Amylin in the insulin resistance of patients with rheumatoid arthritis

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Abstract

Objective

Amylin, which is co-secreted with insulin, plays a role in glycemic regulation and is impaired in type 2 diabetes. In the present study we assess, for first time, the implication of amylin in the development of insulin resistance (IR) in rheumatoid arthritis (RA).

Methods

This was a cross-sectional study involving 361 non-diabetic individuals, 151 patients with RA and 210 sex-matched controls. Insulin, C-peptide, amylin, lipoprotein serum concentrations, and IR indexes by homeostatic model assessment (HOMA2) were evaluated in patients and controls. A multivariable analysis, adjusted for IR-related factors, was performed to determine the differences between patients and controls vis-à-vis amylin and how it is related to IR in RA.

Results

Insulin, C-peptide and HOMA2-IR indexes were higher in RA patients than in controls. Amylin serum levels were found to be upregulated in RA patients compared to controls (1.36 ± 0.81 vs. 1.79 ± 1.51 ng/ml, p=0.011), although this difference was lost after adjusting for covariates (p=0.46). While amylin positively correlated with the presence of rheumatoid factor (beta coef. 0.90 [95%CI -0.23–1.56], p=0.009) and SDAI (beta coef 0.01 [95%CI 0.00–0.03], p=0.034), no significant association with other disease activity scores, glucocorticoid intake, methotrexate use or TNF-alpha inhibitors was found.

Conclusion

IR in RA does not appear to be mediated by amylin. This would imply that the mechanisms associated with IR in RA patients differ from those at work in type 2 diabetes.

Key words

amylin, insulin resistance, rheumatoid arthritis

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Introduction

Amylin, or islet amyloid polypeptide, is a 37-residue peptide hormone co-secreted with insulin from pancreatic β-cells at a ratio of approximately 10:1. Amylin plays a role in glycemic regulation by slowing gastric emptying and promoting satiety, thereby preventing post-prandial spikes in blood glucose levels (1).

Amylin is deficient in type 1 diabetes and relatively deficient in insulin-requiring type 2 diabetes. A greater demand for insulin production, resulting in the simultaneous secretion of proinsulin and proamylin, seems to occur in patients with type 2 diabetes. Interestingly, proamylin has been found to be less toxic than amylin in amyloid accumulation (2). However, in patients with type 2 diabetes, the enzymes responsible for converting these precursors into insulin and amylin cannot keep pace with the high secretion levels of these molecules, ultimately leading to the accumulation of amylin (3). For this reason, it is thought that impaired amylin is an important factor initiating amyloid formation and β-cell death in the pancreas. This is supported by the fact that elevated levels of amylin have been observed in obese adults with impaired glucose tolerance, in patients with type 2 diabetes (4), and in women with polycystic ovary syndrome (5). Recent studies have highlighted the link between amylin and inflammatory markers and the presence of metabolic syndrome (6). However, this amylin-metabolic syndrome association has also been found to occur independently of established risk factors of metabolic syndrome, including obesity, inflammatory markers and insulin resistance. It is still unclear whether amylin plays a causative role in type 2 diabetes and insulin resistance (IR), or if it is merely present in increased amounts due to a defect in insulin secretion.

IR, a condition involving resistance to insulin-mediated glucose uptake by cells, is central to the clustering of multiple metabolic abnormalities and clinical syndromes (7). Several studies have shown an increased prevalence of IR in patients with rheumatoid arthritis (RA) (8-10), which may reflect the level of RA disease activity present (11). It is thought that RA's low-grade inflammation state might contribute to its development (12). This is supported by the fact that in RA patients, IR has also been found to directly correlate with levels of interleukin 6, tumour necrosis factor-(TNF-) alpha, and C-reactive protein (13). In addition, TNF-alpha inhibitor therapies have been shown to improve insulin sensitivity and to reduce IR in RA (14). Diabetes mellitus has also been shown to be a predictor of mortality in RA patients (15). The precise mechanism that links systemic inflammation and IR in RA patients remains unknown.

