"Immunological and molecular characterization of cysticerca antigen of Taenia solium"

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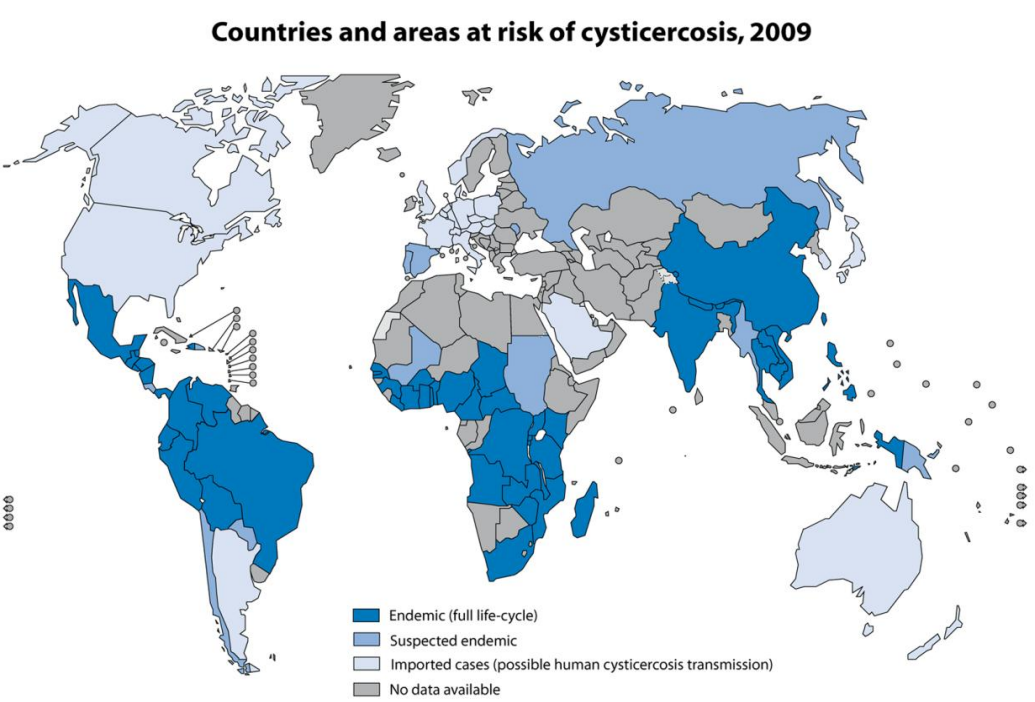
Background and Objetives

- Neuroimaging techniques are expensive and are seldom available to the majority of the population in endemic regions.
- ELISA and EITB assays are used most often to diagnose human cysticercosis , and both those techniques are specific and sensitive.
- However, they have the disadvantage of requiring expensive, specialized equipment that must be run by trained technicians, which limits the use of ELISA and EITB in poorly equipped laboratories in endemic countries. The use of parasitic material.

- To identify immunogenics proteins from *T. solium*
- To establish an easy to use diagnostic test for human cysticercosis

