Metformin Use Is Associated With Longer Progression-Free Survival of Patients With Diabetes and Pancreatic Neuroendocrine Tumors

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Metformin Use Associates With Longer Progression-free Survival of Patients With Diabetes and Pancreatic Neuroendocrine Tumors Receiving Everolimus and/or Somatostatin Analogues

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Abstract:

Background & Aims: Metformin seems to have anti-cancer effects. However, it is not clear whether use of glycemia and metformin affect outcomes of patients with advanced pancreatic neuroendocrine tumors (pNETs). We investigated the association between glycemia and progression-free survival (PFS) of patients with NETs treated with everolimus and/or somatostatin analogues, as well as the association between metformin use and PFS time.

Methods: We performed a retrospective analysis of 445 patients with advanced pNET treated at 24 medical centers in Italy, from 1999 through 2015. Data on levels of glycemia were collected at time of diagnosis of pNET, before treatment initiation, and during treatment with everolimus (with or without somatostatin analogues), octreotide, or lanreotide. Diabetes was defined as prior or current use of glycemia control medication and/or fasting plasma glucose ≥126 mg/dL, HbA1c ≥6.5% (48 mmol/L), or a random sample of plasma glucose ≥200 mg/dL (11.1 mmol/L), with reported classic symptoms of hyperglycemia or hyperglycemic crisis. Patients were assigned to groups based on diagnosis of diabetes before or during anti-tumor therapy. PFS was compared between patients with vs without diabetes. Among patients with diabetes, the association between metformin use and PFS was assessed. We performed sensitivity and landmark analyses, excluding patients who developed diabetes while receiving cancer treatment, and to exclude a potential immortal time bias related to metformin intake.

Results: PFS was significantly longer in patients with diabetes (median 32.0 months) than without diabetes (15.1 months) (hazard ratio for patients with vs without diabetes, 0.63; 95% CI, 0.50–0.80; *P*=.0002). PFS of patients treated with metformin was significantly longer (median PFS, 44.2 months) than for patients without diabetes (hazard ratio for survival of patients with diabetes receiving metformin vs without diabetes, 0.45; 95% CI, 0.32–0.62; *P*<.00001) and longer than for patients with diabetes receiving other treatments (median PFS, 20.8 months; hazard ratio, 0.49; 95% CI, 0.34–0.69; *P*<.0001). In multivariable analysis, adjusted for other factors associated with

outcomes, metformin was associated with longer PFS but level of glycemia was not. Metformin was associated with increased PFS of patients receiving somatostatin analogues and in those receiving everolimus, with or without somatostatin analogues. Sensitivity and landmark analyses produced similar results.

Conclusions: In a retrospective study of patients with pNETs, we found a significant association between metformin use and longer PFS.

KEY WORDS: pancreas, insulin resistance, drug, chemoprevention

Introduction

The incidence of pancreatic neuroendocrine tumors (pNETs) is increasing, and about 50% of patients present with advanced disease at diagnosis[1,2]. Although surgery is the only curative treatment for limited-stage disease[3], the 5-year survival rate is 32% for patients with advanced pNETs[4]. Therapeutic options include liver-directed therapies, chemotherapy, somatostatin analogs (SSAs), the mechanistic target of rapamycin (mTOR) inhibitor everolimus, the multikinase inhibitor sunitinib, and peptide receptor radiotherapy (PRRT)[5-8].

Although type II diabetes mellitus (T2DM) has emerged as a risk factor for the development of pNETs in some studies[9,10], its prognostic role in patients with advanced disease remains unexplored. Indeed, chronic elevation of glycemia may increase the risk of cancer by stimulating tumor anabolism, compensatory hyperinsulinemia, and cell proliferation through stimulation of the mTOR and mitogen-activated protein kinases (MAPK) pathways[11-13]. In many tumors, hyperglycemia and diabetes are associated with higher aggressiveness. In addition, DM is frequently present at diagnosis in advanced pNETs as a consequence of pancreatic involvement by the tumor, rare paraneoplastic syndromes (glucagonomas)[14] or, more often, of surgical[15] (partial or total pancreatectomy) or medical (SSAs or everolimus) treatments[5,8,16-18]. In particular, everolimus induces insulin resistance and hyperinsulinemia due to the combination of impaired insulin secretion and insulin resistance, whereas SSAs inhibit insulin secretion due to induced decrease in pancreatic beta-cell function[17,18].

