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Note

Evaluation of new algorithm using TPLA as an initial syphilis screening test☆

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ABSTRACT

We retrospectively compared the performance of an existing syphilis diagnostic algorithm with a new algorithm that analyzes the results of *Treponema pallidum* latex agglutination (TPLA) tests. Of the 100 clinical blood samples, 51 were classified as positive through both Mediace TPLA and ESPLINE TP; 2/51 were classified as negative by Architect Syphilis TP, whereas 1/51 was negative as per LUMIPULSE Presto TP. The false positive rate when the results of Mediace TPLA and ESPLINE TP were combined was 1.96% versus 0% for both Architect Syphilis TP and LUMIPULSE Presto TP. The sensitivity of Mediace TPLA (98%) was comparable to that of Architect Syphilis TP (98%) but lower than that of LUMIPULSE Presto TP (100%). The specificity of Mediace TPLA was 98.0% versus 100% for Architect Syphilis TP, and versus 100% for LUMIPULSE Presto TP. We conclude that the performance of Mediace TPLA in combination with a reverse algorithm is nearly equal to that of enzyme immunoassay (EIA) or chemiluminescence immunoassay (CIA). Because TPLA is low cost, highly sensitive method for IgM detection, and is easy to operate, we have recommended its adoption for initial syphilis screening tests.

Syphilis remains a worldwide public health concern, as the global incidence of syphilis infection continues to rise [1]. The World Health Organization (WHO) estimated that 6 million new cases of syphilis occur globally each year among adults aged 15–49 years [2]. Although clinical profiling of symptoms is important, serologic tests are still considered the mainstay of syphilis diagnosis [3]. Syphilis serological tests are divided into nontreponemal and treponemal tests, and neither is sufficient alone for diagnosis. To date, numerous assays have been used in clinical settings [4,5]. Two common approaches to the diagnosis of syphilis employ serological tests: the traditional algorithm and the reverse algorithm [6]. In the traditional algorithm, syphilis screening is firstly performed using a nontreponemal test, such as rapid plasma reagin (RPR), and then screen-reactive samples are tested using a treponemal assay. On the other hand, in the reverse algorithm, syphilis screening is first performed using a treponemal test, and then screen-reactive samples are tested using a nontreponemal assay. Historically, the traditional algorithm has been popular; however, recent advancements in instrumentation have enabled more automated and objective approaches to the treponemal test. Currently, however, there are not many laboratories using the reverse algorithm [7]. The Centers for Disease Control and Prevention (CDC) recognizes both approaches, but the reverse algorithm for initial syphilis screening is limited to use with either the enzyme or chemiluminescence immunoassays (EIA and CIA, respectively) [8]. A recently developed TPLA method that involves coating antigenic components of *Treponema pallidum* to latex particles and automatic analysis has now been adopted in several centers [9]. Here, we evaluated a new approach, which uses a modified reverse algorithm with TPLA during the initial syphilis screening, and compared it to the reverse algorithm used with EIA or CIA.

From a total of 3329 cases that were subjected to syphilis serological tests at Sapporo Medical University Hospital between January and June 2017, we analyzed 100 clinical blood samples that were deemed positive by the Mediace TPLA (Sekisui Medical Co,

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Tokyo, Japan) or Mediace RPR (Sekisui Medical Co, Tokyo, Japan). If the sample was reacted by Mediace TPLA, another treponemal test for syphilis was recommended for confirmation of the reactivity; only in these cases, we therefore used ESPLINE TP (Fujirebio INC, Tokyo, Japan) as the confirmatory test. We evaluated four kinds of syphilis serological tests including Architect Syphilis TP (Abbott Diagnostics, Chicago, IL), Lumipulse presto TP (Fujirebio INC, Tokyo, Japan), SERODIA-TPPA (Fujirebio INC, Tokyo, Japan), and the fluorescent treponemal antibody absorption (FTA-ABS) test-SG-KIT (Japan BCG Laboratory, Tokyo, Japan). Architect Syphilis TP and Lumipulse presto TP were CIA based methods. FTA-ABS was used only in cases where the results were discordant between Mediace TPLA, Architect Syphilis TP, Lumipulse presto TP. Mediace TPLA and Mediace RPR were tested using the 7180 automated clinical analyzer (HITACHI High-technologies, Tokyo, Japan). Architect Syphilis TP was used with ARCHITECT i2000SR (Abbott Diagnostics, Chicago, IL), and Lumipulse presto TP was used with Lumipulse L2400 (Fujirebio INC, Tokyo, Japan). ESPLINE TP, SERODIA-TPPA and the FTA-ABS test-SG-KIT were used manually according to the manufacturers' instructions. Samples were stored at −40 °C and thawed immediately prior to analysis. The false-negative and the false-positive cases were determined based on the results from FTA-ABS that is considered as the gold standard for treponemal test [10]. False-negatives were defined as those positive in FTA-ABS but negative in other tests, and false-positives were defined as those negative in FTA-ABS but positive in other tests. The methods used in this retrospective study were approved by the Institutional Review Board (http://web.sapmed.ac.jp/byoin/chiken/irb.html) of Sapporo Medical University Hospital (No. 292-59).