The aim of this study was two-fold: to investigate how amylin is expressed in patients with RA and to study its potential role in the IR observed in these patients compared to controls.

Materials and methods

Study participants

This was a cross-sectional study that included 361 non-diabetic individuals, 151 patients with RA and 210 sex-matched controls. All RA patients were at least 18 years old and fulfilled the 2010 ACR/EULAR diagnostic criteria (16). They had been diagnosed by rheumatologists and were periodically followed-up at rheumatology outpatient clinics. For the purpose of inclusion in the present study, RA disease duration was required to be ≥1 year. Although anti-TNF-alpha treatment has been associated with changes in insulin resistance (8, 17-19), RA patients undergoing TNF-alpha antagonist therapy were not excluded from the present study. The control group consisted of patients recruited from the Spanish Camargo Cohort (20, 21). This cohort was set up between February 2006 and February 2011, and individuals included therein have been followed ever since. The aim was to evaluate the prevalence and incidence of metabolic bone diseases and mineral metabolism disorders. Controls included in the current study were sex-matched subjects without diabetes. Patients and controls with diabetes mellitus were not included in the study. Therefore, none of the patients or controls was receiv-

ing glucose-lowering drugs or insulin therapy. All patients and controls had a glycaemia <7 mmol/l. Patients and controls were excluded if they had a history of myocardial infarction, angina, stroke, a glomerular filtration rate <60 mL/min/1.73m^2, a history of cancer, or any other chronic disease or evidence of infection. None of the controls was receiving glucocorticoids. However, since prednisone is often used in the management of RA, patients taking this drug, or an equivalent dose ≤10 mg/day, were not excluded. The study protocol was approved by the institutional review committees at Hospital Universitario de Canarias and Hospital Universitario Marqués de Valdecilla (both in Spain), and all subjects provided written informed consent.

Data collection

Surveys in RA patients and controls were performed in the same way. Subjects completed a cardiovascular risk factor and medication use questionnaire and underwent a physical examination to determine their anthropometrics and blood pressure. Medical records were reviewed to ascertain specific diagnoses and medications. Waist circumference was measured at the smallest circumference between the rib cage and the iliac crest while the subject was in a standing position. Hip circumference was measured at the widest circumference between the waist and thighs. The waist-to-hip ratio also was estimated. Hypertension was defined as a systolic or a diastolic blood pressure higher than, respectively, 140 and 90 mmHg. Dyslipidaemia was defined if one of the following metrics was present: total cholesterol >200 mg/dl, triglyceride >150 mg/dl, HDL cholesterol <40 in men or <50 mg/dl in women, or LDL cholesterol >130 mg/dl. In patients with RA, disease activity was measured using the Disease Activity Score (DAS28) in 28 joints (22), while disease disability was determined using the Health Assessment Questionnaire (HAQ) (23). Clinical Disease Activity Index (CDAI) (24) and Simple Disease Activity Index (SDAI) (25) scores for RA disease activity were also performed as previously described.

Assessments

The homeostatic model assessment (HOMA) method was performed to determine IR; specifically, in this study we used HOMA2: the updated-computer HOMA model (26, 27). Briefly, this method consists of a structural computer model of the glucose-insulin feedback system in a homeostatic (overnight fasting) state. The model is comprised of a number of nonlinear empirical equations (and precludes an exact algebraic solution), which describe the functions of organs and tissues involved in glucose regulation. This model can be used to determine insulin sensitivity (%S) and β-cell function (%β) from paired fasting plasma glucose and specific insulin, or C-peptide concentrations across a range of 1–2,200 pmol/l for insulin and 1–25 mmol/l for glucose. In our study, we have used both C-peptide and insulin to calculate HOMA2 because there is a logic for using C-peptide data to calculate beta cell function (since C-peptide is a marker of secretion) and for using insulin data to calculate %S (since HOMA-%S is derived from glucose disposal as a function of insulin concentration). This computer model provides an insulin sensitivity value expressed as HOMA2-%S (where 100% is normal). HOMA2-IR (insulin resistance index) is simply the reciprocal of %S.