Metformin, the most widely used drug in the treatment of T2DM, is emerging as a potentially active agent in cancer chemoprevention and treatment[19-23]. Its proposed antitumor mechanisms include the reduction of blood glucose, insulin, and IGF-1 levels as well as cell-autonomous anticancer effects mediated by the inhibition of mitochondrial oxidation, activation of adenosine monophosphate-activated kinase (AMPK), and inhibition of mTOR[20-24]. By reinforcing mTOR

inhibition and preventing activation of the IGF-1 oncogenic axis, metformin could synergize with everolimus and SSAs[25]. In a previous pilot experience, we investigated the prognosis of 31 patients with pNETs treated with everolimus and octreotide LAR; diabetic patients treated with metformin showed increased PFS compared with nondiabetic subjects and diabetics not on metformin[25]. However, the prognostic role of diabetes and metformin use has never been investigated in large populations of patients with advanced pNETs. We performed the multicenter Pancreatic Retrospective Italian MEtformin-NET (PRIME-NET) study to evaluate the association between glycemic status and outcome, measured in terms of PFS and overall survival (OS), in a large population of patients with advanced pNETs. Here we present our findings about the associations among glycemic status, metformin use, and PFS. Data on OS is not yet mature due to the low number of deaths occurring to date; it will be presented in a separate final report.

Patients and methods

Study setting

This was a multicenter, retrospective, independent study of 445 patients with advanced pNETs, treated between 1999 and 2015 at 24 Italian centers. The ethical committee of the coordinating center (Fondazione IRCCS Istituto Tumori Nazionale dei Tumori di Milano, Milan, Italy) approved the study design. All patients signed an informed consent for the use of their personal data for research purposes.

Patients ≥18 years were eligible if they had unresectable (locally advanced or metastatic), well-differentiated (Ki-67 <50%) pNET[26]. Other eligibility criteria were: (i) Eastern Cooperative Oncology Group (ECOG) performance status 0–3; (ii) evaluation of fasting glycemia and/or glycosylated hemoglobin (HBA1c) at diagnosis, before treatment initiation and during treatment; (iii) antitumor treatment with everolimus, everolimus plus SSA (octreotide or lanreotide), or SSA alone. Patients

were ineligible if they had a poorly differentiated neuroendocrine carcinoma or type 1 diabetes mellitus (T1DM).

Glycemic status was assessed at diagnosis, before treatment initiation, and during treatment by standard laboratory tests. There were no predefined time-points for the assessment of glycemia, except for baseline evaluations.

Diabetic patients were defined on the basis of either a documented diagnosis of T2DM before treatment initiation (basal diabetes), or the occurrence of diabetes during oncological therapy (on-treatment diabetes). We considered as diabetics those patients with a medical history of T2DM, previous or current use of antihyperglycemic medication, and, according to international guidelines, those who met one of the following criteria: fasting plasma glucose ≥126 mg/dL (7.0 mmol/L)[27], HbA1c ≥6.5% (48 mmol/L), or random plasma glucose ≥200 mg/dL (11.1 mmol/L) with reported classic symptoms of hyperglycemia or hyperglycemic crisis. Non-diabetics were those subjects who did not meet any of these criteria at any time during the study.

Objectives and design

The primary objective of this study was to investigate a possible association between diagnosis of T2DM and PFS (primary endpoint). Secondary objectives were to evaluate whether an association exists between: (i) diabetes and overall survival (OS, secondary endpoint); (ii) metformin therapy and clinical outcomes (PFS and OS) in diabetic patients; and (iii) diabetes, metformin use, and clinical outcomes (PFS and OS) in patients receiving everolimus and/or SSAs (subgroup analysis).

