Semen Parameters Among Transgender Women With a History of Hormonal Treatment

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OBJECTIVE To report on the semen quality parameters in transgender women who pursued semen cryopreservation either in the presence or absence of gender-affirming hormonal medication.

MATERIALS AND METHODS This retrospective cohort study was conducted as a chart review of consecutive transgender women presenting for semen cryopreservation between January 1, 2012 and March 31, 2018. Demographic data and semen parameters were assessed. Primary outcomes were the semen parameters in subjects with either no exposure, previous exposure or current exposure to gender-affirming hormonal medication.

RESULTS Twenty-eight patients presented for semen cryopreservation and produced 69 specimens. Using a Kruskal-Wallis test, semen analyses were compared between patients who had never used gender-affirming hormonal medication, those who had previously used hormonal medication but discontinued prior to specimen collection, and those who used medication at the time of specimen collection. Median semen parameters for each group were as follows: volume—2.7 mL, 2.1 mL, 0.9 mL, respectively (P = .12); concentration—63.6 M/mL, 39.0 M/mL, 2.4 M/mL, respectively (P < .01); percent motility—51.5%, 34.3%, 15.6%, respectively (P < .01); and the total motile count was 63.2 M,39.1 M, 0.2 M, respectively (P < .01). Fifteen specimens were collected after discontinuing hormonal medication with a mean discontinuation period of 4.4 months.

CONCLUSION Specimens collected in the presence of hormonal medication were associated with abnormal semen parameters. Specimens collected after discontinuation of gender-affirming treatments were comparable to transgender women who had never used hormonal medication. Transgender women should be counseled about the potential impact of gender-affirming hormones and to consider fertility preservation prior to gender-affirming treatments. UROLOGY 124: 136–141, 2019.

Recognition of the importance of fertility preservation for transgender, nonbinary, and nonconforming people has grown in recent years. The Williams Institute estimates that there may be as many as 1.4 million transgender people in the United States.[1] Urologists are the primary surgeons performing gender-affirming orchiectomies for transgender women. The World Professional Association for Transgender Health and the Endocrinology Society Guidelines recommend a discussion of fertility preservation options prior to the initiation of treatment and prior to any fertility altering surgery.[2,3] Transgender female patients have demonstrated a desire for more information about their fertility preservation options and feel that it is important to offer these services to other transgender women.[4,5] Urologists may be tasked with managing the hormonal and fertility considerations, especially for transgender women.[6] Furthermore, transgender women desiring orchiectomy have often been on gender-affirming hormonal treatment for an extended period of time but clinicians have minimal data to utilize when guiding a discussion about fertility preservation in this population.

While semen cryopreservation has been widely utilized for fertility preservation in adult men facing gonadotoxic treatment for a malignancy, there is little information about the adequacy of semen cryopreservation for transgender women. As highlighted by the Endocrine Society Guidelines, "restoration of spermatogenesis after prolonged estrogen treatment has not been studied."[7] No study to date has described the impact of gender-affirming hormonal medication on semen parameters in a cohort of transgender women. We present a cohort of transgender women of whom 10 patients had a history of gender-affirming hormonal medication use by utilizing

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at least 1 mg of estradiol daily. The objective of this study was to describe semen parameters in transgender women in the presence or absence of gender-affirming hormonal treatment.

MATERIALS AND METHODS

Study Population

All patients identifying as transgender women who pursued semen cryopreservation between January 1, 2012 and March 31, 2018 were included in this study. All subjects, except for 1 transgender woman, were able to ejaculate. Specimens were analyzed even if ultimately no sperm were identified.

Gender-affirming hormonal medication usage was defined as the consistent use of estradiol for the purposes of alleviating gender dysphoria or improving feminine physical characteristics congruent with ones identified gender. Subjects were separated into 3 groups: subjects who had never utilized gender-affirming hormonal treatment (n = 18), those who had previously utilized gender-affirming hormonal treatment but discontinued prior to specimen collection (n = 3), and subjects who were on gender-affirming medical treatment at the time of specimen collection (n = 5). The University of California, San Francisco Committee on Human Research (IRB-10-04868) approved this study.

Outcome Measures

We collected the demographic data including age at specimen collection, race, and referral source. Any history of gender-affirming medication use, the medication regimen, the date of initiation, and date of discontinuation were obtained from chart review. Semen parameters including the number of abstinence days, vials stored, volume, total motility, concentration, and total motile count (TMC) were compared between the 3 groups. For subjects who had a subsequent orchiectomy, histologic assessment was reported.

