

Pilot study on evaluation and determination of the prevalence of Polycystic Ovarian Syndrome (PCOS) associated gene markers in the South Indian population

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Abstract

Background: Polycystic ovarian syndrome (PCOS) is typically characterized by a spectrum of manifestations that include menstrual irregularities, anovulation, cysts, hyperandrogenic features like hirsutism, acne, alopecia, and various metabolic complications. The pathology of PCOS is complex and several mechanisms have been potentially involved in the genetic abnormalities/dysfunctions. Hence, the present study aims to examine the prevalence and association of polymorphisms in candidate genes (thyroid adenoma-associated gene [THADA], luteinizing hormone and human chorionic gonadotropin receptor [LHCGR], DENN domain containing 1A [DENND1A], follicle-stimulating hormone receptor [FSHR], Connexin37 [CX37], angiotensin-converting enzyme [ACE], insulin receptor [INSR] and calpain 10 [CAPN10]) in PCOS patients of the South Indian regional population. **Methods:** The study group included 20 PCOS cases and 10 controls, whose deoxyribonucleic acid (DNA) were genotyped by the polymerase chain reaction (PCR), PCR-restriction fragment length polymorphism (RFLP), and PCR product sequencing to determine the prevalence of the DENND1A (rs10818854), LHCGR (rs13405728), FSHR (rs2349415), THADA (rs13429458), CX37 (rs1764391), ACE (rs1799752), INSR (rs1799817), and CAPN10 (rs2975760) polymorphisms. Clinical examinations including anthropometric measurements, biochemical investigations relevant to glucose metabolism, and hormones were measured. **Results:** A significant difference was observed in the DENND1A (rs10818854) polymorphism between the control and PCOS patients ($P = 0.001$). The variants of LHCGR, FSHR, THADA, CX37, ACE, INSR, and CAPN10 were not statistically significant with PCOS. The body mass index (BMI) ($P = 0.01$), triglycerides ($P = 0.01$), and dehydroepiandrosterone sulfate (DHEAS) ($P = 0.05$) were significantly different between the PCOS patients and controls. Significant results were observed in rs1799817 single nucleotide polymorphisms (SNP) of INSR with elevated levels of triglycerides and rs10818854 of DENND1A, rs13429458 of THADA, rs2349415 of FSHR with the high levels of DHEAS. **Conclusion:** In the study population, the presence of rs10818854 of DENND1A polymorphism may be associated with the risk of PCOS and high levels of DHEAS.

Keywords: ACE, CAPN10, CX37, DENND1A, FSHR, INSR, LHCGR, PCR, RFLP, polycystic ovarian syndrome, polymorphism, THADA

INTRODUCTION

Polycystic ovarian syndrome (PCOS) is a complex endocrine disorder estimated to affect 5–10% of women of childbearing age and 3.7–22.5% of Indian women.^[1] PCOS is a multifactorial disorder with different genetic, hormonal, and environmental factors contributing to its etiopathogenesis. Many hypotheses about the pathophysiology of PCOS that have been explained so far include resistant to rupture of follicles, ovarian hyperandrogenism, luteinizing hormone (LH) hypersecretion, hyperinsulinemia, and impaired ovarian follicular development due to increased follicular development blocker paracrine

factors, such as anti-mullerian hormone (AMH).^[2] As a primary causative, genetic variations in PCOS disrupt the signaling mechanisms pertaining to steroid hormone synthesis, regulatory

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mechanisms of gonadotropins, insulin synthesis and secretion, energy balance, and homeostasis.^[3] Familial segregation studies and linkage analysis report that PCOS presents an autosomal mode of inheritance and characteristically exhibits an oligogenic trait, ethnic variations. Cohort studies across the globe (Chinese, European, and other populations) reveal that allelic variants/single nucleotide polymorphisms (SNP) in the genes associated with ovarian steroidogenesis, folliculogenesis, and insulin-regulated glycemic control may disrupt homeostatic signaling mechanisms leading to PCOS.^[2-5] In addition to the limited number of studies, replicating PCOS-associated genetic variations in the Indian population, it is also important to note that studies addressing the prevalence and association of the significantly associated marker genes/genetic variants (inclusive of the genes from chromosomes 2, 9) are extremely limited/non-existent in the Indian/South Indian populations.

