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REVIEW ARTICLE

The Prognostic Value of Heart Type Fatty Acid Binding Protein in Patients with Suspected Acute Coronary Syndrome: A Systematic Review

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Abstract: ***Background*:** Heart type fatty acid protein (HFABP) is a cytosolic protein released early after acute coronary syndrome (ACS) even in the absence of myocardial necrosis.

***Objectives*:** The purpose of this systematic review was to determine whether HFABP levels in patients with suspected, or confirmed ACS, improve risk stratification when added to established means of risk assessment.

***Methods*:** We searched Medline, Pubmed and Embase databases from inception to July 2015 to identify prospective studies with suspected or confirmed ACS, who had HFABP measured during the index admission with at least 1 month follow up data. A prognostic event was defined as all-cause mortality or acute myocardial infarction (AMI).

***Results*:** 7 trials providing data on 6935 patients fulfilled inclusion criteria. There were considerable differences between studies and this was manifest in variation in prognostic impact of elevated HFABP(Odds ratio range 1.2-15.2 for death). All studies demonstrated that HFABP provide unadjusted prognostic information and in only one study this was negated after adjusting for covariates. A combination of both negative troponin and normal HFABP conferred a very low event rate. No study evaluated the incremental value of HFABP beyond that of standard risk scores. Only one study used a high sensitive troponin assay.

***Conclusion*:** There was marked heterogeneity in prognostic impact of HFABP in ACS between studies reflecting differences in sampling times and population risk. Prospective studies of suspected ACS with early sampling of HFABP in the era of high sensitivity troponin are necessary to determine the clinical value of HFABP. HFABP should not currently be used clinically as a prognostic marker in patients with suspected ACS.

Keywords: Acute coronary syndromes, biomarkers, prognosis, systematic review, HFABP, AMI.

1. INTRODUCTION

Risk stratification is crucial to the appropriate management of patients with Acute Coronary Syndrome (ACS) [1]. Current validated methods of risk stratification include the Global Registry of Acute Coronary Events (GRACE) and Thrombolysis In Myocardial Infarction (TIMI) risk scores, both of which use the presence of elevated biomarkers as an adverse risk factor [2]. Studies have demonstrated that incorporating novel non-necrosis biomarkers into standard risk scores can improve risk prediction [3, 4]. Heart Type Fatty Acid Binding Protein (HFABP) is a small cytosolic protein primarily responsible for the transport of long chain fatty acids, which is released rapidly into the serum during myocardial infarction [5, 6]. A number of analyses have demonstrated improved risk stratification of suspected ACS patients when HFABP is used as a biomarker [7].

2. OBJECTIVES

The purpose of this systematic review was to determine the absolute prognostic value of HFABP levels in patients with suspected or confirmed ACS, the incremental prognostic value beyond standard risk stratification and troponin levels, and the clinical utility of measuring HFABP in these patients.

3. METHODS

A systematic review was conducted with adherence to the Preferred Reporting Items for Systematic Reviews and Meta-Analysis (PRISMA) 2009.

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4. STUDY ELIGIBILITY

We included studies investigating the prognostic role of HFABP in patients with suspected or confirmed ACS. Pre-specified inclusion criteria were prospective studies (including post hoc analysis from prospective studies) with human adult patients who had HFABP measured during the index admission with at least 1 month follow up data.

5. STUDY DEFINITIONS

A prognostic event was defined as all-cause mortality or Acute Myocardial Infarction (AMI).

6. SEARCH STRATEGY

The primary search was performed using NHS Evidence to identify suitable English language articles from inception to July 2015 from Medline, Pubmed and Embase databases. The search included HFABP in association with ACS, angina and coronary disease as key terms. Only human studies were allowed. Review articles were not included. Studies were excluded in the absence of complete or potentially extractable data considered necessary to determine the prognostic value of HFABP in patients with suspected or confirmed ACS. Results in abstract-only and poster format were not included. All studies identified had their references hand-searched and scrutinised in order to identify other potential studies for inclusion.

7. DATA EXTRACTION

The primary literature search was performed by an in-house clinical information specialist trained in literature searches and a clinician (JJ). The literature search, scrutiny of abstracts and relevant full texts of all identified studies was undertaken by two authors (JJ and RD) independently and without cross-reference. Disagreements in inclusion or exclusion of articles between the two authors were adjudicated by a 3rd researcher (AK) by reference to the inclusion criteria detailed above.

8. QUALITY OF STUDIES

Each study was assessed for its quality by two reviewers (JJ, AK) using the American Heart Association guidance for the evaluation of novel markers of cardiovascular risk (Table **1**), [8] and, by other quality markers determined by the authors (Table **1**).

The quality of studies was evaluated as to the presence of a clearly defined aim (of determining the prognostic value of HFABP in ACS), if the population studied was similar to a 'real life' suspected ACS population and the appropriateness of sample timing.

Table 1. Study quality based on criteria for evaluation of a novel biomarker.

