# Metformin for Ectopic Fat Deposition and Metabolic Markers in Polycystic Ovary Syndrome (PCOS)

NCT02500147

April 23, 2018

**Title: Body composition and metabolic manifestations of insulin resistance in adolescents with polycystic ovary syndrome. Ectopic fat deposition and metabolic markers: Intervention and follow-up portion**

**Principal Investigator: Aviva Sopher, MD, MS**

**Study Purpose and Rationale:**

Polycystic ovary syndrome (PCOS) affects about 7% of females of reproductive age worldwide and is associated with development of lifelong morbidities including type 2 diabetes mellitus and cardiovascular disease. It commonly first presents in adolescence and young adulthood, which places a significant proportion of this young population at risk for long-term morbidity. Although the underlying etiology of PCOS has not been fully elucidated, insulin resistance (IR) is suggested to be an important underlying causal factor. The association between PCOS and IR further links it to other IR-related disorders including obesity, the metabolic syndrome (MeS) and non-alcoholic fatty liver disease (NAFLD). This constellation of disorders is associated with dyslipidemia, cardiovascular disease and type 2 diabetes mellitus. Recently, NAFLD has been reported to be more prevalent in young individuals with PCOS compared to the general population. Additionally, the progression of hepatic steatosis to non-alcoholic steatohepatitis (NASH) is described to be substantially more rapid in PCOS than in unaffected individuals. Metformin is an insulin sensitizing medication typically used to treat type 2 diabetes mellitus, but also has beneficial effects on IR related conditions such as PCOS and NAFLD; however, its effects on NAFLD in PCOS in the adolescent and young adult populations have not been adequately studied. If, in fact, metformin reduces liver fat in adolescents and young adults with PCOS, their long-term risk of metabolic and cardiovascular disease could be diminished. Additionally, this finding would have both personal and far-reaching beneficial public health benefits. There are limited data that suggest that metformin may also have beneficial effects on bone. If our pilot study demonstrates changes in bone formation markers that corroborate recent proposed mechanisms of action, they could provide proof of concept to support further investigation of the skeletal effects of metformin in a larger randomized clinical trial.

This study proposes to explore the efficacy of metformin to reduce liver fat using novel and minimally invasive imaging and biomarkers in a pilot randomized double-blind placebo-controlled trial.

**Study Design:**

Eligibility Criteria: PCOS subjects who participated in a previous baseline study and have liver fat greater than or equal to 4.8% by MRS or evidence of insulin resistance (on an oral glucose tolerance test) will be invited to participate in this study with the goal of recruiting 44 subjects who are 13 to 25 years at least two years post-menarche. Oligomenorrheic subjects will complete the protocol during the early follicular phase of the menstrual cycle (days one through seven) and amenorrheic subjects will complete the protocol on any day. Exclusion criteria: 1. Presence or history of a medical disorder (e.g. non-classical CAH) or medication known to affect body composition, insulin secretion and sensitivity, or the GH-IGF1 axis (i.e. use of glucocorticoids or thyroid hormone), excessive alcohol use, liver disease (other than NAFLD) indwelling hardware, current or past pregnancy and use of metformin or hormonal contraception within 3 months of enrolling in the previous study.

The protocol will comprise:

a) Randomization: Subjects whose liver fat is 4.8% or greater or have evidence of insulin resistance on an oral glucose tolerance test (n=22) will be randomized to placebo or extended-release metformin (XR) (500 mg (1 tablet) once a day for one week and then 1000 mg (2 tablets) daily for six months). Randomization will be double-blind and will be performed by randomly selected permuted size 2 and size 4 blocks determined by the CUMC Research Pharmacy with the guidance of a statistician and will be stratified based on presence or absence of insulin resistance.

b) Intervention: All subjects will receive identically appearing pill bottles labeled "Study Medication" containing their first three-month supply of either metformin XR or placebo. Subjects will be instructed to take pills after dinner and to take one pill once a day for one week and subsequently two pills once a day for the remainder of the first three months. Subjects will be asked to call with any side effects that they may experience. They will be given a pill diary so that they can keep track of their compliance. They will be instructed to call our office if they experience any adverse effects and will be asked to bring their empty pill bottles to their interim visit to further ensure compliance.

