Research Article

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Sex hormone exposure and reproductive factors in pulmonary arterial hypertension: a case–control study

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Abstract

Pulmonary arterial hypertension (PAH) is a sexually dimorphic disease that for unknown reasons affects women more than men. The role of estrogens, both endogenous and exogenous, and reproductive factors in this female susceptibility is still poorly understood. It has been strongly suggested that sex hormones may influence the development and progression of the disease. We sought to determine whether sex hormone exposures and reproductive factors associate with PAH patients compared to control subjects, using a questionnaire and interview to obtain information regarding these potential risk factors. We conducted a single-center unmatched case–control study. Six hundred and thirty-four women and men with PAH, as well as 27 subjects with BMPR2 mutations but no PAH and 132 healthy population controls were enrolled from the Vanderbilt Pulmonary Hypertension Research Cohort and researchmatch.org. Questionnaires and nurse-led interviews were conducted to obtain information regarding sex hormone exposures and reproductive factors. Additional history was obtained on enrolled patients including disease severity variables and comorbidities. Responses to the questionnaires were analyzed to describe these exposures in this population as well as assess the association between disease severity variables and sex hormone exposures. Reproductive and endogenous factors that determine lifelong estrogen exposure were similar between PAH cases and controls. Patients with associated PAH were significantly more likely to be postmenopausal compared to controls. There were similar rates of "ever-use" and duration of use of oral contraceptive pills and hormone replacement therapy in patients when compared to controls. Disease severity variables were not significantly affected by any exposure after adjusting for PAH sub-group. In contrast to our hypothesis, that a greater exposure to exogenous sources of female sex hormones associates with PAH case status, we found similar rates of endogenous and exogenous sex hormone exposure between PAH patients and unmatched controls.

Keywords

gender issues, hormones, pulmonary arterial hypertension

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Introduction

Pulmonary arterial hypertension (PAH) is a rare disease that results in narrowing of the small pulmonary arteries leading to elevation of pulmonary vascular resistance (PVR), subsequent right ventricular failure, and death. PAH is a sexually dimorphic disease that for unknown reasons affects women more than men. This female predominance has increased over time with PAH cohorts demonstrating a female to male ratio of 1.7:1 in the late 1980s compared to 4:1 in the REVEAL registry in the late 2000s, 2.3:1 in younger patients (18–65 year olds) in COMPERA, and 3.1:1 in more recent

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Chinese registries.[1–4] Female sex appears to be the best established clinical risk factor for most forms of PAH; however, women with PAH have better outcomes than men, leading many in the field to investigate sex hormone influences on the disease.[5–9] Manipulation of estrogens in animal models of pulmonary hypertension (PH) has shown contradictory outcomes; recent data suggest the need to examine the broader sex hormone milieu.[10–12]

Sex hormones may influence the development and progression of PAH. The role of estrogen, both endogenous and exogenous, and reproductive factors in this female susceptibility is still poorly understood. Several observations implicate the sex hormones, particularly estrogens, as modifiers of disease penetrance in subjects with PAH, including the greater penetrance of disease among females with a BMPR2 gene mutation and a lower prevalence for pre-pubertal but higher prevalence for post-pubertal females.[13,14] Irey and Norris described pulmonary vascular intimal lesions in a woman with pharmacologic estrogen exposure, while Morse et al. reported a 64-year-old woman in a family with PAH for whom hormone replacement therapy (HRT) was prescribed and three months later developed clinically significant PH.[15,16] Anecdotally, diagnosis often occurs during or soon after pregnancy when substantial hormonal changes are thought to occur. Contemporary cohorts of PAH patients have also brought to light that the profile of patients being diagnosed with PAH, in addition to being predominately female, are now older and more obese.[5] This suggests that another period of substantial sex hormone fluctuation, menopause, may be important to PAH development.

