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Polycystic ovary syndrome: role of androgen excess self-assessment in diagnosis

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POLYCYSTIC OVARY SYNDROME: ROLE OF ANDROGEN EXCESS SELF-ASSESSMENT IN DIAGNOSIS

by

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POLYCYSTIC OVARY SYNDROME: ROLE OF ANDROGEN EXCESS SELF-ASSESSMENT IN DIAGNOSIS

PASCALINE WANJIRU KARANJA

ABSTRACT

Background: Polycystic ovary syndrome is the most common endocrine disorder affecting reproductive-aged women. It is diagnosed using a combination of menstrual irregularity, clinical and/or biochemical hyperandrogenism and polycystic ovary morphology upon ultrasound. Hyperandrogenism in females may clinically manifest as hirsutism, acne, alopecia, or other masculinization of features. Assessing total/free testosterone, dehydroepiandrosterone sulfate, and 17-hydroxyprogesterone provides biochemical evidence of hyperandrogenism.

Objective: To determine self-reported clinical signs of androgen excess using data from the Ovulation and Menstruation Health (OM) Study, a diverse, multi-ethnic cohort study being conducted at Boston University School of Medicine.

Methods: Data was collected from participants enrolled in the Ovulation and Menstruation Health Study pilot cohort. This epidemiologic survey captured demographics, menstrual cycle patterns, PCOS histories, reproductive histories and manifestations of androgen excess in a diverse patient population. Participants were women ages 18-45 who had the capacity to ovulate/menstruate at the time of the study, had no history of chemotherapy, radiation, or surgical menopause, and were not pregnant at the time of the study. To assess androgen excess, participants were asked to self-report hair growth in nine body areas, acne on the face and back and hair loss on the scalp. The

nine body areas were scored using the modified Ferriman-Gallwey (mFG) scoring system. Reference images created by a medical illustrator were used for hirsutism and alopecia grading while clear descriptions were provided for grading acne severity. Clinical hirsutism was defined as total mFG score of ≥ 8, or ethnic specific cutoff for East Asian (≥ 2) and Southeast Asian (≥ 3) women. Alopecia was defined as scalp hair loss ≥ 2. For participants that consented to medical record validation total, free and bioavailable testosterone lab levels were assessed for biochemical hyperandrogenism evaluation.

Results: Beginning August 9, the day the study opened to the public, 249 participants completed the pilot survey questionnaire. These participants were 66.8% white (n=165), 6.5% Hispanic or Spanish origin (n=16), 10.5% Black or African-American (n=26), 1.6% East Asian (n=4), 2.0% Southeast Asian (n=5), 2.4% South Asian (n=6), and 10.9% were of mixed ethnic backgrounds (n=27). 22.5% (55/245) of these women had clinical hirsutism by total mFG score. Mean total mFG scores were highest in women who were South Asian at 13.8±9.1 (n=6) and Hispanic at 8.6±8.7 (n=16). Moderate-severe acne was reported in 23.6% (58/246) of respondents, 24.8% (41/165) of white women, 26.7% (4/15) of Hispanic women, 15.4% (4/26) of Black women, 0.0% (0/4) of East Asian women, 20.0% (1/5) of Southeast Asian women, 50% of South Asian women (3/6) and 20% (5/25) of women of mixed ethnicities. 9.4% (23/246) of all pilot women reported alopecia, highest in Black (26.9%, 7/26) and East Asian women (25%, 1/4). Among women that had a PCOS diagnosis there was a higher presence of clinical hirsutism, higher acne severity, and higher prevalence of alopecia when compared to non-PCOS

women. In addition, 33%(4/12) of the 44 women that consented to medical record validation had total testosterone levels above the normal range.

Conclusions: This pilot population demonstrated an ethnic dependent pattern of development for hirsutism, acne and alopecia. Additionally, women who had a PCOS diagnosis were more likely to report having the clinical signs of androgen excess than those without a diagnosis.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF ABBREVIATIONS

17OHP 17-hydroxyprogesterone

AE-PCOS ..... Androgen Excess and PCOS society

BMC ..... Boston Medical Center

BUSM .....Boston University School of Medicine

CCCA..... Central Centrifugal Cicatricial Alopecia

CLIAs.....Chemiluminescence immunoassays

DHEA .....Dehydroepiandrosterone

DHEA-S .....DHEA Sulfate

DHT .....Dihydrotestosterone

ELISA ..... Enzyme-linked immunosorbent assay

FT ..... Free testosterone

GDM ..... Gestational diabetes mellitus

GnRH ..... Gonadotropin-releasing hormone agonists

LC-MS/MS .....Liquid chromatography-tandem mass spectrometry

mFG .....modified Ferriman-Gallwey

NCCAH ..... Nonclassic Congenital Adrenal Hyperplasia

OM ..... Ovulation and Menstruation

PCOM .....Polycystic Ovary Morphology

PCOS ..... Polycystic Ovary Syndrome

RIAs ..... Radioimmunoassays

SHBG ..... Sex hormone-binding globulin

TT.....Total Testosterone

INTRODUCTION

Polycystic Ovary Syndrome

Polycystic Ovary Syndrome (PCOS) is one of the most common endocrine and metabolic disorders in reproductive-aged women. The etiology of PCOS is complex since both genetic variants and environmental factors play a role in its development (Azziz, 2017). This disorder affects 6-10% of women globally, however its prevalence varies based on which diagnostic criteria is used when making the diagnosis (Azziz, 2017). There is also an increased prevalence of PCOS in women with oligoovulatory infertility, obesity and/or insulin resistance, either type of diabetes, first-degree relatives with PCOS, premature adrenarche history, use of antiepileptic drugs, and those of certain ethnicities (Azziz, 2017). The three diagnostic criteria most commonly used today are the NIH criteria developed in 1990, Rotterdam criteria developed in 2003, and the Androgen Excess and PCOS society (AE-PCOS) criteria developed in 2006 (Table 1)(Sirmans & Pate, 2013). The most current Guidelines by the Endocrine Society recommend the use of the Rotterdam Criteria for defining PCOS (Legro et al., 2013). This includes the diagnosis of at least two of the following: 1) clinical or biochemical hyperandrogenism, 2) ovary dysfunction resulting in menstrual irregularity, or 3) polycystic ovary morphology on ultrasonography (The Rotterdam ESHRE/ASRM, 2004).

Table 1. Criteria for Polycystic Ovary Syndrome. Adapted from Sirmans & Pate, 2013

1990 NIH Criteria (Include all of the following)	**2003 Rotterdam Criteria** (Include 2 of the following)	**2006 AE-PCOS Criteria** (Include all of the following)
• Clinical and/or biochemical hyperandrogenism • Menstrual dysfunction	• Clinical and/or biochemical hyperandrogenism • Oligo-ovulation or anovulation • Polycystic ovaries	• Clinical and/or biochemical hyperandrogenism • Ovarian dysfunction and/or polycystic ovaries

Patients qualify for a hyperandrogenism diagnosis if they have clinical manifestations and/or biochemical evidence of androgen excess. Hyperandrogenism can present clinically as hirsutism, acne, or alopecia, with hirsutism being the most commonly used clinical sign of androgen excess (Lizneva et al., 2016). Biochemical evidence of hyperandrogenism, which is referred to as hyperandrogenemia, includes elevated serum androgen concentrations as determined by specific lab based thresholds (Lizneva et al., 2016). Most women with PCOS actually have both clinical signs and biochemical evidence of hyperandrogenism (Barbieri & Ehrmann, 2018a). Menstrual irregularity includes either infrequent ovulation (oligoovulation) causing menstrual patterns that consist of either less than 9 menstrual periods in a year or absent ovulation (anovulation) causing no menstrual periods for 3 or more consecutive months (Barbieri & Ehrmann, 2018a). Polycystic Ovary Morphology (PCOM) is the abnormal appearance of the ovaries upon ultrasound imaging. Despite the name, this abnormal appearance is determined based on follicular number and size, not cysts (Barbieri & Ehrmann, 2018a).

These clinical manifestations of PCOS do not only negatively impact the quality of life of the affected patients, but also cause a significant burden on society's health care

resources (Tan et al., 2013). In 2005 the economic burden of PCOS in the United States (U.S.) was calculated to be approximately $4.37 billion annually (Azziz et al., 2005). This burden is largely associated with the increased prevalence of diabetes associated with PCOS (40%), hirsutism (14%), menstrual dysfunction treatments (31%), and use of infertility services (12%) (Azziz et al., 2005). Furthermore, the clinical manifestations of hyperandrogenism alone causes an economic burden estimated at more than $600 million in the U.S. annually (Bode et al., 2012). PCOS is also associated with other long-term health implications such as metabolic syndrome making it a significant public health concern.

Long-term health outcomes associated with PCOS

There are various metabolic issues associated with PCOS. At least half of women with PCOS are also obese, which contributes to worse clinical manifestations of the disorder (Barbieri & Ehrmann, 2018c). Most women with PCOS also have insulin resistance or hyperinsulinemia, which can in turn contribute to hyperandrogenism both directly and indirectly (El Hayek et al., 2016; Barbieri & Ehrmann, 2018c). Hyperinsulinemia can stimulate androgen biosynthesis in the ovarian theca cell and can also have suppressive effects on SHBG production by the liver (Barbieri & Ehrmann, 2018c). Women with PCOS also have a much higher prevalence of metabolic syndrome than the normal population (Sirmans & Pate, 2013). Additionally PCOS puts women at increased risk for nonalcoholic fatty liver disease, impaired glucose tolerance or type II diabetes, sleep apnea, mood disorders, and small dense LDL particles associated with

increased risk of coronary heart disease (Randeva et al., 2012). All these metabolic issues are risk factors for cardiovascular disease (Randeva et al., 2012).

Additionally, when women with PCOS conceive, either naturally or through ovulation induction therapy, the risk of pregnancy complications is increased (Qin et al., 2013). These complications include early pregnancy loss, gestational diabetes mellitus (GDM), pregnancy induced hypertension, preeclampsia, and preterm birth (Qin et al., 2013). For this reason it is important that women with PCOS receive proper screening and treatment interventions to either prevent the occurrence or properly manage these negative health outcomes (Legro et al., 2013; Sirmans & Pate, 2013).

Clinical Manifestations of PCOS and their Diagnosis

Hyperandrogenism

Hyperandrogenism is a common endocrine disorder with a prevalence of 5-10% in the general population (Yildiz, 2006). Clinical signs of hyperandrogenism may include hirsutism, acne, alopecia, and virilization in more severe cases (Yildiz, 2006). However, virilization is rare and often suggests an androgen-secreting tumor (Yildiz, 2006). Hyperandrogenism can present clinically without biochemical evidence of hyperandrogenemia and vice versa. Therefore either clinical manifestations or biochemical evidence of excess androgen can be used to diagnose hyperandrogenism (Yildiz, 2006). Additionally, patient history and physical examination are necessary for the evaluation and diagnosis of hyperandrogenism.

