GAVIN PUBLISHERS

Journal of Digestive Diseases and Hepatology

Liu YY, et al. J Dig Dis Hepatol: JDDH-139

Research Article

DOI: 10.29011/2574-3511. 000039

Renal Injury During Long-Term Oral Antiviral Therapy in Chronic Hepatitis B (CHB)

Ya Yun Liu[1], Wei Liao[1], Qian He[2], Ai Jing Xu [1], Jian Ya Xue[1], Cheng Zhong Li [1], Xue Song Liang[1*]

[1]Department of Infectious Diseases, Changhai Hospital of Navy Military Medical University, Shanghai, China

[2]Department of Health Statistics, Navy Military Medical University, Shanghai, China

[*]**Corresponding author:** Xue Song Liang, Associate Professor, Department of Infectious Diseases, Changhai Hospital of Navy Military Medical University, Shanghai, China. Tel: +8602131161902; Email: liangxuesong2000@163.com

Citation: Liu YY, Liao W, He Q, Xu AJ, Xue JY, et al. (2018) Renal Injury During Long-Term Oral Antiviral Therapy in Chronic Hepatitis B (CHB). J Dig Dis Hepatol: JDDH-139. DOI: 10.29011/2574-3511. 000039

Received Date: 25 November, 2017; **Accepted Date:** 10 January, 2018; **Published Date:** 17 January, 2018

Summary

Aim: To determine the incidence and clinical outcome of long-term nucleot(s)ide analogues (NAs) usage related renal injury in patients with chronic hepatitis B (CHB).

Methods: A real-time prospective observation study was carried out between March 2014 and May 2017. 255 patients with CHB, which have been receiving one or two types NAs for antiviral therapy for more than six months, were enrolled. Serum and urinary β_2 microglobulin (β_2 MG), serum phosphorus and calcium level, serum creatinine (Scr) and estimated glomerular filtration rate (eGFR) were determined for all patients at the enrollment and every 6 months during follow-up. Patients were followed up for 52 weeks.

Results: At the enrollment 27(10.58%)NAs long-term treated CHB patients showed mild or moderate eGFR value decrease, but the mean change of eGFR value among different therapy groups had no significant difference (p=0.925). But at the enrollment 135 (52.55%) NAs long-term treated CHB patients showed serum or urinary β2 MG level abnormality. Among different NAs groups the abnormal rate of urinary β2 MG level of ADV group was the highest and was significantly higher than that of LDT group [69.88% Vs14.81%, p=0.002].Multivariate analysis found that ADV (OR = 1.07, 95% CI=0.38-3.04, p=0.0016) and LDT (OR=0.10, 95% CI=0.03-0.40, p=0.0011) were both independent factors for urinary-β2 MG abnormality rate. Age (OR=1.02, 95% CI=1.00-1.05, p=0.00478) was also the independent risk factors for urinary β2 MG abnormality rate. For eGFR level change, age was the only independent risk factor (t=4.48, p<0.0001) and LDT was the unique drug which related to eGFR change (t=2.33, p=0.0204). At the end of 52-weeks' follow-up urinary β2MG level of patients in ADV-excluding therapy group decreased at an average of 3.94mg/L, and eGFR level increased at an average of 1.7 ml/minute. But for patients in ADV-including group, urinary β2MG level increased at an average of 1.41 mg/L and eGFR level decreased at an average of 5.66 ml/min.Furthermore, at the end of 52-weeks' follow-up the urinary β2MG level in only 7(11.36%) patients recovered completely.

Conclusion: During long-term NAs anti-HBV therapy, CHB patients developed mainly proximal tubular damage. For monitoring NAs related subclinical renal injury low-molecular-weight protein markers such as urinary β2MG was more sensitive than eGFR level. Prolonged NAs especially ADV antiviral therapy may be accompanied with irreversible renal damage even renal failure.

Keywords: Antiviral Therapy;Chronic Hepatitis B (CHB);Estimated Glomerular Filtration Rate (eGFR);Hepatitis B Virus (HBV);Immunohistochemical;Nucleoside/Nucleotide Analogue (NAs); Renal Tubular; Serum creatinine (Scr);Urinary β2microglobulin (β2 MG)

Introduction

Chronic Hepatitis B Virus (HBV) infection is one of the main causes of chronic liver injury. Around the world about 2 billion people has been infected, and more than 3500 million people were chronic HBV carriers [1,2]. Development of anti-HBV Nucleoside/Nucleotide Analogues(NAs) in the last twenty years have changed the history of Chronic Hepatitis B(CHB) treatment [3,4]. To now five NAs have been approved for CHB antiviral therapy including lamivudine (LAM), adefovir dipivoxil (ADV), telbivudine(LDT), entecavir(ETV) and tenofovir (TDF).

