The Role of Wisteria Floribunda (M2BPGi) Serum Level for Diagnosing Liver Fibrosis in Hepatitis B Patient: An Evidence Based Case Report

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ABSTRACT

Aim: *To assess mac-2-binding protein glycosylation isomer (M2BPGi) serum performance compared to liver biopsy in diagnosisng liver fibrosis*

Method: *Literature search using Pubmed®, Ebsco®, ProQuest®, Scopus®, Clinical Key databases, and the Cochrane Library® yield four relevant and full-text articles. The four articles were critically appraised for its validity, importance, and applicability.*

Results: *Sensitivity and specificity in all four studies showed that M2BPGi serum was inadequate to ruling in and ruling out the diagnosis of liver fibrosis in chronic hepatitis B patients. The difference in M2BPGi cut-off value to determine the stage of fibrosis in each study makes this value cannot be used as an accurate standard to determine the advanced stage (F≥3) of liver fibrosis. On the other hand, M2BPGi serum combined with other tests are known to improve the diagnostic accuracy.*

Conclusion: *MBP2Gi serum cannot be used as a diagnostic modality for detecting liver fibrosis in chronic hepatitis B patients.*

Keywords: *hepatits B, fibrosis, mac-2-binding protein glycosylation isomer (M2BPGi), wisteria floribunda, liver biopsy*

ABSTRAK

Tujuan: *Untuk menilai performa diagnosis mac-2-binding protein glycosylation isomer (M2BPGi) dibandingkan dengan biopsi hati.*

Metode: *Pencarian literatur dengan menggunakan database Pubmed®, Ebsco®, ProQuest®, Scopus®, Clinical Key, dan Cochrane Library® yang menghasilkan empat artikel relevan dan full-text. Keempat artikel tersebut ditelaah dengan menggunakan kriteria validity, importance, dan applicability untuk menentukan derajaat kegunaan dalam studi ini.*

Hasil: *Sensitivitas dan spesifisitas pada keempat studi menunjukkan bahwa pemeriksaan M2BPGi masih belum memadai untuk mendeteksi (ruling in) dan menyingkirkan (ruling out) diagnosis fibrosis hati pada pasien hepatitis B kronik. Perbedaan nilai cut off M2BPGi untuk menentukan stadium fibrosis pada tiap studi, membuat nilai tersebut belum dapat langsung dijadikan standar yang akurat untuk menentukan stadium lanjut (F ≥ 3) fibrosis hati. Di sisi lain, pemeriksaan M2BPGi yang dikombinasikan dengan pemeriksaan lain diketahui meningkatkan akurasi diagnostik.*

Simpulan: *MBP2Gi belum dapat dijadikan sebagai modalitas diagnosis untuk mendeteksi fibrosis hati pada pasien hepatitis B kronik.*

Kata kunci: *hepatitis B, fibrosis, mac-2-binding protein glycosylation isomer (M2BPGi), wisteria floribunda, biopsi hati*

INTRODUCTION

Liver fibrosis is an excessive accumulation of extracellular matrix proteins which causes liver tissue turns into scar tissue, causing a decrease in liver function. Liver fibrosis occurs as a result of chronic damage to liver tissue, one of which is caused by chronic hepatitis B.[1] It has become a global concern because more than 240 million people in the world are infected to this virus.[2] 15% to 40% individuals who are untreated can progress to cirrhosis, which may lead to liver failure and liver cancer.[3] Liver fibrosis severity can be categorized into F1-F4 staging.[1] Hence, a clinician must conduct a useful diagnostic method to determine this condition.