Hirsutism

Hirsutism is the most commonly used clinical diagnostic criteria for hyperandrogenism. About 80% of patients with hyperandrogenism also present with hirsutism (Yildiz, 2006). It is prevalent in 7% of women in the general population and is marked by male-pattern excessive terminal hair growth (Yildiz, 2006). Male-pattern growth is deemed the appropriate terminology because men have higher androgen levels, and therefore higher degrees of terminal hair growth in sex-specific areas comparable with what is seen in these women. Androgens determine the type and distribution of hair growth and act on sex-specific areas of the body to convert the small, straight vellus hairs to long, coarse and thick terminal hairs (Bode et al., 2012; Yildiz, 2006). Therefore excess androgen causes excessive growth of terminal hairs in these areas (Bode et al., 2012; Yildiz, 2006)(Figure 1). The most common cause of hirsutism is actually PCOS, accounting for 75% of hirsutism cases (Bode et al., 2012). Other causes of hirsutism include some androgenic medications and other endocrinopathies such as non-classic congenital adrenal hyperplasia, thyroid dysfunction, Cushing syndrome, acromegaly, hyperprolactinemia, androgen-secreting tumors, and neoplasms (Bode et al., 2012; Yildiz, 2006). However, the chance of neoplasm being the cause is rare as it only accounts for 0.2% of hirsutism cases. This form of hirsutism is often associated with rapid onset of symptoms, virilization, and palpable abdominal or pelvic mass (Bode et al., 2012). Hirsutism may also arise spontaneously; 20% of hirsutism cases are caused by

idiopathic hyperandrogenemia associated with normal ovulation cycles and androgen excess. (Bode et al., 2012).

In addition to a physical examination, a medical and family history should be obtained to diagnose hirsutism (Bode et al., 2012). The physical examination includes an evaluation of the patient's body hair to determine if the excess hair is terminal or vellus hair and whether it is growing in a male-pattern (Yildiz, 2006). The physician must also distinguish between hypertrichosis, which is excess hair growth all over the body that is not a result of hyperandrogenism. (Yildiz, 2006) Those determined to have a severe rapid onset of hirsutism should undergo further workup to eliminate or diagnose a possible androgen-secreting tumor (Bode et al., 2012).

The physical evaluation of hirsutism is most commonly done using the modified Ferriman-Gallwey (mFG) score. This is a pictorial scale that shows the distribution and degree of the hair growth. This subjective scale can either be used by the patient for self-assessment or used by a clinician. The presence of hair growth in 9 androgen-sensitive body sites is scored on a scale of 0 to 4. These 9 areas are the upper lip, chin, chest, upper abdomen, lower abdomen, upper back, lower back, upper arms and thighs. A score of 0 indicates an absence of terminal hair growth while a score of 4 indicates excessive terminal hair growth. An individual can have a total mFG score ranging from 0 to 36 (Bode et al., 2012). Typically the cutoff for a hirsutism diagnosis is an mFG score of ≥8 (Bode et al., 2012), however this cutoff may vary depending on ethnicity (Table 2). In summary, this mFG scoring system indicates that individuals with an mFG score less than 8 are considered normal, those with a score between 8 and 15 are considered to have

mild hirsutism and those with a score greater than 15 are considered to have moderate or severe hirsutism (Bode et al., 2012).

Table 2. Ethnic specific Hirsutism Cut-offs. *Summary of studies regarding ethnic specific cut-offs for mFG score.*

Ethnicity	Suggested mFG cut-off	Author, year
Caucasian	≥8	DeUgarte et al., 2006
Hispanic, Latina, or Spanish Origin	≥8	DeUgarte et al., 2006
Black or African American	≥8	DeUgarte et al., 2006
East Asian	≥2	Yildiz et al., 2010
Southeast Asian	≥3	Cheewadhanaraks, 2004
South Asian	≥6	Zargar, 2002

Acne

Acne vulgaris is one of the most common visible skin disorders for individuals between the ages of 15 and 40 (Franik et al., 2018). Although acne is largely known to affect 70% of adolescents during puberty, it is important to note that adults are also affected (Franik et al., 2018; Gold et al., 2009). It is prevalent in about 15% of all age groups (Uysal et al., 2017), with 4% being adult men and 13% being adult women (Tan et al., 2013). During puberty the increased adrenal androgen production is usually accompanied by increased acne severity, which suggests that acne is associated with increased androgen levels (Yildiz, 2006). Additionally, androgens cause sebaceous glands to enlarge increasing sebum production, black heads, and inflammation on the face, neck, chest, shoulders and back (Franik et al., 2018; Yildiz, 2006). Hormonal disorders, such as androgen excess, can cause increased acne severity and should therefore be evaluated (Uysal et al., 2017). This in turn explains the increased acne severity in women with PCOS who also present with hyperandrogenism (Franik et al., 2018).

Although there are many different methods for assessing the severity of acne, there is no universally agreed upon grading system. Most of these methods involve evaluating and counting the number, type and distribution of the lesions (Yildiz, 2006). The types of blackheads present, amount of inflammation, areas of the body affected and how widespread the acne is are all also evaluated when grading acne. The severity type is defined as mild, moderate, or severe (Pochi et al., 1990). Although there is no universally used acne global severity scale, diagnosis commonly occurs using clear descriptions of the different acne categories (Table 3) (Gold et al., 2009).

Table 3. Types of Acne. Adapted from Gold *et al*., 2009

Acne Type	Description of Condition
No acne	Total absence of acne and blemishes
Subclinical acne	Small number of blackheads and whiteheads; barely visible; first sign of blemish
Comedonal acne	Blackheads and whiteheads (slightly inflamed—may be red); blemishes are visible
Mild acne	• Several inflamed pimples—red in color • Less than 20 whiteheads/blackheads or less than 15 inflammatory (red) lesions (pimples) or less than 30 total lesions (pimples) not all inflamed (red in appearance)
Moderate Acne	• Many inflamed pimples (red in color) and pustules (visible accumulation of pus in skin) • 20 to 100 whiteheads/blackheads or 15 to 50 inflammatory (red) lesions (pimples) or 30 to 125 total lesions (pimples) not all inflamed (red in appearance)
Severe nodular acne	• Inflamed pimples and pustules (visible accumulation of pus in skin) with a few deep nodular lesions (solid mass can be felt under skin—can sometimes be raised) • Greater than 5 cysts (solid mass of skin like a knot, can be raised or felt under the skin) or total whiteheads/blackheads count greater than 100 or total inflammatory count greater than 50 or greater than 125 total lesions
Severe cystic acne	Many nodular cystic lesions (with signs of scarring)

Alopecia

Alopecia is the loss of scalp hair and is also known as a male-pattern temporal form of balding (Yildiz, 2006; Barbieri & Ehrmann, 2018c). Although there are several causes of alopecia, androgenic alopecia is the most common cause (Yildiz, 2006). Androgenic alopecia was reported in 67% of women with PCOS (Lizneva et al., 2016). Androgens can affect different stages of the hair follicle growth cycle and different areas of the body. This multi-component effect plays a role in determining whether hair growth or hair loss will occur. Although scalp follicles typically have the longest anagen phases, the phase that primarily determines the length of hair, increased androgen results in a shorter anagen phase (Randall, 2008) (Figure 1). This effect leads to hair loss by replacing the scalp's thick, pigmented terminal hairs with smaller hair follicles and short non-pigmented vellus hairs (Yildiz, 2006) (Figure 2). Androgenic alopecia is not commonly used as a marker of hyperandrogenism. However, if androgenic alopecia also presents with a history of irregular menses, hirsutism, acne, or virilization then an evaluation of hyperandrogenism should be done (Yildiz, 2006).

