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Varicocelectomy in adolescents — Does it safeguard future fertility? A single centre experience

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Summary

Background
There is paucity of literature comparing varicocelectomy to observant management amongst adolescent boys with hormonal and semen abnormalities resulting from high grade unilateral varicoceles and consequent testicular volume loss. Furthermore, it is not known whether surgical correction in such adolescents improves paternity rates in future compared to their non-operated cohort.

Objective
The primary objective was to compare adolescent boys with unilateral high grade varicocele with associated ipsilateral testicular volume loss who were operated versus those who were not, in relation to their fertility markers (hormonal, semen parameters, and testicular volume) over a 5 year follow up period. The secondary objective was to compare the paternity rates in the respective groups over long term.

Study design
This was a single center, retrospective study of a prospectively maintained database conducted from 2010 to 2020, based on a standardized protocol. All adolescent boys >15 years of age (middle and late adolescence), with grade II or III unilateral varicoceles with abnormal fertility markers, who were operated (Group A) and not operated (Group B) were included. The changes in hormonal assay, sonographic assessment, semen analysis at presentation, 1st year and the 5th year follow up amongst both the groups were collated and analysed. Primary paternity rates amongst both the groups was documented by telephonic or email conversations.

Results
Of the 182 boys referred for varicocele management, 110 boys (Group A −70 boys and Group B - 40 boys) satisfied our inclusion criteria and were analysed. Mean age at presentation amongst Group A boys was 16.5 years (15–18 years) and Group B boys was 16 years (15–18 years). Grade III varicoceles were more predominant amongst both the groups. There was a significant improvement in all Group A boys (operated) in the fertility markers from the time at presentation to the 5th year follow up ($p < 0.001$). In Group B, (boys not operated) there was no significant improvement in the above parameters. The testicular catch up growth was 92% at the 5th year follow-up in Group A and 42% in Group B. At long term follow-up, the paternity rate was 80% and 36% in Group A and B respectively.

Discussion and conclusion
In adolescent boys in whom hormonal assay, testicular volumes and semen characteristics are negatively affected by high grade unilateral varicoceles, surgical correction could normalize these values, thereby safeguarding their fertility in the long term.

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Introduction

Varicocele in adolescent boys has an incidence of up to 14–20% and presents as a therapeutic dilemma [1]. The prevalence of varicoceles amongst early adolescent boys (11–14 years) is 7.8%, and amongst middle and late adolescent boys (15–19 years) it is 14.1% [1]. Testicular atrophy secondary to a varicocele occurs in 7.3% of early adolescent boys and in 9.3% of middle and late adolescent boys thus signifying that, the incidence and prevalence of testicular atrophy in presence of varicocele increases with puberty [1]. The effects of varicoceles are long term, as well as progressive, leading to a decrease in testicular volume, in turn resulting in hormonal and semen abnormalities in at least some of the middle and late adolescents, with possible adverse effects on future fertility [2]. Hormonal factors such as serum follicle stimulating hormone, serum total testosterone [3,4]; sonographic details such as testicular volume, testicular atrophy index [5] and semen characteristics such as sperm concentration, % of sperm motility and DNA fragmentation index [4,6] play an important role in assessing these adverse effects. There is paucity of literature on the therapeutic benefits of varicocelectomy in middle and late adolescents with already pre-existing hormonal and semen abnormalities resulting from unilateral high grade varicocele and consequent testicular volume loss in relation to their paternity rates [7]. In published literature, controversy still exists in the management of these adolescent varicoceles with some who support early surgical intervention to others who support conservative management based on the testicular catch up growth [8,9]. Currently there are very few long term comparative studies of surgical treatment versus expectant management of this particular clinical scenario with regard to ultimate paternity rates [10]. Our clinical cohort included orthodox Indian adolescent boys, in whom early age of marriage with reduced usage of contraceptive measures was customary and a follow-up of these adolescent boys could provide us an early insight of the paternity potential after a varicocelectomy.

The primary objective of our study was to compare fertility markers i.e. hormonal assay, sonographic details, and semen characteristics amongst the middle and late adolescent boys with unilateral high grade varicoceles who were operated versus those who were managed conservatively, over a 5 year follow -up period. The secondary objective was to compare the paternity rates in the respective groups over long term.

Material and methods

This was a single center, retrospective study of a prospectively maintained database conducted by the Department of Pediatric urology from 2010 to 2020 based on a standardized protocol (Fig. 1). The recruitment and surgical treatment of these cases occurred between 2010 and 2014 and they were followed up for a period of 5 years from 2015 to 2020. All adolescent boys of >15 years of age (middle and late adolescent), with grade II, III unilateral varicoceles with abnormal fertility markers, who were operated as well as those who were managed conservatively were included. Adolescent boys with sub clinical or Grade I unilateral varicoceles, bilateral varicoceles, recurrent varicoceles, concurrent hydroceles, boys in whom semen samples were not obtained and children <15 years of age were excluded. Bilateral varicoceles were excluded as we wanted to assess the effect of high-grade varicocele on the testicular volume loss of the ipsilateral testes compared to the healthy contralateral testes.

All children with varicoceles who were referred to us for further management, underwent a clinical examination on an outpatient basis, following which they were graded as Grade I (palpable only during Valsalva manoeuvre), Grade II (palpable without Valsalva manoeuvre) and Grade III (visible without need for palpation) as described by Dubin

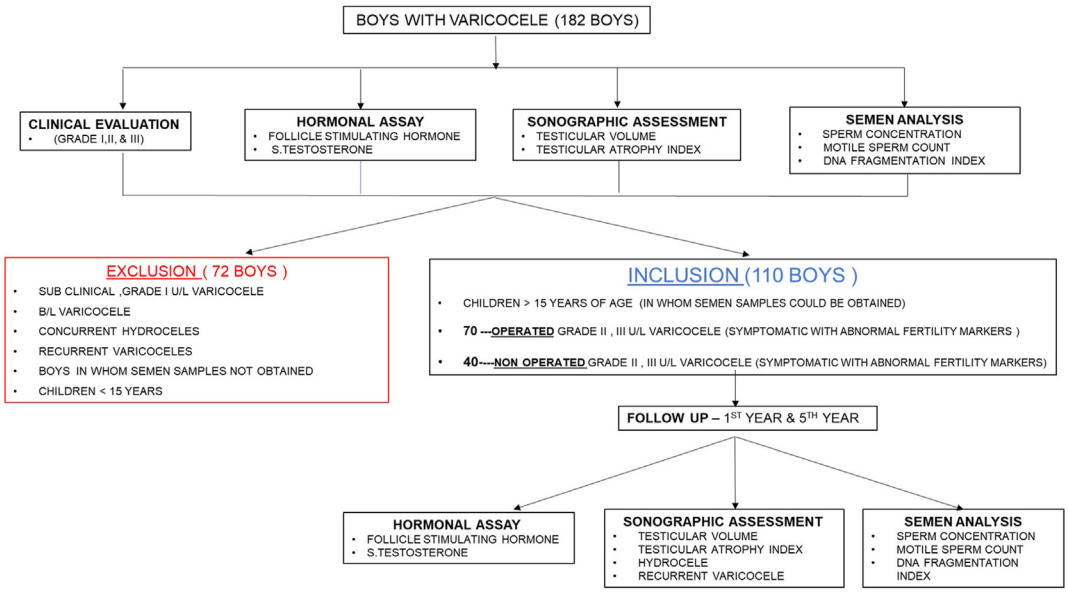

Fig. 1 Protocol for management of adolescent boys with varicoceles.

