## Methods

Male psoriatic patients, aged between 18 and 55 years, were included from those consecutively examined at the Dermatology Department, Catholic University, Rome, Italy. Diagnosis of psoriasis was performed by clinical criteria. Patients enrolled should not have applied any local therapy for at least 1 month and not have been under traditional systemic or biological drug treatment for at least 3 months before the study investigation. Patients with erythrodermic, guttate or inverse psoriasis were excluded. Moreover, exclusion criteria were: history of cryptorchidism, orchitis, testicular torsion or trauma, hypogonadism, occupational chemical exposure, Y chromosome microdeletions, karyotype abnormalities, cystic fibrosis transmembrane conductance regulator mutations and fever and nonsteroidal anti-inflammatory drug intake within 3 months prior to enrollment. Finally, after enrollment, patients with an ultrasound testicular volume <12 mL, follicle-stimulating hormone (FSH) levels >8 mIU/L, azoospermia or a positive semen culture were excluded from any further evaluation. A group of healthy subjects matched for age, BMI and geographic origin and without a history of any of the above-mentioned conditions associated with gonadal dysfunction were included in the study as control group. All subjects signed a written informed consent form at the time of their enrollment.

For each patient and control subject, anamnestic, anthropometric (BMI) and demographic data were recorded. In psoriatic patients, duration of the disease was reported, and severity was assessed by the Psoriasis Area and Severity Index (PASI) score [12]. All enrolled subjects underwent a complete physical and andrological examination, standard semen analysis, complete microbiological analysis (including culture for bacteria, mycoplasms and chlamydia in semen and in prostatic secretion after prostate massage), and ultrasound evaluation.

Ejaculates were collected 3–5 days after sexual abstinence, and spermiograms were immediately made according to World Health Organization guidelines (2010). Semen analysis was performed

by 2 different operators (G.G. and D.M.). Semen infections were diagnosed in the presence of uniform growth of more than  $10^3$  CFU/mL of pathogenic bacteria in cultures of diluted seminal plasma and/or secretions obtained after prostatic massage.

Scrotal ultrasound was carefully performed by means of a linear transducer, in longitudinal, transverse, and oblique scans with the patients in the supine position using the WS80 Elite equipment (Samsung, USA). For each patient and control, ultrasound examination included measurement of testicular volume, documentation of testicular homogeneity and echogenicity, epididymal morphometry, including the craniocaudal diameter of the caput and cauda epididymis, and echogenicity evaluation of multiple cysts and/or of a large hydrocele. The prostate-vesicular region was assessed with rectal ultrasonography using a biplane biconvex transducer with transverse and longitudinal scans. The prostate volume was measured using the planimetric method by scanning the organ at 5-mm intervals in transverse sections. The ultrasound examinations were conducted by the same operator (D.M.). We classified our patients as affected by male accessory gland inflammation according to the ultrasound criteria previously described [13, 14].

After spermiograms, ejaculates were immediately centrifuged (300 g for 10 min), the supernatants removed, and seminal plasma stored at  $-80^{\circ}$ C until tested. Specimens were thawed and analyzed on the same day. Soluble urokinase-type plasminogen activator receptor (suPAR) was assayed, on diluted samples (1:20), using an enzyme-linked immunosorbent assay kit (suPARnostic ViroGates, Birkerød, Denmark), according to the manufacturer's instructions. The intra- and interassay coefficients of variation (CVs) were 2.4 and 5.6% at 3.2 ng/mL and 0.9 and 3.9% at 13.6 ng/mL, respectively.

A blood sample was collected at 8 a.m. for the determination of testosterone, estradiol (E2), sex hormone-binding globulin (SHBG), luteinizing hormone (LH), and FSH. Testosterone and E2

were assayed in duplicate by radioimmunoassay with the use of commercial kits by Radim (Pomezia, Italy). LH, FSH, and SHBG were assayed by immunoradiometric methods on a solid-phase coated tube, which is based on a monoclonal double-antibody technique. Reference values of the studied hormones are reported in Table 1. The intra-assay CVs (%) were 6.1% for testosterone, 2.3% for E2, 6.9% for SHBG, 5.6% for LH, and 6.9% for FSH. The interassay CVs were 9.3% for testosterone, 3.5% for E2, 8.5% for SHBG, 9.1% for LH, and 8.4% for FSH.

Comparison of categorical variables between groups was performed by the  $\chi^2$  test and Fisher exact test (2-tailed) as appropriate. Continuous variables were compared between groups by the Student *t* test (2-tailed). Correlations were assessed with Spearman  $\rho$  analysis. For all tests the value of *p* < 0.05 was considered to be significant.

This study has been approved by the local Ethical Committee and was conducted in accordance with Declaration of Helsinki guidelines.