

# HEART-TYPE FATTY ACID BINDING PROTEIN VERSUS CARDIAC TROPONIN I IN EARLY DETECTION OF MYOCARDIAL INJURY INDUCED BY SCORPION STING.

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## ABSTRACT

**Background:** Scorpion envenomation is an important public health hazard in tropical and sub-tropical regions. Envenomation by scorpions can result in a wide range of clinical effects, including, cardiotoxicity, neurotoxicity and respiratory dysfunction. Biomarkers play an important role in the early diagnosis and management of patients with the cardiotoxicity related to scorpion sting. Diagnosis of myocardial injury at an early stage in the emergency department is often difficult. A recently proposed biomarker, heart fatty acid binding protein (H-FABP) has been found to appear in the circulation superior to that of cardiac troponins in the early hours of scorpion sting. **Objective:** The objective of this study was to evaluate the clinical reliability of heart-type fatty acid-binding protein (h-FABP) in identifying patients with the cardiotoxicity in the early hours following scorpion sting. **Methods:** troponin I (in laboratory) and H-FABP were performed on 246 patients who presented with scorpion sting with duration since onset ranging from 20 minutes to 2 hours. **Results:** Data of the 246 patients were analyzed. H-FABP had a higher sensitivity of 76.40% and a specificity of 96.10% compared with 58.30% and 98% for troponin I respectively (at first 6 hours). **Conclusion:** H-FABP was found to be a better biomarker of cardiac necrosis in the early hours in the diagnosis of non-conclusive ECG in patients with scorpion sting.

**Keywords:** Scorpion ; H-FABP; troponin I; myocarditis; ROC

## INTRODUCTION

Scorpion sting is a distressing endemic public health problem in several developing countries. Complications of Scorpion envenomation are cardiotoxicity, neurotoxicity and respiratory distress (Himmatrao and Pramodini, 2012).

These conditions require early diagnosis and ICU care from few hours to days. High morbidity and mortality occur due to delay in recognition, several studies in recent years revealed the accuracy and efficiency of evaluation of these patients; using ideal blood markers for early diagnosis of

myocardial injury (Bawaskar and Bawaskar, 1992).

The ideal cardiac marker should be highly sensitive and specific; rapidly rise and fall after ischemia, can be performed simply, and not affected by other organs, particularly the kidney (Abdel-Reheim, et al., 2002).

The presence of positive cardiac biomarkers indicates higher risk and worse prognosis (Ishii et al., 1997).

Early diagnosis of myocardial injury is sometimes difficult due to ambiguous electrocardiogram (ECG) changes and delayed detection of cardiac markers such as troponin and

creatine kinase (CK). Cardiac troponin I (cTnI) begins rising in the blood 4-6 hours post infarction. It peaks in 12-24 hours but may take weeks to return to normal (**Bawaskar and Bawaskar, 1992**).

Heart type fatty acid binding protein (H-FABP) is a new marker with a low molecular weight cytoplasmic protein. It is abundant in myocardial tissue; its concentration in skeletal muscle is about 10-20% of its concentration in heart muscle (**Donoghue et al., 2006**).

Several studies revealed that H-FABP is a rapid indicator for evaluation of myocardial damage in patients with scorpion sting related myocarditis. It is more sensitive and becomes positive earlier than troponins (**Glatz et al., 2002**).

Following myocardial cell damage, this small protein diffuses much more rapidly than troponins through the interstitial space (**Ayca et al., 2012**).

It appears in the circulation as early as 90 min after the onset of the symptoms, reaching its peak within six hours and remains positive for nearly 24 hours. These features make h-FABP an excellent indicator for myocardial injury (**Ghani et al., 2000**).

This prospective study was undertaken to evaluate h-FABP versus troponin I as a rapid indicator for assessment of myocardial damage in patients with scorpion sting and non-conclusive ECG.

