,,OVIDIUS" UNIVERSITY CONSTANŢA

CSUD-DOCTORAL SCHOOL OF MEDICINE

THESIS

Hemorheological aspects in diabetic nephropathy

SUMMARY OF THE DOCTORAL THESIS

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Keywords: diabetic nephropathy, hemorrheologic, blood viscosity, erythrocyte aggregability, deformability of red cells, mathematical modeling

Note: Figures and tables inserted in the summary of the thesis retain the original numbering of the paper presented in extenso.The table of contents is also similary with of the thesis.

INTRODUCTION

Diabetes mellitus (DM) is a metabolic disease that has one of the highest prevalence in industrialized countries and is also a public health problem affecting these countries. The prevalence of diabetes in 2014 was about 422 million impairment of individuals globally representing approximately 8.5% of the population, requiring spending about 612 billion. Diabetes tends to be a pandemic in recent years and that any attempt in medical research that can prevent this disease and its many complications that develops can be helpful.

Wilfrid G.Oakley ,, said that man may be master of his fate, and the victim's blood sugar, and this does seem to be included about half a billion patients worldwide and in Romania the number is about two million. Any change in the microcirculation cause or worsen diabetes complications. Therefore, the proposed study sought to highlight the behavior hemorrheologic diabetic patients with renal disease at different stages in an attempt to answer a question that is can reduce the progression of diabetic kidney disease when hemorrheologic factors are known?

It should also be stressed that diabetes costituie leading cause of death worldwide. Diabetic nephropathy is the first cause of terminal chronic renal failure (IRCT) in developed countries. Concerns regarding quality of life and care of patients with kidney disease are becoming increasingly important in all countries. In recent years significant progress has been made regarding hemorrheologic. Here we speak not only the emergence of new techniques rheological measurements as accurate, but also a better understanding of the phenomena associated with this feature blood. Hemorrheologic characteristics are closely linked to both the interactions and the mechanical properties of red blood cells, and can influence a change in the disease.

Following the studies it was noted that not all patients can develop diabetic microvascular complications, especially talking about the kidney, even if blood glucose levels was trying to be totally controlled. The same unpredictable evolution can shape and when switching from different stages of diabetic nephropathy to end chronic renal failure because some individuals are more likely to develop these complications though he was maintaining normal blood sugar levels.

Do not rheologic factors can influence the progression of this disease? This was the motivation from which I began this study. I tried to bring a new approach to diabetic pathology and renal microvascular damage mainly of legally hemorrheologic.

Demonstration implications hemorrheologic characteristics would be a useful way, low-observable in the development and emergence of diabetic nephropathy. That was why we conducted this study.

And last but not least, the low number of national studies regarding the involvement of hemorelogici in various diseases, especially regarding diabetes and its complications microvascular kidney led me to choose this subject in an attempt to use this research still niche to portray pathologic mechanism of diabetic renal disease.

The thesis is divided into two parts, namely Part I showing noţini theoretical current that supported the importance of the theme in terms of diabetic nephropathy (Chapter I), and notions current hemorrheologic (Chapter II) and second part , comprising the personal contribution that is material and methods (Chapter III), results (Chapter IV) and discussions (Chapter V) and conclusions (Chapter V), which went from carrying out scientific research.

Research directions and objectives

The primary motivation of this work is given on the one hand by the increasing incidence of patients diagnosed with diabetes despite specific therapies arrive develop microvascular complications, referring mainly to diabetic retinopathy and diabetic nephropathy and the other their evolution to major events.

Any vision and any new approach can bring multiple benefits to both physicians and quality of life.

Finally, the small number of national studies dealing with in terms hemorrheologic diabetic patients and especially those with diabetic nephropathy, strengthened motivation to achieve this study.

The main objectives were: the description of the clinical and demographic groups of patients (age, gender, origin, duration of illness, etc.); Description of hematological, biochemical reveal that renal damage;

3. Analysis of blood rheologic parameters, represented by blood viscosity, plasma viscosity, and red cell aggregation in the deformability of red blood cells, in conjunction with hematological and biochemical parameters, and the establishment of correlations between these values obtained.

4. Mathematical modeling helps predict how the deformable particles and aggregates such as erythrocytes behave the blood flow and also offers great insights into how the viscosity can turn

these deformable particles and how it can also disrupt the flow sanguin.De we tried as recognized mathematical models to verify the determination indirect and add new correlations - especially those of renal impairment, Germany, mainly to assess how this parameter affects blood flow characteristics.

The general methodology of research

The study was a research descriptive and controlled clinical trial, which focused on the one hand the description of the clinical and biological characteristics of the patients and the control group, and on the other hand the behavior of hemorrheologic both patients with diabetic nephropathy and those in group martor.Studiul proposed included a total of 42 diabetic patients with chronic kidney disease at different stages of evolution, patients in family medicine cabinet records, patients admitted to the Emergency Hospital "St. Andrew "Constanta and also patients hospitalized in the Dialysis Diaverum Constanta.