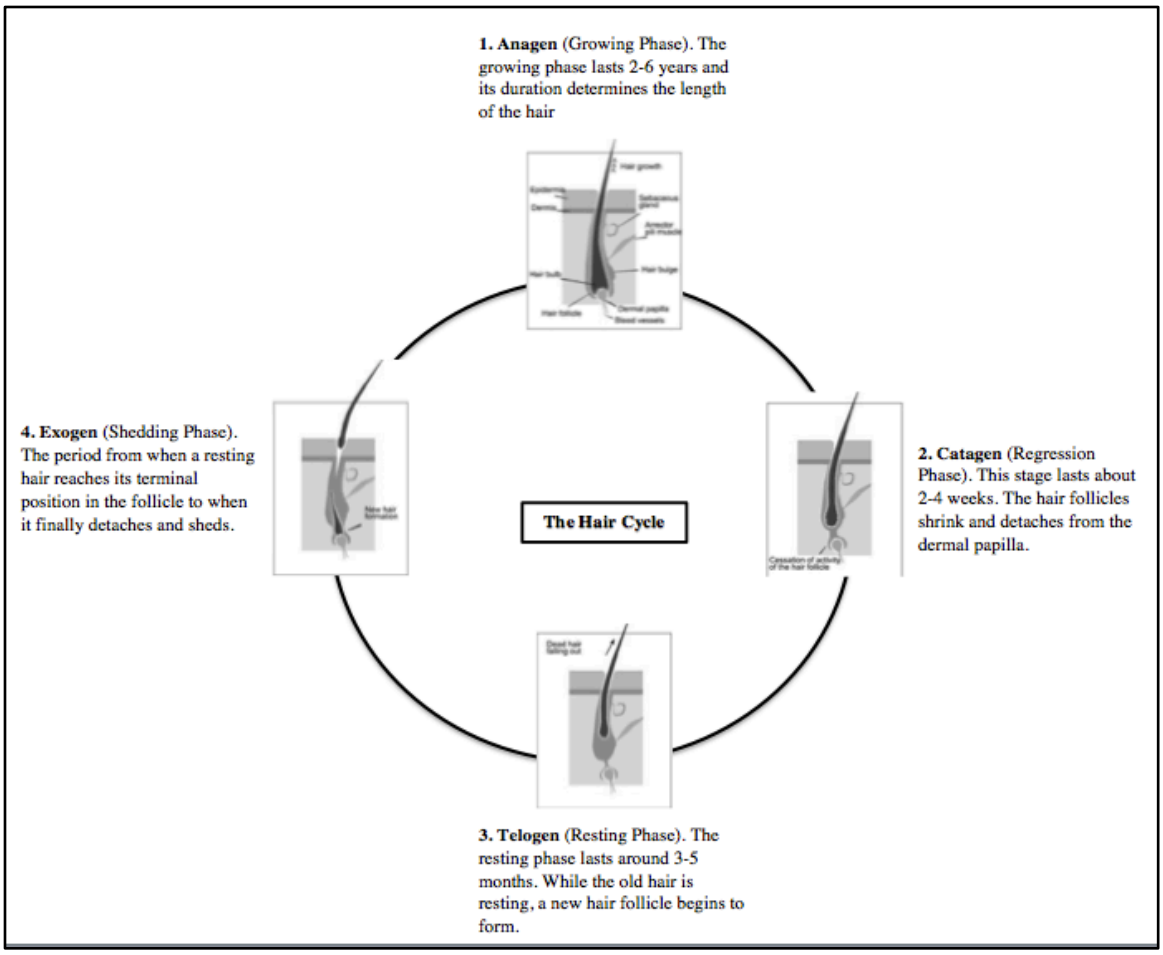


Figure 1. The Hair Cycle. (*Adapted from Hair Again, 2017 and Welch, 2018).*

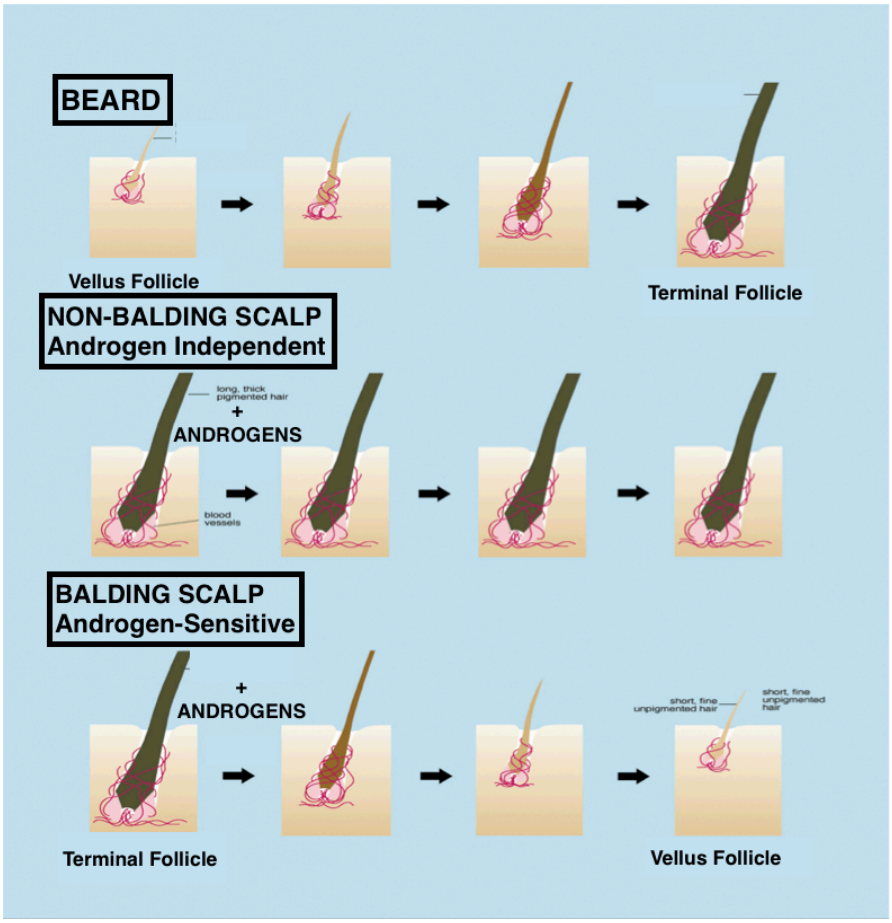


Figure 2. Effect of Androgen on hair follicles *(Adapted from Randall, 2008) Androgen stimulates hair growth in sex-specific areas, but has the opposite effect in scalp areas.*

Evaluation of alopecia includes conducting a hair-pull test to detect hairs in the telogen phase, examining for the presence of hirsutism or acne and assessing the hair loss pattern (Lizneva et al., 2016). The 3 common hair loss patterns are Ludwig's type, Christmas tree, and Hamilton type (Herskovitz & Tosti, 2013). There are seven hair loss classification systems that have been proposed by various researchers for grading female pattern hair loss (Gupta & Mysore, 2016) (Table 4). The hair loss pattern is typically determined using the Ludwig scale or Christmas tree (Olsen) classification systems (Lizneva et al., 2016).

Table 4. Female pattern hair loss classification systems

System	Year and Source	Description
Ebling and Rook	1975 Gupta & Mysore, 2016	• 5-stage system • Centrifugal hair loss with recession of the frontotemporal hair line
Ludwig System	1977 Ludwig, 1977	• 3-stage system • Centrifugal hair loss while preserving the hair line
Savin Scale	1992 Savin, 1992	• 9 computer images • 8 stages of increasing crown balding and 1 image for frontal anterior recession
Olsen	1994 Olsen, 1994	• Similar to Ludwig, but includes frontal-vertical alopecia (Christmas tree pattern)
Bouhana*	2000 Bouhana, 2000	• Multifactorial classification including extent of balding, degree of elasticity, thickness of the scalp, covering power of hair based on density, caliber, shape, length, growth rate, and hair color
Sinclair	2004 Sinclair et al., 2004	• Self-report photographic measure with 5 images • Images include hair parted centrally • Image 1 has no hair loss and severity of hair loss increases in the following images
Basic and specific classification*	2007 Lee et al., 2007	• 4 Basic types represent shape of anterior hair line • 2 Specific types represent density of hair on frontal and vertex areas • Each type includes subtypes of severity • Combination of basic and specific types determines final type

* Used for both female and male hair loss classification

Virilization

Virilization is caused by an overproduction of androgens. It typically results from PCOS or a tumor. Virilization is characterized by androgenic alopecia, amenorrhea, deepening of the voice, increased muscle mass, decreased breast size, and clitoromegaly (Yildiz, 2006). Clitoromegaly is defined as a clitoral index greater than 35mm (Yildiz, 2006). Virilization may occur in patient's experiencing severe or rapid hyperandrogenism. However, it is more commonly associated with an androgen-secreting neoplasm, particularly in cases of rapid onset of symptoms (Yildiz, 2006).

Biochemical Hyperandrogenism

Androgens are hormones primarily produced in the adrenal glands and secondarily in the ovaries. These hormones include, but are not limited to, testosterone, dihydrotestosterone (DHT), dehydroepiandrosterone (DHEA), DHEA Sulfate (DHEA-S), and androstenedione.

For individuals with moderate or severe hirsutism, biochemical tests for androgen excess are usually conducted (Bode et al., 2012). Biochemical tests are done using a variety of assays to test for serum levels of total testosterone (TT), free testosterone (FT), androstenedione, and DHEA-S (Agapova et al., 2014). Serum TT is the best estimate of androgen production in hirsute women (Martin et el., 2018). Although plasma FT is 50% more sensitive than TT, it is expensive and not widely available (Bode et al., 2012). FT tests should only be considered if TT is only moderately elevated (Bode et al., 2012). In women with hyperandrogenic symptoms but normal menstrual cycles, literature suggests measuring only TT levels (Barbieri & Ehrmann, 2018a). For those with hyperandrogenic

symptoms and oligomenorrhea, serum TT, 17-hydroxyprogesterone (17OHP) and other routine labs should be run to identify the cause of the irregular menses (Barbieri & Ehrmann, 2018a). These tests are important since 22 to 85% of women with PCOS have elevated serum androgen levels (Azziz et al., 2009).

When measuring TT the normal limit is between 45-60 ng/dL (1.6 to 2.1 nmol/L), therefore a serum TT level above 60 ng/dL is elevated (Barbieri & Ehrmann, 2018a). If serum TT is over 150 ng/dL, an evaluation for causes such as adrenal-secreting tumors must be conducted due to this extremely high value (Barbieri & Ehrmann, 2018b). These reference values may depend upon the specific laboratory and methodology used. Women with PCOS, on the other hand, may present with either normal or elevated serum TT (The Rotterdam ESHRE/ASRM, 2004). Special considerations must be made when measuring 17-OHP. This hormone must be measured in the morning, since levels are highest during this time, and within the early follicular phase for women that are still ovulating (Martin et al., 2018). For those without cycles, the latter consideration is not necessary and measurements can be taken at any point (Martin et al., 2018). This test is used to eliminate a nonclassic congenital adrenal hyperplasia (NCCAH) diagnosis since 17-OHP levels greater than 200 ng/Dl suggest that the patient has NCCAH (Legro et al., 2013).

In addition to biochemical tests, there are various assays to measure serum TT such as radioimmunoassays (RIAs), enzyme-linked immunosorbent assay (ELISA), chemiluminescence immunoassays (CLIAs) and liquid chromatography-tandem mass spectrometry (LC-MS/MS) (Rosner et al., 2007). Although there is no standardized assay

to measure androgen in women, the Endocrine Society position statement suggests using LC-MS/MS, due to the fact that extraction and chromatography techniques increase sensitivity and accuracy as compared to direct immunoassays (Rosner et al., 2007; Agapova et al., 2014).

Ovulatory Dysfunction

The most common cause of ovulatory dysfunction is PCOS (El Hayek et al., 2016). Both oligoovulation and anovulation cause irregularities of the menstrual cycle resulting in either oligomenorrhea or amenorrhea, respectively (El Hayek et al., 2016; The Rotterdam ESHRE/ASRM, 2004). These irregularities can manifest in a number of different ways. For example, there can be normal or slightly delayed puberty onset followed by irregular cycles, or menstrual cycles that are regular at first but then become irregular (Barbieri & Ehrmann, 2018c). The infrequent ovulation makes it harder for women with PCOS to conceive or results in infertility if she experiences anovulation (Qin et al., 2013; Legro et al., 2013).

Polycystic Ovary Morphology (PCOM)

The ultrasound appearance of PCOS can be evaluated noninvasively using high frequency transvaginal ultrasonography (Legro et al., 2013). The 2003 Rotterdam criteria defines PCOM as 12 or more follicles present in at least one ovary that are 2-9 mm in diameter and/or an ovarian volume greater than 10mL (The Rotterdam ESHRE/ASRM, 2004). This criterion is also age-specific for women below 40 because follicle numbers and ovarian volume decrease with age (Barbieri & Ehrmann, 2018c). It is unnecessary to perform this evaluation for PCOS diagnosis if a woman is already known to have

menstrual irregularity and hyperandrogenism (Barbieri & Ehrmann, 2018a). The ultrasound should only be done if the woman has either hyperandrogenism, but borderline normal menstruation, or menstrual irregularity, but no evidence of hyperandrogenism (Barbieri & Ehrmann, 2018a). Figure 3 highlights the steps for PCOS diagnosis.

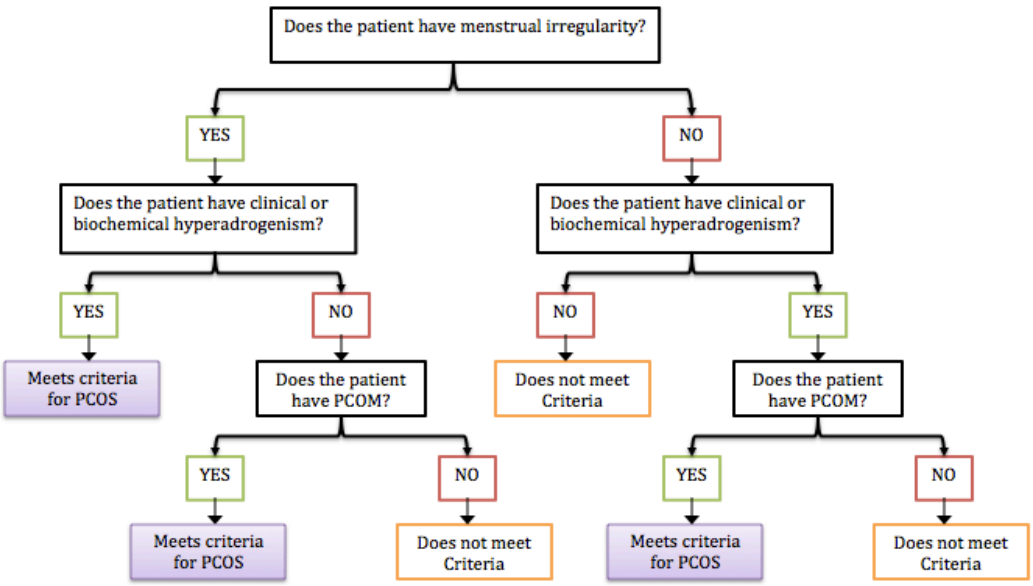


Figure 3. Flow Chart of Rotterdam Criteria to meet a PCOS diagnosis.