Novel marker reported:	Reiter [9]	Viswanathan [12]	McCann [11]	Ilva [14]	Kilcullen [7]	O'Donoghue [13]	Ishii [10]
In accordance with STROBE [19]	++	++	++	+	++	++	++
a) Standard RF, and	+++	+++	+++	+++	+++	+++	+++
b) results of risk model using established factors	-	+++	++	++	+++	+++	+++
a) RR, OR, HR with CI/p value	-	+++	+++	-	+++	+++	+++
b) RR, OR, HR adjusted for RF and CI/p value	+++	+++	+++	-	+++	+++	+++
c) p value for addition of novel marker to standard risk markers.	-	+++	+++	-	+++	-	+++
a) C-index and CL for model with established risk markers	+	+	-	-	-	-	+
b) C-index and CL for model including novel and established risk markers	+	+	-	-	-	-	+
c) Discrimination index/slope or binary R2 for model with and without novel marker.	-	-	-	-	-	-	-
d) Graphic display of predicted cases before and after inclusion of the marker.	-	-	-	-	-	-	-
a) Display observed *vs.* expected event rates without/ with the novel risk marker.	-	-	-	-	-	-	-
b)using generally recognised risk thresholds, subjects reclassified and event rates in reclassified groups	-	-	-	-	-	-	-
Clearly defined aim	Good	Fair	Good	Good	Good	Good	Good
'real-life' population	Good	Good	Good	Good	Fair	Fair	Good
Appropriate sampling period for HFABP release	-	Yes	Yes	Yes	Yes	No	-

+++ Complete adherence, ++reasonable adherence, +partial adherence, - does not report
RF risk factors, CI Confidence interval, CL confidence limits.

9. RESULTS

9.1. Study Selection

The primary search and cross referencing identified 276 manuscripts (Fig. **1**). From the articles identified, 269 were excluded from the review. The majority of papers were excluded as the article was not addressing an ACS population or no prognostic information was provided. See Fig. (**1**) for full list of exclusion criteria. 118 full text articles were reviewed in order to obtain the final 7 trials providing data on 6,935 patients for this systematic review (Fig. **1**).

9.2. Quality of Studies

The majority of studies had clearly defined aims. Reiter *et al*. [9], Ishii *et al*. [10] and McCann *et al*. [11] were prospective observational studies with pre-specified aims. Viswanathan *et al*. [12] had the clear aim of establishing the prognostic value of HFABP in patients with suspected ACS, with intended focus on the low to intermediate risk patients. However patients were recruited, regardless of initial risk/troponin levels. O'Donoghue *et al*. [13] Kilcullen *et al*. [7] and Ilva *et al*. [14] were post-hoc analyses.

9.3. Timing of Sample Acquisition

At the onset of myocardial infarction HFABP rises rapidly and reaches peak levels within 4 to 8 hours, then falls rapidly and return to baseline within 24 hours [15-17]. Each study obtained samples for HFABP at different times (Table **2**). Reiter *et al*. [9] and Ishii *et al*. [10] did not report the timing of sampling. Kilcullen *et al*. [7] measured HFABP between 12-24 hours after symptom onset; samples taken nearer to 24 hours may lead to an underestimation of the risk associated, as levels may have returned to normal by the time sampling had occurred. O'Donoghue *et al*. [13] were those who reported the timings of samples on patients who clearly fall outside of this initial rise of HFABP in ACS; any correlation with HFABP and prognosis in this study is more likely to represent a different pathophysiology, such as ongoing ischaemia.

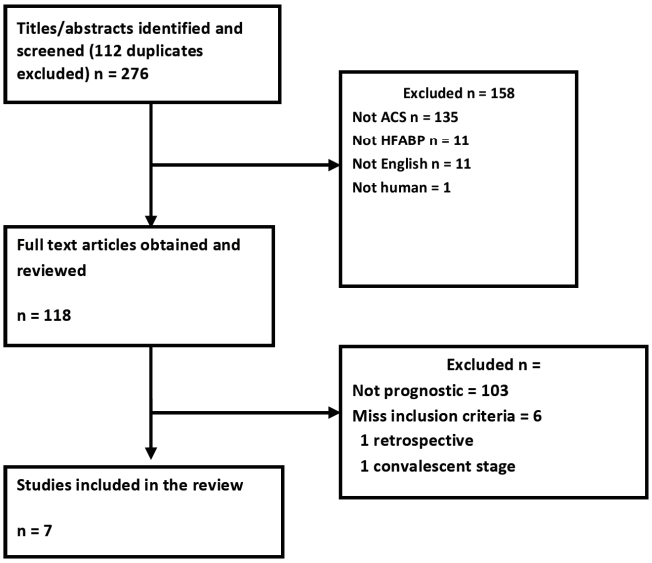

Fig. (1). Flowchart: search strategy and relevant yield of studies included in systematic review.

Table 2. Patient characteristics.

