Title:

Pharmacokinetics and bioavailability of a new testosterone gel formulation in comparison to Testogel® in healthy men

Authors:

H Olsson, R Sandström, A Neijber, D Carrara, L Grundemar

Journal:

Clinical Pharmacology in Drug Development 2014

Original Article

Clinical Pharmacology in Drug Development
3(5) 358–364

DOI: 10.1002/cpdd.110

Pharmacokinetics and Bioavailability of a New Testosterone Gel Formulation in Comparison to Testogel® in Healthy Men

Håkan Olsson[1], Rikard Sandström[1], Anders Neijber[2], Dario Carrara[3], and Lars Grundemar[1]

Abstract

This randomized, open-label, multiple-dose three-way cross-over study compared the pharmacokinetics of a new testosterone gel formulation in two strengths, testosterone gel 1% and testosterone gel 2% (FE 999303), with Testogel® in 11 testosterone-suppressed healthy men. Subjects received one of six treatment sequences; 50 mg of testosterone was administered once daily for 7 consecutive days, with different treatments separated by washout-periods of 6–9 days. Testosterone gel 1% and testosterone gel 2% displayed greater relative bioavailability (2.6- and 1.6-fold, respectively) than Testogel on Day 1, which persisted, to a smaller extent, on Day 7. Initial absorption was highest and most rapid for testosterone gel 1% and 2%, showing apparent first-order absorption kinetics. Maximum serum concentrations (C_{max}) were 6.25 and 2.97 ng/mL, respectively, occurring ~5–6 hours post-application on Day 1 versus C_{max} of 1.71 ng/mL after ~24 hours with Testogel, showing apparent zero-order absorption kinetics. Similar differences were observed on Day 7. All treatments appeared to reach approximately the same steady-state level within the first 24 hours. No application-site skin reactions occurred with any preparation. In conclusion, the new testosterone formulation showed higher bioavailability, and the ability to deliver more testosterone in a smaller volume.

Keywords

gel, testosterone, dihydrotestosterone, pharmacokinetics, bioavailability

Male hypogonadism is a clinical syndrome characterized by testosterone levels below the normal range (<3 ng/mL) and associated with symptoms[1] such as low libido, erectile dysfunction, increased body fat and decreased muscle mass, reduced bone mineral density, and depressed mood.[2] Testosterone supplementation aims to restore serum testosterone to the normal physiological range and has been used in a vast number of men for several decades since the market introduction of testosterone formulations for male hypogonadism. Testosterone supplementation has been associated with a range of benefits in hypogonadal men including improvements in sexual function, bone mineral density, and body composition.[2–4]

The bioavailability of oral testosterone is low due to poor absorption, rapid metabolism, and inactivation by the liver.[5] Transdermal testosterone delivery allows absorption directly into the systemic circulation at a controlled rate, and while a range of testosterone formulations are available, the most commonly used formulations are testosterone gels, the advantages of which include a generally good tolerability profile, ease of use, and dose flexibility.[1,6–8] However, the bioavailability of current gels is usually low and the administered gel volumes are large, which can cause inconvenience to patients. There is also the potential risk of transfer to a partner or child via skin contact with the application site[9–11] which can cause clinical virilisation.[12] Thus, there is a great need for better testosterone gels with higher transdermal bioavailability and lower gel volume per dose.

Two concentrations of a new testosterone gel formulation, testosterone gel 1% and testosterone gel

[1]Ferring Pharmaceuticals A/S, Experimental Medicine, Copenhagen, Denmark
[2]Ferring Pharmaceuticals A/S, Urology and Endocrinology, Copenhagen, Denmark
[3]Ferring Galenisches Labor AG, Allschwill, Switzerland

Submitted for publication 3 July 2013; accepted 13 February 2014

Corresponding Author:
Håkan Olsson, Ferring Pharmaceuticals A/S, Kay Fiskers Plads 11, DK-2100 Copenhagen, Denmark
(e-mail: hakan.olsson@ferring.com)