Genome-wide association studies (GWAS) enable the functional association of the genes and their variants in disease conditions, and thereby, the susceptibility loci for the diseased condition. Based on several GWAS studies in PCOS that report the association of genetic variations impacting ovarian steroidogenesis, folliculogenesis, and insulin signaling,^[6,7] the present study focuses on elucidating the importance and association of such genetic variants in PCOS patients from South India/South Tamil Nadu.

Ovarian steroidogenesis

LHCGR (Luteinizing hormone and human chorionic gonadotropin receptor)

LH is a heterotrimeric protein that plays a pivotal role in reproductive functions such as follicle growth, steroidogenesis, and maturation of oocytes. The G protein-coupled receptor for LHCGR, located in chromosome 2, comprising 11 exons, is expressed in the ovarian theca cells, testicular Leydig cells, and adipose tissue. As a receptor mediating the actions of LH and human chorionic gonadotropin (hCG) on steroid biosynthesis, LHCGR is reported to contain many SNPs that may impact ovarian steroidogenesis. Earlier reports have indicated that exon1 (insLQ) insertional polymorphism and amino acid variations at positions 291, 312 located in exon10 are significantly associated with PCOS. Further, cohort studies from the Chinese, European, and Indian populations independently and cumulatively report the association of rs13405728 in PCOS.^[6,8]

FSHR (Follicle-stimulating hormone receptor)

The FSHR, a G protein-coupled receptor, also located in chromosome 2 (2 p16.3), is expressed in the granulosa cells of the ovary.^[9] Functionally, the follicle-stimulating hormone (FSH)-bound FSHR promotes oogenesis, follicle development, and gametogenesis that results in follicular maturation and granulosa cell proliferation.^[10] Genetic variations in the FSHR gene and the resultant functional implications are observed to arrest follicle development, and thereby, result in primary amenorrhea, hypoplastic ovary, and

high-serum FSH levels. Earlier studies have predicted and demonstrated that the polymorphisms rs6165 and rs6166 in the FSHR gene are associated with the population-specific association of PCOS (South Han Chinese and Koreans). Subsequent phase I, phase II GWAS studies brought to light the significant association of rs2268361 and rs2349415 in the Chinese population and rs1922476, rs2349415 in the European population highlighting that the FSHR gene is a positive candidate marker for PCOS regardless of ethnicity.^[11]

DENND1A (DENN domain containing 1A)

As an activator of small G proteins, guanine nucleotide exchange factor (GEF) plays a critical role in catalyzing the Guanosine diphosphate (GDP) exchange, and thereby, a vital physiological process. DENND1A is one of the four recognized Rab35 GEFs and contains a highly conserved, differentially expressed domain DENN and plays an important role in the clathrin regulatory pathways.^[12,13] Interesting GWAS studies in two independent Chinese populations and a European cohort revealed that DENND1A is significantly associated with PCOS and other studies have also indicated that it is highly expressed in PCOS patients.^[14,15] In particular, the rs10818854, rs2479106 intronic variants of the DENND1A gene have been associated with PCOS.^[16,17]

THADA (Thyroid adenoma-associated gene)

THADA has been attributed a pivotal role in human evolution and GWAS studies have brought to light that it is one of the top risk loci in Type 2 diabetes mellitus (T2DM).^[18] While earlier studies have revealed that THADA is significantly associated with pancreatic β cell response,^[19] further interesting and recent THADA knockout studies have indicated that it is an important regulator/determinant of energy storage and expenditure. Functionally, THADA binds to the Sarco/endoplasmic reticulum (ER) Ca^{2+} adenosine triphosphatase (ATPase) (SERCA) and acts as an uncoupler leading to the observation that THADA knockouts are obese and cold-sensitive.^[20] Further evidence based on familial genetic studies and large cohort studies across the Chinese and European populations also indicate that THADA gene polymorphism rs13429458 may also play a key role in the phenotypic heterogeneity and pathogenic progression of the PCOS patients.^[7] There is a lack of such studies in the South Tamil Nadu population.