Author	Age (years) [mean± SD/ mean range-range] unless stated	Male (%)	Index Diagnosis (%)					Past Medical History (%)						
			AMI	NSTEMI	STEMI	UA	Non ischaemic	Previous MI	DM	Smoking	HTN	Hchol	CHF	CRF
Reiter [9]	Med 64 (IQR 51-76)	67	20	16	4	11	14*	25	19	34	64	45	NR	10
Viswanathan [12]	60.01±15	60.5	20.8	20.8	0	79.2	0	30.5	15.1	24.4	61.2	NR	5.1	NR
McCann [11]	62±13	70.2	52.9	33.6	19.2	26	21.1	36.9	17.3	68.2	54.6	76.6	NR	3.6
Ilva [14]	67.1	61.8	42.0	22.9	19.1	NR	4.1	30.7	17.4	18.8	43.7	58.4	10.2	NR
Kilcullen [7]	Med 72.5± 13	61	87.7	62.9	24.8	12.3	0	27.1	16.9	26.1	NR	NR	7.6	NR
O'Donoghue [13]	38.74% ≥ 65 years	71.9	54.9	22.5	32.4	45.1	0	NR	21.6	35.9	41.9	28.3	5.2	NR
Ishii [10]	64.9 ±10.4	80.5	73.5	NR	47	NR	0	17.7	32.0	54.3	54.9	44.2	0.9	NR

Abbreviations: NR = not reported, AMI= acute myocardial infarction, NSTEMI- non st segment elevation myocardial infarction, STEMI= st segment elevation myocardial infarction, UA= unstable angina, DM= diabetes mellitus, HTN= hypertension, HChol= hypercholesteraema, CHF= chronic heart failure, CRF = chronic renal failure, Med= median, *refers to cardiac non-coronary disease.

Table 3. Study design and endpoints.

Author [reference]	Study design	N	Patients included	Troponin assay	HFABP assay	Time of HFABP sample	Follow up	Study end points relevant to review
Reiter [9]	Prospective observational study	1074	Suspected ACS	Roche. HS cTnT.	Quick-SenshFABP assay	NR	12 months	All-cause mortality
Viswanathan [12]	Prospective observational study	955	Suspected ACS	Siemens Healthcare Diagnostics. Advia TnI /Advia TnI Ultra.	Randox Laboratories	< 12 hours post symptom onset	12 months min; median 18 months	Composite of death and readmission with MI
McCann [11]	Prospective observational study	550	Suspected ACS	Roche, Elecsys, Troponin T mmunoassay	Human H-FABP ELISA test kit, Hycult, biotechnology	Median time 6 hours post symptom onset	12 months	Composite of death and MI
Ilva [14]	Post hoc analysis	293	Suspected ACS	Abbott Diagnostic Division. Architect STAT Troponin I assay.	Innotrac Diagnostics (experimental assaty)	0-24 hours post symptom onset	6 months	Death and readmission with MI
Kilcullen [7]	Post hoc analysis	1448	Confirmed ACS EMMANCE-2 study	Beckman Coulter. Accu TnI assay.	Dainippon Pharmaceutical	12-24 hours post symptom onset	12 months	Death
O'Donoghue [13]	Post hoc analysis	2287	Confirmed ACS OPUS-TIMI 16.	Biosite Diagnostics. Troponin I assay.	Dainippon Pharmaceutical	41±20 hours post randomisation.	10 months	Death, MI
Ishii [10]	Prospective observational study	328	Confirmed ACS	Roche Diagnostics. Elecsys Troponin T immunoassay.	Dainippon Pharmaceutical	NR	6months	Cardiac death and MI

NR = not reported. Revasc = coronary revascularisation, CP= chest pain, CHF= chronic heart failure, MI= myocardial infarction.

9.4. Trial Population and Demographics

Study and participant characteristics were extracted (Table **2, 3**). The trial population was mostly male, in common with most clinical trials. There was considerable variability in the subtype of ACS between studies, reflected in the differences in inclusion criteria. All studies included patients with myocardial infarction and Viswanathan *et al*. [12] alone excluded those with STEMI. Reiter *et al*. [9] and McCann *et al*. [11] were those who specifically described HFABP and outcome in those with a final diagnosis of non-cardiac chest pain. Other clinical characteristics appeared similar between studies. Each study had differing durations of follow up and end points (Table **3**).

9.5. Biomarker Assays

There are currently no international analytical standards for HFABP analysis. However, each study except for those by McCann *et al*. [11] and Ilva *et al*. [14] provided information regarding the precision of the HFABP analysis (Table **3**). Troponin assays varied between studies. There has been considerable change in troponin assays over recent years, particularly with the development of higher sensitive troponin assays. High sensitive troponin are generally understood to be those with a coefficient of variation of 10% or less at the 99th percentile with the ability to detect cardiac troponin in at least 50% of the reference population [18, 19]. Only the study by Reiter *et al.* used troponin assays (Hs Tn T [Roche]) that fulfilled this definition.