PFS was defined as the time between initiation of treatment with everolimus and/or SSAs and disease progression or death for any cause. OS was defined as the time between treatment initiation and death for any cause. Disease progression was defined according to RECIST 1.1 criteria, i.e. on the basis of measurement of tumor lesions, as detected through computed tomography (CT) or magnetic resonance (MRI). As a definition of disease progression, we also

considered those situations in which, even if the RECIST 1.1 criteria were not met, there was progressive deterioration of patient clinical conditions likely due to the disease (e.g. high disease burden in the liver).

PFS analysis was first performed in non-diabetic patients and in those with T2DM. Then T2DM patients were further divided according to their antidiabetic treatment, and PFS was separately analyzed in (i) diabetics on metformin (alone or combined with other antidiabetic therapies); (ii) diabetics on insulin or diet (i.e., not receiving metformin); and (iii) nondiabetic patients (Figure S1).

Statistical analysis

Patients' characteristics were analyzed by descriptive statistics. PFS was defined as time from treatment initiation to disease progression (assessed according to clinical practice at the time of diagnosis), death from any cause, last visit, or lost to follow-up. OS was defined as time from treatment initiation to death for any cause. Risk for disease progression and for overall mortality was compared using the Kaplan-Meier method.

Sample size was calculated *a priori*. To obtain a 90% statistical power, with a two-sided α error of 0.05, assuming that 60% of the subjects were diabetic and 40% were non-diabetic, 267 events (progression or deaths without progression) were needed, and at least 400 patients were to be included to detect a hazard ratio (HR) of progressive disease (PD) of 0.67 for diabetic versus nondiabetic patients. With these numbers, assuming that half of the diabetic patients had received metformin and half had not received metformin, 77% power was anticipated to detect HR 0.67 in each subgroup analysis. Data on OS will be available and the final analysis on survival will be performed when 267 deaths will have occurred.

Given the exploratory intent of the analysis, we did not plan hierarchical testing for multiple endpoints, or alpha error splitting, and we did not correct for multiple testing.

The log-rank test was used to compare the outcomes of different groups. To assess the clinical impact of the parameters under study along with the most relevant known prognostic factors in advanced pNETs (pathological tumor grading [G1–G2 vs. G3]; primary tumor resection; presence of liver, lymph node, and peritoneal metastases), multivariable analysis was performed, using the Cox regression model. Multivariable analysis was stratified by the anticancer treatment received, and an additional multivariable analysis was conducted considering only diabetic patients.

To exclude a relevant effect deriving from the time-on-treatment bias (i.e., the possibility that early interruption of everolimus or SSA therapy due to disease progression might result in lower patient exposure to these drugs and a consequently lower incidence of diabetes in poorly responding patients), we performed a sensitivity analysis, excluding patients who developed on-treatment diabetes from the diabetic group. We also performed a landmark analysis to exclude a potential immortal time bias related to metformin intake, that is, the possibility that patients taking metformin are those who most benefited from the treatment (everolimus plus/minus SSAs or SSAs) and consequently were more likely to develop treatment-related diabetes due to longer treatment exposure. In this landmark analysis, we included only patients without disease progression at 3 months after treatment initiation, thus excluding those patients who were less likely to initiate metformin due to early disease progression and treatment interruption. Patients included in the landmark analysis were then divided into two groups. Group 1 included patients who were taking metformin at 3 months (both those who were already on metformin before treatment initiation and those who started metformin within the first 3 months of therapy); group 2 included patients who were not taking metformin at 3 months (both those who never took metformin and those who started metformin later than 3 months after treatment initiation). In this analysis, patients starting metformin later (i.e., those with treatment-potential immortal time bias) were conservatively evaluated as patients who were not exposed to metformin.

Moreover, to evaluate the association between metformin intake and PFS, we considered for each patient the highest metformin dosage reported in medical records during the entire treatment period. We then defined two patient categories: (i) low dose: patients receiving metformin up to 1000 mg/day; (2) high dose: patients receiving a dose between 1000 and 3000 mg/day, and we compared PFS in the two categories with that of nondiabetic patients.

