Accepted Preprint first posted on 10 January 2017 as Manuscript EJE-16-0653

Metformin prevents metabolic side effects during systemic glucocorticod treatment

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Short title: Metformin and side-effects of glucocorticoids

Keywords: Metformin, prevention, glucocorticoids, side effects

Word count: 2921

Clinicaltrials.gov NCT01187849

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ABSTRACT

Objectives

Patients receiving glucocorticoid treatment are prone to develop metabolic complications. In preclinical studies metformin prevented the development of the metabolic syndrome during glucocorticoid excess. We herein investigated the metabolic effect of metformin during glucocorticoid treatment in non-diabetic patients.

Methods

In a double-blind, placebo-controlled trial, patients starting glucocorticoid treatment (prednisone, prednisolone or methylprednisolone) for four weeks were randomized to concomitantly receive metformin (850mg once daily for one week followed by 850mg twice daily for three weeks) or placebo. All patients underwent a standardized oral glucose tolerance test at baseline and after four weeks. The primary endpoint was change in the 2h area under the curve (AUC) of glucose during the oral glucose tolerance test between baseline and four weeks.

Results

29 of 34 randomized non-diabetic patients completed the trial (17 metformin, 12 placebo). In patients allocated to placebo, median glucose 2h AUC increased from baseline to four weeks (836 [IQR 770-966] to 1202 [1009-1271] mmol l^{-1} min^{-1}; p=0.01). In contrast, glucose levels remained similar to baseline in the metformin group (936 [869-1003] to 912 [825-1011] mmol l^{-1} min^{-1}; p=0.83). This change within four weeks was different between both groups (p=0.005). Glucocorticoid equivalent doses were similar in both groups (placebo: 980.0 [560.0-3259.8]mg/28d; metformin: 683.0 [437.5-1970.5]mg/28d; p=0.26).

Conclusions

In this first randomized, controlled trial of metformin targeting metabolic complications in patients needing glucocorticoid therapy, we observed a beneficial effect of metformin on

glycaemic control. Metformin thus seems to be a promising drug for preventing metabolic side effects during systemic glucocorticoid treatment.

INTRODUCTION

Up to 2.5% of the adult western population receive systemic glucocorticoid therapy, mostly for inflammatory conditions. Diabetes mellitus, dyslipidaemia, central obesity and hypertension are well-known and common side effects of glucocorticoid treatment [1, 2]. Especially, diabetes mellitus is a recurring problem with a reported prevalence of up to 40% in patients receiving long-term glucocorticoid treatment [3-7]. Even if used as an antiemetic drug in cancer patients, glucocorticoids clearly increased the risk of diabetes mellitus [7]. In contrast to other well-known side effects of glucocorticoids, such as gastric ulcer disease, no randomized-controlled evidence exists that has investigated potential therapeutics for the treatment of metabolic side effects of glucocorticoids.

Many of the changes seen in glucocorticoid excess, such as gluconeogenesis, correspond to metabolic steps regulated by adenosine-monophosphate-activated protein kinase (AMPK) [8]. AMPK is a key regulator of energy metabolism and mediator of several hormones affecting appetite and metabolism [9]. Metformin, a widely used drug for prevention and treatment of diabetes mellitus type 2, exerts most of its beneficial effects on metabolism through the activation of AMPK [10, 11]. We have shown previously that glucocorticoid treatment changes AMPK activity in human adipocytes in vitro and reduced AMPK activity is seen in adipose tissue of patients with Cushing's syndrome [12, 13]. Importantly, metformin reversed the effects of corticosteroids on AMPK in vitro both in primary hypothalamic cell culture as well as in adipocytes, suggesting that metformin and glucocorticoids influence the AMPK signalling pathway in opposite ways and that the metformin effect is able to override the cortisol effect [12, 14]. In vivo studies showed that treatment with an AMPK activator prevented glucocorticoid-induced increase in glucose levels, hepatic glycogen production and hepatic steatosis in rats

[15]. Furthermore, metformin efficiently prevented the dexamethasone-induced deterioration of glucose metabolism in mice and horses [16, 17]. These data suggest that metformin treatment could be beneficial in preventing metabolic complications in patients receiving long-term corticosteroid treatment.

In the first double-blind, randomized, placebo-controlled trial we investigated the metabolic effects of metformin during glucocorticoid treatment in non-diabetic patients starting treatment with corticosteroids for at least 4 weeks.

MATERIALS AND METHODS

Study Design

In this randomized, placebo-controlled, double-blind study, we included patients starting glucocorticoid treatment for at least 4 weeks. Participants were recruited at several departments at the University Hospital Basel and the Cantonal Hospital Aarau from August 2010 to March 2015. Patients were randomized in a 1:1 ratio to receive either metformin 850mg daily p.o. for one week followed by 850mg twice daily p.o. for another three weeks or identical placebo (Merck). The study was terminated after four weeks in all patients, also in cases where glucocorticoid treatment was continued. The study was registered at Clinicaltrials.gov NCT01187849.

