Medical University - Varna

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“Assessment and dynamic monitoring of hyperphosphatemia - a predictor of bone mineral disorders in dialysis patients”

Thesis summary

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The present dissertation was developed in the Department of Internal Medicine, Teaching Sector of Nephrology, Hemodialysis and Toxicology at the Faculty of Medicine, Medical University "Prof. Dr. Paraskev Stoyanov" - Varna.

The dissertation contains 164 pages and is illustrated with 45 tables and 29 figures and one appendix. The reference list includes 206 literature sources, of which 8 in Cyrillic and 198 in Latin.

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The official defense of the dissertation will take place on 08.04.2022 at hours in - Varna.

The defense materials are available in the Scientific Department of Medical University - Varna and are published on the website of Medical University - Varna.

Note: In the thesis summary the numbers of tables and figures do not correspond to the numbers in the thesis.

CONTENTS

ABBREVIATIONS USED

25(OH)D	calcidiol
АФ	Alkaline phosphatase
ВхПТ	Secondary hyperparathyroidism
ГФ	Glomerular filtration
ехоКГ	echocardiography
КМН-ХБЗ	Bone mineral disorders in chronic kidney disease
КТ	Computer tomography
М.С.	Medical specialist
ПОЕ	weeks 20 – 27
СЗО	World Health Organization
СК	Vascular calcification
СС	Cardiovascular
ХБЗ	Chronic kidney disease
ХБН	Chronic renal failure
ХД/HD	Hemodialysis
ХДф/ HDF	Haemodiafiltration
ANOVA	Analisys of variance
CaSR	Calcium sensitive receptor
EDTA	Ethylenediaminetetraacetic acid
ELISA	Enzime linked immunosorbent assay
ESKD	End stage kidney disease
FDA	Food and Drug Administration
FGF-23	Fibroblast growth factor -23
HRQoL	Health-related quality of life
JTT-751	Ferric citrate
KDIGO	Kidney Disease: Improving Global Outcomes
KDQoL-SF-36	Kidney Disease Quality of Life -Short Form-36
MBD	Mineral and bone disorders
PTH	Parathyroid hormone
PA21	Sucropheric oxyhydroxide
USRDS	United States Renal Data System

I. INTRODUCTION

Bone and mineral disorders in chronic kidney disease represent particularly severe systemic complications of increasing medico-social importance worldwide.

The KidneyDisease: Improving Global Outcomes (KDIGO) Foundation recognizes that chronic kidney disease involves systemic disorders of mineral and bone metabolism, namely calcium, phosphorus, parathyroid hormone, vitamin D, mineralization, volume, linear growth or bone strength, and vascular or other soft tissue calcifications. These are associated with risk of fractures, cardiovascular disease and death, so regular monitoring is recommended.

BMD-CKD syndrome involves disturbances in the metabolism of calcium, phosphate, parathyroid hormone (PTH) and vitamin D, which cause pathological changes in bone metabolism, vascular and soft tissue calcifications.

Patients with CKD on hemodialysis treatment are at risk of complications, such as anaemia, electrolyte disturbances (e.g. hyperkalaemia, hyperphosphatemia) and BMD-CKD, including secondary hyperparathyroidism, changes in vitamin D activation and renal osteodystrophy.

With the progression of CKD and loss of nephron mass, absolute calcitriol deficiency, phosphate retention - hyperphosphatemia with hypocalcemia occurs, which further enhances PTH production and leads to the development of secondary hyperparathyroidism with hyperplasia of the parathyroid glands and disturbances in bone turnover.

Secondary hyperparathyroidism (SHPT) is a complication of chronic kidney disease and end-stage renal disease, resulting in altered calcium (Ca) and phosphorus (P) homeostasis. In SHPT, excess amounts of parathyroid hormone (PTH) are continuously released from the hyperplastic parathyroid glands, thereby increasing blood PTH levels, and elevated concentrations of circulating PTH are associated with abnormalities in serum Ca, P, and FGF-23 (J. Cunningham et al., 2011). These biochemical changes worsen the progression of SHPT and correlate with abnormal bone histology, increased risk of fractures, vascular calcification, soft tissue calcification and increased mortality (Joy et al., 2007). (98); (100);

Control of elevated serum phosphorus, calcium levels, restoration of vitamin D levels and suppression of PTH production remain targets for effective treatment of BMD-CKD.

Multiple classes of drugs, including phosphate binders, vitamin D analogues and calcimimetics have been developed to directly or indirectly influence markers of BMD-CKD. In particular, within the class of calcimimetics - Cinacalcet (Sensipar, Amgen, Inc.) and etelcalcetide (Parsabiv, Amgen, Inc.) are two drugs available in the EU.

II. PURPOSE AND TASKS

The purpiose of the dissertation is to analyze the diagnostic, clinical and therapeutic aspects of bone mineral metabolism disorders in chronic kidney disease during conservative and hemodialysis treatment. In order to achieve this purpose, we set ourselves the following tasks:

- To investigate the diagnostic and prognostic value of calcium, phosphorus and parathyroid hormone in the development and disturbance of bone-mineral metabolism in patients with chronic kidney disease in the pre-dialysis stage and on hemodialysis treatment.

