Conclusion: Quantitative renal cortical echogenicity using computer-based image analysis might be a useful tool to identify patients with CGN and renal progression related to renal fibrosis.

Keywords: Renal ultrasound, Computer-based image analysis, CKD progression

Introduction

Renal sonography is a reliable and noninvasive diagnostic tool, providing ease of use and valuable information concerning structural changes, especially among patients with chronic kidney disease (CKD).[1] Decreased kidney length, cortical parenchymal thickness and increased parenchymal echogenicity could represent CKD parenchymal damage.[2] Renal sonographic information can assess CKD status, and significant correlation was found between cortical thickness, renal length and estimated glomerular filtration rate (eGFR).[3,4]

Several studies have demonstrated association of sonographic findings with renal histopathologic findings. Initial investigation indicated that renal cortical echogenicity correlated to severity of global sclerosis, tubular atrophy, the number of hyaline casts per glomerulus and focal leukocytic infiltration.[5] One later study, revealed significant correlation between the degree of cortical echogenicity and glomerulosclerosis or tubular atrophy, but without any correlation to interstitial fibrosis.[6] Recently another study confirmed that renal length and cortical thickness predicted renal progression and histopathologic changes, especially, after weighting for cortical echogenicity, when scored by comparison with liver echogenicity.[7]

The reliability of renal sonography is questionable because sonographic dimensions and quantitative measurement are often operator dependent. The use of computer-assisted image analysis has become increasingly accessible in many areas and has also provided greater accuracy in the interpretation of various types of image studies. One recent study indicated that using a comprehensive approach to analyze and classify CKD stages, according to renal sonographic images, when assisted by an image-processing model, identified potential patients with CKD at early stages.[8] However, data about the relationship between renal progression and renal sonographic assessment, using computer-assisted image analysis, among patients with chronic glomerulopathy (CGN) are limited. In this study, we aimed to predict renal progression using computer-based image analysis of renal sonographic findings. We also aimed to investigate association between renal sonographic findings and chronic histopathologic findings.

Methods

Study design

This study employed a prospective cohort design (diagnostic test), conducted on enrolled patients with CGN, attending Phramongkutklao Hospital, during June through to November 2021. The trial was approved by the Ethics Committee of the Institute Review Board at the Royal Thai Army Medical Department (IRBRTA) April 28, 2021, with code R037h/64. This study was registered with Thai Clinical Trials code TCTR202203211001.

Study population

The inclusion criteria comprised patients aged over 18 years with CGN, who were undergoing renal biopsies and could provide informed consent. Patients with obstructive uropathy, renal tumor, single kidney, polycystic kidney disease, transplanted kidney, pregnancy, body mass index more than 30 kg/m^2 and end stage kidney disease were excluded.

Laboratory measurements

Biochemical parameters and albuminuria were measured in serum and urine using a Roche P800 Modular Chemistry Analyzer (Roche Diagnostics, Basel, Switzerland), measurement being performed at baseline and after three months. The eGFR was estimated from serum creatinine using the CKD-EPI (Chronic Kidney Disease Epidemiology Collaboration) formula[9] and CKD progression was defined as an eGFR-CKD-EPI decline of more than 25% from baseline within one year.

Renal ultrasonography

Ultrasonographic studies on kidneys were carried out using a Philips HD3-EXP ultrasound machine with 3.5 MHz convex transducer. Renal sonography was performed by a single nephrologist with 10 years of experience in the field of renal sonography in the outpatient department of our renal unit and the investigator was blinded regarding patients' histories and laboratory results. Renal sonographic parameters including kidney length, parenchymal thickness and parenchymal echogenicity were obtained from both kidneys. The renal sonographic images were captured in both longitudinal and short axis views, 8 to 10 images each. Renal images were collected and compared with liver images, comprising 2 to 5 images for each patient, for renal to liver echogenicity ratio measurement. All pictures were obtained in a .jpg file and all ultrasound images were performed by a single nephrologist.