Diagnostic Challenges of PCOS and Hyperandrogenism

There are certain challenges that come with diagnosing PCOS and its clinical manifestations. First, when diagnosing PCOS, physicians must be careful to eliminate other disorders that mimic the clinical features of this disorder, such as thyroid disease, hyperprolactinemia, and NCCAH (Legro et al., 2013). The diagnostic criterion also has age-specific limitations. In adolescents anovulatory symptoms and PCOM may be normal

in stages of reproductive maturation and not necessarily an indication of PCOS (Legro et al., 2013). Therefore, PCOS diagnosis in adolescents should be based primarily on a hyperandrogenism diagnosis in the presence of persistent oligomenorrhea (Legro et al., 2013). In post-menopausal women, there is a normal presence of amenorrhea and decrease in follicle size further complicating the detection of PCOM (Barbieri & Ehrmann, 2018c). Therefore, in post-menopausal women the Endocrine Society suggests using a well-documented history of amenorrhea and hyperandrogenism during their reproductive years to make a PCOS diagnosis (Legro et al., 2013).

When diagnosing hirsutism, there are limitations in using the mFG scale because it is subjective and doesn't account for all androgenic areas. Furthermore, it does not take into consideration other possible causes of increased hair growth (Bode et al., 2012). When diagnosing hyperandrogenism, acne alone cannot be used. This is because most acne patients do not have hyperandrogenism and data regarding the prevalence of hyperandrogenism in acne patients is limited (Yildiz, 2006). However, if a patient has severe acne along with hirsutism or irregular menstrual periods then hyperandrogenism can be considered (Yildiz, 2006). It is also important to keep in mind that measuring serum androgen levels is not useful for those on estrogen-progestin oral contraceptives, metformin, or spironolactone (Barbieri & Ehrmann, 2018a).

Management and Treatment of PCOS

Early diagnosis and treatment are important for improving the quality of life for women with PCOS. However, most women report having a poor diagnostic experience

associated with long delays and inadequate health information (Barbieri & Ehrmann, 2018a). Lifestyle interventions such as weight loss and decreasing the degree of insulin resistance, or hyperinsulinemia with diet, exercise or medications has decreased androgen levels in women (Barbieri & Ehrmann, 2018c; Legro et al., 2013). Decreasing androgen levels improves some hyperandrogenic symptoms. Exercise, healthy diet and weight loss have been proven to reduce cardiovascular risk factors, diabetes, and reproductive dysfunction in the general population, and are thought to also work in improving these health outcomes in PCOS patients as well (Legro et al., 2013).

Treatment of the hyperandrogenic, clinical manifestations of PCOS involves suppressing androgen production and/or action through the use of hormonal contraceptives, glucocorticoids, insulin-lowering agents and gonadotropin-releasing hormone agonists (GnRH) (Lizneva et al., 2016).

Hormonal contraception is the first line of treatment for menstrual irregularity, hirsutism and acne (Legro et al., 2013). In adolescents with PCOS, hormonal contraception, lifestyle therapy, and metformin, a drug that helps lower blood glucose levels, are the recommended treatment options (Legro et al., 2013). The Endocrine Society suggests metformin be used only in women who also have Type 2 Diabetes or impaired glucose tolerance and as a second line of therapy for women who have menstrual irregularity but cannot take or cannot tolerate hormonal contraception (Legro et al., 2013).

To treat anovulatory infertility the Endocrine Society suggests the use of clomiphene citrate (Legro et al., 2013). This is a drug that stimulates the increase of

hormones that support ovulation. It should be used as the first-line treatment option with metformin as an adjuvant therapy to prevent ovarian hyperstimulation syndrome in PCOS patients undergoing in vitro fertilization (Legro et al., 2013).

Hirsutism treatment should be guided by the severity and the amount of distress it causes the patient (Bode et al., 2012). If the symptoms are mild and menses are normal, then hirsutism can be treated based on a clinical educated guess (Bode et al., 2012). If child bearing is not a current concern, then birth control should be the first line of treatment. Hormonal birth control is the best option as it increases sex hormone-binding globulin (SHBG), which therefore decreases the bioavailable testosterone levels and inhibits ovarian androgen production (Bode et al., 2012). In addition to oral contraceptives, various medications commonly used for the treatment of hirsutism include antiandrogens, insulin-lowering agents and eflornithine. Topical agents such as eflornithine can also be used for excessive hair growth on the face (Bode et al., 2012). GnRH agonists are used only for severe cases that are not responsive to other treatment methods. Ketoconazole may also be used when other therapies have failed (Bode et al., 2012). Regardless of which treatment the patient chooses, it is important to monitor the response to the medication for 6 months before making adjustments (Bode et al., 2012). As an alternative to medication, a lot of women with hirsutism opt to use various methods for cosmetic hair removal. These methods include shaving, chemical depilation, waxing or plucking, electrolysis and laser therapy. Shaving is effective if repeated often, however it is only temporary and the hair regrown is coarser (Bode et al., 2012). Chemical depilation can cause reactive dermatitis, waxing and plucking can cause scarring or

hyperpigmentation, electrolysis is only effective for smaller areas and laser therapy is expensive (Bode et al., 2012).

Acne treatments include both topical and systemic treatments (Lizneva et al., 2016). For mild to moderate acne over the counter benzoyl peroxide preparations and topical cleansers can be used (Gold et al., 2009). For severe acne types, topical or systemic antibiotics provided by health care providers can be used (Gold et al., 2009). Additionally, phototherapy with blue light has been shown to improve skin conditions both in vivo and clinically, which can eliminate the need for prescriptions and harsh chemicals (Gold et al., 2009). These treatments can be used in addition to medications that decrease androgen levels.

Alopecia treatments are typically topical and usually contain 2% topical minoxidil (Lizneva et al., 2016). The treatment involves the application of 1ml minoxidil to the scalp twice daily for at least 12 months (Herskovitz & Tosti, 2013). These treatments are used in addition to medications to decrease androgen levels. If these treatments are not effective then patients can also choose additional cosmetic alternatives such as hair transplantation, wigs, etc.

SPECIFIC AIMS

This thesis project seeks to determine self-reported clinical signs of androgen excess using data from the Ovulation and Menstruation Health (OM) Study, a diverse, multi-ethnic cohort study being conducted at Boston University School of Medicine.

Specifically, this thesis aims to:

1. Describe the distribution of hirsutism within the Ovulation and Menstruation Health Study pilot cohort.
2. Describe the distribution of acne within the Ovulation and Menstruation Health Study pilot cohort.
3. Describe the distribution of alopecia within the Ovulation and Menstruation Health Study pilot cohort.

METHODS

The Ovulation and Menstruation Health (OM) Study

The Ovulation and Menstruation Health (OM) Study is a comprehensive epidemiologic survey that seeks to characterize lifestyle and health characteristics of an ethnically diverse cohort of women. The study collects information regarding menstrual characteristics and manifestations of androgen excess in order to create a multi-ethnic longitudinal database to assess the health risks of PCOS. The goal of the pilot study specifically was to develop the best data collection methods and instruments for the future multi-ethnic, long-term PCOS cohort study that will be conducted in both English and Spanish. The recruitment goal for the pilot study was to have 200 participants complete the online consent form and survey module. A two-minute informational video on the menstrual cycle was created specifically for the OM Study and posted on the study website to be used for recruitment purposes.

Survey Design

The pilot study consisted of an online baseline questionnaire designed by Dr. Shruthi Mahalingaiah, a board-certified reproductive endocrinology/infertility physician-scientist at Boston University School of Medicine (BUSM). The survey was administered through REDCap, a secure web application for building and managing online research surveys and databases. The questionnaire collected information regarding demographics, anthropometrics, menstrual cycle health, contraceptive use, medication and supplement use, PCOS status, androgen excess characteristics, reproductive health, general health,

diet and lifestyle, and pregnancy and birth history. In addition, participants who have received care at Boston Medical Center (BMC) were given the option to have their survey results validated against their medical record.

A board-certified reproductive endocrinology/infertility physician-scientist designed the study questions. To ensure the survey questions were appropriate for a diverse population, particularly with respect to race/ethnicity and education level, the questions were written at an 8th grade reading level. Additionally, a select subset of questions underwent cognitive understandability and usability tests. To help participants correctly answer questions involving a self-reported rate of a condition, Renee Cannon, a licensed medical illustrator, was commissioned to sketch images for questions targeting body shape and hair growth.

Study Population

The OM study pilot included women who were 18-45 years old that still had the capacity to ovulate and menstruate, no prior history of chemotherapy or radiation, and who were not currently pregnant. The study population was not recruited with consideration of a present PCOS diagnosis, which allowed both women with PCOS and non-PCOS controls to be captured. In order to be able to answer the questions, the study subjects were also required to be able to read English and have an email address for receiving the survey link. The pilot study population of the 249 participants who completed the entire survey was used to determine patterns of hirsutism, acne, and alopecia severity.

Recruitment

Participants were actively recruited by multiple methods across multiple cities in the United States starting on August 9th, 2017. Although active recruitment for the pilot study ended October 10th 2017 the survey link is still currently open to additional participants. Initially the participants recruited were BMC patients who received a mailed letter prior to their appointment explaining the study and notifying them of the potential presence of study staff in their doctor's waiting room. Upon arrival to the appointment, these patients were approached in the OB/GYN clinics in the BMC Doctor's Office Building and Yawkey Clinic. Additional participants were recruited in-person at a local community Women's Market in Jamaica Plains, Boston, MA. The pilot recruitment then expanded to include the dissemination of the study website and survey link by posting flyers around Boston University and by advertisements on Facebook and other social-networking and health-related websites.

For in-person recruitment, the individuals were first asked to view an informational video about the study and were then asked to complete a screener if they were interested in participating. Eligible individuals were then prompted to start the survey on REDCap. For those that could not complete the survey in one sitting, follow-up emails were sent via REDCap. The emails provided participants with a code to access and proceed with their survey from the location they left off.