Patients

Inclusion criterion was a newly initiated treatment with prednisone ≥7.5mg or an equivalent glucocorticoid for at least 4 weeks. Glucocorticoid tapering was determined by the treating physicians. Exclusion criteria were preexisting diabetes mellitus (according to the American Diabetes Association criteria); renal insufficiency (estimated glomerular filtration rate using the CKD-EPI formula above 60 ml/min/1.73); severe conditions affecting renal function (e.g. dehydration, fever, severe infection); severe conditions causing tissue hypoxia (e.g. acute cardiac or respiratory insufficiency); scheduled examination using intravascular contrast agent containing iodine; alcohol consumption of more than 40g/d (male) or 20g/d (female); known allergy to metformin; pregnancy or breast feeding; any condition compromising the ability of the subject to give written informed consent.

The study was approved by the ethical committees of the participating hospitals and Swissmedic and was conducted in accordance with the ethical guidelines of the Declaration of Helsinki. Written informed consent was obtained from all participating subjects before randomization.

Study Assessment

At baseline and after four weeks, a standardized 2-hour 75g oral glucose tolerance test was performed. After an overnight fast baseline blood samples for fasting glucose, insulin, HbA1c, a full lipid profile and safety blood measurements were taken directly before ingestion of glucose. Additional blood samples for glucose were taken 30, 60, 90, and 120 minutes thereafter.. Physical examination, urine analysis were performed and doses of glucocorticoids were assessed at both visits. After one week, a telephone call took place to assess compliance, adverse events and dosage of glucocorticoids. Three forms of glucocorticoids were prescribed: prednisone, prednisolone and methylprednisolone (Suppl. Tab. 1). If needed, doses of glucocorticoids, e.g. methylprednisolone, were converted to

equivalent doses of prednisone [18]. Due to glucocorticoid tapering, cumulative glucocorticoid doses were calculated as follows: area under the curve was calculated using glucocorticoid doses at baseline, one and four weeks. The average daily prednisone dose was calculated as the area under the curve of the 28 study days.

Plasma glucose and lipids were measured with enzymatic assays (Cobas® modular analyser, Roche Diagnostics, USA). Serum insulin and c-peptide were assessed using immune assays (Immulite® 2000, Siemens, Germany). Hba1c was analysed in EDTA plasma with high performance liquid chromatography (G8 HPLC Analyzer, Tosho Bioscience, USA). Measurements of all blood parameters were performed in the routine central laboratory unit of the University Hospital Basel. The reported HOMA index was calculated according to Matthews et al. [19]. Body impedance analysis (Bodyimpedance Analyzer Model BIA 101, Akern Srl Florence Italy) was performed to assess body composition and energy expenditure.

A randomization list based on single sequence of random assignments, was created by the Pharmaceutical Unit of the University Hospital Basel. Patients as well as study personnel were blinded to the medication allocation.

Study End Points

The predefined primary endpoint was the change in the area under the concentration-time curve (AUC) for glucose during the 75g oral glucose tolerance test between baseline and four weeks. Predefined secondary endpoints included change in fasting glucose levels, glycated haemoglobin levels (HbA1c), Homeostatis Model Assessment (HOMA)-Index, fasting lipid levels, body mass index, body composition and waist/hip ratio.

Statistical Analysis

According to the protocol, the primary analysis followed the intention to treat principle, i.e. patients with complete follow-up were analysed in the groups to which they were randomized. Patients in the metformin group were expected to have unchanged 2 hour glucose levels after ingestion of 75g glucose, while patients in the placebo group would have an increase of approximately 25%. Based on these assumptions, a sample size of 66 patients (33 per arm) was calculated to detect a significant difference between these distributions with a power of 90% at the two-sided 5% level. Discrete variables are expressed as counts (percentages) and continuous variables as median (interquartile range, IQR). To compare changes across treatment groups the Mann-Whitney-U test was used for continuous data and the Fisher's exact test for categorical data. The Wilcoxon signed-rank test was used for comparisons within subjects. Incremental AUC for glucose values (during 120 minutes of a standardized oral glucose tolerance test) and glucocorticoid doses (during 28 days of study duration) was calculated using the trapezoid rule. To adjust for relevant covariates linear regression analyses were employed. P value <0.05 was defined as significant. Data were analysed using statistical software (Statistical Package for Social Sciences, IBM SPSS Version 22, Chicago IL). Figures were drawn using GraphPad Prism (GraphPad Software Inc., LaJolla, CA).