2% (FE 999303) have been developed with the aim of achieving rapid transdermal absorption and improved testosterone bioavailability.[13] Increased bioavailability could allow administration of a lower dose and/or smaller volume, reducing the potential for interpersonal transfer of unabsorbed gel. This new testosterone gel utilizes a transdermal delivery system based on the Advanced Transdermal Delivery™ (ATD™) gel technology, which has been developed to provide enhanced passive skin permeation.[14] ATD™ technology utilizes a combination of solvents and permeation enhancers that facilitate the rapid passage of active agents across the skin and the new testosterone gel formulation has shown promising in vitro results regarding skin penetration of testosterone.[13]

This study investigated the pharmacokinetic profile, relative bioavailability, and tolerability of the new testosterone gel 1% and testosterone gel 2% formulations in comparison with the marketed product Testogel® (marketed as Androgel® in the USA) in healthy men.

Methods

Study Design

This randomized, open-label, active control, multiple-dose three-way cross-over study was conducted in Germany involving 11 healthy men. All subjects provided written informed consent prior to any study-related procedure. This study was performed according to the Declaration of Helsinki, in compliance with Good Clinical Practice (GCP), and the protocol was approved by an Independent Ethics Committee (Ethikkommission der Ärztekammer Hamburg, Hamburg, Germany).

To minimize interference with pharmacokinetic assessments, endogenous testosterone production was suppressed by the gonadotrophin-releasing hormone receptor agonist, triptorelin. Subjects received an intramuscular injection of 3.75 mg Decapeptyl® N 1-month depot on Day −21, and a second injection on Day 8 in the first treatment period to maintain testosterone suppression throughout the duration of the study.

After the 21-day run-in and down-regulation period, subjects were randomized to one of six treatment sequences. In each sequence, subjects received daily administration for 7 consecutive days of either 5 g testosterone gel 1% (FE 999303; Ferring Galenisches Labor AG, Allschwill, Switzerland), 2.5 g testosterone gel 2% (FE 999303; Ferring Galenisches Labor AG), or 5 g Testogel® (1% testosterone; Laboratories Besins International [for Bayer Vital Gmbh], Drogenbos, Belgium). The constituents of the gels are listed in Table 1. Treatment periods were separated by washout-periods of 6–9 days. Each dose (equivalent to 50 mg testosterone) was applied to the same area of the dry and clean abdomen once daily. The gel was evenly spread over a 1,000 cm^2 area, marked with a skin-marker using a stencil before administration, until completely absorbed. The application site was allowed to dry before being covered with clothes and subjects were not allowed to shower or bathe until at least 6 hours post-application.

Table 1. List of Constituents for Testosterone 1% and 2% Gels and Testogel 1%

Testosterone Gel 1% and 2%	Testogel 1%
Ethanol	Ethanol
Propylene glycol	Testosterone
Diethylene glycol monoethyl ether	Carbomer 980
Testosterone	Isopropyl myristate
Carbomer 980	Sodium hydroxide
Trolamine	
Edetate disodium	

Subjects

Healthy Caucasian men, 18–45 years of age with a body mass index (BMI) of 18.0–30.0 kg/m^2 and body weight of 50–100 kg were eligible for inclusion. Subjects were physically and mentally healthy, as confirmed by examinations prior to enrolment. Endogenous testosterone level had to be $\leq$1 ng/mL on Day −1.

The main exclusion criteria included: skin color (natural or acquired) that prevented evaluation of changes in application-site skin color; skin disease that might affect the absorption of testosterone, or tattoos or scars in the application area, or prior history of skin irritability with transdermal testosterone drug products or their excipients.

Pharmacokinetic Parameters

Individual baseline-corrected pharmacokinetic parameters were calculated by non-compartmental analysis (NCA) using the software WinNonlin®Pro (Pharsight Corporation, Cary, NC, USA) using individual actual sampling time points relative to dosing. The baseline testosterone value was the mean of the values obtained prior to treatment administration. Serum concentrations below the lower limit of quantitation (LLQ) and missing values were excluded from the NCA.