Given that not everyone with a known risk factor for the development of PAH develops disease, it is critical to know whether endogenous or exogenous hormones, in addition to known and unknown genetic susceptibilities, confer additional risk. Pharmacologic estrogen exposure in the form of oral contraceptives (OCPs) and HRT, which have been in common use for the last three to four decades and associate with other female predominant diseases such as breast cancer and cardiovascular disease, may be of concern.[17,18] Previously, a novel but uncontrolled study of PAH patients found that over 80% of women with PAH reported prior use of any hormone therapy.[19]

We sought to determine whether sex hormone exposures and reproductive factors associate with PAH in an unmatched case-control study, using a questionnaire and interview to obtain information regarding these potential risk factors. We hypothesized that a greater exposure to exogenous sources of female sex hormones associates with PAH patients.

Methods

Subjects

Subjects enrolled in the Vanderbilt Pulmonary Hypertension Research Cohort were eligible for inclusion in the study. This included patients followed for clinical care at Vanderbilt University Medical Center (VUMC), members of the Vanderbilt Familial Pulmonary Hypertension Registry (VFPHR), and subjects enrolled directly via the Research Room of the Pulmonary Hypertension Association (PHA) International Conference in both 2012 (Orlando, Florida) and 2014 (Indianapolis, Indiana). Research Room participants were considered eligible for the study if they reported a diagnosis of PH that was confirmed by medical record review (including documentation of cardiac catheterization supportive of PAH diagnosis according to internationally accepted criteria), currently receiving medical treatment for PAH and able to give informed consent. The Institutional Review Board (IRB) approved this study, with all subjects consented for participation (VUMC, IRB Registration #9401 for patient and family members and #111530 for normal controls). We included patients with idiopathic, heritable, or associated PAH as designated by their treating PH physicians and meeting traditional, international diagnostic criteria: mean pulmonary artery pressure (PAP) greater than or equal to 25 mmHg at rest, mean pulmonary capillary wedge pressure less than or equal to 15 mmHg, and PVR greater than three Wood units.[20] Prevalent and incident patients were included. Patients with WHO Groups 2 through 5 disease were excluded. The selection of subjects and sub-group breakdown is further depicted in Fig. 1.

Controls were recruited from VUMC via the ResearchMatch (www.researchmatch.org) program, the nationwide, disease-neutral research volunteer registry. Unaffected *BMPR2* mutation carriers (unaffected mutation carriers) were recruited from the VFPHR. There were no exclusion criteria for controls other than self-reported "healthy" and absence of cardiovascular or respiratory disease. The majority of healthy controls were from the middle Tennessee area, similar to most recruited PAH patients and unaffected mutation carriers.

Data collection

Providers at Vanderbilt developed a questionnaire and conducted a nurse-led interview in order to obtain information regarding reproductive history and sex hormone exposure (androgen and estrogen containing compounds) in patients and healthy volunteers. The design of the questionnaire was guided by previous validated epidemiologic studies in CVD and hormone responsive cancers, such as the Nurses' Health Study I and II. These questionnaires were administered at the initial visit at VUMC, at the 2012 and 2014 PHA International Conferences Research Room, or over the phone if the subject was enrolled from another center. The specific questions regarding reproductive factors and sex hormone exposures can be viewed in the supplemental materials. Subjects were asked to estimate the total duration of therapy for each agent but not about specific dosing or

