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Correlation between glomerular filtration rate produced from the modified gates (*in vivo* method) and the modification of diet in renal disease (*in vitro* method) for patients with renal diseases

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ABSTRACT

Rapid and accurate estimation of the glomerular filtration rate (GFR) is required in the assessment of patients with chronic kidney disease and in order to provide information regarding to the functional status of the kidney. In the present study, the GFR is determined by gamma camera uptake method modified Gates (*in vivo* method) and Modification of Diet in Renal Disease method (MDRD) (*in vitro* method). ^{99m}Tc-DTPA renography is performed by Gates' method (*in vivo*) to obtain GFR and correlated by GFR that predicted by Modification of Diet in Renal Disease equation (*in vitro*) after adjustment different parameters on the evaluation of the glomerular filtration rate of 153 patients with a wide range of renal function, such as: radioactive ^{99m}Tc-DTPA dose that administrated to patient, time of counting pre-injected syringe, post-injected syringe and the distance between syringe and detector of gamma camera. The obtained results show that, comparison of GFR that calculated by Gates' method with that of GFR estimated by MDRD equation in the 153 patients resulted in a significant and good correlation when using the following parameters: administered radioactive dose ranged from 10 to 15 mCi of ^{99m}Tc-DTPA with time of 10 seconds for pre-syringe counts, time of 30 seconds for post-syringe counting and at distance 30 cm from the detector of gamma camera. In addition, the correlation is better in case of chronic renal failure and reduced renal function than in case of healthy group.

KEYWORDS

Glomerular filtration rate (GFR);
^{99m}Tc-DTPA renography;
Gates method, Modification in diet in renal disease method;
Pre-injected syringe count;
Post-injected syringe count.

INTRODUCTION

In accordance with the Kidney Disease Outcomes Quality Initiative (K/DOQI) guidelines, estimates of glomerular filtration rate (GFR) are the best overall indices of the level of renal function[1]. Glomerular filtration rate provides an excellent measure of the filtering capacity of the kidneys. It can be used as an index of functioning renal mass; and changes in GFR which can delineate progression of kidney disease. The level of

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GFR being a strong predictor of the time to onset of kidney failure and the risk of complications of chronic kidney disease (CKD) such as: cardiovascular disease, hypertension, anemia, malnutrition, bone disease, neuropathy, decreased quality of life and death[1,2].

Inulin clearance is proved as the gold standard for glomerular filtration rate (GFR) determination. However; this method is not performed in clinical practice, because of technical complexity and limited availability. The intrinsic creatinine clearance has been widely performed as only alternative to inulin clearance in routine practice. This method, however, is not accurate compared to inulin clearance[3-5].

Urinary clearance of exogenous radioactive markers (^{125}I-iothalamate and ^{99m}Tc-DTPA) and plasma clearance of Iohexol and ^{51}Cr-EDTA have also been used, although these methods are not readily available[6]. Serum cystatin C has been used to estimate the GFR but this method is not in clinical use[7]. Thus, the clearance of endogenous creatinine (Ccr) remains the most commonly used method in estimating the GFR in clinical practice. However, this method is fraught with several shortcomings, first of all the need for timed urine collection. Ccr also overestimates the GFR in renal failure, since serum creatinine is both filtered and secreted by the kidneys.

The determination of serum creatinine is the most widely used and commonly accepted measure of renal function in clinical medicine. Regardless of its widespread use, the accuracy of estimating GFR on the basis of the serum creatinine concentration only is limited, because it is affected by several factors, including body mass, gender, and age. In an attempt to circumvent these limitations, a variety of formulas have been developed, which also take into account age, sex, and body size in their calculation. Among these formulas, the Modification of Diet in Renal Disease equations (MDRD) are widespread, since they are supposed to compensate for the major drawbacks of serum creatinine determination and adequately correlate with GFR measured by the reference method[8].

The gamma camera uptake method with ^{99m}Tc-DTPA is simple and less time consuming for the determination of the glomerular filtration rate[9]. In ^{99m}Tc-DTPA renography, the glomerular filtration rate (GFR) is calculated without blood or urine sampling[10].