Liver biopsy is the gold standard for diagnosing liver fibrosis and its staging. This method provides a clear picture of histopathology analysis. The biopsy is considered to be a gold standard because of its high accuracy.[4] However, liver biopsy is an invasive diagnostic procedure, with 40% incidence of pain in patients and 0.5% incidence of significant complications.[5] Also, sampling errors can also occur primarily in small tissue.[6]

At present, there have been several studies showing that mac-2 binding protein glycosylation isomer (M2BPGi) serum or wisteria floribunda can diagnose liver fibrosis.[7-10] M2BPGi is a serum secreted by the liver during the development of fibrosis. Using M2BPGi serum will reduce the risk of complications and shorten the time in diagnosing liver fibrosis. This report aims to determine the diagnostic performance of M2BPGi/wisteria floribunda in diagnosing liver fibrosis compared to liver biopsy.

CASE ILLUSTRATION

A 25 years-old male came to a hospital four months ago with the chief complaint of epigastric abdominal pain. He described that he often feels nauseous and sometimes vomit. There was also loss of appetite and an enlargement in the stomach. However, the patient never had yellow eyes dan brownish urine. During his adolescence, he used syringe drugs for about three years.

Laboratory and PCR examination results are HBsAg (+), SGOT 155 U/L, SGPT 120 U/L, total bilirubin 0.84 mg/dL, direct bilirubin 0.13 mg/dL, and HBV-DNA (+). The doctor who treated him defines hepatitis B virus infection as his diagnosis. Hence the doctor ordered a liver biopsy examination to determine the patient liver's fibrosis level. However, the patient refused to have a biopsy because it is an invasive procedure. A friend of the patient's who visited him in the hospital told him that in Japan, where she lived, there was a new serum biomarker for detecting liver fibrosis. The doctor decided to study the new biomarker using the evidence-based medicine method.

ANSWERABLE CLINICAL QUESTION

In young adult patients who suspected having liver fibrosis in chronic hepatitis B infection, does M2BPGi serum biomarkers provide accurate and faster results when compared to biopsy methods in detecting liver fibrosis?

METHOD

The article searching was conducted on PubMed®, Ebsco®, ProQuest®, Scopus®, Clinical Key, and Cochrane® using the keywords "chronic hepatitis", "M2BPGi", "biopsy", and "fibrosis" along with its synonyms which is combined using Boolean operations (Table 1). After the article search process completed, then the article selection process is carried out by screening title abstract, filtering duplication, and eligibility criteria as shown in the flowchart (Figure 1).

Table 1. Article searching strategy (done in September 19th, 2018)

Database	Search terms	Results
PubMed	((chronic hepatitis) AND ((M2BPGi OR (wisteria floribunda)))) AND ((biopsy OR (liver histology) AND ((fibrosis OR (fibrosis degree)))	31
Ebsco	((chronic hepatitis) AND ((M2BPGi OR (wisteria floribunda)))) AND ((biopsy OR (liver histology) AND ((fibrosis OR (fibrosis degree)))	18
ProQuest	((chronic hepatitis) AND ((M2BPGi OR (wisteria floribunda)))) AND ((biopsy OR (liver histology) AND ((fibrosis OR (fibrosis degree)))	48
Scopus	((chronic hepatitis) AND ((M2BPGi OR (wisteria floribunda)))) AND ((biopsy OR (liver histology) AND ((fibrosis OR (fibrosis degree)))	9
Clinical Key	chronic hepatitis AND (M2BPGi OR (wisteria floribunda)) AND Fibrosis	1
Cochrane	chronic hepatitis AND (M2BPGi OR (wisteria floribunda)) AND Fibrosis	0

