Investigative Clinical Study on Prostate Cancer Part V: Luteinizing Hormone and the Pituitary-Testicular-Prostate Axis at the Time of Initial Diagnosis and Subsequent Cluster Selection of the Patient Population

ANTONIO B. PORCARO[1], ALDO PETROZZIELLO[2], FILIPPO MIGLIORINI[1], BEATRICE CARUSO[3], CLAUDIO COCCO[3], TEODORO SAVA[4], CLAUDIO GHIMENTON[5], MARIO ROMANO[6], CARMELO MONACO[1] and LUIGI COMUNALE[1]

Departments of [1]Urology, [2]Internal Medicine Endocrinology Section, [3]Laboratory Medicine, [4]Medical Oncology, [5]Pathology and [6]Radiation Oncology, University Integrated Hospitals, Civil Major Hospital, Verona, Italy

Abstract. *Aim: To evaluate Luteinizing hormone (LH) physiopathology along the pituitary testicular prostate axis at the time of initial diagnosis of prostate cancer in relation to the available clinical variables and to the subsequent cluster selection of the patient population. Patients and Methods: Age, percentages of positive cores at Trans Rectal Ultrasound Scan Biopsy (TRUSB) (P+), biopsy Gleason score (bGS), LH, Total Testosterone (TT), Free Testosterone (FT) and Prostate Specific Antigen (PSA) were the continuous clinical variables. All patients had histologically proven carcinoma of the prostate and had not previously received 5α-reductase inhibitors, LH-releasing hormone analogues or testosterone replacement treatment. Correlation analysis was performed for the patient population. Correlation analysis, linear regression and analysis of variance was computed in groups and subgroups of the prostate cancer population. Results: Correlation analysis of the patient population showed that LH was significantly correlated to age (p=0.02) and FT (p=0.01). The population was clustered in LH I (LH≤7.5 IU/l) and LH II (LH>7.5 IU/l). Correlation analysis showed significant LH correlations for TT (p<0.0001) and FT (p=0.0004) for LH I; significant LH correlation to FT (p=0.0001) for LH II. Simple linear regression showed that LH was significantly predicted by both TT (p-Value<0.0001) and FT (p-Value=0.0004) in LH I; but only FT (p-Value<0.0001) in LH II. Multiple linear regression showed that LH was significantly predicted by both TT (p-Value=0.0004) and PSA (p-Value=0.03) in LH I; but only by FT (p-Value=0.003) in LH II. Analysis of variance showed that: a) LH and age were significantly lower in LH I than II; b) LH I expressed higher mean FT levels (p=0.08) and lower mean P+ (p=0.07) than LH II. The LH versus PSA plot was computed for LH group I and 3 sub clusters were created: LH I group A (LH/PSA≤0.25), B (0.25<LH/PSA ≤0.75), and C (LH/PSA>0.75). Correlation analysis showed that LH was significantly correlated to age (p=0.01), TT (p=0.03) and PSA (p=0.0004) in LH IA; LH was significantly correlated to PSA (p<0.0001) in LH IB; and LH significantly correlated to TT (p=0.005), FT (p=0.01), and PSA (p=0.008) in LH 1C. Multiple linear regression showed that LH was significantly correlated to age (p=0.02) and PSA (p=0.01) in LH IA, to TT (p=0.01) and PSA (p<0.0001) in LH IB, and to PSA (p=0.003) and weakly to TT (p=0.09) in LH IC. The groups differed significantly for mean levels of LH (p=0.0004), TT (p=0.005), FT (p=0.01), PSA (p<0.0001), bGS (p=0.003). Analysis of variance between the subgroups of the patient population (LH IA, LH IB, LH IC, LH II) showed significant differences in mean levels for LH (p<0.0001), age (p=0.004), TT (p=0.009), FT (p=0.02), PSA (p<0.0001), PSA/FT (p<0.0001), bGS (p=0.01), but not for P+ (p=0.10). Conclusion: According to LH physiopathology, the prostate cancer population could be clustered into hypo-gonadic and non-hypo-gonadic group at diagnosis. The hypo-gonadic group expresses an aggressive tumor phenotype and might be divided into two more different significant subsets including primary and secondary hypo-gonadic patients: the former (LH II) including older patients with high LH levels, the latter (LH IA) including younger patients with low LH and LH/PSA levels (subgroup LH IA). The non-hypo-gonadic group showed a less aggressive tumor phenotype and according to the LH/PSA ratio might be*

Correspondence to: Antonio Benito Porcaro, Azienda Ospedaliera Universitaria Integrata Verona Dipartimento AD Attività Integrata DI Chirurgia ED Oncologia – Pancreas CenterUnità Complessa DI Urologia D.O., Sede di Borgo Trento - P.le A. Stefani, 1- 37121 Verona, Italy. Tel: +39 045 8123313, Fax: +39 045 8123471, e-mail: drporcaro@yahoo.com

Key Words: Luteinizing hormone, LH, total testosterone, TT, free testosterone, FT, prostate-specific antigen, PSA, prostate cancer.

clustered into LH IB (0.25<LH/PSA≤0.75) and LH IC (LH/PSA>0.75), the former showing a more aggressive tumor phenotype than the latter. Confirmatory studies are necessary.