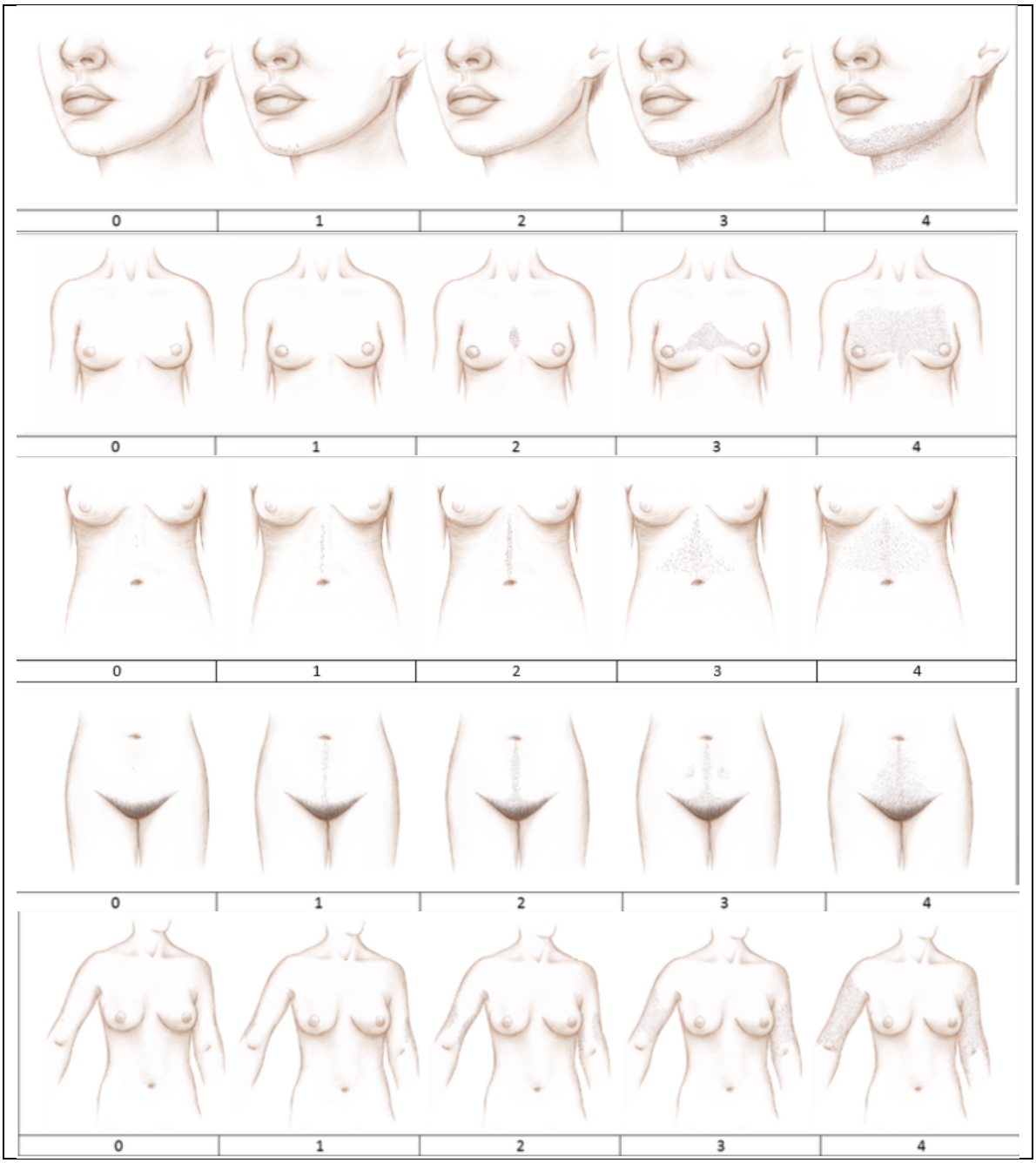
Those accessing the survey online could also choose to watch the informational video on the study website before being directed to the screener and questionnaire.

Individuals who consented to participate in this pilot study were entered into a raffle for a $200 gift card that was awarded at the end of the pilot recruitment period.

Hyperandrogenism analysis

In order to assess hyperandrogenism, questions from the survey measuring androgen excess indicators such as hirsutism, acne and alopecia were pulled from the dataset. Participants were asked to self-report the degree of hirsutism in nine androgen sensitive areas based on the images created by Renee Cannon (Figure 4). They were also asked to rate the severity of the acne based on a very brief description of the acne types (Table 5). Lastly, participants were asked to self-report degree of alopecia using the images illustrated by Renee Cannon (Figure 5).

Assessments of patterns of the clinical manifestations were determined by looking at the frequencies of self-reported degree of hirsutism, acne and alopecia. Also of interest was the data for clinically captured biochemical androgen excess in the subset of women who consented to medical record validation.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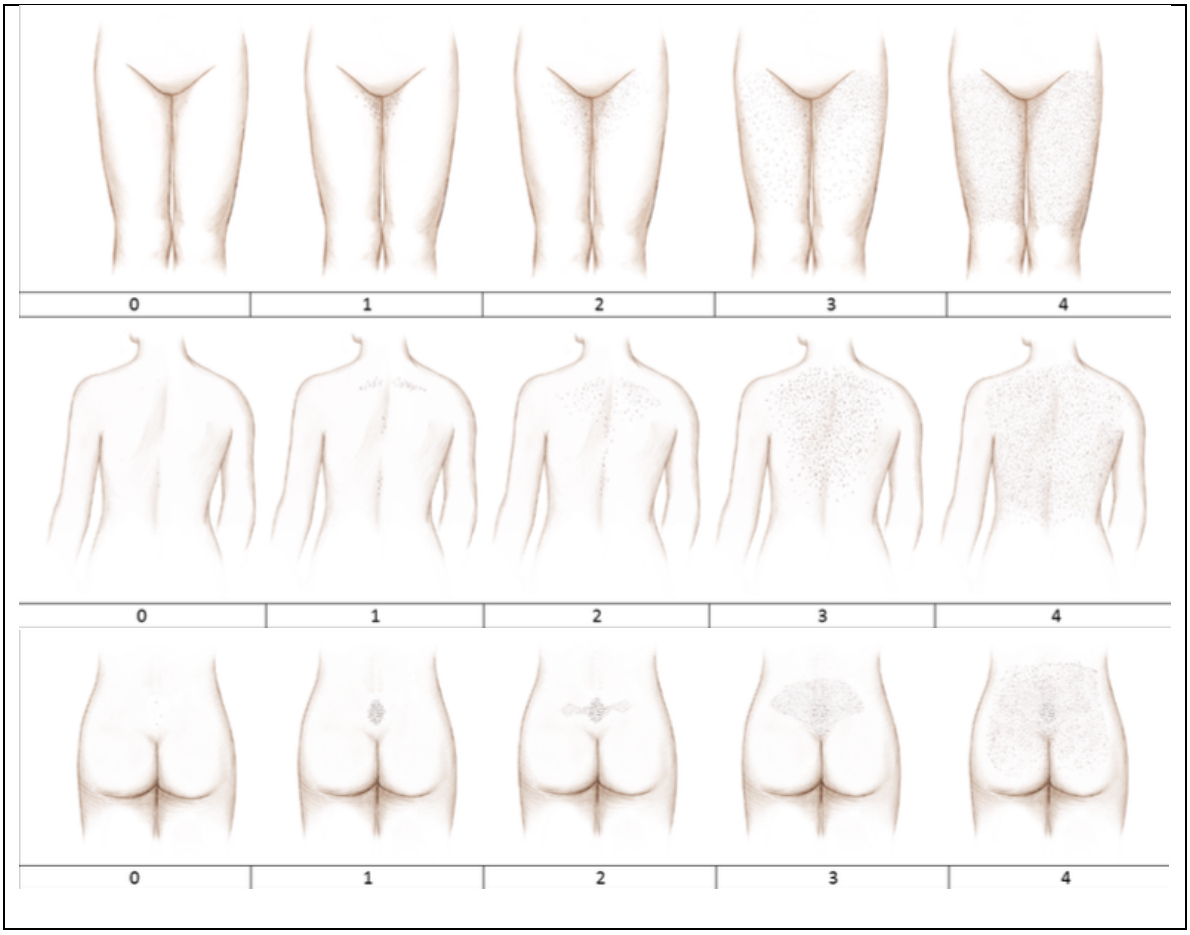

Figure 4. mFG Pictorial Tool. *Medical illustrator Renee Cannon illustrated these images for the OM Study. They were provided as the reference images to the questions asking women to self-report the degree of hirsutism.*

Table 5. Acne Type Descriptions *These were the descriptors provided in the OM study questionnaire asking women to rate the severity of their acne.*

Acne Type	Description
None or rare acne	None to a couple of pimples
Mild acne	4 or more pimples
Moderate acne	4 or more pimples that are red and irritated
Severe acne	4 or more pimples that are red, irritated, and have pus

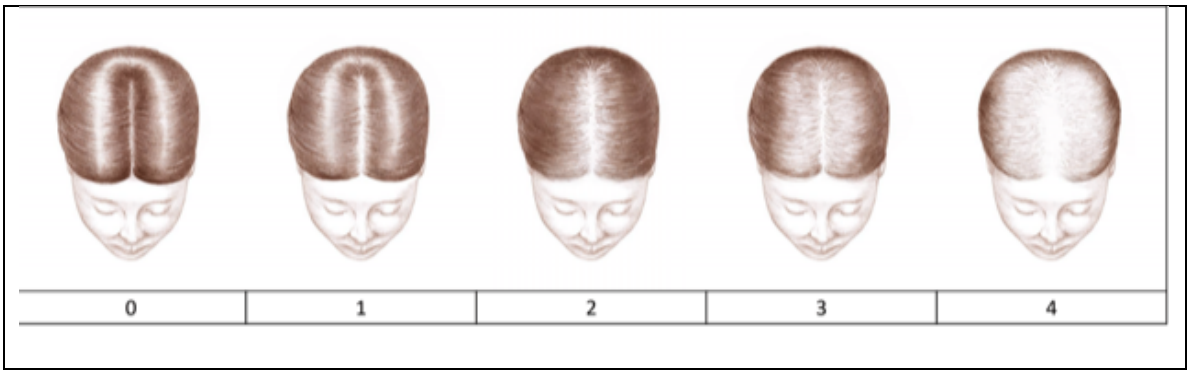

Figure 5. Alopecia Images *Medical illustrator Renee Cannon drew these images based on the Sinclair classification system for the OM Study. They were provided as the reference images to the questions asking women to self-report the degree of Alopecia*

RESULTS

After 2 months of active recruitment, a total of 283 individuals were deemed eligible and started the survey, and 249 of these participants actually completed the survey (Table 6). The majority of participants that completed the survey were recruited online (45.4%). Out of the 247 women that completed the study survey and answered the race/ethnicity question, 66.8% were white, 6.5% were Hispanic or of Spanish origin, 10.5% were Black or African-American, 1.6% were East Asian, 2.0% were Southeast Asian, 2.4% were South Asian, and 10.9% were of mixed ethnic backgrounds (Table 7).