RESULTS

Baseline characteristics

34 individuals were randomly assigned (1:1) to receive metformin (n=20) or placebo (n=14). In the metformin group two patients withdrew from the study due to gastrointestinal symptoms and vertigo, respectively; another patient was lost to follow up after the baseline visit. In the placebo group one patient did not receive glucocorticoids as foreseen and one

patient was lost to follow up. A total of 17 subjects in the metformin group and 12 subjects in the placebo group completed the trial (Fig. 1). Patients in both treatment groups were well matched for baseline characteristics (Tab. 1). Baseline glucocorticoid doses were similar in both groups (metformin: 35.0 (11.3-50.0) mg/d; placebo: 30.0 (20.0-362.5) mg/d; p=0.48). A comparison between patients completing the trial (n=29) and patients dropping out (n=5) showed no difference in baseline criteria except for glucocorticoid doses (complete: 40.0 (20.0-95.0) mg/d; drop out: 12.5 (10.0-26.3) mg/d, p=0.03) (Suppl. Tab. 2). AUC prednisone doses in patients completing the trial remained similar in both groups throughout the study (metformin: 683.0 (437.5-1970.5) mg/28d; placebo: 980.0 (560.0-3259.8) mg/28d, p=0.26). Indications for glucocorticoid treatment are presented in Tab. 2. Concomitant medication with potential effect on glucose and/ or lipid metabolism is listed in Supplemental Table 3. Due to slow study recruitment and time expiry of study drug the study had to be prematurely terminated. This led to fewer study participants than intended and to an unbalanced randomization.

Effect of Metformin on Glycaemia

2h-AUC glucose remained similar from baseline to four weeks in the metformin group (p=0.83), while increasing in the placebo group (p=0.01; Fig. 2A-C). Accordingly, the primary endpoint of 2h-AUC glucose change within four weeks was different between both groups (p=0.005; Tab 3; Fig. 2D). After adjustment for gender, cumulative glucocorticoid dose and HbA1c, treatment group remained strongly associated with 2h-AUC glucose (adjustment for gender: treatment group p=0.006, R^2=0.32; adjustment for glucocorticoid dose: treatment group p=0.003, R^2=0.33; adjustment for HbA1c: treatment group p=0.002, R^2=0.38). Among the secondary endpoints, the change in fasting glucose, fasting insulin and

HOMA-index were different between the two groups (p=0.01, p=0.003, and p=0.035, respectively; Fig. 3A-F). We observed no change in HbA1c in the treatment and placebo groups during the study period (p=0.64; Fig. 3G-H).

Furthermore, we aimed to differentiate between responders and non-responders to metformin. As the 2h AUC glucose increase in the placebo group was 40.3 (18.9-51.0)%, we allocated patients in the metformin group with an increase below 20% to responders, and patients with an increase equal and above 20% to non responders. This resulted in three patients, which were classified as non-responders. Their baseline characteristics are listed in Supplement Table 4.

Effect of Metformin on Lipids

Fasting triglyceride levels did not change during the trial and there was no difference between groups (p=0.30). Total cholesterol levels increased only in the placebo group (p=0.02) while remaining stable in the metformin group (p=0.10). No difference in cholesterol between groups was observed (p=0.15). HDL levels increased in both groups compared to baseline (metformin: p<0.0001; placebo: p=0.003). The HDL increase over the four weeks was more pronounced in the metformin group (p=0.04). LDL levels did not change during the trial and there was no difference between groups (p=0.71; Tab 3).

Effect of Metformin on Body Composition and Energy Expenditure

We identified no change in BMI, waist-hip ratio, basal metabolic rate, fat free mass and fat mass during the study period; there was no difference across treatment groups (Tab. 3).

Adverse events

Gastrointestinal symptoms were present in 20.0% of patients in the metformin and in 21.4% of patients in the placebo group (Suppl. Tab 5). All gastrointestinal symptoms were either

mild or moderate. There was no difference between groups (p=0.99). In the metformin group one subject discontinued the study due to gastrointestinal symptoms, another patient discontinued due to vertigo. One subject in the metformin group was hospitalized for further evaluation of the underlying disease (vasculitis) after study inclusion. The hospitalization was rated as serious adverse event unrelated to the study drug.

DISCUSSION

In this trial with non-diabetic patients receiving systemic glucocorticoids, we demonstrate for the first time that preventive metformin treatment is superior to placebo with respect to glycaemic control as indicated by 2h glucose AUC, HOMA-Index, fasting glucose and fasting insulin. This effect was consistent after adjustment for gender, cumulative glucocorticoid dose and HbA1c. While HDL cholesterol levels increased in both groups during GC treatment, we did not observe a change in triglycerides, LDL, body weight or body composition.