9.6. Prognostic Impact of Elevated HFABP

All included studies demonstrated that HFABP provides (unadjusted) prognostic information (Table **4**). The studies analysing the end points mortality and combined mortality/AMI found higher levels of HFABP were associated with a worse prognosis. All three studies by Mccann [11], O'donoghue [13] and Ishi [10] analysing the end point of

Table 4. **Main study outcomes for entire study cohort.**

Study [ref], year	N	Duration of follow up	Coronary Revascularisation rates	End point	No of events	HFABP cut off for analysis	Events in 'lower' HFABP/total in group	Events in 'higher' HFABP/total in group	Unadjusted risk for 'higher' HFABP levels	Adjusted risk for 'higher' HFABP levels	Covariates used
Reiter [9] 2013	1074	12 months	Not specified	Death	-	5.76µg/L.	-	-	-	aHR 1.017 (1.007 – 1.029) p = 0.002	Age, gender, cardiovascular risk factors
Viswanathan [12] 2010	955	>12 months	8.1%(inpatient revascularisation noted only)	Death or MI	96	6.48µg/l	48/838	48/117	uRR 7.16 (5.05-10.17) p<0.0001 uOR 11.45 (7.16-18.31) p <0.0001	aHR 2.62 (1.3-5.28) p = 0.007	Age, DM, HTN, previous HF, previous MI, admission HR, ST Depression, Creatinine, Tn
McCann [11] 2009	550	12 months	PCI 38% CABG 8%	Death	29	5µg/l	-	-	uOR 21.2 (2.9 – 157.3) p = 0.003	aOR 10.5 (1.4-80.6) p=0.023	Age, gender, risk factors, cardiac history, SBP, Killip class, ECG, eGFR, WCC, Tn, investigational biomarkers.
				Death or MI	54				uOR 5.4(2.4-12.2) p<0.001	aOR 2.7 (1.1-6.4) p = 0.028	
				MI	31				Not significant	Not significant	
Ilva [14] 2008	293	6 months	Not specified	Death or MI	43	10.4µg/l	18/183	25/110	uRR* 2.31 (1.32-4.04) p = 0.0033 uOR* 2.7 (1.39 – 5.22 p = 0.0032	Not significant	Age, gender, DM, Chol, HTN, Smoker, prev MI, Prev revasc, Killip Class, ST deviation, Tn.
Kilcullen [7] 2007	1448	12 months	PCI 7.4% CABG 2.6% (both inpatient revascularisation noted only)	Death	296	5.8µg/l	11/305	285/1143	uRR* 6.91 (3.84 – 12.46) p < 0.0001 uOR* 8.88 (4.79 – 16.45) p <0.0001	-	
O'Donoghue [13] 2006	2287	30 days	PCI for index event 33.9%	MI	-	8µg/l	-	-	uHR 1.9 (1.04-3.4) p=NR	-	Demographics, clinical characteristics, time to randomisation, index diagnosis, creatinine clearance, ST deviation, Biosite Tn
		10 months		Death	102		61/1955	41/332	uHR 4.1 (2.6-6.5) p<0.001	aHR 2.7 (1.5-4.9) p=NR	
		10 months		MI	140		109/1955	31/332	uHR 1.6 (1-2.5) p=0.053	-	
Ishii [10], 2005	328	30 days	PCI 55.5% CABG 5.2%	Cardiac death	14	9.8µg/l	1/164	13/164	uRR* 13 (1.72 – 98.24) p = 0.0129 uOR* 14.03 (1.81 – 108.57) p = 0.0114	-	Age, gender, time from chest pain, STEMI, Tn, Creatinine, Killip class, Anterior AMI, previous MI.
		30 days		Death or MI	18		3/164	15/164	uRR* 4.7 (1.38 – 16.03) p = 0.0136 uOR* 5.42 (1.53 – 19.03) p = 0.0087	-	
		6 months		Cardiac death	15		1/164	14/164	uRR 14.5 (1.91 – 110.5) p = 0.009	aRR 8.92 (1.15 – 69.4) p = 0.04	
		6 months		Death or MI	25		3/164	22/164	uRR 7.7 (2.3 – 25.7) p = 0.0009	aRR 8.96 (2.64 – 30.4) p = 0.0004	
		6 months		MI	10		2/164	8/164	uRR* 4 (0.86-18.55) p=0.076		

*Calculated by author using raw data. - = Not reported.

Table 5. Outcome according to absolute value ranges of HFABP.