In the analysis, 51/100 samples were deemed positive by both Mediace TPLA and ESPLINE TP. On the other hand, 50/100 samples were positive following the Architect Syphilis TP test, and 51/100 were positive following the Lumipulse presto TP test (Table 1a, b). The results from three samples differed depending on whether Mediace TPLA, Architect Syphilis TP or Lumipulse presto TP was used. Those samples were further tested using another syphilis serological test (Table 2). The false positive rate of the combination with Mediace TPLA and ESPLINE TP was 1.96% (1 of 51), whereas that of Architect Syphilis TP and Lumipulse presto TP was 0%. The false-negative rate of Mediace TPLA was 2.04% (1 of 49), whereas that of Architect Syphilis TP was 2.00% (1 of 50), that of Lumipulse presto TP was 0%.

We evaluated the different algorithms with respect to diagnostic accuracy. We confirmed that results guided by the algorithm and clinical diagnosis were consistent. According to the reverse algorithm using Architect Syphilis TP or Lumipulse presto, 55 cases were classified as “syphilis unlikely”, 31 were identified as “current syphilis likely” and 14 were “syphilis likely” (Fig. 1a, b). On the other hand, according to the modified reverse algorithm, the 56 cases were classified as “syphilis unlikely”, 30 cases were identified as “current syphilis likely” and 14 were classified as “syphilis likely” (Fig. 1c). When Architect Syphilis TP or Lumipulse presto TP were used as alternatives to the SERODIA-TPPA treponemal test, the number of cases classified as “syphilis likely” increased to 19 and those classed as “unlikely” decreased to 51 (Fig. 1d).

Although several recent studies have attempted to rank diagnostic algorithms, no definitive answer has been found to date [11–14]. Nontreponemal tests can detect active infections, but have high false-positive rates. On the other hand, treponemal tests provide a more specific detection method, but also detect infections that have already been treated. The results from three samples differed according to whether Mediace TPLA, Architect Syphilis TP or Lumipulse presto TP was used. As shown in Table 2, both Case 1, in which no reaction was observed with Mediace TPLA and Case 2, where no reaction was seen with Architect Syphilis TP, were deemed positive by FTA-ABS. Therefore, Cases 1 and 2 were considered false-negative results with by Mediace TPLA and Architect Syphilis TP, respectively. On the other hand, Case 3, which was positive following the Mediace TPLA test, was scored negative by FTA-ABS. Therefore, we considered that Mediace TPLA produced a false-positive result in this instance. As shown in Fig. 1a–c, the one sample that classified as “current syphilis likely” by reverse algorithm but as “syphilis unlikely” was considered false-negative result with by Mediace TPLA. On the other hand, shown in Fig. 1c and d, 5 samples were classified as “syphilis unlikely” by SERODIA-TPPA but as “syphilis likely” by Lumipulse presto TP. All the 5 samples were reacted by FTA-ABS and diagnosed as past syphilis by their physician. The performance of Mediace TPLA is almost equivalent to that of Lumipulse presto or Architect Syphilis TP. Also, the cost per test of Mediace TPLA is inexpensive compared with the other tests even though the necessity of confirmation test. The cost of TPLA per test is about a half of that of EIA or CIA. The confirmation test by ESPLINE TP need in the samples reacted by Mediace TPLA, not all samples. Furthermore, another study reported that, owing to its ability to detect IgM, Mediace TPLA could detect infection earlier than EIA or CIA [15]. Together with its ease of use, these qualities lead us to recommend Mediace TPLA for use in initial screening tests. In this study, we demonstrated that a new algorithm using TPLA as an initial screening test was useful for syphilis diagnosis. The sensitivity of SERODIA-TPPA is worse than that of EIA or CIA. The performance of FTA-ABS might be almost equal to that of EIA or CIA in our study though FTA-ABS was used only when

Table 1a
The comparison of reactivity of Mediace TPLA and Architect Syphilis TP or Lumipulse presto TP.