Computer assisted image analysis

Sonographic images were analyzed by the department of Computer Science, Faculty of Science, Kasetsart University and were similar to previously used protocols.[8] At the beginning, the systems required users to define the area of interest, focusing on three main areas: the cortical area, calyceal system and whole renal contour. To ensure the precise definitions of the renal contours using an ultrasound image, the boundaries between different parts of the kidney were initially identified in polygonal points, using a LabelMe Program. After that, polygonal masking was performed, by coding in a MATLAB Program, to differentiate renal cortex areas from any renal cyst and calyceal areas (Figure 1).

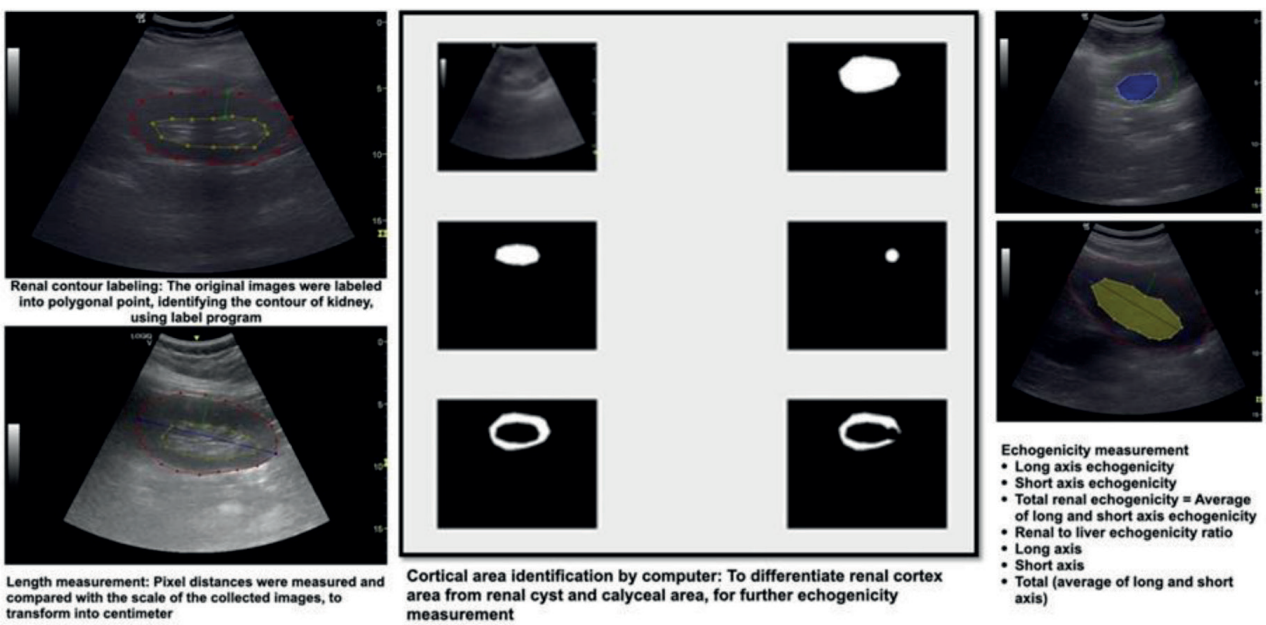


Figure 1 Analysis of sonographic images

The next step was cortical echogenicity measurement. Sonographic images were transformed to small discrete elements called pixels and echogenicity measurements were demonstrated in numerical value, representing the intensity of a particular channel at the location of a pixel. In each patient, renal echogenicity was measured, to obtain an average value, within the sets of pictures. These were classified by the captured view as follows. Long axis echogenicity, short axis echogenicity, total renal echogenicity, which is the average of long and short axis echogenicity and finally the renal to liver echogenicity ratio, which is renal echogenicity compared with liver echogenicity. Length was measured using the longest pixel distances compared with the scale of the collected images, converted to centimeters. Furthermore, deep learning model training to identify renal contour was performed, using YOLO v5 training model (Figure 1).