Insulin (Architect Abbott, 2000I) and C-peptide (Immulite 2000, Siemens) were determined by chemiluminescent immunometric assays. Amylin was assessed via an enzyme-linked immunosorbent assay (ELISA) (Phoenix Pharmaceuticals, California, USA). The assay sensitivity (minimum detectable concentration) for amylin was 0.58 ng/ml. This ELISA does not cross-react with human insulin, glucagon or proinsulin. Precision was estimated as inter-assay <15%, intra-assay <10%. Standard techniques were used to measure plasma glucose, C-reactive protein (CRP), Westergren erythrocyte sedimentation rate (ESR), and serum lipids.

Statistical analysis

Demographic and clinical characteristics were compared between patients with RA and controls using chi-square tests for categorical variables or a T-Student test for continuous variables (data expressed as mean ± standard deviation). For non-continuous variables, either a Mann-Whitney U-test was performed or a logarithmic transformation was made and data were expressed as a median (interquartile range). Differences in glucose homeostasis metabolism molecules and HOMA indexes were studied through three different multivariable regression models: 1) a univariate unadjusted model; 2) a model adjusted for those variable with a *p*-value lower than 0.20 in the differences between patients and controls (age, sex, waist circumference, dyslipidaemia, statins, antihypertensive treatment, and CRP and cholesterol levels); and 3) a model adding glucocorticoid intake as a binary variable and adjusted for the same previously described variables. Univariate association analysis of the relation of several inflammation markers and disease-related data with glucose, insulin, C-peptide, amylin and HOMA indexes was assessed through lineal regression. For all analyses, we used a 5% two-sided significance level, and all analyses were performed using IBM SPSS Statistics v. 21 software (IBM, Armonk, NY, USA) and Stata v. 13/SE software (StataCorp, College Station, TX, USA). A *p*-value <0.05 was considered statistically significant.

Results

Demographic, laboratory and disease-related data

A total of 361 participants, 151 RA patients and 210 controls, with a mean (± standard deviation) age of 53±11 and 58±9 years, respectively, were included in this study. The demographic and disease-related characteristics of the participants are shown in Table I. Although there were no differences between patients and controls with regard to BMI, waist circumference was found to be higher in patients than in controls (92±14 *vs*. 96±13, *p*=0.015). Although the presence of hypertension did not differ between controls and patients, dyslipidaemia was more frequently noted in RA patients. In this sense, RA patients generally displayed lower levels of lipid metabolism molecules. In

Table I. Demographics, laboratory data and disease-related characteristics of RA patients and controls.

	Controls (n=210)	AR (n=151)	*p*
Age, years	58 ± 9	53 ± 11	**0.000**
Female, n (%)	148 (70)	120 (79)	0.052
Body mass index, kg/m^2	28 ± 5	28 ± 5	0.78
Waist circumference, cm	92 ± 14	96 ± 13	**0.015**
Cardiovascular risk factors			
Smoking, n (%)	42 (20)	26 (17)	0.51
Hypertension, n (%)	58 (27)	45 (30)	0.65
Dyslipidaemia, n (%)	33 (15)	54 (36)	**0.000**
Diabetes, n (%)	0 (0)	0 (0)	-
Medication			
Statins	14 (6)	43 (28)	**0.000**
Antihypertensive treatment	35 (16)	45 (30)	**0.003**
Laboratory data			
CRP, mg/dL	1.0 (1.0-3.0)	3.2 (1.5-5.8)	**0.000**
ESR, mm/hour	10 ± 8	34 ± 22	**0.000**
Triglycerides, mg/dL	104 ± 50	144 ± 92	**0.000**
HDL-C, mg/dL	63 ± 18	56 ± 15	**0.000**
LDL-C, mg/dL	135 ± 34	121 ± 31	**0.000**
Total Cholesterol, mg/dL	219 ± 37	206 ± 36	**0.001**
Lipoprotein A, mg/dL	16 (9-39)	35 (11-121)	**0.000**
Apolipoprotein A1, mg/dL	191 ± 35	170 ± 28	**0.000**
Apolipoprotein B, mg/dL	102 ± 24	110 ± 63	0.11
ApoB:ApoA ratio	0.55 ± 0.16	0.66 ± 0.30	**0.000**
Atherogenic index	3.7 ± 4.0	4.0 ± 3.6	0.10
Disease-related data			
Disease duration, years		6.6 (3.3-13.9)	
ACPA, n (%)		89 (59)	
Rheumatoid factor, n (%)		109 (72)	
Erosions, n (%)		55 (36)	
Extrarticular manifestations, n (%)		16 (11)	
DAS 28-ESR		3.7 ± 1.2	
DAS 28-CRP		2.9 ± 1.0	
SDAI		13 (8-21)	
CDAI		81 (33-108)	
HAQ		0.630 (0.380-1.130)	
Current prednisone, n (%)		57 (38)	
Prednisone current doses, mg/day		5 (5-6)	
NSAIDs. n (%)		69 (46)	
DMARDs, n (%)		129 (85)	
Methotrexate, n (%)		113 (75)	
Biologic drugs, n (%)		35 (23)	
Anti-TNF alpha drugs, n (%)		20 (13)	