Pharmacokinetic parameters were estimated from serum concentration–time data for testosterone up to 24 hours after the first administration (Day 1) and up to 48 hours after the last administration (Day 7). Pharmacokinetic parameters calculated were the area under the concentration–time curve from the first and last doses (Days 1 and 7) (AUC_τ), C_{max} (maximum serum concentration), t_{max} (time to C_{max}), and $t_{1/2}$ (elimination half-life) after the first and last doses. The areas under the concentration–time curves were used to calculate the

relative bioavailabilities of the testosterone gel 1% and 2% formulations with reference to Testogel.

Assessments

Blood samples for measurement of serum testosterone were collected immediately prior to and at 2, 4, 6, 8, 12, and 16 hours after the first treatment administration, pre-dose on Days 2–6, and pre-dose and at 2, 4, 6, 8, 12, 16, 24, 48 hours after the last administration. Analyses of serum testosterone and dihydrotestosterone (DHT) were performed by the Department of Bioanalysis at Ferring Pharmaceuticals, and Scope Life Sciences GmbH, respectively, in Good Laboratory Practice (GLP)-compliant facilities. The quantification of testosterone and DHT in human serum was made using validated liquid chromatography tandem-mass spectrometry methods. Samples for testosterone analysis were prepared by supported liquid extraction from 0.25 mL serum using an Isolute SLE+ 400 mg 96-well plate, followed by reconstitution in 20% ethanol in water. After injection of 25 μL to a Waters ACQUITY UPLC BEH C18 1.7 μm column, testosterone was eluted by a 10–100% gradient of methanol/1 mM ammonium bicarbonate, pH 10. The mass spectrometer (Waters Quattro micro triple quadropole mass spectrometer) was operated with positive electrospray ionization monitoring the ions m/z $289.2 \rightarrow 96.7 \pm 1.5$ for testosterone and m/z $292.2 \rightarrow 96.7 \pm 1.5$ for the internal standard [$^2H_3$]testosterone. The lower limit of quantification was 100 pg/mL and the upper limit 15,000 pg/mL. The quality control samples were all within ±6% bias, well below the pre-defined limit ±15%, with a back-calculated bias of the calibration samples (100–15,000 pg/mL) of ±2%.

Samples for DHT analysis were prepared by liquid–liquid tert-butylmethylether extraction from 0.3 mL serum followed by solid-phase extraction on Oasis μElution plates in methanol. A portion of the eluate was injected onto the liquid chromatography column using a mobile phase of 0.05% formic acid, 2 mM ammonium acetate and methanol. The mass spectrometer was operated with positive electrospray ionization and multiple reaction monitoring using the transitions m/z $291 \rightarrow 255$ for DHT and m/z $294 \rightarrow 258$ for the internal standard (5-alpha-androstan-17β-ol-3-one-16,16,17-d3). Instrumentation comprised a Mercury MS Synergi Max-RP column (2 μm, 2 mm × 20 mm, Phenomenex, Torrance, CA, USA) with a Micromass Quattro Ultima detector with electrospray ionization. The calibration range was 0.2–10 ng/mL and the lower limit of quantification was 0.2 ng/mL. In addition to the general safety and tolerability assessments, administration site reactions were specifically monitored.

Statistical Methods

Statistical analyses included descriptive statistics reflecting the explorative nature of the study. Unless otherwise stated, all tests were two-sided with the significance level 5%. Missing values were not imputed.

Pharmacokinetic parameters AUC_τ and C_{max} were compared across treatments using an analysis of variance (ANOVA) model for the log-transformed values. The ratio of the pharmacokinetic parameters was estimated along with 90% confidence limits. As a complement to the testosterone analyses the same exercises were performed with DHT.

Results

Study Subjects

Eleven subjects were randomized to one of six treatment sequences, two to each sequence except for the sequence: Testogel—testosterone gel 1%—testosterone gel 2%, to which only one subject was randomized. One subject withdrew consent during treatment period 2, leaving 10 who completed all three-treatment periods. The 11 randomized subjects were aged 23–45 (mean 36.5) years with a body weight of 76–100 (mean 86.4) kg and BMI of 23.8–29.5 (mean 26.0) kg/m^2. Mean baseline testosterone values were 0.48, 0.39, and 0.34 ng/mL for the testosterone gel 1%, testosterone gel 2%, and Testogel groups, respectively; for DHT, mean baseline values were 0.020, 0.059, and 0.067 ng/mL, respectively.