and Amelar [11]. A hormonal assay comprising of serum follicle stimulating hormone (>10 IU/ml -abnormal) and serum total testosterone (<3.5 ng/ml-abnormal) was performed in all, and an abnormality in any one of these hormones was termed as an abnormal hormonal assay [12,13]. A Sonographic assessment was performed by a single experienced radiologist, to document the testicular volume (both normal and affected testis), using the "Lambert s" formula *i.e. Testicular volume(ml) = length × width × height × 0.71* [14,15]. Testicular atrophy was defined by a difference of > 2 ml in the testicular volume compared to the contralateral testis, which in turn should be within the limits of normal values for that age [14,15]. The Testicular atrophy index of the affected testis was calculated by, *% Testicular Atrophy Index = (contralateral testis volume — affected testis volume)/contra-lateral testis volume × 100*, and an index of > 20% was also considered to confirm testicular atrophy [5,15]. Semen samples were collected by masturbation following 2—4 days of ejaculatory abstinence. After semen liquefaction, analysis was performed based on the World Health Organization criteria where in a sperm concentration of <15 million/ml and % of sperm motility of <40% was considered abnormal [16]. To evaluate the sperm DNA fragmentation index, a Sperm Chromatin Structure Assay was performed on fresh semen samples, and a threshold of >25% was considered to be abnormal, with a high probability of encountering reproductive problems [17,18]. The DNA double helix is opened by a denaturation process using heat or a lower pH, to expose the DNA fragments or potential DNA breaks following which the exposed strands are stained using acridine orange which fluoresces green when it is bound to native DNA and red when bound to broken DNA thereby signifying the damage. The DNA in the total sample was utilised to calculate the DNA fragmentation index, although a Tunnel technique is more direct and can distinguish between vital and total fractions. All the boys underwent a screening ultrasound of the abdomen to rule out the presence of any renal masses. The indications for surgical management amongst these adolescent boys (all tanner stage V), was Grade II or III varicoceles who were symptomatic i.e., pain/orchialgia (not subsiding even with scrotal support); *and* those who had abnormal fertility markers i.e., abnormal hormonal assay *and* difference of > 2 ml in the testicular volume to that of the normal contralateral testis with a testicular atrophy index of > 20%, *and* abnormal semen characteristics. All these adolescent boys who required surgery were counselled, and those who agreed were grouped as ***Group A*** — the *"operated group"* and those who refused comprised of ***Group B*** — the *"non-operated group"*. Amongst all the boys who agreed for surgery, a microscopic subinguinal varicocelectomy was performed as per our departmental protocol. Post operatively the boys were discharged on the next day of the procedure on a course of antibiotics (cephalosporins) and anti-inflammatory medications for 3 days, with application of a scrotal support. The post-operative complications amongst the Group A boys (operated group) were graded based on the Modified Clavien—Dindo Scale for surgical complications [19,20].

Follow up

All the adolescent boys included in our study i.e., Group A (*operated)*, and Group B (*non-operated)* were followed up at one year and then at 5 years from the time at presentation. At each visit, hormonal assay (serum follicle stimulating hormone, serum total testosterone), sonographic assessment (testicular volumes of affected and normal side, testicular atrophy index, testicular catch-up growth) and semen analysis (sperm concentration, % of sperm motility and DNA fragmentation index) were documented. The demographic and clinical details; changes in the hormonal assay, sonographic assessment, semen analysis at presentation, 1st year follow up and the 5th year follow up amongst both the groups, were collated and analysed. A statistical analysis using the paired t-test was performed to compare the changes in fertility markers in both the groups of boys (Group A and Group B) from the time at presentation to the 5th year follow up, and a p value of <0.05 was considered to be significant. The Statistical software namely SPSS 22.0, and R environment ver.3.2.2 was used for the analysis of the data. Microsoft word and Excel were used to generate graphs and tables. For the evaluation of primary paternity rates (i.e., without the need of assisted techniques of fertilisation) amongst both the groups a long term follow-up was carried out by telephonic or email conversations.

This study was approved by the Institutional Ethics Committee and an informed verbal as well as written consent was obtained from all the boys who were included in this study and their parents.

Results

A total of 182 boys with varicoceles were referred to us for further management, of which 110 boys who were symptomatic with abnormal fertility parameters met our inclusion criteria and were retrospectively analysed. Of these 110 boys, 70 boys were categorized as Group A (*operated group*) and 40 boys as Group B (*non-operated group*). The recruitment and surgical treatment of these cases (Group A and Group B) occurred between 2010 and 2014 (2010 — 32 boys, 2011 — 25 boys, 2012 — 14 boys, 2013 — 20 boys, 2014 — 19 boys) following which all the boys completed a 5 year follow up by the end of 2020.

The mean age at presentation amongst the Group A boys was 16.5 years (15—18 years) and Group B boys was 16 years (15—18 years). The mean age at the 1st year of follow up amongst the Group A boys was 17.5 years (16—19 years) and Group B boys was 18 years (16—19 years). The mean age at 5th year of follow up amongst Group A boys was 21.5 years (20—23 years) and Group B boys was 22.5 years (20—23 years). Grade III varicoceles were reported to be 62% in group A and 65% in group B with the left side being predominant in both the groups. Pain/orchialgia was documented in 45/70 (64%) in Group A and in 30/40 (75%) in Group B. All the boys included in our review were of Tanner V, as all were in the middle and late adolescent age group. As per the inclusion criteria, all the 110 boys had an

abnormal hormonal assay at presentation i.e., 50% (55/110) of the boys had an abnormal serum follicle stimulating hormone levels and 77% (85/110) of the boys had an abnormal serum total testosterone level. All the 110 boys had a difference of >2 ml in testicular volume in comparison to the contralateral normal testis with a testicular atrophy index >20% and all had abnormal sperm characteristics at presentation. The demographic and clinical details of Group A (Operated) and Group B (Non operated) adolescent boys have been illustrated in Table 1.