Data expressed as mean (± standard deviation) or median (interquartile range). Dichotomous variables are expressed as n and percentage. DAS28: Disease Activity Score. ACPA: Anti-citrullinated peptide/protein antibody; DMARDs: Disease-modifying Antirheumatic Drug; ESR: erythrocyte sedimentation rate; CRP: C reactive protein; HAQ: Health Assessment Questionnaire; SDAI: Simple Disease Activity Index; CDAI: Clinical Disease Activity Index; HDL-C: high-density cholesterol lipoprotein; LDL-C: low-density cholesterol lipoprotein; NSAIDs: non-steroidal anti-inflammatory drugs; TNF: tumour necrosis factor.

contrast, triglycerides, lipoprotein A and apolipoprotein B were found to be higher in RA patients (Table I).

As expected, the assessment of ESR and CRP revealed statistically significant, higher levels in RA patients than controls. Patients in our series had moderate-active disease as shown by DAS28 (3.7±1.2) and fifty (38%) were on prednisone (an average dose of 5 interquartile range -IQR- 5–6 mg/day). Disease duration was 6.6 (IQR 3.3–13.9) years, and 59% and 72% were respectively positive for the anti-citrullinated protein antibodies and rheumatoid factor. In addition, while 85% of the patients were on disease-modifying antirheumatic drugs, 13% were on anti TNF-alpha treatment and 23% were receiving other biologic therapies.

Differences in carbohydrate metabolism molecules and insulin resistance indexes between RA patients and controls

HOMA2-IR indexes, whether calculated with insulin or C peptide, were different between patients and controls (Table II). In this sense, HOMA2-S% was lower in RA patients than controls after adjusting for traditional IR-related factors and prednisone intake (105±53 *vs*. 108±75, *p*=0.006). Similarly, HOMA2-IR was found to be higher in RA patients after multivariable analysis (1.65±1.69 *vs*. 1.27±0.82, *p*=0.054). In contrast, HOMA2-%B was higher in the univariate analysis. However, this difference was lost after adjusting for covariates (*p*=0.14).

When HOMA2 indexes were calculated using C peptide, the differences between patients with RA and controls were stronger. In this regard, all comparisons showed higher HOMA2-IR and HOMA2-%B indexes and lower HOMA2-%S in RA patients even after multivariate analysis (Table II).

Whereas glucose serum levels did not achieve statistically significant differences between controls and patients, insulin (9.8±6.5 *vs*. 13.0±13.4 U/ml, *p*=0.007) and C peptide serum levels (1.53±0.77 *vs*. 3.37±2.94 ng/ml, *p*=0.000) were found to be upregulated in RA patients. These differences were still present after multivariate adjustment, including glucocorticoid intake.

Amylin serum levels were higher in patients with RA than in controls in the univariate analysis (1.79±1.51 *vs*. 1.36±0.81, *p*=0.011). However, in the multivariate regression analysis, which included glucocorticoid intake, the association with RA was lost (Table II). Similarly, an amylin/insulin ratio comparison between patients and controls was out of the range of significance.