Androgens are essential for the prostate to achieve and maintain normal tissue mass, composition, and secretory function. The interstitial cells of Leydig are responsible for the production of 95% of all circulating androgen in the form of testosterone. The principal control of testosterone synthesis is mediated by the Luteinizing hormone (LH). It has been shown that the prostate contains specific LH receptors that are structurally and functionally similar to those expressed in the gonads, and it is possible that LH may in some way physiologically regulate prostate size (1). Approximately 98% of the circulating androgens are bound to plasma proteins, including a specific beta-globulin, testosterone-binding globulin (TeBG). The free testosterone in the blood is the physiologically important fraction.

Etiological and stimulatory factors of prostate cancer are still being sought. The role of androgens in the pathogenesis of the disease remains unclear, but is thought to be pivotal. According to evidence from the reported literature, prostate cancer is androgen dependent (2), increases the levels of prostate-specific antigen (PSA) (3), and is related to PSA growth rate for its extension and prognosis (4, 5). It has also extensively been shown that pretreatment total testosterone (TT) and free testosterone (FT) serum levels may be abnormal in prostate cancer patients (6-13).

Prostate cancer is an interesting tumor for clinical investigation. Unfortunately, at the moment we lack a complete understanding of the physiopathology of its natural history (14). The pituitary axis in prostate cancer has been investigated extensively and it has been suggested that the tumor may produce a substance that alters the normal function of the pituitary–testicular axis also resulting in abnormal LH and Follicle Stimulating Hormone (FSH) serum levels (6, 10-22). It has also been suggested that the impact of prostate cancer on the hypothalamic–pituitary axis may be more profound in high-grade prostate cancer (20), but this hypothesis has not been confirmed (23).

This study aimed at evaluating LH physiopathology along the pituitary testicular prostate axis at the time of initial diagnosis of prostate cancer in relation to the available clinical variables and to the subsequent cluster selection of the patient population.

Patients and Methods

The study involved 86 individuals diagnosed with prostate cancer. The descriptive statistics of the patient population with treatments performed at the time of this communication are shown in Table I. All the patients had histologically proven carcinoma of the prostate and had not previously received 5α-reductase inhibitors, LH-releasing hormone analogues or testosterone replacement treatment.

Table I. *Summary and descriptive statistics of the patient population (n=86).*

Clinical variable	Mean	Med	SD	Min	Max
Age (years)	65.78	66.68	6.99	50.51	80.41
LH (2.10-10 IU/l)	6.87	4.20	8.53	1.10	48.00
FSH (1.0-14 IU/l)	9.83	6.45	11.85	1.30	54.80
PRL (65-425 mU/l)	201.94	164.50	167.02	70.00	1011.00
TT (9-29 nmol/l)	16.05	14.95	6.69	6.50	40.70
FT (31-163 pmol/l)	33.59	31.95	10.85	14.10	71.90
PSA T (2-4 μg/l)	9.14	6.06	8.31	1.31	44.60
P+	0.35	0.31	0.22	0.06	1.00
bGS	6.53	6.00	0.79	5.00	9.00
Clinical stage	**n**				
cT1c	40				
cT2	43				
cT3	3				
cN0	86				
cM0	86				
Primary treatment	**n**				
Radical prostatectomy	75				
Radiation	10				
Androgen blockade	1				

LH, Luteinizing hormone; FSH, follicle stimulating hormone; PRL, prolactin; TT, total testosterone; FT, free testosterone; PSA T total PSA; P+, % biopsy positive cores; bGS, biopsy Gleason score; cT, tumor clinical staging; cN, clinical node stage; cM, clinical staging for metastases; med median; SD, standard deviation; Var, variance.

The 14-core Trans Rectal Ultrasound Scan (TRUS) guided prostate biopsy technique was routinely used and additional cores were taken when a lesion on either TRUS or digital rectal examination was evident. The biopsy Gleason score was used to grade the tumors. Patients were classified according to primary tumor stage, lymph node and metastatic status, using the TNM categories recommended by the 1997 International Union Against Cancer TNM classification system (24). The total patient population under the testosterone study, still open and progressing, is over 220 individuals, but this communication does not include those patients who were not simultaneously assessed for pituitary gland hormones. After informed signed consent, pretreatment simultaneous serum samples were obtained from a cubital vein, at least one month after TRUSB between 8.00-8.30 a.m. for measuring serum LH, TT, FT and PSA levels. The samples were analyzed at the same laboratory of our hospital. LH (range: 2.0-10 IU/l), TT (normal range: 9-29 nmol/l) and PSA (normal range: 2-4 ug/l) were measured by immunochemiluminescent test performed by ADVIA Centaur XP, Siemens. Free testosterone (FT normal range: 31-163 pmol/l) was measured by immunoradiometric test (DSL, USA).

Statistical analysis. Age, percentage of positive cores at TRUSB (P+), biopsy Gleason score (bGS), LH, TT, FT and PSA were the continuous clinical variables considered. Correlation analysis of the continuous variables of the patient population was performed.