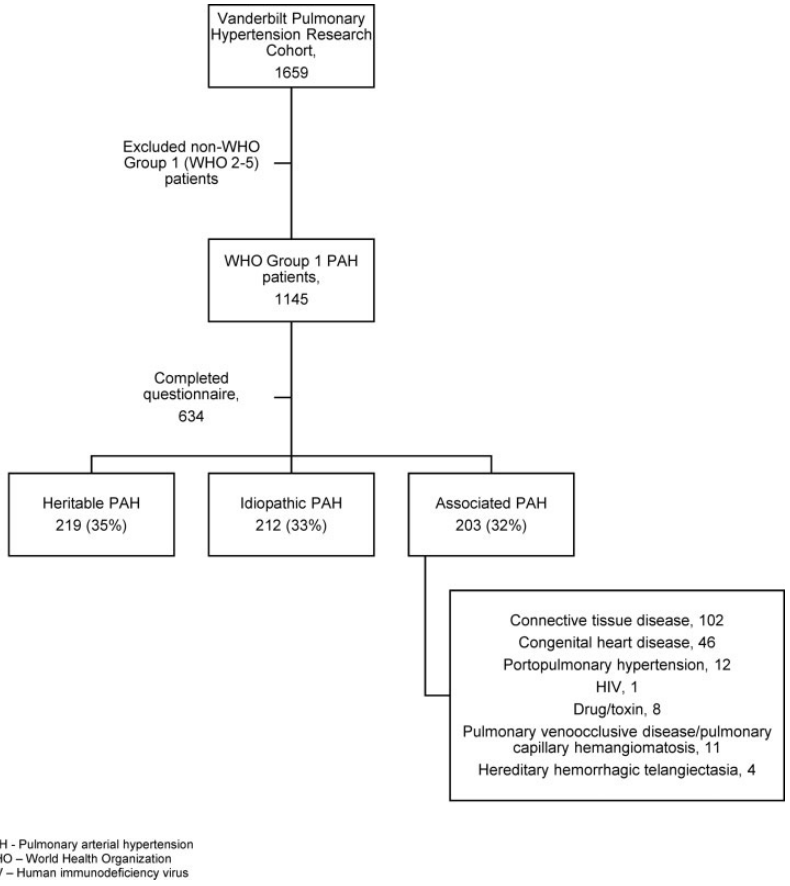

Fig. 1. Subject selection and PAH WHO Group 1 sub-group breakdown. Inclusion criteria include patients with idiopathic, heritable, or associated pulmonary arterial hypertension as designated by their treating physicians and meeting traditional, international diagnostic criteria.

composition of these hormone therapies given anticipated difficulty in accurate recall. The interviewers were not blinded to case status or study hypotheses.

Additional history was obtained on enrolled patients including disease severity variables such as baseline 6-minute walk distance and right heart catheterization data, as well as other relevant medical history including body mass index (BMI) and comorbidities.

Data analyses

Descriptive statistics including mean, standard deviation, median, and inter-quartile range for continuous variables, and percent and frequency for categorical variables were summarized for subject groups. We did not impute missing data. In the analyses, only subjects with non-missing value for the variables are included.

With approximately 200 subjects in each of the PAH groups and 132 in the control group, the study has 80% power to detect an effect size of 0.32 for continuous variables or odds ratio of 2.5 for categorical variables.

The Kruskal–Wallis Test or Pearson chi-square test was used for between group comparisons. Unadjusted logistic regression analysis was used to compare patient groups to healthy controls. This was then adjusted for potential confounders including age, race (white was used as the reference when adjusting for race), and BMI. Linear regression analyses were conducted to assess the association between disease severity variables and sex hormone exposures. This analysis was also adjusted for PAH sub-group (idiopathic, heritable, or associated). The main analyses were performed in female subjects only.

Results

Characteristics of cases and controls

Overall 634 patients, 132 healthy controls, and 27 unaffected mutation carriers completed the questionnaire. The breakdown of PAH patients included 219 with heritable (35%), 212 with idiopathic (33%), and 203 with associated PAH (32%) (Fig. 1 and Table 1).

Females comprised 80% of the patients and 73% of controls. The mean age at time of questionnaire for patients and controls was 46.9 and 38.9 years, respectively. The age of unaffected mutation carriers was higher (57.9 years) compared to patients with heritable disease (38.6 years). The majority of patients and controls self-identified as white.

Table 1. Demographics and baseline characteristics of all participants by study group.