The fertility markers in Group A boys (operated) improved after surgery, which was evident in the 1st year of follow-up itself. The improvement in these parameters at the 5th year follow-up, in comparison to the normal reference range values was statistically significant with a p value of <0.001. The testicular catch-up growth amongst these boys at the 1st year follow up was 78% (55/70) and at the end of the 5th year follow up it was 92% (65/70), which was statistically significant in comparison to the normal growth of the contralateral testis of the Group A cohort. On the contrary, the changes documented in Group B in relation to the similar fertility markers from the time at presentation to the 1st year follow up and to the 5th year follow up was statistically insignificant in comparison to the normal reference range values, with a p value of >0.05. The testicular catch-up growth among these boys at the 1st year follow up was 37% (15/40) and at the end of the 5th year follow up it was 42% (17/40), which was statistically insignificant in comparison to the normal growth of the contralateral testis of the Group B cohort. There was no significant improvement in the hormonal assay and semen characteristics amongst all these boys until the end of the 5th year follow up. The baseline (at presentation), 1st year follow up and 5th year follow up details of the hormonal assay (serum follicle stimulating hormone, serum total testosterone), sonographic assessment (testicular volume-affected and normal testis, testicular atrophy index) and semen analysis (sperm concentration, % of sperm motility, DNA fragmentation index) of Group A (operated) and Group B (non-operated) have been illustrated in Table 2.

Table 1 Demographic & clinical details amongst Group A (Operated) and Group B (Non operated) adolescent boys.

n = 110 Boys		
	Group A (Operated – 70 boys)	Group B (Non-operated – 40 boys)
Mean age at presentation	16.5 years (*15–18 years*)	16 years (*15–18 years*)
Mean age at 1st year follow up	17.5 years (*16–19 years*)	18 years (*16–19 years*)
Mean age at 5th year follow up	21.5 years (*20–23 years*)	22.5 years (*20–23 years*)
Laterality (L/R)	Left – 62 **(88%)** Right – 8 **(12%)**	Left – 34 **(85%)** Right – 6 **(15%)**
Presenting Symptoms (Pain/orchialgia)	45/70 **(64%)**	30/40 **(75%)**
Grade of varicocele	G II – 26 **(37%)** G III – 44 **(62%)**	G II – 14 **(35%)** G III – 26 **(65%)**
Tanner Stage	Tanner V – 70	Tanner V – 40

The overall complication rate was 7.1% (5/70) amongst the Group A boys (operated), which was graded by the modified Clavien–Dindo scale for surgical complications. Garde I complications were noted in 3 boys – 1 boy developed a mild hydrocele which was managed conservatively, and 2 boys had persistent orchialgia which subsided over a period of 6 months with scrotal support. Grade III complications were noted in 2 boys –1 boy with a hydrocele required surgery and 1 boy who developed a hypertrophic scar required surgical excision. None of the boys developed a recurrent varicocele. We document the percentage of resolution of symptoms to be 95% (43/45) amongst Group A boys and 36% (11/40) amongst the non-operated Group B cohort. The post operative complications (Group A -operated), with follow up details amongst Group A (operated) and Group B (non-operated) adolescent boys have been depicted in Table 3.

At the time of drafting the manuscript (2021), 44% (31/70 boys) of Group A boys were married with a paternity rate of 80% (i.e., 25/31) and amongst Group B boys 58% (22/40 boys) were married with a paternity rate of 36% (i.e., 8/22) respectively. Table 4 depicts the paternity details amongst Group A and Group B boys.

Discussion

The evaluation and management of an adolescent boy with a varicocele should be aimed towards identification of possible risk factors associated with long-term fertility. The primary points of evaluation are grade of the varicocele, testicular volumes, endocrine evaluation, and semen characteristics [2]. In published literature, some have argued for early surgical correction if the testicular volume discrepancy is >20% while others have noted that nearly 80% of these volume discrepancies correct over time without any intervention [21]. There is strong evidence that the adolescent varicocele, especially grade II or III, may affect ipsilateral testicular growth, testicular histology, and semen parameters. However, the exact impact of these changes in middle and late adolescent boys (who are operated and non-operated), on future fertility remain to be determined and the assessment of these changes with respect to the paternity rates, brings a new perspective in the management of adolescent varicoceles [22]. We have attempted at depicting that in a highly selective cohort of boys (middle and late adolescent boys i.e., > 15 years of age), with testicular volume loss and hormonal and sperm abnormality, intervention helps in improving these parameters with ultimate positive effect on future fertility compared to expectant management. These criteria will probably also help in avoiding unnecessary intervention as varicoceles are not always associated with abnormality in above parameters. Boys with none of the above associated abnormalities may be observed. These findings are further corroborated by the De Win et al study which showed that varicoceles with low peak resistance flow were not different from boys without a varicocele. Paduch and Skoog summarized the indications for interventions to be testicular growth arrest of more than 2 ml difference between affected and normal testis, abnormal semen analysis,

Table 2 Changes in the hormonal assay, sonographic assessment, and semen characteristics from the time at presentation to the 5th year follow up, amongst *Group A* (operated) and *Group B* (non -operated) adolescent boys with high grade unilateral varicoceles.

n = 110 Boys

Group A — 70 Boys (Operated)			Group B — 40 Boys (Non-Operated)					
Parameters	At Presentation		1st Year Follow Up		5th Year Follow Up		P value	
							Presentation to 5th year	
	Group A	Group B	Group A	Group B	Group A	Group B	Group A	Group B
Hormonal Assay								
Mean FSH (IU/ml)	14.30	14.5	5.30	14	4.40	13.5	***p < 0.001***	***p > 0.05***
Range	*(7—17 IU/ml)*		*(1.5—16 IU/ml)*		*(1.5—17 IU/ml)*			
Mean S. Testosterone (ng/ml)	2.40	2.30	7.10	2.45	8.08	2.8	***p < 0.001***	***p > 0.05***
Range	*(1.5—5.5 ng/ml)*		*(2—9.5 ng/ml)*		*(1.5—10 ng/ml)*			
Sonographic Assesment								
Mean Testicular Volume (ml) (**Affected Side**)	**12.10**	**12.30**	**16.50**	**14.20**	**19.75**	**16.0**	***p < 0.001***	***p > 0.05***
Range	***(11.5—12.5 ml)***		***(14—17 ml)***		***(15.5—20 ml)***			
Mean Testicular Volume (ml) (**Normal Side**)	**15.60**	**15.80**	**17.30**	**17.80**	**20.90**	**20.50**	***p < 0.001***	***p < 0.001***
Range	***(15—16 ml)***		***(17—18 ml)***		***(20—21 ml)***			
Mean TAI * (%)	**22.40**	**22.15**	**4.62**	**20.22**	**5.50**	**21.95**	***p < 0.001***	***p > 0.05***
Range	***(21—25%)***		***(3.5—22%)***		***(4.5—23%)***			
Semen Analysis								
Mean Sperm Concentration (millions/ml)	12.50	12.30	29	13.50	37	13	***p < 0.001***	***p > 0.05***
Range	*(11—15 mil/ml)*		*(13 — 26 mil/ml)*		*(12.5—40 mil/ml)*			
Mean Motile Sperm Count (%)	32.90	31	58.45	35.5	64.60	34.5	***p < 0.001***	***p > 0.05***
Range	*(30—47%)*		*(33—70%)*		*(30—80%)*			
Mean DFI **(%)	36	34.5	21	33	16	33.5	***p < 0.001***	***p > 0.05***
Range	*(33—40%)*		*(20—40%)*		*(15—40%)*			

TAI * — Testicular atrophy index, ***DFI*** ** — DNA fragmentation index.

