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Toxoplasma gondii, cytomegalovirus, and Treponema pallidum
infections in pregnancy: what's new?

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Toxoplasma gondii, cytomegalovirus, and *Treponema pallidum* infections in pregnancy: what's new?

Congenital infections pose specific challenges in diagnostics and medical care. Infections acquired before or during pregnancy can extend to the fetus and lead to adverse outcomes, including fetal/ newborn death and malformation. Certain factors can affect the clinical course, such as the timing of an infection. At the 26th European Congress of Clinical Microbiology and Infectious Diseases (ECCMID), experts discussed the diagnosis and treatment of congenital toxoplasmosis, cytomegalovirus (CMV) infection, and syphilis.

Toxoplasmosis: changes in epidemiology

Toxoplasma gondii is one of the most successful parasites. It shows a world-wide distribution, and toxoplasmosis can be life-threatening in immunocompromised patients and fetus. However, there is no consensus on patient monitoring and treatment, and information on the impact on public health is scarce. Toxoplasma is a food-borne disease, but the risk factors related to cysts vary greatly between countries. "Therefore, prevention should be tailored to the local risk", explained Prof. François Peyron, Institut de Parasitologie Mycologie Médicale, Hôpital de la Croix-Rousse, Lyon, France. In South America, toxoplasmosis was demonstrated to be water-borne as well. As Prof. Peyron indicated, the type of route of infection matters greatly, as it has a dramatic effect on the choice of preventive measures.

Another important epidemiological aspect is the declining seroprevalence. In France, seroprevalence rates dropped from 80 % in the 1960s to 37 % in 2010 (Figure 1), and they are expected to decrease further. The same trend has occurred in the United States. Among the multifactorial causes responsible for this development, those that stand out are improved hygiene and food consumption (e. g., industrial freezing of meat), as well as information programs. Decreases in seroprevalence affect the cost-benefit ratio of screening of pregnant women, rendering these programs more expensive. However, it is not known if declining seroprevalence is a world-wide phenomenon, as information from many countries is lacking.

Virulence might depend on location

T. gondii shows a certain genetic diversity. Almost all European strains belong to the type-II category, which have only low virulence. In South America, on the other hand, a variety of atypical genotypes has been identified. "Importantly, there is a link between clinical virulence and genotypes", Prof. Peyron stated. South American strains tend to have deleterious effects on the retina, which can cause ocular lesions. "When gross malformations occurred in fetuses in France, it was found that these infections had occurred through horse meat that was imported from South America."

According to the traditional pathophysiological knowledge, life-long protective immunity is conveyed by the presence of toxoplasma cysts. Cysts are supposed to persist during the entire life of the hosts. However, the dogma of "once infected, forever protected" is being challenged these days. "Serology can turn negative over time in congenitally infected people," Prof. Peyron observed. The concept of the cyst lasting the entire life of the host should therefore be revisited.

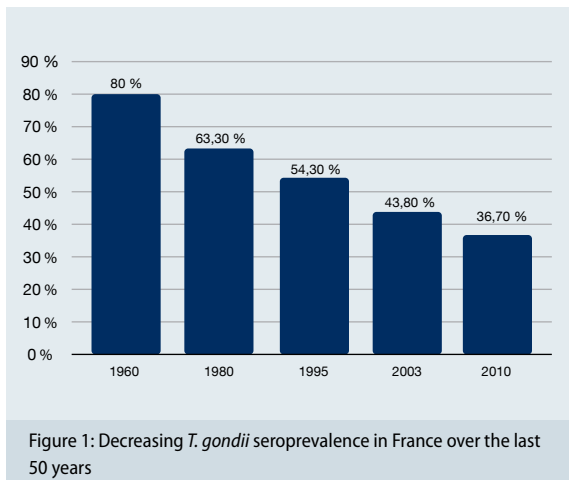
Diagnosis in pregnant women

In antenatal diagnosis, important progress has been made over the last 20 years due to the use of amniocentesis and ultrasound. If amniocentesis is positive, treatment needs to be reinforced, because the fetus will be infected. Ultrasound can reveal gross abnormalities. In France, pregnancy termination is offered only in cases of abnormality detected by ultrasound. In the area of Lyon, this applied to less than 1 % of 2,500 documented maternal infections. "A diagnosis of toxoplasmosis in pregnant women therefore does not mean that pregnancy termination needs to be considered, at least not in Europe," Prof. Peyron emphasized.