Pharmacokinetics

Testosterone absorption was substantially more rapid with the testosterone gel 1% and 2% formulations compared with Testogel, with testosterone gel 1% providing the most rapid absorption (Figure 1). Mean t_{max} occurred at similar time points for testosterone gel 1% and 2% formulations, around 5–6 hours after application, on both Days 1 and 7 (Table 2). In contrast, after Testogel administration the testosterone concentration slowly increased to reach the steady-state level approximately 20 hours after dosing on Day 1, and without a marked t_{max} after administration on Day 7 (albeit a maximal value was recorded at approximately 13 hours) (Figure 1). The corresponding C_{max} for testosterone gel 1% and 2% on Day 1 was 6.25 and 2.97 ng/mL, respectively, compared to 1.71 ng/mL for Testogel (Table 2). The same order of rank, and similar concentrations, were seen on Day 7, the maximal testosterone concentrations being 6.67, 3.16, and 2.22 ng/mL for testosterone gel 1%, 2%, and Testogel, respectively. The ANOVA of baseline-corrected testosterone concentrations demonstrated significantly higher C_{max} with the new testosterone gel formulations versus Testogel on Days 1 and 7 (Table 3).

The mean (geometric) of baseline-corrected AUC_τ for testosterone was substantially greater for the testosterone gel 1% and 2% formulations compared

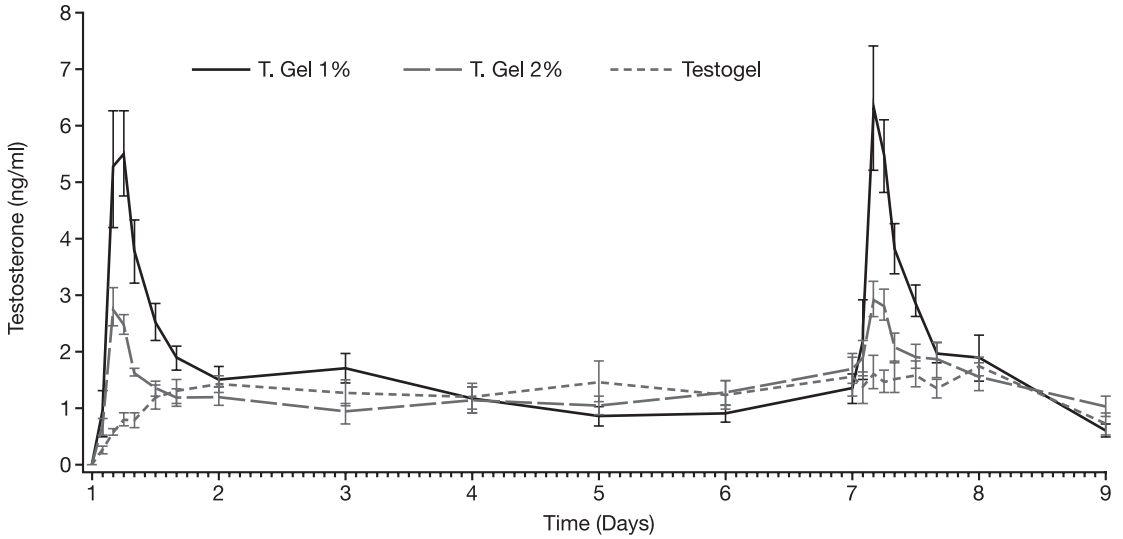

Figure 1. Mean (SE) testosterone concentrations for the testosterone gel 1% (T. Gel 1%) and 2% (T. Gel 2%) formulations and Testogel from Days 1 to 9.

with Testogel, the testosterone gel 1% formulation showing the greatest absorption. The ANOVA of baseline-corrected testosterone concentrations showed that absorption with testosterone gel 1% and 2% was significantly higher compared to Testogel (Table 3). The geometric mean ratios of AUC_τ versus Testogel (i.e., the relative bioavailability) on Day 1 were 2.63 ($P < 0.001$) and 1.61 ($P = 0.001$) for testosterone gel 1% and 2% formulations, respectively.