- To investigate the dynamic influence of phosphorus-binding drugs sevelamer hydrochloride and calcium carbonate on markers of bone-mineral metabolism in dialysis patients.

- To look for a correlation between etelcalcetide and markers of bone mineral metabolism in hemodialysis patients and to monitor its effectiveness and safety in overcoming hyperphosphatemia.

- To compare serum sclerostin levels in pre-dialysis patients and patients undergoing hemodialysis treatment and to evaluate the effect of etelcalcetide (Parsabiv) treatment on serum sclerostin levels in hemodialysis patients.

- To analyze and compare the effect of convection hemodialysis and hemodiafiltration on hyperphosphatemia in dialysis patients.

- To analyze the survival and quality of life in dialysis patients in terms of biochemical markers of BMD-CKD.

Hypothesis

The constellation of diagnostic and therapeutic methods that we have developed contributes both to elucidating etiopathogenetic mechanisms of bone mineral metabolism disorders in patients with chronic kidney disease on conservative and hemodialysis treatment and to increasing the effectiveness of the individualized approach to these patients.

III. MATERIAL AND METHODS

- Material:

A comprehensive diagnostic - treatment study was conducted between 1.II.2019 and 31.I.2022 incl.

The study was carried out with the permission of KENI at MU-Varna with Protocol/Decision No. 110/11.01.2022.

Inclusion Criteria:

- Persons over 18 years of age
- Persons who have signed an informed consent
- Persons with proven CKD - pre-dialysis and dialysis stage

Exclusion criteria:

- Persons under 18 years of age
- Persons who have not signed informed consent

A total of 116 patients - 75 men and 41 women with CKD - pre-dialysis (control group) and on hemodialysis treatment from the Clinic of Nephrology and Dialysis of the University Hospital "St. Marina" were included in the study. They were clinically followed up and examined by routine methods. There were 86 patients undergoing dialysis treatment and 30 patients in pre-dialysis stage. 75 (65%) men and 41 (35%) women were included in the study (Fig. 1). The study patients had a mean age of 63.55±9.69 years and the static age distribution was normal (Kolmogorov-Smirnov test, $p>0.1$).

The mean age of the male group was 53.75±10.50 years, and the statistical distribution of age in the male group was normal (Kolmogorov-Smirnov test, $p=0.052$). Mean age in women was 49.25±10.45 years, and the statistical distribution of age in the women's group was normal (Kolmogorov-Smirnov test, $p>0.1$). There was no statistically significant difference between the age in males and that in females ($p=0.228$).

According to the WHO age distribution, 17 patients (15%) were young (18 to 44 years); 51 patients (44%) were middle-aged (45 to 59 years); 39 patients (34%) were elderly (60 to 74 years); and 9 patients (7%) were old (75 to 89 years) (Figure 2).

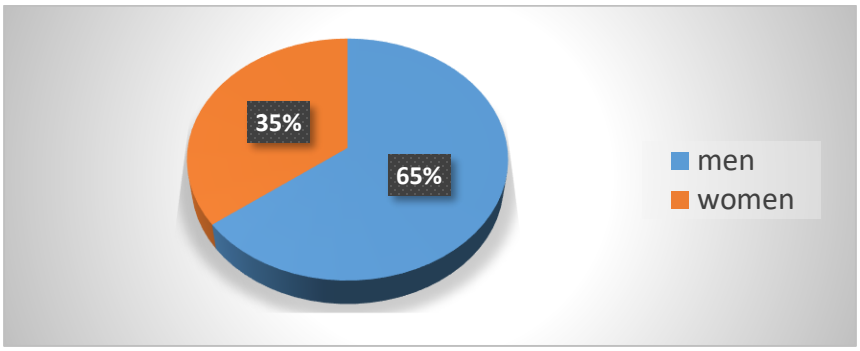


Fig.1 Gender distribution of the studied patient groups

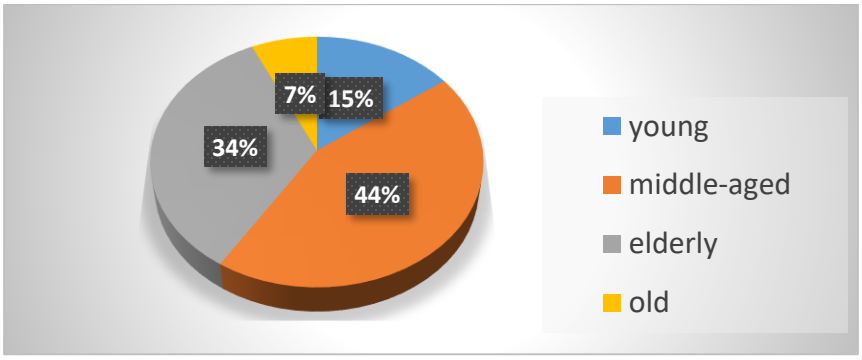


Fig.2 Age distribution of the studied patient groups according to WHO

- Methods:

1. Clinical examinations:

Clinical examinations include a detailed history and objective physical status of patients in the clinic.