Given that the hyperinsulinemic status that frequently occurs in cancer patients with T2DM may contribute to their prognosis[12-13], and that the potential anticancer effects of metformin could be in part mediated by reduced plasma insulin levels and/or improved systemic insulin sensitivity[20], we aimed at comparing the prognosis of patients more likely to be hyperinsulinemic with the PFS of patients more likely to be hypoinsulinemic. To do so, in a further analysis we excluded patients who had undergone partial or total pancreatectomy, who were therefore more likely to have different grades of surgery-induced hypoinsulinism. Thereafter, we defined two patient populations: the former including patients taking everolimus alone (more likely to be hyperinsulinemic) and the latter including patients treated with SSAs alone (more likely to be hypoinsulinemic).

Lastly, since everolimus and SSAs can produce opposite effects on plasma insulin levels, we also separately assessed the impact of T2DM and metformin use in patients treated with only everolimus.

All statistical tests were two-tailed, and *P*-values <.05 were considered significant. Statistical analyses were performed using S-Plus (S-PLUS 6.0 Professional, release 1; Insightful Corporation, Seattle, WA, USA).

Results

Patient characteristics

Characteristics of patients with or without T2DM are summarized in Table 1. In total, 445 patients were evaluated (Table 1), 16 of whom had Multiple Endocrine Neoplasia type 1 (MEN-1) syndrome. Median age was 59 years (interquartile range [IQR] 49–69, range 10–89). Two hundred nine patients (47%) were nondiabetic, whereas 236 (53%) were diabetics, of whom 112 (25%) received metformin; the remaining 124 patients were treated with insulin (20%) or lifestyle recommendations, including diet and physical activity (8%). Among metformin-treated patients, 69 (62%) received metformin alone, 31 (28%) received metformin plus insulin, and 12 (11%) received metformin plus incretins. Among diabetic patients, 179 (76%) had basal T2DM, and 57 (24%) developed on-treatment diabetes (Tables S1, S2).

Overall, diabetic patients were slightly older (median age 60 vs. 57 years), more frequently male (59% vs. 47%), had a G3 tumor in a lower number of cases (3% vs. 9%), presented a higher BMI (24.4 vs. 23.0 kg/m^2), more frequently underwent primary tumor resection (61% vs. 49%), and had less frequent liver involvement at initiation of antitumor therapy (87% vs. 95%) (Table 1). Characteristics of patients with basal or on-treatment diabetes are reported in Table S3.

Among patients with T2DM, those receiving metformin were less likely to have liver (82% vs. 91%) and peritoneal (7% vs. 16%) metastases, and more likely to have lymph node metastases (56% vs. 43%) compared to diabetics not treated with metformin. (Table S4).

Comparison between diabetic and nondiabetic patients

In the overall population, median PFS was 23.4 months. Median PFS was 15.1 months in nondiabetic patients and 32.0 months in diabetic subjects, with an absolute difference of 16.9 months in favor of diabetic patients (Figure 1). The HR for progression in diabetic patients versus non-diabetic patients was 0.63 (95% CI: 0.50–0.80; *P* = .0002).

Comparison between diabetic patients receiving or not receiving metformin

Median PFS was 44.2 months in metformin-treated patients and 20.8 months in otherwise-treated diabetic patients (Figure 2). There was a 55% reduction in the risk of progression or death for metformin-treated patients compared with nondiabetic ones (HR 0.45; 95% CI: 0.32–0.62; *P* < .00001). Conversely, we did not find a significant difference in the risk of disease progression between diabetics not treated with metformin and nondiabetics (HR 0.86, 95% CI: 0.65–1.13; *P* = .26). The hazard ratio for diabetic patients treated with metformin versus diabetic patients not treated with metformin was 0.49 (95% CI: 0.34–0.69; *P* <.0001).