Table 3 Post operative complications (Group A -operated), with follow up details amongst Group A (operated) and Group B (non-operated) adolescent boys.

Post Operative Complications — Group A (*5/70 operated boys*)	
Modified Clavien-Dindo classification (7.1%)	**Grade I** — 3 Boys **Grade III** — 2 Boys
Follow Up Details	
	Group A / **Group B**
Resolution of symptoms (Pain/Orchialgia)	43/45 (95%) / 11/30 (36%)

Table 4 Paternity details amongst Group A (operated) and Group B (non-operated).

	Group A (Operated — 70)	Group B (Non-operated — 40)
Number of boys married	31/70 (***44%***)	22/40 (***58%***)
Mean age at phone call for details	22.5 years (*20—24 years*)	23 years (*20—24 years*)
Mean years of marriage	2.5 years (*1—4 years*)	3 years (*1—4 years*)
Paternity rate (*Without assisted techniques*)	25/31 (***80%***)	8/22 (***36%***)
Mean years to conception after marriage	1.5 years (*1—2 years*)	3.5 years (*2—4 years*)
% Paternity	***80%***	***36%***

abnormal hormonal assays and symptomatic varicoceles, which was similar to our indications for intervention amongst these adolescent boys with high grade unilateral varicoceles [23]. Cayan et al. analysed 36 studies, in order to ascertain the best surgical technique, concluding that the microsurgical subinguinal varicocelectomy technique has higher pregnancy rates and lower postoperative complications than other varicocelectomy techniques [24]. We at our institute have implemented the similar surgical technique in all the 70 boys who were operated.

Assessment of reproductive hormone levels is an integral component of evaluating an adolescent boy with a varicocele. Serum total testosterone and serum follicle

stimulating hormone both play an important role in the pathogenesis of spermatogenesis and in turn fertility. Follicle stimulating hormone is necessary for signalling Sertoli cells to produce factors required for maturation of germ cells into spermatozoa [25]. Kass et al., reported that adolescent boys with varicoceles have an exaggerated increase in follicle stimulating hormone secretion, thereby implying a varicocele effect on the hypothalamic-pituitary-gonadal axis [26]. Guarino et al. in his research, documented that Tanner V adolescents with varicoceles depicting exaggerated levels of follicle stimulating hormones with corresponding abnormal semen analysis, could predict the risk of future infertility thereby acting as an indicator for a surgical intervention. It was also noted that assessment of testicular volumes alone does not predict testicular dysfunction [27]. On the contrary, Zampieri et al. documented the importance of follicle stimulating hormone levels in late adolescent boys (Tanner V) with high grade (Grade III) unilateral varicoceles as a treatment indicator, which correlated with testicular hypotrophy, thereby predicting the risk of future infertility [28]. Both Guarino et al. and Zampieri et al., stress the importance of analysis of follicle stimulating hormone in a selected section of late adolescent boys (Tanner V), with high grade varicoceles and testicular volume loss and abnormal semen analysis, which is similar to the selected cohort of boys in our review. In our study 50% of all the boys had increased levels in mean follicle stimulating hormone at presentation. In Group A the mean follicle stimulating hormone at presentation was 14.30 IU/ml (7—17 IU/ml) and in Group B it was 14.50 IU/ml (7—17 IU/ml). Following varicocelectomy in Group A this reduced to a mean of 5.30 IU/ml (1.5—16 IU/ml) at the 1st year follow-up and to 4.40 IU/ml (1.5—17 IU/ml) at the end of the 5th year follow up. This was comparable to that observed in other studies [12]. However, amongst the Group B boys, the mean follicle stimulating hormone level remained high i.e. 14 IU/ml (1.5—16 IU/ml) and 13.5 IU/ml (1.5—17 IU/ml) at the 1st year and 5th year follow up respectively. Varicoceles are associated with defective testosterone synthesis, and a varicocelectomy has positive effects on Leydig cell function, and in turn spermatogenesis [29]. We observed an abnormally low mean serum total testosterone for age in 77% of all the boys at presentation i.e. 2.40 ng/ml (1.5—5.5 ng/ml) in Group A cand 2.30 ng/ml (1.5—5.5 ng/ml) in Group B. After varicocelectomy in Group A, mean total testosterone value increased to 7.10 ng/ml (2—9.5 ng/ml) at the end of the first-year follow-up and 8.08 ng/ml (1.5—10 ng/ml) at the 5th year follow up. This was comparable to that in published literature [4,13]. Amongst the Group B boys the mean serum total testosterone remained low i.e. 2.45 ng/ml (2—9.5 ng/ml) and 2.80 ng/ml (1.5—10 ng/ml) at the first year and 5th year follow up respectively. Similar to our study, Cayan et al. reported a reduction in follicle stimulating hormone and an increase in serum testosterone levels following varicocelectomy amongst adolescent boys [12]. Fig. 2 graphically represents the changes in the mean hormonal assay amongst the adolescent boys in Group A (operated) and Group B (non-operated) from presentation to the 5th year follow up respectively.