c) Brief interim evaluation: After three months of the intervention phase, subjects will be seen for a general health assessment, which includes weight, blood pressure and a physical examination, and will receive their second three-month supply of their assigned intervention along with a second pill diary. They will again be instructed to call our office if they experience any adverse effects and will be asked to bring their empty pill bottles to their follow-up study visit to further ensure compliance.

d) Follow-up exam: At the end of six months, subjects will undergo laboratory and body composition testing similar to those performed in the baseline testing. Studies that are not repeated in this protocol are transabdominal pelvic ultrasound, MRI of the ovaries and DXA of the hip, spine and wrist

1) History and physical exam: Height, weight, BMI (percentile and z-score for <20 years), blood pressure, physical exam, Ferriman-Gallwey hirsutism scoring, smoking, alcohol and family history;

2) Laboratory evaluation will be between 0800 and 1000 after an overnight fast via an intravenous catheter for:

    a) General endocrine panel: Thyroid function, prolactin, 17-hydroxyprogesterone;

    b) PCOS panel: LH, FSH, estradiol, sex hormone binding globulin, testosterone, free testosterone, androstenedione, dehydroepiandrosterone sulfate (DHEAS);

    c) Liver panel: Liver enzymes (ALT, AST, GGT), apoptosis markers (Fas, cytokeratin-18 fragments, M30), pancreatic polypeptide, irisin

d) Cardiovascular risk panel: Cholesterol, triglycerides, HDL, LDL, free fatty acids, c-reactive protein;

e) IR evaluation: hemoglobin A1C, glucose, insulin, c-peptide and two-hour oral glucose tolerance test (OGTT) after a 75 g glucose challenge with measurement of glucose and insulin at 30, 60, 90 and 120 minutes. IR will be determined by age- and BMI- specific HOMA-IR cutoffs based on a National Health and Nutrition Examination Survey (NHANES) study of 1,164 nonobese adolescents 12 to 19 years old. Other IR indices that will be evaluated are whole body insulin sensitivity index (WBISI) and insulin area under the curve (iAUC);

f) Bone evaluation: 25-hydroxyvitamin D, 1,25-dihydroxyvitamin D, osteocalcin, c-telopeptide, PTH, P1NP

3) MRS of the liver for intrahepatic lipid content (IHL) and of the right soleus muscle for intramyocellular lipid (IMCL) acquired using standard point resolved spectroscopy sequence (PRESS). Percentage liver fat will be calculated.

4) Total body MRI with 10 mm slices and 40 mm interslice gaps for total body adipose tissue , subcutaneous adipose tissue (SAT) and visceral adipose tissue(VAT);

5) MRI for percentage liver fat.

e) Dual-energy x-ray absorptiometry (DXA): Total body DXA will be performed for total body lean and fat mass, percentage body fat and bone mineral density (BMD).

**Statistical Procedures:**

Statistical Analysis:

1. Change in liver fat is defined as percentage liver fat at six months minus percentage liver fat at baseline. The difference in mean change between metformin XR and placebo groups as well as its 95% confidence intervals will be calculated. The two-sample t-test will be used to compare groups. Sample Size and Power Calculations: Over a 1.5 year recruitment period, 44 subjects is a feasible recruitable sample size. We generally see at least 100 PCOS patients within our division per year. We also work closely with physicians in the Division of Adolescent Medicine and the Department of Obstetrics and Gynecology who will also serve as sources of study subjects for us. Forty-four subjects randomized 1:1 to metformin versus placebo provided greater than 80% power to detect a 1.4 standard deviation difference in the group average change from baseline to endpoint assuming a 5% type I error rate and an independent T-test. Preliminary data showing subjects with PCOS have an average of 4.8% liver fat, and assuming the average within subject change is less than 0.5+0.5% in the placebo group, the power calculations indicate the minimum detectable difference attributable to metformin treatment to be a 0.7% absolute, or a 15% relative decline in percentage liver fat. After the intent-to-treat analysis we will analyze the influence of partial compliance by adding cumulative dose exposure in to a multiple regression

model assessment of the group difference. Subjects will be stratified based on presence or absence of insulin resistance.

