

Characterization of NS3 protease from an Egyptian HCV genotype 4a isolate

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Abstract: The role of the NS3 protease in HCV replication was demonstrated by the ability of a protease inhibitor cocktail (10 µg/ml) to abolish the induced cytopathic effect in RAW macrophages upon infection with Egyptian sera. The HCV protease gene was amplified from Egyptian sera by nested PCR and cloned downstream of the CMV promoter in a mammalian expression plasmid, which was then used to transform bacteria. Colonies carrying the gene in the correct orientation were subjected to large-scale plasmid purification followed by sequencing. Phylogenetic comparison of the sequence obtained with published sequences from different genotypes confirmed that our sequence belongs to genotype 4a. Of the other genotypes, the most closely related ones were from genotype 1. Multiple alignments of protease peptides showed that the catalytic triads and binding residues for substrate, Zn²⁺ and the NS4 cofactor are conserved among different isolates, including ours, and confirmed the closer homology between NS3 of genotypes 4 and 1. The HCV-protease-encoding construct was successfully transcribed in both mammalian cells and mice. Mouse antibodies produced against the protease-encoding-construct detected the 18-kDa enzyme in lysates of cells transfected with the construct by Western blotting, and in the media of infected cells by ELISA.

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Discriminant function based on hyaluronic acid and its degrading enzymes and degradation products for differentiating cirrhotic from non-cirrhotic liver diseased patients in chronic HCV infection

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Abstract

Background/aims: The invasive liver biopsy is still considered the gold standard for assessing patients with chronic hepatitis C (CHC). Our aim was to determine the operating characteristics of a non-invasive index based on blood biomarkers for the prediction of cirrhosis in CHC patients.

Methods: Hyaluronic acid level was determined by radioimmuno-assay and its degrading enzymes and degradation products were determined by standard techniques in 153 patients with CHC with and without liver cirrhosis. Statistical analyses were performed by logistic regression, and receiver-operating characteristic (ROC) curves.

Results: The multivariate discriminant analysis (MDA) selected a function based on absolute values of five biochemical markers; $\text{Score} = [1.63 + \text{Hyaluronic acid (}\mu\text{g/l)} \times 0.001 + \text{N-acetyl-beta-D-glucosaminidase (}\mu\text{mol/ml/min)} \times 0.02 + \text{glucuronic acid (}\mu\text{g/dl)} \times 0.015 + \text{glucosamine (}\mu\text{g/dl)} \times 0.006 + \text{AST/ALT ratio} \times 0.04]$. The selected MDA function correctly classified 96% of the cirrhotic patients at a discriminant cut-off score=2.5 (i.e. less than 2.5 indicated CHC without liver cirrhosis and greater than 2.5 indicated liver cirrhosis) with high degrees of sensitivity (95%) and specificity (97%). The positive predictive and negative predictive values were also high (95% and 97%, respectively).

Conclusion: A patient with CHC can be simply and efficiently classified into cirrhotic or non-cirrhotic liver diseased patient using his or her MDA score. (c) 2006 Published by Elsevier B.V.

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Immunochemical identification and detection of a 36-KDa *Toxoplasma gondii* circulating antigen in sera of infected women for laboratory diagnosis

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Abstract

The detection of *Toxoplasma gondii* circulating antigens has been indicated to be a reliable diagnostic approach of active human toxoplasmosis. However, few reports have appeared in the literature regarding the diagnostic potential of *T. gondii* circulating antigens. Here, a specific antibody and western blot analyses were used to demonstrate the presence of a highly reactive antigen of 36-kDa, not only in the extract of *T. gondii* tachyzoites, but also in selected sera of women with confirmed laboratory and clinical signs of recent toxoplasmosis. The 36-kDa *Toxoplasma* antigen was purified from *T. gondii* tachyzoites and human serum using electroelution from preparative polyacrylamide gels. The purified polypeptides showed a single peak at 10.9min when analyzed by capillary zone electrophoresis. Based on the above encouraging results, we have developed an ELISA format for the detection of target *Toxoplasma* antigen (TAg-ELISA) in human serum samples. The TAg-ELISA detected the target antigen in 88% sera of acutely infected women and showed high degree of specificity (91%) among sera from non-infected women. In conclusion, the detection of 36-kDa *Toxoplasma* circulating antigen in human sera appears to be a promising alternative approach for laboratory diagnosis of active *T. gondii* infection.

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Author(s): ARAUJO, FG; REMINGTON, JS

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Source: JOURNAL OF CLINICAL PATHOLOGY-MOLECULAR PATHOLOGY Volume: 51 Issue: 2 Pages: 105-109 Published: APR 1998

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Author(s): Attallah, AM; Karawia, EA; Ismail, H; et al.

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Author(s): Aubert, D; Maine, GT; Villena, I; et al.

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Author(s): Beghetto, E; Buffolano, W; Spadoni, A; et al.

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Author(s): Bhopale, G M; Naik, S R

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Author(s): Cozon, GJN; Ferrandiz, J; Nebhi, H; et al.

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Author(s): Denkers, EY; Gazzinelli, RT

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Author(s): Dubey, JP

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Author(s): Giraldo, M; Cannizzaro, H; Ferguson, MAJ; et al.