Study [ref], year	Covariates included	Risk of higher HFABP in patients with ACS
Viswanathan [12], 2010	Unadjusted	Death or MI HR (95% CI) according to HFABP level HFABP 0.15-3.26 µg/l HR 1, p = <0.001. HFABP 3.27-6.48 µg/l HR 3.41 (1.89 – 6.16) p = 0.001 HFABP 6.49-12.77 µg/l HR 15.67 (8.16-30.07) p = <0.001. HFABP 12.78-151.0 µg/l HR 20.37 (10.38-40.00) p =<0.001.
Kilcullen, [7] 2007	Unadjusted	All-cause mortality HR (95% CI) according to HFABP level HFABP < 6.38µg/l HR 1. HFABP 6.38-12.39µg/l HR 4.45 (2.47-8) p <0.001. HFABP 12.39 – 36.2µg/l HR 8.78 (4.99-15.46) p < 0.001. HFABP > 36.2µg/l HR 11.69 (6.67-20.49) p <0.001.
O Donoghue [13], 2006	Unadjusted	Death rate 10 months (p<0.001) HFABP <8 µg/l 3.1% HFABP 8-16 µg/l 6.9% HFABP >16 µg/l 18% MI rates 10 months (p=0.009) HFABP <8 µg/l 5.6% HFABP 8-16 µg/l 5.5% HFABP >16 µg/l 13.8%
Viswanathan, [12] 2010	Age, DM, HTN, previous HF, Previous MI, HR, ST depression, creatinine, troponin.	Death or MI median 18 month HFABP 0.15 – 3.26 µg/l HR 1, p = 0.003. HFABP 3.27 – 6.48 µg/l HR 0.78 (0.39 – 1.55) p = 0.48. HFABP 6.49 – 12.77 µg/l HR 2.62 (1.30 – 5.28) p = 0.007. HFABP 12.78 – 151.0 µg/l HR 1.54 (0.55 – 4.32) p = 0.41.
Kilcullen, [7] 2007	GRACE risk factors, and inpatient PCI and HS CRP.	HFABP quartiles adjusted for GRACE risk factors plus hs-CRP with TnI as continuous variable. HFABP < 6.38µg/l HR 1. HFABP 6.38-12.39µg/l HR 2.32 (1.25 – 4.30) p = 0.007. HFABP 12.39 – 36.2µg/l HR 3.17 (1.73 – 5.82) p < 0.001. HFABP > 36.2µg/l HR 4.88 (2.67 – 8.93) p <0.001.

acute MI in isolation, found that there was no statistically significant association with HFABP levels, although there was a trend towards significance in the study by O'Donoghue *et al*. [13] (RR 1.7, p=0.053). All studies demonstrated a strong linear relationship between HFABP level, when categorised, in subgroups rather than as a dichotomous variable, and the hazard ratio of hard endpoints (Table **5**).

Analyses were undertaken in all studies to determine the incremental value of abnormal HFABP by adjusting for a range of covariates (Table **6**). No authors looked directly at the incremental value of HFABP beyond calculated traditional risk scores such as the GRACE or TIMI score. Kilcullen *et al*. [7] used GRACE variables plus high sensitivity C reactive protein (hs-CRP) with troponin I as a continuous variable but did not define incremental values against a summative GRACE score. Viswanathan *et al*. [12], Kilcullen *et al*. [7] and O'Donoghue *et al*. [13] demonstrated that HFABP retains prognostic power even when troponins are incorporated in the multivariable regression model.

Troponin and HFABP provided complimentary risk information in the studies by Kilcullen *et al*. [7] and Reiter *et al*. [9]. Both Kilcullen *et al*. [7] and Reiter *et al*. [9] demonstrated the incremental value of HFABP beyond troponin. Both studies [7, 9] revealed zero to 6-month mortality if both HFABP and troponin levels were within normal limits. In patients with raised troponin levels, Kilcullen *et al*. [7] demonstrated elevated HFABP was associated with a 25% mortality over 12 months, compared with <5% mortality for those with HFABP levels within normal range. In comparison, Reiter *et al*. [9] discovered a mortality of 20% and 3% over 2 years in those with elevated HFABP versus HFABP in the normal range respectively. In patients with normal troponin levels, Kilcullen *et al*. [7] demonstrated that an elevated HFABP was associated with a 20% mortality at 1 year (compared to <3% annual mortality with normal HFABP levels). This contrasts with the study by Reiter *et al*. [9] who discovered that HFABP did not differentiate risk in patients with normal troponin levels at 2 years.

Table 6. Unadjusted and covariate adjusted risk of elevated HFABP.

Unadjusted risk			
Subgroup	**Study**	**Risk associated with 'higher' HFABP levels in patients with normal troponin levels**	
Normal troponin levels	Viswanathan, 2010	Death or MI HR (95% CI) according to HFABP level HFABP 0.15-3.26 μg/l HR 1 HFABP 3.27-6.48 μg/l HR 3.46 (1.69 – 7.10) p = 0.001 HFABP 6.49-12.77 μg/l HR 11.20 (4.95-25.36) p <0.001. HFABP 12.78-151.0 μg/l HR 16.64 (2.21-125.51) p = 0.006. HFABP >5.3 ♀ and >5.8 ♂ HR 6.57 (3.05 – 14.11) p 0.0001 Subgroup analysis of 384 patients with additional admission sample taken for HFABP HR 5.08 (1.84 – 14.07) p = 0.002	
Normal troponin levels	Kilcullen, 2007	HFABP < 6.38μg/l HR 1. HFABP 6.38-12.39μg/l HR 6.50 (1.53 – 27.71) p = 0.011 HFABP 12.39 – 36.2μg/l HR 5.79 (1.08 – 31.12) p = 0.041 HFABP > 36.2μg/l HR unable to calculate, as no deaths.	
Covariate adjusted risk			
Subgroup	**Study**	**Covariates included**	**Risk**
Normal troponin levels	Viswanathan, 2010	Age, DM, HTN, HF, previous MI, HR, ST depression, Creatinine, troponin.	HFABP >5.8μg/l adjusted HR (for GRACE Risk Factors and Hs CRP and tnI as continuous variable) aHR 11.35 (2 – 64.34, p = 0.006). Death and MI Adjust HR (age and creatinine): HFABP 0.15 – 3.26 μg/l HR 1, p = 0.01. HFABP 3.27 – 6.48 μg/l HR 1.55 (0.72 – 3.36) p = 0.26 HFABP 6.49 – 12.77 μg/l HR 3.12 (1.11 – 8.76) p = 0.03. HFABP 12.78 – 151.0 μg/l HR 16.67 (2.19 – 127.06) p = 0.007.
Normal troponin levels	Kilcullen, 2007	GRACE risk factors, HS CRP and TnI as additional continuous variable	All-cause mortality HR if HFABP > 5.8 μg/L HR 11.35 (2 – 64.34).
STEMI	Kilcullen, 2007	GRACE risk factors, HS CRP and TnI as additional continuous variable	no deaths in the group with HFABP levels ≤5.8 μg/l. 77 deaths in STEMI subgroup, all HFABP levels >5.8 μg/l. HR assessment not possible
STEMI	Ishii	Age, gender, time from onset chest pain, increased cTnT, creatinine, Kilip class >1, anterior AMI, previous history of MI.	Cardiac death or MI if HFABP >9.8 μg/L RR 11.3, 1.41 – 90.6, p=0.02
NSTEMI	Kilcullen, 2007	GRACE risk factors, HS CRP and TnI as additional continuous variable	All cause mortality aHR 3.11 (1.45 – 6.7) p = 0.004.
NSTEMI	Ishii, 2005		For both UA and NSTEMI patients HFABP >9.8μg/L – aRR 8.31 (1.76 – 39.1) p = 0.007