Table II. *LH correlation analysis of the prostate cancer population.*

Variable	Statistic	LH	Age	TT	FT	PSA	G/b
Age	R	0.33					
	p-Value	0.002					
TT	R	–0.11	0.004				
	p-Value	0.32	0.97				
FT	R	–0.27	–0.06	0.64			
	p-Value	0.01	0.55	<0.0001			
PSA	R	–0.07	0.15	–0.12	–0.05		
	p-Value	0.51	0.16	0.26	0.67		
G/b	R	0.13	0.23	0.02	–0.03	0.21	
	p-Value	0.22	0.03	0.83	0.77	0.04	
P+	R	0.19	0.11	0.03	–0.04	0.10	0.45
	p-Value	0.08	0.30	0.78	0.73	0.35	0.0001

R=Pearson's correlation coefficient.

According to LH serum levels, the patient population was clustered into LH group I (LH≤7.5 IU/l) and LH group II (LH>7.5 IU/l). Analysis of LH correlating to the continuous variables in the two subgroups was computed. Multiple linear regression analysis of the variables predicting LH was also performed. Analysis of variance was computed in order to detect significant differences between group LH I and II.

The LH I patient population was clustered according to the LH/PSA ratio into group A (LH/PSA≤0.25), B (0.25<LH/PSA≤0.75) and group C (LH/PSA>0.75). Correlation analysis, simple and multiple linear regression analysis were performed in each subgroup. Analysis of variance of the continuous variables between the LH 1 subgroups was computed in order to assess significant differences.

Analysis of variance between the different LH subgroups (LH IA, LH IB, LH IC AND LH II) of the patient population was computed, in order to detect significant differences of the investigated clinical variables which also included the PSA to FT ratio (LH, AGE, TT, FT, PSA, PSA/FT, bGS and P+).

Results

The results of correlation analysis of the patient population are reported in Table II. As shown, LH was significantly correlated to age (p=0.02) and FT (p=0.01); also its correlation coefficient for P+ was close to statistical significance (p=0.08). The bGS was significantly correlated to age (p=0.03), PSA (p=0.04) and P+ (p=0.0001). Interestingly, FT was significantly correlated to both LH (p=0.01) and TT (p<0.0001). P+ correlation coefficient to age was close to statistical significance (p=0.08). Figure 1 depicts the scatter plot of LH *versus* age (Figure 1a) and FT (Figure 1b), the regression lines are also reported. As shown by the plots, two clusters of the patient population are suggested according to the LH serum levels, group I (LH≤7.5 IU/l) and II (LH>7.5 IU/l), respectively.

Table III shows the correlation analysis results for the two LH subgroups. As shown, LH I showed significant LH correlations for TT (p=0.0001) and FT (p=0.0004); also the bGS was significantly correlated to PSA (p = 0.03) and P+ (p=0.0001). LH subgroup II results were as follows: a) LH was significantly correlated to FT (p=0.0001); b) FT was significantly correlated to PSA (p=0.04), c) bGS was almost significantly correlated to age (p=0.06). Figure 2 shows the scatter plot of LH *versus* FT of the patient population clustered into groups I and II; the regression lines are also reported. Table IV shows the results related to simple and multiple linear regression analysis (Table IVa), and analysis of variance of LH subgroup I and II (Table IVb). As depicted for LH group I, LH was significantly predicted by both TT (p<0.0001) and FT (p=0.0004) in simple regression, and by both TT (p=0.0004) and PSA (p=0.03) in multiple linear regression. As shown for LH cluster II, LH was only significantly predicted by FT in both simple (p<0.0001) and multiple (p=0.003) analysis. As reported in Table IVb, analysis of variance between the two subgroups showed that LH and age were significantly lower in LH cluster I than II. Also, LH subgroup I showed higher mean FT levels (p=0.08) and lower mean P+ (p=0.07) than group II with a trend approaching statistical significance.

Table III. *Correlation anakysis of the LH I and LH II subgroups.*

Variable	Stat	LH	Age	TT	PSA	FT	P+
LH subgroup I							
Age	R	0.20					
	p-Value	0.09					
TT	R	0.54	0.04				
	p-Value	<0.0001	0.76				
PSA	R	0.13	0.19	-0.14			
	p-Value	0.28	0.11	0.25			
FT	R	0.41	0.04	0.69	-0.17		
	p-Value	0.0004	0.73	<0.0001	0.16		
P+	R	0.12	0.00	0.12	0.16	0.07	
	p-Value	0.34	0.98	0.30	0.19	0.55	
G/b	R	0.04	0.16	0.13	0.34	0.02	0.50
	p-Value	0.30	0.19	0.29	0.003	0.87	0.0001
LH subgroup II							
Age	R	0.17					
	p-Value	0.55					
TT	R	-0.36	0.15				
	p-Value	0.17	0.57				
PSA	R	-0.31	-0.02	-0.05			
	p-Value	0.23	0.93	0.84			
FT	R	-0.92	-0.27	0.33	0.51		
	p-Value	<0.0001	0.31	0.21	0.04		
P+	R	0.10	0.37	-0.23	-0.08	-0.34	
	p-Value	0.70	0.15	0.39	0.76	0.19	
G/b	R	0.08	0.47	-0.23	-0.11	-0.11	0.27
	p-Value	0.76	0.06	0.38	0.69	0.69	0.30

