**Methodology**

**Study design**: Cross-sectional observational study.

**Study duration**: One year.

**Study center**: SMS Medical College and Hospital, Jaipur, India.

**Ethics:** The study was approved by the Institutional Ethics committee and written informed consent was obtained from all subjects before recruitment.

**Subjects**: Thirty women of reproductive age (15–45 years) with HAIR-AN syndrome were recruited in Group A, and 30 BMI-matched women of same age group with PCOS were recruited in Group B. HAIR-AN syndrome was diagnosed on the basis of the clinical triad of hyperandrogenism, insulin resistance, and acanthosis nigricans.PCOS was diagnosed as per the Rotterdam criteria. Hyperprolactinemia, congenital adrenal hyperplasia, Cushing syndrome, or virilizing adrenal/gonadal tumor was ruled out prior to recruitment. Women with a history of using oral contraceptives or other drugs altering the glucose and insulin metabolism within the last 3 months were excluded.

**Clinical Evaluation:**

* A detailed clinical evaluation of patients including age, height, weight, BMI, severity of acne, hirsutism, acanthosis nigricans, and menstrual irregularity was conducted. Hirsutism was evaluated using the Ferriman-Gallwey score and a score of 38 was taken as diagnostic of hirsutism [41]. Acne was graded as mild, moderate, and severe as described by Pochi et al. [42].Acanthosis nigricans was graded as per Burke et al. [43].
* Baseline blood samples were drawn after overnight fasting. Serum levels of follicle stimulating hormone, luteinizing hormone, prolactin, testosterone, dehydroepiandrosterone sulphate, 17-hydroxy progesterone, cortisol, fasting plasma glucose, and insulin were determined.
* Patients in both groups were assessed for serum levels of adiponectin, leptin, TNF-α, and IL-6.
* The methods are summarized in a flowchart as shown in Figure 1.

**Laboratory measurements**: Biochemical parameters like serum glucose, the lipid profile, triglycerides, LDL, HDL, and VLDL were measured on a Kopran AU/400 fully automated analyzer. Serum hormone levels were measured by chemiluminescent immunoassay (ADVIA Centaur XP). Serum insulin was measured using a chemiluminescent immunometric assay (Immulite 2000 machine; sensitivity 0.5 mU/L, CV% 7.5). Leptin (Lab Systems Diagnostic Oy, Finland; sensitivity <62.5 pg/mL, CV% <12), adiponectin (Lab Systems Diagnostic Oy; sensitivity 0.185 ng/mL, CV% <10), IL-6 (Lab Systems Diagnostic Oy), and TNF-α (Lab Systems Diagnostic Oy) were estimated by the ELISA method. Insulin resistance was calculated using the homeostasis model assessment insulin resistance index (HOMA-IR) score: FBG(mg/dL) x fasting Insulin (mU/L)/405. A HOMA-IR cut-off of 2.5 was taken as normal [44].

**Statistical analysis**: Quantitative variables are expressed as the mean ± SD and the qualitative variables are expressed in percentages and proportions. Analysis was done using the SPSS version 23.0 (IBM SPSS Statistics Inc., Chicago, Illinois, USA) software program. Categorical variables were compared using the χ2 test. All values were not normally distributed (assessed using the Shapiro-Wilk test and visual histogram and box plots), the Mann-Whitney U test was used for comparison of numerical data, and correlations were derived using Spearman’s coefficient (rho). Statistical significance was assigned at a *p* value <0.05.