Due to limitation of references methods, it is recommended to estimate glomerular filtration rate (GFR) by serum creatinine–based equations[11]. Therefore, simple and accurate determination of the GFR is still a challenge clinically[12]. Estimation of the glomerular filtration rate (GFR) is required in the assessment of patients with chronic kidney disease (CKD) in order to provide information regarding the functional status of the kidneys[13].

In the present study, ^{99m}Tc-DTPA renography is performed by Gates' method (*in vivo*) to obtain glomerular filtration rate (GFR) and correlated by GFR that predicted by Modification of Diet in Renal Disease equation (MDRD) (*in vitro*) after adjustment different parameters on the evaluation of GFR of 153 patients with a wide range of renal function, such as: radioactive ^{99m}Tc-DTPA dose that administrated to patient, time of counting pre-injected syringe, post-injected syringe and the distance between syringe and detector of gamma camera.

SUBJECTS AND METHODS

Patients

In the present work, 153 patients (97 males and 56 females) ranging in age from 18 to 76 years (mean ± SD, 46 ± 14) are included in the study. The patients are referred for evaluation of renal function and pathophysiology in routine practice. They are given a wide variety of clinical diagnosis including chronic renal failure, reduced renal function and healthy persons for donation.

The correlation between the GFR that calculated by Gates' method and GFR that estimated by MDRD equation by the new protocol after adjustment the different parameters - administered radioactive dose range 10-15 mCi of ^{99m}Tc-DTPA with time of 10 seconds for pre-syringe counts, time 30 seconds for post-syringe counting and distance 30 cm from the detector of gamma camera - on 153 patients suspected of having different renal diseases are referred to Nuclear Medicine Department of King Fahd Centre, Cairo University Hospitals, Egypt.

Calculation of GFR by gates method

^{99m}Tc-DTPA was prepared in Radioisotope Laboratories in King Fahd Center, Cairo University Hospitals, Egypt, using a commercially available freeze-dried kit[14,15]. The dose is ranged from 10 to 15 mCi and is

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administered to 153 patients with different renal disease and healthy persons. Prior to the administration, the pre-injection syringe with straight needle is counted by two different devices: (1) Dose calibrator (ATOMLAB 100) and (2) Gamma camera (Siemens, Orbit, Single head), which is attached to a Low-Energy General-Purpose Parallel-Hole Collimator for 10 seconds. The patient is hydrated with 300-500 ml of water 30 minutes prior to the examination. The patient lay down on a bed in the supine position at distance 30 cm from the detector of gamma camera and the image will acquired a posterior except one patient with ectopic kidney lay down on a bed in the prone position. ^{99m}Tc-DTPA is given through a butterfly needle into vein and is followed by infusion of 20 ml of normal saline then 2 ml lasix. Frames of 128 × 128 matrix are recorded with an online-computer, initially at one second for one minute and then at 10 seconds for 20 minutes. The post-injection syringe with a straight needle which is detached before the injection is again counted 30 seconds by a gamma camera in the same way as pre-injection. Region of interest (ROI) over each kidney is assigned manually on the frame added from 1 to 3 minutes following injection. The semi-lunar background ROI around each kidney was defined. The background corrected time-activity curve is generated, and the renal uptake of individual kidney for one minute from 2 to 3 minutes after the injection is calculated. The GFR (GFR Gates) is automatically estimated by a commercially available computer (Oddesey Pegasis Labratorias, Adac) according to the Gates' algorithm.

Calculation of GFR by MDRD equation

For measuring serum creatinine, it was withdrawn 3 ml sample of blood from patients. The serum creatinine is done on Auto Analyzer Model Hitachi 912 (Japan) and using the simplified MDRD equation[16]:

$$\text{eGFR (ml/min/1.73 m}^2\text{)} = 186.3 \times [\text{serum creatinine (mg/dl)}]^{-1.154} \times [\text{age (years)}]^{-0.203} \times [0.742 \text{ (female) or } 1.210 \text{ (black)}] \quad (1)$$

The simplified MDRD equation allows the classification renal function with acceptable precision and requires only minimal information about the patient. It has therefore been included as the primary GFR marker in the Practice Guide-lines for Chronic Kidney Disease, published in 2002 by the Kidney Disease Outcomes Quality Initiative of the National Kidney Foundation (K/DOQI) and the more recent KDIGO guidelines[1,2].