In those patients with CGN, admitted for renal biopsy, the histopathologic grading of chronic changes in native renal biopsy samples were evaluated by a single renal pathologist, focusing on glomerulosclerosis, interstitial fibrosis and tubular atrophy (IFTA), using a modified NIH lupus nephritis activity and chronicity scoring system.[10]

Statistical analysis

The results were expressed as mean ± standard deviations or as median with interquartile range (IQR) according to data distribution. Difference between groups was analyzed using Chi-square, Mann–Whitney U and Student's t tests. Univariate analysis was performed to explore relationships between renal progression and renal histologic findings and other ultrasonographic variables, employing a Pearson correlation test for normally distributed data and a Spearman Rank correlation test for nonparametric data. We also conducted multivariate analysis for CKD progression prediction. Results underwent characteristic (ROC) analysis and the areas under the curves (AUCs) were estimated to investigate the role of each renal sonographic parameter to determine CKD progression or renal fibrosis. Statistical significance was defined as P-value < 0.05.

Results

A total of 37 patients with CGN, 12 males and 25 females, were enrolled. Their mean age was 42.6 ± 13.2 years. As shown in Table 1, baseline characteristics of patients with CKD progression and no CKD progression did not differ. As recorded, 6 (16.2%) patients presented with diabetes mellitus, and 16 (43.2%) patients had lupus nephritis. The correlation of long axis echogenicity, systolic blood pressure, baseline estimated GFR, and urine albumin to creatinine ratio (UACR), to GFR progression, among patients with CGN undergoing renal biopsy is illustrated in Figure 2.

Renal sonography and CKD progression

Total renal echogenicity (50.59 ± 10.33 vs. 43.61 ± 9.45, P = 0.049) and long axis echogenicity (50.18 ± 11.58 vs. 41.91 ± 8.62, P = 0.02), were significantly higher among patients with CKD progression in the CGN group (Table 2). Multivariate analysis was performed to determine their relative contributions to echogenicity. When adjusted for underlying diabetes mellitus, prednisolone use and baseline UACR, renal progression showed significant independent contributions to both total renal echogenicity (adjusted HR 1.13, 95% CI, 1.01 to 1.25) and long axis echogenicity (adjusted HR 1.14, 95%CI, 1.02 to 1.29) (Table 3).

Table 1 Baseline characteristics

Variable	CKD progression (N=12)	Non-CKD progression (N=25)	P-value
Male (%)	25	40	0.371
Age (year)	42.1 ± 12.1	43.4 ± 14.5	0.781
BMI (kg/m^2)	24.3 ± 5.3	24.7 ± 5.1	0.807
Underlying diseases N (%)			
• Type 2 diabetes	4 (33.3)	2 (8)	0.050
• Hypertension	4 (33.3)	7 (28)	0.740
• Lupus nephritis	4 (33.3)	12 (48)	0.399
Medications N (%)			
• Prednisolone	7 (58.3)	21 (84)	0.088
• Other immunosuppressive agents	5 (41.7)	15 (60)	0.295
• ACEs/ARBs	7 (58.3)	20 (80)	0.165

UACR; Urine albumin to creatinine ratio, SBP; systolic blood pressure, DBP; diastolic blood pressure, eGFR; estimated glomerular filtration rate; ACEs; Angiotensin converting enzyme inhibitors, ARBs; Angiotensin receptor blockers.

Table 2 Computer-based sonographic findings in CGN biopsies

Sonographic findings	CKD progression (N=12)	Non-CKD progression (N=25)	P-value
Total renal echogenicity	50.59 ± 10.33	43.61 ± 9.45	0.049
Total renal/liver echogenicity ratio	1.24 ± 0.25	1.13 ± 0.24	0.238
Long axis echogenicity	50.18 ± 11.58	41.91 ± 8.62	0.020
Long axis renal/liver echogenicity ratio	1.22 ± 0.25	1.09 ± 0.23	0.117
Short axis echogenicity	51.36 ± 9.69	45.84 ± 10.70	0.140
Short axis renal/liver echogenicity ratio	1.26 ± 0.26	1.19 ± 0.27	0.452
Cortical thickness (cm)	1.78 ± 0.36	1.75 ± 0.30	0.788
Renal length (cm)	10.39 ± 0.35	11.06 ± 0.30	0.083