	Heritable PAH	Idiopathic PAH	Associated PAH	Unaffected mutation carriers	Healthy controls
Subjects, N	219	212	203	27	132
Age at questionnaire, years[a]	38.6 ± 15.5	49.3 ± 14.4	51.6 ± 14.3	57.9 ± 16.5	38.9 ± 18.5
Gender[a]					
Female	157 (72)	180 (85)	170 (84)	17 (63)	97 (73)
Male	62 (28)	32 (15)	33 (16)	10 (37)	35 (27)
Race[a]					
White	207 (95)	181 (85)	176 (87)	25 (93)	101 (77)
African American (AA)	7 (3)	17 (8)	15 (7)	1 (3.5)	16 (12)
Non-White or AA[b]	5 (2)	14 (7)	12 (6)	1 (3.5)	15 (11)
BMI (kg/m^2)[a]	27.8 ± 6.3	28.6 ± 5.6	28.0 ± 6.8	28.1 ± 5.5	26.9 ± 6.3

Note: Total number of subjects who completed questionnaire is 634. Subjects with PAH are further delineated by WHO Group 1 sub-group. PAH: pulmonary arterial hypertension; BMI: body mass index in kilograms per meter squared.
[a]Data are reported as mean ± standard deviation for continuous variables and count (%) for categorical variables.
[b]Additional races include those identified as Asian, American Indian/Alaska Native, more than one race, and Unknown.

Table 2. Endogenous estrogen exposures in female participants by study group.

	Heritable PAH	Idiopathic PAH	Associated PAH	Unaffected mutation carriers	Healthy controls	P-value[a]
Total subjects, N	157	180	170	17	97	
Age at menarche, years[b]	12.3 ± 1.9	12.5 ± 1.4	12.8 ± 1.8	12.8 ± 1.5	12.4 ± 1.7	0.23
Pregnancies, n[b]	2.0 ± 2.1	1.8 ± 1.5	2.0 ± 1.6	2.4 ± 1.6	1.7 ± 1.7	0.35
Time breastfeeding, months[b]	2.7 ± 7.5	2.8 ± 6.7	3.0 ± 7.0	1.6 ± 2.8	5.8 ± 7.9	0.12
Oophorectomy[b,c]	8 (89)	14 (67)	8 (67)	2 (100)	6 (67)	0.62
Post-menopausal[b,c]						
No	96 (73)	73 (44)	45 (28)	3 (20)	64 (72)	
Yes	16 (12)	57 (35)	72 (44)	10 (67)	24 (27)	
Unknown	20 (15)	35 (21)	45 (28)	2 (13)	1 (1)	
Unadjusted OR (95% CI)	0.45 (0.22–0.90)	2.08 (1.17–3.78)	4.57 (2.53–8.48)			
Adjusted OR (95% CI)[d]	0.64 (0.14–2.66)	1.36 (0.50–3.81)	*4.54* (1.67–12.97)			

Note: Subjects with PAH are further delineated by WHO Group 1 sub-group. Questionnaire can be seen in supplemental materials. PAH: pulmonary arterial hypertension; OR: odds ratio; CI: confidence interval.
[a]Pearson or Kruskal–Wallis Test performed for between group comparisons.
[b]Data are reported as mean ± standard deviation for continuous variables and count (%) for categorical variables. Only subjects with non-missing values were included in the analyses.
[c]The number of study subjects answering each exposure question was not consistent; therefore, % is of the total subjects in each study group that answered the question.
[d]Logistic regression analyses use healthy control group as reference; adjusted for age, race, and BMI.

The average BMI of patients was 28.2 kg/m^2 and healthy controls was 26.9 kg/m^2.

There were differences between the female patient groups when subcategorized according to the underlying cause of their PAH, heritable versus idiopathic versus associated (Supplemental Table 1). Consistent with prior cohort reports, heritable patients were younger and had more severe disease at diagnosis as indicated by higher mean PAP and PVR, as well as lower cardiac index.[21]

Reproductive factors and endogenous sex hormone exposure

Subjects were asked about previously identified reproductive and endogenous factors that determine lifelong estrogen exposure (Table 2). Female patients and controls were similar in terms of menarche age and number of pregnancies. There was a higher duration of breastfeeding in controls (~5.8 months) despite the number of pregnancies

being similar. There was no difference between patients and controls regarding rates of oophorectomy but number of total responses to this question was low (53).

There was a difference between the groups in regard to menopausal status. Patients with associated disease were much more likely (OR 4.5) to be post-menopausal when compared to healthy controls, even when adjusted for age, race, and BMI.