2. Anthropometric methods:

Measurement of blood pressure under standard conditions: blood pressure was measured after a period of rest, at the level of the arm, in the sitting/lying position before the HD procedure, during and after dialysis in patients undergoing hemodialysis. In pre-dialysis patients, arterial blood pressure was measured after a period of rest, at arm level, in the sitting position. The middle of the arm was at the level of the heart, the back was supported and the feet were firmly on the floor.

3. Laboratory tests:

We examined the following laboratory parameters:

- Parathyroid hormone (PTH)

- Calcium

- Phosphorus

- Alkaline phosphatase

- Vitamin D

- Sclerostin

Serum calcium concentrations were quantified by the automated Arsenazo III photometric method.

Serum concentrations of inorganic phosphorus were quantified by the automated phosphomolybdate/UV method adapted on a fully automatic ADVIA 1800 biochemical analyzer, Siemens.

Serum concentrations of modified intact parathyroid hormone were quantified by the two-step automated chemiluminescent immunoassay (CLIA) using two polyclonal antibodies as an application of LIAISON/ DIASORIN.

Serum bone-specific alkaline phosphatase concentrations were quantified with the DiaSorin LIAISON® BAP OSTASE® assay, adapted from CLSI EP17-A, according to the chemiluminescent immunoassay (CLIA) technology.

Serum concentrations of 25-OH-vitamin D and some of its other hydroxylated metabolites were quantified with the LIAISON® 25 Vitamin D TOTAL assay according to chemiluminescent immunoassay (CLIA) technology.

The assay used to measure sclerostin levels was performed by ELISA (Enzyme-linked immunosorbent assay) on fully automated equipment and using the manufacturer's protocols (Biomedica Medizinprodukte GmbH & Co, Wien, Austria).

4. Statistical methods:

4.1) non-parametric Kolmogorov-Smirnov test to check the type of frequency distribution;

4.2) non-parametric chi-square test (X2) - comparing different categories of variables;

4.3) parametric method - Independent t-test - determines whether there is a statistically significant difference between two independent groups;

4.4) parametric method - Paired Samples t-test, comparing the means of the pre- and post-treatment measurements;

4.5) Analysis of variance - ANOVA test - for repeated measurements. The test compares mean between male and female indicators, but also accounts for repeated measurements every 3 months;

4.6) non-parametric method - Fieldman test - the test compares mean between male and female indicators, but takes into account repeated measurements every 3 months;

4.7) non-parametric method - Wilcoxon test comparing the mean of pre- and post-treatment measurements;

4.8) graphical analysis - to visualize the results obtained.

Statistical processing of the obtained data was carried out using BM SPSS v.25, and Jamovi v.2.1.1. descriptive indicators for quantitative and qualitative variables were used and presented in tabular and graphical form. The values of the specific variables at $p \leq 0.05$ were considered statistically significant.

The individual quality of life of 86 hemodialysis patients was investigated with the specialized questionnaire for quality of life in patients with kidney disease with 36 questions (Kidney Disease Quality of Life - Short Form-36, KDQOL-36) after modification by S. Staykova (2018) in order to adapt it to the conditions in our country. The applied method of quality of life assessment includes direct questionnaires.

IV. RESULTS

Task 1. To investigate the diagnostic and prognostic value of calcium, phosphorus and parathormone in the development and disturbance of bone mineral metabolism in patients with chronic kidney disease in the pre-dialysis stage and on hemodialysis treatment.

We studied 86 patients on HD treatment from the Clinic of Nephrology and dialysis, including 56 men with a mean age of 52.13 (standard deviation +-10.2) and 30 women with a mean age of 50.10 (standard deviation 11.1). They are on dialysis treatment 3 times a week for 4 hours, with a duration of dialysis treatment of up to 5-7 years.

A control group of 30 pre-dialysis CKD patients was studied - 19 males mean age 52.10 (standard deviation 11.12) and 11 females mean age 65.5 (standard deviation 13.53).

Table 1.1 shows descriptive statistics of biochemical parameters based on mean and frequency analysis in predialysis patients.

Table 1.1 Descriptive characteristics of biochemical parameters in pre-dialysis patients

	Number	Min value	Max value	Arithmetic mean	Standard deviation
calcium	30	2.05	2.40	2.2380	0.09423
phosphorus	30	1.40	2.30	1.9633	0.21891
parathormone	30	150.00	780.00	385.6667	203.02341

A comparative gender analysis in the pre-dialysis group of patients was done regarding calcium, phosphorus and parathormone values. The observed differences in the percentage distribution was not gender related and no statistically significant difference was found between males and females in the pre-dialysis stage (Tables 1.2; 1.3; 1.4).