Statistical Analysis

The data were determined to be non-normally distributed; therefore, nonparametric tests were applied for analysis. Mean semen parameters of subjects who utilized gender-affirming treatment before, during, or after semen specimen collection were compared using a Kruskal Wallis test. Significant differences between the 3 groups were determined with a Dunn's pairwise comparison. All testing was performed at the 0.05 level of significance. Statistical analysis was performed using Stata *14.2*. Figures were created using R 3.4.3 and represent specimen data.

RESULTS

Patient Characteristics

Thirty patients presented for cryopreservation. Two patients were excluded from analysis; 1 patient had a vasectomy reversal 3 months prior to specimen collection and inadequate information about her gender-affirming treatment plan and the other patient was unable to ejaculate. The 28 remaining patients who presented for semen cryopreservation were included in the analysis. These 28 subjects produced a total of 69 specimens. The majority of subjects (n = 18) presented prior to the initiation of gender-affirming medication. Three subjects had previously used gender-affirming medication but discontinued prior to semen cryopreservation. The mean discontinuation period for these subjects was 4.4 months. Seven subjects used gender-affirming medication at the time of specimen collection. There was a difference in the age at production between the 3 groups (P = .04). Subjects who had never used gender-affirming medication were younger with a median age of 22.3 years compared to 31.3 years in subjects with a history of medication usage although this difference was not statistically significant (P = .17). Demographics are summarized in Table 1.

The majority of subjects (n = 16) were established patients within our academic medical center. Referrals for cryopreservation came from primary-care, a pediatric and adolescent gender clinic and urology. A smaller proportion of subjects were self-referred (n = 5). One patient presented for fertility consultation with her partner but chose to cryopreserve sperm before restarting hormonal medication and attempting to conceive spontaneously (Table 1).

Usage Patterns of Gender-Affirming Medication

Subjects with a history of gender-affirming hormonal medication use utilized a combination of an estrogen and antiandrogen. Estrogen preparations included oral estradiol 2-6 mg, transdermal estradiol 300 mcg, or conjugated estrogen 0.625 mg daily. Subjects used spironolactone 50-100 mg twice daily and variably used finasteride 2.5-5 mg or micronized progesterone 100 mg daily. The median length

Table 1. Demographics

	Never Used Gender Affirming Medication (n = 18)	Previously Used Gender Affirming Medication (n = 3)	Current Gender Affirming Medication Use (n = 7)	*P* Value
Age* (y)	22.3 [18-27.6]	31.3 [23.4-39.9]	28.9 [25.0-30.1]†	.04
Race				
Caucasian (Non-Hispanic)	33% (6/18)	67% (2/3)	71% (5/7)	
Caucasian (Hispanic)	0%	0%	14% (1/7)	
Asian	28% (5/18)	33% (1/3)	0%	
African American	0%	0%	0%	
Not disclosed	39% (7/18)	0%	14% (1/7)	
Number of visits*	2 [2-3]	2 [2-3]‡	1 [1-3]	.03
Referring provider				
Primary care	78% (14/18)	67% (2/3)	29% (2/7)	
Urology	11% (2/18)	0%	57% (4/7)	
Self	11% (2/18)	33% (1/3)	14% (1/7)	

* Reported as median value and interquartile range, some percentages do not sum to 100% due to rounding.
† Pairwise comparison to never used P value = .09; previously used P = .93.
‡ Pairwise comparison to subjects who never used medication P value < .01; currently using medication P = .19.

Table 2. Usage of gender affirming hormonal regimens

	Gender-Affirming Medication					Semen Parameters				
Current Users	Estradiol (mg)	Sprl.(mg)	Other	Time On (mo)	Time Off (mo)	Samples (n)	Conc.(M/mL)	Vol. (mL)	Motility %	TMC
1	1	100		7		1	11.92	2.8	42	14.01
2	4	100	Prg. 100	49		5	0.01	0.4	14.8	<0.01
3	4	200		12		1	n/a	0.1	n/a	n/a
4	1*	100	Fin 5.0	66		1	n/a	0.02§§	n/a	n/a
5	6	200	Fin. 2.5	Unk		1	n/a	0.2	n/a	n/a
6	4	0		2		2	33.5	4.2	42.5	54.4
7	2	50		13		2	13	1.3	30.5	4.94
Previous Users	Estradiol	Sprl.	Other	Time On (mos)	Time Off (mos)	Samples (n)	Conc. (M)	Vol.	Motility %	TMC
1	0.3†	100		41	6.5	4	40.95	2.85	32.5	31.78
2	0.6††	100		52	6	5	67	2.1	27	39.12
3	4	100		42	3	6	29.9	1.5	45.5	19.89