Testicular function directly correlates with testicular volume, as seminiferous tubules and germinal elements comprise 98% of testicular mass [30]. Reduction in testicular volume is caused by primary dysplasia or secondary damage to the seminiferous tubules and germinal elements, thus resulting in disturbed spermatogenesis and in turn fertility [31]. In our study, the mean testicular volume at presentation was 12.10 ml (11.5—12.5 ml) in Group A and 12.30 ml (11—12.5 ml) in Group B, which was reduced for age, with $>$2 ml difference to the contralateral normal testis in all. Following the varicocelectomy amongst the Group A boys at the 1st year follow up the mean testicular volume increased to 16.50 ml (14—17 ml) with a testicular catch up of 78% and at the end of the 5th year follow up the mean testicular volume increased to 19.75 ml (15.5—20 ml), with a testicular catch up of 92% which was significant in comparison to that of the normal contralateral testis and to the normal values in published literature [14,15]. Amongst the Group B boys, we noted a poor testicular catch-up growth of 37% at the 1st year follow up, and furthermore the catch-up growth at the end of the 5th year was 42%. Lemack et al., reported that there was 66% increase in testicular volume of the affected side, following a varicocelectomy in the adolescent boys [32]. Similarly Paduch et al., reported that varicocele repair in adolescent boys with grade II and III varicoceles, reversed testicular growth arrest and resulted in catch-up growth within 1 year of surgery [33]. Seo et al. and Zampieri et al. also reported a catch-up growth in 65% and 80% of their patients, respectively, demonstrated 18—24 months postoperatively [34,35]. Moursy et al., compared surgical versus nonsurgical management of unilateral varicoceles in adolescents boys and the catch-up testicular growth occurred in 70% of surgically managed boys, and in only 50% of the boys managed without any surgery [36]. We documented a mean testicular atrophy index at presentation to be 22.40% (21—25%) amongst Group A cohort, and 22.15% (21—25%) %amongst Group B cohort. This index normalized amongst all the boys in the Group A cohort i.e. $<$ 20% at the 1st year follow up and remained constant at the 5th year follow up which is similar to that reported by Fiogbe et al. [5]. However, amongst all the boys in Group B cohort, the index remained $>$20% until the 5th year follow up, thereby signifying testicular hypotrophy. Fig. 3 graphically represents the mean sonographic changes amongst the adolescent boys in Group A (operated) and Group B (non-operated) from the time at presentation to the 5th year follow up respectively.

Semen analysis has the potential in serving as a useful adjunct along with testicular volume estimation in determining who may benefit from the treatment of varicocele. Guzick et al. and Ku et al., reported a reduced sperm concentration of $<$15 million/ml in adolescent boys with varicoceles which improved significantly following a microscopic subinguinal varicocelectomy [37,38]. In our study the mean sperm concentration at presentation was 12.50 mil/ml (11—15 mil/ml) in Group A and 12.30 mil/ml (11—15 mil/ml) in Group B. Following varicocelectomy, i.e. in Group A, the mean sperm concentration increased to 29 mil/ml (13—26 mil/ml) at the first year follow-up and further increased to a mean of 37 mil/ml (12.5—40 mil/ml) at the end of the 5th year follow up which is normal as per WHO guidelines [16]. On the contrary, all the boys in the Group B cohort continued to have an abnormal mean sperm concentration until the 5th year follow-up. Cayan et al.,

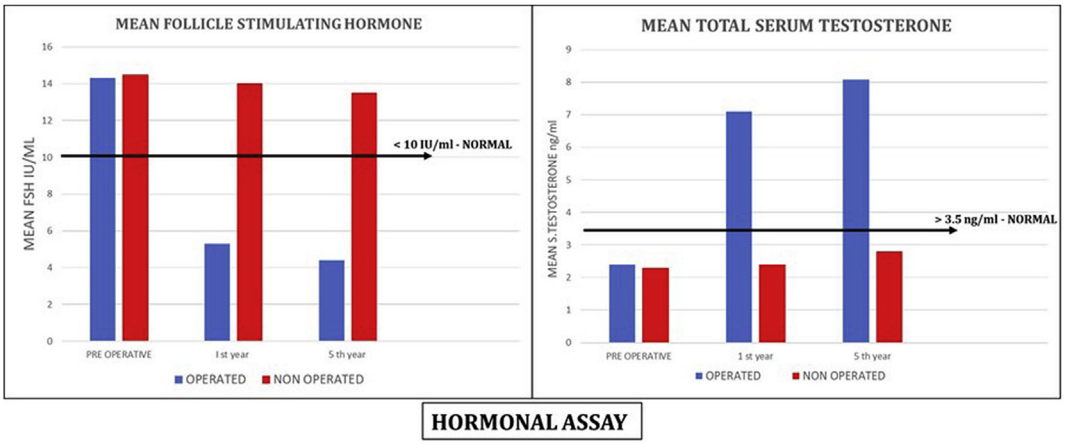

Fig. 2 Graphical representation of the mean hormonal assay changes amongst the adolescent boys(Highly selective middle and late adolescents with high grade unilateral varicoceles and ipsilateral testicular volume loss with hormonal and seminal abnormalities) in Group A (operated) and Group B (non-operated) from the time at presentation to the 5th year follow up.

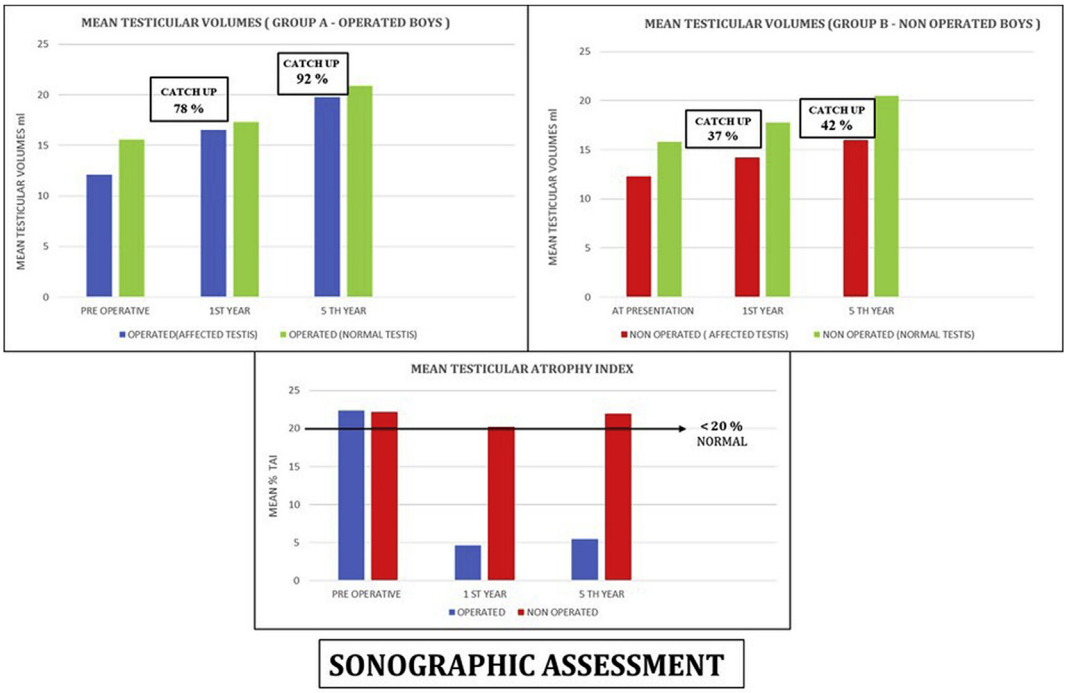

Fig. 3 Graphical representation of the mean sonographic changes amongst the adolescent boys (Highly selective middle and late adolescents with high grade unilateral varicoceles and ipsilateral testicular volume loss with hormonal and seminal abnormalities) in Group A (operated) and Group B (non-operated) from the time at presentation to the 5th year follow up.