Cysticercosis Prevalence



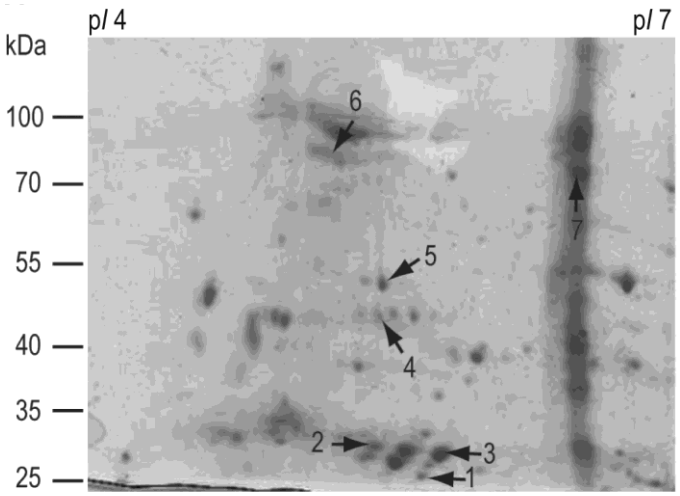
Humans - 20%
Pigs - 33%

Humans - 12%
Pigs - 2%

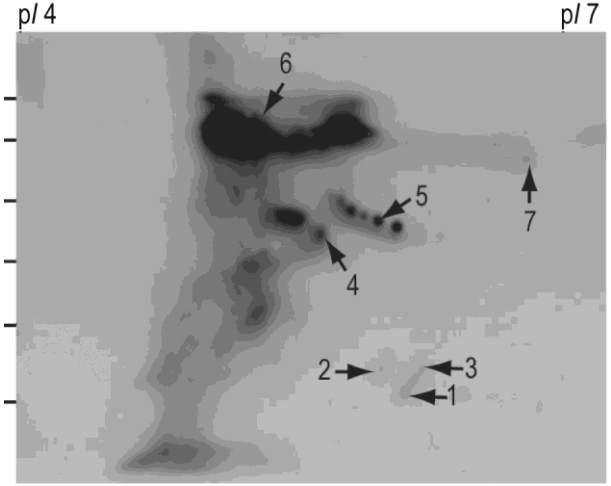
Identification

- Proteins were recognized by sera obtained from patients with NCC
- Comparasion of the western blot and 2D gel stained allowed the localization of antigenic proteins
- Sequence were found within an EST library

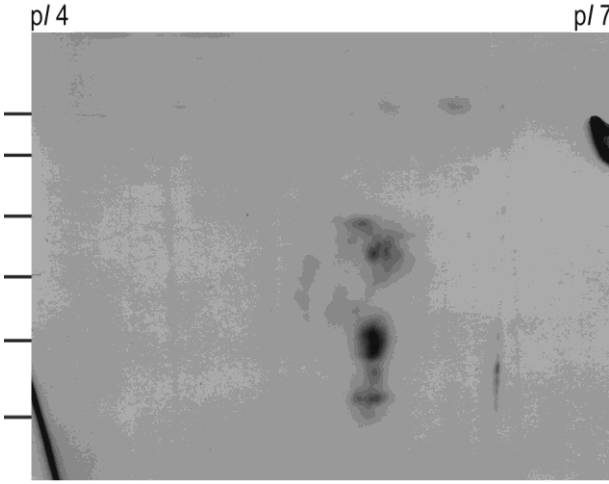
Identification of immunogenic proteins Nicaragua