The study population was separated into three groups of diabetic patients with varying degrees of renal impairment with the following distribution:

- Lot. 1, which includes 10 diabetic patients with mild renal impairment, involving classes with 0/1 stage chronic renal disease with a GFR> 90ml / min / 1.73m2 and those with chronic kidney disease in a GFR stage 2 60 - 90ml / min / 1.73m^2;
- Lot. 2, which includes 18 patients with diabetic nephropathy and diabetic degree of mild and moderate renal impairment corresponding to a GFR of 30 - 60 ml / min / 1.73m^2;
- Lot. 3, which includes 14 patients with severe kidney disease or chronic renal failure or end-stage chronic severe with a GFR <30 ml / min / 1.73m^2.

Inclusion criteria: Caucasians, patients diagnosed with type 2 diabetes, with varying degrees of renal impairment, the patient's consent to cooperate in conducting the study by sampling, and use of data by signing consimţământului informed.

The criteria for exclusion were represented by: signing the consent of the patient refusal, acute metabolic disorders; in the list of permanent presence of potentially nephrotoxic drugs; where the diagnosis of hypertension preceding the diagnosis of chronic renal diabetic; patients with associated urinary tract infection; febrile patients or patients suffering from other acute infections.

For the accuracy of the results was used a control group from the general population, consisting of 21 healthy individuals who have given their consent to personal data and the results of biological samples, selected in accordance with the basic features of the control group, having

the following exclusion criteria: abnormal glucose measured dynamic curve; renal pathology independent of diabetes; patients with hepatic pathology; use of medication; acute or chronic inflammatory disease.

Informed consent was read and signed by each participant in the study, in knowingly, after being informed on all aspects relevant in terms of achievement. The study was conducted in accordance with ethical principles, following the Declaration of Helsinki and was consistent with clinical practice guidelines and regulations both national and international laws respecting the right to confindenţialitate, integrity and ability of the patient to always opt to participate in the study.

STATISTICAL ANALYSIS

For qualitative and quantitative analysis of data from patients enrolled in the study, we used Microsoft Excel and SPSS 20.0. Primary data processing and to obtain descriptive indicators for calculating averages, standard deviation and graphing were used statistical functions of Microsoft Excel. *T - student* test was used to compare the dependent variables. It is considered statistically significant at a p value <0.05.Pentru assess the correlation Simple linear regression was used and also lineară.The multiple regression ANOVA test was used for determining specific linear regression relationship.

RESULTS

The average age of patients was 66.52 +/- 7.71 years, with a report cvasiomogen women / men 1.62 representing a total of 26 women and 16 men, seeing a slightly higher incidence among women (up 23.82 % to men).

Table no.X Parameters hematogical of lots study

Haematological parameters	**VALUES Lot 1**	**VALUES Lot 2**	**VALUES Lot 3**	**VALUES control group**	**p**
Hb mean +/- SD (g / dL)	14.5 +/- 1.68	13.13 +/- 1.01	9.54 +/- 2.01	+/- 14.79 0.74	.5588 0.0001 <0.0001
Ht average +/- SD (%)	44.28 +/- 3.86	+/- 39.29 2.39	+/- 28.47 6.51	+/- 45.06 1.70	.5218 <0.0001 <0.0001

CHEM mean +/- SD (%)	33.74 +/- 1.63	33.4 +/- 0.75	33 +/- 1.77	33 +/- 0.76	.1641 .2199 .9908
VEM average +/- SD (fL)	88.2 +/- 2.43	+/- 90.03 6.08	+/- 90.21 3.6	+/- 89.81 1.98	.1396 .8663 .7237
Leukocytes Mean +/- SD (elem / mm3)	7324 + / 2931.02	7812.22 + / 2038.87	9611.57 +/- 3696.72	6603.3 +/- 1382.1	.8574 .0797 .0059
Platelets average +/- SD (elem / mm3)	223 600 +/- 36018.05	+/- 224,444.44 33604.72	215,571.43 +/- 72716.01	282 500 +/- 42973.65	0.0109 .0016 .0086
ESR average +/- SD (mm / hr)	16.6 +/- 8.47	24.55 +/- 13.45	+/- 57.28 27.25	+/- 8.5 3.05	0.0022 <0.0001 <0.0001
Fibrinogen Mean +/- SD (mg / dl)	446.6 +/- 72.78	420.2 +/- 120.80	651.9 +/- 53.32	+/- 297.22 53.70	<0.0001 0001 <0.0001

From Table no.X, which are described hematological parameters we can see that the hemoglobin in the three groups was 14.5 +/- 1.687 for group 1, of 13.13 +/- 1.01 for patients in group 2 and 9.54 +/- 2.01 for group 3, with statistically significant level, especially for lots 2 and 3 (p = 0.0001, respectively p <0.0001 compared to control group) of mean hematocrit .Valorile were 44.28 +/- 3.86, 39.29 + / - 2.39, 28.47 +/- 6.51, 45.06 +/- 1.70 (almost similar values between group 1 and control group - p = 0.5218, but with the important meaning from the other two groups with severe renal impairment and the control group < 0.0001). Inflammation test values, namely those of ESR and fibrinogen.