Though NAs can validly inhibit HBV replication and postpone disease progression, these NAs could not eliminate HBV replication template covalently closed circular DNA (cccDNA) and HBsAg clearance is also very rare. As a result, the majority of CHB patients will have to receive long-term or even lifelong NAs treatment to achieve disease remission or serologic endpoints, especially patients with advanced liver disease and cirrhosis [5-6]. As NAs' wide application, safety data of NAs were of paramount importance in clinical practice. Generally, the security of these NAs is good during registration trials [8-13], but there have been reports of serious adverse events including myopathy, neuropathy, pancreatitis and renal impairment during post marketing surveillance [14-18].

Nephrotoxicity may occur in a small, yet significant, proportion of patients receiving nucleotide analogs. In ADV 5 years registration study, 3%-4% patients developed Serum creatinine (Scr) elevation at average of ≥0.5mg/dL every year and that of controls was 0%[19]. In TDF 5 years registration study, 1% patients also developed similar renal toxicity [20]. Along prolonged ADV application in clinical practice, more and more nephrotoxicity had been reported, including Fanconi syndrome [21-23].

Registration reports of these NAs suggested that the renal injury was largely reversible with dose adjustment or cessation of therapy. But post marketing surveillance found that renal tubular dysfunction is partially reversible with changing to other antivirals in patients received prolonged ADV therapy [24]. In order to evaluate the incidence and outcome of long-term NAs usage related renal tubular dysfunction, 255 CHB patients having been receiving NAs for anti-HBV for more than 6 months were enrolled. Urinary and serum β_2 microglobulin (β_2-MG), serum calcium, serum phosphorus and Scr were monitored. Estimated glomerular filtration rate (eGFR) was calculated by using the CKD-EPI equation [25]. Then patients who developed renal injury during antiviral therapy were intervened and followed up prospectively.

Methods

Patients

The observational study was carried out on all patients with CHB who were admitted to the outpatient of the Department of Infectious Diseases of Shanghai Changhai hospital. The study was done during the 3-years period from March 2014 to May 2017.

Patient Management and Follow-Up

All patients were subjected to complete history taking by a specialist from the liver diseases center including general social data, antiviral therapy history, viral data, biochemical data, manifestations and renal function data at the start of antiviral treatment. The major inclusion criteria included HBV mono-infection and having been receiving NAs antiviral therapy for more than 6 months at screening.Exclusion criteria included combining with diabetes, chronic renal insufficiency, rheumatic disease, hypertension and other cardiovascular diseases which may cause renal injury.The subjects assigned as disease controls were patients with CHB and all of them did not receive antiviral therapy 6 months before screening and were matched to study groups by age, sex, and clinical diagnosis.

Laboratory Tests

Qualitative tests for albuminuria and levels of urine β_2 MG, urine micro-protein, Serum creatinine (Scr), serum calcium, serum phosphorus, HBV DNA load, and alanine aminotransferase (ALT) were conducted. Quantitative analyses of HBsAg, HBeAg, and HBeAb were also performed. Using the data for Scr levels, we calculated the eGFR by using the CKD–EPI equation.

Renal function data at the start of antiviral treatment was determined as the most recent creatinine measurement before antiviral treatment, if available, or the first value obtained on the first three month of antiviral therapy.

Treatment Adjustment and Follow-Up

All patients with one or more abnormal renal function indicators at the enrollment were given antiviral strategy adjustment according to the Guidelines for Treatment of Chronic Hepatitis B [26,27] and were followed up for another 52 weeks. In brief, the patients without history of drug-resistant mutation were shifted to other non-cross-resistant antivirals, whereas those with drug-resistant mutations were shifted to ETV 1.0 mg/day or ADV plus LDT.

Ethical Aspects

This study was in compliance with Helsinki Declaration and was approved by the Medical Ethics Committee of Shanghai Changhai Hospital. All the enrolled patients gave their written in-

formed consent.

Statistical Methods

X¯All data were analyzed with SAS9.4 (SAS Institute Inc., USA). A two-sided p-value < 0.05 was considered as statistically significant. Categorical variables were expressed as number (%), and continuous variables were expressed as mean ± standard deviation ($\bar{X}$± SD) or median (Q1-Q3). Categorical variables were processed using x^2 test. Continuous variables were compared using one-way Kruskal-Wallis or ANOVA analysis in accordance with the normal test result. The logistic regression model or general linear model analysis was used to estimate the risk factors of categorical variables or continuous various respectively.