Folliculogenesis

ACE (Angiotensin-converting enzyme)

ACE is a key factor that is involved in the conversion of angiotensin I to angiotensin II and is expressed in several tissues including the ovaries. Physiologically, ACE regulates blood pressure, fluid balance, and ovarian functions through follicular development, oocyte maturation, ovulation, and follicular atresia.^[21-27] The intron 15 of the ACE gene corresponds to a 287bp DNA sequence that contains an insertion (I)/deletion (D) polymorphism. Although several reports indicate the association of rs1799752 I/D polymorphisms in PCOS susceptibility and the consequent clinical manifestations such

as insulin resistance, testosterone levels, a few reports also indicate a negative association of the polymorphism with PCOS. However, a significant association of the homozygous deletion (DD) ACE gene polymorphism in the PCOS patients of the Indian population indicates that further studies would enable a better understanding of the polymorphism in the prediction of PCOS.^[28]

CX37 (Connexin37)

Follicular development in the female reproductive cycle critically requires proper intercellular communication between the oocyte and the peripheral granulosa cells through gap junctions.^[29] In particular, the essential multicellular unit termed as mammalian cumulus–oocyte complex requires CX37, a gap junction protein to facilitate communication between the oocytes and the granulosa cells. Loss/lack of the function of CX37 has been demonstrated to arrest follicular development, and its subsequent ovulation.^[30,31] While a single study has reported and identified a CX37 variant rs1764391 (C1019T) to be associated with PCOS in the Indian population, replicative studies that bring to light the importance of the variant in southern Tamil Nadu patients is still lacking.

Insulin signaling pathway

INSR (Insulin receptor)

Insulin resistance is a prominent PCOS-associated dysfunction that is primarily attributed to INSR that is located on chromosome 19. Linkage disequilibrium studies and other genetic studies have revealed a C/T SNP at His1058 in exon17 to be significantly associated with the Chinese and Caucasian PCOS patients. Other studies have also indicated that a T/C polymorphism at Cys1008 in exon17 is also strongly associated as a predictor gene for PCOS.^[32-34] Subsequent studies in the Indian population have interestingly identified that the C/T SNP at His1058 was significantly associated only in PCOS women with a lean phenotype, with insulin resistance and hyperandrogenemia in the assessed population.^[35] Such studies together indicate that exon17 polymorphisms in the INSR gene could serve to predict the risk of incidence of PCOS, and thereby, serve as a genetic marker. While conflicting reports still prevail on the influence of INSR genetic variants in PCOS, the present study efforts are focused to reveal the importance and association of the INSR rs1799817 polymorphism in the chosen PCOS patient population.

CAPN10 (Calpain 10)

CAPN10 located in the 2q chromosome, encodes for a cysteine protease, functionally associated with insulin synthesis and secretion, and thereby, a candidate gene for Type 2 diabetes mellitus (T2DM).^[36] While earlier reports from different groups have also associated CAPN10 polymorphisms with features of PCOS such as fasting insulin, blood glucose levels, and hirsutism,^[37] subsequent studies have not been able to replicate the observed data indicating that population-/ethnicity-based differences could also attribute to the observed differences in the PCOS-associated genotype.^[38-40] Recent reports have also brought forward the fact that the intron 3 polymorphic

variant C (UCSNP44/rs2975760) of CAPN10 is found to be significantly associated with PCOS in the Indian population.^[41]

Hence, the present study, as a first of a kind, proposes to examine the predominant PCOS-associated genetic variants [Figure 1] in ovarian steroidogenesis (LHCGR, DENND1A, FSHR, THADA), folliculogenesis (ACE, CX37), and insulin signaling pathway (INSR, CAPN10) in the South Indian population. The study results would reveal the prevalence, association of the genetic variants/SNPs with PCOS in the regional population, and may also enable unique variations pertaining to the population.