Classification of practical results

The practical results are classified into 3 categories: The first category contains patients which suffering from chronic renal failure while the second category contains people that come to King Fahd Centre, Cairo University Hospitals, Egypt, to donate. In the third category, there are patients which suffering from reduced renal function. The first category consists of 49 patients of serum creatinine ranging from more than 1.6 to 9.5 mg/dl. GFR *in vivo* is measured by gamma camera in Nuclear Medicine Department of King Fahd Centre, Cairo University Hospitals, Egypt, by injection of radioactive dose (^{99m}Tc-DTPA) ranging from 10 to 15 mCi that given through butterfly in vein. While the GFR *in vitro* is carried out by chemical analysis by taking blood samples from patients then measure the serum creatinine in mg/dl. Then substitute by serum creatinine and age of patient in MDRD equation to measure GFR *in vitro*. For the second category – 19 patients come to Fahd Center to donate - the same practical work was done. These patients have normal serum creatinine ranging from 0.4 to 1.5 mg/dl. The GFR *in vivo* is ranging from 80-130 ml/min while the GFR *in vitro* are more than 80 ml/min. For the third category, 85 patients suffering from reduced renal function, the GFR *in vivo* is measured by gamma camera and the GFR *in vitro* that calculated by MDRD equation is measured by the previous method as in case of renal failure and donors. The serum creatinine of these patients is ranging from 0.4 to 1.5 mg/dl like normal patients.

Statistical methods

Statistical analysis of the results are performed by using the Analysis of Variance (ANOVA) to determine the effect of radioactive doses, time of counting and distance between the detector of gamma camera and the syringe and their interaction on glomerular filtration rate (GFR), means at significance level of 0.05. Correlation and regression analyses are also estimated to compute the correlation coefficient (R) for the GFR that measured by Gate's method and GFR that calculated by MDRD equation. All statistics and illustrations (scatter plots) are carried out using Statistical Analysis Systems[17] program Ver. 9.1, SAS Institute in Corporation Cary, NC 27513 USA.

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RESULTS

Patient characteristics after adjustment the parameters

One hundred and fifty three (153) patients (97 males and 56 females) ranging in age from 18 to 76 years with varying levels of kidney function are studied. The patients are referred to Nuclear Medicine Department, Fahd Center, Faculty of Medicine, Cairo University, Egypt, for evaluation of renal function in routine practice. They are given a wide variety of clinical diagnosis including 49 chronic renal failure, 85 reduced renal function and 19 healthy persons that come to the center for donation. TABLE 1 contains the mean age of patient in years, the mean weight in kg, the mean body mass index and the mean serum creatinine in mg/dl (the serum creatinine of each patient ranged from 0.48 to 7.74 mg/dl).

Glomerular filtration rate

The correlation between GFRs which measured by the modified Gates (*in vivo* method) in ml/min/1.73 m^2 and the GFRs determined by Modification of Diet in Renal Disease equation (MDRD) (*in vitro* method) in ml/min/1.73 m^2 for 153 patients is shown in Figure 1. From the figure, it is clear that, a linear correlation between modified Gates' and MDRD–predicted GFR is detected and the regression equation is: $y = 1.167 x + 7.689$ ($R = 0.876$, $p < 0.0001$). This means that the *in vivo* method correlates well with that of the *in vitro* method after the adjustment of the parameters that affect on GFR *in vivo*.