Results

Demographic and Clinical Data Among Patients with Chronic Hepatitis B

In this study 380 patients with CHB and having been receiving NAs antiviral therapy for more than 6 months were screened. 125 of them were ruled out for being accompanied with hypertension, diabetes, chronic kidney disease or history of liver cancer respectively. At last 255 patients were included in this study. 176 patients were receiving different NAs mono-therapy respectively (ADV, 83 patients; ETV, 39 patients; LAM, 27 patients and LDT, 27 patients) and the other 79 patients were receiving LAM or LDT or ETV plus ADV antiviral therapy at the enrollment. 46of those patients who were receiving NAs combined therapy had drug- resistant mutation history during the long-term antiviral therapy. A total of 25 patients with CHB and first-time disease flare and matched with the treatment groups by age, sex, and clinical diagnosis but did not receive any antiviral therapy prior to the study were included as disease controls.

As shown in Table 1, patients in different treatment groups had good comparability in the gender proportion, liver hardness and Scr level or eGFR value before antiviral treatment. All the treatment groups showed good comparability in age and gender with the disease controls. Patients in LDT therapy group were significantly younger than those in LAM or ADV therapy group (P=0.06, 0.03). Therapy duration of these patients varied from 6 months to 132 months. The median therapy duration of ADV, or LAM or NAs combined therapy group was longer than that of LDT or ETV mono therapy group (all P value <0.05). All patients in therapy groups obtained good viral inhibition and ALT recovery when been enrolled in this study.

Characteristics	Patient Groups					Diseases control	P value
	ADV	LDT	LAM	ETV	NAs combine therapy		
number	83	27	27	39	79	25	/
Age, (Md, Q1~Q3)	44(40~51)	36(30~44)	50(37~59)	39(34~47)	41(36~50)	41(34~46)	0.0038
Male sex	66	22	17	34	69	19	0.097
Duration of therapy, months (Md, Q1~Q3)	36(24~72)	18(12~24)	48(24~72)	24(17~42)	36(24~60)	0(0~0)	<0.0001
Scr level before antivial therapy(μmol/L) (X ± SD)	68.68 ±11.34	63.43±10.43	65.20±9.03	70.62 ±15.54	67.09±12.51	/	0.1888
eGFR before antivial therapy(mL/minute) (X ± SD)	114.27 ±19.05	129.46±26.0 1	114.27 ±28.38	116.12 ±24.14	119.58 ±22.83	/	0.0660
Liver hardness(Kpa)	10 (9~15)	10(10~14)	12(10~15)	10(10~14)	10(10~14)	9 (7–19)	0.442
ALT(IU/ml)	25.87±10.27	23.5±9.59	27.58±16.48	25.52±24.55	28.97±17.53	479.08± 150.41	0.001
AST(IU/ml)	25.65±6.42	27.14±7.59	26.75±8.87	23.83±7.41	26.123±8.23	312.36±99.41	0.001
TBI(umol/L)	15.69±6.00	18.68±8.56	17.23±7.48	17.28±8.96	13.48±5.40	76.09±25.10.	0.001
ALB(g/ml)	45.59±2.84	45.96±2.26	44.82±2.91	45.83±2.16	45.70±2.95	36.76±5.99	0.001

HBVDNA (Log IU/ml) (X ± SD)	2.44±0.32	2.75±1.05	2.50±0.50	2.44±0.72	2.44±0.71	6.83 (2.00~7.70)	0.001

Md: median ;lamivudine(LAM), adefovir dipivoxil (ADV), telbivudine(LDT), entecavir(ETV); NAs combine therapy including LAM or LDT or ETV plus ADV; Scr: Serum creatinine; eGFR: estimated Glomerular Filtration Rate

Table 1: Demographic and clinical data of all patients accepting long-term nucleos(t)ide analogue antiviral therapy.

Renal Function Characteristics of Patients with CHB and Being Receiving Long-Term NAs Therapy

To correct a deviation of age and treatment duration among different therapy groups, general linear model or logistic regression model analysis was used. As showed in Table 2, adjusting influence of age and treatment duration, the mean change of eGFR value after long-term NAs antiviral therapy among different therapy groups had no obvious difference(p=0.925). But there were significant difference of serum β_2 MG and urinary β_2 MG abnormal rate among different therapy groups (p<0.0001 Vs p=0.002). Further comparison between any two groups found that there were higher urine β2MG abnormal rate in ADV therapy group than that in LDT therapy group (p=0.032), but there was no significant difference between ADV group and any other therapy groups [LAM(p=0.179), ETV(p=0.203) and NAs combine therapy(p=0.221)]. Unlike urine β_2MG, serum β_2MG mean level in ADV group was higher than both that in LAM therapy group (p=0.041) and ETV group (p=0.007), but there was no significant difference between ADV and LDT therapy group (p=0.45).