Table no.XI Parameters indicating impaired renal of lots study

Renal parameters	**VALUES lot 1**	**VALUES lot2**	**VALUES lot 3**	**VALUES control group**	**p**
BUN mean +/- SD (mg / dll)	35.6 +/- 13.75	+ 49.65 / 13.62	108.9 +/- 44.92	33.38 + / 5.41	.5776 0.0001 <0.0001

Creatinine Mean +/- SD (mg / dL)	0.92 +/- 0.18	1.61 +/- 0.47	3.74 +/- 1.48	0.85 +/- 0.10	.3262 <0.0001 <0.0001
GFR average +/- SD (ml / min /1.73m^2)	+ 71.28 / 13.71	46.81 +/- 9.04	19.28 +/- 5.64	104.55 + / 15.87	0.0003 <0.0001 <0.0001

The true measure of renal impairment, ie glomerular filtration rate, show significantly lower values in all groups of patients, with values of p = 0.0003 for group 1, p <0.0001pentru group 2, respectively p <0.0001 for group 3.

Glucose levels for patients in the control group were significantly lower than those of batches diabetic patients (p <0.0001), but differences between the three groups have shown poor target in terms of statistical significance (p =0.46, 0.23, and 0.28, respectively).

Table nr.XII Carbohydrate and lipid metabolism parameters of lots study

Parameters of carbohydrate metabolism	**VALUES lot 1**	**VALUES lot 2**	**VALUES lot 3**	**VALUES control group**	**p**
Blood glucose Mean +/- SD (mg / dL)	158 + / 39.53	159.55 +/- 28.89	172.28 +/- 41.17	90.25 +/- 5.67	<0.0001 <0.0001 <0.0001
HbA1c Mean +/- SD (%)	6.8 +/- 0.6	7.92 +/- 1.18	9.31 +/- 2.14	5.36 +/- 0.33	<0.0001 <0.0001 <0.0001
Cholesterol total Mean +/- SD (mg / dL)	184.6 +/- 50.15	151.625 +/- 44.88	138 +/- 20.38	180.22 +/- 25.33	.8291 .1210 .0029

In terms of the values of glycated hemoglobin (as can be seen from Table XII), it is noted statistically significant differences between groups of patients and the control group (p <0.001), slightly poor if the comparison between the three groups (p = 0.037468, 0.060227, 0.015235).

Blood viscosity - results

Blood viscosity values obtained at different shear rates as described in Table started from 39.92 +/- 6.38 cP, a value obtained at a low shear rate of 18.8 sec -1, which is found in accordance with the studies have shown that elevated in diabetic patients.

Table nr.XIII The values of blood viscosity in patients

Shear rate	**Viscosity lot 1 Mean +/- SD (CP)**	**Viscosity lot 2 Mean +/- SD (CP)**	**Viscosity lot 3 Mean +/- SD (CP)**	**Viscosity control group Mean +/- SD (CP)**	**p**
18.8 sec $^{-1}$	15.04 +/- 5.44	22.95 +/- 3.13	39.92 +/- 6.38	-	
22.5 sec $^{-1}$	13.31 +/- 3.66	22 +/- 9.76	22.15 +/- 3.99	8.82 +/- 3.53	**0.05** **0.08** **0.05**
30 sec $^{-1}$	10.53 +/- 2.70	17.29 +/- 14.09	17.42 +/- 10.67	8,77+ -2.34	0.34 0.24 0.21
37.5 sec $^{-1}$	8.34 +/- 3.14	10.87 +/- 6.41	16.99 +/- 3.83	8.57 +/- 2.86	0.39 0.48 0.16
45 sec $^{-1}$	7.82 +/- 3.20	9.27 +/- 4.23	12.83 +/- 1.09	2.4 +/- 7.65	0.46 0.19 0.17
60 sec $^{-1}$	6.97 +/- 3.75	8.52 +/- 4.77	13.86 +/- 3.32	5.94 +/- 1.74	0.28 0.11 0.13
75 sec $^{-1}$	6.62 +/- 3.70	6.97 +/- 3.88	11.48 +/- 1.43	5.04 +/- 1.22	**0.08** **0.06** **0.04**
150 sec $^{-1}$	5.31 +/- 2.18	6.16 +/- 3.39	11.27 +/- 6.5	3.4 +/- 1.55	**0.02** **0,005** **0.011**
225 sec $^{-1}$	5.73 +/- 1.53	4.63 +/- 1.48	5.39 +/- 4.47	3.00 +/- 1.26	**0.0003** **0.0036** **0.055**
300 sec $^{-1}$	5.0 +/- 1.14	3.69 +/- 0.62	2.9 +/- 4.24	2.79 +/- 1.12	**0.0004** **0.023** **0.08**
450 sec $^{-1}$	4.21 +/- 0.71	3.26 +/- 0.68	2.05 +/- 0.14	2.49 +/- 0.92	**0.0004** **0.02** 0.46
600 sec $^{-1}$	3.42 +/- 0.13	3.12 +/- 0.87	1.92 +/- 0.21	2.36 +/- 0.82	**0.05** **0.02** 0.15

The shear rate of 100 sec $^{-1}$ range up to 200 s $^{-1}$, the viscosity of the blood measured at a temperature of 370C is about 4-5 mPa.s or centipoise, and is relatively insensitive to any increase above this limit shear .