The difference in GFRs measured by the modified Gates (*in vivo* method) and the GFRs determined by Modification of Diet in Renal Disease equation (MDRD) (*in vitro* method) in ml/min/1.73 m^2 against the mean GFR of the two methods for 153 patients after adjustment the different parameters is shown in Figure 2. Difference in the GFR (GFR_{MDRD} - GFR_{gates}) was 18.21 ±

TABLE 1 : Clinical parameters for 153 patients with different renal diseases

Parameter	Mean ± SD
Age, years	45.90 ± 14.00
Weight, kg	74.84 ± 17.64
Body mass index, kg/m^2	27.80 ± 6.20
Serum creatinine, mg/dl	1.69 ± 1.33
Mean GFR *in vivo*, ml/min/1.73 m^2	62.94 ± 33.00
Mean GFR *in vitro*, ml/min/1.73 m^2	81.15 ± 43.97

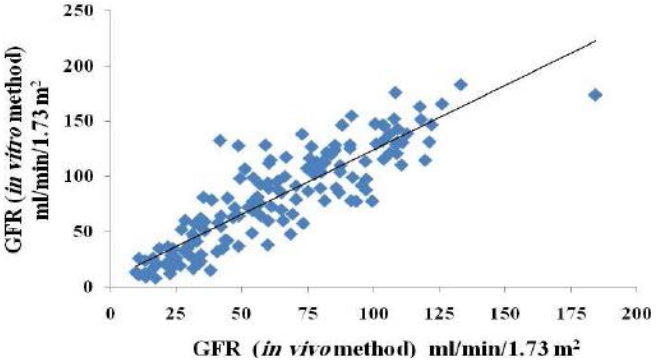

Figure 1: Scatter plots of GFRs determined by the modified Gates (*in vivo* method) against that calculated by MDRD equation (*in vitro* method) after adjustment the parameters that affect on GFR *in vivo* in 153 studied patients with different renal diseases.

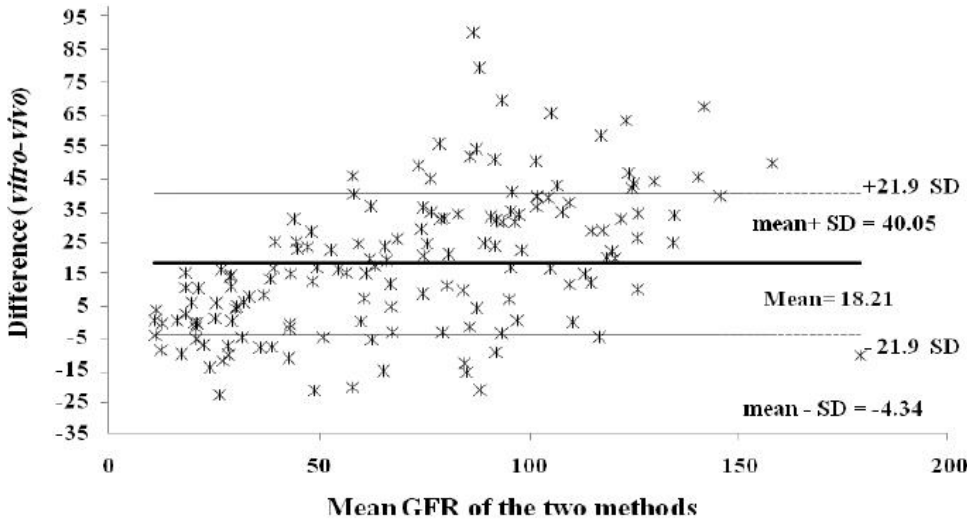

Figure 2 : Plots showing the difference in GFRs by the MDRD equation method (*in vitro*) and the modified Gates' method (*in vivo*) against the mean GFR of the two methods after adjustment of the parameters that affect on GFR.

21.9 ml/min/1.73 m^2. This means that MDRD results are much higher than that obtained with modified gates. Indeed, in most subjects in 115 out 153, the MDRD is higher than that of the modified gates. In these individuals, the MDRD-gates difference has positive values.

To study the best correlation between GFR that measured by gamma camera and the GFR that calculated by MDRD equation in 153 patients, the practical results classified into 3 categories to show the best correlation in which category.