Characteristics			**Patient groups**					
		ADV	**LDT**	**LAM**	**ETV**	**NAs combine therapy**	**Unexposed**	**P value**
Urinary β2 MG(mg/L) (Md, Q1~Q3)		0.655 (0.22~2.1)	0.15 (0.10~0.25)	0.345 (0.21~0.46)	0.285 (0.125~0.65)	0.35 (0.12~0.83)	0.64(0.15~1.02)	0.163
Urinary β2MG abnormal rate(%)	**Urinary β2 MG ≥ 10×UNL**	10/58(17.24%)	0/4(0.00%)	0/15(0.00%)	0/18(0.00%)	8/39 (20.51%)	0/15(0.00%)	<0.001
	10×UNL>urinary β2MG≥5×UNL	18/58(31.03%)	0/4(0.00%)	0/15(0.00%)	2/18(11.11%)	2/39 (5.12%)	1/15(6.67%)	
	5×UNL>urinary β2 MG >1×UNL	30/58(51.72%)	4/4(100%)	15/15(100%)	16/18(88.88%)	29/39 (74.35%)	14/15(93.33%)	
Serum β2 MG (mg/L) (Md, Q1~Q3)		2.01 (1.71~2.36)	1.71 (1.57~1.99)	1.84 (1.55~2.23)	1.72 (1.53~1.96)	1.97 (1.73~2.23)	2.49 (1.87–2.83)	<0.0001
Serum calcium(mmol/L) (Md, Q1~Q3)		2.40 (2.34~2.46)	2.39 (2.35~2.48)	2.40 (2.35~2.48)	2.41 (2.35~2.48)	2.43 (2.35~2.48)	2.21 (2.13~2.28)	0.099
Serum phosphorus(mmol/L) (Md, Q1~Q3)		1.01 (0.93~1.09)	1.07 (1.01~1.18)	1.05 (0.88~1.16)	0.995 (0.88~1.16)	0.995 (0.88~1.16)	1.22 (1.05~1.33)	0.219
Scr change (μmol/L) (X ± SD)		5.51±9.89	3.22±7.05	5.04±12.34	2.46±11.01	5.12±12.29	/	0.971
eGFR change (mL/minute) (X ± SD)		-11.47±19.49	-7.08±17.92	-10.29±27.30	-5.73±23.90	-9.43±19.45	/	0.925

Md: median;UNL: upper normal limit; LNL: lower normal limit; eGFR: estimated Glomerular Filtration Rate; Scr: Serum creatinine; Urinary β2 MG: Urine β2 Micro globulin; Serum β2 MG: Serum β2 Micro globulin

Table 2: Renal function of patients with nucleos(t)ide antiviral therapy for more than 6 months.

Relative Risks of Renal Injury During Long-Term NAs Antiviral Therapy in Patients with CHB

Renal injury risk factors for patients with CHB during long-term NAs therapy were analyzed by using general linear model or logistic regression model. The influence factors assessed including age, antiviral strategy and treatment duration.We found that both ADV (OR=1.07,95%CI=0.38-3.04,p=0.0016) and LDT (OR= 0.10,95%CI=0.03-0.40,p=0.0011) therapy were related to urine β2 MG abnormal rate. But only LDT therapy was related to eGFR value (t=2.33, p=0.0204) (Table 4).Age(OR=1.02, 95%CI=1.00-1.05,p=0.0478) was related to both urine β2 MG abnormal rate (Table 3) and eGFR value (t=-4.88, p<0.0001) (Table4).Influence factors included in this study did not show obvious correlation to other renal tubular index, such as serum calcium and serum phosphorus level.

Characteristics	Coefficient B	Standard Error	Wald Chi-Square	Pr > ChiSq	OR		
					Point estimate	95%CI	
						Lower limit	Upper limit
Age	0.0233	0.01	3.92	0.0478	1.02	1	1.05
Treatment duration	0.01	0.01	1.5	0.221	1.01	1	1.02
NAs combine therapy	-0.05	0.25	0.04	0.8449	0.45	0.16	1.28
LDT	-1.54	0.47	10.65	0.0011	0.1	0.03	0.4
LAM	0.03	0.38	0.01	0.9282	0.49	0.14	1.74
ETV	0.01	0.31	0	0.9761	0.48	0.17	1.41
ADV	0.81	0.26	9.96	0.0016	1.07	0.38	3.04

ADV: adefovir dipivoxil ; LAM: lamivudine, LDT: telbivudine, ETV: entecavir

Table 3: Risk factors for urinary β2micro globulin abnormality rete in patients with CHB on nucleos(t)ide antiviral therapy.