In our study, no statistically significant differences between the studied groups were made up to a shear rate of 450 sec $^{-1}$.

Analyzing the relationship between blood viscosity and haematological parameters and the renal damage most important - the number of leukocytes and platelets, fibrinogen, glucose, urea, ESR, creatinine rate glomerular filtrate - watched highlighting a correlation indicating its contribution (variable independent) value determined blood viscosity.

Table no.XIV Statistical correlations (R^2) for the whole group of patients

Parameters correlated	**Low shear rates (Under 100 sec^{-1})**	**High shear rates (100 sec^{-1})**
Fibrinogen	**0333**	**0370**
Leukocyte	0029	0010
Platelets	**0124**	0040
Glycemia	**0132**	**0165**
BUN	**0438**	**0164**
GFR	**0334**	**0311**

From the analysis results shown in Table no.X blood viscosity existence of dependencies to the following parameters of the 6 analyzed, namely fibrinogen, BUN, GFR, glucose.

At low shear rates there is a good correlation of fibrinogen, urea and glomerular filtrate rate with the value of the correlation coefficient R^2= 0.333 fibrinogen (see Annex 2) and a value of correlation R^2= Urea 0.438, R^2= 0334 to GFR, and less close correlations, but worthy of mention, the platelet count and blood glucose, R^2 = 0.124 and 0.132 respectively.

At high shear rates correlation with the fibrinogen is the most closely - R^2 = 0.37, and the GFR, with a close correlation value, i.e. R^2 = 0.3113. Other parameters specific for urea and glucose, correlations were weak - R^2 = 0.1653, 0.1641.

From the analysis performed it can be seen that the number of white blood cells is not correlated with any of the values of viscosity at shear rates studied.

Correlations with blood viscosity parameter erythrocyte

Because red blood cell parameters of particular importance in terms of rheological behavior of blood is shown in Table. correlation of these parameters with the values of all diabetic patients blood viscosity at different shear rates.

Table nr.XV Correlations between blood viscosity and erythrocyte parameters in diabetic patients

Viscosity according Shear rate	Spearman's rho	Hb	Spearman's rho	Ht	Spearman's rho	ESR
30 sec $^{-1}$	.	p = 0.969		**p = 0.037**	-	-
225 sec $^{-1}$	**0489**	**p = 0.025**	**0605**	**p = 0.004**	**-0.498**	**p = 0.022**
300 sec $^{-1}$	**0511**	**p = 0.021**	**0633**	**p = 0.003**	-	p = 0.12
450 sec $^{-1}$	**0697**	**p = 0.001**	**0794**	**p <0.0001**	**-0.681**	**p = 0.002**
600 sec $^{-1}$	**0590**	**p = 0.021**	**0545**	**0036**	**-0.547**	**p = 0.035**

Through a simple correlation method in the general group of diabetic patients with renal there is a close connection to both the values of hemoglobin, hematocrit values and ESR values, especially at high shear rates. The correlations were statistically significant for all three parameters described high shear rates of 225, 450 and 600 sec $^{-1}$, at low rates while no correlation only with the hematocrit (p = 0.037). For the ESR significant correlation is observed, but the reverse high shear rates (-0.498, -0.681, -0.547).

Regarding erythrocyte indices were observed little correlation. And these correlations were only CHEM - more specifically in the group 3 with severe renal disease, where the values of viscosity obtained at high shear rates (225sec^{-1} and 300 sec^{-1}) had a significant correlation - p = 0.027, p = 0.021, respectively, but inversely proportional (Spearman's rho coefficient of -0.811, -0.829).

Correlations performed on groups of patients

We analyzed per group of patients and the control group the degree of correlation between blood viscosity values of the most important parameters previously analyzed the whole group of patients where we achieved the high correlation that is the hematocrit, glucose, FRG and fibrinogen.