Table 1.2 Comparative analysis of calcium (Ca) values by gender in pre-dialysis patients

			Ca		
X^2=0.889, p=0.346			< 2.2 mlmol/l	2,2 - 2,5 mlmol/l	total
gender	men	number	7	12	19
		%	36.8%	63.2%	100.0%
	women	number	6	5	11
		%	54.5%	45.5%	100.0%
total		number	13	17	30
		%	43.3%	56.7%	100.0%

Table 1.3 Comparative analysis of phosphorus (P) values by gender in pre-dialysis patients

X^2=3.701, p=0.054			**P**		
			0,8 - 1,6 mlmol/l	Над 1,6 mlmol/l	total
gender	men	number	0	19	19
		%	0.0%	100.0%	100.0%
	women	number	2	9	11
		%	18.2%	81.8%	100.0%
total		number	2	28	30
		%	6.7%	93.3%	100.0%

Table 1.4 Comparative analysis of parathormone (PTH) values by gender in pre-dialysis patients

X^2=3.589, p=0.166			**PTH**			
			150 – 300 pg/ml	300 - 600 pg/ml	over 600 pg/ml	total
gender	men	number	7	8	4	19
		%	36.8%	42.1%	21.1%	100.0%
	women	number	8	2	1	11
		%	72.7%	18.2%	9.1%	100.0%
total		number	15	10	5	30
		%	50.0%	33.3%	16.7%	100.0%

Tables 1.5; 1.6 and 1.7 demonstrate the comparative analysis of calcium, phosphorus and parathyroid hormone values by gender in the group of patients undergoing dialysis treatment.

Table 1.5 Comparative analysis of calcium (Ca) values by gender in dialysis patients

			Ca			
X^2=0.943, p=0.624			< 2.2 mlmol/l	2,2-2,5mlmol/l	>2,5 mlmol/l	total
gender	men	number	7	20	29	56
		%	12.5%	35.7%	51.8%	100.0%
	women	number	2	13	15	30
		%	6.7%	43.3%	50.0%	100.0%
number		number	9	33	44	86
		%	10.5%	38.4%	51.2%	100.0%

Table 1.6 Comparative analysis of phosphorus (P) values by gender in dialysis patients

			P		
***X^2*=1.662, p=0.197**			0,8 - 1,6 mlmol/l	over 1,6 mlmol/l	total
gender	men	number	3	53	56
		%	5.4%	94.6%	100.0%
	women	number	4	26	30
		%	13.3%	86.7%	100.0%
	total	number	7	79	86
		%	8.1%	91.9%	100.0%

Table 1.7 Comparative analysis of parathormone (PTH) values by gender in dialysis patients

			PTH			
***X^2*=1.098, p=0.577**			150 - 300 pg/ml	300 - 600 pg/ml	over 600 pg/ml	total
gender	men	number	16	2	38	56
		%	28.6%	3.6%	67.9%	100.0%
	women	number	9	0	21	30
		%	30.0%	0.0%	70.0%	100.0%
total		number	25	2	59	86
		%	29.1%	2.3%	68.6%	100.0%

The results of the analysis again showed no statistically significant difference between men and women in the group of patients undergoing dialysis.

A correlation was made between the two groups - pre-dialysis and dialysis patients in terms of calcium, phosphorus and parathormone values (Table1.8).

Table 1.8. comparative analysis of calcium, phosphorus and parathormone values between the two groups

	group	number	Arithmetic mean	Standard deviation	Standard error
calcium	Dialysis group	86	2.2588	0.20038	0.02161
	Pre-dialysis group	30	2.4380	0.09423	0.01720
phosphorus	Dialysis group	86	2.2007	0.24444	0.02636
	Pre-dialysis group	30	1.9633	0.21891	0.03997
parathormone	Dialysis group	86	893.2558	576.73409	62.19084
	Pre-dialysis group	30	385.6667	203.02341	37.06683

The HD group had higher phosphorus and parathyroid hormone values and lower calcium values compared to the control group - pre-dialysis patients (Table 1.9).

Table 1.9. comparison of biochemical parameters between the two groups

Independent Samples Test										
	Levene's Test for Equality of Variances		t-test for Equality of Means						
								95% confidence interval	
	F	p	t	df	p	Difference in average values	Standard error	lower	upper
calcium	22.777	0.000	5.804	114	0.001	0.22084	0.03805	0.14546	0.29621
			7.996	104.199	0.001	0.22084	0.02762	0.16607	0.27561
phosphorus	0.110	0.074	0.074	114	0.046	0.03736	0.05051	-0.06269	0.13742
			0.078	56.093	0.438	0.003736	0.04788	-0.05854	0.13327
parathormone	10.553	0.002	4.708	114	0.000	507.58915	107.80619	294.02591	721.15238
			7.011	113.964	0.000	507.58915	72.39924	364.16632	651.01198

From the analysis of the results, statistical significance was found in all three biochemical parameters (calcium p=0.001; phosphorus p=0.046; parathormone p=0.001).

Table 1.10 shows the descriptive characteristics of the biochemical parameters by gender in both groups.

Table 1.10. descriptive characterization of biochemical parameters by sex in both groups

	gender	N	Arithmetic mean	Standard deviation	Standard error
calcium	men	75	2.4023	0.20117	0.02323
	women	41	2.4007	0.20979	0.03276
phosphorus	men	75	2.0235	0.21833	0.02521
	women	41	1.9317	0.26214	0.04094
parathormone	men	75	770.1333	554.80090	64.06289
	women	41	747.0732	556.93466	86.97858

Table 1.11. demonstrates and compares the biochemical indices by Independent t-test.

