Open Forum Infectious Diseases

MAJOR ARTICLE

Incidence and Predictors of Serological Treatment Response in Early and Late Syphilis Among People Living With HIV

Vincenzo Spagnuolo,[1,2] Andrea Poli,[2] Laura Galli,[2] Silvia Nozza,[2] Simona Bossolasco,[2] Massimo Cernuschi,[2] Myriam Maillard,[2] Hamid Hasson,[2] Nicola Gianotti,[2] Monica Guffanti,[2] Adriano Lazzarin,[2] and Antonella Castagna[1,2]

[1]Vita-Salute San Raffaele University, Faculty of Medicine and Surgery, Milan, Italy; [2]IRCCS San Raffaele Scientific Institute, Clinic of Infectious Diseases, Milan, Italy

Background. Few studies have investigated predictors of serological response to syphilis treatment in people living with HIV (PLWH).

Methods. This was a retrospective, longitudinal study on PLWH who were diagnosed with and treated for syphilis who had an assessable serological response between January 2004 and June 2016. Serological treatment response (TR) was defined as a ≥4-fold decline in rapid plasma reagin (RPR) titers or a reversion to nonreactive (if RPR ≤1:4 at diagnosis) 12 months after treatment for early syphilis and 24 months after treatment for late syphilis. Factors associated with a TR were assessed with multivariate Cox proportional hazard models for recurrent events.

Results. A total of 829 episodes of syphilis (686 early, 143 late) in 564 patients were recorded. TR was observed in 732 (88%) syphilis episodes. The proportion of TR differed between early and late syphilis (89% vs 83%, respectively; $P = .045$). For early syphilis, TR was associated with a higher nadir CD4+ cell count (adjusted hazard ratio [AHR], 1.06; $P = .029$), an RPR titer >1:32 at diagnosis (AHR, 1.26; $P = .009$), secondary syphilis (AHR, 1.29; $P = .008$), and cases of syphilis diagnosed in more recent calendar years (AHR, 1.36; $P < .0001$). In late syphilis, TR was more likely to occur for first infections (AHR, 1.80; $P = .027$), for episodes that occurred in more recent years (AHR, 1.62; $P = .007$), and for RPR titers >1:32 at diagnosis (AHR, 2.04; $P = .002$). TR was not associated with the type of treatment regimen in early and late syphilis.

Conclusions. Higher RPR titers at diagnosis and a diagnosis of syphilis that was made in more recent years were associated with TR in early and late syphilis.

Keywords. early syphilis; late syphilis; serological response; HIV; antibiotic treatment.

In Western countries, there has been a resurgence of syphilis since 2000 [1], with a peak prevalence in men who have sex with men (MSM) living with HIV [2, 3]. HIV infection has been associated with lower rates of serologically defined treatment responses, compared with the rates that have been observed in the general population [4, 5], although conflicting results on this topic have been reported [6, 7].

A few studies [4–10] have investigated for predictors of treatment response of syphilis in people living with HIV (PLWH), showing controversial results [4–11].

Different guidelines [12, 13] have recommended the same treatment regimens for PLWH and the general population, regardless of the stage of syphilis; however, concerns regarding the treatment of syphilis in PLWH remain. A high proportion of clinicians who care for patients with HIV continue to use 3 doses of benzathine penicillin G (BPG) to treat early syphilis [9, 14] because of possible concerns regarding the lower efficacy of the recommended 1-dose BPG regimen. Additionally, limited data [15, 16] are available on the efficacy of the alternative regimen, doxycycline, especially for the treatment of late latent syphilis.

Therefore, the aims of this study were to evaluate the response rate of treatment of syphilis in a large cohort of PLWH and to identify treatment response predictors (including different treatment regimens) in early and late stages of syphilis.

METHODS

This retrospective, longitudinal study was approved by the Ethics Committee of the San Raffaele Scientific Institute (approval No. 34–2017) and evaluated data from the HIV Clinic of San Luigi Center, San Raffaele Hospital, in Milan, Italy.

Received 3 October 2018; editorial decision 24 November 2018; accepted 28 November 2018 ; Published online November 30, 2018.

Correspondence: Vincenzo Spagnuolo MD, Vita-Salute San Raffaele University, via Stamira d'Ancona 20, 20127 Milan, Italy (spagnuolo.vincenzo@hsr.it).

Open Forum Infectious Diseases®

DOI: 10.1093/ofid/ofy324

In the present study, we included PLWH who were diagnosed with and treated for syphilis between January 2004 and June 2016 (freezing date). A nontreponemal test (rapid plasma reagin [RPR], RPR Plus, Diesse Srl, Monteriggioni (SI), Italy) and a treponemal test (*Treponema pallidum* hemagglutination assay [TPHA], Mycrosyph TPHA, Axis Shield, Dundee, Scotland) were used for the diagnosis of syphilis.

The detection of positive titers in patients without a history of previously treated syphilis and with previous negative RPR and TPHA titers indicated a patient's first diagnosis of syphilis. Among patients with documented previous treatment(s), a seroconversion of the RPR titer (if previously negative) or a ≥4-fold increase in RPR titer was considered a new episode of syphilis (re-infection).

The syphilis stages were classified as early (primary, secondary, and early latent) or late (tertiary and late latent) according to the clinical examination, patient's history, and timing of infections. Primary syphilis was diagnosed in the presence of RPR and TPHA titers that were compatible with a syphilis episode, and the concomitant presence of a single or multiple genital, perineal, perianal, or rectal (if an anoscopy/rectoscopy was performed) ulcers. A case of secondary syphilis was diagnosed in the presence of treponemal and nontreponemal titers that were suggestive of a syphilis episode and a concomitant maculopapular skin rash and/or mucous patches/condylomata lata and/or constitutional symptoms (fever, diffuse lymphadenopathy, malaise). Patients with positive RPR and TPHA titers and cardiovascular involvement or gummas were diagnosed with tertiary syphilis.