Characteristics	Coefficient B	Standard Error	t	P
Age	-0.51	0.10	-4.88	<.0001
Treatment duration	-0.02	0.05	-0.50	0.6161
NAs combine therapy	7.16	4.85	1.48	0.1413
LDT	12.67	5.43	2.33	0.0204
LAM	3.74	5.91	0.63	0.5272
ETV	3.12	5.06	0.62	0.5388
ADV	1.20	4.86	0.25	0.8052
ADV: adefovir dipivoxil ; LAM: lamivudine, LDT: telbivudine, ETV: entecavir				

Table 4: Risk factors for estimated Glomerular Filtration Rate (eGFR)in patients with CHB on nucleos(t)ide antiviral therapy.

Clinical Course of Patients Who Developed Renal Tubular Dysfunction During Long-Term NAs Antiviral Therapy

In this study 135/255(52.94%) patients with CHB developed renal tubular injury and 37 of them (27.40%) were accompanied with mild to moderate eGFR value decrease during long-time antiviral therapy. Only one of all these patients was diagnosed as Fanconi syndrome who with hypophosphatemia and clinical manifestation including fatigue and low limbs ache. 117 of the 135 CHB patients finished the 52-weeks' follow-up and were included into the last data analysis. 59 of these patients continued ADV-including antiviral therapy for another 52 weeks (25 mono therapy and 34 ADV plus LAM or ETV or LDT), and the other 58 patients continued any ADV-excluding NAs antiviral therapy or ceasing antiviral therapy.

As showed in (Table 5), patients in the two antiviral therapy groups had good comparability in the gender, age and liver hardness proportion. Compared to the baseline, urine β_2 MG level of patients in ADV-excluding group decreased at average of 3.94mg/L per year, and eGFR value increased at average of 1.7mL/minute per year. On the contrary, urine β_2 MG level and eGFR value of patents in ADV-including group both deteriorated compared to the baseline (Table 5).

Group	Age, (Md, Q1~Q3)	Mail Sex	Liver har dness (Kpa)	renal function at the begin of follow-up						change of renal function indicators			
				Urinary β2 MG (mg/L)	Serum β_2 MG (mg/L)	Serum calc ium (mmo l/L)	Serum phosp horus (mm ol/L)	eGFR (mL/ min)	Urinary β_2 MG (mg/L)	Serum β_2 MG (mg/L)	Serum calc ium (mm ol/L)	Serum phos phorus (mmo l/L)	eGFR (mL/ min)
ADV-including	46 (39~53)	49	11 (10~15)	4.29± 10.20	2.19± 0.44	2.42± 0.09	1.02± 0.13	106.52± 24.73	-1.41± 10.82	-0.13± 0.55	-0.07± 0.18	-0.13± 0.55	-5.66± 17.71
ADV-excluding	44 (37~56)	47	12 (10~15)	4.09± 9.83	2.27± 0.701	2.32± 0.11	0.96± 0.155	96.72± 18.14	-3.94± 10.82	-0.24± 0.77	-0.06± 0.13	-0.01± 0.55	1.70± 15.81
P value	0.965	0. 776	0.496	0.698	0.321	0.322	0.068	0.317	0.157	0.423	0.628	0.528	0.166

Md: median; ADV: adefovir dipivoxil ; LAM: lamivudine, LDT: telbivudine, ETV: entecavir

Table 5: Clinical course of patients developed renal dysfunction during nucleos(t)ide long-term antiviral therapy.

Unfortunately, we found that although the renal tubular function indexes of these CHB patients in ADV-excluding group improved markedly compared to the baseline, but at the end of 52-weeks' follow-up the urine β_2MG level recovered completely in only 11.36% patients. In this study we found that the renal function of most patients who developed renal injury during long-term anti-HBV therapy could not recover completely during the 52-weeks' ADV-excluding therapy, and especially in those patients who developed both renal tubular function indexes abnormal and eGFR value decrease.