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Author(s): HASSL, A; ASPOCK, H

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Author(s): LINDENSCHMIDT, EG

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Author(s): LOWRY, OH; ROSEBROUGH, NJ; FARR, AL; et al.

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Author(s): Montoya, JG; Liesenfeld, O

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Author(s): Suzuki, Y; Ramirez, R; Press, C; et al.

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Author(s): TOWBIN, H; STAEHELIN, T; GORDON, J

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Author(s): TURUNEN, HJ

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Author(s): VANKNAPEN, F; PANGGABEAN, SO

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Author(s): Wilson, M; Remington, JS; Clavet, C; et al.

Source: JOURNAL OF CLINICAL MICROBIOLOGY Volume: 35 Issue: 12 Pages: 3112-3115 Published: DEC 1997

Use of a novel enzyme immunoassay based on detection of circulating antigen in serum for diagnosis of *Helicobacter pylori* infection

[Attallah, AM](#) (Attallah, AM); [Ismail, H](#) (Ismail, H); [Ibrahim, GG](#) (Ibrahim, GG); [Abdel-Raouf, M](#) (Abdel-Raouf, M); [El-Waseef, AM](#) (El-Waseef, AM); [Abdel-Wahab, M](#) (Abdel-Wahab, M)

Abstract

Recently, noninvasive diagnostic tests for *Helicobacter pylori* infection have gained in significance. We have developed a sensitive and specific noninvasive immunoassay based on the detection of an *H. pylori* circulating antigen (HpCA) in sera from *H. pylori*-infected individuals. Monospecific antibody and Western blot analyses were used to demonstrate the presence of the target antigen in *H. pylori* cell lysate and serum samples. A novel enzyme-linked immunosorbent assay (ELISA) was developed for the detection of HpCA in serum. Endoscopic biopsy specimens from the gastric antra of 221 individuals (143 males and 78 females) with dyspeptic symptoms were evaluated for *H. pylori* infection, with culture used as a "gold standard" for diagnosis. The target *H. pylori* antigen was identified at 58 kDa. HpCA has been detected by ELISA with high degrees of sensitivity, specificity, and efficiency (>90%), and ELISA results show no significant difference ($P > 0.05$) from results of *H. pylori* culture of gastric biopsy specimens. The test's positive and negative predictive values were also high (95 and 86%, respectively). In conclusion, a sensitive and specific immunoassay was developed for the detection of HpCA in human serum. This test can be applied for noninvasive laboratory and field diagnoses of *H. pylori* infection.

Source: CLINICAL AND DIAGNOSTIC LABORATORY IMMUNOLOGY Volume: 11 Issue: 4 Pages: 775-779 DOI: 10.1128/CDLI.11.4.775-779.2004 Published: JUL 2004

KeyWords Plus: CYTOTOXIN; PROTEINS; FECES; SEROLOGY; CATALASE; BINDING; ARTICLE; BREATH; STOOL; URINE

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Publisher: AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC 20036-2904 USA

Web of Science Categories: Immunology; Infectious Diseases; Microbiology

Research Areas: Immunology; Infectious Diseases; Microbiology

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Author(s): Archimandritis, A; Giontzis, A; Smilakou, S; et al.

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Author(s): Garner, JA; Cover, TL

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Author(s): Howden, CW; Hunt, RH

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Author(s): Kabir, S

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Author(s): Kokkola, A; Rautelin, H; Puolakkainen, P; et al.

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Author(s): LAEMMLI, UK

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Group Author(s): Gastrointestinal Physiology Work

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Author(s): TELFORD, JL; GHIARA, P; DELLORCO, M; et al.

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Author(s): TOWBIN, H; STAEHELIN, T; GORDON, J

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Author(s): Vaira, D; Vakil, N

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Author(s): VAIRA D

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Author(s): vanZwet, AA; Thijs, JC; Roosendaal, R; et al.

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Author(s): Westblom, TU; Bhatt, BD

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Author(s): Zhu, YL; Lin, J; Li, D; et al.

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Times Cited: 2 (from All Databases)

**Placental and oral delivery of Schistosoma mansoni antigen from infected mothers
to their newborns and children**

Attallah, AM (Attallah, AM); Ghanem, GE (Ghanem, GE); Ismail, H (Ismail, H); El Waseef, AM
(El Waseef, AM)

Abstract

A 63-kD *Schistosoma mansoni* antigen was detected in 149 (86%) of 174 umbilical cord blood sera from infected women at delivery. Specific IgG antibodies to this antigen were also detected in 80% of cord blood sera. The 63-kD antigen showed the same molecular mass by Western blotting when isolated from cord blood, maternal blood, breast milk, and urine from women infected with *S. mansoni*. This antigen was detected in the urine of 25 infants born to infected mothers and followed for 18-24 months after delivery. It was also detected in some infants up to 21 days after parturition and then disappeared at 28 days, demonstrating the influence of breast-feeding on persistence of antigen in infants born to infected women. Thus, exposure to *Schistosoma* antigens and maternal antibodies to this organism may influence the developing immune responses to natural infection or vaccination of children born in endemic areas.

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KeyWords Plus: TOLERANCE; PROTEINS; VACCINE

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