Comparison between diabetic patients receiving or not receiving metformin according to everolimus and/or SSAs treatment

The improved outcome associated with metformin was consistent across different subgroups of patients, stratified according to treatment: everolimus (with or without SSAs) or SSAs alone (Figure 3 and Table 2). Compared with nondiabetic status, diabetes was associated with improved outcome both in patients treated with everolimus with or without SSAs (HR 0.64, 95% CI: 0.47–0.87) and in those treated with SSAs alone (HR 0.57, 95% CI: 0.38–0.84) (*P* for interaction, 0.64). Moreover, compared with nondiabetic patients, the PFS of diabetic patients receiving metformin was longer both in everolimus with or without SSAs and SSA-treated patients: HR 0.45 (95% CI: 0.30–0.68) and 0.38 (95% CI: 0.21–0.67), respectively (*P* for interaction, 0.67). Conversely, we did not find any significant PFS difference between nondiabetic patients and diabetic patients not treated with metformin, both in everolimus with or without SSAs and SSA-treated patients: HR 0.89 (95% CI: 0.62–1.26) and 0.77 (95% CI: 0.49–1.20), respectively (*P* for interaction, 0.56).

Multivariable analysis

At multivariable analysis stratified by treatment, several known prognostic factors in advanced pNETs, such as tumor grading (G1/G2 vs. G3) and liver metastases, were confirmed to be prognostic (Table 3). Glycemic status was not associated with prognosis. Conversely, metformin

use was associated with improved prognosis after adjustment for other prognostic factors, with an HR for PFS of 0.53 (95% CI: 0.34–0.82; *P* = .004) in the overall population. The same finding was reported in a multivariate analysis of the subgroup of diabetic patients (HR 0.46; 95% CI: 0.29–0.72; *P* = .001) (Table S5).

Sensitivity and landmark analysis

After excluding patients with on-treatment diabetes, metformin use in diabetics remained associated with improved PFS compared with the cohort of nondiabetic patients (Figure S2 and Table S6). Moreover, the landmark analysis performed at 3 months showed longer survival in patients who started metformin before or within 3 months from treatment initiation compared with patients who never took metformin or who started it later, with PFS of 43.7 and 23.3 months, respectively (HR 0.64, 95% CI: 0.43–0.93, *P* = .02) (Figure S3 and Table S7).

Influence of metformin dosage

According to available data, 45 patients received low-dose metformin (median 1000 mg, interquartile range 850–1000), and 60 patients received high-dose metformin (median 2000 mg, interquartile range 1500–2000). At survival analysis, we found no evidence of a trend in PFS differences according to metformin dosage; indeed, median PFS was 45.9 months for patients receiving low metformin dosages and 36.1 months for patients receiving high dosages, which were both significantly longer than the median PFS of 15.7 months observed in nondiabetic patients (HR for low metformin group vs. nondiabetic, 0.44; 95% CI: 0.28–0.71, *P* <.001; HR for high metformin group vs. nondiabetic group, 0.70; 95% CI: 0.56–0.86, *P* = .001).

Effect of T2DM and metformin use in patients treated with everolimus alone.

Since everolimus and SSAs cause hyperglycemia through different mechanisms, we performed a separate analysis to investigate the potential impact of T2DM diagnosis and metformin use on the PFS of patients treated with everolimus alone (Table S8). Only 37 patients received everolimus

alone, which limited the statistical power of this analysis. We found no significant differences between the PFS of non-diabetic and diabetic patients (p=0.45), as well as between diabetics receiving or not receiving metformin and non-diabetic patients (p=0.1 and p=0.28, respectively).

Effect of metformin in patient subgroups with potentially different plasma insulinemic status

Overall, 16 patients treated with everolimus alone did not receive any pancreatic surgery; 12 of them did not receive metformin, and 4 were treated with metformin. Median PFS was 18.4 months in patients who did not receive metformin, and it was not reached in patients who received metformin (HR 0.26, 95% CI: 0.03–2.47).

Among patients treated with SSA alone, 61 did not receive any pancreatic surgery; of them, 46 did not receive metformin and 15 received it. Median PFS was 11.9 and 44.2 months in these subgroups, respectively (HR 0.46, 95% CI: 0.20–1.08).