Asymptomatic subjects with RPR and TPHA titers suggestive of a syphilis episode were diagnosed with early latent syphilis if a previous syphilis serology was available, was not compatible with an active infection, and was performed no later than 1 year before the current syphilis episode. Otherwise, asymptomatic patients were diagnosed with late latent syphilis.

All syphilis diagnoses that were without a documented history of treatment or an available serologic follow-up or neurosyphilis cases (because of the different treatments and diagnostic approaches) were excluded from the analysis. Serological treatment response was defined as a ≥4-fold decline—2 dilutions (eg, from 1:64 to 1:16)—in RPR titers or a reversion to nonreactive (if RPR ≤ 1:4 at diagnosis) 12 months after treatment for early syphilis or 24 months after treatment for late syphilis in the absence of documented re-infection [12].

Serofast status [11] was diagnosed in subjects who experienced a single episode of syphilis during the study period and who were successfully treated but did not revert their RPR titers to nonreactive by the end of the study.

Data Analysis

Results were described as median (interquartile range [IQR]) or frequency (%). Continuous and categorical variables were compared with the nonparametric Kruskal-Wallis test, Mann-Whitney rank-sum test, and the chi-square or Fisher exact test.

The Cochran-Mantel-Haenszel test was applied to assess linear trend over time of BPG use.

Different definitions of the study follow-up period were used in relation to the considered outcome. With regards to the estimate of the incidence rates of syphilis, the study follow-up accrued from the date of the first visit to the last available visit, or the freezing date minus the days that were required to treat syphilis (ie, total follow-up time minus time to treat syphilis).

With regards to the estimate of the incidence rates of treatment response, the follow-up time was accrued from the date of syphilis diagnosis to (1) the date of treatment response or (2) in case of a lack of response to treatment, 12 or 24 months from diagnosis of early and late syphilis episodes, respectively. To evaluate for serofast status, the time of follow-up was accrued from the date of syphilis diagnosis to the date of sero-reversion, or the date of the last available visit.

The crude incidence rates of syphilis (95% confidence intervals [CIs]) were expressed per 1000 person-years of follow-up (PYFU) and calculated as the total number of episodes divided by the cumulative study follow-up at risk for syphilis, contributed by all subjects. The crude incidence rates of treatment response (95% CIs) were expressed per 1000 person-months of follow-up (PMFU) and calculated as the total number of episodes of treatment response divided by cumulative follow-up at risk contributed by all subjects.

Incidence rates were estimated and compared by use of Poisson regression models. The Andersen-Gill extension of the Cox regression model for recurrent events was used to model treatment success on the individual level with a robust (sandwich) variance estimator to account for the matched nature of the sample [17–19]. The adjusted hazard ratios (HRs) of serofast response and the corresponding 95% CIs were reported.

Further multivariate Cox proportional hazard models were calculated to identify factors that were associated with a serofast response in the overall sample and in the early syphilis subgroup. The factors that were associated with serofast response among subjects with late syphilis were not calculated because of the low sample size (9 serofast responses among 58 patients with late syphilis). All these models included the same covariates that were used in the models on treatment response.

All of the statistical tests were two-sided at the 5% level and were performed using SAS software (release 9.4; SAS Institute).

RESULTS

During 44 543 PYFU, 863 episodes of syphilis were recorded in 584 subjects, for an overall incidence rate of 19.4 (18.1–20.7)/1000 PYFU, with a higher incidence of syphilis observed in more recent years (before 2010: 12.9 [11.5–14.4]/1000 PYFU; 2010–2012: 24.6 [21.8–27.4]/1000 PYFU; after 2012: 27.0 [23.9–30.1]/1000 PYFU; $P < .0001$).

Thirty-four diagnoses in 20 subjects were excluded because of the lack of RPR titers obtained after treatment or the absence of a documented treatment after diagnosis, or if there was a diagnosis of neurosyphilis.

Thus, data were analyzed from 564 subjects for a total of 829 diagnoses of syphilis with assessable serological responses. In these subjects, the median number of concomitant RPR and TPHA titer determinations per year was 2.8 (2.3–3.5).

After syphilis treatment, the first RPR determination was performed 3.9 (2.7–5.4) months after therapy.

Among the 829 episodes, 686 (83%) were diagnosed as early syphilis, 97 (14%) were primary syphilis, 239 (35%) were secondary syphilis, and 350 (51%) were early latent syphilis. Among the 143 late syphilis, 136 (95%) cases of late latent and 7 (5%) cases of tertiary syphilis were observed. Characteristics of syphilis episodes according to disease stage are reported in Table 1, whereas Figure 1 shows the calendar-year distribution of syphilis diagnoses, according to syphilis type.

Two hundred thirty-seven (29%) episodes of syphilis were first infections, 592 (71%) were re-infections, and 507 (61%) episodes had an RPR titer ≤1:32 at diagnosis.

BPG was used for the treatment of 738 (89%) cases, and doxycycline was administered in 73 (9%) episodes. For early syphilis, a 1-dose BPG regimen was administered in 175 (26%) cases, a 3-dose BPG regimen was administered in 435 (63%) cases, and doxycycline was administered in 55 (8%) cases. Use of the 1-dose BPG regimen for the treatment of early syphilis increased over time; the 1-dose BPG regimen was used in 22% of diagnoses that were recorded before 2010, in 24% during the 2010–2012 period, and in 41% after 2012 ($P < .0001$ by the Cochran-Mantel-Haenszel test). During the 2015–2016 period, the 1-dose BPG regimen was used for treatment in 61% of early syphilis episodes.

After treatment, a serological response was observed in 732 (88%) syphilis episodes. The overall incidence rate of treatment response was 110.8 (102.7–118.8) per 1000 PMFU, with a significant increase in the more recent years (before 2010: 80.9 [70.5–91.2]/1000 PMFU; 2010–2012: 126.3 [110.9–141.7]/1000 PMFU; after 2012: 142.6 [124.6–160.6]/1000 PMFU; $P < .0001$).