Materials

The prevalence and risk of disease incidence pertaining to the genetic variations impacting ovarian steroidogenesis (LHCGR, DENND1A, FSHR, and THADA), folliculogenesis (ACEI/D, CX37), and insulin signaling (INSR, CAPN10) were aimed to be determined in a pilot population of 20 PCOS patients and 10 controls from a tertiary care center in South Tamil Nadu, India. The study was conducted over 24 months (February 2019 to February 2021) with the approval of the Institutional Ethical Committee and upon obtaining informed consent from the participants. PCOS was defined according to the Rotterdam PCOS consensus criteria. Ten healthy controls with normal cycles, normal ovary ultrasound sonography (USG), and without any clinical/biochemical hyperandrogenism were enrolled. Routine clinical assessments including anthropometric measurements, biochemical tests relevant to glucose metabolism, lipid profile, and hormonal tests such as levels of FSH, luteinizing hormone (LH), dehydroepiandrosterone sulfate (DHEAS), thyrotropin (TSH), thyroxine (FT4), prolactin and estradiol (E2) in the patients with PCOS were measured according to the standard protocols.

Genomic DNA was extracted from the peripheral blood samples using a Qiamp DNA Blood Mini Kit (QIAGEN India Pvt. Ltd., New Delhi, India) according to the manufacturer's protocol. Quantitative and qualitative (260/280 nm absorbance ratio) assessments of the DNA samples were carried out using a nanodrop spectrophotometer, Thermo Fisher Scientific, USA.

The determination of the presence of polymorphism in THADA (rs13429458), LHCGR (rs13405728), DENND1A (rs10818854), CX37 (rs1764391), and INSR (rs1799817) was carried out by Polymerase Chain Reaction (PCR) amplification of a region corresponding to the specific PCR product. The PCR products were visualized after electrophoresis using a Ultra Violet illuminated (UV) gel documentation system (Medicare, Chennai, India). Further, the gel was extracted, purified, and utilized for direct sequencing (Agrigenome, Kochi, Kerala). CAPN10 polymorphism rs2975760 and FSHR polymorphism rs2349415 were also identified by PCR-based restriction fragment length polymorphism (RFLP). The genotyping of Angiotensin-converting enzyme Insertion/Deletion Polymorphism (ACEI/D), PCR was performed for the analysis of the insertion/deletion polymorphism of the

GENETIC MARKERS FOR PCOS



Figure 1: Pathophysiology of PCOS susceptible gene markers.

ACE gene (rs1799752). To ensure the adequate quality of genotyping, we performed random duplicates in 50% of the samples.

Statistical analysis

The allele and genotype frequencies were determined by the gene-counting method. Student's t-test and Fisher's exact test were used to compare the genotype and allele distributions in the study. The relative association between the patients and controls for genotype and allele frequencies was assessed by Pearson's χ^2 test. The clinical variables were compared using a one-way analysis of variance (ANOVA). The Hardy-Weinberg equilibrium (HWE) of genotypes in the patients and control groups was assessed. A strong association (significance) was assumed at $P < 0.05$. The statistical analyses were performed mainly with the help of SPSS 26.0 statistical software, (SPSS, Inc., Chicago, IL, USA).

RESULT

A total of 20 patients and 10 controls were enrolled. Among the study population, 80% of the patients belonged to the age group between 26 and 30 years. The mean age of the patients in our study population was 26.9 ± 5.46 and 26.9 ± 6.34 in the controls. The body mass index (BMI) of the PCOS patients (30.4 ± 6.4) was significantly ($P = 0.01$) higher when compared with the control (23.4 ± 4.7). There was a notable significant difference in the triglyceride ($P = 0.01$) and DHEAS levels ($P = 0.05$) of the patients when compared with the control group [Table 1].