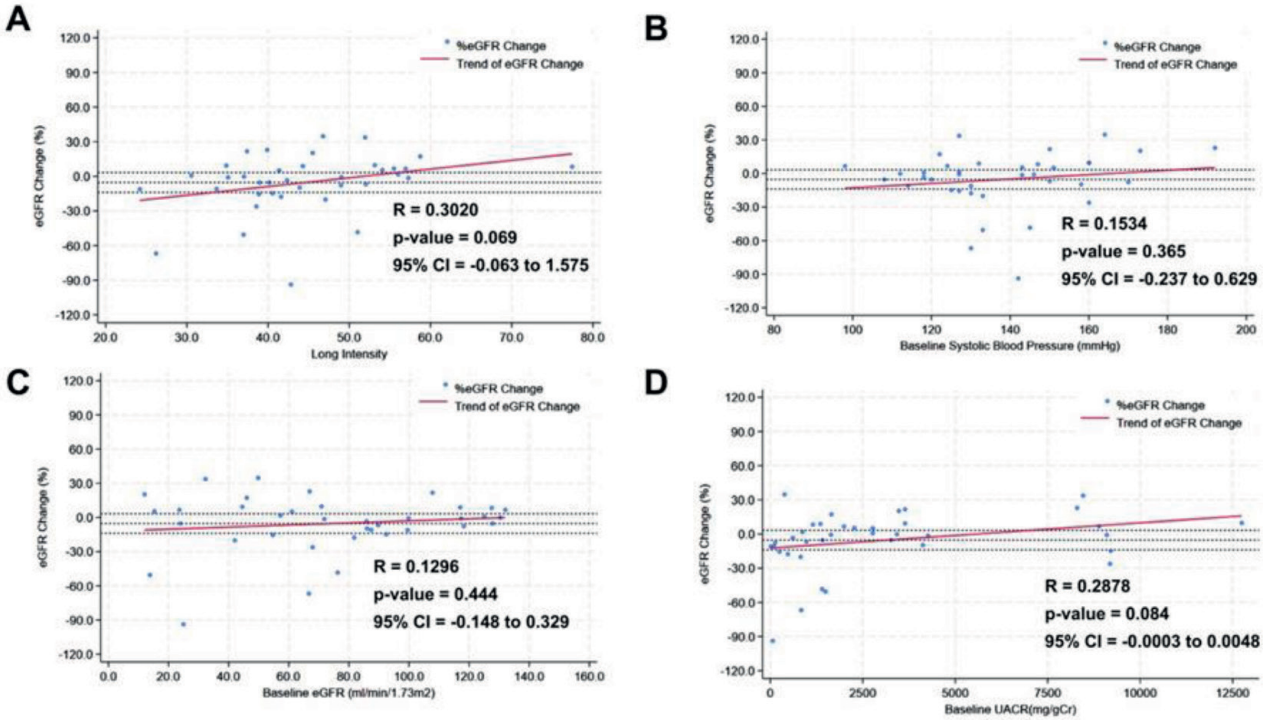


Figure 2 Correlation of long axis echogenicity, systolic blood pressure, baseline estimated GFR and urine albumin creatinine ratio with GFR progression among patients with CGN undergoing renal biopsy

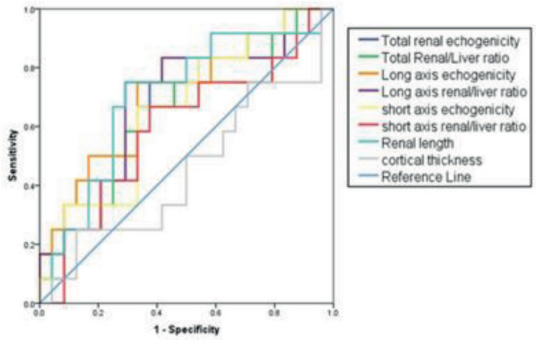
Table 3 Multivariate analysis to predict CKD progression among patients with CGN undergoing renal biopsy