Semen parameters are reported as median values for participants with multiple specimens.
Conc, concentration; Fin., finasteride Unk—start date not specified in chart review; Medications Sprl, spironolactone; Prg., micronized progesterone; TMC, total motile count.
* Estradiol valerate 10 mg IM every 10 days.
† Estradiol 300 mcg transdermal
†† Premarin 0.625 mg.
§§ One drop of specimen visualized in collection cup, diluted with media, no sperm identified.

of medication usage in subjects who continued their medication during specimen collection was 30 months compared to 42 months for prior users. Three subjects discontinued gender-affirming medication prior to specimen collection. The median time of discontinuation was 4.4 months with discontinuation periods ranging from 3 to 6.5 months. Treatment data are summarized in Table 2.

Impact of Gender-Affirming Medication on Semen Analyses

Sixty-nine samples were collected in the following manner: 41 prior to gender-affirming medication, 15 after a period of discontinuation of gender-affirming medication, and 13 while on gender-affirming medication. The median specimen volume for subjects never exposed to gender-affirming hormones was 2.7 mL [25% = 1.6; 75%=3.7], for subjects who previously used hormonal medication it was 2.1 mL [1.6-2.7] and for subjects on hormonal medication at the time of specimen collection, the median volume was 0.9 [0.1-2.8]. There was no significant difference in the volume of specimens between the 3 groups ($P = .12$; Table 3).

There was a difference in the semen concentration between the 3 groups; subjects never exposed to gender-affirming hormones had a median concentration of 63.6 M/mL [31.9-79.0], those with a history of prior gender-affirming hormone use had a concentration of 39.0 M/mL [38.0-66.4] and current users had a median concentration of 2.4 M/mL [0-13] ($P < .01$). Differences were also seen in the percent of motile sperm between the 3 groups with subjects who never used gender-affirming medication having a motility of 51.5% [36.8-58.5], prior hormonal medication users having a motility of 34.3% [32.4-46.8], and current users having a motility of 15.6 [0-42.0] ($P < .01$). Finally, there were clear differences in the TMC between the 3 groups with those who had never used gender-affirming medication having the highest TMC 63.2 M [36.0-134.5], an intermediate value for prior hormone users of 39.1M [31.0-41.1] and a lower TMC of 0.2 M [0-14.0] ($P < .01$) among current hormone users (Table 3; Fig. 1).

On pairwise comparison, differences were attributable to lower values of specimens collected in the presence of gender-affirming medication compared to those with no prior exposure to gender-affirming medication. Additionally, there was a significant difference in semen concentrations between transgender women who previously used gender-affirming medication and transgender women using gender-affirming medication at specimen collection. Specimens collected in the presence of gender-affirming medication also had a lower number of vials stored ($P < .01$; Table 3).

Impact of Gender-Affirming Hormones on Testicular Pathology

Most subjects with follow-up at our academic center commenced usage of gender-affirming medications or surgery after fertility preservation was complete. Two subjects presented for specimen collection within 24 hours of their scheduled orchiectomies. One subject was treated with estradiol 6 mg, spironolactone 200 mg, and finasteride 2.5 mg for 7 years. On pathology, testicular specimens were noted to have partially reduced spermatogenesis. Furthermore, a 4.5 cm adrenocortical rests was identified adjacent to the left spermatic cord. A second patient was treated with estradiol valerate IM, spironolactone 100 mg and finasteride 5 mg for 66 months prior to specimen collection. Her testicular pathology demonstrated atrophy, fibrosis, and hypospermia bilaterally. Evaluation of a TESE specimen identified no sperm.

Table 3. Semen parameters in transgender women in the presence or absence of gender-affirming hormonal medication

	Never Used Gender Affirming Medication (n = 18)	Previously Used Gender Affirming Medication (n = 3)	Current Gender Affirming Medication Use (n = 7)	*P* Value
Abstinence days	3.5 [3-4]	3.0 [2.75-4]	4.1 [3-5]	.50
Vials stored	4 [3-4.5]	3.4 [3-4]	1.6 [0-4.0]*	< .04
Percent motility	51.5 [36.8-58.5]	34.3 [32.4-46.8]	15.6[0-42.0]†	< .01
Concentration (M/mL)	63.6 [31.9-79.0]	39.0 [38.0-66.4]	2.4 [0-13]‡	< .01
Volume (mL)	2.7 [1.6-3.7]	2.1 [1.6-2.7]	0.9 [0.1-2.8]	.12
Total motile count (millions)	63.2 [36.0-134.5]	39.1 [31.0-41.1]	0.2 [0-14.0]§	< .01