Exogenous sex hormone exposure

All subjects were asked about previous exogenous sex hormone exposures, including androgen and estrogen containing compounds. Most female patients and controls reported having taken OCPs in their lifetime, 61–83% as depicted in Table 3. The only significant difference across the groups in terms of ever taking OCPs was between unaffected mutation carriers and controls, with an OR of 0.13 when corrected for age, race, and BMI. A smaller number of both patients and controls reported ever taking HRT, with 25% ever-use in idiopathic and 35% ever-use in associated PAH (APAH) when compared to healthy controls at 18%.

There was no difference between the groups in terms of duration of exogenous hormone exposure, OCPs, or HRT (Fig. 2a and b). Duration of OCP use was similar across the groups with the majority of patients and controls using for between one and five years. The duration of HRT use, albeit in a much smaller number of patients and controls, was similar across the groups with the majority using this therapy for one to five years.

Very few participants (male or female) reported androgen or body building drug use, eight controls (seven women and one man) and one subject (female) with APAH.

Exposures and disease severity variables at diagnosis

Table 4 demonstrates the sex hormone exposure variables as predictors of disease severity at diagnosis in female patients. None of the disease severity variables, including age at diagnosis, mean RA pressure, mean PA pressure, PVR, thermodilution cardiac index, or time to death, was significantly affected by any exposure variable after adjusting for PAH sub-group.

Discussion

This is an unmatched case–control study in PAH describing various reproductive factors and sex hormone exposures, including hormone-containing pharmaceuticals, and evaluation of their potential association with disease status and subject characteristics at diagnosis. As reflected in contemporary registry studies of PAH, there is a distinct female predominance in the patients of this cohort.

Patients and controls in this study had similar exposure to previously identified reproductive and endogenous factors that determine lifelong estrogen exposure. We did find that those patients with associated PAH were older (~50 years) and more likely to be post-menopausal when compared to controls. Patients that reported being post-menopausal at diagnosis had significantly lower PAP and PVR but these correlations were not statistically significant when adjusted for PAH sub-group, heritable versus idiopathic versus associated. The overall trend toward older, post-menopausal patients deserves more investigation as it may have important implications given that significant hormonal fluctuations occur in this age group, especially in the perimenopause transition.[22] There are hormonal influences on exercise capacity and functional status that may be the

Table 3. Exogenous estrogen exposures in female participants by study group.

	Heritable PAH	Idiopathic PAH	Associated PAH	Unaffected mutation carriers	Healthy controls
Total subjects, N	157	180	170	17	97
Taken OCPs[a]					
Yes	59 (61)	138 (83)	122 (78)	10 (62)	72 (78)
No	30 (31)	26 (16)	35 (22)	6 (38)	20 (22)
Unadjusted OR (95% CI)	0.55 (0.28–1.05)	1.47 (0.76–2.81)	0.97 (0.51–1.79)	0.46 (0.15–1.50)	
Adjusted OR (95% CI)[b]	1.43 (0.50–4.74)	1.10 (0.52–2.31)	0.74 (0.35–1.52)	*0.13* (0.02–0.69)	
Taken HRT[a]					
Yes	20 (20)	43 (25)	54 (35)	5 (31)	17 (18)
No	79 (79)	125 (74)	100 (65)	11 (69)	75 (82)
Unadjusted OR (95% CI)	1.12 (0.54–2.31)	1.52 (0.82–2.91)	2.38 (1.30–4.54)	2.01 (0.57–6.33)	
Adjusted OR (95% CI)[b]	2.10 (0.64–6.74)	0.90 (0.38–2.20)	1.12 (0.48–2.67)	0.15 (0.01–1.17)	

Note: Subjects with PAH are further delineated by WHO Group 1 sub-group. Questionnaire can be seen in supplemental materials. PAH: pulmonary arterial hypertension; OCP: oral contraceptive pill; HRT: hormone replacement therapy; OR: odds ratio; CI: confidence interval.

[a]Data are reported as count (%) for categorical variables. The number of study subjects answering each exposure question was not consistent; therefore, % is of the total subjects in each study group that answered the question.

