				
Intermediate vs Favourable	0.57 (0.24–1.35)	0.20	0.78 (0.43–1.42)	0.42
Poor vs Favourable	1.52 (0.62–3.71)	0.36	1.28 (0.64–2.56)	0.49
Number of metastasis				
1 vs ≥ 2	0.42 (0.19-0.97)	0.043	0.84 (0.45-1.57)	0.59
ADA nivolumab at week 6				
Yes vs No	0.87 (0.30–2.56)	0.80	0.54 (0.23–1.26)	0.15
ADA iplimumab at week 6				
Yes vs No			1.33 (0.41–4.28)	0.64
Corticosteroids intake				
Yes vs No	0.83 (0.38–1.79)	0.80	1.43 (0.84–2.42)	0.19
Albumin				
≥35 vs <35 g/L	0.48 (0.19–1.19)	0.11	0.78 (0.35–1.74)	0.55
CRP (per mg/L increase)				
≥10 vs < 10 mg/L	1.44 (0.72–2.87)	0.30	0.69 (0.38–1.22)	0.22
Nivolumab C_{min} at week 6 (median)[a]				
Low vs high	2.80 (1.38–5.68)	0.0044	1.06 (0.63–1.79)	0.83
Ipilimumab C_{min} at week 6 (median)				
<4.9 vs ≥ 4.9 µg/mL			1.77 (1.04–3.03)	0.036
Multivariate analysis				
IMDC				
Intermediate/poor vs favourable	0.60 (0.25–1.44)	0.25	0.93 (0.54–1.62)	0.81
Albumin				
≥35 vs < 35 g/L	0.50 (0.18–1.38)	0.61		
Number of metastasis				
1 vs ≥ 2	0.44 (0.17–1.13)	0.087		
Age (per 1 year increase)			0.97 (0.94-0.99)	0.016
Nivolumab C_{min} (median)				
<56.5 vs ≥ 56.5 µg/mL	2.03 (0.93–4.44)	0.076		
Ipilimumab C_{min} (median)				
<4.9 vs ≥ 4.9 µg/mL			1.77 (1.03–3.05)	0.040

95% CI 95% confidence interval; *ADA* antidrug antibody, C_{min} trough concentration; *CRP* C–reactive protein, *ECOG* Eastern Cooperative Oncology Group, *IMDC* International Metastatic Renal Cell Carcinoma Database Consortium.
[a]Median value corresponds to 56.5 µg/ml and 30.7 µg/mL in the Nivo and Ipi/Nivo groups, respectively.

PFS was observed between patients with IPI-ADA and those without (11.8 [95% CI, 7.8 to NR] vs 10.9 [95% CI, 8.8 to 21.8] months, respectively; log rank-test $p = 0.64$) (Supplementary Fig. 6C). In univariate Cox regression analyses, number of metastasis ($p = 0.043$) and nivolumab C_{min} below the median (i.e. <56.5 µg/mL, $p = 0.0044$) were significantly associated with shorter PFS in the Nivo monotherapy group, as well as age ($p = 0.013$) and ipilimumab C_{min} below the median (i.e. <4.9 µg/mL, $p = 0.036$) in the Ipi/Nivo group (Table 2). In the multivariate analyses, nivolumab C_{min} below the median (i.e. <56.5 µg/mL) was not identified as an independent progression risk factor in the Nivo monotherapy group (HR 2.03, 95% CI [0.93 to 4.44]; $p = 0.076$). Interestingly, ipilimumab C_{min} below the median (i.e. <4.9 µg/mL) was independently associated with a worse PFS in the Ipi/Nivo group (HR 1.77, 95% CI [1.03 to 3.05]; $p = 0.040$) (Table 2).

The median OS was 44.5 [95% CI, 33.7 to NR] months and not reached [95% CI, 47.1 months to NR] in the Nivo monotherapy and Ipi/Nivo groups, respectively. Patients with a nivolumab C_{min} below the median (i.e. <56.5 µg/mL) had a worse median OS than those with a high nivolumab C_{min} (26.9 [95% CI, 15.8 to NR] vs NR [95% CI, 37.4 to NR] months, respectively; log rank-test $p = 0.014$) in the Nivo monotherapy group (Fig. 3a). This association was not

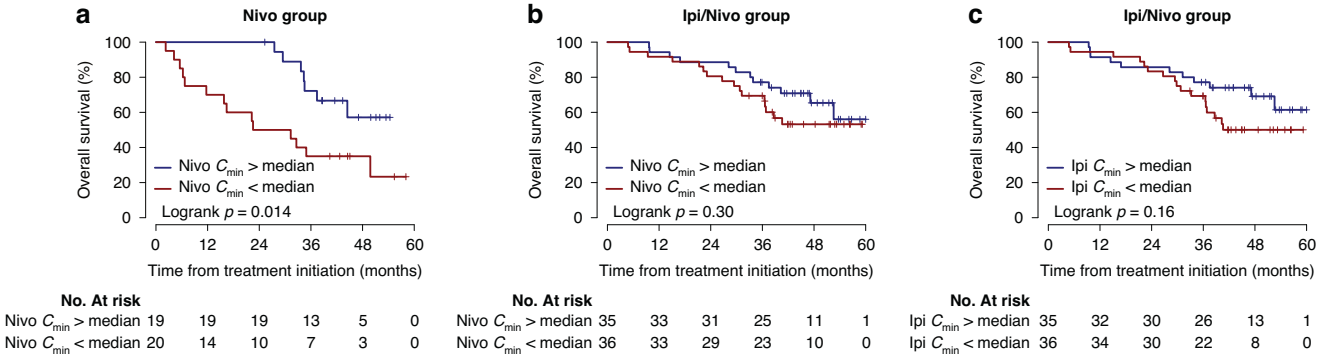

Fig. 3 Kaplan–Meier curves for overall survival (OS) according to trough concentrations (C_{min}) of nivolumab and ipilimumab in the Nivo monotherapy group and the Ipi/Nivo group. Panel **a** shows OS according to nivolumab C_{min} in the monotherapy group. Panels **b** and **c** show OS according to nivolumab and ipilimumab C_{min}, respectively, in the combination group.