Reported as median values [25th percentile-75th percentile].
* Pairwise comparison of vials stored between current users to never used *P* value = .04; current users to previously used *P* = .48.
† Pairwise comparison of percent motility between current users to never used *P* value < .01; current users to previously used *P* = .29.
‡ Pairwise comparison of concentration between current users to never used *P* value < .01; current users previously used *P* = .04.
§ Pairwise comparison of total motile count between current users to never used *P* value < .01; current users to previously used *P* = .19.

DISCUSSION

Use of gender-affirming hormonal medication at the time of specimen collection is negatively associated with parameters of semen quality. The majority of subjects in this cohort presented before commencement of gender-affirming hormonal medication in accordance with recommendations from World Professional Association for Transgender Health, the Endocrine Society, and the American Society of Reproductive Medicine.[7,3] The data clearly demonstrate that patients who cryopreserved sperm prior to the initiation of gender-affirming treatment had better semen parameters compared to transgender women on medication at the time of specimen collection. These data support many organization consensus statements which recommend fertility preservation prior to the commencement of hormonal medication.

Subjects utilizing gender-affirming hormonal medication during specimen collection had a spectrum of sample usability. Three subjects were azoospermic with extremely low volumes. One subject produced 5 specimens which were abnormal on all parameters but each time, sperm could be identified. It is conceivable that such specimens could be used for intracytoplasmic sperm injection in the future. Finally, 3 subjects produced specimens with semen parameters appropriate for intrauterine insemination or in vitro fertilization. Given the small sample size of our cohort, no conclusions can be made about a relationship between the length of gender-affirming medication usage and semen parameters. Subjects that discontinued gender-affirming medication for any period of time had specimens within the WHO reference range. The majority of specimens collected after a break from gender-affirming hormonal medication were appropriate for future use with intrauterine insemination.

The biologic plausibility for our results is supported by several studies on the impact of estrogen on testicular function. Gonadotropin suppression inhibits spermatogenesis as has been demonstrated in a study of 4 men with metastatic prostate cancer who used a GnRH agonist for at least 1 year followed by orchiectomy.[8] Some data suggest that long-term gonadotropin suppression in this

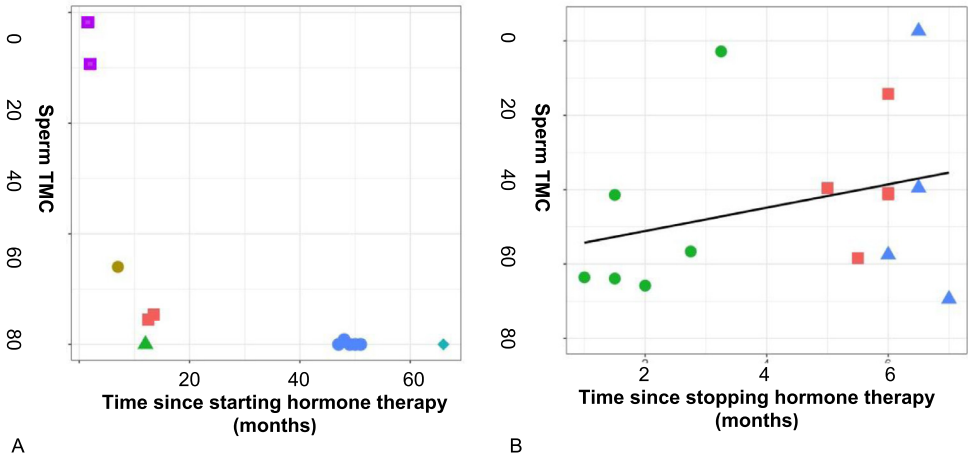

Figure 1. Scatter plot of specimen level total motile counts. Each shape/color represents a different subject; each data point represent a specimen collected from a given patient (**A**) dependent on the length of time gender-affirming medications were used among current users (**B**) dependent on the length of time gender-affirming medications had been discontinued at the time of specimen collection. (Color version available online.)

population may result in irreversible changes to testicular histology.[9] The data on testicular histology among transgender women are variable. Payer et al examined the testicular histology of 6 transgender women who used supplemental estrogen for 1-8.5 year. The authors demonstrated variable effects on Leydig cell morphology and at least partially arrested spermatogenesis.[10] In a review of 11 publications on the impact of gender-affirming medication on testicular function and histology, some data suggested a negative impact of treatment on spermatogenesis but 8 other publications were inconclusive.[11] One prior study from Hamada et al investigated the semen parameters of a cohort of transgender women who had yet to initiate gender-affirming medication. The authors demonstrated that even prior to gender affirmation, abnormalities of semen quality were identified.[12] Though the data are limited, there seems to be an association between estrogen exposure, androgen deprivation, and altered testicular function.