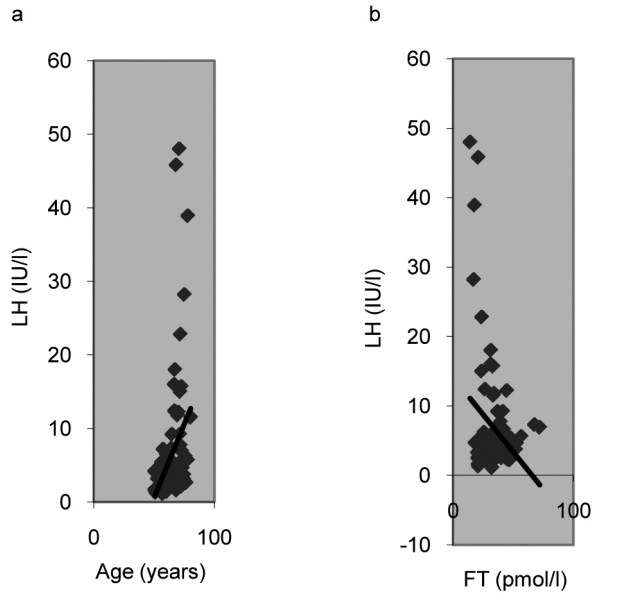

Figure 1. *Scatter plot of LH versus age (a; p-Value=0.002) and FT (b: p-Value=0.01) in the patient population.*

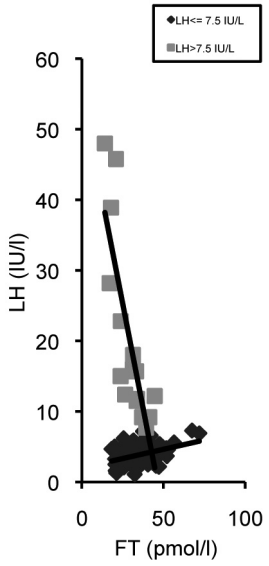

Figure 2. *Scatter plot of LH to FT in subgroups LH I and II of the patient population. LH cluster I, p-Value=0.0004. LH cluster II, p-Value=0.0001.*

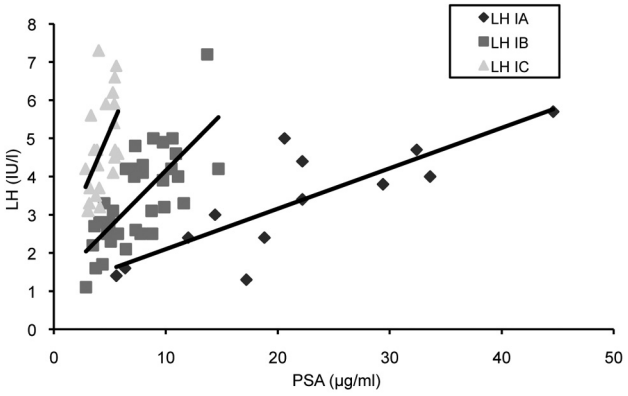

Figure 3. *LH I population with relative subgroups (A, B, C) according to the LH/PSA ratio.*

Since LH in subgroup I was significantly correlated to PSA in multiple linear regression, the LH *versus* PSA plot was computed and three clusters were selected: LH I group A (LH/PSA≤0.25), B (0.25<LH/PSA≤0.75), and C (LH/PSA>0.75). The scatter plot and regression lines of the three LH I clusters (A, B, C) are plotted in Figure 3, which clearly depicts three different subgroups. Correlation analysis of the clinical variables (LH, age, TT, FT, PSA, bGS, P+) of LH I subgroups was computed and the results are reported in Table V. LH subgroup IA showed that: a) LH was significantly correlated to age (p=0.01), TT (p=0.03) and PSA (p=0.0004); b) age was significantly correlated to PSA (p=0.04); c) FT was significantly correlated to bGS (p=0.04); d) G/b was significantly correlated to FT (p=0.04) and P+ (p=0.02). The results for LH I subgroup B showed that: a) LH was significantly correlated to PSA (p<0.0001); b) TT was significantly correlated to FT (p=0.0001); c) FT was significantly correlated to TT (0.0001) and P+ (p=0.05); d) P+ was significantly correlated to FT (p=0.05) and G/b (p=0.01). The results for LH 1 group C were as follows: a) LH significantly correlated to TT (p=0.005), FT (p=0.01), and PSA (p=0.008); b) TT was highly correlated to FT (p=0.0001), P+ (p=0.03) and weakly with bGS (p=0.08); c) FT was significantly correlated to PSA (p=0.05) and P+ (p=0.05); d) P+ was also significantly correlated to bGS (p=0.01). Multiple linear regression showed that LH was significantly correlated to: a) age (p=0.02) and PSA (p=0.01) in LH I subgroup A; b) TT (p=0.01) and PSA (p<0.0001) in LH I subgroup B; c) PSA (p=0.003) and weakly to TT (p=0.09) in LH I subgroup C. Analysis of variance between the different LH I subgroups A,B,C was computed and the results are also reported in Table VI. As shown the groups differed significantly for mean levels of LH (p=0.0004), TT (p=0.005), FT (p=0.01), PSA (p< 0.0001), bGS (p=0.003). Interestingly, LH I group A had significantly higher mean PSA (21,49 mg/l) and bGS (7), and lower mean levels for LH (3.32), TT (13.38) and FT (30.68) than the other two groups.