For all three preparations, the pharmacokinetic profiles after the last administration on Day 7 were very similar to those after the first administration on Day 1, and similar differences in amount absorbed were observed between the formulations on Day 7, albeit to a slightly smaller extent than on Day 1. The corresponding relative bioavailabilities on Day 7 were 2.00 ($P = 0.001$) and 1.39 ($P = 0.05$). All three treatments appeared to reach about the same steady-state level (1–1.5 ng/mL) within the first 24 hours.

The terminal half-life was similar for the two new testosterone gel formulations on Day 1 (~15 hours) but on Day 7, $t_{1/2}$ was more than twice as long for the testosterone gel 2% formulation (~21 hours) than for the 1% formulation (~9 hours) (Table 2). On both Days 1 and 7, $t_{1/2}$ for Testogel was considerably longer than for either of the new testosterone gel formulations (~53 and

Table 2. Summary of Pharmacokinetic Parameters: Maximum Serum Concentration (C_{max}), Time to C_{max} (t_{max}) and Elimination Half-Life ($t_{1/2}$)

Parameter	Day	Testosterone Gel 1% (n = 11)	Testosterone Gel 2% (n = 10)	Testogel (n = 11)
C_{max} (ng/mL)				
Mean (SD)	1	6.25 (3.12)	2.97 (0.85)	1.71 (0.78)
Range		2.34–12.3	1.76–4.34	0.82–3.16
Mean (SD)	7	6.67 (3.45)	3.16 (0.98)	2.22 (0.93)
Range		2.18–13.15	1.65–4.69	0.81–3.60
T_{max} (h)				
Median	1	6	5	24
Range		4–6	4–12	12–24
Median	7	4	5	10
Range		4–6	2–12	4–24
$t_{1/2}$ (h)				
Mean (SD)	1	15.2 (17.9)	14.6 (8.5)	52.5 (53.4)
Range		3.1–67.3	3.5–28.7	8.5–154
Mean (SD)	7	9.4 (6.5)	21.0 (13.3)	71.6 (66.4)
Range		4.2–28.1	7.3–50.3	8.5–155

SD, standard deviation.

Table 3. Intra-Individual Comparison of Serum (a) Testosterone and (b) Dihydrotestosterone Geometric Mean and Mean Ratios for AUC_τ and C_{max} for the Testosterone Gel 1% and 2% Formulations Versus Testogel for Days 1 and 7

Parameter	Day	Testosterone Gel 1% (n = 11) Geometric Mean	Testosterone Gel 2% (n = 10) Geometric Mean	Testogel (n = 11) Geometric Mean	Mean Ratio	90% CI	*P*-Value
(a) Testosterone							
AUC_τ (ng/mL h)	1	57.1	—	21.7	2.63	2.18; 3.17	<0.001
	7	66.7	—	33.4	2.00	1.53; 2.60	0.001
	1	—	35.0	21.7	1.61	1.33; 1.96	0.001
	7	—	46.6	33.4	1.39	1.06; 1.83	0.050
C_{max} (ng/mL)	1	5.49	—	1.59	3.45	2.66; 4.49	<0.0001
	7	5.74	—	2.07	2.78	2.07; 3.73	<0.0001
	1	—	2.88	1.59	1.81	1.38; 2.38	0.0015
	7	—	3.18	2.07	1.54	1.14; 2.08	0.0238
(b) Dihydrotestosterone							
AUC_τ (ng/mL h)	1	15.7	—	8.8	1.79	1.48; 1.16	<0.0001
	7	16.2	—	11.5	1.42	1.08; 1.85	0.0376
	1	—	11.3	8.8	1.29	1.06; 1.58	0.0415
	7	—	13.5	11.5	1.18	0.89; 1.57	0.3266
C_{max} (ng/mL)	1	1.0	—	0.6	1.71	1.39; 2.11	0.0003
	7	1.1	—	0.7	1.63	1.31; 2.02	0.0013
	1	—	0.7	0.6	1.22	0.98; 1.54	0.1375
	7	—	0.8	0.7	1.16	0.92; 1.46	0.2750