reported an increase in the % of sperm motility from 22% to 64% following a microscopic subinguinal varicocelectomy in adolescents [39]. In our study we similarly documented a mean % sperm motility at presentation to be 32.90% (30—47%) amongst the Group A cohort and 31% (30—47%) amongst the Group B cohort. Following varicocelectomy amongst the Group A boys at the 1st year of follow up the mean % sperm motility increased to 58.45% (33—70%), which further increased to 64.60% (30—80%) at the 5th year follow-up, which is normal as per WHO guidelines [16]. Amongst all the Group B boys, the % sperm motility at the 1st year to the 5th year follow up remained below the reference range. Varicocele is characterized by venous stasis, heat stress, hypoxia, and accumulation of toxic metabolites in the testes which lead to spermatogenic arrest. Increased nitric oxide levels and sperm lipid peroxidation has been demonstrated amongst adolescents, which demonstrates seminal oxidative stress and in turn testicular apoptosis causing increased sperm nuclear DNA fragmentation which has been demonstrated to be a cause for male factor infertility [6]. A DNA fragmentation index >25% signifies a probability of male subfertility and the surgical repair of these varicoceles helps in reduction of this index [17].In our study we documented a mean % DNA

fragmentation index at presentation to be 36% (33—40%) amongst Group A cohort, and 34.5% (33—40%)in the Group B cohort. Following varicocelectomy amongst the Group A boys it normalized to <25% at the 1st year follow up and remained constant at the end of 5 years. However, amongst all the boys of Group B the index continued to be >25% even at the 5th year follow up. Similarly, Kadioglu et al. and Telli et al. also reported a significant decrease in DNA fragmentation index after varicocelectomy amongst adolescent boys with high grade varicoceles [40,41]. De Win et al. and Bertolla et al. compared late adolescents without varicocele to those with higher grade of varicoceles and documented that the latter had a higher percentage of cells with DNA fragmentation [6,42]. De Win et al, identified peak resistance flow on doppler sonography as an objective non -invasive tool to identify varicocele patients at risk for high sperm DNA fragmentation. Browo et al., reported that varicocelectomy reduced DNA fragmentation and improved sperm concentration, progressive motility, and morphology in late adolescent boys which was in accordance with our findings [17]. However, because of the different techniques used in different articles, it's difficult to compare one study with another.Fig. 4 graphically represents the changes in the mean semen characteristics amongst the adolescent boys in Group A (operated) and Group B (non-operated) from the time at presentation to the 5th year follow up respectively.

The ultimate goal in the management of an adolescent varicocele would be safeguarding the long-term paternity potential. Based on our results, we document an 80% paternity rate amongst Group A boys (operated) in comparison to a 36% paternity rate amongst the Group B boys (non-operated). This was documented by telephonic conversation or email during long term follow-up. This phase of the study started after the 5th year of in-clinic assessment of the boys in each group. This phase of the study is still ongoing. However, at the time of writing this manuscript, the mean age was 22.5 years (20—24 years) amongst the Group A boys and 23 years (20—24 years) amongst the Group B boys. 44% of group A boys were married, compared to 58% in group B. The mean years of marriage amongst the boys in Group A was 2.5 years (1—4 years) and Group B was 3 years (1—4 years). The mean years to paternity (without assisted techniques of fertility) amongst the Group A boys was 1.5 years (1—2 years) and amongst the Group B boys it was 3.5 years (2—4 years). This could probably signify that the cohort of adolescent boys who were operated could achieve paternity much earlier than those not operated. Cyan et al., reported paternity rates of 77.3% amongst the operated group and 48.4% amongst the non-operated group in boys with important risk factors, which could be compared to the paternity rates amongst the boys in our study [43]. Similarly Salzhauer et al., demonstrated high paternity rates following a varicocelectomy amongst adolescent boys who were followed up into adulthood [10]. However on the contrary, Bogaert et al. reported a paternity rate of 85% in those boys who were conservatively followed up in relation to a paternity rate of 78% in the boys who were treated, thereby concluding that screening has no effect on the paternity rates as most varicoceles probably don't need treatment [44]. Bogart et al reported about most varicoceles encountered in clinical practice, whilst in our study and Cyan et al the findings were reported in a highly selective group of adolescent boys . The possible reason for the contrary findings of the Bogaert study in comparison to ours

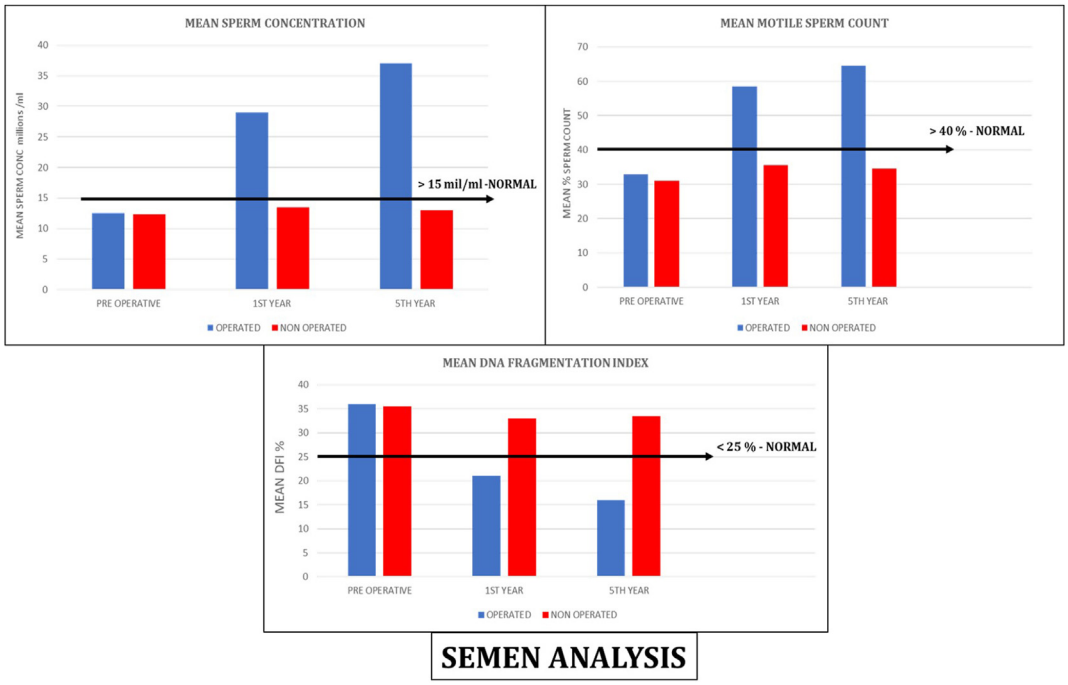

Fig. 4 Graphical representation of the mean semen characteristics changes amongst the adolescent boys (Highly selective middle and late adolescents with high grade unilateral varicoceles and ipsilateral testicular volume loss with hormonal and seminal abnormalities) in Group A (operated) and Group B (non-operated) from the time at presentation to the 5th year follow up.

and Cyan et al, is that the Bogaert study included all screening identified varicoceles regardless of severity, whereas our study included a highly selective cohort of high grade varicoceles with testicular volume loss and hormonal and seminal abnormalities. Fig. 5 Graphical represents the primary paternity rates achieved amongst the adolescent boys in Group A (Operated) and Group B (non-operated).