Univariate relation of inflammatory markers, disease-related data, and disease activity scores with amylin

The associations between laboratory markers, disease activity and therapy with amylin are shown in Table III. CRP

Table II. Differences in carbohydrate metabolism molecules, including amylin, and insulin resistance indices between RA patients and controls.

	Controls (n=210)	Rheumatoid arthritis (n=151)	Univariate model *p*	Adjusted model *p*	Adjusted model + GC *p*
Glucose, mg/dL	90 ± 10	88 ± 19	0.21	0.37	0.71
Insulin, U/mL	9.8 ± 6.5	13.0 ± 13.4	**0.007**	**0.037**	**0.046**
C-peptide, ng/mL	1.53 ± 0.77	3.37 ± 2.94	**0.000**	**0.000**	**0.000**
Amylin, ng/mL	1.36 ± 8.81	1.79 ± 1.51	**0.011**	0.96	0.46
Amylin:insulin ratio	0.18 ± 0.14	0.24 ± 0.27	0.051	0.39	0.09
HOMA2 insulin					
HOMA2-IR	1.27 ± 0.82	1.65 ± 1.69	**0.011**	**0.036**	0.054
HOMA2-%S	108 ± 75	105 ± 53	0.65	**0.019**	**0.006**
HOMA2-%B	111 ± 45	134 ± 69	**0.000**	0.33	0.14
HOMA2 C-peptide					
HOMA2-IR	1.13 ± 0.58	2.49 ± 2.35	**0.000**	**0.000**	**0.000**
HOMA2-%S	115 ± 75	67 ± 41	**0.000**	**0.000**	**0.000**
HOMA2-%B	104 ± 36	180 ± 82	**0.000**	**0.000**	**0.000**

HOMA: Homeostasis Model Assessment, IR: insulin resistance; %S insulin sensitivity; %B: beta cell production; GC: glucocorticoids.
Adjusted model for age, sex, waist circumference, dyslipidaemia, statins, anti-hypertensive treatment, and CRP and cholesterol levels. Analytical data represent the unadjusted values.

tended to be associated with amylin (beta coef. 0.01 [95%CI -0.00-0.01], *p*=0.07). DAS28 was not related with amylin serum levels (this association was not found either using categorised DAS28). No statistically significant association of ACPA or rheumatoid factor with amylin was found. Amylin was associated with SDAI (beta coef. 0.01 [95%CI 0.00-0.03], *p*=0.034) but not with other disease activity scores.

Glucocorticoids intake was positively associated with insulin (beta coef. 2.56 [95%CI -0.31-5.42], *p*=0.080), C-peptide (beta coef. 1.69 [95%CI 1.09-2.28], *p*=0.000), HOMA-IR (beta coef. 0.34 [95%CI -0.02-0.70], *p*=0.067] and HOMA2-%B-C peptide (beta coef. 51 [95%CI 31-70], *p*=0.000). However, this relation was not found with amylin. Methotrexate, TNF-alpha inhibitors, and others non-TNF biologic agents use were not related to glucose homeostasis molecules including amylin. Furthermore, the group of DMARDs including methotrexate, and the whole group of biologic therapies, did not disclose relation with amylin.

Other data showing the relations of inflammatory markers and disease activity scores with insulin, C-peptide serum levels and HOMA indexes are shown in Table III.

Discussion

In the present study we assessed, for the first time, amylin in a large series of patients with RA. Amylin was not related to disease characteristics. Our data indicate that the mechanisms underlying IR in RA may be different from those involved in type 2 diabetes, where amylin plays a central role.

Our data regarding IR levels in RA are in agreement with previous studies. In this regard, higher insulin, C peptide serum levels and HOMA2 indexes have been described in patients with RA (9, 10, 14). In fact, the degree to which the levels of these molecules and indexes are abnormal is proportional to the severity of the inflammatory burden; this holds true for other inflammatory chronic diseases as well (28). Our data concerning cholesterol levels reduction in patients with RA is also in agreement with previous knowledge regarding differences in lipid profile in these patients (29).