Table nr.XVI The correlation coefficients in different groups of patients with renal disease and diabetic parameters

Shear rate	**Parameter**	**R^2 control group**	**R^2 lot 1**	**R^2 lot 2**	**R^2 lot 3**
22.5 sec^{-1}	Ht	**.3786**	**.4848**	**.1062**	**.3300**
	GFR	**.9399**	.0316	**.2318**	**.2578**
	glucose	**.8954**	**.2526**	**.1180**	**.3828**
	fibrinogen	.0075	.0524	**.1038**	**.5330**
150 sec^{-1}	Ht	**.1217**	**.2013**	.0026	**.4985**
	GFR	.0209	**.8384**	**.2598**	.0921
	glucose	.0452	0.0002	**.1493**	.0693
	fibrinogen	.0323	.0057	.0175	.0463
450 sec^{-1}	Ht	.0150	.0845	.0464	.0953
	GFR	0.0002	**.6438**	0.0008	.0171
	glucose	.0427	.0534	.0894	.0327
	fibrinogen	.0282	**.1503**	**.1073**	**.5800**

The blood circulation, blood viscosity varies with shear rate fluctuations, specifically the blood becomes less viscous at high shear rates. It was carried out the correlation between hematocrit, GFR, glucose and fibrinogen between groups of patients and controls. It is noted in small and medium shear rates a significant correlation between hematocrit and blood viscosity for all the studied groups (R^2= 0.3786, 0.4848, 0.1062, 0.3300), but at high shear rates this correlation is very weak.

At high shear rates fibrinogen influence it is important, especially in patients with existing renal impairment, observing a high degree of correlation for groups of diabetics (R^2= 0.1503, 0.1073, 0.5800) compared with the control group, where the influence is very weak fibrinogen (R^2 = 0.0282).

Between glycemic parameters and shear rates are closely correlated especially at low shear (R^2= 0.8954, 0.2526, 0.118, 0.3828), while the high rate, this correlation is absentă. The modifications of the values of hematocrit and fibrinogen diabetes it seems to affect the rheological behavior of diabetic patients, as demonstrated by previous studies. In our study we found

significant correlation parameter, however, renal most relevant, namely GFR with viscosity particularly at low shear rates, observing a degree of correlation R^2= 0.2318 for those of group 2, respectively R^2= 0.2578 for group 3.

Erythrocyte aggregability-results

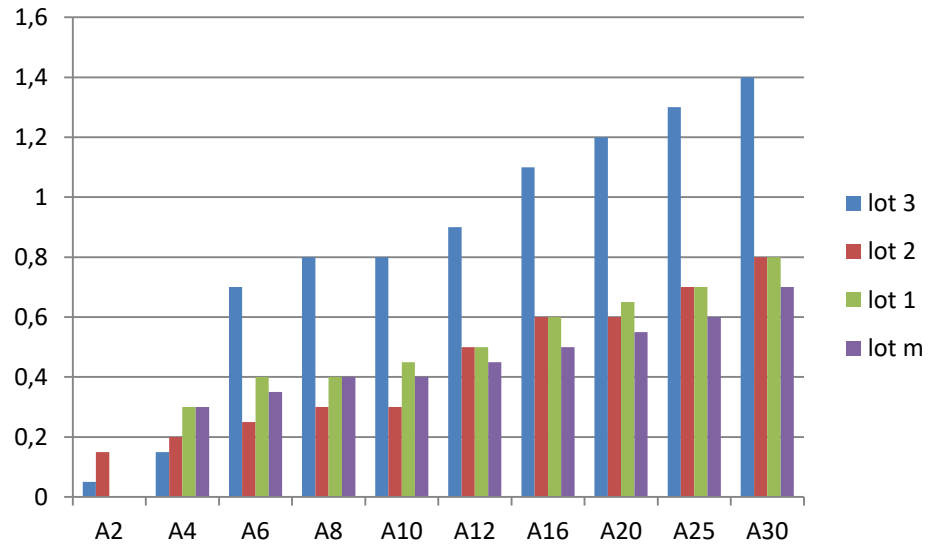

Figure No. 31 Erythrocyte aggregability values plots diabetic patients with varying degrees of renal impairment and the control group

There is an increase in red blood cell aggregation particularly in groups of patients with advanced renal impairment and severe, which contributes to increasing the viscosity of these groups of patients particularly at low shear rates. This increase in red cell aggregation is most evident in group 3 patients, the most severe renally impaired, which contributes to increasing the viscosity of these groups of patients particularly at low shear rates.

Results red cell deformability-results

Erythrocyte deformation demonstrates a unique ability to repeat this enabling its movement to the reduced-diameter vessels, the microcirculation. Erythrocyte deformability is a change that occurs passively in the form of erythrocyte as a result of the action of shearing forces. Deformability is considered a major determinant of blood flow rheological function.