The proportions of treatment response were significantly different between early and late syphilis (89% vs 83%, respectively; $P = .045$). With regards to the comparison of early syphilis that was treated with 1 vs 3 doses of BPG, no differences

Table 1. Characteristics of the 829 Syphilis Episodes (Early and Late) With an Assessable Treatment Response Observed During the Study Period (January 1, 2004–June 30, 2016) and Included in the Analysis; HIV Clinic of San Luigi Center, San Raffaele Hospital, Milan, Italy

	Early Syphilis				Late Syphilis					
Characteristics	Primary (n=97)	Secondary (n=239)	Early Latent (n=350)	All Early Syphilis (n=686)	Tertiary (n=7)	Late Latent (n=136)	All Late Syphilis (n=143)	All Syphilis (n=829)	Overall[a] *P*-value	Early vs Late *P*-value
First infection	30 (31%)	88 (37%)	75 (21%)	193 (28%)	1 (14%)	43 (32%)	44 (31%)	237 (29%)	0.001[d]	0.542[d]
RPR titer at diagnosis										
≤1:32	67 (69%)	100 (42%)	239 (68%)	406 (59%)	2 (29%)	99 (73%)	101 (71%)	507 (61%)	<.0001[d]	0.011[d]
TPHA titer at diagnosis										
>1:5120	70 (72%)	194 (81%)	259 (74%)	523 (76%)	7 (100%)	92 (68%)	99 (69%)	622 (75%)	0.020[d]	0.089[d]
Treatment regimens										
One-dose BPG	25 (26%)	48 (20%)	102 (29%)	175 (26%)	0	0	0	175 (21%)	<.0001[d]	<.0001[d]
Three-dose BPG	54 (56%)	165 (69%)	216 (62%)	435 (63%)	7 (100%)	119 (87%)	126 (88%)	561 (68%)		
Doxycycline	12 (12%)	18 (8%)	25 (7%)	55 (8%)	0	17 (13%)	17 (12%)	72 (9%)		
Other	6 (6%)	8 (3%)	7 (2%)	21 (3%)	0	0	0	21 (3%)		
ART use at the time of diagnosis	83 (87%)	193 (84%)	279 (84%)	555 (81%)	5 (71%)	112 (87%)	117 (82%)	672 (85%)	0.856[d]	0.599[d]
CD4+ (cells/μL)	632 (489–752)	557 (420–808)	630 (476–794)	613 (456–794)	378 (180–803)	580 (445–739)	577 (439–741)	609 (453–784)	0.181[b]	0.155[c]
CD4+/CD8+ ratio	0.59 (0.42–0.78)	0.57 (0.37–0.79)	0.59 (0.43–0.81)	0.58 (0.41–0.80)	0.44 (0.15–0.60)	0.56 (0.40–0.80)	0.56 (0.40–0.79)	0.58 (0.41–0.80)	0.235[b]	0.405[c]
HIV-RNA <50 copies/mL	65 (67%)	156 (66%)	262 (75%)	483 (70%)	5 (83%)	93 (69%)	98 (69%)	581 (70%)	0.155[d]	0.919[d]
Treatment response	90 (93%)	219 (92%)	304 (87%)	613 (89%)	5 (83%)	114 (84%)	119 (83%)	732 (88%)	0.045[d]	0.045[d]

Results are described by median (Q1–Q3) or frequency (%), as appropriate.

Abbreviations: Q1, quartile 1; Q3, quartile 3; RPR, rapid plasma regain; TPHA, *Treponema pallidum* hemagglutination assay; BPG, benzathine penicillin G; ART, antiretroviral therap.

[a]overall p-values refer to the simultaneous comparison of primary vs. secondary vs. early latent vs. tertiary vs. late latent syphilis.

[b]by Kruskal-Wallis test.

[c]by Mann-Whitney test.

[d]by chi-square or Fisher's exact test.