observed in the Ipi/Nivo group when using a median cutoff of 30.7 µg/mL (NR [95% CI, 36.6 to NR] vs NR [95% CI, 52.6 to NR], respectively; log rank-test $p = 0.30$) (Fig. 3b). The median OS was not significantly shorter in patients with ipilimumab C_{min} below the median (i.e. <4.9 µg/mL) than in those with C_{min} above the median in the Ipi/Nivo group (NR [95% CI, 36.6 to NR] vs NR [95% CI, 52.6 to NR, respectively; log rank-test $p = 0.16$) (Fig. 3c). No statistical difference in median OS was observed between patients with positive and negative ADA status, regardless of the ICI and the treatment group (Supplementary Fig. 6D–F). In the multivariate Cox regression analyses, only albuminemia ≥35 g/L was identified as an independent protector factor of death in the Nivo monotherapy group (HR 0.29, 95% CI [0.10 to 0.87]; $p = 0.027$) (Table 3).

Exposure-safety relationship

Grade ≥ 3 TRAEs occurred in 7/39 (18%) and 30/71 (42%) of patients in the Nivo monotherapy and Ipi/Nivo groups, respectively. The median nivolumab C_{min} was not statistically different between patients with and without grade ≥ 3 TRAEs in the Nivo monotherapy group (47.5 [IQR 44.9–59.2] vs 56.6 [IQR 38.2–62.0] µg/mL, respectively; $p = 0.9$) and in the Ipi/Nivo group (32.0 [IQR 23.3–37.9] vs 28.5 [IQR 20.2–35.4] µg/mL, respectively; $p = 0.32$), nor did ipilimumab C_{min} (5.73 [IQR 2.88–6.76] vs 4.79 [IQR 2.80–6.90] µg/mL, respectively; $p = 0.71$) (Supplementary Fig. 7). In the univariate analysis including all patients ($n = 110$), concomitant administration of ipilimumab was identified as the single risk factor of grade ≥ 3 TRAEs (HR 3.34, 95% CI [1.30 to 8.59]; $p = 0.012$) (Supplementary Table 3). Finally, the Supplementary Fig. 8 shows the frequency of immune-related adverse events (irAE) according to severity and quartile exposure for each ICI.

DISCUSSION

The ipilimumab/nivolumab combination is one of the recommended frontline treatments for IMDC intermediate/poor risk m-ccRCC patients. However, the identification of biomarkers to predict efficacy and immune-related toxicity in this population remains a challenge. Among the potential biomarkers, plasma exposure to these ICIs may be a candidate. To the best of our knowledge, the present study is the first to characterise the E-R relationship (including ADA status) in m-ccRCC patients treated in frontline with either single-agent nivolumab or nivolumab/ipilimumab combination within a prospective randomised trial.

Different PK studies have shown that cancer patients with advanced cachexia exhibit increased ICI clearance, resulting in reduced plasma exposure to nivolumab and ipilimumab [9, 11, 15, 27]. In our study, low baseline albumin (<35 g/L) and ADA positivity were associated with reduced nivolumab plasma exposure, indicating increased clearance. This effect was not observed with ipilimumab. After adjusting for biological and clinical variables including albumin level, a nivolumab $C_{min} < 56.5$ µg/mL at week 6 was not an independent predictor of progression in the Nivo monotherapy group. These findings align with the NIVOREN study, where $C_{min} > 48$ µg/mL at week 6 did not predict PFS in mRCC patients treated with single-agent nivolumab [12]. Notably, both studies found shorter OS in patients with low nivolumab C_{min} (<median), but this association lost significance after adjusting for cachexia, a known poor prognostic factor in m-ccRCC [28]. Prior PK/PD studies suggest that increased nivolumab clearance reflects underlying disease severity rather than reduced drug efficacy [9, 12, 13, 15]. Moreover, a phase II trial in m-ccRCC showed no dose-response relationship across a broad dosing range (0.3–10 mg/kg) [6], reinforcing that the worse survival of patients with $C_{min} < 56.5$ µg/mL in the Nivo monotherapy group was due to the worse prognostic status of the patients rather than a direct PK-related impact of nivolumab in our study. Taken together, these results suggest that low nivolumab C_{min} is more likely a reflection of disease biology rather than a modifiable determinant, and therefore does not support pharmacokinetically-guided dose adjustments of nivolumab during the early phase of treatment. Nevertheless, several studies reported prognostic value of early nivolumab clearance in identifying patients at higher risk of death [12, 29]. If such strategy is applied, PK sampling could help to identify patients with high clearance. Different clinical studies, such as the MOIO phase 3 trial (a non-inferiority study of PFS for patients responding to 6 months of ICI-based treatment) [30] are ongoing to assess the efficacy of extended dosing intervals versus the standard administration of anti-PD1/PDL1 agents. In this setting, the use of therapeutic drug monitoring could be helpful to individualise the dosing interval of nivolumab in an era of precision medicine [31, 32]. If the nivolumab de-escalation strategy is successful, it could be used in daily clinical practice, with particular relevance in resource-limited settings. Finally, although the pharmacokinetic data of nivolumab from our study cannot be extrapolated to ICI–TKI combinations such as cabozantinib–nivolumab, a dose and/or dosing frequency de-escalation strategy should be considered in patients concomitantly treated with cabozantinib.