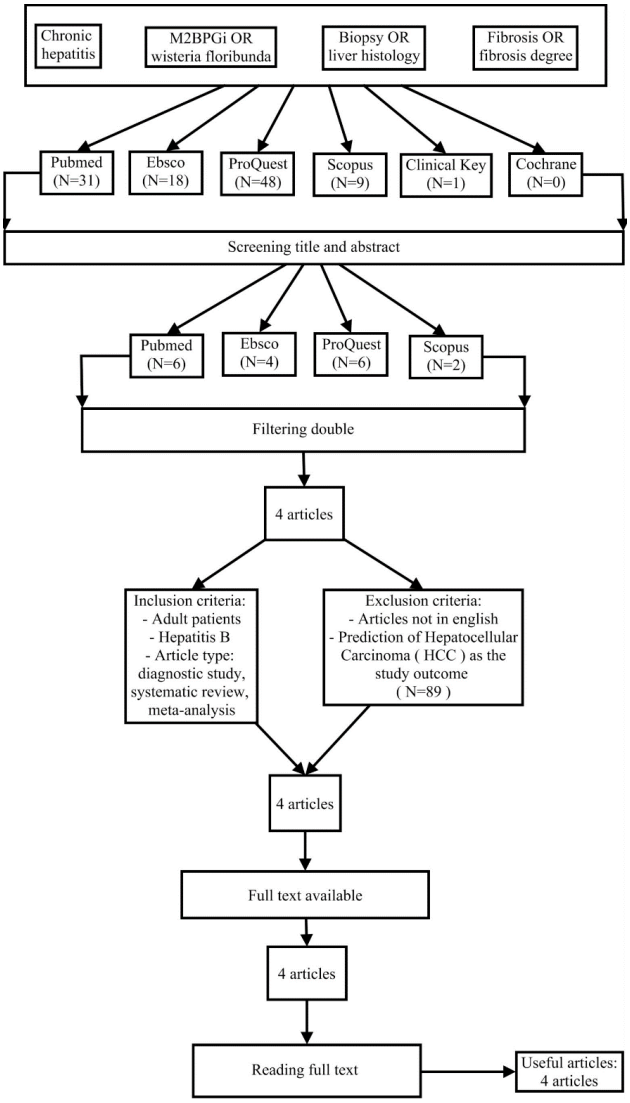

Figure 1. Flow of Article Searching (done in September 19th, 2018)

After the selection, critical appraisal was done using critical appraisal kit for diagnostic study and systematic review based on Center for Evidence-based-medicine (CEBM) University of Oxford.

RESULTS

In Mak L et al study, there are 327 chronic hepatitis B patients with 54 patients have stage 3 or more liver fibrosis (≥ F3). The scoring system which is used to differentiate the stage of liver fibrosis is Ishak scoring system. The optimal cut off for serum M2BPGi is 0.45 cut off index (COI) with sensitivity value 69.6%, specificity value 74.1%, positive predictive value (PPV) 23.1%, and negative predictive value (NPV) 95.5%.[7]

In Jekarl DW et al study, there are 151 chronic hepatitis B patients with 15 patients have stage 3 liver fibrosis (F3). The scoring system which is used to differentiate the stage of liver fibrosis is Knodell scoring system. Cut off for serum WFA-M2BP is 0.7 with sensitivity value 73.3%, specificity value 62.5%, PPV 66.1%, and NPV 70.1%.[8]

In Nakamura M et al study, there are 91 chronic hepatitis B patients with 20 patients have stage 3 liver fibrosis (F3). The scoring system is also Knodell. The cut off value for serum WFA(+)-M2BP is >1 COI with sensitivity value 67%, specificity value 70%, PPV 53%, and NPV 81%. Also in this study, a combination of serum WFA(+)-M2BP and serum miR-122 is perfomed which results in an increase of PPV and specificity value to 64% and 87%.[9]

In Ito K et al meta-analysis, 21 studies were written from January 2013 to September 2016. All these studies are in accordance with the determined eligibility criteria and sourced from the PubMed/ Medline and Cochrane databases.[10] The quality of each study in this meta-analysis is assessed based on the quality assessment of studies of diagnostic accuracy included in systematic review (QUADAS-2). From