Previous studies assessing insulin sensitivity and β-cell function in RA have relied predominantly on fasting parameters, such as HOMA. Although these model-derived indexes have been well validated, they do not account for the

Table III. Inflammatory markers, disease-related data, and disease activity scores: univariate relation with insulin and C-peptide serum levels, HOMA indices and amylin.

	beta coefficient (95% confidence interval), p				
	Insulin, U/mL	C-peptide, ng/mL	Amylin, ng/mL	HOMA2-IR insulin	HOMA2-%B C peptide
CRP, mg/dL	0.02 (-0.05-0.08), 0.61	0.01 (-0.02-0.03), 0.09	0.01 (-0.00-0.01), 0.07	0.00 (-0.01-0.01), 0.61	**0.54 (0.16-0.96), 0.013**
ESR, mm/hour	-0.01 (-0.10-0.09), 0.88	0.00 (-0.02-0.03), 0.67	-0.00 (-0.02-0.01), 0.76	-0.00 (-0.01-0.01), 0.90	0.08 (-0.50-0.66), 0.79
Disease duration, years	-0.03 (-0.27-0.20), 0.78	0.01 (-0.05-0.06), 0.82	0.02 (-0.02-0.05), 0.37	-0.00 (-0.03-0.03), 0.82	-0.51 (-1.95-0.94), 0.49
ACPA	**-4.57 (-9.09--0.04), 0.048**	**-1.11 (-2.10--0.11), 0.029**	0.11 (-0.54-0.76), 0.73	-0.57 (-1.14-0.01), 0.053	**-32.23 (-60.02--4.44), 0.023**
Rheumatoid factor	-1.41 (-6.45-3.63), 0.58	-0.10 (-1.21-1.01), 0.85	**0.90 (-0.23-1.56), 0.009**	-0.13 (-0.77-0.50), 0.68	-18.37 (-49.31-12.57), 0.24
DAS 28-ESR	-0.49 (-2.32-1.35), 0.60	-0.01 (-0.41-0.40), 0.98	-0.02 (-0.30-0.27), 0.91	-0.06 (-0.29-0.18), 0.64	-0.79 (-12.11-10.53), 0.89
DAS 28-CRP	-0.22 (-2.43-2.00), 0.85	0.08 (-0.41-0.57), 0.75	0.06 (-0.28-0.40), 0.72	-0.02 (-0.30-0.26), 0.88	3.33 (-10.31-16.97), 0.63
SDAI	-0.01 (-0.11-0.10), 0.92	0.00 (-0.02-0.03), 0.72	**0.01 (0.00-0.03), 0.034**	-0.00 (-0.01-0.01), 0.91	0.34 (-0.32-0.99), 0.31
CDAI	0.01 (-0.04-0.05), 0.73	0.00 (-0.01-0.01), 0.41	0.00 (-0.01-0.01), 0.90	0.00 (-0.01-0.01), 0.72	0.12 (-0.16-0.39), 0.41
Glucocorticoids intake	2.56 (-0.31-5.42), 0.080	**1.69 (1.09-2.28), 0.000**	0.11 (-0.30-0.52), 0.59	0.34 (-0.02-0.70), 0.067	**51 (31-70), 0.000**
Methotrexate	-9.9 (-44-25), 0.57	-0.11 (-0.47-0.25), 0.55	0.59 (-0.14-1.31), 0.11	-0.19 (-0.82-0.44), 0.55	-5 (-36-25), 0.73
Anti-TNF-alpha drugs	28 (-16-72), 0.21	0.21 (-0.26-0.67), 0.39	0.10 (-0.87-1.06), 0.84	0.54 (-0.27-1.34), 0.19	-12 (-51-27), 0.55

ACPA: Anti-citrullinated peptide / protein antibody; TNF: tumour necrosis factor; GC: glucocorticoids; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein. DAS28: Disease Activity Score; SDAI: Simple Disease Activity Index; CDAI: Clinical Disease Activity Index; HOMA: Homeostasis Model Assessment, HOMA2-IR: calculated with insulin, HOMA2-%B: beta cell production calculated with C peptide.