In comparison with the Nivo monotherapy group, the association between nivolumab C_{min} below median (i.e. <30.7 µg/mL) and worse PFS was not confirmed in univariate analysis in the Ipi/Nivo group. However, comparing exposure–efficacy between two groups remains exploratory, as BIONIKK trial was not designed for this purpose. The difference in sampling occasion (C_{min} after 3 doses in the Nivo monotherapy group versus 2 doses in the Ipi/Nivo group) limits the interpretation and cross-arm comparison of our nivolumab pharmacokinetic results. Indeed, the difference in

Table 3. Univariate and multivariate Cox proportional hazard analysis of risk factors for death in the two treatment groups.

	Nivo monotherapy group ($n = 39$)		Ipi/Nivo group ($n = 71$)	
Variable	**Hazard ratio (95% CI)**	***p*-value**	**Hazard ratio (95% CI)**	***p*-value**
Univariate analysis				
Age (per 1 year increase)	0.98 (0.93–1.02)	0.34	0.99 (0.95–1.03)	0.57
Sex				
Male vs female	0.99 (0.38–2.56)	0.99	0.60 (0.24–1.48)	0.27
Nephrectomy				
Yes vs No	1.07 (0.41–2.76)	0.90	0.60 (0.26–1.37)	0.23
IMDC				
Intermediate vs favourable	2.08 (0.57–7.56)	0.27	1.52 (0.63–3.66)	0.36
Poor vs favourable	3.36 (0.89–12.76)	0.08	2.41 (0.87–6.68)	0.09
Number of metastasis				
1 vs ≥2	0.42 (0.14–1.26)	0.12	0.44 (0.15–1.27)	0.13
ADA nivolumab at week 6				
Yes vs No	1.79 (0.52–6.12)	0.35	0.66 (0.20–2.18)	0.49
ADA iplimumab at week 6				
Yes vs No			2.70 (0.81–8.97)	0.11
Corticosteroids intake				
Yes vs No	1.22 (0.49–3.02)	0.67	1.22 (0.49–3.02)	0.67
Albumin				
≥35 vs <35 g/L	0.22 (0.08–0.60)	0.003	0.32 (0.12–0.89)	0.03
CRP (per mg/L increase)				
≥10 vs <10 mg/L	2.28 (0.96–5.45)	0.06	0.73 (0.31–1.74)	0.48
Nivolumab C_{min} at week 6 (median)[a]	2.98 (1.20–7.42)	0.019		
Low vs high			1.48 (0.70–3.13)	0.31
Ipilimumab C_{min} at week 6 (median)				
<4.9 vs ≥ 4.9 µg/mL			1.72 (0.80–3.68)	0.16
Multivariate analysis				
IMDC				
Intermediate/poor vs favourable	1.35 (0.33–5.46)	0.68	1.65 (0.68–3.99)	0.27
Albumin				
≥35 vs < 35 g/L	0.29 (0.10–0.87)	0.027	0.37 (0.13–1.02)	0.055
Nivolumab C_{min} (median)				
<56.5 vs ≥ 56.5 µg/mL	2.45 (0.93–6.44)	0.07		

95% CI 95% confidence interval; *ADA* antidrug antibody, C_{min} trough concentration, *CRP* C–reactive protein; *ECOG* Eastern Cooperative Oncology Group; *IMDC* International Metastatic Renal Cell Carcinoma Database Consortium.
[a]Median value corresponds to 56.5 µg/ml and 30.7 µg/mL in the Nivo and Ipi/Nivo groups, respectively.

nivolumab E-R relationship may be related to differences in the dynamics of clearance changes between patients treated with nivolumab monotherapy and those receiving the combination with ipilimumab. Nivolumab clearance is known to decrease with improving disease dynamics over time in RCC patients [9]. Furthermore, Zhang et al. reported that the mean time to half of the maximal change in CL over time is 70 days in patients treated with nivolumab monotherapy and 109 days in those treated in combination with ipilimumab [33]. This suggests that the timing of the sampling occasion for assaying nivolumab concentration is a critical factor in the analysis of the E-R relationship. PK sample at week 6 (42 days) might be therefore a better marker of clearance change over time in the Nivo monotherapy group than in the Ipi/Nivo group. In this context, the lack of a later PK sampling time point represents a limitation of our analysis.