Discussion

NAs had become first-line anti-HBV drugs in clinical practice by possessing the advantages of convenient, efficient and well tolerated [26,28-30]. However, because of the rare HBV eradication and high recurrence rate after drug withdrawal, the majority of CHB patients will require long-time antiviral treatment even the lifetime [26,28]. Therefore, the Adverse Drug Reactions Monitoring (ADRM) of these drugs, especially the effect on renal function, are of paramount importance for ensuring long term usage. Drugs such as ADV tend to be more harmful to the tubular than to the glomerular cells, and in this case the decrease in GFR value develops presumably secondarily after tubular injury, being thereby a relatively late event [31] Serum and urine Low-Molecular-Weight Proteins (LMWP) level, such as β_2 MG exhibit renal handling compatibility to that of an "ideal" marker of GFR. Hence, determining the serum concentrations of various LMWPs has been proposed as a useful approach to evaluate GFR impairment [32-36]. This is a study, which evaluated both GFR values estimated with CKD–EPI equation and serum or urine LMWP level. 52.94% CHB patients developed urine β_2 MG increase during long-term NAs therapy but in these patients the abnormal rate of eGFR value was only 27.40%. The results suggested that urine LMWP level such as β_2 MG level was the more sensitive indicator for evaluation of NAs induced renal tubular dysfunction in patients with CHB. Recently Takagi J's report got the same result [37]. In this study we found not only patients who received long-term NAs antiviral therapy, but also those untreated disease controls showed high urine β_2 MG abnormal rate. It indicated that there were subclinical renal dysfunctions in most CHB patients even before NAs therapy. Previous reports also indicated that renal injury was common in patients with CHB because of immune complex glomerulopathy and HBV direct renal injury [38-41]. Unfortunately, in this study we did not obtain the baseline urine β2 MG data of patients with long-term antiviral therapy. Thus, in our study the exact impact of the hepatic disease on renal function and the specific tubular toxicity of NAs cannot be reliably appreciated.

In addition, in this study we found after adjusting influence of age and treatment duration only urine β2MG abnormal rate in ADV therapy group was significantly higher than that in LDT long-term therapy group. And an increase of more than 10×UNL of urine β2 MG was only detected in the ADV mono or combine therapy group (Table 2). These results suggested that antivirals may be one of the risk factors for renal injury during long-term NAs therapy. When investigating potential factors associated with urine β2 MG abnormal and eGFR decrease we identified age as the risk factor for both eGFR decrease and urine β2 MG increase and ADV was the unique drug which was related to the urine β2 MG abnormal during therapy. We also found that LDT had a protect effect on eGFR value and urine β2 MG, which is also in line with the previous study [42-44]. Although we found patients in ADV mono-therapy group having high risk of urineβ2MG abnormal, we

observed ADV combined therapy did not increase the risk of renal injury compared to ADV monotherapy. The result was in line with previous study [44].

Unfortunately, in our study we observed the majority of those patients with CHB who developed renal injury during long-term antiviral therapy can't completely recover after adjusting antiviral to ADV-excluding treatment at the end of the 52-weeks' follow-up. The result was not consistent with the registration study [14,19]. The possible reason was first, most patients in our study had received longer time treatment than those in the registration study and the majority of them in clinical practice have to continue NAs treatment instead of drug withdrawal; second, the kidney injury of these patients was ignored because of low sensitivity of eGFR, and then delayed the drug adjusting time in practice; third, the prolonged NAs treatment may cause renal interstitial fibrosis. When investigating the potential factors associated with renal function improvement, we found that the renal function level before therapy adjustment was the unique significant factor. The result suggested that early detection of injury determined the improvement degree of renal function.

The limitation of this study is the small size and the short time of the prospective observation follow up. As a result, clinical trials with large sample size and long-term follow-up are needed to confirm and investigate the optimal therapy for patients with obvious tubular damage, especially for patients with history of drug resistance mutation and need long-time antiviral therapy.

Conclusion

In summary, the results of this clinical study suggest that many patients developed renal injury during prolonged ADV-including anti-HBV therapy and urinary β2 MG are more sensitive to monitor NAs antiviral therapy related renal tubular injury. Prolonged ADV-including antiviral therapy maybe cause irreversible renal tubular damage in CHB patients.

Acknowledgement

We thank all patients for their understanding and participation.