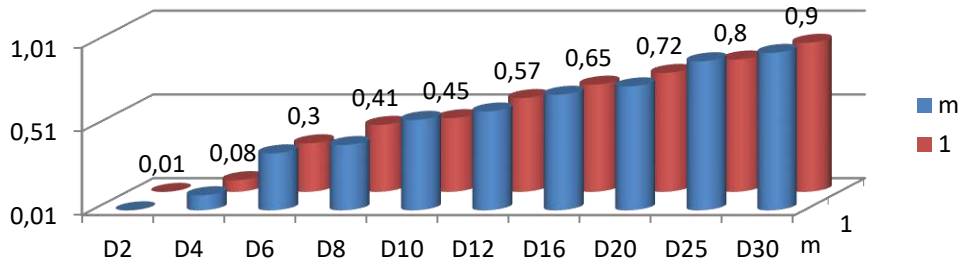

Fig. 33 The values of the erythrocyte deformability mild renal impairment batch (lot 1) as compared with control group

The deformability values for the group 1 of patients are comparable with those of the control group but with the progression of renal disease is observed a marked decrease in the other two groups of patients. Reducing red cell deformability is evident in the groups with advanced renal impairment (lots 2 and 3) compared with controls, whereas in group 1 patients deformability is observed behavior similar to that of the control group.

Using indirect methods of determining the parameters hemorrheologic and design of mathematical relationships.

The indirect determination of the yield

Applying the formula of Zydney (14) the values obtained by us study clearly observed a reduction in the yield with the progression of renal disease.

$$= 0.71\ [H\text{-}0.05]\ 3\ \tau_{\gamma} \qquad (14)$$

In lots of patients in our study applying the formula of Zydney we obtained the following values for yield strength through indirect determination.

The amount of standard considered it was 0.04 dyne / cm2 is observed that is found in our study in the control group, but also there is a marked decrease thereof with the progression of renal disease as in group 3 patients the limit of elasticity 0.011 is very low dyne / cm2.

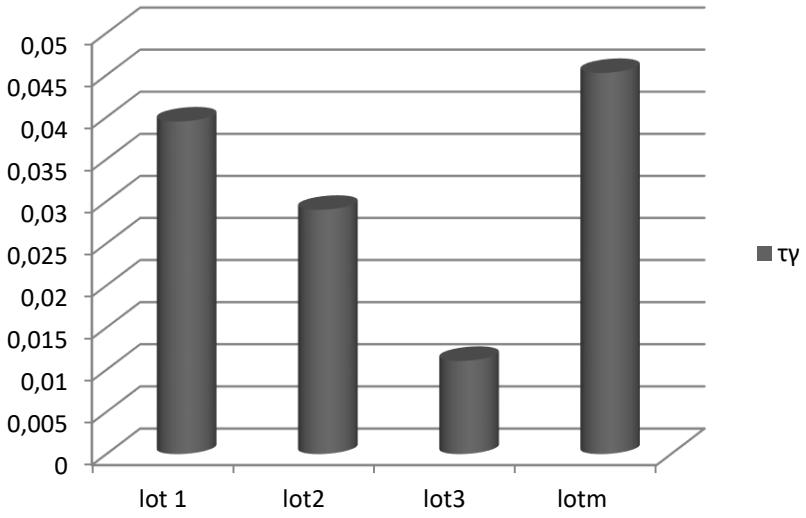

Figure No. 37 The values of the yield stress from the formula of Zydney

For the same parameter, namely the determination of the yield applied to the formula I described by Chien (15)

$$\tau_{\gamma} = 0.27\ HT\ 3 \qquad (15)$$

The reference value determined by Chien was 0.025, which is almost the same as obtained in our study of patients from the control group (0.024506809), but with a marked reduction values of up to 0.0078 for patients with severe renal impairment and degree of terminal.

Applying both Chien and his formula of Zydney values obtained in our study clearly observed a reduction in the yield with the progression of kidney disease.

The values obtained in our study according to the equation Zydnei and equation Chien showed values in accordance with the values obtained by the authors to the control groups, those in group 1 there was a decrease by 24% according to the equation Zydney and a 12% decrease as equation Chien, for patients in group 2 there was a decrease by 36% according to the equation Zydney, and 28% according to Chien and the group 3 patients a decrease of 87% according to the equation Zydney, respectively by 68% according to the equation described in the Chien.

Determination of erythrocyte rigidity- index of Taylor

For greater accuracy in terms of getting their results and reproducibility we have used the equation deformability erythrocyte rigidity index given Taylor's formula or equation Dintenfass.

$$T_k = \frac{\eta_r^{0,4} - 1}{\eta_r^{0,4} * Ht} \quad (16)$$

$$\eta_r = \frac{\eta_{sg}}{\eta_{pl}}$$

η_r it is the relative viscosity

η_{sg} it is blood viscosity

η_{pl} it is plasma viscosity

Characterized Tk indirectly stiffness index erythrocyte deformation, and obtain high values indicate a higher stiffness due to low red blood cell deformability.

Using patient data have erythrocyte rigidity index calculated according to the equation described above and have obtained the rigidity index values Tk.

Applying this formula we have achieved Taylor index elevated loads especially in patients 2 and 3 was 0.37, 0.39, confirming that equation.