We propose that, middle and late adolescents with high grade unilateral varicocele and abnormal fertility markers may be considered for surgical correction to safeguard their future fertility and paternity rates rather than managing them conservatively. However, our study includes a highly selective cohort of middle and late adolescent boys (>15 years) with unilateral high grade symptomatic varicoceles. Furthermore, only those high-grade varicoceles with ipsilateral testicular volume loss and hormonal and seminal abnormalities were included in the study. This is probably different from most community identified varicoceles in adolescents which are generally noted by the primary care provider and are asymptomatic, unilateral, and generally seen in early or mid-puberty. Many of such varicoceles may not be associated with hormonal and sperm parameter abnormalities. We also report our findings in late puberty and these findings cannot be extrapolated to patients in early puberty where testicular growth spurt is more rapid and testicular asymmetry may also manifest in normal boys without varicocele(15). Thus, regular testicular measurements are suggested in affected adolescents . Another important point to note is that, although all our boys who underwent surgery were in late puberty, it was still not too late to reverse the abnormal parameters as is evident from our results. The corollary of this, could probably be to offer expectant line of management in early puberty with regular testicular measurements.

The limitations of our study are that it is a retrospective study with a small sample size. Furthermore, the ultimate effect on fertility which is the "*take home baby rate*" could not be assessed in the entire cohort of boys due to the short-term follow-up. Also, information on frequency of trying to conceive was not available. Randomization of the entire cohort could not be performed due to ethical considerations creating a selection bias. A multivariate analysis with a larger sample size and longer term follow up is ongoing.

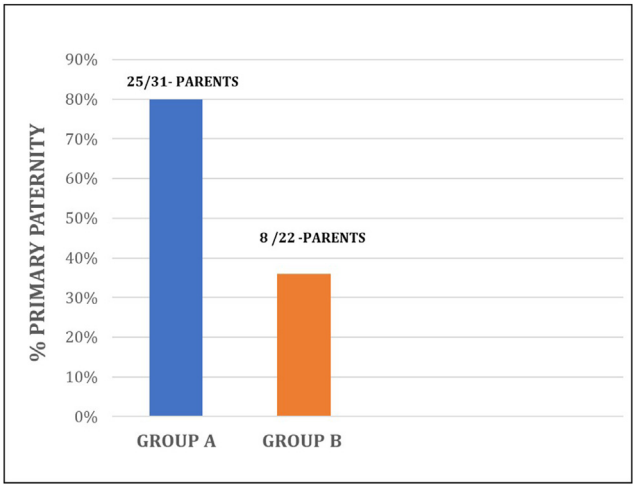

Fig. 5 Graphical representation of primary paternity rates achieved amongst the adolescent boys (Highly selective middle and late adolescents with high grade unilateral varicoceles and ipsilateral testicular volume loss with hormonal and seminal abnormalities) in Group A (Operated) and Group B (non-operated).

Conclusion

In middle and late adolescent boys in whom hormonal assay, testicular volumes and semen characteristics are negatively affected by high grade unilateral varicoceles, surgical correction could normalize these values, thereby safeguarding their fertility in the long term.

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Ethical approval and consent to participate

This study was approved by the Institutional Ethics Committee, an Informed verbal as well as written consent for publication of the data and photos have been obtained from the parents of the children included in this study as per the institutional rules.

Conflicts of interest

None declared.

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References

[1] Akbay E, Cayan S, Doruk E, Duce MN, Bozlu M. The prevalence of varicocele and varicocele-related testicular atrophy in Turkish children and adolescents. BJU Int 2000;86(4):490—3.
[2] Kolon TF. Evaluation and management of the adolescent varicocele. J Urol 2015;194(5):1194—201.
[3] Gordetsky J, van Wijngaarden E, O'Brien J. Redefining abnormal follicle-stimulating hormone in the male infertility population. BJU Int 2012;110(4):568—72.
[4] Çayan S, Akbay E, Bozlu M, Doruk E, Erdem E, Acar D, et al. The effect of varicocele repair on testicular volume in children and adolescents with varicocele. J Urol 2002;168(2):731—4.
[5] Fiogbe MA, Alao MJ, Biaou O, Gbenou SA, Yekpe P, Sossou R, et al. Ultrasound diagnosis of varicocele in the adolescent: our experience from Benin. Afr J Paediatr Surg 2013;10(4):295.
[6] Bertolla RP, Cedenho AP, Hassun Filho PA, Lima SB, Ortiz V, Srougi M. Sperm nuclear DNA fragmentation in adolescents with varicocele. Fertil Steril 2006;85(3):625—8.
[7] Locke JA, Noparast M, Afshar K. Treatment of varicocele in children and adolescents: a systematic review and meta-