2. The association of percent liver fat with IR, dyslipidemia, VAT, total body adipose tissue, and markers of NAFLD and NASH will be estimated with correlation/regression and change in IR, dyslipidemia, and markers of NAFLD and NASH with change in percent liver fat following metformin with multiple regression. Sample Size and Power Calculations: 44 subjects provide 80% power and 5% alpha to detect a baseline association of liver fat with continuous measures of liver fat markers with r-values > + 0.46 when tested against Ho of r = + 0.10. 20 subjects in the longitudinal study provides 80% power and 5% alpha to detect an incremental increase of 0.60 in model $R^2$ when percentage liver fat is added to treatment assignment to predict change in markers.
3. The current state of conflicting or absent data on BMD and bone turnover markers in adolescents with PCOS and IR means that the data collected here are vital to providing preliminary estimates of between group differences and rates of change for design of future studies. A recent study showed changes in bone markers within 14 weeks of treatment of rosiglitazone. We thus predict that we will see changes in bone markers within SIX months of treatment with metformin (13 weeks). (Glintborg D, Andersen M, Hagen C, Heickendorff L, Hermann AP. 2008). Association of pioglitazone treatment with decreased bone mineral density in obese premenopausal patients with polycystic ovary syndrome: a randomized, placebo-controlled trial. J Clin Endocrinol Metab 93:1696-1701).

**Potential Risks:**

Protection Against Risk:

MRI and MRS: These magnetic resonance and nuclear magnetic resonance studies are without known associated risks except claustrophobia while in the magnet or injury from a metal object or indwelling metal from prior surgery. All subjects will be fully acquainted with the magnet and will be certified as "MRI-safe" before entering.

Dual Energy X-Ray Absorptiometry (DXA): There is no discomfort associated with DXA. There is minimal radiation exposure, but it is well within the limits allowed for this type of research. Over 1500 children have had whole body DXA for research purposes in the Body Composition Unit.

Two-hour Oral Glucose Tolerance Test (OGTT): There may be some bruising at the site of needle insertion, but this will resolve spontaneously.

Metformin: Metformin is rarely associated with development of lactic acidosis in patients with preexisting liver disease, kidney disease, heart failure and alcohol abuse. Subjects will have screening for these disorders by checking medical history and liver and kidney function by blood tests The liver and kidney function blood tests will also be checked the 6 month "follow up visit", which will occur after six months of the study intervention. Study subjects are instructed to call with any new symptoms while they are taking the IP.

Pregnancy: Study subjects may begin ovulating or menstruating during the study as a response to metformin or naturally. Even if the study subject is not getting her period regularly or at all she will be instructed to always use a birth control method if she is sexually active. She cannot take a birth control pill if she is participating in this study so she should use a barrier method of birth control if she is sexually active. It is important to note that metformin is a category B drug in pregnancy, which means that its effects on a fetus are not fully known. Study subjects are instructed to call the study team as soon as possible if they think they may have become pregnant during the study.

Adverse events:

A serious adverse event associated with metformin use is lactic acidosis; however, this most commonly occurs in patients with congestive heart failure, renal failure and liver failure. We will be screening patients by questionnaire and with laboratory data in order to exclude those with underlying medical conditions. We will instruct patients to call immediately if they develop unexplained hyperventilation, myalgia. Malaise, unusual somnolence or other nonspecific symptoms.

We will be tracking the follow adverse event data at the 3 month and 6 month visits based on known common adverse events associated with metformin use: diarrhea, abdominal pain, nausea/vomiting, gas/bloating and any other side effect the subject may experience.

**Potential Benefits:**

Families and subjects will be provided with detailed analysis of body composition, OGTT and cardiovascular risk factor results and will be counseled about the implications for future metabolic complications, as well as on how to improve health through diet and exercise. All results will be provided to each subject's primary care physician and/or pediatric endocrinologist if directed by the subject's parents at time of study or later date.