In the first trimester, the probability of congenital infection with a positive polymerase chain reaction (PCR) test result obtained from the amniotic fluid is 64.0 %; in the second trimester this rises to 95.4 %, and in the third trimester, to 98.2 % (Table 1) [1]. Here, it can be assumed that the fetus is infected, and so the treatment and monitoring of the woman need to be adapted. If PCR testing is negative, the probabilities of congenital infection across the trimesters are 1.0 %, 10.0 % and 22.6 %, respectively.

Established and new testing systems

Many diagnostic tests are available, with new kits entering the market also at present. Recently, serological testing has tended to focus on the use of recombinant antigens. "We should use a cocktail of recombinant antigens instead of only one or two, because we are targeting a very small epitope," Prof. Peyron noted. "If genetic drift is present, testing might miss the target." By dating maternal infection, it is possible to differentiate acute from chronic infection. How-



ever, interpretation of values within the grey zones of the kits is still a problem. When mass screening for IgG is performed, false-positive results must be avoided. In IgM screening, conversely, the same applies to false-negative results. Avidity testing is helpful here, but it does not solve the problem entirely. A comparison of anti-toxoplasma IgG avidity kits was performed on 84 clinically defined sera, including the Architect, LIAISON®, and VIDAS kits. “For chronic infection, the majority of sera displayed high avidity, especially with LIAISON®,” Prof. Peyron said. Low avidity, on the other hand, does not necessarily relate to acute infection.

A new diagnostic aspect arises from cellular immunity testing. “Cellular immunity has been disregarded in the clinic for many years, because it was considered too complicated,” Prof. Peyron explained. “Fortunately, testing can be performed now at low cost.” This technique, which can be very useful in pregnant women and newborns, is based on the specific secretion of interferon γ . Sensitivity and specificity are 96 % and 91 %, respectively, for the discrimination between infected and non-infected individuals. Other new tests that might be interesting for the screening of pregnant woman and newborns involve rapid diagnostic tests, saliva tests, and the Multiplexed Anti-*Toxoplasma* IgG, IgM and IgA Assay on plasmonic gold chips. Saliva testing is easy to perform and cheap, but sensitivity and specificity need to be improved. For the Multiplexed Assay, high rates of sensitivity and specificity of almost 100 % have been obtained [2].

Does toxoplasma infection influence behavior?

The potential for induction of behavioral changes by *T. gondii* is a controversial topic. Discussions started when researchers observed that infected mice were attracted by cat urine,

thus promoting the completion of the parasite life cycle, as these mice will be more likely to be eaten by cats. Manipulation of the host might likewise occur in human beings. “It is assumed that cysts located in the brain cause local inflammation that alters neurotransmitters,” Prof. Peyron noted. Increased levels of dopamine have been found in infected mouse brains. Also, tyrosine hydroxylase, which induces dopamine, is encoded in the parasite genome. However, a recent study identified no relationship between seropositivity and depression in 6,663 adults [3]. Prof. Peyron and his team also conducted a study using a questionnaire in 102 congenitally infected adults, which did not show any behavioral abnormalities [4].

Nonetheless, a possible social impact of *T. gondii* infection is conceivable. “Positive serology might prevent candidates from applying for sensitive jobs, such as school bus drivers or airline pilots.” Also, a person accused in court might claim to be not guilty because of seropositivity. Prof. Peyron warned against disregarding the issue of toxoplasmosis and behavioral changes because of the danger of misuse, with a clear-headed view called for. Comprehensive studies need to be conducted, and stigmatization of seropositive people should be avoided.