Table 6. Summary of Survey Completion *The number of participants in the OM Study pilot cohort that completed various parts of the study survey.*

Survey Completion	Initiated Consent Form	Completed Consent Form	Consented	Completed Screener	Screened In (Eligible)	Started Survey	Completed Survey
Participants	438	388	384	384	350	283	249

Table 7. Cohort Study Population

Characteristics	Cohort
Number of women	249
Mean age (years)	27.1 yrs (n=249)
White (%)	66.8%(165/247)
Hispanic, Latina, or Spanish Origin (%)	6.5% (16/247)
Black or African American (%)	10.5 (26/247)
East Asian (%)	1.6% (4/247)
Southeast Asian (%)	2.0% (5/247)
South Asian (%)	2.4% (6/247)
More than 1 race**	10.9% (27/247)
Clinical hirsutism based on mFG score cutoff (%)	22.5% (55/245)
Alopecia (scalp hair score >2)	9.35% (23/246)
Acne (moderate-severe acne)	23.6 (58/246)
PCOS diagnosis by physician (%)	23.1% (57/247)
Any use of hormonal contraceptives (%)	84.3% (210/249)
Permission granted for medical record validation (n)*	44/62 (71%)
Androgen excess based on medical record review (n)	12/44 (27.3%)

*Among women who answered they have received care at BMC

Hirsutism

Clinical hirsutism was defined as total mFG score of ≥2 for East Asian women, ≥3 for Southeast Asian women, or ≥8 in all other race/ethnic categories. These race/ethnic specific cutoffs for mFG scores were based on the currently available clinical

literature. Based on these cutoffs 22.5% of women in the study cohort met the criterion for a clinical hirsutism diagnosis. Furthermore, there was a higher presence of clinical hirsutism between South Asian (66.7%) and Hispanic (43.8%) women. Mean total mFG scores were also highest in South Asian (13.8±9.1) and Hispanic (8.6±8.7) women (Table 9). Among women with PCOS the mean total mFG scores were higher (9.6 ± 7.6) than in non-PCOS women (3.7 ± 3.6). In addition there was a higher presence of clinical hirsutism among the population that had a PCOS Diagnosis (49.1%) when compared to non-PCOS women (14.7%) (Table 8).

Table 8. PCOS Diagnosis and Androgen Excess clinical manifestations *Both self-diagnosed and MD PCOS diagnosis were used.*

PCOS Diagnosis	Total mFG score (mean)	Clinical Hirsutism (%)	Acne severity score (mean)	Moderate to Severe Acne (%)	Alopecia severity (mean)	Alopecia ≥2 (%)
Yes	9.6 ± 7.6	49.1% (27/55)	2.0 ± 1.0	33.9% (19/56)	0.1 ± 0.4	14.3% (8/56)
No	3.7 ± 3.6	14.7% (28/190)	1.8 ± 0.9	20.5% (39/190)	0.1 ± 0.3	7.9% (15/190)

Table 9. Race/Ethnicity and Self-Reported mFG scores and Perception of Hairiness.

Patients reported on hair growth using the mFG score (0 being absent hair growth, 4 being excess hair growth) based on references images provided with questionnaire. Race/ethnic cutoffs for mFG scores were used and specified based on current available clinical literature. Where information is conflicting the most commonly used cutoff or average of available cutoffs was utilize. Where primary data is unavailable, the cutoff score of ≥8 was utilized. Total mFG score was calculated by sum of scores of nine body areas.

	mFG cutoff for hirsutism (≥ n)	Clinical hirsutism (%)	Total mFG score (mean)	mFG score for upper lip (mean)	mFG score for chin (mean)	mFG score for chest (mean)	mFG score for upper abdomen (mean)	mFG score for lower abdomen (mean)	mFG score for upper arms (mean)	mFG score for thighs (mean)	mFG score for back (mean)	mFG score for buttocks (mean)
Total	8	22.5 % (55/245)	5.0±5.4	0.6± 0.9	0.7± 1.0	0.6± 0.7	0.3± 0.8	1.2± 1.2	0.4± 0.8	0.8± 1.1	0.2± 0.6	0.3± 0.8
Race												
White	8	18.9 % (31/164)	4.4±4.5	0.5± 0.9	0.6± 0.9	0.6± 0.7	0.3± 0.6	1.0± 1.1	0.3± 0.6	0.8± 1.1	0.1± 0.4	0.3± 0.7
Hispanic, Latina, or Spanish Origin	8	43.8 % (7/16)	8.6±8.7	0.9± 1.3	0.8± 1.2	0.8± 0.7	0.7 ±1.0	1.6± 1.5	0.9± 1.3	1.4± 1.4	0.7± 1.1	0.8± 1.1
Black or African American	8	19.2 % (5/26)	4.7±3.6	0.5± 0.8	1.0± 1.2	0.2± 0.6	0.3± 0.7	1.4± 1.2	0.7± 1.1	0.6± 0.9	0.1± 0.3	0.1± 0.4
East Asian	2	25% (1/4)	1.8±2.2	0.3± 0.5	0 ± 0	0.5± 0.6	0.3± 0.5	0.3± 0.5	0 ± 0	0 ± 0	0.5± 1.0	0 ± 0
Southeast Asian	3	20% (1/5)	5.4±9.9	0.2± 0.5	0.2± 0.5	0.4± 0.6	0.8± 1.8	1.0± 1.7	0.8± 1.3	0.6± 1.3	0.6± 1.3	0.8± 1.8
South Asian	8	66.7 % (4/6)	13.8± 9.1	1.7± 1.5	1.3± 1.5	1.5± 1.4	1.8± 1.7	2.2± 1.7	1.3± 1.2	2.2± 1.0	1.0± 1.3	0.8± 1.0
Multiple race/ethnicities	8	25% (6/24)	4.8±5.3	0.6± 1.0	0.5± 0.7	0.6± 0.8	0.2± 0.7	1.4± 1.2	0.3± 0.7	0.9± 1.2	0.2± 0.5	0.3± 0.8

Acne

Mean population acne severity score (from 1 to 4) was 1.9 ± 0.9 (n=246). Moderate-severe acne was reported in 23.6%(58/246) of all surveyed women. Moderate-severe acne was also highest in South Asian Women (50%, 3/6) and lowest in East Asian women (0%) (Table 10). Women with PCOS (33.9%) had a higher prevalence of moderate-severe acne than non-PCOS (20.5%) women. Additionally women with a PCOS diagnosis had a higher acne severity mean (2.0 ± 1.0) than non-PCOS women (1.8 ± 0.9 (Table 8).

Table 10. Race/Ethnicity and Self-Reported Acne Severity. *Patients reported acne severity from scale of 1-4 (1 being none, 4 being severe) based on references images provided with questionnaire.*

	Acne severity score (mean)	Report no or rare acne (%)	Report mild acne (%)	Report moderate acne (%)	Report severe acne (%)
Total	1.9 ± 0.9	43.9% (108/246)	32.5% (80/246)	17.5% (43/246)	6.1% (15/246)
Race					
White	1.9 ± 0.9	40% (66/165)	35.1% (58/165)	20 % (33/165)	4.9% (8/165)
Hispanic, Latina, or Spanish Origin	1.9 ± 1.1	46.7% (7/15)	26.7% (4/15)	13.3% (2/15)	13.3% (2/15)
Black or African American	1.6 ± 0.9	61.5% (16/26)	23.1% (6/26)	7.7% (2/26)	7.7% (2/26)
East Asian	1.3 ± 0.5	75% (3/4)	25% (1/4)	0% (0/4)	0% (0/4)
Southeast Asian	1.8 ± 1.3	60% (3/5)	20% (1/5)	0% (0/5)	20% (1/5)
South Asian	2.5 ± 1.4	33.3% (2/6)	16.6% (1/6)	16.7% (1/6)	33.3% (2/6)
Multiple race/ethnicities	1.8 ± 0.8	44% (11/25)	36% (9/25)	20% (5/25)	0% (0/25)

Alopecia

Alopecia was defined as moderate to severe hair loss, a score ≥2, in women that reported hair loss. On a severity scale of 1-4 for alopecia, 9.4%(23/246) of all women met this criterion for an alopecia classification. The highest prevalence of alopecia was in black or African American (26.9%, 7/26), East Asian (25%, 1/4), and mixed ethnicity (20%, 5/25) women. The mean population alopecia severity was 0.4±0.7 in the overall population and highest among East Asian (1±0.8) and Black women (0.8±1.1) (Table 11). Among women with PCOS (14.3%) the prevalence of alopecia was higher than in non-PCOS women (7.9%)(Table 8).

Table 11. Race/Ethnicity and Self-Reported Alopecia Severity. *Patients reported alopecia severity from scale of 0-4 (0 being absent, 4 being severe) based on references images provided with questionnaire.*

	Alopecia severity (mean)	Report alopecia (scalp hair loss classified as ≥2) (%)
Total	0.4±0.7	9.4% (23/246)
Race		
White	0.3±0.6	5.5% (9/165)
Hispanic, Latina, or Spanish Origin	0.3±0.6	6.3% (1/16)
Black or African American	0.8±1.1	26.9% (7/26)
East Asian	1±0.8	25% (1/4)
Southeast Asian	0.2±0.5	0% (0/5)
South Asian	0.3±0.5	0% (0/6)
Multiple race/ethnicities	0.6±0.8	20% (5/25)

Biochemical Hyperandrogenism

44 women consented to medical record validation (71%, 44/62) and 12 of those women had serum androgen levels on file. These 12 women had a total of 58 medical lab values for testosterone. The most recent lab values for bioavailable testosterone, free testosterone, and total testosterone are reported for analysis in Table 12. Total testosterone testing is done using LC-MS/MS by Quest Diagnostics and the reference range reported by Quest diagnostics is 2-45 ng/dL in females 18 years and older. Of the 12 females with medical lab values, 33% (4/12) had total testosterone levels above this normal reference range.

Table 12. PCOS diagnosis and serum levels for participants consenting to medical record review (n=58)

	n	**% or Mean ± SD (min, max)**
Self-Report		
Diagnosis of PCOS by Physician, %	48	
Yes	12	25.0%
No	36	75.0%
Self-Diagnosis of PCOS for participants not diagnosed by physician, %	36	
Yes	2	5.6%
No	34	94.4%
Medical Record Review		
Diagnosis of PCOS in Medical Record, %	46	
Yes	11	23.9%
No	35	76.1%
Serum lab values, Mean ± SD **		
Bioavailable Testosterone	11	10.9 ± 10.3 (1.0, 32.7)
Free Testosterone	11	5.4 ± 5.2 (0.5, 15.9)
Total Testosterone	12	39.5 ± 27.9 (6.0, 87.0)

*% missing (n): Diagnosis by physician: 17.2% (10); Diagnosis in medical record: 20.7% (12); Bioavailable testosterone: 81.0% (47); Free testosterone: 81.0% (47); Total testosterone: 79.3% (46);
**most recent medical record lab values reported per participant

DISCUSSION

The association between hyperandrogenism and polycystic ovary syndrome is well established (The Rotterdam ESHRE/ASRM, 2003). It is also well defined that androgen excess may manifest clinically as hirsutism, acne, alopecia, and other masculinization features. The Endocrine Society has indicated that androgen excess assessment is a key feature in diagnosing PCOS (The Rotterdam ESHRE/ASRM, 2003). Androgen excess is often validated by biochemical evaluation of serum TT, FT, DHEA-S, and 17OHP (Lizneva et al., 2016). This study evaluated the distribution of self-reported clinical signs of hyperandrogenism in a diverse, multi-ethnic cohort of women. Additionally, the study sought to determine differences between presentation of hirsutism, acne, and alopecia in women diagnosed with PCOS and those without a PCOS diagnosis.