analysis of randomized controlled trials. J Pediatr Urol 2017; 13(5):437—45.
[8] Lenzi A, Gandini L, Bagolan P, Nahum A, Dondero F. Sperm parameters after early left varicocele treatment. Fertil Steril 1998;69(2):347—9.
[9] Preston MA, Carnat T, Flood T, Gaboury I, Leonard MP. Conservative management of adolescent varicoceles: a retrospective review. Urology 2008;72(1):77—80.
[10] Salzhauer EW, Sokol A, Glassberg KI. Paternity after adolescent varicocele repair. Pediatrics 2004;114(6):1631—3.
[11] Dubin L, Amelar RD. Varicocele size and results of varicocelectomy in selected subfertile men with varicocele. Fertil Steril 1970;21(8):606—9.
[12] Cayan S, Kadioglu A, Orhan I, Kandirali E, Tefekli A, Tellaloglu S. The effect of microsurgical varicocelectomy on serum follicle stimulating hormone, testosterone and free testosterone levels in infertile men with varicocele. BJU Int 1999;84(9):1046—9.
[13] Kanakis GA, Tsametis CP, Goulis DG. Measuring testosterone in women and men. Maturitas 2019;125:41—4.
[14] Oehme NHB, Roelants M, Særvold Bruserud I, Madsen A, Eide GE, Bjerknes R, et al. Reference data for testicular volume measured with ultrasound and pubic hair in Norwegian boys are comparable with Northern European populations. Acta Paediatr 2020;109(8):1612—9.
[15] Vaganée D, Daems F, Aerts W, Dewaide R, van den Keybus T, De Baets K, et al. Testicular asymmetry in healthy adolescent boys. BJU Int 2018;122(4):654—66.
[16] Cooper TG, Noonan E, Von Eckardstein S, Auger J, Baker HW, Behre HM, et al. World Health Organization reference values for human semen characteristics. Hum Reprod Update 2010; 16(3):231—45.
[17] Birowo P, Wijaya JR, Atmoko W, Rasyid N. The effects of varicocelectomy on the DNA fragmentation index and other sperm parameters: a meta-analysis. Basic Clin Androl 2020;30(1):1—9.
[18] De Iuliis GN, Thomson LK, Mitchell LA, Finnie JM, Koppers AJ, Hedges A, et al. DNA damage in human spermatozoa is highly correlated with the efficiency of chromatin remodeling and the formation of 8-hydroxy-2′-deoxyguanosine, a marker of oxidative stress. Biol Reprod 2009;81(3):517—24.
[19] Dindo D, Demartines N, Clavien P-A. Classification of surgical complications: a new proposal with evaluation in a cohort of 6336 patients and results of a survey. Ann Surg 2004;240(2):205.
[20] Clavien PA, Barkun J, De Oliveira ML, Vauthey JN, Dindo D, Schulick RD, et al. The Clavien-Dindo classification of surgical complications: five-year experience. Ann Surg 2009;250(2): 187—96.
[21] Kolon TF, Clement MR, Cartwright L, Bellah R, Carr MC, Canning DA, et al. Transient asynchronous testicular growth in adolescent males with a varicocele. J Urol 2008;180(3):1111—5.
[22] Minevich E, Wacksman J, Lewis AG, Sheldon CA. Inguinal microsurgical varicocelectomy in the adolescent: technique and preliminary results. J Urol 1998;159(3):1022—4.
[23] Paduch DA, Skoog SJ. Current management of adolescent varicocele. Rev Urol 2001;3(3):120.
[24] Çayan S, Shavakhabov S, Kadioğlu A. Treatment of palpable varicocele in infertile men: a meta-analysis to define the best technique. J Androl 2009;30(1):33—40.
[25] Walker WH, Cheng J. FSH and testosterone signaling in Sertoli cells. Reproduction 2005;130(1):15—28.
[26] Kass EJ, Freitas JE, Salisz JA, Steinert BW. Pituitary gonadal dysfunction in adolescents with varicocele. Urology 1993; 42(2):179—81.
[27] Guarino N, Tadini B, Bianchi M. The adolescent varicocele: the crucial role of hormonal tests in selecting patients with testicular dysfunction. J Pediatr Surg 2003;38(1):120—3.
[28] Zampieri N. Hormonal evaluation in adolescents with varicocele. J Pediatr Urol 2021;17(1). 49—e1.
[29] Takeyama M, Honjoh M, Kodama M, Sakaguchi H, Koh E, Kondoh N, et al. Testicular steroids in spermatic and peripheral veins after single injection of hCG in patients with varicocele. Arch Androl 1990;24(2):207—13.
[30] Takihara H, Cosentino MJ, Sakatoku J, Cockett AT. Significance of testicular size measurement in andrology: II. Correlation of testicular size with testicular function. J Urol 1987; 137(3):416—9.
[31] Riebel T, Herrmann C, Wit J, Sellin S. Ultrasonographic late results after surgically treated cryptorchidism. Pediatr Radiol 2000;30(3):151—5.
[32] Lemack GE, Uzzo RG, Schlegel PN, Goldstein M. Microsurgical repair of the adolescent varicocele. J Urol 1998;160(1):179—81.
[33] Paduch DA, Niedzielski J. Repair versus observation in adolescent varicocele: a prospective study. J Urol 1997; 158(3):1128—32.
[34] Seo JW, Cho KS, Han SW. Ipsilateral testicular catch-up growth after varicocelectomy in adolescents. Korean J Urol 2007; 48(7):731—4.
[35] Zampieri N, Mantovani A, Ottolenghi A, Camoglio FS. Testicular catch-up growth after varicocelectomy: does surgical technique make a difference? Urology 2009;73(2):289—92.
[36] Moursy EES, ElDahshoury MZ, Hussein MM, Mourad MZ, Badawy AA. Dilemma of adolescent varicocele: long-term outcome in patients managed surgically and in patients managed expectantly. J Pediatr Urol 2013;9(6):1018—22.
[37] Guzick DS, Overstreet JW, Factor-Litvak P, Brazil CK, Nakajima ST, Coutifaris C, et al. Sperm morphology, motility, and concentration in fertile and infertile men. N Engl J Med 2001;345(19):1388—93.
[38] Ku JH, Kim SW, Park K, Paick J-S. Benefits of microsurgical repair of adolescent varicocele: comparison of semen parameters in fertile and infertile adults with varicocele. Urology 2005;65(3):554—8.
[39] Cayan S, Acar D, ÜLger Sül, Akbay E. Adolescent varicocele repair: long-term results and comparison of surgical techniques according to optical magnification use in 100 cases at a single university hospital. J Urol 2005;174(5):2003—7.
[40] Kadioglu TC, Aliyev E, Celtik M. Microscopic varicocelectomy significantly decreases the sperm DNA fragmentation index in patients with infertility. BioMed Res Int 2014;2014.
[41] Telli O, Sarici H, Kabar M, Ozgur BC, Resorlu B, Bozkurt S. Does varicocelectomy affect DNA fragmentation in infertile patients? Indian J Urol IJU J Urol Soc India 2015;31(2):116.
[42] De Win G, De Neubourg D, De Wachter S, Vaganée D, Punjabi U. Peak retrograde flow a potential objective management tool to identify young adults with varicocele 'at risk'for a high sperm DNA fragmentation. J Pediatr Urol December 2021;17(6): 760.e1—9.
[43] Çayan S, Şahin S, Akbay E. Paternity rates and time to conception in adolescents with varicocele undergoing microsurgical varicocele repair vs observation only: a single institution experience with 408 patients. J Urol 2017;198(1): 195—201.
[44] Bogaert G, Orye C, De Win G. Pubertal screening and treatment for varicocele do not improve chance of paternity as adult. J Urol 2013;189(6):2298—304.

Appendix A. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.jpurol.2021.11.020.