[b]Logistic regression analyses use healthy control group as reference; adjusted for age, race, and BMI. Only subjects with non-missing values were included in the analyses.

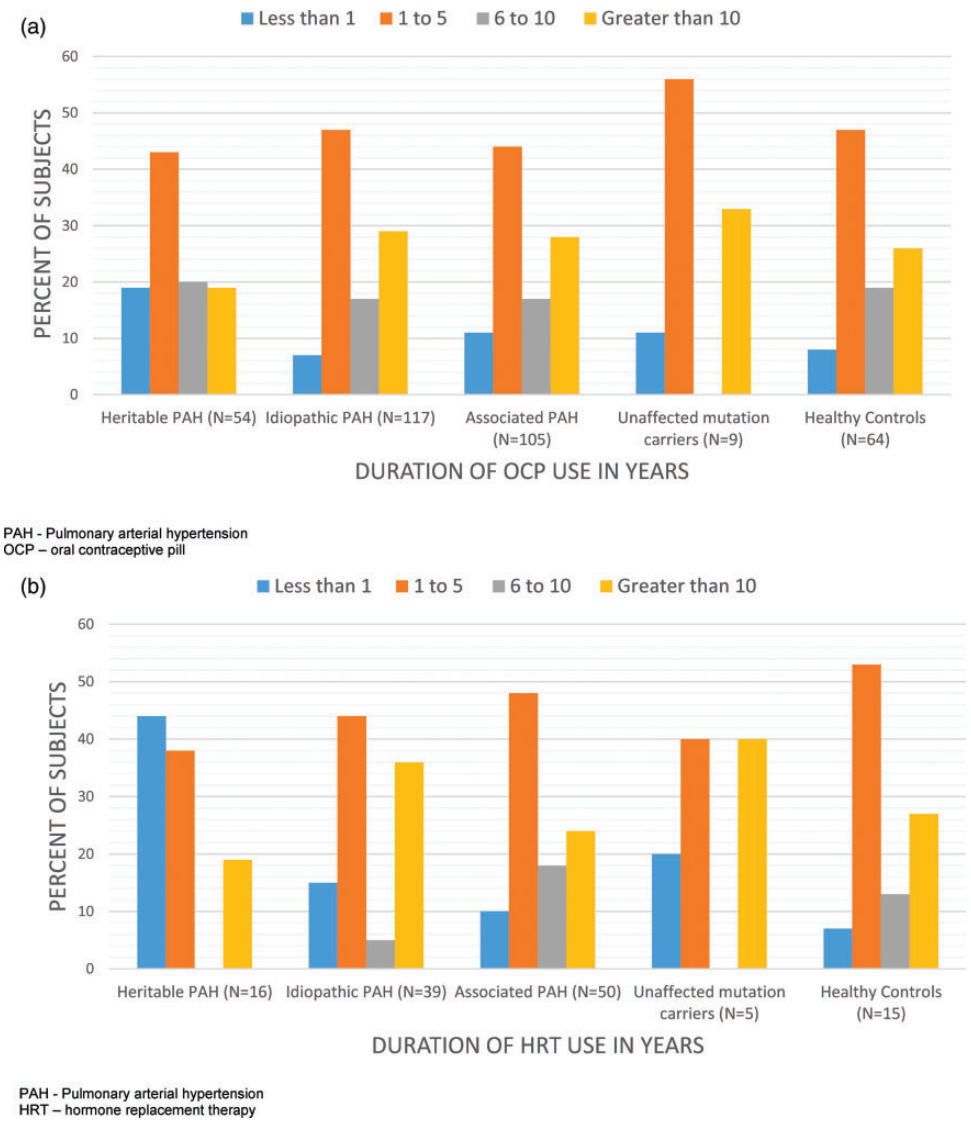

Fig. 2. (a) Duration of oral contraceptive use in female participants by study group. Subjects with PAH are further delineated by WHO Group I sub-group. Subjects were asked to estimate the total duration of therapy for each agent but not about specific dosing or composition of these hormone therapies given anticipated difficulty in accurate recall. (b) Duration of hormone replacement therapy use in female participants by study group. Subjects with PAH are further delineated by WHO Group I sub-group. Subjects were asked to estimate the total duration of therapy for each agent but not about specific dosing or composition of these hormone therapies given anticipated difficulty in accurate recall.

result of changes in endothelial function and/or RV performance, which could impact the timing and severity of presentation of PAH.[23,24] Alternatively, the observation that menopause is associated with more favorable hemodynamics may be due to sicker patients dying before menopause.