MI = Myocardial infarction.

Three studies analysed outcome by absolute HFABP values rather than as a dichotomous variable. They revealed increasing risk with increasing HFABP values. They revealed unadjusted and adjusted prognostic outcome of subgroups of ACS according to HFABP levels (Table **6**). Prognostic information is present across the entire spectrum of ACS.

9.7. Coronary Revascularisation

Coronary revascularisation rates could confound prognostic assessment of biomarkers including HFABP. Reiter *et al*. and Ilva *et al*. [9, 14] did not describe revascularisation rates (Table **4**). Of the others only O'Donoghue *et al*. [13] and Kilcullen *et al*. [7] described revascularisation rates according to HFABP level. There was no apparent difference in coronary revascularisation rates between HFABP subgroups in these 2 studies. However, there was a numerically lower revascularisation rates in the highest quartile of HFABP compared to the lowest quartile in the study by Kilcullen *et al*. [7].

9.8. Assessment of Heterogeneity and Publication Bias Inclusion Criteria (Fig. 2)

Study end points and length of follow up and method of reporting varied considerably between studies, making pooling of data or direct comparison difficult. For the purpose of visually assessing the effect sizes between studies we constructed a funnel plot. The odd ratios for the mortality associated with elevated HFABP are illustrated in Fig. (**2**) from 6 of the 7 studies where individual mortality data was able to be discriminated [7, 9, 10, 12-14]. The point estimate with 95% confidence intervals in Fig. (**2**) was derived from a weighted combination of risk derived from normal versus elevated HFABP populations in these 6 studies. (Review-manager 5.3, Cochrane informatics and knowledge management department). There is an evidence of marked heterogeneity in odds ratio with these studies. The very large confidence limits with data by Ishii *et al.* [10] and McCann *et al*. [11] reflect mainly a small number of events and imply uncertainty over the true hazard risk associated with an abnormal HFABP. The odd ratios of death conferred by an abnormal HFABP in the study by Kilcullen *et al*. [7] and O'Donoghue *et al*. [13] appear more robust with narrower confidence intervals. This difference in odds ratio and the marked differences in point estimates outside the 95% confidence interval in these studies is more likely a reflection of methodological differences and population risk rather than publication bias.

10. DISCUSSION

We have systematically reviewed the role of HFABP as a prognostic biomarker in patients with suspected ACS. As far as we are aware, no previous such analysis has been undertaken to gain further insight into the potential clinical utility of HFABP as a prognostic biomarker for suspected ACS. We report 3 major findings from this systematic review: The evidence for the use of HFABP as a biochemical marker for risk stratification in acute coronary syndromes is weak with heterogeneous studies and lack of consistency in both timing of measurement post-acute coronary syndrome and precision of assay used. Its incremental value beyond 5th generation high sensitive troponins has been evaluated in only one study. Currently there is insufficient evidence to consider its use as a prognostic marker in acute coronary syndrome.

Heart-type fatty acid-binding protein (hFABP) is a small soluble cytosolic protein involved in the transportation of long-chain fatty acids into the cardiomyocyte. It may enter the vascular system directly *via* endothelium because of its small size. It is released rapidly into the circulation in response to cardiomyocyte injury. Due to its solubility, HFABP can be released more rapidly than structurally bound molecules like cardiac troponins and therefore it is an early marker of myocardial ischaemia (for rule-out myocardial infarction in combination with troponins) [20]. However, it is not certain whether release into the circulation in the event of myocardial ischaemia/ necrosis is earlier than high sensitive troponins. HFABP raised in acute myocardial ischaemia even in the absence of myocyte necrosis (troponin negative-4th generation troponin) and therefore it is proposed as a powerful prognostic tool in acute coronary syndrome (and in particular unstable angina) [7, 12].