Analysis of variance between the subgroups of the patient population (LH IA, LH IB, LH IC, LH II) showed significant mean levels of the clinical variables investigated (LH, AGE, TT, FT PSA, PSA/FT, bGS) except for P+ (p=0.10) (see Table VII).

Table IV. *Linear regression and analysis of variance of LH group I and II of the patient population.*

a) Simple and multiple linear regression analysis.

LH ≤7.5 IU/l				LH>7.5 IU/l			
		p-Value				*p*-Value	
Variable	R	Univariate	Multivariate	Variable	R	Univariate	Multivariate
Age (years)	0.20	0.09	0.11	Age	0.16	0.55	0.91
TT (9-29 nmol/l)	0.54	<0.0001	0.0004	TT	–0.36	0.17	0.76
FT (31-163 pmol/l)	0.41	0.0004	0.55	FT	–0.82	<0.0001	0.003
PSA T (2-4 ug/l)	0.13	0.28	0.03	PSA	–0.31	0.23	0.46
P+	0.11	0.34	0.39	P+	0.10	0.70	0.25
G/b	0.04	0.75	0.14	G/b	0.08	0.76	0.80

b) Analysis of variance between LH group I (N=70) and II (N=16).

Groups	Mean	Variance	*p*-Value
LH I (2.10-10 IU/l)	3.84	2.02	<0.0001
LH II (2.10-10 IU/l)	20.15	172.51	
Age I (years)	64.56	48.46	0.0005
Age I (years)	71.08	17.05	
TT I (9-29 nmol/l)	16.4	44.87	0.30
TT II (9-29 nmol/l)	14.48	43.87	
FT I (31-163 pmol/l)	34.57	121.60	0.08
FT II (31-163 pmol/l)	29.33	83.33	
PSA T I (2-4 µg/l)	9.07	64.27	0.88
PSA T II (2-4 µg/l)	9.42	95.31	
P+ I	0.33	0.04	0.07
P+ II	0.44	0.06	
bG I	6.48	0.51	0.23
bG II	6.75	1.13	

Discussion

In the present study, we have shown that at diagnosis LH was significantly correlated to age and FT (see Figure 1) and the correlation to P+ was close to statistical significance, suggesting that LH might be related to tumor biology. According to these results, the prostate cancer population was separated into the LH I and LH II subgroups, the latter accounting for 19% of the patients (see Figure 2).

Subgroup LH II showed a more aggressive tumor phenotype and poorer testicular function than LH subgroup I (see Table IVb). Interestingly, LH subgroup 2 showed features which were similar to those reported by Harper *et al*. and Chen *et al*. who showed that high pretreatment plasma concentration of LH and low concentrations of testosterone in plasma at the time of diagnosis were related to a poor prognosis (12, 13). An interesting finding of the study was that FT always significantly predicted LH and it was significantly correlated to PSA, but not to TT as in LH subgroup I. In our opinion, the LH subgroup II shows tumor features which are suggestive of castration-resistant prostate cancer.

LH subgroup I accounts for 81% of the patient population and shows features indicating a well-functioning pituitary–testicular axis, with tumor variables correlated to each other (see Figure 2, Tables III and IV). Interestingly, multiple linear regression showed that LH was significantly predicted only by TT and PSA, indicating a functional relationship between the pituitary–testis–prostate cancer axis (see Table IV). This striking finding forced us to compute the scatter plot relating LH to PSA, and according to the LH/PSA ratio, three significant LH I subclusters (A, B, C) were selected (see Figure 3). Surprisingly, LH IA subgroup expressed features indicating poorer pituitary–testicular axis function and a more aggressive tumor phenotype than subgroups LH IB and LH IC (see Tables V and VI). Interestingly, LH IA subgroup also showed features which were similar to those of LH subgroup II except for LH serum levels. Also, the LH IC subgroup showed a well functioning pituitary–testicular–prostate axis since LH was significantly correlated to PSA, TT and FT (see Tables V and VI).

Analysis of variance between the subgroups of the patient population (LH IA, LH IB, LHIC, LH II) assessed significant differences of the clinical variables investigated except for P+. As shown in Table VII, the subgroups of the patient population can be ranked according to tumor phenotype (proceeding from the most to the less aggressive) as follows: LH IA, LH II, LH IB, LH IC. Interestingly, LH IA had the lowest mean for LH and TT, and the highest mean PSA, PSA/FT and bGS, while LH II had the highest mean for LH and age and lowest for FT. Moreaver, the P+ mean was higher in the LH II subgroup (0.44) than LH 1A (0.40), LH1b (0.29) and LH 1C (0.36), but the trend did not reach statistical significance (p=0.10), this may be explained by the small numbers of patients in the subgroups.

