70. EVALUATION OF THE WESTERN BLOT DENSITOMETRY FOR THE DIAGNOSIS OF CONGENITAL TOXOPLASMOSIS Luis Felipe Mosquera Solano¹, María Antonia Restrepo Duque¹, Jocelyn Alexandra Arredondo Torres¹, Sebastián Serna Rivera¹, Juan David Fandiño López¹, Jorge E. Gómez M². ¹ Second year Medical Student. University of Quindío, Armenia, Colombia.

² MD. PhD Professor, University of Quindío, Armenia, Colombia.

BACKGROUND: Congenital toxoplasmosis can cause severe ocular and neurological sequels such hydrocephaly, seizures, and chorioretinitis, leading to permanent visual deficit and multiple neurological deficits, if left untreated. An early diagnosis is crucial to prevent the development of these consequences and western blot is the most important diagnostic tool for this disease in newborns, because can distinguish between antibodies transferred from the mother or those produced by the newborn. However, the reading of the results still being done by subjective analysis by expert clinical laboratory specialists, reaching a maximum of 73% of sensitivity. The digitalization of the image of the results of western blot and its analysis by a software quantifying the densitometry of the bands obtained by western blot assay can potentially improve the diagnostic performance of the technique. METHODS: Was evaluated the sensitivity and specificity by digitalizing pictures of western blot results from the laboratory of Biomedical Research at the University of Quindío and published in the scientific literature. For the research,

GelAnalyzer 19.1 software was used to digitalize the bands from western blot membranes from mother and child serum and the molecular weight of protein bands was calculated. The images were from 15 cases of newborns during the first month of life (six from our laboratory and nine from literature), and five negative controls (three from the laboratory and two from literature). Definition of true cases were the persistence of IgM/IgA after day 10 of life, mother with IgG and IgM antibodies positive and newborn with positive IgG plus symptoms, or no decrease in IgG in follow-up. The true negative cases were all newborn whose become negative for IgG anti Toxoplasma before the month 10 of life in absence of treatment, indicating that were maternal passively transferred antibodies. RESULTS: When comparing the immunological profiles of the mothers and their children for IgG by densitometry, congenital infections were detected in 80% (12 out of 15) of the cases (sensitivity) in serum samples obtained during the first month of life. All controls were negative (100% specificity). There was a percentage of 53% (8 out of 15) of children not identifiable with the visual analysis performed by two undergraduate researchers, likewise, the specificity with observation was 57% (3 out of 5) contrasted with the present study. Finally, three bands were identified as the most common in newborns with congenital infection: 49 KDa, 98-97 KDa and 72 KDa proteins (in order of frequency). **CONCLUSION**: Digitalization of western blot images increase sensitivity for the diagnosis of newborn with congenital toxoplasmosis during the first month of life.

Figure. Densytometric Analisis of IgG anti Toxoplasmabands Profile Obtained by Western Blot Assay.

Mother's western blot				Newborn's western blot			
and # Mama 🔻	Rf v	Raw volum 🔻	MW -	Band #Hija	Rf2 -	Raw volume -	MW4
1.	0,04	69	235	1.	0,016	289	243
				2.	0,133	139	202
				3.	0,259	102	157
				4.	0,302	35	143
2.	0,354	72	124	5.	0,353	249	125
3.	0,392	238	111	6.	0,386	310	113
4.	0,474	170	82	7.	0,463	558	86
				8.	0,499	79	73
				9.	0,525	232	64
				10.	0,57	130	48
				11.	0,612	109	33
5.	0,696	972	4	12.	0,674	950	11
6.	0,75	307	0	13.	0,725	221	0
7.	0,823	1132	0	14.	0,789	1253	0
8.	0,873	272	0	15.	0,838	344	0
9.	0,901	733	0	16.	0,863	616	0
10.	0,936	191	0	17.	0,905	138	0
	88 9		8 9 10	250		8 8 60 63 63 69 64	16 15 12
50 50 50 50 50 50 50 50 50 50	, i i	225 250 275 100 125 8		200- 6150- 100- 50-	i i j	ini ini i	
0 25 50 75 100	125 150 175 200 Pix		0 3/5 400	0 25 50 75 10	125 150 175 200 ;	225 250 275 300 325 35	0 375 400 425 4

Densytometric Analisis of IgG anti Toxoplasmabands Profile Obtained by Western Blot Assay

-Peaks of 157,143. 73, 64, 48, 33 present in newborn but in mother.

-**Conclusion:** specific IgG produced by newborn, but not by the mother, indicating congenital infection.

Key words: Congenital Toxoplasmosis; Western Blot; Diagnosis of Toxoplasmosis (Source: MeSH-NLM).