Screening: the issue of cost-effectiveness

There is no consensus across countries with respect to toxoplasmosis screening of pregnant women, while at the same time, early diagnosis and effective treatment can make an enormous difference. The outcome is very good when antenatal screening reveals the infection in time and treatment is applied to the pregnant woman and to the newborn. On the other hand, fetal loss and malformation can occur if no measures are taken. “Nowadays, we have strong evidence that antenatal treatment works,” Prof. Peyron stressed. Studies show that antenatal treatment is efficient when given within 4 weeks of maternal infection [5], and that it reduces the risk of transmission from mother to child [6]. In France, monthly screening was introduced in 1992, and fetal infections decreased afterwards [7]. Severe neurological sequelae were reduced when treatment started during pregnancy [8].

“The challenge is how to reduce the costs of screening,” Prof. Peyron said. This might be achieved by new tests, like saliva tests, and also by information programs for pregnant women and care providers. The burden of disease should be evaluated for each country. Other measures include the implementation of information programs and reference centers, and the release of homogeneous guidelines, at least for Europe. As Prof. Peyron pointed out, it is important to attract the attention of policymakers. “Congenital toxoplasmosis must not become a neglected disease.”

Indication	1 st trimester	2 nd trimester	3 rd trimester
Pretest probability of congenital infection	2.2 %	23 %	56 %
Positive likelihood ratio	79	69	43
Probability of congenital infection with a positive test (90 % CI)	64 % (39 %–100 %)	95.4 % (91 %–100 %)	98.2 % (96.2 %–100 %)

Tab. 1: Evolution of the probability of congenital infection according to positive PCR



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What is the value of the toxoplasma IgM?

Prof. Peyron: IgM is very important, because it is a marker of acute infection. At first IgM rises, which could be specific or not, and afterwards IgG levels rise. Then one can be sure that the woman has been recently infected. According to the timing of infection during pregnancy, the risk of fetal infection and the severity of infection vary considerably.

How does avidity work?

Prof. Peyron: Avidity should be considered as an interesting tool for dating infection. When the antibody response matures, the highly variable region of the antibody changes, and the affinity to the specific epitope increases. With recent infection, IgG avidity is usually low, but many pregnant women keep low avidity for a long time. Therefore we only rely on high avidity to rule out recent infection. Also, avidity is kit sensitive.

How do toxoplasma screening policies vary between countries?

Prof. Peyron: In France it is mandatory, while in Italy it is highly recommended, and in Germany, it depends on the region. In the United States, there is no screening at all despite lobbying. Switzerland dropped screening 15 years ago. At that time, there was no clear evidence that antenatal treatment worked. Policymakers must ask themselves how much money they want to invest in order to treat one case. Cost-benefit studies are difficult, because the costs of child care in institutions must be considered as well. Information campaigns are very important. In France, we attempted to pass this task to obstetricians, but this did not work, because physicians usually do not have enough time for patient education. Screening and information are very important.

Cytomegalovirus: watch out for primary infection in pregnancy

The overall birth prevalence of congenital CMV infection is 0.64 %. In pregnant women, CMV infection can be primary or non-primary (i.e., as reactivation or reinfection). Transmission to the fetus can occur in both cases. The rate of transmission remains constant throughout pregnancy, with the risk being considerably higher in primary than in non-primary infection (40 %–50 % vs. 0.5 %–1 %). Accordingly, significant handicaps at birth (e. g., mental retardation, bilateral hearing loss) are seen more frequently in cases of primary infection (30 %–40 %, vs. 1 %–4 % in non-primary infection) [9]. Although, increasing observations demonstrate the risk for symptomatic infection at birth and sequelae, especially hearing loss, are similar upon primary and non-primary maternal CMV infection [10].

“Routine antenatal screening for CMV has never been recommended by any public health authority in any country,” said Prof. Tiziana Lazzarotto, Laboratory of Virology, Operative Unit of Clinical Microbiology, St. Orsola Malpighi University of Hospital, University of Bologna, Italy. Nevertheless, *de-facto* screening is performed through extensive testing in Europe and Israel, and in some States in the United States.