The findings of the present study might explain the majority of the controversial results in literature relating LH and TT to prostate cancer. LH IA might be related to high grade (20) and advanced prostate cancer (19); it also might

Table V. *Correlation analysis of the different subgroups (A, B, C) of the LH I population.*

Variable	Stat	LH	Age	TT	FT	PSA	G/b
LH I - GROUP A							
Age	R	0.77					
	p-value	0.001					
TT	R	0.59	0.37				
	p-value	0.03	0.20				
FT	R	0.46	0.17	0.48			
	p-value	0.11	0.50	0.09			
PSA	R	0.82	0.56	0.42	0.17		
	p-value	0.0004	0.04	0.15	0.57		
G/b	R	0.35	0.22	0.42	0.55	0.22	
	p-value	0.23	0.45	0.14	0.04	0.45	
P+	R	0.20	0.08	–0.08	0.29	0.14	0.62
	p-value	0.50	0.78	0.78	0.32	0.64	0.02
LH I - GROUP B							
Age	R	0.01					
	p-Value	0.10					
TT	R	0.28	0.01				
	p-Value	0.54	0.93				
FT	R	0.10	0.03	0.60			
	p-Value	0.54	0.83	0.0001			
PSA	R	0.71	–0.07	–0.09	–0.10		
	p-Value	<0.0001	0.66	0.60	0.55		
G/b	R	–0.12	0.01	–0.06	–0.28	–0.12	
	p-Value	0.49	0.95	0.69	0.10	0.70	
P+	R	0.08	–0.24	–0.17	–0.33	0.12	0.40
	p-Value	0.63	0.17	0.31	0.05	0.48	0.01
LH I . GROUP C							
Age	R	0.17					
	p-Value	0.42					
TT	R	0.55	–0.05				
	p-Value	0.005	0.81				
FT	R	0.48	0.01	0.70			
	p-Value	0.01	0.95	0.0001			
PSA	R	0.53	0.15	0.09	0.027		
	p-Value	0.008	0.47	0.67	0.05		
G/b	R	0.17	0.19	0.36	0.22	–0.10	
	p-Value	0.41	0.37	0.08	0.29	0.63	
P+	R	0.13	0.20	0.44	0.39	0.10	0.51
	p-Value	0.53	0.35	0.03	0.05	0.63	0.01

R=Pearson's correlation coefficient.

Table VI. *Analysis of variance between the subgroups (A, B, C) of the LH I patient population.*

Group	Variable	Count	Mean	Variance	F-test	*p*-Value
A	LH (2.10-10 IU/l)	13	3.32	2.06	8.55	0.0004
B		34	3.43	1.54		
C		23	4.75	1.59		
A	Age (years)	13	66.07	45.60	0.54	0.58
B		34	63.77	50.54		
C		23	64.89	48.91		
A	TT (9-29 nmol/l)	13	13.38	31.21	5.66	0.005
B		34	15.2	21.61		
C		23	19.89	70.94		
A	FT (31-163 pmol/l)	13	30.68	46.72	4.27	0.01
B		34	32.55	90.01		
C		23	39.74	177.71		
A	PSA T (2-4 ug/l)	13	21.49	125.98	47.52	<0.0001
B		34	7.53	9.08		
C		23	4.33	1.00		
A	G/b	13	7.07	0.91	6.33	0.003
B		34	6.32	0.28		
C		23	6.39	0.43		
A	P+	13	0.40	0.03	1.02	0.36
B		34	0.30	0.04		
C		23	0.34	0.05		

explain LH dynamics after radical prostatectomy (10, 20, 22). LH I subgroups B and C might explain the results reported in metastatic prostate cancer patients under hormonal treatment, irrespective of tumor grading, where higher testosterone and lower LH levels were good prognostic factors (13). LH subgroup II might be related to the findings reported by Harper *et al*. who showed that high LH and low TT concentrations in plasma at the time of diagnosis related to a poor survival prognosis (12).

An interesting finding of the present study was that the PSA/FT was significantly higher in LH IA (0.72) and LH II (0.31) than LH IB (0.26) and LH IC (0.12). This result relates to our preceding reports where we showed that the PSA/FT ratio is growth rate parameter expressing different biological patterns as well as assessing different groups of prostate cancer patients (25); it is strongly associated with pathological prognostic factors (pT and pGS) and when ≥0.40, is strongly associated with large extensive and high-grade tumor (26); and is an effective parameter in clustering the prostate cancer population (27). Interestingly, LH I showed a significant high correlation coefficient relating TT to FT (see Table III), which means that TT is a significant predictor of FT; also we have shown that the FT/TT ratio was a significant parameter in clustering the prostate cancer patient population (28). In our opinion, aggressive prostate tumors might be related to hypo-gonadic patients in whom, as a theory, it might explain the development of castration resistance since in such patients TT no longer regulates cancer growth. According to the findings of the present report, the hypo-gonadic prostate cancer population might be clustered into primary (or testicular) and secondary (or pituitary) groups with low LH serum levels in younger

Table VII. *Analysis of variance between the different LH groups of the prostate cancer patient population.*