Discussion

In this multicenter, retrospective study of 445 patients with advanced pNETs, we found that diabetes, either diagnosed before treatment initiation, or emerging during everolimus therapy with or without SSA, was associated with longer PFS regardless of the specific anticancer treatment received. Moreover, when stratifying diabetic patients according to antidiabetic treatment, those receiving metformin had longer PFS than nondiabetic ones, whereas no differences were observed between nondiabetic patients and those with T2DM treated only with insulin or diet modifications. The benefit associated with metformin was independent of the antitumor treatment. The results of multivariable analysis represent in our opinion the most relevant finding of the study, because they suggest that it is metformin use—rather than glycemic status—that is associated with an improved prognosis in advanced pNETs patients.

Given that everolimus, SSAs, or both can induce diabetes, the observed improved prognosis in the group of diabetic patients could simply reflect longer exposure to an effective anticancer treatment (immortal time bias). However, the sensitivity analysis that we performed by eliminating patients who developed on-treatment diabetes seems to exclude this possibility and reinforces the conclusion that metformin use correlates with improved patient prognosis. These findings are further strengthened by the landmark analysis, which showed longer survival in patients who started metformin before or within 3 months from treatment initiation than patients who never took metformin or who started it later.

Our findings are consistent with recent retrospective evidence, including a meta-analysis of 20 retrospective studies that showed a 38% reduced risk of death in metformin-receiving cancer patients with T2DM[28]. Several prospective studies are testing the efficacy of metformin in combination with standard treatments in many solid cancers. These studies are investigating metformin also in nondiabetic patients, who represent the majority of cancer patients.

To date, only three prospective randomized studies of patients with unresectable pancreatic exocrine tumors have been published[29-31]. However, these studies failed to demonstrate any advantage from combining metformin with standard chemotherapy treatments.

This discrepancy may stem from the following factors. (i) In retrospective studies, metformin is taken only by those patients diagnosed with diabetes. For various reasons, including specific metabolic or tumor biology profiles, these patients could benefit from metformin, although nondiabetic ones could not. Because in prospective studies metformin is given to both diabetic and nondiabetic patients, it could prove ineffective to improve prognosis in the overall population. (ii) Many patients included in retrospective studies started metformin several months, or even years, before tumor diagnosis and treatment. Because metformin could affect the tumorigenesis process by altering systemic metabolism or proliferation of tumor precursor cells, malignancies

evolving under chronic metformin exposure may display less aggressive behavior. This could result in a clinical advantage for patients under metformin treatment in retrospective studies, whereas this compound could be ineffective when given to patients at the initiation of oncological treatment. (iii) Retrospective studies are subject to poor reporting bias, which can affect the assessment of diabetes duration, the use and dosage of antidiabetic drugs, or both.

In this study, we did not disclose any significant association between metformin dose and PFS. However, this analysis presents major limitations. First, patients on metformin had received this drug at any time during their clinical history; therefore, there was no predefined time point for the assessment of metformin dose during the course of diabetes. Second, given that treatment for hyperglycemia can change over time, we cannot rule out that metformin dosage has been frequently changed in evaluated patients on the basis of diabetes control, emerging comorbidities, or need for the association of other antidiabetic drugs during the course of disease. Therefore, we believe that, due to the prolonged PFS reported in many patients, there could have been considerable variation in metformin dosage, and the highest dosage may not well reflect global exposure to metformin during the treatment period. Moreover, the reported metformin dosage for individual patients may not necessarily mirror the average exposure dosage over months or years of diabetes management. Therefore, we believe that the absence of a dose–effect relationship regarding metformin use cannot be considered definitive. Prospective trials with detailed information about metformin dosage and its changes during the treatment are, however, required to investigate this major issue.

It is still unclear whether potential metformin anticancer effects are mediated by changes in systemic metabolism (blood glycemia and insulinemia), through cell-autonomous anticancer effects, or through a combination of both[17,18]. Our finding that the glycemic status was not

associated with patient outcome regardless of metformin use suggests that the role of metformin in reducing glycemia is likely poorly relevant in patients with advanced pNETs.