Table 2. Validity, relevance, and level of evidence

			Validity			Importancy				Applicability
						Statistic value				
Author	Year of pub.	Level of evidence	Representative spectrum patients	Application reference standard	Independent Blinding Conduct	Sens. (%)	Spec. (%)	PPV	NPV	Sufficient Details of Conduct
Mak et al	2018	2b	+	+	?	69,9	74,1	23,1	95,5	+
Jekarl et al	2018	2b	+	+	+	73,3	62,5	66,1	70,1	+
Nakamura et al	2017	2b	+	+	-	67	70	53	81	+

+ yes – no ? not mentioned

Author	Year of pub.	Level of evidence	PICO	Important studies imculed	Appropriate eligibility criteria	Validity of studies	Result similarity	Result's forest plot
Ito et al	2017	1	+	-	+	+	-	+

+ yes – no ? not mentioned

total 21 studies (4772 patients), there are 5 studies of hepatitis B patients (1066 patients) with 163 patients have stage 3 liver fibrosis (F3). The METAVIR evaluation system is used to differentiate the stage of liver fibrosis in this study. The mean sensitivity and specificity values of WFA^+-M2BP levels for stage 3 liver fibrosis (F3) are 0.615 and 0.709. The cut off values for detecting hepatitis B patients' WFA^+-M2BP levels are different for each study including 1.42 in Nishikawa study (2016), 1.40 in Ishii study (2016), and 1.26 in Ichikawa study (2016).

DISCUSSION

All four studies exhibit similarities in terms of patient spectrum and the reference test used. Patients in all studies also exhibit variety in terms of age range and stages of liver fibrosis diagnosed (ranging from F0 to F4), with biopsy and histopathological grading as the golden standard for diagnosis. However, unlike the other two studies, it was not clear whether blinding was conducted in the study by Mak et al and Nakamura et al, increasing the risk of bias in those two studies.[7,9]

Table 3. Characteristics of literatures

Literature	Number of samples	Characteristics of samples	Index test	Reference test	Outcome	Advantages	Disadvantages
Mak et al[7]	327 (≥F3 n=54)	Mean age 38.1 years old, men (70%), HbeAg + (62.1%), advanced fibrosis or early stage of cirrhosis (15.3%)	HISCL M2BPGi reagent kit on automated immunoanalyzer HISCL-800 (Sysmex, Hyogo, Japan)	Liver biopsy	Diagnosis of stage ≥3 liver fibrosis in chronic hepatitis B patient	- Detect the effects of antiviral to M2BPGi levels, - Examination is clearly presented and can be applied - Large sample size - Selection bias is minimal because of randomization - Asian population	M2BPGi test is not certain to be done blindy
Jekarl et al[8]	151 (F3 n=15)	Mean age 40 years old, men (72.5%), and HBeAg-positive (78%)	Automated chemiluminescence enzyme immunoassay analyzer (HISCL-5000, Sysmex, Kobe, Japan)	Liver biopsy	Diagnosis of stage ≥3 liver fibrosis in chronic hepatitis B patient	- Examination is clearly presented and can be applied - Large sample size - Selection bias is minimal because of consecutive admissions - Liver biopsy and M2BPGi test are performed at one time (blind) - Asian population	There are exclusion criterias which make hepatitis B patients are not all examined
Nakamura et al[9]	91 (F3 n=20)	Mean age 40 years old (between 25-68 years old), men (72.5%), HbeAg + (78%)	HISCL M2BPGi assay kit (Sysmex Co., Kobe, Japan)	Liver biopsy	Diagnosis of stage ≥3 liver fibrosis in chronic hepatitis B patient	- Examination is clearly presented and can be applied - Selection bias is minimal because of consecutive admissions - Asian population	- Small sample size - Not all samples are examined by both liver biopsy and M2BPGi test
Ito et al[10] (meta-analysis)	21 studies (4772 patients) with 5 studies about hepatitis B (942 patients and 163 F3)	- Ishii et al.: 62% men, mean age 44 years old - Ichikawa et al.: 64% men, mean age 47 years old - Heo et al.: 73% men, mean age 51 years old - Nishikawa et al.: 62% men, mean age 16 years old - Zou et al.: 68% men and mean age 38 years old (*training*); 62% men and mean age 37 years old (*validation*)	HISCL M2BPGi assay kit	Liver biop-sy	Diagnosis of stage ≥3 liver fibrosis in chronic hepatitis B patient	- The aim of meta-analysis is clear - Large sample size - The quality of studies is measured by QUADAS-2 criteria - Evaluate the ability of WFA^+-M2BP to predict HCC and survival rate of HCC patients - Forest plot is understand-able	- There are inclusion and exclusion criterias which make literatures of hepatitis B patients and WFA^+-M2BP examination can not be used entirely - Not only Asian population - There is a significant heterogeneity (evaluated by Q Cochran test)