differences observed in hepatic and peripheral insulin sensitivity or for insulin secretion. Intact and split proinsulin have been found to be upregulated in patients with RA (10), indicating that disproportionate hyperproinsulinaemia is present in RA a reflection of β-cell dysfunction. In this report, disproportionate hyperproinsulinaemia was observed in patients with RA after multivariable analysis, including glucocorticoids. However, we did not find hyperamylinaemia in patients with RA after multivariable analysis. Taking this in account, it is possible that amylin plays no role in the development of IR in RA. Therefore, we believed that the pathophysiological pathways related to amyloid deposit may not be preponderant in the mechanisms leading to IR in RA.

In the univariate analysis, we found a positive relation of amylin serum levels with SDAI and rheumatoid factor. In addition, we noted a positive trend in the relation with CRP, although statistical significance was not reached. This relation with disease activity was not observed for the other disease activity scores. This trend for a relation with CRP is consistent with a recent report (6) in which plasma amylin concentrations were positively associated with CRP, IL-6, BMI, waist circumference, blood pressure, fasting glucose, insulin, amylin/insulin ratio, HOMA-IR, LDL cholesterol and triglycerides, while being negatively associated with HDL cholesterol. In this report, after multiple adjustments, the risk of metabolic syndrome was significantly higher (odds ratio 3.71 [95%CI 2.53–5.46]) in comparisons between the highest and lowest amylin quartiles. The association remained significant even further controlling for BMI, inflammatory markers, insulin or HOMA-IR (6).

To the best of our knowledge, our study is the first in the literature to assess amylin in a chronic inflammatory disease. Remarkably, in our series, although insulin, C peptide serum levels and HOMA2 indices were higher in patients treated with prednisone, this was not the case with amylin. It is believed that glucocorticoids induce an increase in hepatic glucose production and peripheral resistance to insulin action, though it is not known if the mechanisms involved in glucocorticoid-related IR include a defect in β-cell secretion. In fact, the role of hepatic insulin extraction in humans treated long-term with glucocorticoids has not been investigated. The effect of dexamethasone on beta cell secretion and amylin has only been addressed in a single study (30). In this report, frequently sampled intravenous and oral glucose tolerance tests were performed in healthy subjects before and after 5 days of oral dexamethasone administration (4 mg/day). An increase in overall beta-cell activity was observed during both tests based on analysis of oral glucose tolerance test profiles of amylin, the secretion of which was higher following glucocorticoid treatment. Our results, however, do not go in the same direction. We believe that the effects of short-term use of dexamethasone is not equivalent to those driven by prolonged use of low doses of glucocorticoids. Our findings suggest that amylin is not involved in the IR induced by glucocorticoids, in which a peripheral phenomenon *versus* a direct β-cell dysfunction or a defect in secretion has previously been suggested. However, glucocorticoids exert pleiotropic metabolic effects that may be difficult to interpret in a chronic inflammatory disease setting, such as RA. For this reason, future studies are needed to clarify the relation of glucocorticoids with amylin.

In conclusion, we have assessed for the first time how amylin is related to IR in a large population of patients with RA. Our finding that amylin, a critical molecule involved in type 2 diabetes, is not implicated in the IR of RA patients may well support the contention that IR in inflammatory arthritis may be due to a peripheral mechanism related to inflammation and not stem from a mechanism whereby β-cell secretion of amylin is disrupted.

Authors' contributions

IFA and MAGG had full access to all of the data in the study and take responsibility for the integrity of the data and the accuracy of the data analysis. IFA and MAGG conceived and designed the study. BTS, AdVG, BU, RLM, JMO, JLH, and IFA acquired the data. BTS, AdVG, BU, RLM, JMO, JLH, MAGG, and IFA analysed and interpreted the data. All authors were involved in drafting the manuscript or critically revising it for important intellectual content. All authors read and approved the final manuscript.

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