Coomassie



Positive sera



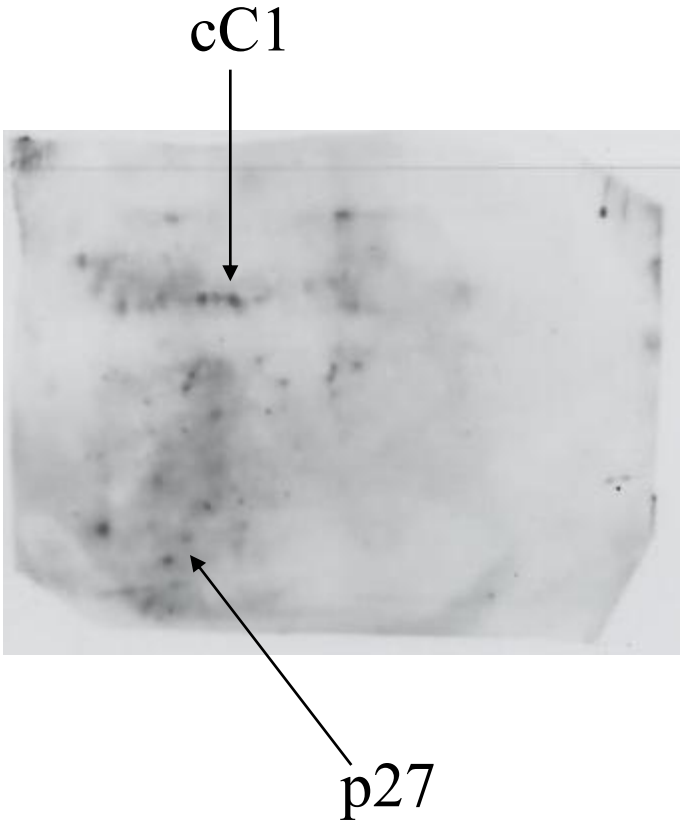
Negative sera

Amino acid identification and gene identification

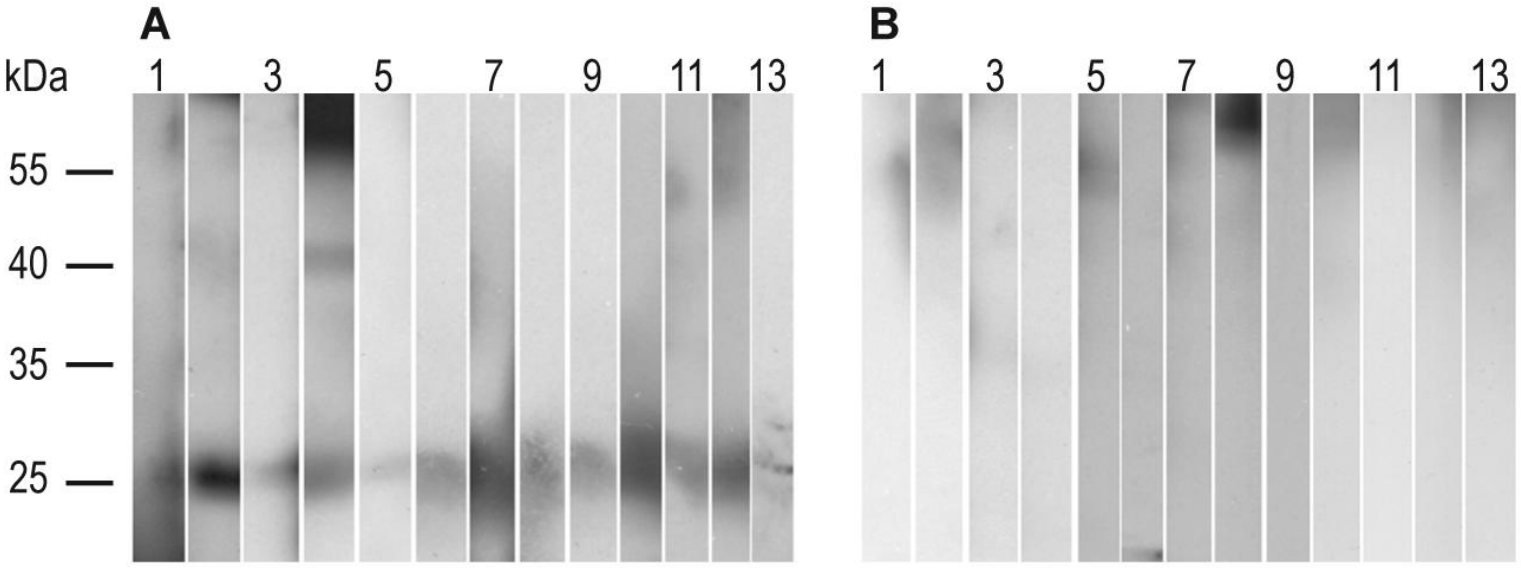
Spot number	Description	M.W.
1,3,	Hydatid disease diagnostic antigen P 29 OS *Echinococcus granulosus*	27 079 Da
5	14-3-3 protein homolog 2 Echinococcus multilocularis	27 778 Da
11	OS *Taenia solium* GN ACTIN 1 PE 3 SV 1	41 717 Da
15	Paramyosin OS *Taenia solium* GN PMY PE 1 SV 2	98 839 Da
15	phosphoenolpyruvate carboxykinase, putative [*Schistosoma mansoni*]	71 314 Da
7	Small heat shock protein p36 *Taenia saginata*	35 572 Da

Identification of immunogenic protei
Mozambique

- cC1 and p27 proteins were not recognized in human negative sera and these proteins will be evaluated.



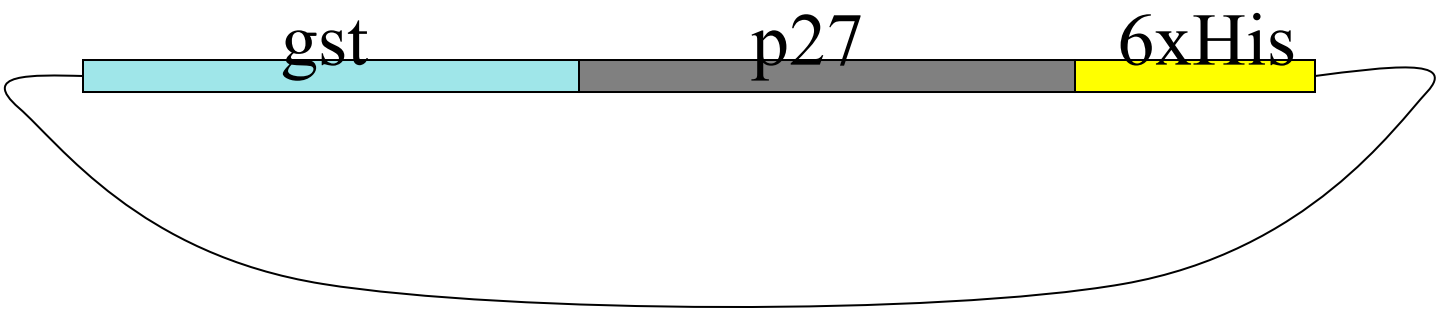
P29 become p27 *T. solium* expressed as a recombinant protein