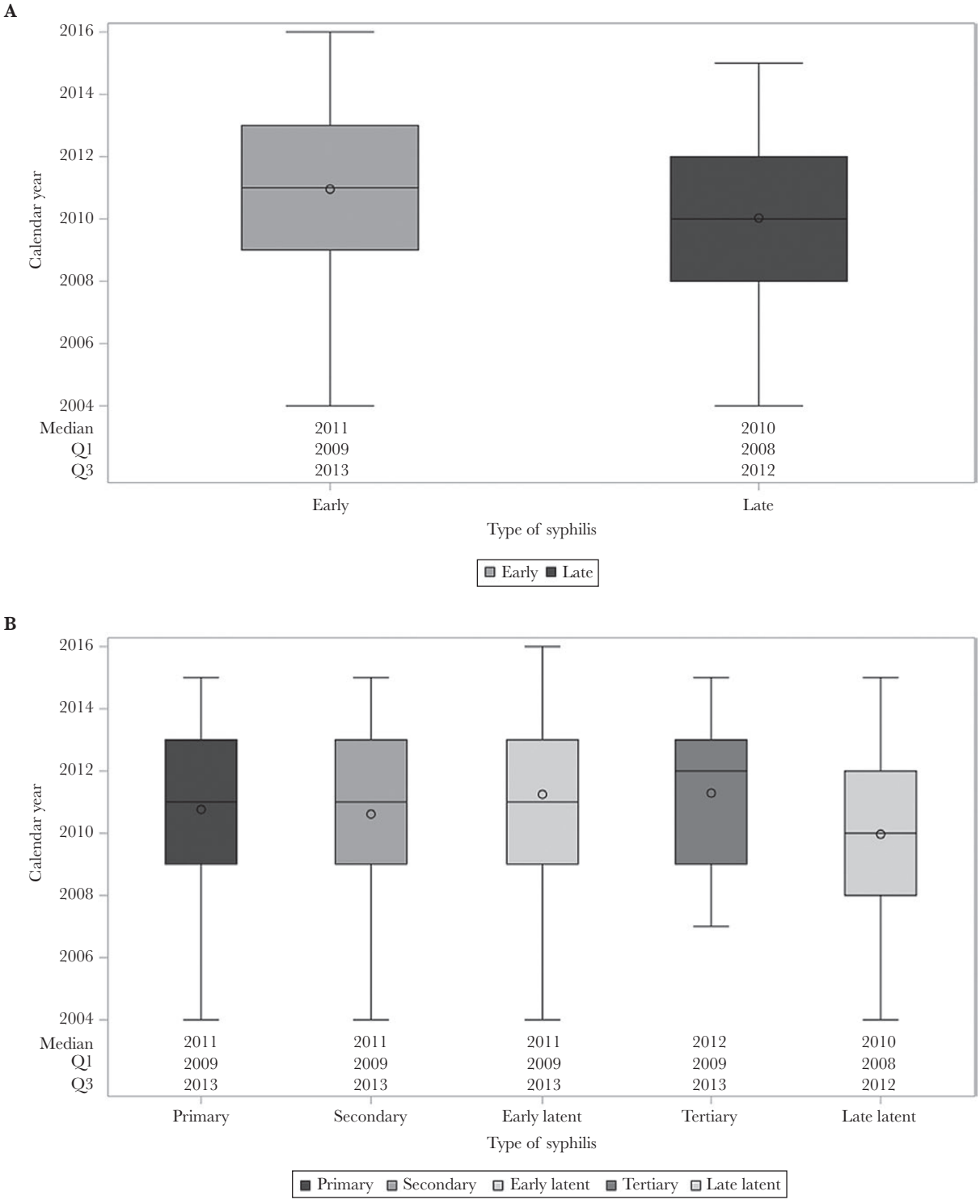

Figure 1. Calendar-year distribution of syphilis diagnoses according to syphilis type.

were observed in the frequency of treatment response for any type of syphilis. In primary syphilis, treatment response was observed in 23/25 (95%) episodes that were treated with 1 dose vs 50/54 (93%) episodes that were treated with a 3-dose BPG regimen ($P = .997$); in secondary syphilis, a treatment response was observed in 46/48 (97%) episodes that were treated with 1 dose, compared with 151/165 (91%) episodes that were treated with the 3-dose BPG regimen ($P = .471$); in early latent syphilis: treatment response in 90/102 (88%) episodes that were treated with 1 dose, vs 193/216 (89%) episodes that were treated with a 3-dose BPG regimen ($P = .841$); in all early syphilis episodes: 159/175 (91%) episodes that were treated with 1 dose,

vs 394/435 (91%) that were treated with a 3-dose BPG regimen ($P = .999$).

The crude incidence rate of treatment response in early syphilis episodes was 120.8 (111.3–130.4) per 1000 PMFU as compared with 77.4 (63.5–91.3)/1000 PMFU among late syphilis episodes ($P < .0001$). A significant increase in the crude incidence rate of treatment response was found, over time, for early syphilis episodes (before 2010: 88.5 [75.7–101.3]/1000 PMFU; 2010–2012: 135.8 [117.6–153.9]/1000 PMFU; after 2012: 151.3 [131.1–171.5]/1000 PMFU; $P < .0001$), whereas a marginally significant increase was observed for late syphilis (before 2010: 61.1 [44.0–78.2]/1000 PMFU; 2010–2012: 94.2 [66.4–122.0]/1000 PMFU; after 2012: 96.9 [59.6–134.1]/1000 PMFU; $P = .056$).

Subjects' characteristics at their first syphilis episode during the study period are detailed in Table 2 according to syphilis stage.

The median age of the 564 subjects included in the study was 41.6 (36.2–47.6) years, 561 (99.6%) were males, 499 (88%) were MSM, they were HIV-infected for 7.6 (3.4–13.1) years, 82 (14%) were hepatitis C virus– or hepatitis B virus–coinfected, 104 (19%) were antiretroviral therapy (ART)–naïve, and 364 (65%) had HIV-RNA <50 copies/mL, a CD4+ level of 584 (438–754) cells/mm^3, and a nadir CD4+ of 310 (202–411) cells/mm^3.

The CD4+ cell count at syphilis diagnosis increased over time: a median of 551 (411–749) cells/mm^3 for syphilis episodes that were diagnosed before 2008, 568 (415–766) cells/mm^3 for episodes that were diagnosed between 2008 and 2011, and 644 (506–823) cells/mm^3 for syphilis episodes that were diagnosed after 2011 ($P = .001$ by Kruskal-Wallis test).

Three hundred seventy-four (66%) subjects had a single episode of syphilis diagnosis during their follow-up, 132 (23%) had 2 episodes, 43 (8%) had 3, 13 (2%) had 4, and 2 patients had 5 syphilis diagnoses during their follow-up.

Results of multivariate analyses on factors that were associated with treatment response for early and late syphilis are detailed in Table 3. For episodes of early syphilis, a response to treatment was independently associated with a higher nadir CD4+ cell count (adjusted hazard ratio [AHR], 1.06; 95% CI = 1.01–1.12; $P = .029$), an RPR titer >1:32 (AHR, 1.26; 95% CI, 1.06–1.51; $P = .009$), and a diagnosis that had been made in more recent calendar years (AHR, 1.36; 95% CI, 1.17–1.59; $P < .001$). Additionally, subjects with early latent syphilis had a similar risk of treatment response compared with those with primary syphilis but a lower risk when compared with those with secondary syphilis (AHR, 0.78; 95% CI, 0.64–0.93; $P = .008$).

Treatment response for late syphilis was higher in first infections (AHR, 1.80; 95% CI, 1.07–3.03; $P = .027$) and in more recent calendar years (AHR, 1.62; 95% CI, 1.14–2.31; $P = .007$). Additionally, treatment response for late syphilis was associated with a higher RPR titer at diagnosis (AHR, 2.04; 95% CI, 1.29–3.23; $P = .002$). Interestingly, the risk of treatment response was not found to be associated with any of the treatment regimens in all the estimated models (overall, early, and late syphilis diagnoses).