AUC_τ, area under the concentration–time curve from the last dose (Day 7) and 24 hours post-dose; C_{max}, maximum serum concentration. Endpoints are ln-transformed before analysis, and results are transformed back and presented as ratios. The model is a mixed linear model and includes treatment and period as fixed effects, and subject as random effect. The mean is the geometric least square means estimated from the model. The *P*-value is based on a two-sided test of the difference; 90% confidence intervals (CI) are given.

72 hours, respectively). However, due to the shallow slopes in the terminal parts of the curve, the half-lives should be treated with caution.

The time-course of serum DHT concentrations followed the serum testosterone time-course closely, albeit with smaller concentration amplitudes after administration (Figure 2). Consequently, the ANOVA of baseline-corrected DHT showed smaller differences in AUC_τ and C_{max} between testosterone gel 1% or 2% formulations and Testogel (Table 3). Nevertheless, for testosterone gel 1% both AUC_τ and C_{max} were significantly higher than Testogel on Day 1 and 7. For testosterone gel 2% the AUC_τ on Day 1 was significantly higher than Testogel ($P < 0.05$) but for AUC_τ on Day 7 and C_{max} on Days 1 and 7 the difference between these treatments was not statistically significant.

Safety

There were no administration-site reactions with any treatment, and no serious or severe adverse events. Single treatment-related adverse events occurred with all treatments with similar low frequency, and no clinically significant changes in vital signs, ECG, hematology, urinalysis or clinical chemistry values occurred.

Discussion

In this trial in healthy men with suppressed testosterone, the new testosterone gel 1% and 2% formulations (FE 999303) exhibited higher testosterone bioavailability and faster absorption compared to a commonly used product. Consequently, testosterone reached levels close to normal within a shorter time frame subsequent to application of the testosterone gel 1% and 2% formulations compared with Testogel.

The time–concentration profiles of testosterone demonstrated that the three preparations had different absorption patterns during the 24 hours following application. The new testosterone gel 1% and 2% formulations showed a rapid first-order like absorption pattern with only a few hours to C_{max}, while Testogel displayed a slower absorption rate resembling a zero-order absorption pattern with a much longer time to reach maximal concentration. Since the intrinsic half-life of testosterone is within 1 hour, the elimination of all three preparations is determined by the absorption rate, the slower absorption of Testogel being reflected in a substantially longer half-life. Since the new testosterone gel 1% and 2% formulations both showed the same overall time–concentration profile, albeit with a different