Despite the very frequent use of glucocorticoids and the well-known detrimental impact on glucose metabolism, hardly any randomized-controlled trials have investigated the prevention of glucocorticoid-induced diabetes [20-23]. In one of these trials, troglitazone prevented deterioration of glucose metabolism during glucocorticoid treatment, while pioglitazone and metformin had no effect [24]. Noteworthy, troglitazone can no longer be used as it was withdrawn from the market. Compared to our study, duration of metformin and steroid treatment was very short and metformin dose was low. Two other randomized controlled trials targeting the GLP-1 pathway produced heterogenous results [25, 26]. Importantly, all three studies were performed in individuals without inflammatory disease, thus not representing the

patients in need of glucocorticoid treatment. As inflammation is a known mediator of insulin resistance, it is important to investigate potential benefits of metformin in an appropriate study population [27]. Therefore, more convincing strategies to prevent metabolic side effects of glucocorticoid treatment in patients indeed suffering from inflammatory diseases are needed.

From a pathophysiological point of view, metformin is an attractive preventive treatment strategy in patients receiving corticosteroids. Metformin's mode of function has been extensively discussed and several mechanisms such as inhibition of glycerolphosphate dehydrogenase, enhanced action of glucagon-like-peptide 1 or antagonism of glucagon have been proposed [28-31]. Overall, activation of AMPK seems to play an important role [10, 11, 32, 33]. AMPK is generally considered to be a master regulator of energy metabolism, sensing energy depletion and activating energy-generating pathways [9]. Glucocorticoids have been shown to inhibit AMPK activity and, importantly, metformin was able to reverse this inhibitory effect of glucocorticoids on AMPK in vitro and in animal studies [12, 13, 15].

In accordance with these experimental data, our study showed that metformin favourably influences several side effects of glucocorticoid therapy. We found that metformin prevented an increase of 2h glucose AUC indicating preservation of glucose tolerance. The HOMA-Index, a marker of insulin resistance, clearly improved in the metformin group while deterioration was observed in the placebo group. Fasting glucose levels decreased in the metformin group while increasing in the placebo group during the study period. Moreover, change in fasting insulin was different between groups. Still, we could not identify a difference in HbA1c. However, our study was conducted over four weeks while HbA1c reflects average blood glucose over the previous 8 to 12 weeks [34]. Therefore, we speculate that a longer study duration could show a beneficial effect on HbA1c.

Compared to glucose metabolism, the role of glucocorticoids in lipid metabolism is more controversial. Patients with endogenous overproduction of glucocorticoids are prone to

develop dyslipidaemia [1]. Similarly, glucocorticoid administration has been associated with deterioration of lipid metabolism [35]. Interestingly, in a large observational study, glucocorticoids were associated with higher HDL levels and glucocorticoid treatment was shown to normalize HDL levels in rheumatoid arthritis [36-38]. This positive effect of glucocorticoids may be due to the reduction of the inflammatory burden rather than a direct impact on lipid metabolism.

While the role of glucocorticoids on lipids remains unclear, metformin presumably has a beneficial effect by decreasing triglycerides and LDL cholesterol while increasing HDL cholesterol independent of glucose metabolism [39-41]. In our trial, we did not observe a change in triglycerides nor LDL; however, HDL cholesterol levels increased in both study groups. This finding may be due to a direct effect of glucocorticoids or rather an indirect effect of lowering the inflammatory status.

Central obesity is another characteristic feature of chronic high dose glucocorticoid exposure [42, 43]. In the Diabetes Prevention Program Study metformin reduced body weight for around 2kg during a two year study period in diabetic patients [44]. Thus, metformin exerts opposite effects to glucocorticoids regarding weight.

In our trial, four weeks of glucocorticoid treatment did not result in change of body composition or waist/hip ratio in either study groups. Consequently, no effect of metformin could be observed. Possibly, the study duration was too short and the sample size too small; longer treatment duration with corticosteroids and metformin or placebo, respectively, may provide different results.

Gastrointestinal adverse events occurred in similar number in both treatment groups. Several other studies found metformin to be safe and well tolerated [45].

Our study has some limitations. First, the study was prematurely terminated which led to a rather small sample size. This was due to a combination of slow and difficult recruitment and time to expiry of the trial drug. Nevertheless, due to higher than expected effect of metformin the sample size was sufficient to demonstrate a significant effect on the primary and several secondary endpoints. Since we show a highly significant result, lack of statistical power is not an issue. Second, more and predominantly male patients were in the metformin group. Third, causes of glucocorticoid administration were very variable and the study design did not allow stratification of diseases. While overall glucocorticoid doses were not different between groups, some participants in the placebo group received the highest doses. Importantly, variability of indications and administration of glucocorticoid treatment mirror real life practice, and make the results more generalizable. Fourth, baseline HbA1c was slightly higher in the placebo group, potentially putting these patients at higher risk for development of diabetes. Importantly, the difference in HbA1c was not significant between groups, and the primary endpoint remained highly significant after adjustment for HbA1c.