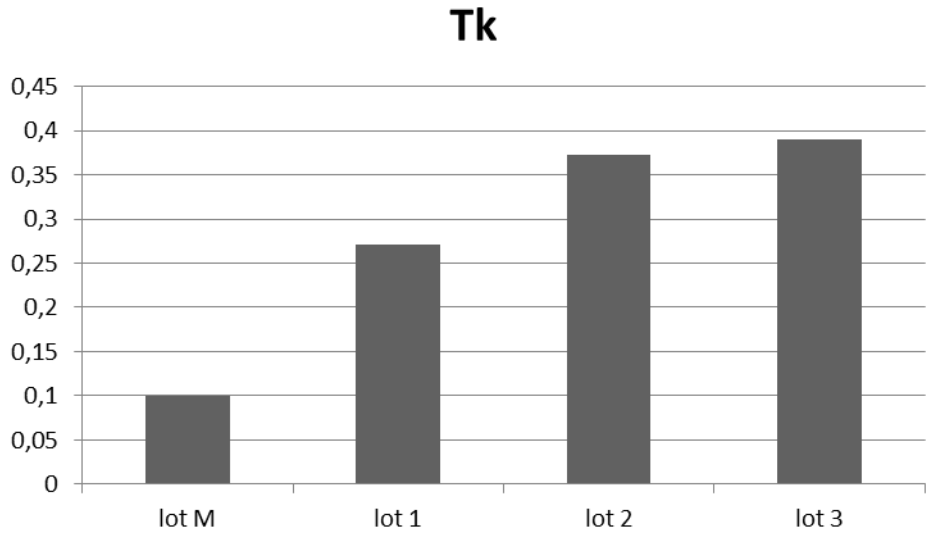

Figure No. 39 Values obtained for lots of study by stiffness index Taylor

Determining a relationship between blood parameters and blood viscosity

Simple linear regression model is used to find a relationship between parameters that provide the best data.

On the other hand, multiple linear regression model is used when at least two independent variables used in an attempt to find the best relationship between them. In multiple linear regression model is used least squares regression to estimate coefficients. The regression coefficients of the independent variables illustrates the contributions that are not dependent on the dependent variables. Unlike the simple linear regression to be originally calculated correlation levels between each of the independent variables.

From the analysis of the correlations described above revealed that there is a dependent blood viscosity to the following parameters of the analyzed namely:

- Hematocrit
- Fibrinogen
- GFR
- Glucose

To these parameters can conduct to a multiline regression relation to blood viscosity at low shear rates. We chose values of blood viscosity shear rate of 22.5 sec^{-1} because low shear rates is determined by the blood viscosity parametrii.Iniţial we selected most important parameters established after relations correlation results described in nr.XVI table.

In the second stage of the analysis, effecting the expression of blood viscosity () (expressed in cP) into two or more variables, as follows: $\vartheta_{sânge}$

Determination of multiline regression study for group 3 of patients

For lot 3 with the highest degree of renal impairment danalysis correlation coefficients shown in our study resulted interrelatedness of the four parameters, namely hematocrit, GFR, blood glucose and fibrinogen in patients with severe renal impairment and degree of terminal.

We determine the linear regression function for the four parameters, namely:

X1 = Hematocrit

X2 = FRG

X3 = Glucose

X4 = Fibrinogen

in relation to the viscosity of the blood at a shear rate of 22.5 sec-1 VS3 / sr22.5.

Such regression function is obtained:

$$\vartheta_{\frac{sânge_{vs3}}{sr22.5}} = -72{,}822 + 1{,}021Ht_{vs22.5} - 0{,}613rfg_{vs22.5} + 0{,}142Glicemie_{vs22.5} + 0{,}077Fibrinogen_{vs22.5}$$

and coeficientele correlation are found in the table. XX

Table nr.XX - Correlation coefficients for lot 3

Statistics Regression	
R - multiple correlation coefficient	0.911030677
R^2 - coefficient of multiple determination	0.829976895
R_c^2 - corrected coefficient of multiple determination	0.489930685

Conclusion: Statistical analysis shows that there is a strong dependency between the four parameters analyzed.

Determination of multiline regression study for group 2 of patients

For patients in group 2 analyzed we could observe dThe correlation coefficients in the analysis results shown below influence the strength of the four parameters, namely hematocrit, Germany, glucose and fibrinogen

We determine the linear regression function for the four parameters, namely:

X1 = Hematocrit

X2 = FRG

X3 = Glucose

X4 = Fibrinogen

in relation to the viscosity of the blood at a shear rate of 22.5 sec-1 (VS3 / sr22.5).

Such regression function is obtained:

$$\vartheta_{\frac{sânge_{vs3}}{sr22.5}} = 319{,}211 + 6{,}47Ht_{vs3} - 4{,}525rfg_{vs3} - 0{,}976Glicemie_{vs3} - 0{,}435Fibrinogen_{vs3}$$

and coeficientele correlation are found in the table. XXI

Table nr.XXI -Correlation coefficients for lot 2

Statistics Regression	
$\boldsymbol{R}$ **- multiple correlation coefficient**	0.933001514
$\boldsymbol{R^2}$ **- coefficient of multiple determination**	0.870491826
$\boldsymbol{R_c^2}$ **- corrected coefficient of multiple determination**	0.611475477

Conclusion: The statistic analysis shows that there is a strong dependency between the four parameters analyzed.