	Unadjusted HR	95% CI	P-value	Adjusted HR	95% CI	P-value
Model 1						
Total renal echogenicity	1.08	(0.99, 1.17)	0.060	1.13	(1.01, 1.25)	0.026
Type 2 diabetes	5.75	(0.88, 37.62)	0.068	8.11	(0.65, 100.9)	0.104
Prednisolone	0.27	(0.56,1.28)	0.099	0.30	(0.40, 2.31)	0.250
UACR	1	(1.00,1.00)	0.075	1.00	(1.00,1.00)	0.230
Model 2						
Long axis echogenicity	1.10	(1.01, 1.20)	0.037	1.14	(1.02, 1.29)	0.026
Type 2 diabetes	5.75	(0.88, 37.62)	0.068	8.83	(0.65, 100.9)	0.098
Prednisolone	0.27	(0.56,1.28)	0.099	0.33	(0.40, 2.31)	0.295
UACR	1	(1.00,1.00)	0.075	1.00	(1.00,1.00)	0.265

UACR; Urine albumin to creatinine ratio, HR; Hazard ratio, Adjusted; T2DM, prednisolone and baseline UACR.

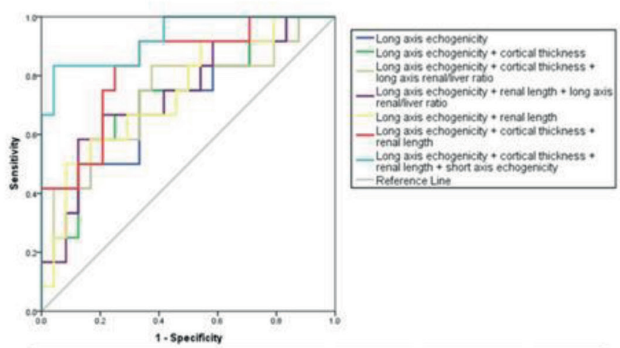
ROC analysis was performed to identify the best ultrasonographic parameters able to discriminate CKD progression from non-CKD progression in the CGN group. Long axis echogenicity was the only sonographic parameter that significantly predicted renal progression (AUC 0.71; 95% CI, 0.52 to 0.89) and when combined with other sonographic findings, including cortical thickness, renal length and short axis echogenicity, was able to achieve a better score to predict CKD progression in the CGN group (AUC 0.93; 95% CI, 0.84 to 1.00) (Figure 3).

A. Area under the curve of predictive factors for CKD progression



Sonographic findings	AUC	95% CI	*p*-value
Cortical thickness (cm)	0.451	(0.24, 0.61)	0.638
short axis renal/liver echogenicity ratio	0.597	(0.39, 0.80)	0.347
Total renal/liver echogenicity ratio	0.635	(0.44, 0.83)	0.191
Short axis echogenicity	0.653	(0.47, 0.84)	0.140
Total renal echogenicity	0.681	(0.49, 0.87)	0.081
Long axis renal/liver echogenicity ratio	0.687	(0.49, 0.88)	0.070
Renal length (cm)	0.701	(0.52,0.89)	0.052
Long axis echogenicity	0.705	(0.52, 0.89)	0.048

B. Area under the curve of long axis echogenicity combined with other parameters for CKD progression



Sonographic findings	AUC	95% CI	*p*-value
Long axis echogenicity	0.705	(0.52, 0.89)	0.048
Long axis echogenicity + cortical thickness	0.726	(0.54, 0.91)	0.029
Long axis echogenicity + cortical thickness + long axis renal/liver ratio	0.736	(0.55, 0.92)	0.022
Long axis echogenicity + renal length + long axis renal/liver ratio	0.740	(0.56, 0.92)	0.021
Long axis echogenicity + renal length	0.743	(0.57, 0.92)	0.019
Long axis echogenicity + cortical thickness + renal length	0.830	(0.69, 0.97)	0.001
Long axis echogenicity + cortical thickness + renal length	0.931	(0.84,1.00)	< 0.001