Table 1.11. comparison of biochemical indices by gender in the two groups

Independent Samples Test										
	Levene's Test for Equality of Variances		t-test for Equality of Means						
								95% confidence interval	
	F	p	t	df	p	Mean Difference	Std. Error Difference	lower	upper
calcium	0.268	0.606	0.039	114	0.969	0.00153	0.03967	-0.07705	0.08012
			0.038	79.466	0.970	0.00153	0.04016	-0.07840	0.08147
phosphorus	3.963	0.049	2.013	114	0.056	0.09176	0.04557	0.00148	0.18204
			1.908	70.601	0.060	0.09176	0.04808	-0.00412	0.18764
parathormone	0.059	0.809	0.214	114	0.831	23.06016	107.90212	-190.69311	236.81344
			0.213	82.109	0.831	23.06016	108.02466	-191.83103	237.95136

The results of the analysis showed no statistical significance in any of the biochemical parameters (Ca: p=0.969; P: p=0.060; PTH: p=0.831), regarding the sex characteristics between the patients of the two groups (Table 1.11).

Task 2. To investigate the dynamic influence of phosphorus-binding drugs - sevelamer hydrochloride (Renagel) and calcium carbonate on markers of bone mineral metabolism in dialysis patients.

The therapeutic influence of phosphorus-binding drugs - sevelamer hydrochloride and calcium carbonate on the markers of bone mineral metabolism in dialysis patients was studied in the Clinic of Nephrology and Dialysis at the University Hospital "St. Marina"-Varna for a period of 2 years. For this purpose, 43 haemodialysis patients were included, 18 of them on calcium carbonate treatment and 25 on sevelamer hydrochloride (Renagel) treatment. In patients on sevelamer hydrochloride treatment, calcimimetic (Cinacalcet) treatment was added to compare the effect on parathyroid hormone levels in patients on hemodialysis treatment.

Table 2.1. Descriptive characteristics of biochemical parameters before and after treatment with calcium carbonate

Descriptive statistics						
	Number of patients	Rank	Min. value	Max.value	Arithmetic mean	Standard deviation
Calcium prior to treatment	18	0.20	2.10	2.30	2.2161	0.06118
Phosphorus prior to treatment	18	0.65	1.65	2.30	1.9722	0.17923
Parathormone prior to treatment	18	1900.00	890.00	2500.00	1343.8889	674.79094
Calcium after treatment	18	0.40	2.20	2.60	2.4367	0.09530
Phosphorus after treatment	18	0.65	1.35	2.00	1.7333	0.20123
Parathormone after treatment	18	1960.00	890.00	2500.00	1252.7778	574.98650
Valid N (listwise)	18					

Table 2.2. Comparison of biochemical parameters before and after treatment with calcium carbonate

Comparison of average values				
	Arithmetic mean	Number	Standard deviation	Standard error
Calcium prior to treatment	2.2161	18	0.06118	0.01442
Calcium after treatment	2.4367	18	0.09530	0.02246
Phosphorus prior to treatment	1.9722	18	0.17923	0.04225
Phosphorus after treatment	1.7333	18	0.20123	0.04743
Parathormone prior to treatment	1343.8889	18	674.79094	159.04975
Parathormone after treatment	1252.7778	18	574.98650	135.52562

Table 2.3. Results of comparison of biochemical parameters before and after treatment with calcium carbonate

Paired Samples Test								
	Paired Differences							
				95% confidence interval				
	Mean difference	Standard deviation	Standard error	Lower interval	Upper interval	t	df	p
calcium before- after treatment	-0.22056	0.09052	0.02134	-0.26557	-0.17554	-10.337	17	0.001
phosphorus before- after treatment	0.23889	0.12107	0.02854	0.17868	0.29909	8.371	17	0.001
parathormone before- after treatment	91.1111	171.76667	37.62624	106.23590	475.98633	0.622	17	0.09

The results of the comparison of calcium values before and after calcium carbonate treatment showed a statistically significant difference in the two measurement periods (t= -10.337, p=0.001). The mean calcium values before treatment were lower (2.21 ±0.61) compared to the values after treatment (2.43 ±0.95).

From the results obtained comparing serum phosphorus values before and after calcium carbonate treatment, there was a statistically significant difference in the two periods (t= 8.371, p=0.001). Mean phosphorus values before treatment were higher (1.97 ± 0.18) compared to values after calcium carbonate treatment - (1.73 ± 0.2).

Calcium carbonate treatment showed no statistical significance in both periods: pre-treatment (1343.89±674.79) and post-treatment (1252.78±574.99) on parathormone values (t= 0.622, p=0.09).