Our results indicate that metformin prevents deterioration of glucose metabolism if treatment is timed with initiation of glucocorticoids. This study provides the basis for metformin as a preventive treatment in patients newly receiving glucocorticoid therapy. Further studies are needed to test if occurrence of glucocorticoid-induced diabetes can be reduced, and if metformin has similar beneficial effects in patients with continuous glucocorticoid treatment. As our patient number was too small to identify unique characteristics distinguishing responders from non-responders, this remains to be investigated in future studies.

In summary, this is the first randomized-controlled trial showing that metformin has a beneficial preventive effect on glycaemic control in non-diabetic patients receiving systemic glucocorticoid therapy.

DECLARATION OF INTEREST

All authors declare no conflict of interest.

FUNDING

This work was supported by the Swiss National Research Foundation

AUTHOR CONTRIBUTIONS

M.C.-C. designed the study. E.S., S.M., T.K. N.N., M.B. conducted the experiments. E.S. analysed the data. E.S., S.M., M.C.-C. wrote the manuscript. T.K., I.P., P.S., B.M., M.K. reviewed and edited the manuscript. M.C.-C. is the guarantor of the study and, as such, takes responsibility for the contents of this article.

ACKNOWLEDGMENT

We are grateful for Merck&Co to provide study drug and matching placebo; Dr Blerina Kola for her contribution in the initial planning of the study; Prof Matthias Briel for statistical support; Dr Thomas Daikeler, Dr Peter Haeusermann, Dr Christof Rottenburger, Prof Viviane Hess for their support regarding recruitment; Susanne Ruesch and Cemile Bathelt for their support in conducting the trial. We especially thank Marc Donath for supporting the study.

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FIGURE LEGENDS

Fig. 1. Enrolment of participants

Fig. 2. Change in glucose during oral glucose tolerance test

A) Plasma glucose values during oral glucose tolerance test at baseline and after four weeks in placebo treated patients. B) Glucose values during oral glucose tolerance test at baseline and after four weeks in patients treated with metformin. C) 2h-AUC glucose in both study groups at baseline and after 4 weeks. D) Differences in 2h-AUC glucose between baseline and four weeks in each study group. Data represent median values error bars indicate interquartile ranges. * indicates p-value <0.05.

Fig. 3. Change in HOMA-Index, fasting glucose, fasting insulin and HbA1c

A) HOMA-Index at baseline and after four weeks for both study groups. B) Differences in HOMA-Index between baseline and four weeks in each study group. C) Fasting glucose at baseline and after four weeks in each study group. D) Differences in fasting glucose between baseline and four weeks in each study group. E) Fasting insulin at baseline and after four weeks in each study group. F) Differences in fasting insulin at baseline and after four weeks in each study group. G) HbA1c at baseline and after four weeks in each study group. H) Differences in between baseline and four weeks in each study group. Data represent median values, error bars indicate interquartile ranges. * indicates p-value <0.05.

Tab. 1 Baseline characteristics (including 5 patients with missing outcome variables); median values (IQR)

	Placebo (n=14)	**Metformin (n=20)**	**P-Value**
Male sex (%)	35.7	70.0	0.08
Age (years)	56.5 (46.5-67.8)	58.0 (35.8-74.3)	0.69
BMI (kg/m^2)	25.7 (20.6-27.5)	24.2 (21.6-28.6)	0.69
Waist/hip ratio	0.9 (0.8-1.0)	1.0 (0.9-1.0)	0.24
Systolic blood pressure (mmHg)	129 (120-147)	132 (116-139)	0.96
Diastolic blood pressure (mmHg)	80 (71-86)	75 (70-80)	0.26
HbA1c (%)	5.7 (5.4-5.9)	5.4 (5.3-5.8)	0.32
HbA1c (mmol/mol)	39.0 (36.0-40.0)	36.0 (34.0-40.0)	0.32
Fasting glucose (mmol/l)	5.0 (4.6-5.3)	4.8 (4.6-5.3)	0.77
Fasting insulin (mIU/L)	5.8 (2.5-11.1)	8.6 (4.3-14.8)	0.29
HOMA Index	1.0 (0.5-2.0)	1.9 (1.0-3.4)	0.18
Glucose 2h AUC ($mmol\ l^{-1}\ min^{-1}$)	864.8 (782.6-1012.1)	937.5 (872.3-991.1)	0.34
Triglycerides (mmol/l)	1.1 (0.9-1.2)	1.3 (0.9-1.7)	0.32
Total cholesterol (mmol/l)	4.8 (4.4-5.2)	4.8 (4.3-5.6)	0.64
HDL cholesterol (mmol/l)	1.4 (1.0-1.7)	1.2 (1.0-1.4)	0.48
LDL cholesterol (mmol/l)	2.9 (2.6-3.1)	3.1 (2.5-3.8)	0.27
Creatinine (umol/l)	67.0 (60.8-75.5)	79.0 (59.8-87.3)	0.27
Prednisone dosage (mg/d)	30.0 (20.0-362.5)	35.0 (11.3-50.0)	0.48
Basal metabolic rate (kcal)	1665 (1423-1923)	1730 (1593-1823)	0.60
Fat free mass (kg)	57.0 (47.2-62.9)	57.9 (50.6-63.3)	0.70