Prevention: prenatal treatment and hygienic intervention

No active CMV vaccines have been established yet. Passive immunization of pregnant women with CMV-specific hyperimmune globulin treatment (e.g., Cytotect®, Cytogam®) is currently being investigated. In 2005, Nigro et al. showed that CMV-specific hyperimmune globulin is safe and might be effective in the treatment and prevention of congenital CMV infection [11]. However, these results were not confirmed by the randomized, double-blind, placebo-controlled, phase IIb CHIP study [12]. An open, single-arm phase III trial has been investigating Cytotect® in pregnant women who have experienced seroconversion in the first trimester. Publication of these results is expected soon. A randomized,

double-blind, placebo-controlled phase III study is assessing Cytogam® in pregnant women with primary infection. This trial will be completed in December 2018.

A controlled study provides evidence that behavioral measures and hygiene counselling of CMV-seronegative pregnant women can prevent maternal CMV infection [13]. The rates of seroconversion for the interventional group and the control group were 1.2 % and 7.6 %, respectively ($p < 0.001$). The number of newborns with congenital infection was 3 vs. 8. Young children are the primary source of CMV infection for pregnant women; the virus is shed in urine, saliva and tears. “Therefore, it is very important to offer information regarding hygienic and behavioral measures.”

Diagnosis of maternal infection

Maternal CMV infection can be ascertained in a reliable way by means of serological diagnosis if IgG and IgM screening tests are performed before 12 weeks of gestation. With regard to the results, four categories are possible (Table 2). Depending on the combined results and the period of pregnancy, different measures are required.

“It is very important to offer avidity testing before 16 weeks of gestation, because during this period, the sensitivity for the identification of an elevated risk of fetal transmission is 100 %,” said Prof. Lazzarotto [14]. “After 21–22 weeks, it decreases to 60 %.” A high avidity index before 16 weeks of gestation indicates no current or recent primary infection, whereas a low or moderate avidity index suggests acute or recent primary infection. IgG avidity assays, such as LIAISON® CMV IgG Avidity, show high sensitivity and specificity. “Serological tests vary from one laboratory to another,” Prof. Lazzarotto cautioned. “Therefore, the method used and its reference values must be carefully assessed.” The reference values for low, moderate and high avidity depend on the type of commercial kit. Moreover, false high avidity might be measured with automated test systems at very low CMV IgG levels. Virological diagnosis can complement the picture here; real-time PCR is used to identify viral DNA in body fluids (i. e., whole blood, saliva, urine).

Categories	Period of pregnancy	What does this mean?	What should be done?
IgG- IgM-	Just before pregnancy or during pregnancy	Non-immune women. High risk of acquiring primary infection	Information on hygiene and behavioral measures
IgG+ IgM-	Before 16 weeks of gestation	Past infection	No further diagnostic investigation required
IgG- IgM+	Before or during pregnancy	Indicative of primary infection or IgM false-positive result	Serological testing performed in the same laboratory after 10–15 days
IgG+ IgM+	Before 16 weeks of gestation	Indicative of <ul style="list-style-type: none"> • Active infection • Primary infection • Non-primary infection 	Advanced diagnosis → avidity IgG test

Tab. 2: Primary CMV infection in pregnant women: interpretation and measures to be taken

Diagnosis of fetal infection

As Prof. Lazzarotto stressed, it is important to offer antenatal CMV diagnosis. This is reliable, with amniotic fluid being the most appropriate material. Fetal blood does not offer any additional diagnostic value; also, fetal blood assessment carries a high risk of fetal demise and should therefore be avoided. Prenatal diagnosis is indicated 6–8 weeks after the onset of maternal infection. Amniocentesis should be performed at 20–21 weeks of gestation. The fetus excretes CMV via urine into the amniotic fluid, and a sufficient amount of fetal diuresis is produced only at that time. “Amniocentesis must be accompanied by ultrasound examination and counselling,” Prof. Lazzarotto said. The sensitivity, specificity, and predictive values of PCR tests for CMV-DNA detection in amniotic fluid are very high. In the newborn, the gold standard of diagnosis is real-time PCR on urine or saliva.