Table 2.4. Descriptive characteristics of biochemical parameters before and after treatment with sevelamer hydrochloride and cinacalcet

Descriptive statistics						
	Number of patients	Rank	Min. value	Max.value	Arithmetic mean	Standard deviation
Calcium prior to treatment	25	0.40	2.30	2.70	2.5076	0.10698
Phosphorus prior to treatment	25	0.80	1.65	2.45	2.0720	0.20486
Parathormone prior to treatment	25	1850.00	650.00	2500.00	1353.6000	607.08237
Calcium after treatment	25	0.30	2.20	2.50	2.3148	0.09862
Phosphorus after treatment	25	0.80	1.30	2.10	1.7504	0.21603
Parathormone after treatment	25	1670.00	530.00	2200.00	964.4000	468.96233

Table 2.5 Comparison of biochemical parameters before and after treatment with sevelamer hydrochloride and cinacalcet

Comparison of average values				
	Arithmetic mean	Number	Standard deviation	Standard error
Calcium prior to treatment	2.5076	25	0.10698	0.02140
Calcium after treatment	2.3148	25	0.09862	0.01972
Phosphorus prior to treatment	2.0720	25	0.20486	0.04097
Phosphorus after treatment	1.7504	25	0.21603	0.04321
Parathormone prior to treatment	1353.6000	25	607.08237	121.41647
Parathormone after treatment	964.4000	25	468.96233	93.79247

Table 2.6. Results of comparison of biochemical parameters before and after treatment with sevelamer hydrochloride and cinacalcet

Paired Samples Test								
	Paired Differences							
				95% confidence interval				
	Mean difference	Standard deviation	Standard error	Lower interval	Upper interval	t	df	p
calcium before- after treatment	0.19280	0.10872	0.02174	0.14792	0.23768	8.866	24	0.001
phosphorus before- after treatment	0.32160	0.10302	0.02060	0.27907	0.36413	15.608	24	0.001
parathormone before- after treatment	389.20000	271.12297	54.22459	277.28594	501.11406	7.178	24	0.001

Treatment with sevelamer hydrochloride and cinacalcet showed statistical significance in both periods in the values of calcium, phosphorus and parathormone.

From the results obtained comparing serum calcium values before and after treatment with sevelamer hydrochloride and sinacalcet, there was a statistically significant difference in both measurement periods (t= 8.866, p=0.001). Pre-treatment calcium values were lower (2.51 ± 0.11) compared to post-treatment values with sevelamer hydrochloride - (2.31 ± 0.01).

Comparison of serum phosphorus values, before and after treatment with sevelamer hydrochloride and cinacalcet revealed statistical significance in both periods (t= 15.608, p=0.001). Pre-treatment phosphorus values were higher (2.07 ±0.2) compared to post-treatment values (1.75 ±0.22).

The results of the comparison of PTH values, before and after treatment with sevelamer hydrochloride and cinacalcet, showed a statistically significant difference in the two measurement periods (t= 7.178, p=0.001). The mean PTH values before treatment were higher (1353.6 ±607.08) compared to the values after treatment (964.4 ±468.96).

From the study conducted, it was proved that treatment with both calcium carbonate and combination of sevelamer and cinacalcet showed statistical significance in terms of serum calcium and phosphorus in dialysis patients, pre-treatment values were similar with post-treatment values with both drugs. Parathormone values remained without significant change with calcium carbonate treatment, and were significantly lower after combined treatment with sevelamer hydrochloride and a calcimimetic (cinacalcet).

Task 3. To look for a correlation between etelcalcetide and markers of bone mineral metabolism in hemodialysis patients and to monitor its effectiveness and safety in overcoming hyperphosphatemia.

Twenty-seven patients (12 women and 15 men) on hemodialysis treatment with etelcalcide (Parsabiv i.v.) were followed up for 12 months at the Clinic of Nephrology and Dialysis, University Hospital "St. Marina"- Varna.

Patients are on hemodialysis 3 times a week for 4 hours - bicarbonate dialysis procedure. The study patients had high baseline Ca, P and PTH values before starting their treatment. Parsabiv was administered at a dose of 12.5 mg/week, given intravenously at the end of each dialysis session.

The present analysis of the results obtained is based on the comparison of Ca, P and iPTH values every 3 months over a 12-month period of treatment with Parsabiv.

Comparing serum calcium values :

The results of the analysis showed statistically significant differences when comparing calcium values for the study period (Table 3.1) and when comparing by gender.

Table 3.1. comparison of serum calcium values during treatment with Parsabiv i.v.

Comparison (F=7.14, p<0.014)							
RM Factor 1		**RM Factor 1**	**Mean Difference**	**SE**	**df**	**t**	**p**
Calcium prior to treatment	-	Calcium 3m after treatment	0.40492	0.0220	25.0	18.3981	<0.001
	-	Calcium 6m after treatment	0.23300	0.0239	25.0	9.7419	<0.001
	-	Calcium 9m after treatment	0.38092	0.0227	25.0	16.7644	<0.001
	-	Calcium 12m after treatment	0.37967	0.0156	25.0	24.2888	<0.001
Calcium 3m after treatment	-	Calcium 6m after treatment	-0.17192	0.0228	25.0	-7.5289	< 0.001
	-	Calcium 9m after treatment	-0.02400	0.0210	25.0	-1.1424	0.783
	-	Calcium 12m after treatment	-0.02525	0.0193	25.0	-1.3074	0.689
Calcium 6m after treatment	-	Calcium 9m after treatment	0.14792	0.0216	25.0	6.8394	<0.001
	-	Calcium 12m after treatment	0.14667	0.0215	25.0	6.8088	< 0.001
Calcium 9m after treatment	-	Calcium 12m after treatment	-0.00125	0.0157	25.0	-0.0795	1.000