Determination of multiline regression study for group 1 of patients

For diabetic patients with mild renal impairment who were classified in group 1dThe correlation coefficients in the analysis presented above resulting strength of the two parameters, namely the influence of hematocrit and glucose.

We determine the linear regression function for the 2 parameters that matter, namely:

X1 = Hematocrit

X2 = Glucose

in relation to the viscosity of the blood at a shear rate of 22.5 sec-1 VS3 / sr22.5.

Such regression function is obtained:

$$\vartheta_{sânge_{vs3}/sr22.5} = 24{,}111 - 0{,}445Ht_{vs3} + 0{,}053Glicemie_{vs3}$$

and coeficientele correlation are found in the table. XXII.

Table nr.XXII -Correlation coefficients for lot 1

Statistics Regression	
$\boldsymbol{R}$ **- multiple correlation coefficient**	0.724205732

R^2 - coefficient of multiple determination	0.524473942
R_c^2 - corrected coefficient of multiple determination	-0.426578173

Conclusion: The statistic analysis shows that there is a good dependency between those two parameters analyzed.

Using regression calculation multiline tried to develop a mathematical relationship that includes biological parameters most closely correlated with blood viscosity at low shear rates. This mathematical relationship we hope to be a real help in the diagnosis of diabetic patients with renal impairment, because trying to modify these biological parameters of the equation, namely the rate of glomerular filtrate, hematocrit, fibrinogen or blood sugar - by appropriate therapy and treatment hygiene and adequate dietary - we to maintain viscosity values within normal limits, which would improve the quality of life of patients with a reduction of microvascular complications or a slowdown in their appearance.

CONCLUSIONS

Based on the objectives we have set in the personal thesis and Relating to the currently existing studies in the literature, we can draw the following conclusions of this scientific research:

1. Kidney damage was the selection criteria of patients in our study following the most faithful indicator ie glomerular filtration rate, which was significantly decreased in all groups of patients.
2. The viscosity of the blood at shear rates of multiple batches studied showed different characteristics depending on the degree of renal impairment.
3. At low shear rates found increased blood viscosity in all patients included in the study compared with controls.
4. With increasing rates, the behavior is different, demonstrating a significant reduction in blood viscosity of Lot 3, with severe renal impairment presenting even lower values than the control group.

3. If patients with mild renal impairment influence biological parameters is given only hematocrit values with the progression of renal damage there is a correlation with renal and glycemic parameters, and with the fibrinogen.

4.Viscosity at low shear rates is mainly determined by red blood cell aggregability. Increased erythrocyte aggregation was demonstrated by performing the test filterability where we observed elevated, particularly in groups of patients with advanced renal impairment and severe, according to previous studies.

6. Blood viscosity at high shear rates is influenced by red cell deformability. Reducing red cell deformability was evident in the groups with advanced renal impairment (lots 2 and 3) compared with controls, whereas in group 1 patients deformability is observed behavior similar to that of the control group.

7. To further enhance the accuracy of the results obtained in terms of the deformability of the method we have accomplished indirect determination has shown the same behavior.

8. Regarding plasma viscosity 's an increase of it in the group with severe renal impairment, which probably influenced the increased fibrinogen of patients in this group.

9. Determination of the elasticity was carried out using equations known in the art and have observed a clear reduction in that with the progression of kidney disease by applying the Chien's formula Zydney.

10. Formulating a regression equation multiline tried to develop a mathematical relationship that includes biological parameters most closely correlated with blood viscosity at low shear rates. This mathematical relationship we hope to be a real help in monitoring diabetic patients with renal impairment.

11. Hemorheological behavior is different with the progression of renal disease and a better understanding of it can help contribute to the attempt to reduce the complications of diabetes and of minimizing the devastating consequences of such determination.

12. In determining all parameters were followed in the study described fully reproducible methods we used.

Originality of the thesis and objectives proposed

The study conducted by us, I think that is a first in Romania as it addresses issues hemorrheologic in patients with diabetic nephropathy, depending on the rate of progression of renal damage.

Relationship diabetus mellitus- diabethic nephropathy - hemorrheologic is still a challenge. This study wanted to give a new perspective in terms of diabetes, its complications kidney default in terms hemorrheologic.

In most studies observed rheological aspects of diabetic patients were viewed as a whole, without trying to define the complications of the disease. With this doctoral thesis I tried to show how rheological parameters evolve differently depending on the rate of progression of renal damage.

The following considerations may contribute to the originality of this thesis:

- I realized determining blood viscosity, erythrocyte aggregation and deformability and plasma viscosity in patients with varying degrees of renal impairment
- We used mathematical models recognized in the determination of rheological parameters by indirect methods to demonstrate compliance with the values obtained by direct method, which can be useful in medical practice.
- Making a prediction relationship through a multiple regression equation in assessing biological changes involving rheological parameters.

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