Syphilis: a disease is back

Treponema pallidum is the causative pathogen of syphilis, which remains a public health concern. In 2011, the World Health Organisation (WHO) estimated that 36.4 million adults are infected worldwide. At the same time, a successful test for this chronic bacterial infection has been available since the early 1900s, and penicillin treatment was introduced in the 1940s. “After the introduction of penicillin, there was a

marked decline in reported syphilis cases,” noted Dr. Pascale Huynen, Clinical Microbiology Department, University Hospital of Liège, Belgium. “Over the last decade, however, this rate increased again, notably in Russia, China and the United States.” In developed countries, increases in syphilis rates are seen mainly for the high-risk groups, such as the homosexual male population.

Congenital syphilis is based on transplacental transmission mainly during the second half of pregnancy or at birth [15]. The risk is maximal in cases of recent maternal infection, within the previous year. Also, the transmission rate depends on the stage of maternal syphilis: it is 60 % to 80 % if primary or secondary syphilis is present. In late syphilis, the risk decreases to 8 %. “Approximately 1.5 million pregnant women are infected with active syphilis every year”, reported Dr. Huynen [16, 17]. “Half of them remain untreated, which results in adverse outcomes.” These include early fetal loss, stillbirth, perinatal death, prematurity or low birth weight, and serious neonatal infection, which is analogous to the secondary stage of acquired syphilis. Clinical signs of congenital syphilis at birth include hepatosplenomegaly, abnormal facial features, edema, abdominal distension, pallor, skin lesions, and fever. However, approximately half of infected newborns are asymptomatic.

Are automated tests useful for high-volume routine?

Prof. Lazzarotto: Yes. At my hospital, the microbiological laboratory performs CMV-testing on 150 samples a day. This is impossible without automated systems. Also, they are important regarding the prevention of human errors. The bar code is recognized by the system, and the history of the vial is fully traceable. Interpretation by laboratory technicians, which can be wrong due to misreading, is not necessary.

The Nigro study results have not been confirmed, and the CMV vaccine has been demonstrated not to be successful. Are there any other drugs in development with promising results?

Prof. Lazzarotto: Some international companies are currently working on a new vaccine. The previous vaccine included only one glycoprotein; this was not enough in terms of efficacy. This new vaccine includes other glycoproteins and

further proteins involved in the pathogenesis of CMV. At present, antiviral treatment is not available in pregnancy. Foscavir and cidofovir are nephrotoxic and cannot be administered in pregnant women. For ganciclovir, we do not have strong evidence that it crosses the placenta. Another problem relating to ganciclovir is a decrease in fetal-fertility, which was demonstrated in the animal model.

Which aspects would you deem especially important for daily practice?

Prof. Lazzarotto: Even in the absence of preventive treatment or vaccines, we can resort to hygienic measures. It was shown that behavioral measures are very effective for the prevention of CMV. The education of pregnant women is therefore very important. Also, we have the possibility to treat the newborn. Randomized controlled trials established that it is mandatory to start treatment before one month of age.



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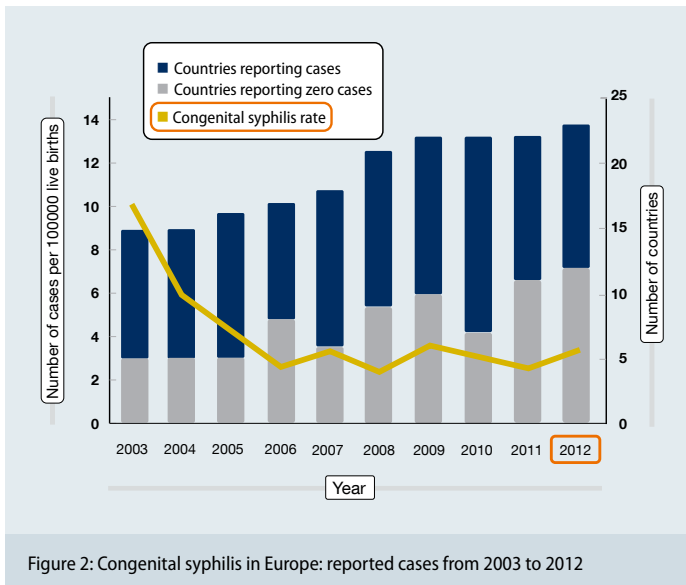


Figure 2: Congenital syphilis in Europe: reported cases from 2003 to 2012

Congenital syphilis is particularly prevalent in developing countries. According to WHO estimates, 96 % of maternal syphilis infections and 98 % of adverse outcomes occur in low-income and middle-income areas. In Europe, the figures pertaining to congenital syphilis have remained stable over the last 10 years (Figure 2) [18], but it is suspected that there is considerable underreporting.