## **MATERIAL & METHODS**

The study was conducted from January 2010 to December 2014 in Clinical toxicology Department, Faculty of Medicine, Minia university hospital. This paper highlights the diagnostic performance of h-FABP which was

compared with that of troponin I, the standard test in clinical practice. Total 246 patients suffering from Scorpion sting satisfying inclusion criteria were selected. All patients had a 12-lead ECG and biochemical markers which included troponin I and h-FABP. After the initial assessment, venous blood was drawn from patients at time of presentation and at six hours from onset of sting then after 24hrs from the onset, into plain tubes (without anticoagulants) and the serum sample was stored at -20 °C until biochemical parameters were determined.

### **Inclusion criteria**

- Patients with history of scorpion sting.
- The age below 18 years for both genders.
- Time window of admission: onset of sting within 2 hours.
- Patient with signs suggestive of envenomation including local pain of varying degrees, repeated vomiting, centripetal hyperthermia, priapism, agitation and shock.

### **Exclusion criteria**

- Above 18 years of age.
- Known renal disease (serum creatinine level 2.0 mg/dl or more): as Heart type fatty acid binding protein (H-FABP) level is usually elevated in patients with renal insufficiency, due to clearance of h-FABP through the kidney.
- Traumatic injury
- Patients who are not in position to give consent.

### **Biochemical Analyses**

Serum level of Heart type fatty acid binding protein (h-FABP) was measured using a recently developed ELISA, Oxis Research Inc, USA. The sensitivity of h-FABP assay was 17.7 ng/ml. Serum cTnI level was determined using Immulite troponin I

Kit which is a solid-phase, two-site chemiluminescent enzyme immuno-metric assay for use with the immulite automated analyzer (Cat No. LKTI, Diagn. Products Corp, USA). The detection limit of cTnI assay was approximately  $\geq 0.1$  ng/ml. The diagnostic performance of h-FABP was compared with that of troponin I, the standard test in clinical practice. The diagnosis of myocardial injury (myocarditis) was confirmed with Echocardiography (wall motion abnormalities,  $\downarrow$ ejection fraction) and positive troponin at time point 24hr from onset of sting. Data were analyzed for clinical sensitivity, specificity and negative predictive value.

#### **Statistical method**

The collected data were coded, tabulated, and statistically analyzed using SPSS program (Statistical Package for Social Sciences) software version 20. ROC curve analysis was used to determine area under the curve, sensitivity, specificity, positive predictive value, negative predictive value and accuracy. The level of significance was taken at ( $P$  value  $\leq 0.05$ ).

### **RESULTS**

A total of 246 patients (144 males and 102 females) were included in this study. ECG findings (ST segment elevation in leads II, III, Avf and V5, V6) were enrolled also in the study. Analysis of the data showed repeated troponin testing 6 hours after the onset of envenomation showed positive results in 84 patients, while Heart type fatty acid binding protein (h-FABP) diagnosed 110 stinged patients with myocardial injury as compared to 84 patients by troponin I in the initial 6-

hour period; i.e. 26 more patients were diagnosed by h-FABP.