References

1. Trepo C, Chan HL, Lok A (2014) Hepatitis B virus infection. Lancet 384: 2053-2063.
2. Ott JJ, Stevens GA, Groeger J, Wiersma ST (2012) Global epidemiology of hepatitis B virus infection: new estimates of age-specific HBsAg seroprevalence and endemicity. Vaccine 30: 2212-2219.
3. Kwon H, Lok AS (2011) Hepatitis B therapy. Nat Rev Gastroenterol Hepatol 8: 275-284.
4. Lok AS, McMahon BJ, Wong JB, Ahmed AT, Farah W, et al. (2016) Antiviral therapy for chronic hepatitis B viral infection in adults: A systematic review and meta-analysis. Hepatology 63: 284-306.
5. Liver. EAftSot (2012) EASL Clinical Practice Guidelines: management of chronic hepatitis B virus infection. J Hepato l57: 167-185.
6. Liaw YF, Kao JH, Piratvisuth T, Piratvisuth T, Gane E, et al. (2012) Asian-Pacific consensus statement on the management of chronic hepatitis B: a 2012 update. Hepatol Int 6: 531-561.
7. Terrault NA, Bzowej NH, Chang KM, Hwang JP, Jonas MM (2016) AASLD Guidelines for Treatment of Chronic Hepatitis B. Heptanol 63: 261-283.
8. Marcellin P, Chang TT, Lim SG, Tong MJ, Sievert W, et al. (2003) Adefovir Dipivoxil for the Treatment of Hepatitis B e Antigen-Positive Chronic Hepatitis B. New England Journal of Medicine 348: 808-816.
9. Lai CL, Chien RN, Chang TT, Guan R, Tai DI, et al. (1998) A One-Year Trial of Lamivudine for Chronic Hepatitis B. New England Journal of Medicine 339: 61-68.
10. Chang TT, Marcellin P, Lim SG, Wolfson MS, Fry J, et al. (2003) Adefovir Dipivoxil for the Treatment of Hepatitis B e Antigen-Negative Chronic Hepatitis B. New England Journal of Medicine 348: 800-807.
11. Lai CL, Shouval D, Lok AS, Chang TT, Cheinquer H, et al. (2006) Entecavir versus Lamivudine for Patients with HBeAg-Negative Chronic Hepatitis B. New England Journal of Medicine 354: 1011-1020.
12. Lai CL, Gane E, Liaw YF, Hsu CW, Thongsawat S, et al. (2007) Telbivudine versus Lamivudine in Patients with Chronic Hepatitis B. New England Journal of Medicine 357: 2576-2588.
13. Marcellin P, Heathcote EJ, Buti M, Gane E, Krastev Z, et al. (2008) Tenofovir Disoproxil Fumarate versus Adefovir Dipivoxil for Chronic Hepatitis B. New England Journal of Medicine359: 2442-2455.
14. Fontana RJ (2009) Side effects of long-term oral antiviral therapy for hepatitis B. Hepatology 49: 185-195.
15. Zou XJ, Jiang XQ, DY T (2011) Clinical features and risk factors of creatine kinase elevations and myopathy associated with telbivudine. J Viral Hepat 18: 892-896.
16. Coca S, Perazella MA (2002) Acute renal failure associated with tenofovir: evidence of drug-induced nephrotoxicity. Am J Med Sci 324: 342-344.
17. Wang M, Da Y, Cai H, Lu Y, Wu L, et al. (2012) Telbivudine myopathy in a patient with chronic hepatitis B. International journal of clinical pharmacy 34: 422-425.
18. Khungar V, Han SH (2010) A Systematic Review of Side Effects of Nucleoside and Nucleotide Drugs Used for Treatment of Chronic Hepatitis B. Current hepatitis reports 9: 75-90.
19. Hadziyannis SJ, Tassopoulos NC, Heathcote EJ, Chang TT, Kitis G, et al. (2006) Long-term therapy with adefovir dipivoxil for HBeAg-negative chronic hepatitis B for up to 5 years. Gastroenterology 131: 1743-1751.
20. Marcellin P, Gane E, Buti M, Afdhal N, Sievert W, et al. (2013) Regression of cirrhosis during treatment with tenofovir disoproxil fumarate for chronic hepatitis B: a 5-year open-label follow-up study. The Lancet 381: 468-475.
21. Kim YJ, Cho HC, Sinn DH, Gwak GY, Choi MS, et al. (2012) Frequency and risk factors of renal impairment during long-term adefovir dipivoxil treatment in chronic hepatitis B patients. J Gastroenterol Hepatol 27: 306-312.

22. Mederacke I, Yurdaydin C, Grosshennig A, Erhardt A, Cakaloglu Y, et al. (2012) Renal function during treatment with adefovir plus peginterferon alfa-2a vs either drug alone in hepatitis B/D co-infection. J Viral Hepat 19: 387-395.

23. Ha NB, Ha NB, Garcia RT, Trinh HN, Vu AA (2009) Renal dysfunction in chronic hepatitis B patients treated with adefovir dipivoxil. Hepatology 50: 727-734.

