Tropical diseases conference 2019 Nano-ELISA in the Diagnosis of Experimental Toxoplasmosis Mervat Zakaria El Azzouni¹, Lobna Abd El-Aziz El Zawawy¹, Doaa El-Said Said Ahmed¹, Wegdan Ramadan Ahmed², Maha Mohamed Gomaa³

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Background: Toxoplasmosis is a worldwide endemic disease. In congenitally infected infants and immunocompromised patients, toxoplasmosis causes high rates of morbidity and mortality. In these cases antibody detection is not valuable and detection of parasite antigen could be useful. It can also provide specific diagnosis of acute infection. Gold nanoparticles can be conjugated with antibodies affording them promising applications in signal enhancement of bio-chemical detection of parasite antigen. Gold nano-ELISA was evaluated in the current work in the detection of Toxoplasma SAG1 antigen in immunocompetent and immunosuppressed mice, infected with RH strain or Me49 strain of T. gondii, for its potential as an early sensitive diagnostic technique.

Introduction:

Toxoplasmosis is a zoonotic parasite sickness in animals which resulting from a parasitic protozoan called Toxoplasma gondii (Tavassoli et al., 2013). This parasite has a excessive capability for infection the homoeothermic beings and it may be hidden after influencing in lots of species. Toxoplasma is a parasite from the Apicomplexa branch that is chargeable for the contamination of a wide variety of vertebrates. Human is stricken by this pathogen in two methods which might be, obtained and congenital (Dubey, 2008). As AIDS (received immune deficiency syndrome) incidence large, Toxoplasma as an opportunistic pathogen has been diagnosed as the main agent of death among AIDS sufferers. Toxoplasma triggered Encephalitis is the crucial complications of Toxoplasmosis in AIDS patients. About 20% up to 47% of people who've antibodies in opposition to Toxoplasma, face with Toxoplasma Encephalitis (Cox and Wakelin, 2010). Toxoplasmosis ends in large damages to each people and animals. Congenital Toxoplasmosis that is the end result of feticide, birth of toddlers with complications together with Hydrocephalus, Microcephaly, low intelligence, severe abnormalities in organs, blindness, physical problems, intellectual disorders inclusive of Schizophrenia results in monetary damages annually. Patient treatment inside the United States fee about \$ zero.Four to \$ eight.Eight and this fee in England anticipated to one.2 up to 12 million bucks (Markell et al., 2006). Rodents, ruminants such as sheep and cattle, birds, and pigs are some of the hosts of this parasite and cats are the very last host of this parasite (Mosallanejad et al., 2012). The enlargement of these hosts and the close relationship between these

animals and human beings are a chief motive of the incidence of Toxoplasmosis in humans. As a result of this reality, 500 to one million humans, who're approximately one-0.33 of the sector's overall populace, are confronted with this ailment (Shirbazou et al., 2013). Existence of appropriate sanitary situations within the society, particularly in high danger groups, prevention of intellectual disorders which caused by feticide, stillbirth, preterm delivery, congenital difficulties, show the significance of consistent monitoring of this not unusual disease in urban and rural groups. Three Diagnosis of Toxoplasmosis may be completed by using a few methods in addition to, molecular approach, Immunoblotting techniques, tissue biopsy and serology. These methods consist of Sabin-Feldman dye take a look at, indirect hemagglutination (IHA), oblique fluorescence antibody (IFA), ELISA (Meganathan et al., 2010). Another take a look at which known as the changed agglutination take a look at (MAT) that still has a high sensitivity and specificity seems on Tachyzoite (Gamble et al., 2005). Polymerase chain response (PCR) is likewise one of the excellent strategies for trying out Toxoplasmosis, which has a excessive sensitivity and specificity (Koloren, 2013). Among the above methods, ELISA method has excessive sensitivity and specificity. In this case, such a way which has industrial kits can broadly use in laboratories all around the international to measure IgM, IgG and IgE antibodies (Montoya, 2002). Unacceptable effective and negative outcomes can be the most important issue in case of these kits. Many research have been performed to perceive compounds of Toxoplasma as antigens to be used in serologic diagnostic techniques to enhance their diagnostic importance (Gamble et al., 2005; Glor et al., 2013). In this regard, high interest has been focused to the E/S antigens of Tachyzoite of Toxoplasma (Suzuki, 2002). Regarding to characteristics which are said for these antigens, it appears they are appropriate for detecting antibodies that act against the parasite in the serum (Nishikawa et al., 2002). Several research have shown that ELISA method which makes use of E/S antigens of Toxoplasma,

Methods: 192 laboratory bred Swiss Albino mice were used. A control non-infected group (group I) was formed of 60 mice; 30 mice were immunocompetent and the other 30 were experimentally immunosuppressed. An experimental group (group II) was formed of 132 mice divided into two subgroups. Subgroup IIa involved 60 T. gondii RH strain- infected mice; 30 mice were immu-

nocompetent and the other 30 were experimentally immunosuppressed. Subgroup IIb included 72 T. gondii Me49 strain- infected mice; 36 mice were immunocompetent and the other 36 were experimentally immunosuppressed. Six mice from each subgroup were sacrificed on days zero, 1, 2, 7 and 14 post infection (PI). The remainder six mice in subgroup IIb were kept up to 60 days PI, and then they were sacrificed to take their brain tissue to detect T. gondii cysts as a proof of infection. Blood samples were collected and serum was separated to be assayed for T. gondii SAG1antigen by nano-ELISA formats employing monoclonal anti-SAG1 as the capture antibody. Nano-ELISA was further compared to modified form of sandwich ELISA 'capture ELISA'.

Results: T. gondii SAG1 antigen was detected in RH strain- infected animals, whether immunocompetent or immunosuppressed, as early as the 1st day PI by nano-ELISA with a sensitivity of 83.3%. It was detected till the end of the experiment with a sensitivity of 100%. In Me49 strain- infected animals, whether immunocompetent or immunosuppressed, nano-ELISA detected the antigen starting from the first day PI with a low sensitivity (16.7%). While this assay recorded 83.3% sensitivity on the second day PI and 100% on the seventh and 14th days PI. Capture ELISA detected the antigen in mice infected with RH or Me49 strain of T. gondii, whether immunocompetent or immunosuppressed, only in the seventh and 14th day PI with a sensitivity of 83.3% and 100% respectively. There were no false positive results in the control subgroups neither by capture nor by nano-ELISA (100% specificity).

Conclusion: Nano-ELISA is a promising sensitive method for an early and specific diagnosis of acute phase of toxoplasmosis especially under immunosuppressive conditions.

Keywords: T. gondii SAG1 antigen, nano-ELISA, immunosuppression.

Biography:

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