The genotypic and allelic frequencies of rs10818854 were statistically different in the patients and control ($P = 0.001$) and the genotypic frequencies were in accordance with the HWE ($P = 0.356$). It reflects the positive relationship between

rs10818854 in DENND1A and increased risk of PCOS. The genotypic distributions were not statistically significant for rs13405728 in LHCGR ($P = 0.197$), rs13429458 in THADA ($P = 0.301$), rs2349415 in FSHR ($P = 0.091$), rs1764391 in CX37 ($P = 0.114$), rs1799752 in ACE ($P = 0.504$), rs1799817 in INSR ($P = 0.329$), and rs2975760 in CAPN10 ($P = 0.301$) among the patients and controls [Table 2].

The incidence of suspected SNPs in the study population using PCR amplification, direct sequencing, and PCR-based RFLP method is depicted in Figure 2. Together, the results obtained demonstrate the reliability and feasibility of the utilization of PCR-based strategies, PCR product sequencing in the determination of rs10818854, rs13405728, rs13429458, rs1764391, rs1799817, and rs2975760 in molecular diagnosis.

Further analysis of the anthropometric and clinical parameters were compared in the subgroups of patients with genetic variants among the studied population, DENND1A (rs10818854), LHCGR (rs13405728), THADA (rs13429458), FSHR (rs2349415), CX37 (rs1764391), ACEI/D (rs1799752), INSR (rs1799817), and CAPN10 (rs2975760) wild type and hetero/homozygous carriers. The association analysis for determining the influence of the SNPs in BMI, lipid metabolism, and the parameters associated with glucose metabolism, the hormone levels associated with folliculogenesis, and ovarian steroidogenesis between the carriers and non-carriers of the study participants revealed significant differences for triglycerides levels in INSR (rs1799817), dehydroepiandrosterone sulfate (DHEAS) levels in DENND1A (rs10818854), THADA (rs13429458), FSHR (rs2349415), and CAPN10 (rs2975760), LH/FSH levels in DENND1A (rs10818854).

DISCUSSION

The present study is the first to investigate the association of polymorphisms impacting ovarian steroidogenesis (DENND1A, LHCGR, FSHR, and THADA) folliculogenesis (ACE, CX37), and insulin signaling (INSR, CAPN10) in South Indian women with PCOS. It is well-documented that PCOS women are more vulnerable to health problems like

diabetes, hypertension, cardiovascular disorders, anovulation, infertility, difficulties in conception, and adverse pregnancy outcomes.^[42,43] Our results have shown a statistically significant association of higher BMI ($P = 0.01$) and elevated levels of triglycerides ($P = 0.01$) with PCOS. Our findings are in line with the previous studies done by Thathapudi *et al.*^[44] in South Indian women which shows the BMI to be statistically significant ($P < 0.01$).^[45,46] In the present study, 85% of the PCOS patients are overweight (BMI >23 kg/m²).^[47] However, Haider *et al.*^[48] and Zhang *et al.*^[49] reported no significant differences in the BMI. Similar to the present findings, Macut *et al.*^[50] have also reported elevated triglycerides levels in PCOS women. The characteristic dyslipidemia profile associated with insulin resistance is the most common metabolic abnormality in PCOS. The blood levels of DHEAS appear to be slightly higher in women with PCOS.^[51]

In the present study, the prevalence of rs10818854 polymorphism in DENND1A is observed to be significantly high in PCOS patients ($P = 0.001$) when compared with the control. This variant is present in the intron of the DENND1A gene and it is similar to the related intronic gene variants, which may influence disease association possibly by introducing structural alterations within the upstream or downstream chromosomal regions.^[14] Shi *et al.*^[7] also reported a correlation between rs10818854 variant allele and elevated PCOS risk in GWAS 1. Cui *et al.*,^[52] Goodarzi *et al.*,^[53] and Welt *et al.*^[54] also identified an increased risk which is similar to the present study but not in accordance with the study done by Gammoh *et al.*^[55] In essence, it may be concluded that DENND1A rs10818854 polymorphism may increase the risk of PCOS in the South