Figure 3 Area under the curve of predictive factors for CKD progression

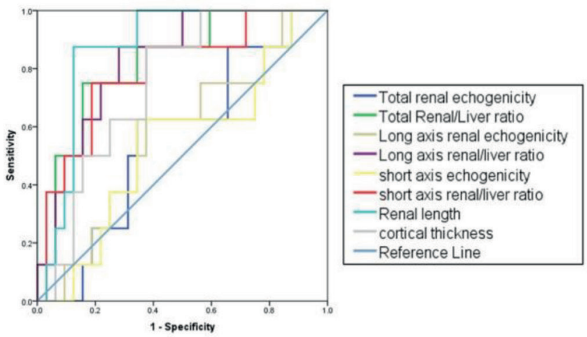
Renal sonography and histopathologic findings

The median percentage of glomerulosclerosis was 31.67% (IQR 8.71 to 57.29), median of percentage of IFTA was 20% (IQR 5 to 40) and median of chronicity score was 4.5 (IQR 2.5 to 7.0). Percentage of IFTA positively correlated to total renal to liver echogenicity ratio (R = 0.399, P = 0.014), long and short renal to liver echogenicity ratio (R = 0.36, P = 0.023), short axis renal/liver echogenicity ratio (R = 0.40, P = 0.011) and negatively correlated to cortical thickness (R = -0.39, P = 0.013) and kidney length (R = -0.50, P= 0.001). The percentage of glomerulosclerosis and chronicity scores also negatively correlated with cortical thickness and renal length (Table 4).

ROC analysis of ultrasonographic parameters used to determine IFTA >50% among patients with CGN is illustrated in Figure 4. AUC to diagnose IFTA > 50% using kidney length, long axis renal to liver echogenicity ratio, total renal/liver echogenicity ratio, short axis renal/liver echogenicity ratio and cortical thickness were 0.87 (95% CI, 0.76 to 0.98), 0.83 (95% CI, 0.67 to 0.98), 0.82 (95% CI, 0.67 to 0.98), 0.79 (95% CI, 0.62 to 0.97) and 0.75 (95% CI, 0.59 to 0.91), respectively. The optimal long axis renal to liver echogenicity ratio was determined as 1.138 with a sensitivity of 87.5% and specificity of

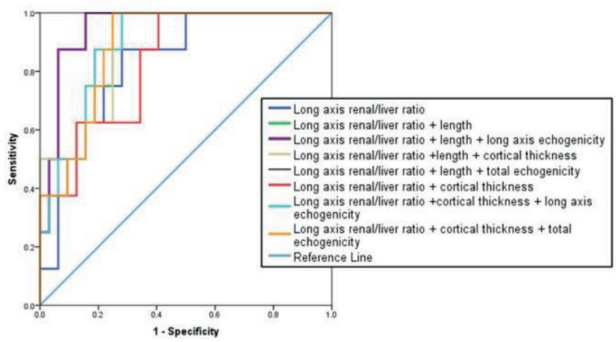
71.9% (AUC, 0.83; 95% CI, 0.67 to 0.98). The cutoff levels of long axis renal to liver echogenicity ratio, used to indicate IFTA > 50%, are also demonstrated in Table 5. In addition, combined long axis renal to liver echogenicity ratio with kidney length and long axis echogenicity achieved a perfect score predicting IFTA > 50% in the CGN group such as (AUC 0.95; 95% CI, 0.88 to 1.00).