Diagnostic options

Syphilis can be identified via direct diagnosis from the ulcer or lesion in the pre-serological phase, or via detection of antibodies to *T. pallidum* in serum, cerebrospinal fluid (CSF), or whole blood. Direct diagnosis is performed using dark-field microscopy, antigen detection via fluorescence microscopy, and molecular biology. The molecular biology approach has high sensitivity and specificity, and it can also be performed on amniotic fluid and material from the placenta and umbilical cord. However, serology is the main laboratory tool for screening and follow-up of treatment [19]. Also, serology is the only method that can identify latent syphilis.

A wide range of tools is available. Non-treponemal tests provide information on the activity of the disease, while treponemal tests are more specific, and can thus be used to confirm reactive non-treponemal test results. However, unlike the non-treponemal tests, treponemal tests cannot distinguish between active and previously treated infection. "A combination of the two test types is recommended," Dr. Huynen said. No serological test that can discriminate between syphilis and non-venereal treponematosi has been established to date.

Non-treponemal and treponemal tests

Non-treponemal tests, such as Rapid Plasma Reagin (RPR)/Veneral Disease Research Laboratory (VDRL) tests have the advantage of being inexpensive and quantitative. "They are the only tests recommended to follow the course of the disease during and after treatment," noted Dr. Huynen. Testing is simple and rapid, and can also be performed on CSF (i. e., for the diagnosis of neurosyphilis). As the tests are not

specific for *T. pallidum*, false-positive results due to cross-reactivity represent the main limitation. The specificity is 93 % to 98 %.

Treponemal tests include fluorescent treponemal antibody absorbed test (FTA-Abs), *Treponema pallidum* hemagglutination assay (TPHA)/ *Treponema pallidum* particle agglutination assay (TPPA), enzyme immunoassays (EIAs)/chemiluminescent immunoassays (CLIAs)/ micro-bead immunoassays (MBIAs), rapid tests, and Western blotting. TPHA and TPPA are specific for *T. pallidum*, and are quantitative for serum and CSF. The sensitivity ranges from 85 % to 100 %, and specificity from 98 % to 100 %. "Reactivity usually persists over the lifetime, even after treatment," Dr. Huynen explained. The fluorescent treponemal antibody absorbed test is based on immunofluorescence. It has high sensitivity (70 %–100 %) and specificity (94 %–100 %), but it remains positive even after treatment. Indications include assessment of congenital syphilis (IgM) and confirmation of recent infection demonstrated with RPR/TPHA. Western blotting is a confirmatory test that can contribute to the diagnosis of congenital syphilis.

Rapid tests are based on immunochromatography, and these can be used in developing countries, as they are cheap and require only minimal training. A small amount of whole blood collected by a finger prick is sufficient. However, the test cannot distinguish between active and past infection. "More than 20 tests are available," Dr. Huynen said. "Most of them exhibit high sensitivity and specificity."

What can enzyme immunoassays do?

In recent years, enzyme immunoassays (i. e., EIA/ CLIA/ MBIA) have been developed for the screening of syphilis. Their sensitivity and specificity is comparable to those of other tests (82 %–100 %, 97 %–100 %, respectively), and they allow for IgM and/or IgG detection. IgM assessment is very sensitive in early infections and congenital infections. As enzyme immunoassays are qualitative, their only indication is screening. Test results do not correlate with disease activity, and they remain positive even after treatment, except in 15 %–25 % of patients who are treated early (primary stage).