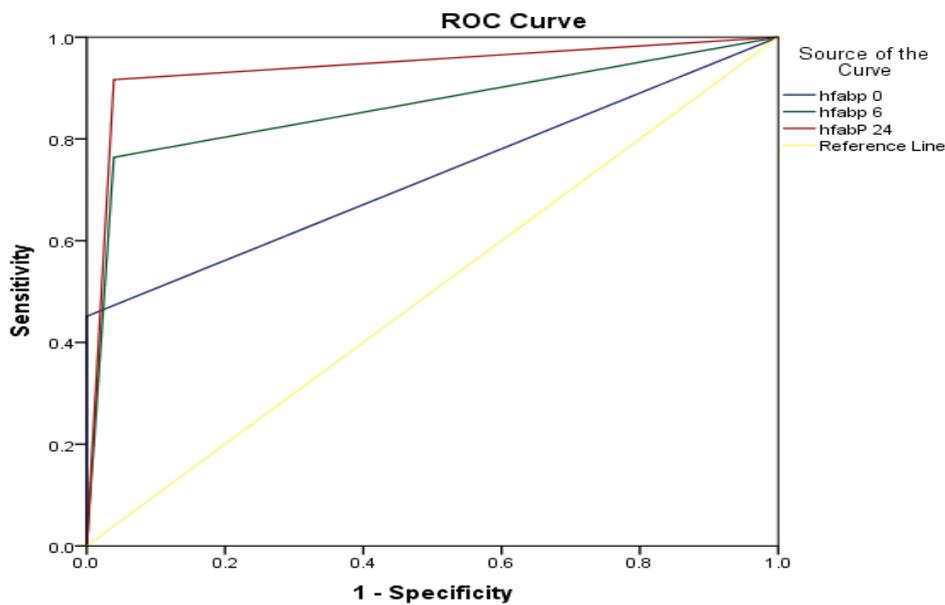
As shown in Table 1, the sensitivity and specificity of troponin I for the diagnosis of myocardial injury at hospital admission were 28.5% and 100%, respectively, the sensitivity and specificity of H-FABP were 45.1% and 100%, which were significantly higher than the sensitivity of troponin I ( $P < 0.001$ ), but there was no statistical difference in specificity between them. Also another analysis showed that the sensitivity and specificity of troponin I for the diagnosis of myocardial injury in the initial 6 hours after the onset of sting, were 58.59% and 98%, respectively, the sensitivity and specificity of H-FABP were 76.40% and 96.10% which were significantly higher than the sensitivity of troponin I ( $P < 0.001$ ). Sensitivity could have been higher if h-FABP was repeated as the other biochemical markers if the first test was negative. Heart type fatty acid binding protein (h-FABP) was found to have a higher negative predictive value when compared with that of troponin I. The negative predictive values of h-FABP and troponin I were 74.2%, % and 62.5% respectively.

Receiver Operator Characteristic Curve (ROC) analysis was done to demonstrate H-FABP and troponin I levels (Fig 1, 2). From the ROC, the optimum cut-off value which H-FABP can be considered positive was found to be 17.7 ng/ml, while the detection limit of cTnI assay was approximately  $\geq 0.1$  ng/ml. The area under the ROC curves for H-FABP was observed to be 0.862 ( $P < 0.001$ ) and it was the highest among the markers, which was significantly greater than troponin I (0.782)

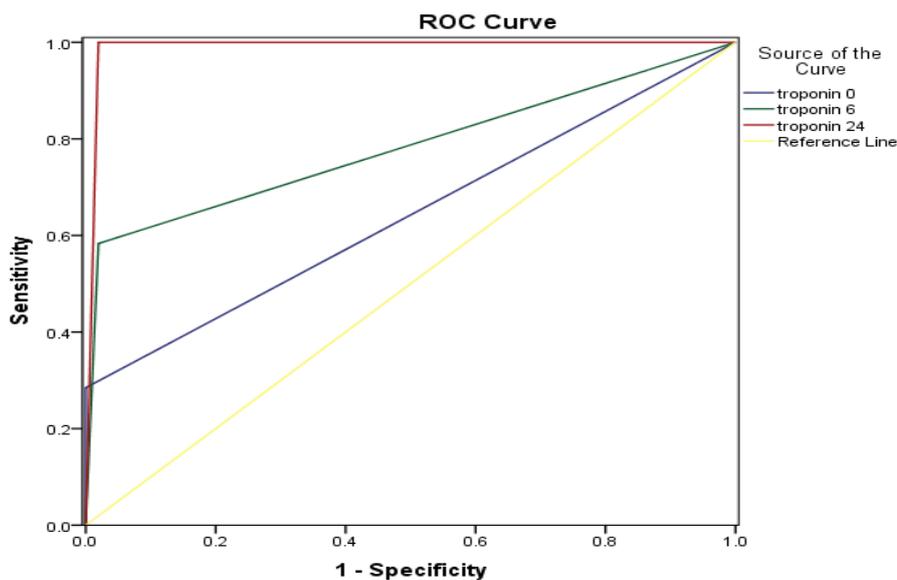
**Table (1):** Comparison of Serum Levels of H-FABP with Troponin I in Different Study Groups