The main finding of our study is the identification of a low ipilimumab C_{min} (i.e. <4.9 µg/mL) at week 6 as an independent risk factor for progression in the Ipi/Nivo group. Furthermore, OS in patients with $C_{min} < 4.9$ µg/mL tended to be poorer but without reaching the statistical significance. Consistent with our findings, several studies have reported an exposure-survival relationship for ipilimumab in patients treated in monotherapy for advanced melanoma or in combination with nivolumab for advanced HCC [20–22]. In the present study, although adjusted for confounders like albumin and CRP, it is unclear whether the E–R relationship for PFS is genuine or influenced by hidden factors. Large randomized studies with multiple dose levels can help clarify the true exposure–efficacy relationship, because dose–response analyses are less affected by prognostic confounders [34]. In 217 patients with advanced melanoma, a randomised, phase 2, dose-ranging study of ipilimumab at 0.3, 3 and 10 mg/kg as monotherapy

reported a dose-dependent effect on efficacy, with an improvement in best overall response with an increased dose [4]. Subsequently, a randomised, double, blind, phase 3 post-marketing trial including 727 patients with advanced melanoma showed a longer median OS in patients treated in monotherapy with the 10-mg/kg dose compared to those treated with the 3-mg/kg dose (HR 0.84, $p = 0.04$) [5]. In patients with advanced HCC treated with ipilimumab (1 or 3 mg/kg) plus nivolumab (3 or 1 mg/kg), Sangro et al. observed a significant dose-effect of ipilimumab with respect to OS [20]. These results show an improved efficacy with increased ipilimumab doses, which supports an E-R relationship for efficacy in these two tumour types. However, Hammers et al. reported a similar 2-year OS in m-ccRCC patients treated with ipilimumab 1 or 3 mg/kg in combination with nivolumab (67.3% vs 69.6%, respectively) [7], which precludes the conclusion on a clear E-R relationship of ipilimumab. In this context, our 4.9 µg/mL threshold must be validated in larger prospective cohorts or phase 3 trials before clinical implementation. If validated, early drug monitoring of ipilimumab could be integrated in routine practice, leading to different therapeutic strategies, such as dose escalation for subsequent infusions or extended induction at a dose of 1 mg/kg, for patients with low plasma exposure (<4.9 µg/mL) and a good safety profile. Finally, this efficacy threshold cannot be generalised to other tumours, such as advanced non-small-cell lung cancer and microsatellite-instability–high metastatic colorectal cancer, when 1 mg/kg of ipilimumab is used in combination with nivolumab.

Identifying the risk factors for the development of grade ≥ 3 TRAEs is a challenge in preventing treatment discontinuation of ICIs in m-ccRCC patients. As expected, we identified concomitant administration of ipilimumab as an independent risk factor for grade ≥ 3 TRAE, but no association between increased ipilimumab C_{min} and the occurrence of grade ≥ 3 TRAEs. A meta-analysis showed that the incidence of grade ≥ 3 TRAEs in metastatic melanoma patients is dependent on ipilimumab dose, with a 27% reduced risk with lower doses (3 vs 10 mg/kg) [35]. Besides, Feng et al. found an exposure-safety relationship in melanoma patients treated with single-agent ipilimumab at doses of 0.3 to 10 mg/kg [22]. According to their modelling, the probability of experiencing grade ≥ 3 irAE would be ~10% and 30% for steady-state C_{min} of 10 and 50 µg/mL, respectively. In melanoma patients treated with single-agent ipilimumab (3 mg/kg), Kogushi et al. reported higher ipilimumab C_{min} at week 7 in patients who developed irAEs than in those who did not [21]. In their cohort, the median ipilimumab C_{min} was 16.3 (IQR 11.4–20.9) µg/mL. Among patients with HCC treated in combination with nivolumab, a higher incidence of grade ≥3 immune-mediated adverse events has been observed in those receiving ipilimumab at 3 mg/kg (mean average concentration during cycle 1 :16.7 µg/mL) compared to 1 mg/kg (5.1 µg/mL) [20]. Collectively, these findings indicate that ipilimumab-related toxicity is dose-dependent. In our cohort, the median ipilimumab C_{min} at week 6 was 4.9 (IQR 2.8–6.8) µg/mL, consistent with the low 1 mg/kg dose, and suggesting that plasma exposure may remain below the threshold associated with the occurrence of grade ≥3 TRAEs. Overall, these results suggest that the high incidence of grade ≥ 3 TRAEs in the Ipi/Nivo group was mainly due to the immunological synergy between nivolumab and ipilimumab. Regarding nivolumab, the present study also shows a lack of E-R for safety in agreement with a recent study conducted in m-ccRCC patients [12]. Furthermore, Motzer et al. reported no significant effect of doses of 0.3 to 10 mg/kg on grade ≥ 3 TRAEs incidence in m-ccRCC patients treated in monotherapy [6]. Plasma exposure in patients treated with 10 mg/kg was more than three times higher than that observed in our cohort, which supports our findings. Finally, several PK/PD studies in other tumour types have reported a lack of exposure-response for safety in patients treated with nivolumab monotherapy (3 mg/kg, 240 mg every 2 weeks or 20 mg every 3 weeks) [13, 14, 16, 18, 36, 37], confirming the flat exposure-response over the range of exposures achieved in our patients.