Enzyme immunoassays offer advantages over treponemal tests because they can be automated, allowing for high throughput, with diminished laboratory occupational hazard. Manual pipetting is not necessary, and false-negative results due to the prozone reactions are avoided.

LIAISON® was the first CLIA available for *T. pallidum* antibody screening (for total antibodies) of serum and CSF. This system uses the TpN17 recombinant specific *T. pallidum* antigen. "It has been well demonstrated in several studies, but also in our routine practice, that these tests exhibit high sensitivity and specificity of more than 99 %," Dr. Huynen reported [20, 21]. LIAISON® XL is fully automated, and offers the advantages of a random-access system and full traceability. The reagents are also stable over long periods of time (≥ 4 weeks).

Recommendations for follow-up

Different algorithms are used for serological screening and diagnosis. These include the algorithm provided by the American Centers for Disease Control and Prevention, and

the recommendations by WHO and European Guidelines on the Management of Syphilis 2014 (Figure 3) [22]. In this case a single treponemal test is used as screening. For confirmation both treponemal and non treponemal tests are needed to assess if the infection is active or not. If discordant results are obtained, then another treponemal test such as western blot can be used. Full interpretation of the test results requires a synopsis of different tests (Table 3). For follow-up, quantitative testing (e. g., RPR) is recommended every 2 to 3 months. If 4-fold decreases in titers occur, it can be assumed that the treatment has worked. Increasing titers, on the other hand, hint at reinfection or reactivation.

At birth, the same serological profile (IgG) is present in the mother and newborn. IgM detection can be performed to highlight antibody production by the newborn. Quantitative RPR serologic titers that are 4-fold higher in the baby than the mother are highly indicative of congenital syphilis. For the follow-up of an infected child, RPR testing is recommended every 2 to 3 months until the test becomes seronegative. "A neonate with negative RPR born to a seropositive mother should be retested at 3 months to rule out incubating congenital syphilis at birth," Dr. Huynen explained. Importantly, treponemal tests should not be used in newborns, because passive transfer of maternal IgG might persist for more than 15 months.

Treatment of the mother and the newborn

"In pregnancy, all patients with a positive test should be treated," Dr. Huynen emphasized. Penicillin G is the only known effective drug for preventing maternal transmission and treating fetal infection. Benzathine penicillin G is recommended at a dose of 2.4 million units intramuscularly. Pregnant women should receive penicillin appropriate to their stage of infection. Primary, secondary and early latent syphilis call for a single dose, while three doses should be administered for the other stages. Neurosyphilis requires treatment for 10 to 14 days.

Neonates born to women showing reactive serologic tests for syphilis should be examined thoroughly for evidence of congenital syphilis. In cases of proven, highly probable, or possible congenital syphilis, either aqueous crystalline penicillin G (100,000-150,000 units/kg/day, intravenously) or procaine penicillin G (50,000 units/kg/day, intramuscularly) should be applied for 10 days. If congenital syphilis is less likely, a single dose of benzathine penicillin G (50,000 units/

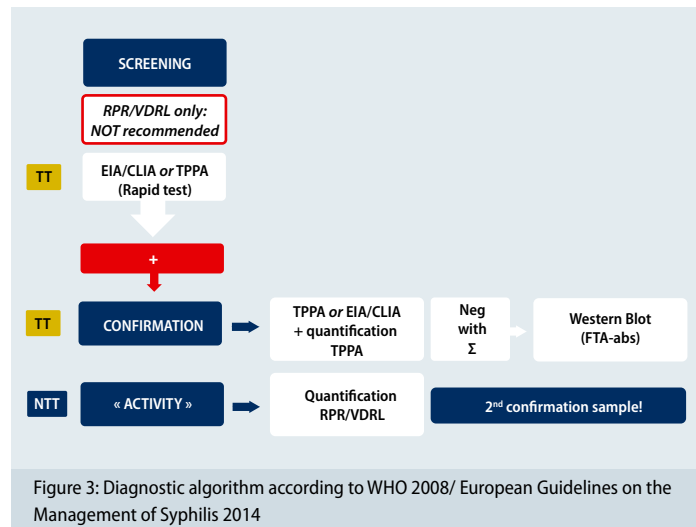


Figure 3: Diagnostic algorithm according to WHO 2008/ European Guidelines on the Management of Syphilis 2014

kg, intramuscularly) is indicated. Neonates who show persistent RPR titers by 6 to 12 months should be re-evaluated and treated with 10 days of penicillin G. Also, CSF examination should be included in the work-up.