	Sensitivity	Specificity	NPV	AUC	P value
<b>h-FABP 0</b>	45.1%	100%	56.4	0.726	<0.001*
<b>h-FABP 6</b>	76.4%	96.1%	74.2	0.862	<0.001*
<b>h-FABP 24</b>	91.7%	96.1%	89.1	0.939	<0.001*
<b>Troponin 0</b>	28.5%	100%	49.8	0.642	<0.001*
<b>Troponin 6</b>	58.3%	98%	62.5	0.782	<0.001*
<b>Troponin 24</b>	100%	98%	100	0.990	<0.001*

Area under the Curve (AUC),  
Negative Predictive Value (NPV)



**Figure (1) :**Receiver Operator Characteristic Curve (ROC) Analysis for the assay of H-FABP



**Figure (2):** Receiver Operator Characteristic Curve (ROC) Analysis for the assay of cTnI.

## **DISCUSSION**

Scorpion venom is a complex mixture of neurotoxins, cardiotoxins, nephrotoxins and haemolysins. Venom can cause myocardial damage by several pathogenetic mechanisms, coronary artery vasospasm by release of histamine, serotonin and leukotrienes) (Ho-pang et al., 2009), toxic myocarditis by reduction of Na-K-ATPase (Rahav and Weiss, 1990), release of allergenic proteins causes anaphylactic shock leading to hypotension, also Scorpion venom inhibits angiotensin converting enzyme (ACE), resulting in pulmonary oedema (Bawaskar and Bawaskar, 1999).

Despite the development of assays for highly sensitive and specific biochemical markers of myocardial necrosis, myocardial injury remains a challenge. A sensitive and specific earlier marker of myocardial cell injury might be helpful in treatment decisions (Ayca et al., 2012).

The principal characteristics that would make an ideal marker for early detection of myocardial injury include, a small size of molecular marker is more rapidly released into the circulation allowing early detection of myocardial damage, also its absence in the circulation under physiological conditions, thus its detection will be abnormal with even minimal increase of the marker in the plasma and absolute specificity for the myocardium (Donoghue et al., 2006).

H-FABP's advantage is dominant in the early phase of myocardial infarction due to: its smaller size and weight, its abundance in the heart tissue and its release and clearance kinetic (Okamoto et al., 2000; Tsuji et al., 1993).

The combination of a sensitive marker such as h-FABP for early detection, with a cardio-specific marker such as troponin for later confirmation can be recommended to provide optimal diagnostic performance, to minimize the risk of false exclusions of patient with myocarditis (Katrukha et al., 1999).

Our results demonstrate that the sensitivity of h FABP is higher when compared to other biochemical markers troponin by approximately 18% and around the same specificity in comparison with other biomarkers in the early hours. In our study, the sensitivity of Troponin I at hospital admission was 28.5%, which was very low compared with previous reports in which sensitivities ranged from 38.1% to 100%, (Sayre et al., 1998; Ruzgar et al., 2006; Zarich et al., 2001; Hetland et al., 1996).

Sensitivities of H-FABP assays in patients at hospital admission and 6 hours after the onset of sting were significantly higher than those of Troponin I assays, but sensitivities of H-FABP were significantly lower than sensitivities of Troponin I 24 hours after the onset of sting, which means H-FABP are much more useful than Troponin I in patients admitted at hospital and 6hours from the onset of sting in detection of myocardial Injury. The sensitivity of H-FABP was the highest markers in patients at hospital admission and 6 hours after the onset of sting, due to its low specificity. This result is similar to the results that showed the sensitivity and specificity of H-FABP were 89% and 52% in patients admitted within 2 hours from the onset of symptoms (Seino et al., 2003), also the sensitivity and specificity of H-FABP were 89.7% and

72.2% within 3 hours after onset of symptoms (**Okamoto et al., 2000**) and the sensitivity and specificity of H-FABP were 73% and 71% within 4 hours after onset of symptoms (**McCann et al., 2008**).