The first category: Glomerular filtration rate for renal failure patients

In the first category, the serum creatinine of each patient is ranging from more than 1.6 to 9.5 mg/dl. The correlation between GFRs measured by the modified Gates (*in vivo* method) that measured in ml/min/1.73 m^2 and the GFRs determined by Modification of Diet in Renal Disease equation (MDRD) (*in vitro* method) in ml/min/1.73 m^2 in 49 patients is shown in Figure 3. From the figure, it is clear that, a linear correlation between modified Gates' and MDRD–predicted GFR is detected and the regression equation is: $y = 0.642 x + 11.24$ ($R = 0.66$, $p < 0.0001$). This means that the *in vivo* method correlates with that of the *in vitro* method after the adjustment of the parameters that affect on GFR *in vivo* in renal failure patients.

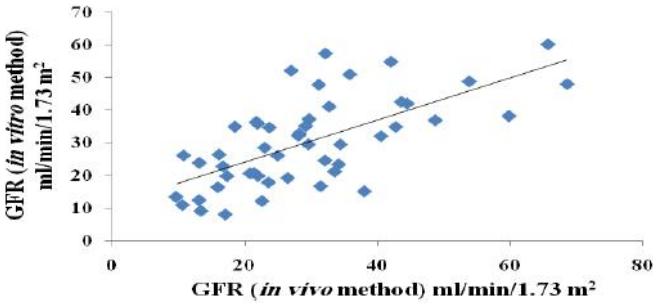

Figure 3 : Scatter plots of GFRs determined by the modified Gates (*in vivo* method) against that calculated by MDRD equation (*in vitro* method) in 49 patients of renal failure.

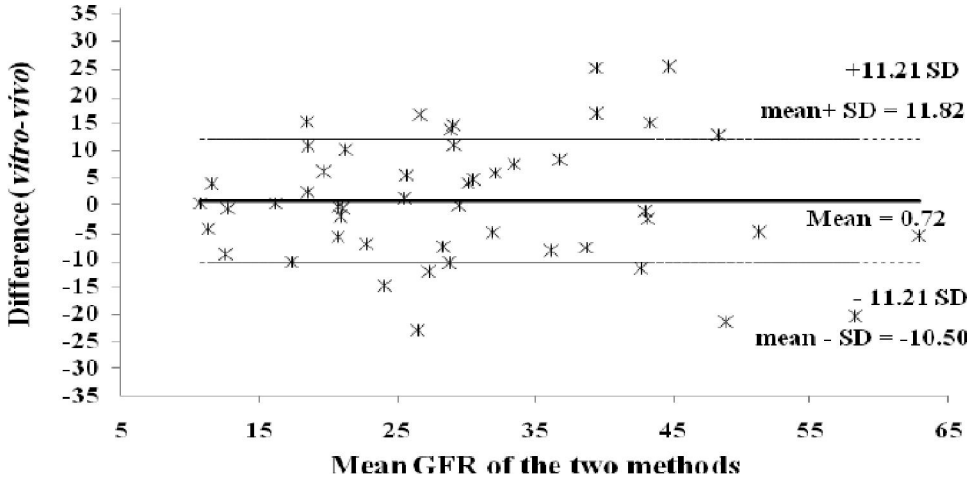

Figure 4 : Plots showing the difference in GFRs by the MDRD equation method (*in vitro* method) and the modified Gates' method (*in vivo*) against the mean GFR of the two methods after adjustment of the parameters that affect on GFR in 49 patients of renal failure.

The difference in GFRs measured by the modified Gates' (*in vivo* method) and the GFRs determined by Modification of Diet in Renal Disease equation (MDRD) (*in vitro* method) in ml/min/1.73 m^2 against the mean GFR of the two methods for 49 patients that suffering of chronic renal failure is shown in Figure 4. Difference in the GFR (GFR_{MDRD}-GFR_{gates}) was 0.72 ± 11.21 ml/min/1.73 m^2. This means that MDRD results are not higher than that obtained with modified gates in most patients that suffering of renal failure. Indeed, in fewer subjects (in 24 out 49) the MDRD is higher than that of the modified gates in case of renal failure.