Our understanding of the impact of ADAs on plasma drug concentrations and clinical outcomes in ICI-treated metastatic cancer patients remains limited. Consistent with CHECKMATE trials (1.1%–8.5% incidence) [38], we found low IPI-ADA incidence, though Kneverland et al. reported higher rates (26%) [39], possibly due to differences in ADA detection methods. In line with findings in melanoma patients treated with ipilimumab single-agent [39], we did not observe any impact of IPI-ADA positivity on IPI plasma concentrations. In contrast, we could not confirm an association between ADA positivity and worse OS, possibly due to differences in tumour type or treatment regimen (monotherapy versus combination). NIVO-ADA incidence ranged from 11.2% to 12.7% in single-agent nivolumab patients, regardless of the tumour type [38, 40], which is consistent with the percentage observed in our study monotherapy group (13%). The FDA packaging insert notes higher NIVO-ADA rates (21.9%) when combined with 3 mg/kg ipilimumab versus 10.0% in monotherapy [41]. In our study, coadministration of low-dose ipilimumab (1 mg/kg) did not significantly increase NIVO-ADA incidence, suggesting a dose-dependent effect on NIVO-ADA production. Using a population approach, Agrawal et al. reported a 14% increase in nivolumab clearance in NIVO-ADA-positive patients receiving single-agent nivolumab [40]. In the Nivo monotherapy group, the median dose-normalized nivolumab C_{min} was 30% lower in patients with ADA compared to that in patients without ADA but this difference was not statistically significant. In contrast, it was approximately twofold lower and statistically significant in NIVO-ADA positive patients compared to those without NIVO-ADA in the Ipi/Nivo group, and NIVO-ADA positivity was identified as an independent determinant of nivolumab C_{min}. The lack of statistical power in the Nivo monotherapy group ($n = 39$) could explain this result. Furthermore, the greater ipilimumab-induced immune stimulation could also result in a higher production of NIVO-ADA per patient and therefore a more significant effect on nivolumab clearance. However, our qualitative assay could not address this hypothesis. Furthermore, the inability of our assay to distinguish between neutralizing and non-neutralizing antibodies also introduces a bias in the interpretation of our pharmacokinetic results. Our exploratory results require confirmation through the use of more robust and specific assays. Agrawal et al. showed no significant effect of NIVO-ADA positivity on survival in 1,086 patients treated for various types of metastatic cancer [40], which supports our results. As far as we know, no association between worse survival and neutralizing ADA positivity has been still described. As noted earlier, a limitation of our study was our inability to differentiate neutralizing from non-neutralizing ADAs for both ipilimumab and nivolumab, though their positivity frequencies are low in the literature (0–1.6% and 0–2.8%, respectively) [40, 41]. Future studies should use more robust ADA assays able to differentiate neutralizing from non-neutralizing antibodies to precisely evaluate the clinical relevance of ADA types.

Our study has several limitations. The first limitation of our PK/PD study is the relatively small sample size, with only 110 patients with plasma samples available at week 6, representing 63% of the cohort treated with ICIs in the BIONIKK study. Compared to industrial studies, academic studies such as BIONIKK use broader inclusion criteria to better reflect real-world patients, but their less stringent biological sample collection conditions lead to lower patient enrolment. Missing samples were mainly attributed to non-compliance with collection protocols at certain centres. A second limitation is the unbalanced randomisation between patients receiving single-agent nivolumab and those receiving the combination with ipilimumab, potentially leading to differences in levels of immune cell infiltration and inflammation in the tumour microenvironment. A third limitation is that, unlike ICI-tyrosine kinase inhibitor combinations, nivolumab monotherapy is

not a standard first-line treatment. These ICI-tyrosine kinase inhibitor combinations were not represented in this study. Finally, the timing of PK assessment at week 6 was not optimal since PK metrics derived from the first dose would be more relevant to explore the E-R relationship because of the time-varying clearance of ICIs [9]. The BIONIKK trial was not originally designed for PK/PD analyses; however, ancillary studies were subsequently implemented to investigate early plasma biomarkers of treatment response. Week 6 was selected to align with the dosing schedules of both nivolumab (every 2 weeks) and the nivolumab–ipilimumab combination (every 3 weeks), thereby maximising likelihood of sample availability. Nevertheless, the present study has also clinical and analytical strengths related to the clinical trial design: two arms of treatment (nivolumab in monotherapy or in combination with ipilimumab) with randomisation, the long follow-up time, standardised collection of clinical data, uniform collection and storage of samples and the use of rigorously validated multiplex LC-MS/MS method [24].

CONCLUSION

The present study suggests an E-R relationship for efficacy of ipilimumab in m-ccRCC patients treated in combination with nivolumab. A lower ipilimumab C_{min} (i.e. <4.9 µg/mL) at week 6 may be associated with a shorter PFS. As our results are exploratory, prospective validation of the efficacy threshold in larger studies or phase 3 trials is essential before implementing a pharmacokinetically guided strategy.

DATA AVAILABILITY

The datasets used and analysed during the current study are available from the corresponding author upon reasonable request.

