

## Principles of Diagnosis of Lyme Borreliosis in Croatia

**Gordana Blagojević Zagorac\***

*Department of Physiology and Immunology, Medical Faculty, University of Rijeka, Rijeka, Croatia*

**\*Corresponding Author:** Gordana Blagojević Zagorac, Department of Physiology and Immunology, Medical Faculty, University of Rijeka, Rijeka, Croatia.

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Lyme borreliosis is an infectious multisystem inflammatory disease caused by bacteria *Borrelia burgdorferi* and disease is transmitted from animals to humans through a bite from an infected tick. Lyme borreliosis has early and late manifestations and beside pathognomonic erythema migrans rash which occurs in about 50 - 70% patients, symptoms are not specific and can mimic many other diseases. Therefore diagnosis of Lyme disease is quite challenging.

In Croatia, like in many other countries, diagnosis of Lyme disease is based on appearance of the pathognomonic erythema migrans, and on detection of *Borrelia burgdorferi* specific antibodies in a patient's blood. The standard procedure of antibody testing includes initial (screening) ELISA testing and confirmatory Western blot testing in the case of positive or equivocal ELISA results. Other tests like PCR and cultivation of *Borrelia burgdorferi* are rarely used and test like SeraSpot or EliSpot that can be done in some foreign laboratories are not yet available in Croatia.

In the cases where pathognomonic erythema migrans is not proven, there are many traps in this two-step approach that must be taken in consideration when dealing with a patient with suspicion of a Lyme disease. First of all, with this approach the patient is consider not to have Lyme disease if screening result of ELISA testing is negative and in that case no further testing is advised and antibiotic therapy is not recommended. Although it is consider for ELISA testing being low specific but high sensitive it must be noticed that there are cases in which patients with Lyme borreliosis are negative in ELISA testing (e.g. if testing is done too early and there was no time for antibodies to develop) and therefore diagnosis of Lyme disease cannot be ruled out only on the basis of ELISA testing and further tests have to be done if clinical picture rises doubts of Lyme disease. If ELISA test is positive or equivocal it is recommended that Western blot for both IgG and IgM antibodies should be done. Western blot is consider to be more specific then ELISA due to algorithms that are used in interpretation of Western blot testing, i.e. Western blot is consider to be positive only if at least 5 of 10 bands are read as positive. In the case of positive ELISA and negative Western blot, Lyme borreliosis is again ruled out from differential diagnosis and ELISA is consider to be false positive due to their low specificity. But it must be considered that Western blot can be negative despite of presence of anti-*Borrelia burgdorferi* antibodies because Western blot has lower sensitivity then ELISA. Furthermore, by this approach only IgM positivity does not prove Lyme disease if there is no seroconversion to IgG four weeks after infection and it is also consider as false positive result although it is shown that in some cases seroconversion from IgM to IgG does not occur even in the later stages of infection.

We can conclude that although the diagnostic approach of Lyme borreliosis that is used in Croatia is reliable, in some cases it may result in a person with Lyme disease not to be recognized. Namely, beside patients with proven erythema migrans, by using these approach only patients with positive or equivocal ELISA test and at the same time IgG positive Western blot will be recognized as patients with Lyme borreliosis. Considering that early diagnosis and early treatment of Lyme borreliosis are very important, it is necessary to have in mind all pitfalls of this approach and besides antibody testing to give more importance to clinical presentation.

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