As our results were demonstrating an H-FABP area under the curve of 0.862 with 76.4% sensitivity and 96.1% specificity, so our results were in complete agreement with the above studies demonstrating an area under the curve of 0.965 with 87% sensitivity and 93% specificity. These results indicate the fact that H-FABP is well suited for early detection of myocarditis associating scorpion sting. Another study not agree with the above results which reported an increased area under the curve for Troponin I than H-FABP, as H-FABP levels were measured at a later period, whereas H-FABP is useful especially in the early hours after infarction (**Glatz et al., 2002**).

Heart-type fatty acid-binding protein (h-FABP) supplies diagnostic safety in the first and decisive hours. Heart-type fatty acid-binding protein (h-FABP) was found to be a superior biomarker of cardiac necrosis in the diagnosis of myocardial infarction due to its higher myocardial content and low plasma concentration (1.8g/l).

### **CONCLUSION**

Heart-type fatty acid-binding protein (h-FABP) is a superior biomarker of cardiac necrosis in the diagnosis of scorpion sting with toxic myocarditis in patients in the early hours due to higher sensitivity and higher negative predictive value in comparison with other biomarkers, namely troponin I.

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الملخص العربي**دور الاحماض الدهنيه المرتبطه ببروتين القلب H-FABP ضد التروبونين في تشخيص اصابه عضله القلب في الساعات الأولى في المرضى الذين يعانون من لدغة العقرب****\*عمر و رضا زكي ، \*\*مصطفى ابو طالب احمد****\* قسم الطب الشرعي والسموم الاكلينيكيه كليه الطب البشري – جامعه بني سويف.****\*\* قسم الطب الشرعي والسموم الاكلينيكيه كليه الطب البشري – جامعه المنيا.**

تمثل لدغه العقرب خطرا على كبريا على الصحة العامة في المناطق المدارية الاستوائية وشبه الاستوائية. من الممكن ان تؤدي لدغه العقرب إلى مجموعة واسعة من الآثار السريرية مثل اختلال وظيفي في الجهاز الدوري والعصبي و التنفسي. المؤشرات الحيوية تلعب دورا هاما في التشخيص المبكر في المرضى الذين يعانون من اختلال وظيفي في الجهاز الدوري عقب حدوث لدغة العقرب. تشخيص إصابة عضلة القلب في مرحلة مبكرة في قسم الطوارئ غالبا ما يكون صعبا. فقد تم اكتشاف احماض دهنيه مرتبطه ببروتين القلب H-FABP ، ووضح تفوقه على التروبونين في التشخيص خلال الساعات الاولي من لدغة العقرب. وكان الهدف من هذه الدراسة هو تقييم مدي كفاءه الاحماض الدهنيه المرتبطه ببروتين القلب في تحديد المرضى الذين يعانون من مشكله في عضلة القلب في الساعات الأولى لللدغة العقرب. تم اجراء تحليل التروبونين الأول و H-FABP على 246 مريضا الذين حضروا الي المستشفى بداعي لدغة عقرب في مده تتراوح من 20 دقيقة إلى ساعتين. وقد تم تحليل النتائج الخاصه ب 246 مريضا: اوضحت النتائج الاتي ان حساسيه وخصوصيه H-FABP هي 76.40% و 96.10% وهي اعلي مقارنة مع 58.30% و 98% للتروبونين (في أول 6 ساعات من حدوث اللدغه). الملخص : تم التوصل الي ان H-FABP من افضل العلامات البيولوجية في تشخيص اصابه القلب في الساعات الأولى في المرضى الذين يعانون من لدغة العقرب.