REFERENCES

1. Albiges L, Powles T, Staehler M, Bensalah K, Giles RH, Hora M, et al. Updated European Association of Urology Guidelines on renal cell carcinoma: immune checkpoint inhibition is the new backbone in first-line treatment of metastatic clear-cell renal cell carcinoma. Eur Urol. 2019;76:151–6.
2. Motzer RJ, Rini BI, McDermott DF, Arén Frontera O, Hammers HJ, Carducci MA, et al. Nivolumab plus ipilimumab versus sunitinib in first-line treatment for advanced renal cell carcinoma: extended follow-up of efficacy and safety results from a randomised, controlled, phase 3 trial. Lancet Oncol. 2019;20:1370–85.
3. Kato R, Obara W. Persisting challenges in the development of predictive biomarkers for immuno-oncology therapies for renal cell carcinoma. Expert Rev Anticancer Ther. 2025;25:97–103.
4. Wolchok JD, Neyns B, Linette G, Negrier S, Lutzky J, Thomas L, et al. Ipilimumab monotherapy in patients with pretreated advanced melanoma: a randomised, double-blind, multicentre, phase 2, dose-ranging study. Lancet Oncol. 2010;11:155–64.
5. Ascierto PA, Del Vecchio M, Robert C, Mackiewicz A, Chiarion-Sileni V, Arance A, et al. Ipilimumab 10 mg/kg versus ipilimumab 3 mg/kg in patients with unresectable or metastatic melanoma: a randomised, double-blind, multicentre, phase 3 trial. Lancet Oncol. 2017;18:611–22.
6. Motzer RJ, Rini BI, McDermott DF, Redman BG, Kuzel TM, Harrison MR, et al. Nivolumab for metastatic renal cell carcinoma: results of a randomized phase II trial. J Clin Oncol. 2015;33:1430–7.
7. Hammers HJ, Plimack ER, Infante JR, Rini BI, McDermott DF, Lewis LD, et al. Safety and efficacy of nivolumab in combination with ipilimumab in metastatic renal cell carcinoma: the CheckMate 016 study. J Clin Oncol. 2017;35:3851–8.
8. Leven C, Padelli M, Carré JL, Bellissant E, Misery L. Immune checkpoint inhibitors in melanoma: a review of pharmacokinetics and exposure–response relationships. Clin Pharmacokinet. 2019;58:1393–405.
9. Liu C, Yu J, Li H, Liu J, Xu Y, Song P, et al. Association of time-varying clearance of nivolumab with disease dynamics and its implications on exposure response analysis. Clin Pharmacol Ther. 2017;101:657–66.
10. Sanghavi K, Zhang J, Zhao X, Feng Y, Statkevich P, Sheng J, et al. Population pharmacokinetics of ipilimumab in combination with nivolumab in patients with advanced solid tumors. CPT Pharmacometrics Syst Pharmacol. 2020;9:29–39.
11. Bajaj G, Wang X, Agrawal S, Gupta M, Roy A, Feng Y. Model-based population pharmacokinetic analysis of nivolumab in patients with solid tumors. CPT Pharmacometrics Syst Pharmacol. 2017;6:58–66.
12. Maritaz C, Combarel D, Dalban C, Blondel L, Broutin S, Marabelle A, et al. Nivolumab plasma concentration and clearance associated with overall survival in patients with renal cell carcinoma. J Immunother Cancer. 2025;13:e010059.
13. Feng Y, Wang X, Bajaj G, Agrawal S, Bello A, Lestini B, et al. Nivolumab exposure-response analyses of efficacy and safety in previously treated squamous or nonsquamous non-small cell lung cancer. Clin Cancer Res. 2017;23:5394–405.
14. Bellesoeur A, Ollier E, Allard M, Hirsch L, Boudou-Rouquette P, Arrondeau J, et al. Is there an exposure-response relationship for nivolumab in real-world NSCLC patients? Cancers. 2019;11:1784.
15. Hurkmans DP, Basak EA, van Dijk T, Mercieca D, Schreurs MWJ, Wijkhuijs AJM, et al. A prospective cohort study on the pharmacokinetics of nivolumab in metastatic non-small cell lung cancer, melanoma, and renal cell cancer patients. J Immunother Cancer. 2019;7:192.
16. Basak EA, Koolen SLW, Hurkmans DP, Schreurs MWJ, Bins S, Oomen De Hoop E, et al. Correlation between nivolumab exposure and treatment outcomes in non–small-cell lung cancer. Eur J Cancer. 2019;109:12–20.
17. Wang X, Feng Y, Bajaj G, Gupta M, Agrawal S, Yang A, et al. Quantitative characterization of the exposure-response relationship for cancer immunotherapy: a case study of nivolumab in patients with advanced melanoma. CPT Pharmacometrics Syst Pharmacol. 2017;6:40–8.
18. Zhao Y, Tsujimoto A, Ide T, Zhang J, Feng Y, Gao L, et al. Model-based population pharmacokinetic and exposure response analyses for safety and efficacy of nivolumab as adjuvant treatment in subjects with resected oesophageal or gastroesophageal junction cancer. Br J Clin Pharmacol. 2024;90:2920–30.
19. Agrawal S, Feng Y, Roy A, Kollia G, Lestini B. Nivolumab dose selection: challenges, opportunities, and lessons learned for cancer immunotherapy. J Immunother Cancer. 2016;4:72.
20. Sangro B, Yau T, El-Khoueiry AB, Kudo M, Shen Y, Tschaika M, et al. Exposure-response analysis for nivolumab plus ipilimumab combination therapy in patients with advanced hepatocellular carcinoma (CheckMate 040). Clin Transl Sci. 2023;16:1445–57.
21. Koguchi Y, Iwamoto N, Shimada T, Chang SC, Cha J, Curti BD, et al. Trough levels of ipilimumab in serum as a potential biomarker of clinical outcomes for patients with advanced melanoma after treatment with ipilimumab. J Immunother Cancer. 2021;9:e002663.
22. Feng Y, Roy A, Masson E, Chen TT, Humphrey R, Weber JS. Exposure-response relationships of the efficacy and safety of ipilimumab in patients with advanced melanoma. Clin Cancer Res. 2013;19:3977–86.
23. Vano YA, Elaidi R, Bennamoun M, Chevreau C, Borchiellini D, Pannier D, et al. Nivolumab, nivolumab-ipilimumab, and VEGFR-tyrosine kinase inhibitors as first-line treatment for metastatic clear-cell renal cell carcinoma (BIONIKK): a biomarker-driven, open-label, non-comparative, randomised, phase 2 trial. Lancet Oncol. 2022;23:612–24.
24. Marin C, Khoudour N, Millet A, Lebert D, Bros P, Thomas F, et al. Cross-validation of a multiplex LC-MS/MS method for assaying mAbs plasma levels in patients with cancer: a GPCO-UNICANCER study. Pharmaceuticals. 2021;14:796.
25. Millet A, Khoudour N, Bros P, Lebert D, Picard G, Machon C, et al. Quantification of nivolumab in human plasma by LC-MS/HRMS and LC-MS/MS, comparison with ELISA. Talanta. 2021;224:121889.
26. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). Eur J Cancer. 2009;45:228–47.
27. Abe K, Shibata K, Naito T, Otsuka A, Karayama M, Maekawa M, et al. Impacts of cachexia progression in addition to serum IgG and blood lymphocytes on serum nivolumab in advanced cancer patients. Eur J Clin Pharmacol. 2022;78:77–87.
28. Aslan V, Kılıç ACK, Sütcüoğlu O, Eraslan E, Bayrak A, Öksüzoğlu B, et al. Cachexia index in predicting outcomes among patients receiving immune checkpoint inhibitor treatment for metastatic renal cell carcinoma. Urol Oncol. 2022;40:494.e1–494.e10.
29. Otten LS, Piet B, van den Haak D, Schouten RD, Schuurbiers M, Badrising SK, et al. Prognostic value of nivolumab clearance in non-small cell lung cancer patients for survival early in treatment. Clin Pharmacokinet. 2023;62:1749–54.
30. Gravis G, Marino P, Olive D, Penault-LLorca F, Delord JP, Simon C, et al. A non-inferiority randomized phase III trial of standard immunotherapy by checkpoint inhibitors vs. reduced dose intensity in responding patients with metastatic cancer: the MOIO protocol study. BMC Cancer. 2023;23:393.
31. Ratain MJ, Goldstein DA. Time is money: optimizing the scheduling of nivolumab. JCO. 2018;36:3074–6.
32. Puszkiel A, Bianconi G, Pasquiers B, Balakirouchenane D, Arrondeau J, Boudou-Rouquette P, et al. Extending the dosing intervals of nivolumab: model-based simulations in unselected cancer patients. Br J Cancer. 2024;130:1866–74.