Sex hormone-containing pharmaceuticals, both androgens and estrogens, provide the most obvious and quantifiable environmental exposure in PAH patients. As we found very low rates of androgen exposure in both cases and controls, there is a focus on estrogen as the most female-specific exogenous sex hormone exposure in our study. There were no differences between patients and controls in "ever use" of OCPs, ranging from 61 to 83%. This is similar to national data from the Centers for Disease Control and Prevention (CDC); from 2006 to 2010 the amount of sexually experienced women age 15–44 who have "ever used" birth control pills was 89% in non-Hispanic whites, 78% in non-Hispanic blacks, and 73% in Hispanics. The percent of women aged 15–44 currently using contraception in the U.S. from 2011 to 2013 included ~26% using an OCP.[25] A notable percent of patients with both idiopathic (29%) and associated (28%) PAH reported a greater than 10-year duration of use of OCPs, but this was similar to 26% of controls reporting >10 years of use.

There were relatively high rates of HRT "ever use" in patients with idiopathic (25%) and associated (35%) PAH, as well as controls (18%) in this study population but no significant difference between the groups when corrected for age, race, and BMI. The use of HRT dramatically

Table 4. Relationship between disease severity variables at diagnosis and sex hormone exposures in female subjects with WHO Group 1 PAH.

Variable	Estimate of exposure	P-value
Age at diagnosis (years)		
Menarche age	−0.05	0.88
Pregnant within six months of diagnosis	−3.52	0.14
Menopausal	3.08	0.14
OCP ever use	1.30	0.38
HRT ever use	−0.48	0.75
RA pressure (mmHg)		
Menarche age	−0.24	0.39
Pregnant within six months of diagnosis	−3.02	0.15
Menopausal	−0.80	0.64
OCP ever use	1.70	0.19
HRT ever use	0.57	0.66
PA pressure (mmHg)		
Menarche age	0.03	0.97
Pregnant within six months of diagnosis	−6.91	0.11
Menopausal	−3.35	0.38
OCP ever use	4.70	0.10
HRT ever use	−2.17	0.44
PVR (Wood units)		
Menarche age	0.03	0.91
Pregnant within six months of diagnosis	−0.82	0.67
Menopausal	−1.81	0.38
OCP ever use	0.78	0.61
HRT ever use	−0.23	0.87
Thermodilution cardiac index (L/min/m^2)		
Menarche age	0.02	0.70
Pregnant within six months of diagnosis	0.41	0.17
Menopausal	0.01	0.98
OCP ever use	0.02	0.93
HRT ever use	0.03	0.88
Time to death (years)		
Menarche age	0.04	0.94
Pregnant within six months of diagnosis	1.64	0.84
Menopausal	−3.97	0.53
OCP ever use	1.40	0.45
HRT ever use	1.06	0.60

Note: Linear regression was used to assess the association between disease severity variable and sex hormone exposure factors (predictors). This was adjusted for PAH sub-group (idiopathic, heritable, CTD-associated, or other associated). Dichotomous variables were compared yes versus no. PAH: pulmonary arterial hypertension; OCP: oral contraceptive pill; HRT: hormone replacement therapy; RA: right atrium; PA: pulmonary artery; PVR: pulmonary vascular resistance.

rose in the U.S. from 16 million dispensed prescriptions in 1966 to 90 million in 1999, but then declined after the publication of the Women's Health Initiative study in 2002 and by 2005 sales were down by 50%. National Health and Nutrition Examination Survey (NHANES) data from 1988 to 1994 revealed 44% of women "ever used" HRT and 22% with current use, whereas more recent data from 2009 to 2010 suggests only 5% of women are currently using HRT.[26] There is limited contemporary data available on "ever use" of HRT, but the use in our current patient and control populations appears higher than the most recent national data, suggesting this may be a unique exogenous risk factor, especially for idiopathic or associated PAH.