Variable	Group	Count	Sum	Mean	Variance	*p*-Value
LH (2.10-10 IU/l)	LH1A	13	43.10	3.32	2.06	<0.0001
	LH1B	32	110.50	3.45	1.63	
	LH1C	25	115.20	4.61	1.71	
	LH2	16	322.40	20.15	172.51	
Age (years)	LH1A	13	858.85	66.07	45.60	0.004
	LH1B	32	2036.18	63.63	52.73	
	LH1C	25	1624.46	64.98	45.86	
	LH2	16	1137.29	71.08	17.06	
TT (9-29 nmol/l)	LH1A	13	173.90	13.38	31.21	0.009
	LH1B	32	482.60	15.08	22.74	
	LH1C	25	491.80	19.67	65.62	
	LH2	16	231.60	14.48	43.87	
FT (31-163 pmol/l)	LH1A	13	398.86	30.68	46.73	0.02
	LH1B	32	1052.40	32.89	93.01	
	LH1C	25	968.30	38.73	176.11	
	LH2	16	469.20	29.33	83.34	
PSA T (2-4 ug/l)	LH1A	13	279.35	21.49	125.99	<0.0001
	LH1B	32	248.22	7.76	8.83	
	LH1C	25	107.71	4.31	0.94	
	LH2	16	150.78	9.42	95.32	
PSA/FT	LH1A	13	9.30	0.72	0.17	<0.0001
	LH1B	32	8.34	0.26	0.002	
	LH1C	25	3.10	0.12	0.00	
	LH2	16	4.95	0.31	0.05	
bGS	LH1A	13	92.00	7.08	0.91	0.01
	LH1B	32	201.00	6.28	0.27	
	LH1C	25	161.00	6.44	0.42	
	LH2	16	108.00	6.75	1.13	
P+	LH1A	13	5.22	0.40	0.04	0.10
	LH1B	32	9.14	0.29	0.04	
	LH1C	25	8.95	0.36	0.06	
	LH2	16	7.09	0.44	0.06	

patients, the former being related to the aging process (andropause) while the latter to unknown factors which have yet to be investigated.

The major limitations of the study are the small size of the prostate cancer population, as well as the lack of confirmatory studies.

Conclusion

According to LH physiopathology, the prostate cancer population at diagnosis may be clustered into hypo-gonadic and non-hypo-gonadic groups. The hypo-gonadic group has an aggressive tumor phenotype and might be divided into two more different significant subsets including primary and secondary hypo-gonadic patients, the former (LH II) including older patients with high LH levels, the latter (LH IA) including younger patients with low LH and LH/PSA levels (subgroup LH IA). The non-hypo-gonadic group shows a less aggressive tumor phenotype and according to the LH/PSA ratio might be clustered in LH IB ($0.25<LH/PSA\leq0.75$) and LH IC ($LH/PSA>0.75$), the former showing a more aggressive tumor phenotype than the latter. Confirmatory studies are needed.

References

1 Reiter E, McNamara M, Closset J and Hennen G: Expression and functionality of luteinizing hormone/chorionic gonadotropin receptor in the rat prostate. Endocrinology *136*: 917-923, 1995.

2 Huggins C and Hodges CV: Studies on prostate cancer. I: The effect of castration, of estrogen and of androgen injection on serum phosphatases in metastatic carcinoma of the prostate. Cancer Res *1*: 293-297, 1941.

3 Stamey TA, Yang N, Hay AR, McNeal JE, Freiha FS and Redwine E: Prostate-specific antigen as a serum marker for adenocarcinoma of the prostate. N Engl J Med *317*: 909-916, 1987.

4 Stenman UH, Abrahamsson PA, Aus G, Lilja H, Bangma C, Hamdy FC, Boccon-Gibod L and Ekman P: Prognostic value of serum markers for prostate cancer. Scand J Urol Nephrol Suppl *216*: 64-81, 2005.