Androgen Excess and Demographics

It has been determined that there is ethnic variation in the presentation and manifestations of PCOS, particularly in the clinical presentation of hyperandrogenism (Bozdag et al., 2016). The pilot study data supports this claim as it demonstrated considerable ethnic variability in hirsutism, acne and alopecia. Based on mean total mFG score, South Asian women self-reported the greatest degree of hair growth, whereas East Asian women reported the lowest degree (Table 9). This observed ethnic variability in hair growth is due to the number of hair follicles per unit skin area and rate of hair growth (Yildiz, 2006). Furthermore, baseline hair distribution patterns vary between races. East Asian and Native American women have the least body hair. Whereas, White and

African-American women have an intermediate amount of hair and Mediterranean, South Asian, and Middle Eastern women have substantially more body hair than all other races/ethnicities (Barbieri & Ehrmann, 2018a; Monash University, 2018).

When considering the androgen excess marker of acne in the OM pilot, South Asian women self-reported the most severe cases (Table 10). Although studies have shown that the expression of acne differs between skin types, these studies are limited in establishing a clear pattern between race/ethnicity and acne caused by hyperandrogenism (Davis & Callender, 2010). There seemed to be an association between acne severity and race/ethnic variability in this study, however a larger study is needed to confirm these findings.

When evaluating the frequency of self-reported hair loss, African-American women had the highest number of alopecia cases when frequencies were compared to non-African American women (Table 11). This cohort data reveals that alopecia is an important indicator of androgen excess in African American women. Data presented at the 2016 American Academy of Dermatology's 74th annual meeting demonstrated that African-American women are more prone to hair loss. There are thought to be three major types of alopecia in this population including: androgenic alopecia, central centrifugal cicatricial alopecia (CCCA), and traction alopecia. CCCA is caused by inflammation and destruction of hair follicles and traction alopecia occurs due to hair styling practices (American Academy of Dermatology, 2016). Despite identifying major types of alopecia, studies have not been able to determine how much of this hair loss is due to androgenic alopecia specifically.

Androgen Excess and PCOS

In the analysis of the pilot cohort data, hirsutism, acne, and alopecia were far more common among women diagnosed with PCOS than in non-PCOS women (Table 8). This data supports the current understanding of hyperandrogenism as a criterion for PCOS diagnosis (The Rotterdam ESHRE/ASRM, 2003). Further data collection must be done in order to establish a relationship between biochemical hyperandrogenism as only 33% of the participants that had their medical record validated had evidence of TT excess (Table 12). The small sample size makes it uncertain whether these values are representative of the pilot cohort, and the general population as a whole. Recruiting more participants willing to consent to medical record validation in the larger OM study will allow us to more appropriately validate the use of biochemical evidence for PCOS diagnosis.

Strengths, limitations, and future considerations

The data from this paper was derived from a large multi-ethnic pilot cohort. This allowed for a representative evaluation of clinical hyperandrogenism in a more racially/ethnically diverse cohort of women than existing PCOS studies would have allowed. However, the study is limited by the subjective nature of asking participants to self-report hair growth, acne and hair loss. Including images for hair growth and hair loss along with the descriptors provided participants with a visual guide to help improve the accuracy of their response. Although Renee Cannon also illustrated images for acne, an executive decision was made not to include them in the questionnaire. It was believed that including the images would not have the same effect as it did for hirsutism and

alopecia responses. Instead clear descriptions were included with each acne type; past studies have shown that if subjects are given clear descriptions of acne type, they are capable of self-diagnosing their acne (Gold et al., 2009).

Although research findings have shown that hirsutism cutoffs are race/ethnicity dependent, the available literature is not representative of the scope of this variability. Furthermore, current studies may be biased since different regions, countries, or sample sizes may show different cutoffs in similar ethnicities. Conducting a larger study with an increased sample size and targeting race/ethnicities that are not currently represented in the pilot cohort may improve this issue. Additionally, despite the mFG scale being the most commonly used diagnostic tool for hirsutism, it has not been validated in patient populations for androgen excess self-assessment. Future studies should evaluate whether self-assessment of hair and skin using the mFG scale is consistent with clinical examination for the purposes of PCOS diagnosis. The information gathered would be used to inform physicians on how to diagnose and treat individuals with hirsutism and PCOS.

Similarly, there is a wide selection of more than 25 measures for acne available. However, most of these tools are subjective. One study evaluated 18 current acne global grading scales (AGGS) serving as a step towards harmonizing the scales and creating a foundation for the development of a universal grading scale (Tan et al., 2013). It is necessary to come up with a consistent universal standard for grading acne severity in order to strengthen the use of acne as a diagnostic feature for hyperandrogenism.

In order to have a more diverse and accurate representation of the global population, the OM survey can be translated into different languages and more targeted recruitment efforts can be made. Preliminary steps have been made in making these changes as Dr. Shruthi Mahalingaiah's Lab has started translating the questionnaire into Spanish in order to expand accessibility to Spanish speakers. Having a large cohort representative of a global population will increase our understanding on the presentation of PCOS features and support the ethnic variability patterns observed in the cohort population.

APPENDIX

Questionnaire from Ovulation and Menstruation (OM) Study Pilot Survey

Medical record validation

Section III: About you

10. Have you ever received care at Boston Medical Center?
    a. Yes
    b. No

11. We would like to look at your medical records in order to study other factors associated with menstrual health. All study data is protected by HIPAA-compliant equipment and software.
    a. Yes, it is OK to check my medical record
    b. No, please do not check medical record

Demographic Information

Section IV: Baseline Questionnaire

4. Which categories describe you? Select all that apply. You may select more than one group.
    a. White
       For example, German Irish, English, Italian
    b. Hispanic, Latina, or Spanish Origin
       For example, Mexican, Mexican American, Puerto Rican, Cuban, Salvadoran, Dominican, Colombian, etc
    c. Black or African American
       For example, African American, Jamaican, Haitian, Nigerian, Ethiopian, Somali, etc.
    d. East Asian
       For example, Chinese, Japanese, Korean, Mongolian, Taiwanese, etc.
    e. Southeast Asian
       For example, Burmese, Cambodia, Hmong, Indonesian, Laotian, Malaysian, Filipino, Thai, Vietnamese, etc.
    f. South Asian
       For example Asian Indian, Bangladeshi, Bhutanese, Maldivian, Nepalese, Pakistani, Sri Lankan, etc.
    g. American Indian or Alaskan Native
       For example, Navajo Nation, Blackfeet Tribe, Mayan, Aztec, Native Village of Barrow Inupiat Traditional Government, Nome Eskimo Community, etc.
    h. Middle Eastern or North African
       For example, Lebanese, Iranian, Egyptian, Syrian, Moroccan, Algerian, etc.

i. Native Hawaiian or Other Pacific Islander
For example, Native Hawaiian, Samoan, Chamorro, Tongan, Fijian, Marshallese, etc.
j. Some other race, ethnicity, or origin.

Contraceptive History

Section VII: Contraceptive History

1. Hormonal contraceptives include the Pill, patch, implants, or injectables. Have you ever used hormonal contraceptives?
    a. Yes
    b. No

Androgen Excess Questions

Section VIII: Health and Body

5. Thinking about your face or back, how would you rate your acne? If your acne changes during menstrual cycle, please think about the acne at its worst.
    a. None or rare acne (none to a couple pimples)
    b. Mild acne (4 or more pimples)
    c. Moderate acne (4 or more pimples that are red and irritated)
    d. Severe acne (4 or more pimples that are red, irritated, and have pus)

For the next questions, we want to know about how much coarse or thick hair you have on different parts of your body. When you answer, please think about your natural body state – that means how much hair you have when you are not using hair removal procedures or treatment. You can use the images as a guide, but remember your own hair pattern may be slightly different.:

13. How much coarse or thick hair do you have on your upper lip?
    [Upper Lip hair growth image included]
    a. 0 No thick hairs
    b. 1 Few scattered thick hairs
    c. 2 several scattered thick hairs
    d. 3 moderate amount of thick hairs
    e. 4 A lot of hair
14. How much coarse or thick hair do you have on your chin?
    [Chin hair growth image included]
    a. 0 No thick hairs
    b. 1 Few scattered thick hairs
    c. 2 several scattered thick hairs
    d. 3 moderate amount of thick hairs
    e. 4 A lot of hair
15. How much coarse or thick hair do you have on your chest and nipple area?
    [Chest and Nipple hair growth image included]

a. 0 No thick hairs
b. 1 Few scattered thick hairs
c. 2 several scattered thick hairs
d. 3 moderate amount of thick hairs
e. 4 A lot of hair

16. How much coarse or thick hair do you have on your upper abdomen?
[Upper abdomen hair growth image included]
    a. 0 No thick hairs
    b. 1 Few scattered thick hairs
    c. 2 several scattered thick hairs
    d. 3 moderate amount of thick hairs
    e. 4 A lot of hair
17. How much coarse or thick hair do you have on your lower abdomen – that is the area from your belly button to your pubic area?
[Lower abdomen hair growth image included]
    a. 0 No thick hairs
    b. 1 Few scattered thick hairs
    c. 2 several scattered thick hairs
    d. 3 moderate amount of thick hairs
    e. 4 A lot of hair
18. How much coarse or thick hair do you have on your upper arms?
[Upper arms hair growth image included]
    a. 0 No thick hairs
    b. 1 Few scattered thick hairs
    c. 2 several scattered thick hairs
    d. 3 moderate amount of thick hairs
    e. 4 A lot of hair
19. How much coarse or thick hair do you have on your thighs?
[Thighs hair growth image included]
    a. 0 No thick hairs
    b. 1 Few scattered thick hairs
    c. 2 several scattered thick hairs
    d. 3 moderate amount of thick hairs
    e. 4 A lot of hair
20. How much coarse or thick hair do you have on your back?
[Back hair growth image included]
    a. 0 No thick hairs
    b. 1 Few scattered thick hairs
    c. 2 several scattered thick hairs
    d. 3 moderate amount of thick hairs
    e. 4 A lot of hair
21. How much coarse or thick hair do you have on your buttocks?
[Buttocks hair growth image included]
    a. 0 No thick hairs

b. 1 Few scattered thick hairs
c. 2 several scattered thick hairs
d. 3 moderate amount of thick hairs
e. 4 A lot of hair

22. Using the images below, how would you rate your current scalp hair thickness? You can use the images as a guide, but your own hair pattern may be slightly different. [Scalp hair loss image included]
    a. 0 Thick and full hair
    b. 1 Slightly reduced hair with widening part
    c. 2 Reduced hair with widening part and some scalp showing
    d. 3 Significantly reduced hair and more scalp showing
    e. 4 Scalp mostly visible

Polycystic Ovary Syndrome Questions

Section IX: Polycystic Ovary Syndrome

1. Polycystic Ovary Syndrome is a health condition involving irregular periods, excess testosterone, increased acne, body and facial hair, and many small cysts in the ovaries. Some women also experience hair loss on the scalp. Has a doctor ever diagnosed you with Polycystic Ovary Syndrome or PCOS?
    a. Yes
    b. No
2. Do you think you might have PCOS?
    a. Yes
    b. No

LIST OF JOURNAL ABBREVIATIONS

AFP	American Family Physician
Clin Epidemiol	Clinical Epidemiology
Front Physiol	Frontiers in Physiology
Hum Reprod	Human Reproduction
Int J Endocrinol Metab	International Journal of Endocrinology and Metabolism
J Clin Aesthet Dermatol	The Journal of Clinical and Aesthetic Dermatology
J Clin Endocrinol Metab	The Journal of Clinical Endocrinology & Metabolism
J Cutan Aesthet Surg	Journal of Cutaneous and Aesthetic Surgery
J Med Assoc Thai	Journal of the Medical Association of Thailand
Semin Reprod Med	Seminars in Reproductive Medicine

REFERENCES

Agapova SE, Cameo T, Sopher AB, Oberfield SE. Diagnosis and Challenges of Polycystic Ovary Syndrome in Adolescence. *Semin Reprod Med*. 2014;32(3):194-201. doi:10.1055/s-0034-1371091

American Academy of Dermatology. Survey: Almost half of African-American women have experienced hair loss. March 4, 2016. https://www.aad.org/media/news-releases/hair-loss-in-african-american-women. Accessed March 1, 2019.