All of the studies evaluated in this systematic review, indicate that HFABP does provide some prognostic information in patients with suspected ACS to varying degrees. Only one of the seven studies concluded that the significance of this prognostic information was not present after adjusting for covariates.

However, before contemplating the merits of HFABP, it is important to consider the parameters necessary to consider

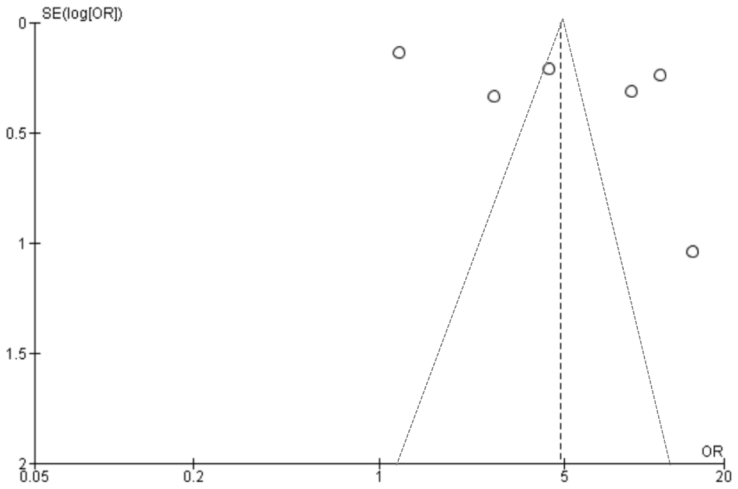

Fig. (2). Funnel plot of Standard error of odds ratio against odds ratio of death with elevated HFABP in [suspected] ACS ('weighted' point estimate of 6 studies [7, 9, 10, 12-14]).

a biomarker as a clinically useful and cost-effective tool in clinical practise. Statistical methods including odds ratios, risk ratios and hazard ratios may not be the most appropriate technique for determining the clinical utility of a biomarker. The desirable features for a prognostic biomarker of atherosclerotic cardiovascular disease have already been proposed [21]. Although HFABP appears to add to the clinical assessment of patients, it is not known whether HFABP can alter management or lead to an improvement in health outcome

Perhaps the most clinically interesting aspects identified in this review was the incremental value of HFABP beyond troponin as demonstrated by Kilcullen *et al*. [7] and Reiter *et al*. [9]. HFABP elevation in the presence of a normal troponin may reflect myocardial ischaemia and could lead to greater potential myocardial salvage if an early interventional strategy is adopted. Patients with normal troponin and HFABP levels appear to predict a very low risk population group, which may benefit from early hospital discharge. However, both of these hypotheses are untested.

Studies investigating whether HFABP leads to risk reclassification beyond internationally recognised validated risk scores and contemporary high sensitivity troponin assays, would be welcomed by the authors. Moreover, randomised studies comparing the measured health outcome for patients who have HFABP determined and receive an intervention tailored to HFABP levels, with those who do not have HFABP measured and receive standard intervention would be required before the assay can be considered for routine clinical practice.

10.1. Limitations

We conducted an extensive comprehensive review, nevertheless a number of limitations persist. The primary search was performed using extremely large and reliable databases, which leads to the potential introduction of database bias. Differences in the designs of the trials, with differing sampling intervals and cut-of points, precluded a meta-analysis or an easy summation of evidence thus reducing the overall power of this analysis.

10.2. Publication Bias

Fig. (**2**) and Table **4** suggest marked heterogeneity in odds ratio relative risk with elevated HFABP for our outcome measures. This could indicate publication bias with a preponderance of 'positive studies' for HFABP. However it could also be explained by a difference in methodology, timing of samples and troponin assays used.

CONCLUSION

Each of the studies identified in this review concluded that HFABP provides some prognostic information in patients with suspected or confirmed ACS. Only one of the seven studies concluded that the significance of this prognostic information was not present after adjusting for covariates. The data suggest that regardless of the subtype of ACS, patients with 'high' HFABP levels are at higher risk of death or myocardial infarction at any time during the follow-up compared with those with 'low' HFABP levels. However, there is insufficient evidence to currently recommend its uptake, as a clinical tool, in decision-making patients with suspected ACS. Additional studies, particularly randomised control studies are required to investigate the outcome for early discharge of patients who are high sensitive troponin negative and HFABP negative against standard care (high sensitive troponin alone with clinical judgement and ECG). Also it would be prudent to investigate intermediate risk patients with randomisation to an invasive strategy in event of HFABP positivity against routine care. Such studies would add greatly to the evidence base and also allow a determination of cost-effectiveness of a HFABP 'enabled' strategy.