33. Zhang J, Sanghavi K, Shen J, Zhao X, Feng Y, Statkevich P, et al. Population pharmacokinetics of nivolumab in combination with ipilimumab in patients with advanced malignancies. CPT Pharmacometrics Syst Pharmacol. 2019;8:962–70.
34. Kawakatsu S, Bruno R, Kågedal M, Li C, Girish S, Joshi A, et al. Confounding factors in exposure–response analyses and mitigation strategies for monoclonal antibodies in oncology. Br J Clin Pharmacol. 2021;87:2493–501.
35. Rahma OE, Reuss JE, Giobbie-Hurder A, Shoja E Razavi G, Abu-Shawer O, Mehra P, et al. Early 3+3 trial dose-escalation phase I clinical trial design and suitability for immune checkpoint inhibitors. Clin Cancer Res. 2021;27:485–91.
36. Wang X, Ludwig EA, Passarell J, Bello A, Roy A, Hruska MW. Population pharmacokinetics and exposure - safety analyses of nivolumab in patients with relapsed or refractory classical Hodgkin lymphoma. J Clin Pharmacol. 2019;59:364–73.
37. Patil VM, Noronha V, Menon N, Rai R, Bhattacharjee A, Singh A, et al. Low-dose immunotherapy in head and neck cancer: a randomized study. JCO. 2023;41:222–32.
38. https://www.ema.europa.eu/en/documents/rmp/yervoy-epar-risk-management-plan_en.pdf.
39. Kverneland AH, Enevold C, Donia M, Bastholt L, Svane IM, Nielsen CH. Development of anti-drug antibodies is associated with shortened survival in patients with metastatic melanoma treated with ipilimumab. OncoImmunology. 2018;7:e1424674.
40. Agrawal S, Statkevich P, Bajaj G, Feng Y, Saeger S, Desai DD, et al. Evaluation of immunogenicity of nivolumab monotherapy and its clinical relevance in patients with metastatic solid tumors. J Clin Pharmacol. 2017;57:394–400.
41. Galle P, Finn RS, Mitchell CR, Ndirangu K, Ramji Z, Redhead GS, et al. Treatment-emergent antidrug antibodies related to PD-1, PD-L1, or CTLA-4 inhibitors across tumor types: a systematic review. J Immunother Cancer. 2024;12:e008266.

ACKNOWLEDGEMENTS

The authors thank the patients, their families, and their caregivers for participating in this trial. We also thank Sylvie Decarpentries for her technical assistance to assay plasma concentrations of nivolumab and ipilimumab, as well as the level of anti-drug antibody.