A. Area under the curve of predictive factors for IFTA>50%



Sonographic findings	AUC	95% CI	*p*-value
Short axis echogenicity	0.539	(0.32, 0.76)	0.735
Total renal echogenicity	0.563	(0.36, 0.77)	0.589
Long axis echogenicity	0.570	(0.36, 0.78)	0.543
Cortical thickness (cm)	0.746	(0.59, 0.91)	0.033
short axis renal/liver echogenicity ratio	0.793	(0.62, 0.97)	0.011
Total renal/liver echogenicity ratio	0.820	(0.67, 0.98)	0.006
Long axis renal/liver echogenicity ratio	0.828	(0.69, 0.97)	0.005
Renal length (cm)	0.871	(0.76,0.98)	0.001

B. Area under the curve of Long axis renal/liver echogenicity ratio combined with other parameters for IFTA > 50%



Sonographic findings	AUC	95% CI	*p*-value
Long axis renal/liver echogenicity ratio	0.828	(0.69, 0.97)	0.005
Long axis renal/liver ratio + cortical thickness	0.887	(0.78, 0.99)	0.001
Long axis renal/liver ratio + length + cortical thickness	0.891	(0.78, 0.99)	0.001
Long axis renal/liver ratio + cortical thickness	0.891	(0.79, 0.99)	0.001
Long axis renal/liver ratio + length	0.895	(0.79, 0.99)	0.001
Long axis renal/liver ratio + length + total echogenicity	0.941	(0.87,1.00)	< 0.001
Long axis renal/liver ratio + length + long axis echogenicity	0.949	(0.88, 1.00)	< 0.001

Figure 4 Area under the curve to determine IFTA > 50%

Table 4 Correlation between sonographic findings and renal pathology

Sonographic findings	Glomerulosclerosis (%)	IFTA (%)	Chronicity score
Total renal echogenicity	0.16 P = 0.324	0.02 P = 0.918	-0.05 P = 0.738
Total renal/liver echogenicity ratio	0.31 P = 0.055	0.39 P = 0.014	0.30 P = 0.065
Long axis echogenicity	0.13 P = 0.420	-0.01 P = 0.953	-0.09 P = 0.599
Long axis renal/liver echogenicity ratio	0.29 P = 0.072	0.36 P = 0.023	0.26 P = 0.102
Short axis echogenicity	0.17 P = 0.308	0.04 P = 0.830	-0.03 P = 0.838
Short axis renal/liver echogenicity ratio	0.30 P = 0.059	0.40 P = 0.011	0.31 P = 0.053
Cortical thickness (cm)	-0.51 P = 0.001	-0.39 P = 0.013	-0.41 P = 0.009
Renal length (cm)	-0.45 P = 0.004	-0.50 P = 0.001	-0.47 P = 0.002

Table 5 Cutoff value of long axis renal/liver echogenicity ratio to predict IFTA > 50%

Long axis renal/liver echogenicity ratio	Sensitivity	Specificity	PPV	NPV	+ LR	- LR
1.102	87.5 %	56.3 %	33.33	94.74	2.00	0.22
1.123	87.5 %	68.8 %	41.18	95.65	2.80	0.18
1.138	87.5 %	71.9 %	43.75	98.83	3.11	0.17
1.157	75.0 %	71.9 %	40.00	92.00	2.67	0.33
1.189	62.5 %	78.1 %	41.67	89.29	2.89	0.48
1.239	62.5 %	84.4 %	50.00	90.00	4.00	0.44
1.353	50.0 %	90.6 %	57.14	87.88	5.33	0.55

Discussion

In this study, we indicated that ultrasonographic parameters, including kidney length, parenchymal thickness and quantitative renal echogenicity, using computer-based image analysis, were associated with renal progression and fibrosis in renal histopathology among patients with CGN. Using computer-based image analysis, we defined new ultrasonographic parameters, such as long axis echogenicity, that significantly correlated to renal progression, as well as long axis renal to liver echogenicity ratios that correlated to the degree of renal fibrosis. Also, ROC curve analysis, to determine IFTA > 50%, showed that combining the long axis renal to liver echogenicity ratio with kidney length and long axis echogenicity, provided the best parameter, exhibiting the highest AUC.