POTENTIAL CONFLICTS OF INTEREST

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REFERENCES

[1] Anderson JL, Adams CD, Antman EM, *et al*. ACC/AHA 2007 guidelines for the management of patients with unstable angina/non-ST-Elevation myocardial infarction: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Writing Committee to Revise the 2002 Guidelines for the Management of Patients With Unstable Angina/Non-ST-Elevation Myocardial Infarction) developed in collaboration with the American College of Emergency Physicians, the Society for Cardiovascular Angiography and Interventions, and the Society of Thoracic Surgeons endorsed by the American Association of Cardiovascular and Pulmonary Rehabilitation and the Society for Academic Emergency Medicine. J Am Coll Cardiol 2007; 50: e1-157.

[2] D'Ascenzo F, Biondi-Zoccai G, Moretti C, *et al*. TIMI, GRACE and alternative risk scores in Acute Coronary Syndromes: a meta-analysis of 40 derivation studies on 216,552 patients and of 42 validation studies on 31,625 patients. Contemp Clin Trials 2012; 33: 507-14.

[3] Widera C, Pencina MJ, Meisner A, *et al*. Adjustment of the GRACE score by growth differentiation factor 15 enables a more accurate appreciation of risk in non-ST-elevation acute coronary syndrome. Eur Heart J 2012; 33: 1095-104.

[4] Eggers KM, Kempf T, Venge P, Wallentin L, Wollert KC, Lindahl B. Improving long-term risk prediction in patients with acute chest pain: the Global Registry of Acute Coronary Events (GRACE) risk score is enhanced by selected nonnecrosis biomarkers. Am Heart J 2010; 160: 88-94.

[5] Storch J, Thumser AE. The fatty acid transport function of fatty acid-binding proteins. Biochim Biophys Acta 2000; 1486: 28-44.

[6] Glatz JF, van BM, Paulussen RJ, Veerkamp JH, Van der Vusse GJ, Reneman RS. Release of fatty acid-binding protein from isolated rat heart subjected to ischemia and reperfusion or to the calcium paradox. Biochim Biophys Acta 1988; 961: 148-52.

[7] Kilcullen N, Viswanathan K, Das R, *et al*. Heart-type fatty acid-binding protein predicts long-term mortality after acute coronary

syndrome and identifies high-risk patients across the range of troponin values. J Am Coll Cardiol 2007; 50: 2061-7.

[8] Hlatky MA, Greenland P, Arnett DK, *et al*. Criteria for evaluation of novel markers of cardiovascular risk: a scientific statement from the American Heart Association. Circulation 2009; 119: 2408-16.

[9] Reiter M, Twerenbold R, Reichlin T, *et al*. Heart-type fatty acid-binding protein in the early diagnosis of acute myocardial infarction. Heart 2013; 99: 708-14.

[10] Ishii J, Ozaki Y, Lu J, *et al*. Prognostic value of serum concentration of heart-type fatty acid-binding protein relative to cardiac troponin T on admission in the early hours of acute coronary syndrome. Clin Chem 2005; 51: 1397-404.

[11] McCann CJ, Glover BM, Menown IB, *et al*. Prognostic value of a multimarker approach for patients presenting to hospital with acute chest pain. Am J Cardiol 2009; 103: 22-8.

[12] Viswanathan K, Kilcullen N, Morrell C, *et al*. Heart-type fatty acid-binding protein predicts long-term mortality and re-infarction in consecutive patients with suspected acute coronary syndrome who are troponin-negative. J Am Coll Cardiol 2010; 55: 2590-8.

[13] O'Donoghue M, de Lemos JA, Morrow DA, *et al*. Prognostic utility of heart-type fatty acid binding protein in patients with acute coronary syndromes. Circulation 2006; 114: 550-7.

[14] Ilva T, Lund J, Porela P, *et al*. Early markers of myocardial injury: cTnI is enough. Clin Chim Acta 2009; 400: 82-5.

[15] Kleine AH, Glatz JF, Van Nieuwenhoven FA, Van der Vusse GJ. Release of heart fatty acid-binding protein into plasma after acute myocardial infarction in man. Mol Cell Biochem 1992; 116: 155-62.

[16] Glatz JF, Kleine AH, Van Nieuwenhoven FA, Hermens WT, van Dieijen-Visser MP, Van der Vusse GJ. Fatty-acid-binding protein as a plasma marker for the estimation of myocardial infarct size in humans. Br Heart J 1994; 71: 135-40.

[17] Van Nieuwenhoven FA, Kleine AH, Wodzig WH, *et al*. Discrimination between myocardial and skeletal muscle injury by assessment of the plasma ratio of myoglobin over fatty acid-binding protein. Circulation 1995; 92: 2848-54.

[18] Reichlin T, Hochholzer W, Bassetti S, *et al*. Early diagnosis of myocardial infarction with sensitive cardiac troponin assays. N Engl J Med 2009; 361: 858-67.

[19] Keller T, Zeller T, Peetz D, *et al*. Sensitive troponin I assay in early diagnosis of acute myocardial infarction. N Engl J Med 2009; 361: 868-77.

[20] Body R, Carley S, McDowell G, *et al*. The Manchester Acute Coronary Syndromes (MACS) decision rule for suspected cardiac chest pain: derivation and external validation. Heart 2014; 100: 1462-8.

[21] Vasan RS. Biomarkers of cardiovascular disease: molecular basis and practical considerations. Circulation 2006; 113: 2335-62.