Fat mass (kg)	16.9 (9.3-22.1)	14.6 (8.5-21.2)	0.77

Tab. 2 Indications for glucocorticoid treatment (including 5 patients with missing outcome variables

Diagnosis	**Placebo (n=14)**	**Metformin (n=20)**
Arthritis	2	2
Vasculitis	1	3
Polymyalgia rheumatica	1	2
Eosinophilic fasciitis	1	
Lupus erythematodes	1	
Sarcoidosis		2
Sclerosing Lymphadenopathy	1	
Cutaneous sclerosis		1
MorbusWegener	1	
Alopecia areata		1
Pemphigus	2	1
Eczema		1
Metastatic prostate carcinoma		1
Astrocytoma	1	
Organizing Pneumonia		1
Allergic bronchopulmonary aspergillosis		1
Myasthenia gravis		1
Endocrine Orbitopathy	3	2
Scleritis		1

Tab. 3 Primary and secondary endpoints; median values (IQR); for each parameter, change from baseline was compared between groups (metformin vs. placebo) using the Mann-Whitney-U test and within-groups using the Wilcoxon signed-rank test ([a] Prednisone dosage was calculated as area under the curve using glucocorticoid doses at baseline, one and four weeks).

	Placebo	**Metformin**	**Between-group p**
Glucose 2h AUC (mmol l^{-1} min^{-1}; 17 patients on metformin vs. 8 on placebo)			
Baseline	835.5 (769.9-966.0)	936.0 (869.3-1002.8)	0.005
4 weeks	1202.3 (1008.8-1270.9)	912.0 (825.0-1011.0)	
Within-group p	0.01	0.83	
HOMA-Index (17 vs. 9)			
Baseline	1.0 (0.4-1.4)	2.2 (1.0-3.6)	0.035
4 weeks	1.5 (0.8-2.0)	1.1 (0.6-2.7)	
Within-group p	0.07	0.04	
Fasting glucose (mmol/l; 17 vs. 11)			
Baseline	4.8 (4.4-5.3)	4.8 (4.6-5.3)	0.01

4 weeks	5.3 (4.5-5.6)	4.6 (4.2-5.0)	
Within-group p	0.07	0.04	
Insulin (mIU/L; 17 vs. 10)			
Baseline	5.4 (2.3-8.3)	9.3 (4.5-15.6)	
4 weeks	6.8 (4.0-13.4)	5.7 (3.3-13.4)	0.003
Within-group p	0.07	0.06	
HbA1c (%; 16 vs.12)			
Baseline	5.7 (5.3-5.9)	5.4 (5.3-6.0)	
4 weeks	5.8 (5.3-5.9)	5.5 (5.3-6.0)	0.64
Within-group p	0.19	0.48	
HbA1c (mmol/mol; 16 vs. 12)			
Baseline	39.0 (34.0-41.0)	36.0 (34.0-42.0)	
4 weeks	40.0 (34.0-41.0)	37.0 (34.0-42.0)	0.64
Within-group p	0.19	0.48	
Triglycerides (mmol/l; 17 vs. 11)			
Baseline	1.1 (0.8-1.1)	1.3 (0.9-1.6)	
4 weeks	1.2 (0.9-1.3)	1.2 (1.0-1.4)	0.30
Within-group p	0.17	0.65	
Total cholesterol (mmol/l; 17 vs. 11)			
Baseline	4.8 (4.5-5.1)	4.8 (4.2-5.7)	0.15
4 weeks	5.6 (4.8-6.9)	5.4 (4.6-6.4)	