Table 1: Baseline characteristics of the control and PCOS women

Parameter	Control (n=10)	Patients (n=20)
Age (years)	26.9±6.34	26.9±5.46
BMI (kg/m ²)	23.4±4.7	30.4±6.4**
RBS (mg/dL)	107.1±11.9	109.5±41.3
T.CHO (mg/dL)	159.5±31.65	175.7±29.8
TGL (mg/dL)	109.28±25.2	205.9±77.6**
HDL (mg/dL)	33.9±7.48	41.4±11.4
LDL (mg/dL)	83.57±8.26	92.7±26.4
TSH (mIU/mL)	2.96±0.955	4.2±3.8
FT4 (ng/dL)	1.1±0.321	1.2±0.4
LH (mIU/mL)	7.25±1.9	11.0±7.0
FSH (mIU/mL)	4.9±1.74	5.8±1.8
Prolactin (ng/mL)	10.7±3.07	15.2±7.6
E2 (pg/mL)	88±15.8	80.1±48.1
DHEAS (µg/dL)	185.8±63.3	266±128.9*

Values are mean±SD of triplicate experiments. *Significant at $P < 0.05$, ** significant at $P < 0.01$. BMI-Body mass index; RBS-Random blood sugar, T.CHO-Total cholesterol; TGL-Triglyceride levels; HDL-High density lipid; LDL-Low density lipid; TSH-Thyroid stimulating hormone; FT4-Thyroxine; LH-Luteinizing hormone; FSH - Follicle-stimulating hormone; E2-Estradiol; DHEAS - Dehydroepiandrosterone sulfate

Table 2: Genotypic and allelic frequencies of PCOS susceptible gene polymorphisms

	Cases				Control				χ^2 (P)	HWE χ^2 (P)
	Genotype % (n)	Allele %	Genotype % (n)	Allele %	Genotype % (n)	Allele %				
DENND1A (rs10818854)	GG 90 (18)	G 95	GG 100 (10)	G 100	GA+AA 10 (2)	A 5	GA+GG 0 (0)	A 0	13.76 (0.001*)	2.065 (0.356)
LHCGR (rs13405728)	AA 50 (10)	A 75	AA 50 (5)	A 75	AG+GG 50 (10)	G 25	AG+GG 50 (5)	G 25	1.667 (0.197)	2.297 (0.130)
THADA (rs13429458)	AA 80 (16)	A 90	AA 80 (8)	A 85	AC+CC 20 (4)	C 10	AC+CC 20 (2)	C 15	1.071 (0.301)	1.154 (0.183)
FSHR (rs2349415)	AA 10 (2)	A 40	AA 0 (0)	A 15	AG+GG 90 (18)	G 60	AG+GG 100 (10)	G 85	2.857 (0.091)	11.627 (0.001*)
CX37 (rs1764391)	CC 70 (14)	C 82.5	CC 50 (5)	C 70	CG+GG 30 (6)	G 17.5	CG+GG 50 (5)	G 30	2.500 (0.114)	8.64 (0.003*)
ACEI/D (rs1799752)	II 30 (6)	I 57.5	II 10 (1)	I 25	ID 55 (11)	D 42.5	ID 30 (3)	D 75	3.333 (0.504)	1.568 (0.457)
INSR (rs1799817)	CC 45 (9)	C 70	CC 20 (2)	C 45	CT+TT 55 (11)	T 30	CT+TT 80 (8)	T 55	2.222 (0.329)	12.150 (0.001*)
CAPN10 (rs2975760)	TT 65 (13)	T 82.5	TT 80 (8)	T 90	TC+CC 35 (7)	C 17.5	TC+CC 20 (2)	C 10	1.071 (0.301)	1.875 (0.171)