Finally, serofast status was diagnosed in 82/335 (25%) of the patients with a single diagnosis of syphilis during the study period (early syphilis, 73/277 [26%]; late syphilis, 9/58 [16%]; $P = .094$). Predictors of serofast status are reported in Table 4.

DISCUSSION

In our cohort of PLWH, 863 episodes of syphilis in 584 subjects were observed between January 2004 and June 2016. More recent years of the study reported a higher incidence of syphilis, similar to other studies [8, 20].

Almost all of the subjects were males, and approximately 90% were MSM. The high prevalence of syphilis in MSM living with HIV has been described [21] and is associated with a reduction in the awareness of sexually transmitted infections and high-risk sexual behaviors in this population [22]. With regards to these issues, in this study, 71% of the infections occurred in individuals with a history of syphilis (re-infections) and 34% of the subjects had more than 1 syphilis diagnosis during the follow-up period, indicating the recurrence of high-risk sexual behaviors in our patients.

Overall, subjects included in our study showed good immune-virological status. At the time of diagnosis of syphilis, the median CD4+ cell count was approximately 600 cells/mm^3, 81% had received some form of antiretroviral treatment, and 80% of treated subjects had a viremia of <50 copies/mL.

Despite the fact that the 3-dose BPG regimen remains the more frequently prescribed treatment, including among early syphilis diagnoses (63% of the total episodes), the 1-dose BPG treatment has recently become the preferred regimen for treatment of early syphilis, in accordance with guidelines and recommendations [12, 13].

Treatment failure was observed in 12% of episodes that were included in the study, which is a proportion that is similar to what has been observed in other studies regarding uninfected individuals [11] and PLWH in the post-ART era [9, 10].

Additionally, differences were observed in the proportions of subjects who responded to treatment when diagnosed with early and late syphilis. This finding is similar to what has been reported by other studies [7, 23] and confirms that subjects with late syphilis diagnoses are at a higher risk for serologically defined treatment failure, although, in our study, different time points were used to evaluate for a response in early and late syphilis, as suggested by the Centers for Disease Control and Prevention [12]. These time points, although established, are arbitrary. Therefore, it is possible that if the period of testing for treatment response was extended, the proportion of responders would increase.

Table 2. Characteristics at First Syphilis Episode During the Study Period (January 1, 2004–June 30, 2016) of the 564 People Living With HIV who Were Included in the Study; HIV Clinic of San Luigi Center, San Raffaele Hospital, Milan, Italy

		Early Syphilis				Late Syphilis				
Characteristics	All Syphilis (n = 564)	Primary (n = 72)	Secondary (n = 175)	Early Latent (n = 206)	All Early Syphilis (n = 453)	Tertiary (n = 3)	Late Latent (n = 108)	All Late Syphilis (n = 111)	Overall[a] *P* Value	Early vs Late *P* Value
Age, y	41.6 (36.2–47.6)	42.7 (37.2–49.2)	40.1 (34.9–47.2)	41.6 (35.7–47.0)	41.3 (35.5–47.3)	42.9 (39.6–46.5)	43.4 (38.3–49.7)	43.4 (38.4–49.6)	.028[b]	.008[c]
Ethnicity									.391[d]	.397[d]
White	497 (88)	61 (85)	153 (87)	188 (91)	402 (89)	3 (100)	92 (85)	95 (86)		
Other	67 (12)	11 (15)	22 (13)	18 (9)	41 (9)	0	16 (15)	16 (14)		
Male gender	562 (99.6)	72 (100)	173 (99)	206 (100)	451 (99.5)	3 (100)	108 (100)	108 (100)	.347[d]	.999[d]
HIV risk factor									.064[d]	.109[d]
MSM	499 (88)	64 (89)	150 (86)	193 (94)	407 (90)	2 (67)	90 (82)	92 (83)		
PWID	7 (1)	0	4 (2)	1 (1)	5 (1)	0	2 (2)	2 (2)		
Heterosexual	26 (5)	2 (3)	8 (5)	7 (3)	17 (4)	1 (33)	8 (7)	9 (8)		
Other/unknown	32 (6)	6 (8)	13 (7)	5 (2)	24 (5)	0	8 (9)	8 (7)		
Years since HIV diagnosis	7.6 (3.4–13.1)	8.5 (3.2–14.3)	6.7 (3.0–12.4)	6.7 (2.8–12.1)	6.8 (3.0–12.4)	4.1 (0.4–23.0)	10.9 (5.7–14.5)	10.4 (5.6–14.5)	.012[b]	.001[c]
Years of ART	4.9 (1.0–10.6)	5.3 (0.8–12.0)	3.8 (0.9–9.7)	4.2 (0.7–10.2)	4.0 (0.8–10.2)	4.0 (0–17.6)	7.1 (2.9–11.4)	7.0 (2.8–11.4)	.097[b]	.011[c]
Naïve	104 (19)	12 (17)	34 (19)	41 (20)	87 (19)	1 (33)	16 (15)	17 (15)	.746[d]	.413[d]
Nadir CD4+, cells/μL	310 (202–411)	330 (225–407)	319 (198–425)	307 (204–433)	315 (207–425)	31 (19–164)	299 (193–356)	294 (189–356)	.023[b]	.012[c]
Previous AIDS diagnosis	62 (11)	5 (7)	18 (10)	23 (11)	46 (10)	1 (33)	15 (14)	16 (14)	.440[d]	.235[d]
HCV antibodies									.056[d]	.243[d]
Negative	492 (87)	63 (87)	158 (90)	175 (85)	396 (87)	2 (67)	95 (88)	97 (87)		
Positive	53 (9)	4 (6)	14 (8)	27 (13)	45 (10)	1 (33)	7 (6)	8 (7)		
Unknown	18 (3)	5 (7)	3 (2)	4 (2)	12 (3)	0	6 (6)	6 (5)		
HBsAg									.167[d]	.225[d]
Negative	404 (72)	51 (71)	139 (80)	140 (68)	330 (73)	3 (100)	72 (67)	75 (68)		
Positive	29 (5)	4 (6)	6 (3)	15 (7)	25 (6)	0	4 (4)	4 (4)		
Unknown	130 (23)	17 (22)	30 (17)	51 (25)	98 (22)	0	32 (29)	32 (29)		
CD4+, cells/μL	584 (438–754)	614 (472–727)	550 (413–784)	610 (445–758)	597 (442–764)	102 (24–180)	564 (441–695)	558 (427–693)	.112[b]	.254[c]
CD4+/CD8+ ratio	0.55 (0.38–0.78)	0.58 (0.38–0.76)	0.55 (0.34–0.77)	0.57 (0.42–0.80)	0.56 (0.38–0.78)	0.11 (0.07–0.15)	0.52 (0.39–0.76)	0.50 (0.38–0.76)	.053	.309[c]
HIV-RNA <50 copies/mL	364 (65)	47 (65)	100 (58)	148 (72)	295 (65)	1 (33)	69 (65)	70 (63)	.067[d]	.999[d]