Prevention of congenital syphilis is worthwhile

Worldwide, congenital syphilis is responsible for the death of more than one million babies every year. As syphilis infection can be asymptomatic, detection is often delayed. "Unlike many neonatal infections, congenital syphilis is a preventable disease," Dr. Huynen pointed out. Infected mothers should be identified and treated before the middle of the second trimester. "Serological screening in all pregnant women – not only in those being perceived as a high-risk group – and treatment are feasible, even in low-resource settings," Dr. Huynen said. Information on the importance of treating the women, the newborn, and the partners should be provided to the patients.

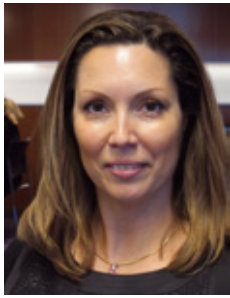
For pregnant women, screening is recommended at the first prenatal visit. In the case of increased risk, testing should be repeated at 28 weeks of gestation and at delivery. Information concerning risk behaviors and treatment of sex partners should be obtained to assess the risk of reinfection. For newborns, routine screening is not recommended. If they are born to mothers with syphilis, quantitative non-treponemal

Enzyme immunoassays (EIA/CLIA)*	Rapid Plasma Reagin (RPR)	Treponema pallidum particle agglutination assay (TPPA)	Interpretation
Positive	Negative**	Positive	Primary syphilis OR treated syphilis OR latency
Positive	Positive**	Negative	Primary syphilis OR false positive result
Positive	Positive	Positive	Active syphilis OR treated syphilis (RPR < 32)

*EIA, enzyme immuno assays; CLIA, chemiluminescent immunoassays

** FTA-Abs IgM if active infection suspected

Tab. 3: Interpretation of the results of syphilis testing



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How do you rate the development of the syphilis cases diagnosed in your laboratory?

Dr. Huynen: The incidence used to be low in the past, but now the numbers are increasing. Our hospital contains a large center for AIDS patients, and another center for hepatitis C patients. Also, there are refugee centers in the vicinity of the hospital.

How would you judge the performance of the LIAISON® Treponema assay as used in your laboratory?

Dr. Huynen: We use this test only for screening, because it is semi-quantitative. It cannot be used for the follow-up. It is a very sensitive test; for early pregnancy, the results show that the test is surprisingly sensitive. We had one case of a patient in whom only the LIAISON® TP screening

test and the PRP were positive. The other treponemal tests were negative. The clinician was sure that the patient did not have syphilis, but one week later the diagnosis was confirmed. Also, the LIAISON® TP test is highly specific at higher antibody levels. At very low results close to the cut-off, false-positive results can occur, and so the testing of a second serum sample is required to confirm the diagnosis. However, we only once observed positive results with the LIAISON® screening that turned out to be negative with TPHA testing. This was a patient who was infected with hepatitis C and HIV. Six weeks later the syphilis test result was highly positive with both treponemal tests and RPR. In some cases in whom the interpretation of the TPHA results is difficult, we use LIAISON® TP screening of the CSF to confirm the results.

mal testing is called for. "No mother or newborn should leave the hospital without determination of the maternal serological status at least once during pregnancy, and again at delivery if the risk is increased," Dr. Huynen stated.

Screening and treatment for syphilis in pregnant women are cost-effective antenatal interventions, and are strongly promoted by the WHO, as large reductions in congenital syphilis are feasible. Between 2008 and 2012, a 33 % decrease in maternal infections and adverse pregnancy outcomes was noted. India alone represented 37 % of this decline due to improvements in data quality and efforts towards the control of sexually transmitted infections. "This is a promising sign that efforts to control syphilis in pregnant women are having a true public health impact."

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