From the results obtained, statistical significance was found in both men and women after the third month of treatment with Parsabiv 12.5mg i.v./week (p<0.001). Serum calcium concentrations decreased in 62% of patients, after which treatment with calcium carbonate 600mg 2x600 mg/day was included with adjustment in dialysis solution from A11 - 1.25 Ca2+ to A13.0 - 1.50 Ca2+ twice weekly and A13.1 - 1.75 Ca2+ once weekly. The effect of the

adjustment and the combination treatment was noted after the 6th month and serum calcium reached reference values.

Comparison of serum phosphorus values:

At the third month of treatment with Parsabiv i.v., there was no statistical significance in both females (p=0.568) and males (p=0.092) regarding serum phosphorus values. Sevelamer 800mg 3x1tb/day p.os. was also started in the 3rd month, and after the 6th month, 80% of the patients achieved normal serum phosphorus values. The results of the analysis showed statistically significant differences after the 6th,9th and 12th month of treatment (p<0.001); (Table 3.2).

Table 3.2. comparison of serum phosphorus values during treatment with Parsabiv i.v.

Comparison (F=6.29, p<0.001)							
RM Factor 1		**RM Factor 1**	**Mean Difference**	**SE**	**df**	**t**	**p**
Phosphorus prior to treatment	-	Phosphorus 3m after treatment	0.105	0.0212	25.0	4.956	1 .000
	-	Phosphorus 6m after treatment	0.411	0.0290	25.0	14.196	<0 .001
	-	Phosphorus 9m after treatment	0.809	0.0481	25.0	16.808	<0 .001
	-	Phosphorus 12m after treatment	0.637	0.2980	25.0	2.139	0.0235
Phosphorus 3m after treatment	-	Phosphorus 6m after treatment	0.307	0.0176	25.0	17.408	< 0.001
	-	Phosphorus 9m after treatment	0.704	0.0446	25.0	15.788	< 0.001
	-	Phosphorus 12m after treatment	0.533	0.3019	25.0	1.764	0.415
Phosphorus 6m after treatment	-	Phosphorus 9m after treatment	0.397	0.0369	25.0	10.764	< 0.001
	-	Phosphorus 12m after treatment	0.226	0.3071	25.0	0.736	0.946
Phosphorus 9m after treatment	-	Phosphorus 12m after treatment	-0.171	0.3040	25.0	-0.563	0.979

In the comparative analysis of serum phosphorus values in terms of gender characteristics in the group of patients on Parsabiv 12.5mg i.v/week, no statistically significant difference ($p<0.396$) was demonstrated between males and females.

Comparing iPTH values :

In comparing the PTH values, the results of the analysis showed statistically significant differences $p<0.001$ (Table 3.3.), and no statistical significance was found $p=0.354$ by gender.

Table 3.3. comparison of PTH values during treatment with Parsabiv i.v.

Comparison (F=87.22, p=0.001)							
RM Factor 1		**RM Factor 1**	**Mean Difference**	**SE**	**df**	**t**	**p**
PTH treatment	-	PTH 3m after treatment	295.5	50.1	25.0	5.89	< 0.001
	-	PTH 6m after treatment	547.3	80.3	25.0	6.82	< 0.001
	-	PTH 9m after treatment	845.1	85.9	25.0	9.83	< 0.001
	-	PTH 12m after treatment	937.1	94.7	25.0	9.89	< 0.001
PTH 3m after treatment	-	PTH 6m after treatment	251.8	39.6	25.0	6.36	<0.001
	-	PTH 9m after treatment	549.6	49.4	25.0	11.13	<0.001
	-	PTH 12m after treatment	641.6	58.7	25.0	10.92	<0.001
PTH 6m after treatment	-	PTH 9m after treatment	297.8	22.0	25.0	13.52	<0.001
	-	PTH 12m after treatment	389.8	30.6	25.0	12.73	<0.001
PTH 9m after treatment	-	PTH 12m after treatment	92.0	16.1	25.0	5.72	<0.001

The results showed that more than 50% of patients on etelcalcetide (Parsabiv) achieved more than a 30% decrease in PTH in the first trimester of treatment, and after 12 months, 70% of patients achieved a 60% decrease in PTH.

Task 4. To compare serum sclerostin levels in pre-dialysis patients and patients undergoing hemodialysis treatment and to evaluate the effect of etelcalcetide (Parsabiv) treatment on serum sclerostin levels in hemodialysis patients.

In the clinic of Nephrology and dialysis of University Hospital "St. Marina" Varna a comparative analysis of serum sclerostin levels in pre-dialysis patients (control group) and patients undergoing dialysis treatment was performed. A total of 89 patients were studied - 59 on HD and 30 - control group.

The role of etelcalcetide (Parsaviv) on serum sclerostin concentration in dialysis patients was also analyzed, and they were divided into two groups, 27 patients on HD and Parsabiv treatment and 32 patients on HD without Parsabiv treatment.