The second category: Glomerular filtration rate for healthy people

In the second category (people that come to Fahd Center to donate), the serum creatinine is ranging from 0.4 to 1.5 mg/dl. The GFR (*in vivo*) is ranging from 80 to 130 ml/min/1.73 m^2 whiles the GFR (*in vitro*) is more than 80 ml/min/1.73 m^2.

The correlation between GFRs measured by the modified Gates (*in vivo* method) that measured in ml/min/1.73 m^2 and the GFRs determined by Modification of Diet in Renal Disease equation (MDRD) (*in vitro* method) in ml/min/1.73 m^2 for 19 healthy people is

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shown in Figure 5. From the figure, it is clear that, a linear correlation between modified Gates' and MDRD–predicted GFR is detected and the regression equation is: y = 0.673 x + 59.11 (R = 0.628, p = 0.004). This means that the *in vivo* method correlates with that of the *in vitro* method after the adjustment of the parameters that affect on GFR *in vivo* in donor's people.

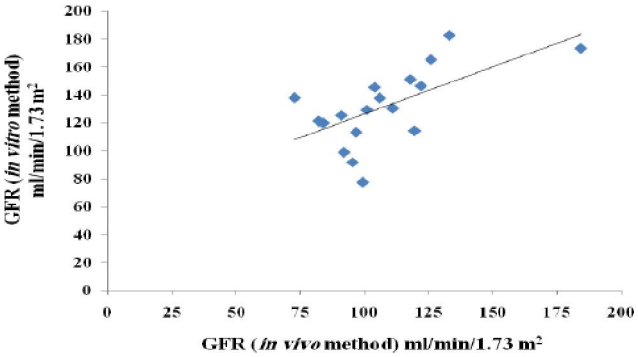

Figure 5 : Scatter plots of GFRs determined by the modified Gates (*in vivo* method) against that by the Modification of Diet in Renal Disease equation (MDRD) (*in vitro* method) in 19 donor's people.

The difference in GFRs measured by the modified Gates (*in vivo* method) and the GFRs determined by modification of diet in renal disease equation (MDRD) (*in vitro* method) in ml/min/1.73m^2 against the mean GFR of the two methods for 19 donors' people is shown in Figure 6. The difference in the GFR (GFR_{MDRD} - GFR_{gates}) is 24.40 ± 22.41 ml/min/1.73m^2. This means that MDRD results were much higher than that obtained with modified gates in donor's people. Indeed, in most subjects (in 15 out 19) the MDRD is higher than that of the modified gates. In these individuals, the MDRD-gates difference has positive values. In healthy people the GFR that obtained by MDRD equation always higher than that obtained by modified gates.

The third category: Glomerular filtration rate for reduced renal function

In reduced renal function of 85 patients (category 3), the GFR (*in vivo*) is measured by gamma camera and the GFR (*in vitro*) that calculated by MDRD equation as in cases of chronic renal failure (category 1) and donors people (category 2). The serum creatinine of is ranging from 0.4 to 1.5 mg/dl like healthy people.