Results are described by median (Q1–Q3) or frequency (%), as appropriate.

Abbreviations: ART, antiretroviral therapy; HBsAg, hepatitis B surface antigen; HCV, hepatitis C virus; MSM, men who have sex with men; PWID, people who inject drugs.

[a]Overall *P* values refer to the simultaneous comparison of primary vs secondary vs early latent vs tertiary vs late latent syphilis.

[b]By Kruskal-Wallis test.

[c]By Mann-Whitney test.

[d]By chi-square or Fisher exact test.

According to the different clinical characteristics and criteria for serological response, we decided to separately analyze early and late syphilis to evaluate predictors of treatment response.

Treatment response in episodes of early syphilis was independently associated with a higher nadir CD4+ cell count, an RPR titer >1:32, and a diagnosis made in more recent calendar years. Additionally, episodes of early latent syphilis had a lower risk of treatment response when compared with episodes of secondary syphilis.

Immunosuppression has been associated with treatment failure in syphilis, as reported in previous studies on PLWH [10, 22] and subjects with an impaired immune system have been reported to respond more slowly to syphilis treatment, compared with those with a good immunological status [10, 24]. In this study, higher RPR titers were associated with treatment response both in early and late syphilis. This finding has been described in HIV-negative and HIV-infected individuals [11]. Higher RPR titers may reflect a more potent immunological response, which is associated with a more efficacious clearance of *Treponema pallidum* [25]. Additionally, subjects with lower RPR titers may have a longer duration of infection, with lower metabolic activity of the microorganism and a delayed response to treatment [26]. Finally, a 4-fold decline in the RPR titer (the definition of treatment response) is more easily obtained in

Table 3. Multivariate Cox Proportion Hazards Models for Recurrent Events: Factors Associated With Treatment Response

	Overall		Early Syphilis		Late Syphilis	
Characteristic	AHR (95% CI)	*P* Value	AHR (95% CI)	*P* Value	AHR (95% CI)	*P* Value
Age, per 5 y longer	0.97 (0.92–1.03)	.331	0.97 (0.91–1.03)	.325	1.00 (0.88–1.13)	.945
Years of antiretroviral treatment, per 5 y longer	0.98 (0.90–1.05)	.539	1.00 (0.92–1.09)	.950	0.81 (0.64–1.02)	.071
Nadir CD4+, per 100 cells/μL higher	1.04 (1.0–1.09)	.078	1.06 (1.01–1.12)	.029	1.04 (0.91–1.19)	.611
Type of syphilis						
Early vs late	1.25 (0.98–1.58)	.070			Not included	-
Primary vs early latent	Not included	-	1.23 (0.93–1.64)	.152	Not included	-
Secondary vs early latent	Not included	-	1.29 (1.07–1.56)	.008	Not included	-
Primary vs secondary	Not included	-	0.95 (0.71–1.28)	.749	Not included	-
Calendar year of diagnosis, per 1 y longer	1.41 (1.22–1.62)	<.0001	1.36 (1.17–1.59)	<.0001	1.62 (1.14–2.31)	.007
First infection, yes vs no	1.28 (1.04–1.57)	.021	1.15 (0.92–1.45)	.224	1.80 (1.07–3.03)	.027
RPR titer at diagnosis, >1:32 vs ≤1:32	1.41 (1.19–1.64)	<.0001	1.27 (1.06–1.52)	.009	2.04 (1.30–3.23)	.002
CD4+/CD8+ ratio, per 20% higher	1.02 (0.97–1.06)	.456	1.01 (0.96–1.07)	.687	1.01 (0.91–1.13)	.833
HIV-RNA, per $\log_{10}$ copies/mL higher	1.00 (0.92–1.09)	.944	1.03 (0.94–1.12)	.547	0.80 (0.63–1.01)	.061
Treatment regimens						
Doxycycline vs BPG	1.15 (0.80–1.66)	.445	Not included		Not included	-
1-dose BPG vs 3-dose BPG	Not included		0.97 (0.73–1.60)	.811	Not included	-
Doxycycline vs 3-dose BPG	Not included		1.08 (0.89–1.77)	.719	1.48 (0.80–2.73)	.216
Doxycycline vs 1-dose BPG	Not included	-	1.11 (0.72–1.69)	.648	Not included	-

Abbreviations: AHR, adjusted hazard ratio; BPG, benzathine penicillin G; CI, confidence interval; RPR, rapid plasma reagin.

patients with higher titers at the time of diagnosis of syphilis [10].

The calendar year of syphilis diagnosis was another predictor of treatment response both in early and late syphilis. Episodes that were diagnosed in less recent years had a lower risk of response, compared with the more recent diagnoses. This finding is probably related to the lower CD4+ cell count observed in subjects with a diagnosis in less recent years, and the possible consequent delay in treatment response.