In diagnosing advanced liver fibrosis (≥ F3) among chronic hepatitis B patients, serum WFA-M2BPGi in all four studies show similar yet rather poor sensitivity and specificity (61,5-73,3% and 62,5-74,1%, consecutively). In other words, advanced liver fibrosis was successfully detected in only 61.5-73.3% patients and ruled out in only 62.4-74.1% patients, while the remaining patients were incorrectly diagnosed (false negative or false positive). This indicates that M2BPGi has yet to be a good modality in ruling on nor ruling out advanced liver fibrosis in chronic HBV patients.

Nevertheless, study by Nakamura et al shows an increase in M2BPGi's specificity to 87% when used in combination with serum microRNA-122 examination in diagnosing advanced liver fibrosis. This shows that combining M2BPGi with other diagnostic test may improve the overall sensitivity and specificity in diagnosing advanced liver fibrosis.[9]

In addition, studies by Mak et al, Jekari et al, and Nakamura et al show a variation in the PPV and NPV of M2BPGi (23.1-66.1% and 70.1-95.5% respectively). The lowest PPV (23.1%) and the highest NPV (95.5%) were found in the study by Mak et al, but this was due to the small sample size for the stage F ≥ 3 compared to other stages. Furthermore, despite being conducted in Asia (Korea and Japan), none of the studies actually states the prevalence of advanced liver fibrosis in chronic HBV patients in their respective countries. Comparison to the prevalence in Indonesia hence cannot be made and the predictive values cannot be used as reference in determining the diagnostic value of M2BPGI in the Indonesian population.[7,8,9]

Despite these findings, it is worth mentioning that the histopathological scoring system used differs in each study. Jekari et al and Nakamura et al used the Knodell scoring system, while Ito et al and Mak et al used METAVIR and Ishak scoring system respectively.[7,8,10] Although these scoring systems overlap, there might be some discrepancies that affect the overall sensitivity and specificity. The cut-off values of serum M2BPGi used to determine each stage of liver fibrosis in each study also differs. Stage F ≥ 3 was defined by the cut-off values of 1.26, 1.40, and 1.42 in the meta-analysis by Ito et al., ≥ 0,45 COI in the study by Mak et al, ≥0.7 COI in the study by Nakamura et al, and ≥ 1 COI in the study by Nakamura et al. Scoring system and the range of cut-off values used need to be standardized in future studies to obtain more accurate sensitivity and specificity.

CONCLUSION

WFA-M2BPGi is novel serum marker currently being developed to diagnose various stages of liver fibrosis in chronic hepatitis B patients. Despite its growing availability in Indonesia, particularly in Cipto Mangunkusumo Hospital in Jakarta, based on the available studies, serum M2BPGi is yet to be a good modality in ruling in and ruling out advanced liver fibrosis (F ≥ 3) due to its rather poor sensitivity and specificity. The studies appraised also used different histopathological scoring systems as reference tests and varying, non-standardized M2BGPi cut-off values for each staging, compromising the accuracy of its sensitivity and specificity. However, combining serum M2BPGi with other examination may increase the overall sensitivity and specificity in diagnosing advanced liver fibrosis, as when M2BPGi was used in combination with serum microRNA-122 examination.

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