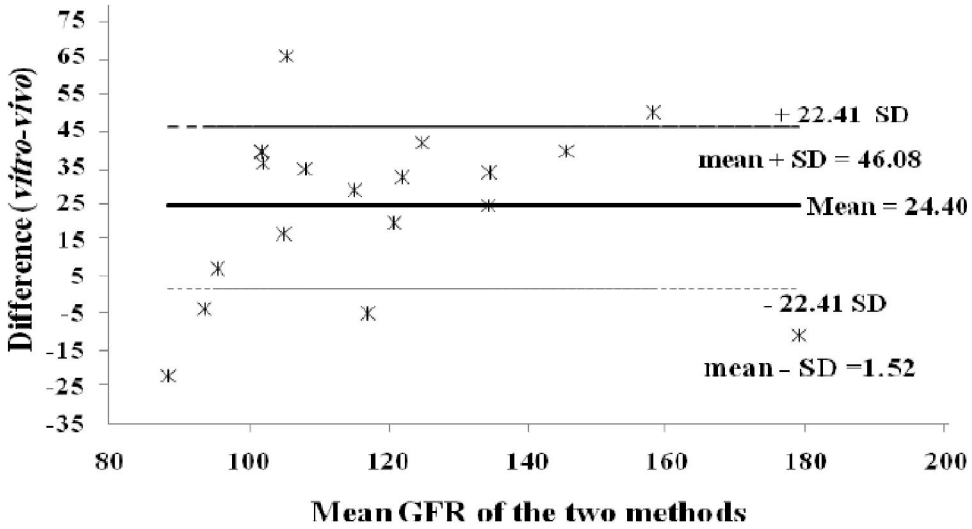

Figure 6 : Plots showing the difference in GFRs by the MDRD equation method (*in vitro* method) and the modified Gates' method (*in vivo*) against the mean GFR of the two methods after adjustment of the parameters that affect on GFR in 19 healthy donor's people.

The correlation between GFRs measured by the modified Gates (*in vivo* method) that measured by ml/min/1.73 m^2 and the GFRs determined by Modification of Diet in Renal Disease equation (MDRD) (*in vitro* method) in ml/min/1.73 m^2 for 85 patients that have moderate renal function is shown in Figure 7. From the figure, it is clear that, a linear correlation between modified Gates' and MDRD–predicted GFR is detected and the regression equation is: y = 0.864 x + 36.75 (R = 0.706, p < 0.0001). This means that the *in vivo* method correlates well with that of the *in vitro* method after the adjustment of the parameters that affect on GFR *in vivo* in patients that have moderate renal function.

The difference in GFRs measured by the modified Gates (*in vivo* method) and the GFRs determined by MDRD equation (*in vitro* method) in ml/min/1.73m^2 against the mean GFR of the two methods for 85 pa-

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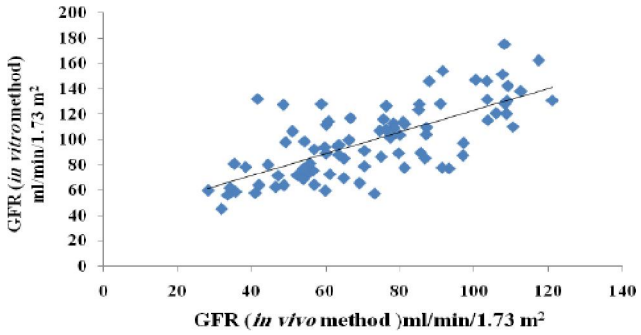

Figure 7 : Scatter plots of GFRs determined by the modified Gates (*in vivo* method) against that by the Modification of Diet in Renal Disease equation (MDRD) (*in vitro* method) in 85 patients of moderate renal function.

tients that have moderate renal function is shown in Figure 8. The difference in the GFR (GFR_{MDRD} - GFR_{gates}) is 26.91 ± 20.61 ml/min/1.73m^2. This means that MDRD results are much higher than that obtained with modified gates in patients with moderate renal function. Indeed, in most subjects (in 76 out 85) the MDRD iss higher than modified gates. In these individuals, the MDRD-gates difference has positive values.