		Mediace TPLA + ESPLINE TP	
		(+)	(−)
Architect Syphilis TP	(+)	49	1
	(−)	2	48

Table 1b

		Mediace TPLA + ESPLINE TP	
		(+)	(−)
Lumipulse presto TP	(+)	50	1
	(−)	1	48

Table 2
Details of distinct syphilis serological test results obtained with three different treponemal test kits.

	Mediace TPLA ((+): ≧10.0 T.U.)	Architect Syphilis TP ((+): ≧1.0 S/CO)	Lumipulse presto TP ((+): ≧1.0 C.O.I)	Mediace RPR	Serodia-TP PA	FTA-ABS	Classification
Case 1	(−) 3.5	(+) 2.52	(+) 4.3	(+)	(−)	(+)	Current syphilis likely
Case 2	(+) 11.0	(−) 0.85	(+) 1.0	(−)	(−)	(+)	Syphilis likely
Case 3	(+) 10.2	(−) 0.52	(−) 0.6	(−)	(−)	(−)	Syphilis unlikely

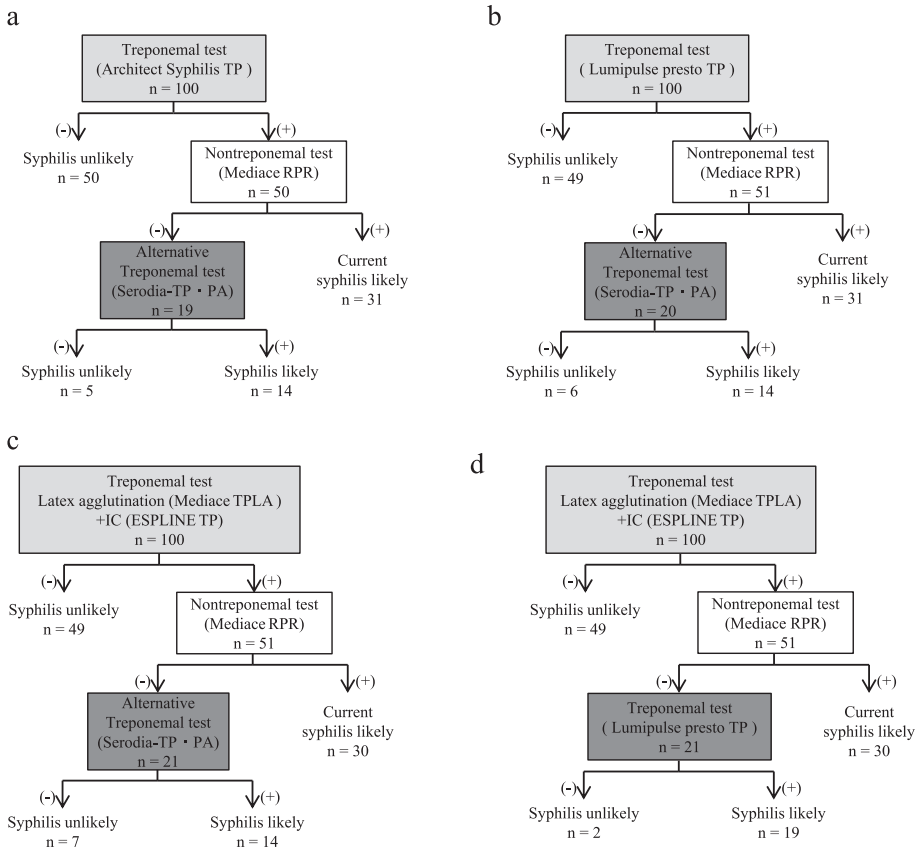

Fig. 1. Comparison of the original and modified reverse algorithms. All samples were classified the syphilis infection status guided by the algorithm using Architect Syphilis TP (a), Lumipulse presto TP (b), and both Mediace TPLA and ESPLINE TP (c) as an initial test. (d) In addition to using both Mediace TPLA and ESPLINE TP as an initial test, samples were classified guided by the algorithm using Lumipulse presto TP as an alternative treponemal test instead of the SERODIA-TPPA.

discordant results between treponemal tests were obtained. Furthermore, performing the test is complicated by handling and the need for technical skills and experience. From the above, in a new algorithm using TPLA as an initial screening test we recommend the treponemal test by EIA or CIA as the alternative treponemal test following RPR test, according to our result. Therefore, we believe that the new modified reverse algorithm using TPLA as an initial screening test has clear advantages over other algorithms. However, we have to concern the limitation of this retrospective study because of the use of frozen serum which have been stored for some time may affect the results.

Conflicts of interest

None.

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