Within-group p	0.02	0.10	
HDL (mmol/l; 17 vs. 11)			
Baseline	1.5 (1.0-1.6)	1.3 (1.1-1.5)	0.04
4 weeks	2.0 (1.7-2.8)	1.7 (1.3-1.9)	
Within-group p	0.003	<0.0001	
LDL (mmol/l; 17 vs. 11)			
Baseline	2.9 (2.6-3.1)	3.0 (2.3-3.9)	0.71
4 weeks	3.0 (2.7-3.4)	3.0 (2.5-3.8)	
Within-group p	0.53	0.83	
BMI (kg/m^2; 17 vs. 12)			
Baseline	25.7 (21.9-26.4)	23.7 (20.9-28.7)	0.30
4 weeks	25.5 (21.4-27.4)	23.6 (21.1-28.7)	
Within-group p	0.72	0.26	
Waist-hip ratio (16 vs. 9)			
Baseline	0.9 (0.8-1.0)	1.0 (0.9-1.0)	0.36
4 weeks	1.0 (0.9-1.0)	1.0 (0.9-1.0)	
Within-group p	0.17	0.93	
Basal metabolic rate (kcal; 14 vs. 10)			
Baseline	1665 (1523-1888)	1730 (1550-1835)	0.95

4 weeks	1620 (1418-1952)	1745 (1513-1820)	
Within-group p	0.65	0.55	
Fat free mass (kg; 14 vs. 10)			
Baseline	57.0 (47.5-62.3)	58.3 (52.9-63.9)	
4 weeks	54.7 (41.6-63.2)	57.7 (51.5-64.7)	0.38
Within-group p	0.37	0.95	
Fat mass (kg; 14 vs. 10)			
Baseline	16.9 (10.4-21.3)	14.6 (9.8-22.3)	
4 weeks	19.2 (12.1-22.8)	17.1 (9.5-22.9)	0.98
Within-group p	0.59	0.35	
AUC Prednisone dosage (mg/28d: 17 vs 12) [a]	980.0 (560.0-3259.8)	683.0 (437.5-1970.5)	0.26

Fig 1)

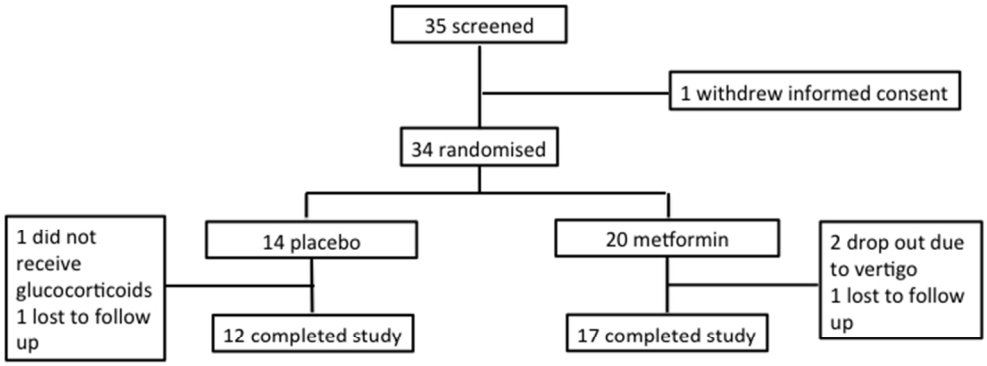

Fig. 1. Enrolment of participants

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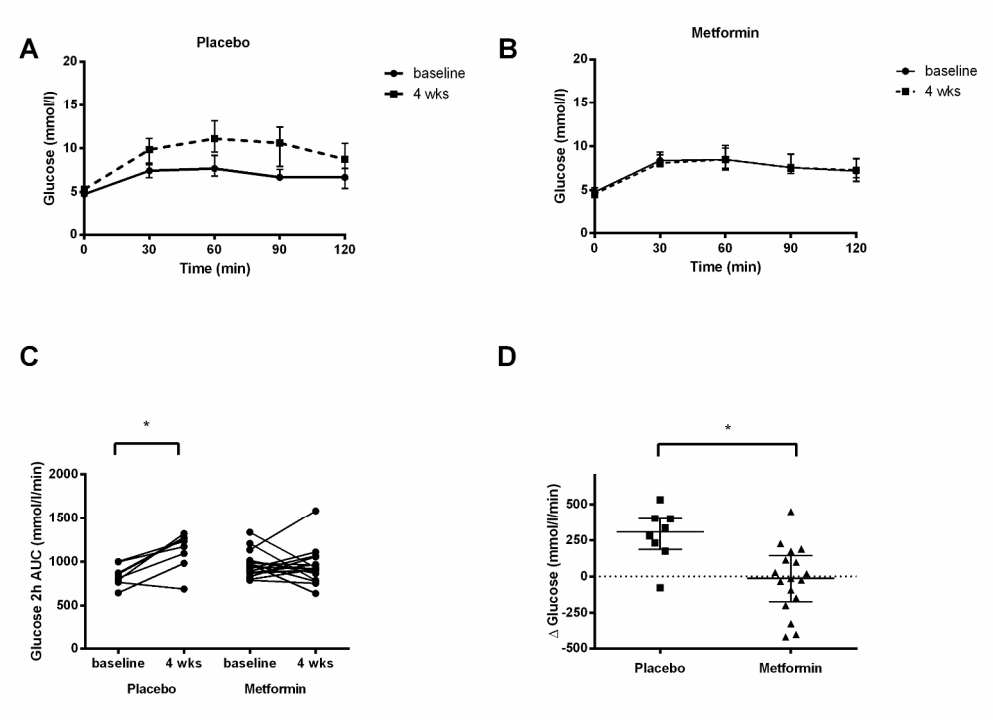