5 Monda JM, Myers RP, Bostwick DG and Oesterling JE: The correlation between serum prostate-specific antigen and prostate cancer is not influenced by the serum testosterone concentration. Urology *46*: 62-4, 1995.
6 Imamoto T, Suzuki H, Fukasawa S, Shimbo M, Inahara M, Komiya A, Ueda T, Shiraishi T and Ichikawa T: Pretreatment serum testosterone level as a predictive factor of pathological stage in localized prostate cancer patients treated with radical prostatectomy. Eur Urol *47*: 308-12, 2005.
7 Mearini L, Costantini E, Zucchi A, Mearini E, Bini V, Cottini E and Porena M: Testosterone levels in benign prostatic hypertrophy and prostate cancer. Urol Int *80*: 134-140, 2008.
8 Yano M, Imamoto T, Suzuki H, Fukasawa S, Kojima S, Komiya A, Naya Y, Ichikawa T: The clinical potential of pretreatment serum testosterone level to improve the efficiency of prostate cancer screening. Eur Urol *51*: 375-80, 2007.
9 Ide H, Yasuda M, Nishio K, Saito K, Isotani S, Kamiyama Y, Muto S and Horie S: Development of a nomogram for predicting high-grade prostate cancer on biopsy: the significance of serum testosterone levels. Anticancer Res *28*: 2487-92, 2008.
10 Miller LR, Partin AW, Chan DW, Bruzek DJ, Dobs AS, Epstein JI and Walsh PC: Influence of radical prostatectomy on serum hormone levels. J Urol *160*: 449-453, 1998.
11 Olsson M, Ekstrom L, Schulze J, Kjellman A, Akre O, Rane A and Gustafsson: Radical prostatectomy: influence on serum and urinary androgen levels. Prostate *70(2)*: 200-5, 2010.
12 Harper ME, Pierrepoint CG and Griffiths K: Carcinoma of the prostate: relationship of pretreatment hormone levels to survival. Eur J Cancer Clin Oncol *20(4)*: 477-82, 1984.
13 Chen SS, Chen KK, Lin AT, Chang YH, Wu HH and Chang LS: The correlation between pretreatment serum hormone levels and treatment outcome for patients with prostatic cancer and bony metastasis. BJU Int *89(7)*: 710-3, 2002.
14 Walsh PC: Chemoprevention in prostate cancer. N Engl J Med *362(13)*: 1237-1238, 2010.
15 Hammond GL, Kontturi M, Maattala P, Puukka M and Vihko R: Serum FSH, LH and prolactin in normal males and patients with prostatic diseases. Clin Endocrinol *7(2)*: 129-135, (1977).
16 Kumar VL, Wafhwa SN, Kumar V and Farooq A: Androgen, estrogen, and progesterone receptor contents and serum hormone profiles in patients with benign hypertrophy and carcinoma of the prostate. J Surg Oncol *44(2)*: 122-128, 1990.
17 Anderson SO, Adami HO, Bergstrom R and Wide B: Serum pituitary and sex steroid hormone levels in the etiology of prostatic cancer – a population-based case control study. Br J Cancer *68*: 97-102, 1993.
18 Schatzl G, Madersbacher S, Thurridl T, Waldmuller J, Kramer G, Haitel A and Marberger M: High-grade prostate cancer is associated with low serum testosterone levels. Prostate *47(1)*: 52-58, 2001.
19 Hilz H, Graefen M, Noldus J, Hammerer P, Knabbe C, Huland E and Huland H: Advanced prostate cancer is associated with a decrease in serum luteinizing hormone. Eur Urol *38(3)*: 243-249, 2000.
20 Madersbacher S, Shatzl G, Bieglmayer C, Reiter BW, Gassner C, Berger P, Zidek T and Marberger M: Impact of radical prostatectomy and TURP on the hypothalamic-pituitary-gonadal axis. Urology *60*: 869-874, 2002.
21 Sofikerim M, Eskicorapaci S, Oruc O and Ozen H: Hormonal predictors of prostate cancer. Urol Int *79*: 913-918, 2007.
22 Imamoto T, Suzuki H, Yano M, Kawamura K, Kamiya N, Araki K, Komiya A, Naya Y, Shiraishi T and Ichikawa T: Does the presence of prostate cancer affect serum testosterone levels in clinically localized prostate cancer patients? Prostate Cancer Prostatic Dis *12(1)*: 78-82, 2009.
23 Fodstad P, Bjoro T, Torlakovic G and Fossa SD: No association of serum gonadal or pituitary hormone with prognostic parameters in stages T1 to T3 pN0M0 prostate cancer. J Urol *168*: 1188-1192, 2002.
24 Flemming ID, Cooper JS, Henson DE, Hutte RVP, Kennedy BJ, Murphy GP, O'Sullivan B, Sobin LH, Yarbro JN (ed.): American Joint Committee on Cancer Staging Manual, 5th ed. Philadelphia, JP Lippincott, pp. 219-222, 1997.
25 Porcaro AB, Migliorini F, Petrozziello A, Antoniolli SZ, Rubilotta E, Lacola V, Sava T, Ghimenton C, Caruso B, Monaco C and Comunale L: Investigative clinical study on prostate cancer: on the role of the pretreatment total PSA to free testosterone ratio in selecting different biology groups of prostate cancer patients. Int Urol Nephrol *42(3)*: 673-681, 2009.
26 Porcaro AB, Monaco C, Romano M, Petrozziello A, Rubilotta E, Lacola V, Sava T, Ghimenton C, Caruso B, Antoniolli SZ, Migliorini F and Comunale L: Investigative clinical study on prostate cancer part II: on the role of the pretreatment total PSA to free testosterone ratio as a marker assessing prostate cancer prognostic groups after radical retropubic prostatectomy. Urol Int *85(2)*: 152-158, 2010.
27 Porcaro AB, Petrozziello A, Romano M, Sava T, Ghimenton C, Caruso B, Migliorini F, Zecchini Antoniolli S, Rubilotta E, Lacola V, Monaco C and Comunale L: Investigative Clinical Study on Prostate Cancer Part III: Exploring totale PSA and free testosterone distributions and linear correlations in groups and subgroups of operated prostate cancer patients according to the total PSA/FT ratio. Urol Int *85(4)*: 407-409, 2010.
28 Porcaro AB, Petrozziello A, Migliorini F, Lacola V, Romano M, Sava T, Ghimenton C, Caruso B, Zecchini Antoniolli S, Rubilotta Emonaco C and Comunale L: Investigative Clinical Study on Prostate Cancer Part IV. On exploring functional relationships of total testosterone predicting free testosterone and total prostate specific antigen (PSA) in operated prostate cancer patients. Urol Int, in press.

Received December 15, 2010
Revised February 14, 2011
Accepted February 15, 2011