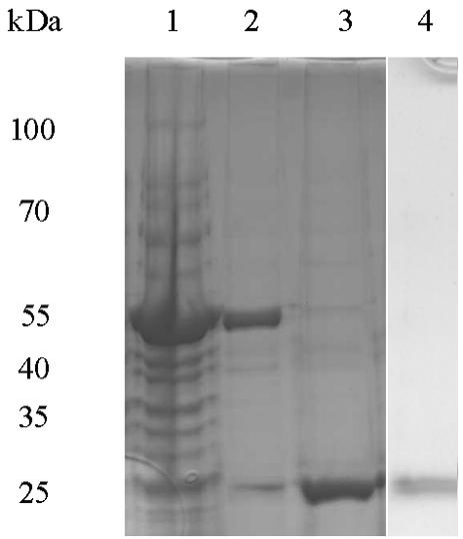
Positive Negative

sera from Nicaragua

Production of a pure recombinant protein



pGEX

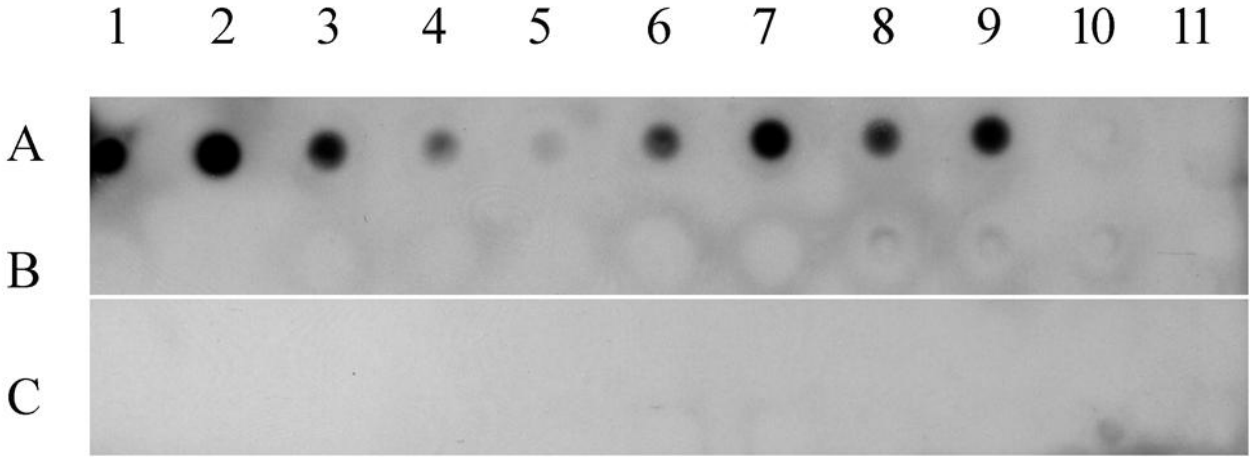


To increase the efficacy and reduce the background, we purified the Tsol-p27 in two steps using glutathione-agarose and his-selected nickel beads.

The dot blot assay

- The results obtained using our immunodot blot technique with the highly purified Tsol-p27 antigen showed a markedly decreased background compared to what could be achieved by Western blot analysis using one-step purified Tsol-p27 antigen.
- Tested with sera from > 200 people from Nicaragua
- Compared with ELISA and EITB
- Easy to use and
- Cheap
- Example

Dot blot



- (A) Probing done with sera from two NCC -positive controls (1 and 2), seven positive epileptic cases (3–9), and two negative controls (10 and 11).
- (B) Probing done with negative serum samples from epileptic cases.
- (C) Probing done with sera from patients positive (1–6) or negative (7–11) for Chagas disease.

Results, using EITB as gold standard

The immunodot blot Tsol-p27 test was comparable to the Western blot Tsol-p27 assay with respect to specificity (97.8% versus 95.6%) but not sensitivity (86.7 versus 76.4%), and it was similar to ELISA regarding both specificity (97.8% versus 94.6%) and sensitivity (86.7 versus 86.7).

Collaboration

Sweden.

Genomic projects: Scilife, KI

Jonas Lundström, Björn Anderson

Diagnostics: Swedish Institute for Comunicable Disease

Jessica Beser, Marianne Lebbad

Nicaragua

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Thank you