With respect to insulinemia, existing evidence on the potential oncogenic role of insulin suggests that even physiological concentrations of insulin could stimulate cancer growth[32]. In our study, measurements of blood insulin concentration were not available; therefore, we could not conclude that patients under insulin therapy actually had higher blood insulin concentrations than nondiabetic ones. Nevertheless, the fact that diabetic patients receiving insulin did not have reduced PFS suggests that insulin therapy is not associated with a worse clinical outcome, as confirmed also by the multivariable analysis.

Based on the lack of an association between blood glucose levels and insulin intake with patient prognosis, we believe that metformin might be associated with an improved prognosis in patients with pNET by displaying direct, cell-autonomous anticancer effects. Furthermore, metformin was associated with longer PFS in both patients treated with everolimus, which is known to reduce peripheral tissue sensitivity to insulin and to cause hyperinsulinemia, and in those receiving SSAs, which can reduce blood insulin levels[17,18]. This finding may further support the notion that mechanisms other than the reduction of circulating insulin levels might contribute to the prolonged PFS in patients on metformin. Another argument in favor of this hypothesis consists in results of our subgroup analysis, which, although performed in a small number of patients, suggests that metformin-associated effects do not seem mediated by the reduction of blood glucose concentrations, whereas cell-autonomous, antitumor effects could be more prominent.

Given pNET dependence on the IGF1R–PI3K-AKT-mTOR axis, the biological rationale for combining metformin with everolimus (i.e., strengthening of mTOR pathway inhibition through the AMPK-TSC1/2-mTOR axis) or SSAs (through synergistic inhibition of the IGF-1 receptor/PI3K/AKT/mTOR pathways) may exist (Figure 4)[25].

Conclusions

With all the limitations of retrospective studies, our results showed, for the first time, that in a population of patients with advanced pNETs treated with everolimus, SSAs, or both, diabetic subjects receiving metformin had statistically and clinically meaningful prolonged PFS compared with both nondiabetic patients and diabetics treated with insulin or diet.

Although causal relationships cannot be retrieved at the moment, these findings suggest that metformin could have some antitumor effects in the treatment of patients with advanced pNETs. Based on our results, two prospective, pilot, and phase II studies are currently ongoing at the Istituto Nazionale Tumori (Milan, Italy) to assess metformin in combination with both SSAs and everolimus in the treatment of advanced pNETs (MetNET-1 trial, NCT 02294006) and in combination with SSAs in lung and small bowel NETs (MetNET-2 trial, NCT02823691).

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Figure legends

Figure 1. Kaplan-Meier plot of PFS between patients with type 2 diabetes mellitus and nondiabetic patients

Figure 2. Kaplan-Meier plot of PFS among nondiabetic patients, diabetics treated with metformin, and diabetics not receiving metformin but treated with insulin or diet

Figure 3. Forest plot showing the effect of glycemia on PFS in patient subgroups according to the

oncological treatment administered.

Figure 4. Interplay between IGF-1/IGF1R/PI3K/Akt/mTOR and AMPK pathways. Potential synergistic activity among somatostatin analogs, everolimus, and metformin may derive from inhibition of the IGF-1/IGF1R/PI3K/Akt/mTOR axis at different levels of the cascade. Modified by Pusceddu S et al. Fut Oncol 2016; 12:1251–1260[25].

Figure S1. PRIME-NET study design

* Well-differentiated pNETs = well-differentiated pancreatic neuroendocrine tumors with Ki-67 <50% according to 2017 pNET World Health Organization (WHO) classification

** Type 2 diabetes = patients with a level of fasting plasma glucose ≥126 mg/dL (7.0 mmol/L) or HbA1c ≥6.5% (48 mmol/L) or random plasma glucose ≥200 mg/dL (11.1 mmol/L) with reported classic symptoms of hyperglycemia or hyperglycemic crisis; EVE = everolimus; SSA = somatostatin analogs; GEP-NETs = gastro-entero-pancreatic neuroendocrine tumors

Figure S2. Kaplan-Meier plot of PFS among metformin recipients, patients on insulin or diet, and nondiabetic subjects, with the exclusion of patients developing on-treatment diabetes (sensitivity analysis)

Figure S3. Kaplan-Meier plot of PFS among patients receiving metformin vs. patients not receiving metformin three months after treatment initiation (landmark analysis)