Renal sonography is a useful diagnostic tool for kidney diseases, but the results of the test mainly depend on the physician's experience and can vary among operators. Thus, data about the best ultrasonographic parameters in evaluating CKD remain conflicting. We established a model of an image-processing system to evaluate and measure sets of renal images among patients with CGN to provide a more precise, accurate and reliable detection system.

Regarding the process of image analysis, we focused mainly on the measurement of echogenicity. Because of the lack of standardized measuring methods or cutoff value of the cortical echogenicity, many studies only visually evaluated this component, by comparing it with liver echogenicity and translating the results only to scaling scores and not by direct measurement. Several studies showed association between renal function and ultrasonographic parameters, including kidney length, parenchymal thickness and renal parenchymal echogenicity.[11,12] Our study notably found that both total renal echogenicity and long axis echogenicity predicted renal progression, and the combination of long axis echogenicity along with other sonographic findings achieved better parameters for the prediction of renal progression in patients with CGN.

Tubulointerstitial changes have been proven to better predict renal progression and prognosis in patients with CGN and diabetic nephropathy.[13] Based on the assumption that evidence of chronicity change would be a useful to guide therapy in CGN, a sonographic test, that is able to avoid unnecessary renal biopsies in severe CGN, would be a desirable diagnostic tool. One study showed that cortical renal echogenicity was related to tubular atrophy and interstitial inflammation.[6]

However, the level of renal parenchymal echogenicity is a subjective assessment. Our study found that renal echogenicity, augmented by computer based-imaging analysis, correlated well to tubulo-interstitial fibrosis, predicting irreversible impairment of renal function. Moreover, we confirmed that renal echogenicity levels, especially combined with other parameters, improved scoring for prediction of IFTA > 50% in the CGN group. For long axis renal to liver echogenicity ratio, the cutoff value 1.138 provided a sensitivity 87.5%, specificity 71.9%, negative predictive value 98.8% and likelihood ratio 0.17, which again could be helpful in predicting severe chronic change and therefore further refine the decision-making process for renal biopsy indication. Similar to related studies, quantitative renal echogenicity by kidney/liver ratio strongly determined irreversible kidney injury by renal histopathology score[14] and has also been shown to reflect the severity of damage in pediatric renal cases.[15]

Several limitations were encountered in this study. Firstly, this was a single-center referral care center study. Secondly, the number of patients was relatively few with a relatively short time to evaluate CKD progression. Thirdly, the study still requires internal and external validity to confirm its applicability because the renal ultrasonographic data was collected by a single nephrologist. However, the strength of our computer-based image analysis was that it required less time for quantitative echogenicity measurement, for example it only took 10 minutes for the analysis of 1,400 images, of great use in analysis of large data sets in future clinical trials.

Conclusion

Quantitative renal cortical echogenicity, using computer-based image analysis, might be a useful tool to identify patients with CGN at risk of renal progression. Renal cortical echogenicity and thickness exhibited a close relationship to the degree of chronic tubulointerstitial changes among patients with CGN, who were referred for renal biopsy. The ultrasonographic parameters, using computer-based image analysis defined in this study, could provide more objective data in assessing CKD.[16] However, further large-scale clinical and research studies in CKD populations are needed to confirm our study results.

Conflicts of interest

All authors have no conflict of interest to declare.

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This study did not receive any funding in any form.

Data sharing statement

The data presented in this study are available on request from the corresponding author.

Authors' contribution

Conceptualization: NC, AC, NN, PT, NV, PT, PI, WK, OS, BS

Data curation: NC, AC, NN, PT, OS

Formal analysis: NC, BS, OS

Methodology: NC, AC, BS

Project administration: NC

Writing-original draft: NC, AC, BS

Writing-review & editing: NC, AC, NN, PT, NV, PT, PI, WK, OS, BS

All authors read and approved the final manuscript.

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org/education/kidneyweek/2022/ program abstract. aspx? Control Id=3767577

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