*Significant at $P < 0.05$; χ^2 : Pearson's Chi-square test to assess the relative association between the patients and controls for genotype and allele frequencies; HWE χ^2 indicates the result from the Hardy-Weinberg distribution of genotypes in the PCOS and control groups; DENND1A - DENN domain containing 1A; LHCGR - Luteinizing hormone/chorionic gonadotropin receptor. FSHR - follicle-stimulating hormone receptor; CX37 - Connexin37; ACE I/D - Angiotensin-converting enzyme insertion/deletion polymorphism; INSR - Insulin receptor; CAPN10 - Calpain-10

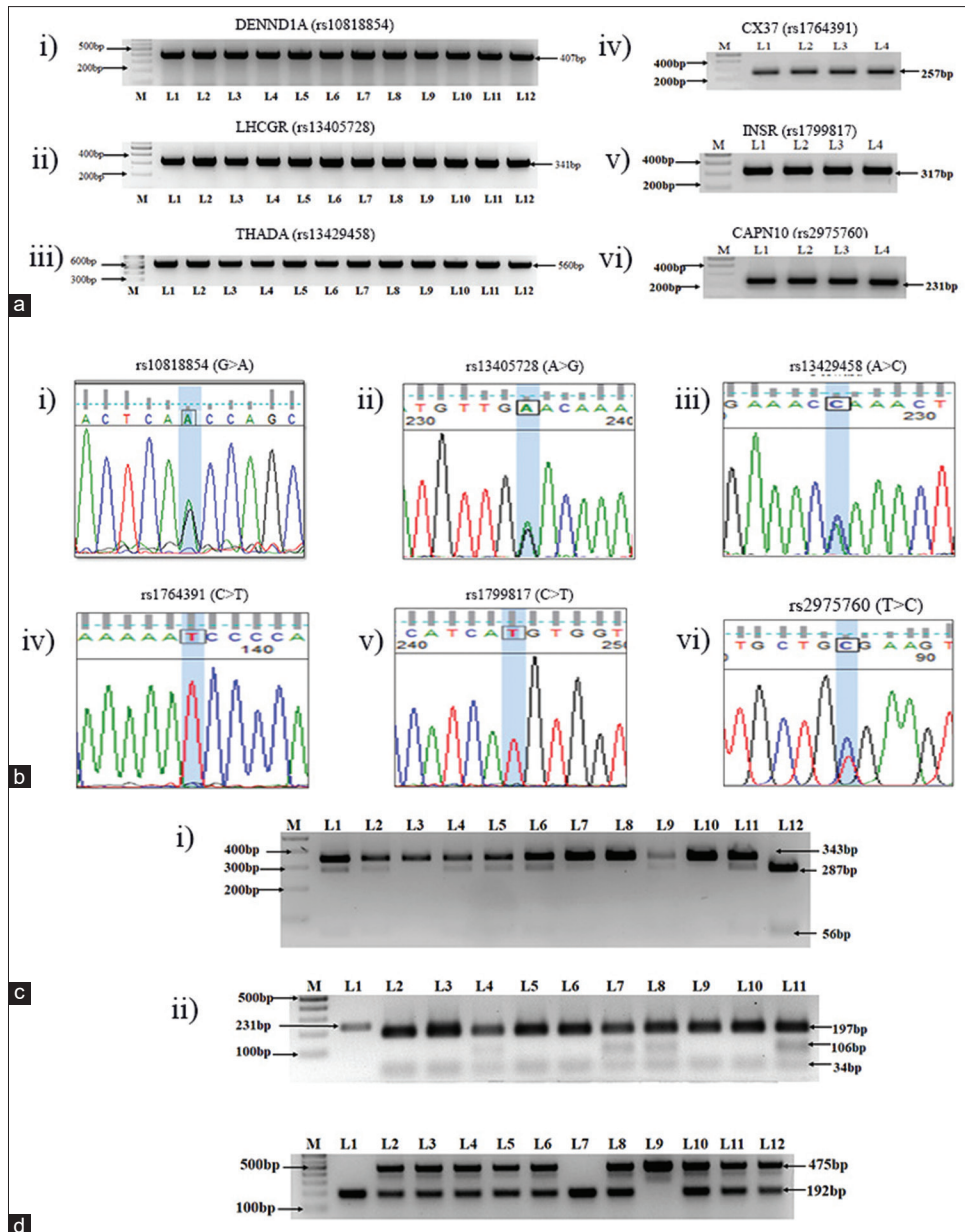


Figure 2: Determination of the incidence of the studied SNPs in controls and PCOS patients (a) Separation of PCR product encompasses SNP, electrophoretically in a 2% agarose gel (b) Chromatographs show the variants of study genes (c) RFLP analysis (i) rs2349415 SNP of FSHR with BspPI enzyme (ii) rs2975760 SNP of CAPN10 with Fnu4HI enzyme (d) Analysis of the insertion/deletion polymorphism of the ACE gene.

Indian population. We examined the phenotypic correlations of the tested DENND1A variants with associated PCOS features. In the present study, we have confirmed that there is a positive association with the level of DHEAS ($P = 0.01$). McAllister *et al.*^[15] reported that the intronic variants in DENND1A lack apparent functions that would favor splicing of V2 over V1 and these DENND1A V2 are overexpressed in the PCOS theca cells. These differential expressions are correlated to increased androgen (DHEAS) production, which is a measure of the hyperandrogenism associated with PCOS. The increased LH/FSH level that we observed in the DENND1A variant carriers suggests that DENND1A also plays a role in the regulation of gonadotropin secretion.^[11] Apparently, the CC genotype

in rs13429458 of the THADA gene and the GG genotype in rs2349415 of the FSHR gene are also significantly associated with the elevated level of DHEAS. Our results are consistent with the previous studies showing that the triglyceride levels are increased in the context of insulin resistance.