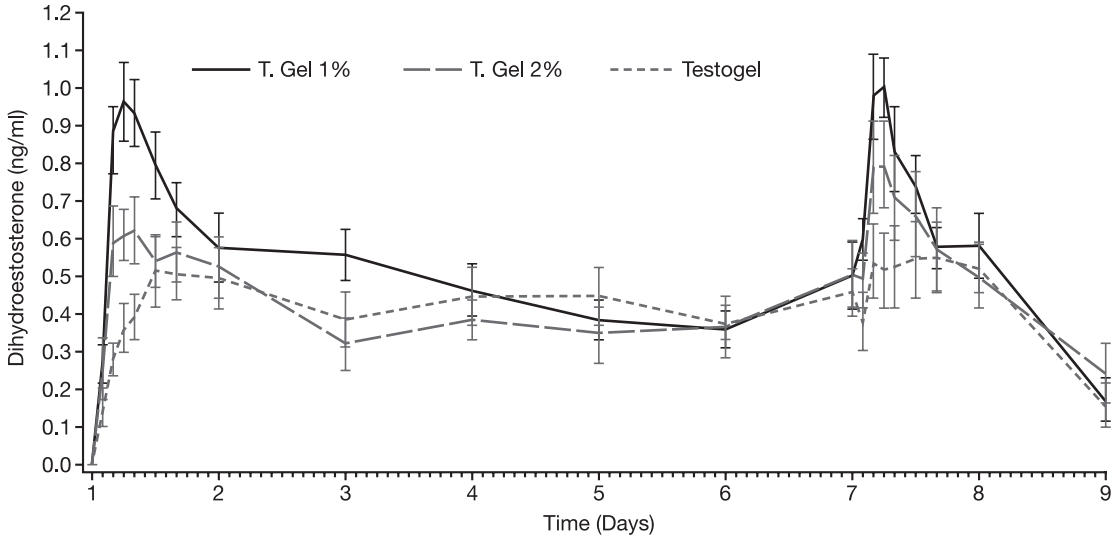

Figure 2. Mean (SE) dihydrotestosterone concentrations for the testosterone gel 1% (T. Gel 1%) and 2% (T. Gel 2%) formulations and Testogel from Days 1 to 9.

magnitude, it is likely that the difference between these new formulations and Testogel is a result of differences in composition. In accordance with other transdermal testosterone products, the three formulations in the present study produced a rise in DHT levels concomitant with increased testosterone levels, reflecting metabolic conversion via 5 α-reductase in the skin.[15]

The exact mechanism behind the substantially higher testosterone absorption with the testosterone gel 1% versus the 2% formulation is as yet unknown. The composition of the formulations were the same, the factors differing being the amount of gel and the testosterone concentration. Since a higher concentration in diffusion driven absorption would be expected to produce a higher absorption rate, the hypothesis is that the amount of gel applied has an impact on the absorption of testosterone. In vitro skin permeation studies have shown that increasing the amount or volume of gel applied per cm^2 increases testosterone permeation.[12] The greater application volume of the 1% formulation (5 vs. 2.5 g with the 2% formulation) may thus provide an explanation for the higher absorption.

Testosterone steady state was reached after the first dose with all three preparations. Moreover, the rapid establishment of persistent and similar pre-dose testosterone levels from Days 2 to 7, strongly indicated that there was no accumulation of testosterone with any preparation. For the new testosterone gel 1% and 2% formulations this was also supported by the similar time courses for the testosterone concentration on Days 1 and 7.

Endogenous testosterone production and release shows a circadian rhythm with higher testosterone levels in the morning, although this circadian pattern can become blunted with aging.[16,17] The new testosterone gel 1% and 2% formulations, with their more rapid absorption and the testosterone peaks clearly above that of the steady-state level, may thus provide a circadian-like rhythm resembling the natural diurnal variation in testosterone.

Unabsorbed testosterone from gels poses a risk of transfer to the skin of partners and/or children. Indeed, topical androgens can increase testosterone levels in exposed women and children[9] and cause hyperandrogenism.[10,18–22] In the current study, the testosterone gel 1% and 2% formulations delivered a higher proportion of the testosterone dose to the circulation compared to Testogel, and so may reduce the risk of testosterone transfer to partners/children. Moreover, the amount of testosterone contained in the skin, and never reaching the circulation, might be reduced with the new formulations, contributing to a decrease in the amount available for contact transfer.

In conclusion, the new testosterone gel 1% and 2% formulations achieved greater and more rapid absorption of testosterone, with a more circadian-like concentration profile, than Testogel without any application-site reactions.

Declaration of Conflicting Interests

Håkan Olsson, Rikard Sandström, Anders Neijber, Dario Carrara and Lars Grundemar are all employees of Ferring Pharmaceuticals.

Funding

This study was funded by Ferring Pharmaceuticals. Medical writing assistance (funded by Ferring Pharmaceuticals) was provided by Thomas Lavelle of Bioscript Medical.