DISCUSSION

Estimation of the glomerular filtration rate (GFR) is

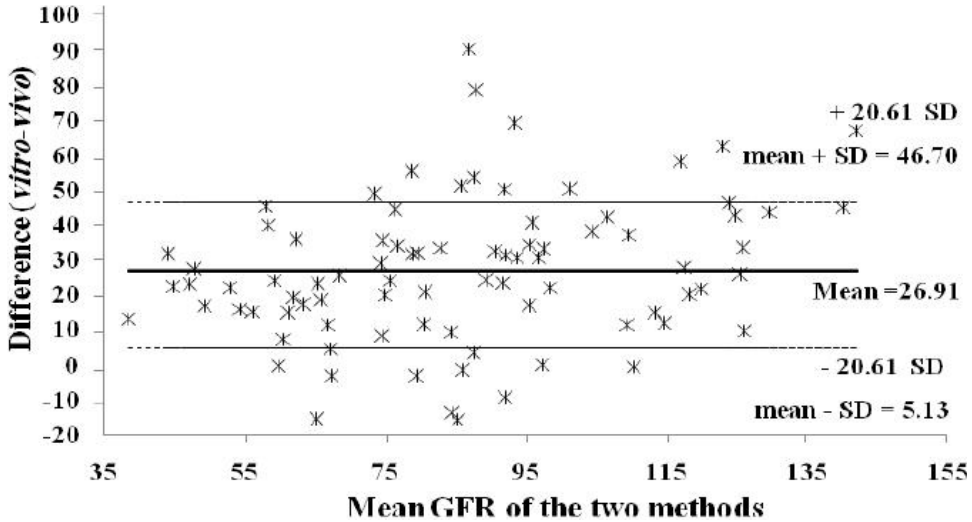

Figure 8 : Plots showing the difference in GFRs by the MDRD equation method (*in vitro* method) and the modified Gates' method (*in vivo*) against the mean GFR of the two methods after adjustment of the parameters that affect on GFR in 85 patients of moderate renal function.

required in the assessment of patients with chronic kidney disease (CKD) in order to provide information regarding the functional status of the kidneys. Current guidelines advocate the use of prediction equations, such as the Cockcroft–Gault (CG) formula and the Modification of Diet in Renal Disease (MDRD) study-derived equations, over clearance of endogenous creatinine (Ccr) in achieving this aim[13-15]. The Gates correlated well with the plasma sample method. The significant correlation of the renal uptake of ^{99m}Tc-DTPA against the 24-hours creatinine clearance has promoted this method for clinical application in routine practice[18]. However; the Gates was proved to be inaccurate and less precise than the CG for predicting the GFR. In addition, the Gates tended to overestimate the GFR. These results were consistent with previous reports[19,20]. It has debated whether the Gates' method is accurate for predicting the GFR[21]. Several sources of errors in the estimation of GFR by scintigraphy are recognized: background correction, decay statistics, attenuation correction, and estimation of arterial plasma activity, volume measurements and radiopharmaceutical quality[22].

Even if ^{99m}Tc-DTPA renography is not precise as a measurement of global renal function, it provides notable information such as quantitative individual renal function and pathophysiological changes of the kidney in renovascular hypertension, hydronephrosis and renal transplant. It is suggested that isotopic renography is likely to be overtaken by competing technologies which can provide one test to give simultaneous information about both structure and function[23].

There are other non-physical parameters like age, body mass index (BMI), serum creatinine and gender may affect on the measurements of glomerular filtration rate (GFR). It is found that there are strong correla-

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tions between the serum creatinine in mg/dl and the GFR. In the same time there is very weak correlation between the age and measurement of GFR while there is not correlation between BMI and measurement of GFR and the gender does not effect on the GFR *in vivo* value.

In the present study, studying the different factors that affect on the GFR measurement, the others that have not effect on GFR; and the GFR that measured by Gates method (*in vivo* method) are well correlated with the GFR that calculated by MDRD equation (*in vitro* method) (R H" 0.71) for 153 patients with different renal diseases after adjustment radioactive dose, time of counting and distance between the detector of gamma camera and syringe.

In conclusion, after adjustment the different parameters such as: radioactive ^{99m}Tc-DTPA for patient, time of counting of radioactive syringe and distance between syringe and detector of gamma camera, this study will optimize Gates technique to make it possible to compute accurate GFR coincident with scan of kidney. It was found that, the ^{99m}Tc-DTPA renography will become more accurate in measurement of GFR, if these parameters are corrected and thus measurement of global renal function become more precise. Moreover, in case of classification of 153 patients into three categories according to serum creatinine and GFR, it is found that the correlation between the two methods is very strong in case of renal failure patients and reduced renal function, but the correlation is not strong in case of donor's people.

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