AUTHOR CONTRIBUTIONS

YAV, WHF, CSF, and SO designed the study. YAV, SO and CSF supervised the study. BB, AP, AJ, MB, DM, DB, BL, DP, MGG, CC, PB, CT, EC, GG, EL, WHF, CSF, SO, CMS, YAV participated in the collection and assembly of data. AJ performed the statistical analysis. BB, AP, AJ, and YAV analyzed and interpreted the data. BB, AP, AJ, and YAV drafted the manuscript. All authors revised the work, approved its final version, and agreed to be accountable for all its aspects.

FUNDING

The princeps trial was funded by Bristol Myers Squibb (New-York, NY, USA) and ARTIC (Paris, France) (BIONIKK contract (R17169DD). Funders had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The ancillary study was supported by SIRIC CARPEM (INCa-DGOS-Inserm-ITMO Cancer_18006).

COMPETING INTERESTS

BB has received consulting and speaking fees (Esteve, Eisai, GSK, Pierre Fabre Oncologie and Pfizer), as well as support for travel and accommodation to attend meetings (Pierre Fabre). AP has received speaking fees (Eisai, Bristol Myers Squibb and Pierre Fabre Oncologie). MB has received consulting fees (MSD, Ipsen, Astellas, Janssen, and Merck). DM has received fees for consulting or advisory roles (BMS, Merck, Eisai, MSD Oncology, Pfizer, Ipsen, J&J, Astellas, AstraZeneca, Bayer, Novartis/3 A), as well as support for travel and accommodation to attend meetings (BMS, Johnson & Johnson, Bayer Novartis, Merck). BL has received consulting and speaking fees (Astellas, Pfizer, Ipsen, Bayer, Janssen-Cilag, Astellas, Eisai, BMS), as well as support for travel and accommodation to attend meetings (Ipsen, AstraZeneca, Pfizer, and MSD Oncology, BMS and Novartis). MGG has received consulting and speaking fees (BMS, MSD Oncology, Pfizer, Ipsen, Roche, Bayer/Onyx, Amgen, Sanofi, Janssen-Cilag, Astellas Medivation, AstraZeneca, and Gilead Sciences Inc.), as well as support for travel and accommodation to attend meetings (Bayer, Roche, Ipsen, AstraZeneca, Astellas Pharma, Pfizer, Janssen-Cilag, and MSD Oncology) and research funding (from Pfizer, MSD Oncology, Roche, Ipsen, Janssen-Cilag, AstraZeneca, Merck, and BMS) paid to her institution. PB has received consulting and speaking fees (BMS, Janssen-Cilag, Pfizer, Merck, Novartis/AdaCaP, MSD, IPSEN, Astra Zeneca, Astellas, EISAI, Gilead, BAYER), as well as support for travel and accommodation to attend meetings (AstraZeneca, Bayer, Ipsen, MSD, Merck and Pfizer). CT has received consulting and speaking fees (Pfizer, MSD, Servier, Bayer, Astra-Zeneca). GG has received consulting and speaking fees paid to her institution (BMS, Janssen, Pfizer, Alliance Merck-Pfizer, Novartis, MSD, IPSEN, Astra Zeneca, Astellas, EISAI, AMGEN, BAYER), as well as support for travel and accommodation to attend meetings (AstraZeneca, Bayer, Bristol Myers Squibb, Janssen, Ipsen, MSD, and Pfizer). GG has also received fees related to a data safety monitoring board or advisory board, paid to her institution (Alliance Merck-Pfizer, AstraZeneca, Bayer, Bristol Myers Squibb, Eisai, Ipsen, Janssen, Novartis and Pfizer). WHF has received fees as member of scientific boards (Anaveon, Geneta, IGI, Mestag, OSE immunotherapeutics and Oxford biotherapeutics). CSF has received consulting fees (Mestag). SO has received consulting and speaking fees (Amgen, Astellas, AstraZeneca, BMS, Bayer, Eisai, Ipsen, Janssen, MSD Oncology, Novartis, Pfizer, Roche, Sanofi), as well as support for travel and accommodation to attend meetings (Bayer, Ipsen, Astellas, Janssen, Pfizer, and MSD Oncology) and research funding (Bayer, BMS, AstraZeneca, Ipsen, Janssen and Pfizer) paid to the research team association named ARTIC. YAV has received honoraria (Bristol Myers Squibb, MSD, Merck, Ipsen, Pfizer, Roche and Eisai), as well as support for travel and accommodation to attend meetings (MSD, Merck, Pfizer, Ipsen and EISAI) and research grants (Bristol Myers Squibb and Ipsen). EC is an employee of AstraZeneca. EC has received consulting and speaking fees (BMS, Eisai, AstraZeneca, Pfizer, Janssen, Astellas, MSD, Novartis/Adacap). AJ, CC, DB, DP, EL and CMS declare no conflict of interest.

ETHICS APPROVAL AND CONSENT TO PARTICIPATE

The BIONIKK trial protocol and subsequent amendments were approved by the French Health authorities and ethics committee. The trial was conducted in accordance with Good Clinical Practice guidelines. All patients provided written informed consent.

CONSENT FOR PUBLICATION

All authors had full access to the data and control of the final approval and decision to submit the manuscript for publication.

ADDITIONAL INFORMATION

Supplementary information The online version contains supplementary material available at https://doi.org/10.1038/s41416-026-03340-1.

Correspondence and requests for materials should be addressed to Benoit Blanchet.

Reprints and permission information is available at http://www.nature.com/reprints

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.