In late syphilis, re-infections were associated with a higher risk of treatment failure than in patients at their first infection. This result is similar to that described in PLWH by Jinno et al. [10], although it is unclear why this association was not observed for early syphilis cases in this study.

Table 4. Multivariate Cox Proportion Hazards Models: Factors Associated With Serofast Response

	Overall		Early Syphilis	
Characteristic	AHR (95% CI)	*P* Value	AHR (95% CI)	*P* Value
Age, per 5 y longer	0.98 (0.81–1.19)	.849	0.93 (0.75–1.17)	.537
Years of antiretroviral treatment, per 5 y longer	1.38 (1.06–1.79)	.020	1.30 (0.95–1.77)	.098
Nadir CD4+, per 100 cells/μL higher	0.80 (0.66–0.98)	.019	0.75 (0.59–0.96)	.020
Type of syphilis				
Early vs late	3.55 (1.50–8.40)	.040		
Primary vs early latent	Not included	-	1.91 (0.96–3.80)	.066
Secondary vs early latent	Not included	-	0.81 (0.42–1.56)	.532
Primary vs secondary	Not included	-	2.35 (1.04–5.31)	.040
Calendar year of diagnosis, per 1 y longer	0.67 (0.59–0.76)	<.0001	0.59 (0.50–0.70)	<.0001
First infection, yes vs no	0.73 (0.42–1.27)	.268	0.71 (0.38–1.32)	.281
RPR titer at diagnosis, >1:32 vs ≤1:32	1.01 (0.61- 1.65)	.982	1.35 (0.80–2.28)	.266
CD4+/CD8+ ratio, per 20% higher	1.01 (0.87–1.17)	.924	1.15 (0.92–1.43)	.230
HIV-RNA, per $\log_{10}$ copies/mL higher	1.13 (0.89–1.43)	.325	1.10 (0.86–1.42)	.448
Treatment regimens				
Doxycycline vs BPG	1.32 (0.38–4.59)	.664	Not included	
1-dose BPG vs 3-dose BPG	Not included	-	2.40 (1.25–4.62)	.009
Doxycycline vs 3-dose BPG	Not included	-	1.59 (0.48–5.22)	.446
Doxycycline vs 1-dose BPG	Not included	-	0.66 (0.20–2.25)	.510

The multivariate model on factors associated with serofast response among subjects with late syphilis was not calculated because of the low sample size (9 serofast responses among 58 late syphilis patients).

Abbreviations: AHR, adjusted hazard ratio; BPG, benzathine penicillin G; CI, confidence interval; RPR, rapid plasma reagin.

The risk of treatment response was not found to be associated with the treatment regimens in early and late syphilis. In other studies [9, 10, 14], which are in agreement with these results, a 1-dose BPG regimen for PLWH showed an efficacy that was similar to that of the 3-dose BPG regimen; it should be considered the firstline regimen in treatment of episodes of early syphilis among PLWH, in accordance with current international guidelines.

In our study, doxycycline demonstrated a similar efficacy as BPG, also in patients with late latent syphilis, although this result should be interpreted with caution because of the small number of patients that were treated with doxycycline at this disease stage.

The optimal management of patients who failed to demonstrate a treatment response is unclear. Because treatment failure might be the result of central nervous system involvement, cerebrospinal fluid (CSF) examination can be considered [12]. If the CSF examination is negative, retreatment should be administered, although serologic titers might not decline despite repeated courses of therapy [27].

Of interest is the finding that 25% of the patients did not serovert their nontreponemal test titers during the follow-up period (serofast status), despite an appropriate decline of their RPR titers. These subjects had a serologically defined cure, although the persistence of a positive RPR titer may represent a concern for the clinician because of the paucity of data regarding the long-term management and outcomes of these patients.

Data regarding serofast status, especially among PLWH, are lacking. In this study, a lower nadir CD4+, a longer duration of antiretroviral treatment, a diagnosis of early syphilis as compared with late syphilis, and a less recent calendar year of syphilis infection were associated with serofast status. Interestingly, the restricted analysis on early syphilis episodes showed that a serofast status was related to a diagnosis of primary syphilis, as compared with a secondary syphilis diagnosis; it was also related to the use of a 1-dose BPG treatment, as compared with a 3-dose BPG schedule.

Our study has several limitations. First, we cannot exclude the possibility of selection bias, as 4% of the total syphilis episodes were excluded because of incomplete information or loss to follow-up. Second, given the retrospective nature of the study, the included subjects had different follow-up durations and were differently distributed over time. Third, in our definition of re-infection, syphilis episodes that occurred in PLWH who seroconverted their RPR titers (if previously negative) were included. However, HIV infection has been associated with false-positive, low RPR titers [28], and thus syphilis re-infections may have been overdiagnosed, especially among those who seroconverted with a low RPR titer (≤1:2) and were diagnosed with latent syphilis (early and late). Therefore, we performed additional sensitivity analyses after the exclusion of these episodes (n = 30) by recalculating the multivariable analyses of Table 3 after excluding these 30 cases. The results (Supplementary Table 1) were similar to those already reported herein. Finally, it was not ensured that the subjects did not utilize other antibiotics that were active against *Treponema pallidum* in between their follow-up visits. Nonetheless, this is one of the largest studies that has investigated predictors of treatment response in PLWH diagnosed with syphilis, and it had a long follow-up period.

In conclusion, a higher incidence of syphilis was observed in the more recent years of this study, with an overall serological response of 88%. For episodes of early syphilis, factors that were associated with the serological treatment response were a higher nadir CD4+ cell count, an RPR titer >1:32, a diagnosis that was recorded in more recent years, and a secondary syphilis diagnosis (when compared with early latent syphilis). In subjects that were diagnosed with late syphilis, a serological treatment response was associated with first infection, a diagnosis in more recent years, and higher RPR titers.

Acknowledgments

We would like to thank JournalEdit (https://www.journaledit.com/) for editing and reviewing this manuscript for English language.

Financial support. This study did not receive external funding.