A strength of this study is that it captures patients at different points of their transition and represents the spectrum of patients that may present for gender-affirming medication or orchiectomies. To date, there are no data on semen quality among transgender women with a history of estrogen and antiandrogen exposure.

This study may have a positive impact on fertility management among transgender female patients. Our data suggest that within months of discontinuation of gender-affirming hormonal medication, transgender women may produce a specimen that can be used for intrauterine insemination or to potentially conceive spontaneously in the future. For patients in whom discontinuation of gender-affirming medication is intolerable but who are able to ejaculate, it may be possible to produce specimens appropriate for future use via in vitro fertilization. However, some subjects with prolonged use of gender-affirming hormonal medication in this cohort experienced azoospermia. Given the intrapatient variation in semen parameters between samples, it is hard to predict whether ejaculation itself would be a predictive of active spermatogenesis. Therefore, it should be emphasized that sperm cryopreservation prior to the commencement of gender-affirming hormones is ideal.

The retrospective design of this study limited the ability to collect data about the hormonal milieu at specimen collection. Future studies should ascertain whether thresholds of serum estrogen or testosterone levels may predict semen quality. In the present study, the lack of consistent data about sex steroid levels at specimen collection leaves the question of causality open for further investigation. In a 2015 study from Schneider et al the authors demonstrated that in a cohort of 108 transgender women seeking gender-affirming orchiectomies, serum testosterone levels were not associated with the histologic evidence of spermatogenesis. Though all transgender women had utilized gender-affirming hormones with a maximum of 6 weeks of discontinuation prior to surgery, 24% of testicular specimens showed evidence of normal spermatogenesis.[13] The precise mechanisms involved in the altered semen parameters cannot be determined from this study; however the clinical association with reduced total motile counts is clear. The authors do raise the possibility of medication adherence as a reason for variation in sex steroid hormones and subsequently testicular histology and spermatogenesis within their population. Serum sex steroid hormone levels in our cohort would be helpful to understand the role that medication adherence may have on varied semen parameters in our treatment groups.

Another limitation of this study is that all but one of the subjects who presented to our clinic was able to ejaculate. This is likely not representative of all transgender women. It is likely that because the majority of subjects were referred from within our academic center, that there may have been an element of provider bias and self-selection bias toward transgender women who could ejaculate. Though evidence of spermatogenesis in transgender women has been previously described, no studies have described the ability of this population to produce useful semen samples in a manner that is applicable for clinicians, surgeons, and patients. One question that arises from this data is what would be the optimal time of discontinuation of gender-affirming medication prior to fertility preservation. While it is plausible that a period of 60-70 days, the time needed for completion of spermatogenesis in nonexposed sperm, may be sufficient, our data are limited to 3 subjects who discontinued medication thus no absolute recommendations can be made from our data.

Finally, our data are limited in that we were unable to adjust for potential confounders that may impact semen parameters. Other etiologies to consider include lifestyle exposures such as drug use and nonmedical gender-affirming behaviors such as maintaining the penis and testes in an inguinal position,[14] often referred to as "tucking."[15] Additionally, our patient population may not be representative of the general population of transgender women in the United States. Previous studies have suggested that transgender people may have less access to routine health services as a consequence of anticipated stigma and actual discrimination.[16] Future studies should describe fertility preservation practices and semen parameters in a community setting that may be more representative of the general population.

CONCLUSION

Transgender women using gender-affirming hormonal medication at the time of fertility preservation have abnormal semen parameters and may be azoospermic, especially when the ejaculate volume is low. Discontinuation of gender-affirming medication may be associated with an improvement in semen parameters. Clinicians treating transgender women should counsel patients about the potential impact of gender-affirming hormones and to consider fertility preservation prior to the initiation of treatment.

SUPPLEMENTARY MATERIALS

Supplementary material associated with this article can be found, in the online version, at https://doi.org/10.1016/j.urology.2018.10.005.

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