Azziz R.Epidemiology and genetics of the polycystic ovary syndrome in adults. In: Barbieri RL, Martin KA. ed. *UpToDate*. Waltham, MA.: UpToDate; 2017. www.uptodate.com. Accessed December 17, 2018.

Azziz R, Carmina E, Dewailly D, et al. The Androgen Excess and PCOS Society criteria for the polycystic ovary syndrome: the complete task force report. *Fertility and Sterility*. 2009;91(2):456-488. doi:10.1016/j.fertnstert.2008.06.035

Azziz R, Marin C, Hoq L, Badamgarav E, Song P. Health Care-Related Economic Burden of the Polycystic Ovary Syndrome during the Reproductive Life Span. *J Clin Endocrinol Metab*. 2005;90(8):4650-4658. doi:10.1210/jc.2005-0628

Barbieri RL, Ehrmann DA. Diagnosis of polycystic ovary syndrome in adults. In: Crowley WF, Martin KA. ed. *UpToDate*. Waltham, MA.: UpToDate; 2018a. www.uptodate.com. Accessed November 27, 2018.

Barbieri RL, Ehrmann DA. Evaluation of premenopausal women with hirsutism. In: Snyder PJ, Crowley WF, Martin KA. ed. *UpToDate*. Waltham, MA.: UpToDate; 2018b. www.uptodate.com. Accessed November 17, 2018.

Barbieri RL, Ehrmann DA. Clinical Manifestations of polycystic ovary syndrome in adults. In: Snyder PJ, Crowley WF, Martin KA. ed. *UpToDate*. Waltham, MA.: UpToDate; 2018c. www.uptodate.com. Accessed November 17, 2018.

Bode DV, Seehusen DA, Baird D. Hirsutism in Women. *AFP*. 2012;85(4):373-380.

Bouhanna P. Multifactorial Classification of Male and Female Androgenetic Alopecia. *Dermatologic Surgery*. 2000;26(6):555-561. doi:10.1046/j.1524-4725.2000.00009.x

Bozdag G, Mumusoglu S, Zengin D, Karabulut E, Yildiz BO. The prevalence and phenotypic features of polycystic ovary syndrome: a systematic review and

meta-analysis. *Hum Reprod*. 2016;31(12):2841-2855. doi:10.1093/humrep/dew218

Cheewadhanaraks S, Peeyananjarassri K, Choksuchat C. Clinical diagnosis of hirsutism in Thai women. *J Med Assoc Thai*. 2004;87(5):459-463.

Davis EC, Callender VD. A Review of Acne in Ethnic Skin. *J Clin Aesthet Dermatol*. 2010;3(4):24-38.

DeUgarte CM, Woods KS, Bartolucci AA, Azziz R. Degree of Facial and Body Terminal Hair Growth in Unselected Black and White Women: Toward a Populational Definition of Hirsutism. *J Clin Endocrinol Metab*. 2006;91(4):1345-1350. doi:10.1210/jc.2004-2301

El Hayek S, Bitar L, Hamdar LH, Mirza FG, Daoud G. Poly Cystic Ovarian Syndrome: An Updated Overview. *Front Physiol*. 2016;7. doi:10.3389/fphys.2016.00124

Franik G, Bizo A, Włoch S, Kowalczyk K, Biernacka-Bartnik A, Madej P. Hormonal and metabolic aspects of acne vulgaris in women with polycystic ovary syndrome. *European review for medical and pharmacological sciences*. 2018; 22:4411-4418.

Gold MH, Andriessen A, Biron J. Self-diagnosis of Mild-to-Moderate Acne for Self Treatment with Blue Light Therapy. *J Clin Aesthet Dermatol*. 2009;2(4):40-44.

Gupta M, Mysore V. Classifications of Patterned Hair Loss: A Review. *J Cutan Aesthet Surg*. 2016;9(1):3-12. doi:10.4103/0974-2077.178536

Hair Again. Growth Cycle. http://www.fresnohair.com/2/post/2017/05/growth cycle.html. Published May 6, 2017. Accessed March 11, 2019.

Herskovitz I, Tosti A. Female Pattern Hair Loss. *Int J Endocrinol Metab*. 2013;11(4). doi:10.5812/ijem.9860

Lee W-S, Ro BI, Hong SP, et al. A new classification of pattern hair loss that is universal for men and women: Basic and specific (BASP) classification. *Journal of the American Academy of Dermatology*. 2007;57(1):37-46. doi:10.1016/j.jaad.2006.12.029

Legro RS, Arslanian SA, Ehrmann DA, et al. Diagnosis and Treatment of Polycystic Ovary Syndrome: An Endocrine Society Clinical Practice Guideline. *J Clin Endocrinol Metab*. 2013;98(12):4565-4592. doi:10.1210/jc.2013-2350

Lizneva D, Gavrilova-Jordan L, Walker W, Azziz R. Androgen excess: Investigations

and management. *Best Practice & Research Clinical Obstetrics & Gynaecology*. 2016;37:98-118. doi:10.1016/j.bpobgyn.2016.05.003

Ludwig E. Classification of the types of androgenetic alopecia (common baldness) occurring in the female sex. *British Journal of Dermatology*. 1977;97(3):247-254. doi:10.1111/j.1365-2133.1977.tb15179.x

Martin KA, Anderson RR, Chang RJ, et al. Evaluation and Treatment of Hirsutism in Premenopausal Women: An Endocrine Society Clinical Practice Guideline. *J Clin Endocrinol Metab*. 2018;103(4):1233-1257. doi:10.1210/jc.2018-00241

Monash University, 2018, The International evidence-based guideline for the assessment and management of Polycystic Ovary Syndrome (PCOS) 2018, Monash University, Australia, ISBN-13:978-0-646-98332-5. Available at: monash.edu/medicine/sphpm/mchri/pcos

Olsen EA. Androgenetic alopecia. In: Olsen EA, editor. Disorders of Hair Growth: Diagnosis and Treatment. New York: McGraw-Hill; 1994. pp. 257–83.

Pochi PE, Shalita AR, Strauss JS, et al. Report of the consensus conference on acne classification: Washington, D.C., March 24 and 25, 1990. *Journal of the American Academy of Dermatology*. 1991;24(3):495-500. doi:10.1016/S0190-9622(08)80076-X

Qin JZ, Pang LH, Li MJ, Fan XJ, Huang RD, Chen HY. Obstetric complications in women with polycystic ovary syndrome: a systematic review and meta-analysis. *Reprod Biol Endocrinol*. 2013;11:56. doi:10.1186/1477-7827-11-56

Randall VA. Androgens and hair growth. *Dermatologic Therapy*. 2008;21(5):314-328. doi:10.1111/j.1529-8019.2008.00214.x

Randeva HS, Tan BK, Weickert MO, et al. Cardiometabolic Aspects of the Polycystic Ovary Syndrome. *Endocr Rev*. 2012;33(5):812-841. doi:10.1210/er.2012-1003

Rosner W, Auchus RJ, Azziz R, Sluss PM, Raff H. Utility, Limitations, and Pitfalls in Measuring Testosterone: An Endocrine Society Position Statement. *J Clin Endocrinol Metab*. 2007;92(2):405-413. doi:10.1210/jc.2006-1864

Savin RC. A method for visually describing and quantitating hair loss in male pattern baldness. *Journal of Investigative Dermatology*. 1992;98:604.

Sinclair R, Jolley D, Mallari R, Magee J. The reliability of horizontally sectioned scalp biopsies in the diagnosis of chronic diffuse telogen hair loss in women.

Journal of the American Academy of Dermatology. 2004;51(2):189-199. doi:10.1016/S0190-9622(03)00045-8

Sirmans SM, Pate KA. Epidemiology, diagnosis, and management of polycystic ovary syndrome. *Clin Epidemiol*. 2013;6:1-13. doi:10.2147/CLEP.S37559

Tan JKL, Jones E, Allen E, Pripotnev S, Raza A, Wolfe B. Evaluation of essential clinical components and features of current acne global grading scales. *Journal of the American Academy of Dermatology*. 2013;69(5):754-761. doi:https://doi.org/10.1016/j.jaad.2013.07.029

The Rotterdam ESHRE/ASRM sponsored PCOS consensus workshop group. Revised 2003 consensus on diagnostic criteria and long-term health risks related to polycystic ovary syndrome. *Fertility and Sterility*. 2004;81(1):19-25. doi:10.1016/j.fertnstert.2003.10.004

Uysal G, Sahin Y, Unluhizarci K, et al. Is acne a sign of androgen excess disorder or not? *European Journal of Obstetrics & Gynecology and Reproductive Biology*. 2017;211:21-25. doi:10.1016/j.ejogrb.2017.01.054

Welch C. Understanding the Hair Growth Cycle. Toppik Blog. https://www.toppik.com/hairtoppiksblog/understanding-hair-growth-cycle/. Published May 8, 2018. Accessed March 11, 2019.

Yildiz BO, Bolour S, Woods K, Moore A, Azziz R. Visually scoring hirsutism. *Hum Reprod Update*. 2010;16(1):51-64. doi:10.1093/humupd/dmp024

Yildiz BO. Diagnosis of hyperandrogenism: clinical criteria. *Best Practice & Research Clinical Endocrinology & Metabolism*. 2006;20(2):167-176. doi:10.1016/j.beem.2006.02.004

Zargar AH, Wani AI, Masoodi SR, Laway BA, Bashir MI, Salahuddin M. Epidemiologic and etiologic aspects of hirsutism in Kashmiri women in the Indian subcontinent. *Fertility and Sterility*. 2002;77(4):674-678. doi:10.1016/S0015-0282(01)03241-1

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