Fig. 2. Change in glucose during oral glucose tolerance test
A) Plasma glucose values during oral glucose tolerance test at baseline and after four weeks in placebo treated patients. B) Glucose values during oral glucose tolerance test at baseline and after four weeks in patients treated with metformin. C) 2h-AUC glucose in both study groups at baseline and after 4 weeks. D) Differences in 2h-AUC glucose between baseline and four weeks in each study group. Data represent median values error bars indicate interquartile ranges. * indicates p-value <0.05.

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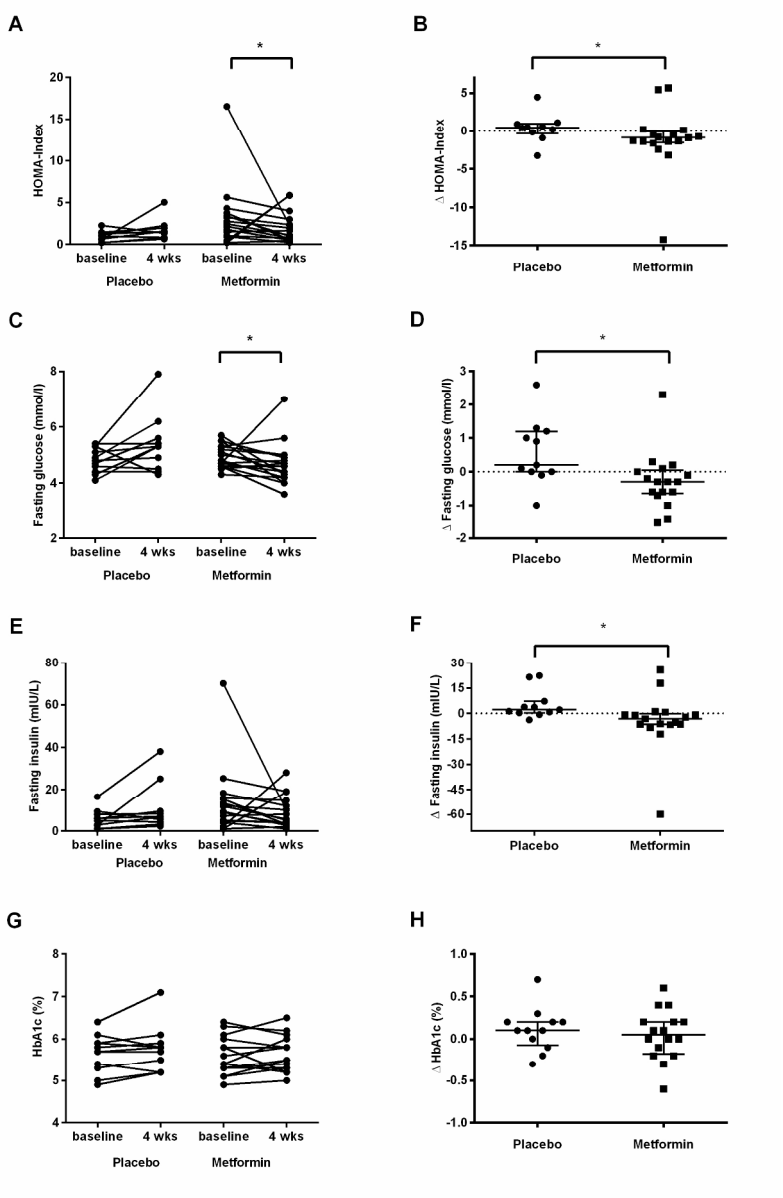

Fig. 3. Change in HOMA-Index, fasting glucose, fasting insulin and HbA1c
A) HOMA-Index at baseline and after four weeks for both study groups. B) Differences in HOMA-Index between baseline and four weeks in each study group. C) Fasting glucose at baseline and after four weeks in each study group. D) Differences in fasting glucose between baseline and four weeks in each study group. E) Fasting insulin at baseline and after four weeks in each study group. F) Differences in fasting insulin at baseline and after four weeks in each study group. G) HbA1c at baseline and after four weeks in each study group. H) Differences in between baseline and four weeks in each study group. Data represent median values, error bars indicate interquartile ranges. * indicates p-value <0.05.

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