^[56] To the best of our knowledge, this is the first study from South Tamil Nadu, India, providing evidence for DENND1A (rs10818854) polymorphism, which is positively correlated with an elevated level of DHEAS and draws attention to the fact that the PCOS patients with DENND1A polymorphism confer an increased risk. Identifying the genetic variants in the regional population would help to predict the predominant clinical manifestations in PCOS patients. This can be a useful tool to evaluate the

genotype and phenotype correlations that will help in further clinical manifestation and decide the treatment strategy. The main limitation of this study is the small number of participants, the patients and controls did not have comparable BMI because our aim was predominantly to look at the relevance and association of the genetic variants/SNPs with PCOS in the regional population. Although the differences in various populations observed can be attributed to diverse ethnic and geographic differences, our study might be considered as a preliminary analysis which can be further proved strongly with larger cohort size.

CONCLUSION

This is the first study from South Tamil Nadu, India, which has reported that the DENND1A (rs10818854) polymorphism may be associated with the risk of PCOS and is positively correlated with an elevated level of DHEAS. This SNP may be a useful marker in determining the genetic susceptibility to the pathogenesis of PCOS. INSR (rs1799817) genotype shows association with an elevated level of triglycerides and rs13429458 of THADA, rs2349415 of FSHR which is also significantly associated with high levels of DHEAS while, rs13405728 of LHCGR, rs1764391 of CX37, rs1799752 of ACE, and rs2975760 of CAPN10 did not show any significant association. Studies on genetic predisposition to PCOS in the regional population would help to predict the predominant clinical manifestations, and to evaluate the genotype and phenotype correlations that help in further clinical manifestation in the PCOS patients. Further, large-scale studies in the regional population may offer better insight into the analyzed susceptible gene variants and their potential role in the incidence of PCOS.

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Conflicts of interest

There are no conflicts of interest.

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