References

1. Lunenfeld B, Arver S, Moncada I, Rees DA, Schulte HM. How to help the aging male? Current approaches to hypogonadism in primary care. *Aging Male.* 2012;15:187–197.
2. Wang C, Nieschlag E, Swerdloff RS, et al. ISA, ISSAM, EAU, EAA and ASA recommendations: investigation, treatment and monitoring of late-onset hypogonadism in males. *Aging Male.* 2009;12:5–12.
3. Liverman CT, Blazer DG, eds. *Testosterone and Aging: Clinical Research Directions*. Washington, DC: National Academies Press; 2004.
4. Miner M, Canty DJ, Shabsigh R. Testosterone replacement therapy in hypogonadal men: assessing benefits, risks, and best practices. *Postgrad Med.* 2008;120:130–153.
5. Wang C, Swerdloff RS, Iranmanesh A, et al. Transdermal testosterone gel improves sexual function, mood, muscle strength, and body composition parameters in hypogonadal men. *J Clin Endocrinol Metab.* 2000;85:2839–2853.
6. Dandona P, Rosenberg MT. A practical guide to male hypogonadism in the primary care setting. *Int J Clin Pract.* 2010;64:682–696.
7. McNicholas T, Ong T. Review of Testim gel. *Expert Opin Pharmacother.* 2006;7:477–484.
8. Bhasin S, Cunningham GR, Hayes FJ, et al. Testosterone therapy in men with androgen deficiency syndromes: an Endocrine Society clinical practice guideline. *J Clin Endocrinol Metab.* 2010;95:2536–2559.
9. Lakshman KM, Basaria S. Safety and efficacy of testosterone gel in the treatment of male hypogonadism. *Clin Interv Aging.* 2009;4:397–412.
10. de Ronde W. Hyperandrogenism after transfer of topical testosterone gel: case report and review of published and unpublished studies. *Hum Reprod.* 2009;24:425–428.
11. Surampudi PN, Wang C, Swerdloff R. Hypogonadism in the aging male diagnosis, potential benefits, and risks of testosterone replacement therapy. *Int J Endocrinol.* 2012;2012: Article ID 625434; doi: 10.1155/2012/625434.
12. Grenier A, Carrara DN, Ferring BV. International patent classification A61 K 9/06, A61 K 31/568, A61 K 47/10, A61 K 31/58, A61 P 5/26, A61 K 9/00. World Intellectual Property Organization, International Publication Number WO 2012/101016 A1; International Publication Date 2 August 2012.
13. Alberti I, Grenier A, Kraus H, Carrara DN. Pharmaceutical development and clinical effectiveness of a novel gel technology for transdermal drug delivery. *Expert Opin Drug Deliv.* 2005;2:935–950.
14. Täuber U, Schröder K, Düsterberg B, Matthes H. Absolute bioavailability of testosterone after oral administration of testosterone-undecanoate and testosterone. *Eur J Drug Metab Pharmacokinet.* 1986;11:145–149.
15. Swerdloff RS, Wang C, Cunningham G, et al. Long-term pharmacokinetics of transdermal testosterone gel in hypogonadal men. *J Clin Endocrinol Metab.* 2000;85:4500–4510.
16. Bremner WJ, Vitiello MV, Prinz PN. Loss of circadian rhythmicity in blood testosterone levels with aging in normal men. *J Clin Endocrinol Metab.* 1983;56:1278–1281.
17. Tenover JS, Matsumoto AM, Clifton DK, Bremner WJ. Age-related alterations in the circadian rhythms of pulsatile luteinizing hormone and testosterone secretion in healthy men. *J Gerontol.* 1988;43:M163–M169.
18. Yu YM, Punyasavatsu N, Elder D, D'Ercole AJ. Sexual development in a two-year-old boy induced by topical exposure to testosterone. *Pediatrics.* 1999;104:e23.
19. Kunz GJ, Klein KO, Clemons RD, Gottschalk ME, Jones KL. Virilization of young children after topical androgen use by their parents. *Pediatrics.* 2004;114:282–284.
20. Brachet C, Vermeulen J, Heinrichs C. Children's virilization and the use of a testosterone gel by their fathers. *Eur J Pediatr.* 2005;164:646–647.
21. Bhowmick SK, Ricke T, Rettig KR. Sexual precocity in a 16-month-old boy induced by indirect topical exposure to testosterone. *Clin Pediatr (Phila).* 2007;46:540–543.
22. Merhi ZO, Santoro N. Postmenopausal virilization after spousal use of topical androgens. *Fertil Steril.* 2007;87:976e.13–976e.15.