24. Gara N, Zhao X, Collins MT, Chong WH, Kleiner DE (2012) Renal tubular dysfunction during long-term adefovir or tenofovir therapy in chronic hepatitis B. Alimentary pharmacology & therapeutics 35: 1317-1325.

25. Levey AS, Stevens LA, Schmid CH, Zhang YL, Feldman HI, (2009) A New Equation to Estimate Glomerular Filtration Rate. Annals of Internal Medicine 150: 604-612.

26. Terrault NA, Bzowej NH, Chang KM, Hwang JP, Jonas MM, et al. (2016) AASLD Guidelines for Treatment of Chronic Hepatitis B. Hepatology 63: 261-283.

27. WHO Guidelines Approved by the Guidelines Review Committee. (2015) Guidelines for the Prevention, Care and Treatment of Persons with Chronic Hepatitis B Infection. Geneva: World Health Organization Copyright (c) World Health Organization.

28. Sarin SK, Kumar M, Lau GK, Abbas Z, Chan HL, (2016) Asian-Pacific clinical practice guidelines on the management of hepatitis B: a 2015 update. Hepatol Int 10: 1-98.

29. Dienstag JL (2009) Benefits and risks of nucleoside analog therapy for hepatitis B. Hepatology 49: 112-121.

30. Merriman RB, Tran TT (2016) AASLD practice guidelines: The past, the present, and the future. Hepatology 63: 31-34.

31. Donadio C, Lucchesi A, Ardini M, Giordani R (2001) Cystatin C, B2-microglobulin, and retinol-binding protein as indicators of glomerular filtration rate: comparison with plasma creatinine. J Pharm Biomed Anal; 24: 835-842.

32. Bökenkamp A, Domanetzki M, Zinck R, Schumann G, Byrd D, et al. (1998) Cystatin C-a new marker of glomerular filtration rate in children independent of age and height. Pediatrics 101: 875-881.

33. Le Bricon T, Thervet E, Benlakehal M, Bousquet B, Legendre C, et al. (1999) Changes in plasma cystatin C after renal transplantation and acute rejection in adults. Clin Chem 45: 2243-2249.

34. Schardijn GH, SvE LW (1987) Beta 2-microglobulin: its significance in the evaluation of renal function. Kidney Int 32: 635-641.

35. Tramonti G, Donadio C, Ferdeghini M, Annichiarico C, Norpoth M, (1996) Serum tumour-associated trypsin inhibitor (TATI) and renal function. Scand J Clin Lab Invest 56: 653-656.

36. Donadio C (2010) Serum and urinary markers of early impairment of GFR in chronic kidney disease patients: diagnostic accuracy of urinary-tract protein. Am J Physiol Renal Physiol 299: 1407-1423.

37. Takagi J, Morita H, Ito K, Ohashi T, Hirase S, et al. (2016) Urinary beta-2 Microglobulin Levels Sensitively Altered in an Osteomalacia Patient Receiving Add-on Adefovir Dipivoxil Therapy for Hepatitis B Virus Infection. Internal medicine 55 1599-1603.

38. Hong L, Zhang J, Min J, Lu J, Li F (2010) A role for MHBst167/HBx in hepatitis B virus-induced renal tubular cell apoptosis. Nephrology, dialysis, transplantation 25: 2125-2133.

39. Takekoshi Y, Tochimaru H, Nagata Y, Itami N (1991) Immunopathogenetic mechanisms of hepatitis B virus-related glomerulopathy. Kidney international Supplement 35: 34-39.

40. He XY, Fang LJ, Zhang YE, Sheng FY, Zhang XR, (1998) *In situ* hybridization of hepatitis B DNA in hepatitis B-associated glomerulonephritis. Pediatric nephrology 12: 117-120.

41. Lai KN, Ho RT, Tam JS, Lai FM (1996) Detection of hepatitis B virus DNA and RNA in kidneys of HBV related glomerulonephritis. Kidney international 50: 1965-1977.

42. Gane EJ, Deray G, Liaw YF, Lim SG, Lai CL, et al. (2014) Telbivudine improves renal function in patients with chronic hepatitis B. Gastroenterology 146: 138-146.

43. Lee M, Oh S, Lee H, Yeum T, Lee J (2014) Telbivudine protects renal-function in patients with chronic hepatitis B infection in conjunction with adefovir-based combination therapy. J Viral Hepat 21: 873-881.

44. Qi X, Wang J, Chen L, Huang Y, Qin Y, et al. (2015) Impact of nucleos(t)ide analogue combination therapy on the estimated glomerular filtration rate in patients with chronic hepatitis B. Medicine 94: 646.