Potential conflicts of interest. The authors have no conflicts of interest to declare. All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

References

1. Peterman TA, Su J, Bernstein KT, Weinstock H. Syphilis in the United States: on the rise? Expert Rev Anti Infect Ther **2015**; 13:161–8.
2. Peterman TA, Furness BW. The resurgence of syphilis among men who have sex with men. Curr Opin Infect Dis **2007**; 20:54–9.
3. de Voux A, Kidd S, Grey JA, et al. State-specific rates of primary and secondary syphilis among men who have sex with men—United States, 2015. MMWR Morb Mortal Wkly Rep **2017**; 66:349–354
4. Ghanem KG, Erbelding EJ, Wiener ZS, Rompalo AM. Serological response to syphilis treatment in HIV-positive and HIV-negative patients attending sexually transmitted diseases clinics. Sex Transm Infect **2007**; 83:97–101.
5. Rolfs RT, Joesoef MR, Hendershot EF, et al. A randomized trial of enhanced therapy for early syphilis in patients with and without human immunodeficiency virus infection. The Syphilis and HIV Study Group. N Engl J Med **1997**; 337:307–14.
6. Knaute DF, Graf N, Lautenschlager S, et al. Serological response to treatment of syphilis according to disease stage and HIV status. Clin Infect Dis **2012**; 55:1615–22.
7. Dionne-Odom J, Karita E, Kilembe W, et al. Syphilis treatment response among HIV-discordant couples in Zambia and Rwanda. Clin Infect Dis **2013**; 56:1829–37.
8. González-López JJ, Guerrero ML, Luján R, et al. Factors determining serologic response to treatment in patients with syphilis. Clin Infect Dis **2009**; 49:1505–11.
9. Ganesan A, Mesner O, Okulicz JF, et al; Infectious Disease Clinical Research Program HIV/STI Working Group. A single dose of benzathine penicillin G is as effective as multiple doses of benzathine penicillin G for the treatment of HIV-infected persons with early syphilis. Clin Infect Dis **2015**; 60:653–60.
10. Jinno S, Anker B, Kaur P, et al. Predictors of serological failure after treatment in HIV-infected patients with early syphilis in the emerging era of universal antiretroviral therapy use. BMC Infect Dis **2013**; 13:605.
11. Seña AC, Zhang XH, Li T, et al. A systematic review of syphilis serological treatment outcomes in HIV-infected and HIV-uninfected persons: rethinking the significance of serological non-responsiveness and the serofast state after therapy. BMC Infect Dis **2015**; 15:479.
12. Workowski KA, Bolan GA; Centers for Disease Control and Prevention. Sexually transmitted diseases treatment guidelines, 2015. MMWR Recomm Rep **2015**; 64:1–137.

13. Janier M, Hegyi V, Dupin N, et al. 2014 European guideline on the management of syphilis. J Eur Acad Dermatol Venereol **2014**; 28:1581–93.
14. Andrade R, Rodriguez-Barradas MC, Yasukawa K, et al. Single dose versus 3 doses of intramuscular benzathine penicillin for early syphilis in HIV: a randomized clinical trial. Clin Infect Dis **2017**; 64:759–64.
15. Tsai JC, Lin YH, Lu PL, et al. Comparison of serological response to doxycycline versus benzathine penicillin G in the treatment of early syphilis in HIV-infected patients: a multi-center observational study. PLoS One **2014**; 9:e109813.
16. Antonio MB, Cuba GT, Vasconcelos RP, et al. Natural experiment of syphilis treatment with doxycycline or benzathine penicillin in HIV-infected patients. AIDS. **In press**.
17. Amorim LD, Cai J. Modelling recurrent events: a tutorial for analysis in epidemiology. Int J Epidemiol **2015**; 44:324–33.
18. Andersen PK, Gill R. Cox's regression model for counting processes: a large sample study. Ann Stat **1982**; 10:1100–1120.
19. Cook RJ, Richard J, Lawless JF. The Statistical Analysis of Recurrent Events. New York: Springer-Verlag; **2007**.
20. Novak RM, Ghanem A, Hart R, et al. Risk factors and incidence of syphilis in HIV-infected persons, the HIV Outpatient Study, 1999–2015. Clin Infect Dis. **2018**; 67:1750–9.
21. Refugio ON, Klausner JD. Syphilis incidence in men who have sex with men with human immunodeficiency virus comorbidity and the importance of integrating sexually transmitted infection prevention into HIV care. Expert Rev Anti Infect Ther **2018**; 16:321–31.
22. Pufall EL, Kall M, Shahmanesh M, et al; Positive Voices study group. Sexualized drug use ('chemsex') and high-risk sexual behaviours in HIV-positive men who have sex with men. HIV Med **2018**; 19:261–70.
23. Tong ML, Lin LR, Liu GL, et al. Factors associated with serological cure and the serofast state of HIV-negative patients with primary, secondary, latent, and tertiary syphilis. PLoS One **2013**; 8:e70102.
24. Ghanem KG, Moore RD, Rompalo AM, et al. Antiretroviral therapy is associated with reduced serologic failure rates for syphilis among HIV-infected patients. Clin Infect Dis **2008**; 47:258–65.
25. Seña AC, Wolff M, Martin DH, et al. Predictors of serological cure and serofast state after treatment in HIV-negative persons with early syphilis. Clin Infect Dis **2011**; 53:1092–9.
26. Cox DL, Akins DR, Porcella SF, et al. Treponema pallidum in gel microdroplets: a novel strategy for investigation of treponemal molecular architecture. Mol Microbiol **1995**; 15:1151–64.
27. Seña AC, Wolff M, Behets F, et al. Response to therapy following retreatment of serofast early syphilis patients with benzathine penicillin. Clin Infect Dis **2013**; 56:420–2.
28. Rompalo AM, Cannon RO, Quinn TC, Hook EW 3rd. Association of biologic false-positive reactions for syphilis with human immunodeficiency virus infection. J Infect Dis **1992**; 165:1124–6.