Given the suggested increase and perhaps longer duration of use of OCPs and HRT in PAH patients, it will be critical to know whether these exogenous hormone sources, in addition to previously recognized and yet unknown genetic susceptibilities, truly confer additional risk of developing pulmonary vascular disease. We speculate that exogenous and/or endogenous exposures that perturb a normal physiologic hormonal balance may be harmful over time, although the mechanism of this impact is unclear. Various studies in women with other vascular diseases including coronary artery disease (CAD), venous thromboembolism, and preeclampsia have also demonstrated association with estrogen receptor (ER) polymorphisms.[27–30] There are also data to suggest certain ER polymorphisms in post-menopausal women with CAD have an augmented response of high-density lipoprotein (HDL) cholesterol to HRT.[31] Additionally, polymorphisms in the genes involved in the synthesis (CYP19A1) and metabolism (CYP1A1, CYP1B1) of estrogen have been implicated in a range of female predominant diseases including hereditary PAH and breast cancer.[32,33] Future investigations into how genetic variability, in combination with reproductive factors and sex hormone exposures, plays a role in both onset and severity of PAH are necessary. This study was not designed to explore this interplay but larger multi-center studies, such as the ongoing United States Pulmonary Hypertension Scientific Registry, may provide important advances in that regard.

Limitations

Despite its strengths, this study contains several limitations. Almost 50% of eligible patients did not complete the questionnaire, introducing some potential for selection bias. As this is an unmatched case–control study, there is risk of confounding from many factors. While we attempted to control for age, race, and BMI, there is still the potential for confounders that limit the assessment of causal association of these exposures. The generalizability of the results is also affected by the few minority patients and controls. Although the enrollment of cases and controls, including unaffected mutation carriers, of similar demographics minimizes recall bias, such bias remains a concern for any historical study. The significant lag time between potential exposures and disease may also contribute to recall bias, although this is relatively similar for cases and controls. In addition, there were known limitations in the questionnaire employed, which pragmatically balanced the depth and number of questions with the likelihood of completion. As a result, there was a

lack of documentation in the questionnaires regarding some key factors that may influence lifetime exposure to sex hormones, such as menopausal age and age at first pregnancy. There was no clarification of current use of OCPs or HRT, which may be important as many of the associations between use of these exogenous compounds and increased risk of breast cancer or CAD are with current usage. Patients may be aware that estrogen exposure, OCPs and HRTs, has been linked to PAH development and therefore may overestimate their prior exposures compared to controls. Another factor that must be noted is that most controls and heritable patients were from the mid-South region of the U.S.—there may be regional variations in prescribing practices of OCPs and HRT over the years that influences the exposures these subjects may have. In addition, the potency of OCPs and HRT in terms of estrogen type and dose has declined over time, introducing another potential source of bias. Finally, there is an over representation of subjects with heritable PAH in this study, relative to the generally expected percentage of other PAH forms.

Conclusion

It has been established that female sex is a major risk factor for the development of PAH but some males have worse survival. Endogenous and exogenous exposure to sex hormones, such as estrogens, may modify the risk of developing PAH or severity of disease. This large, single-center case–control study found no major differences in endogenous or exogenous sex hormone exposures between patients and controls. However, post-menopausal status or HRT use may have implications on disease severity. Sex differences and hormonal influences on the right ventricle likely account for some of the variation in severity and survival. Large, multi-center studies are needed to further evaluate the impact of OCP and HRT use in this population. Such studies may also benefit from a careful integration of sex hormone exposures with inherent risk factors, including genetic composition.

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Conflict of interest

The author(s) declare that there is no conflict of interest.

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Supplemental material

Supplemental material for this article is available online.

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