Table 4.1 shows the descriptive characteristics of sclerostin values in the study groups.

Table 4.1 Descriptive statistics of serum sclerostin in control group and group on HD treatment

		N	Arithmetic mean	Mediana	Standard deviation	Standart error
Sclerostin value pmol/l	HD	59	207	201	52.9	6.89
	Control group	30	61.5	63.5	17.5	3.20

In comparing the sclerostin values in the control and HD treatment groups, a statistically significant difference (p<0.001) was found (Table 4.2).

Table 4.2. comparison of serum sclerostin values in control group and group on HD treatment

Independent Samples T-Test

							95% Confidence Interval	
		Statistic	**df**	**p**	**Mean difference**	**SE difference**	**Lower**	**Upper**
Sclerostin value pmol/l	Student's t	14.6 [a]	87.0	< 0.001	145	9.95	125	165

[a] Levene's test is significant (p <0 .05), suggesting a violation of the assumption of equal variances

The results showed that patients undergoing extracorporeal treatment had up to 3 times higher serum sclerostin values compared to the control group - pre-dialysis patients (Figure 4.3).

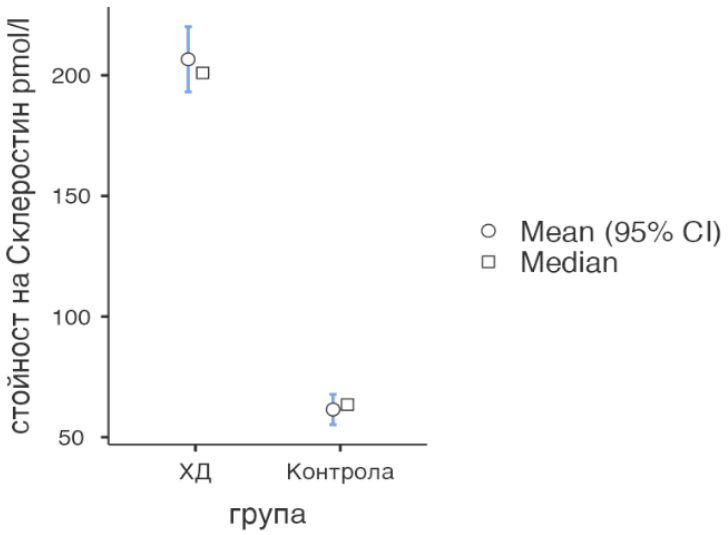


Fig. 4.3. comparison of serum sclerostin values in control group and group on HD treatment

Table 4.4 demonstrates the descriptive characteristic of sclerostin values by gender in the study groups.

Table 4.4 Descriptive statistics of serum sclerostin by gender in control group and HD group

		N	Arithmetic mean	Mediana	Standard deviation	Standart error
Sclerostin value pmol/l	women	40	132	156	54.7	8.65
	men	49	179	215	94.0	13.4

The comparative analysis of serum sclerostin values by gender showed statistical significance ($p<0.001$) (Table 4.5).

Table 4.5 Comparison of serum sclerostin values by gender

						95% Confidence Interval	
		Statistic	p	Mean difference	SE difference	Lower	Upper
Sclerostin value pmol/l	Mann-Whitney U	594	0.001	-56.0		-82.0	-30

From the analysis of the results, it was found that in terms of sex characteristics, men had higher serum sclerostin values than women. (Fig.4.6.)

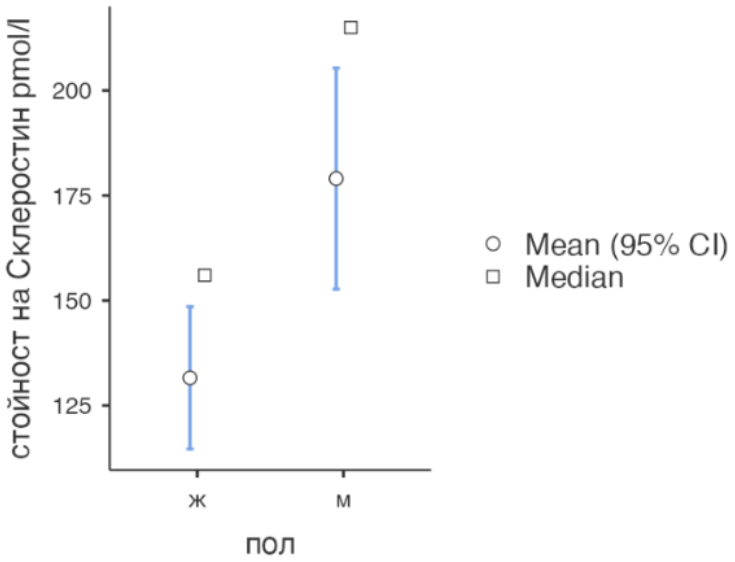


Fig. 4.6. comparison of serum sclerostin values by gender

The effect of treatment with etelcalcetide (Parsabiv) on serum sclerostin levels in hemodialysis patients was evaluated.

Table 4.7 provides a descriptive characterisation of sclerostin levels in haemodialysis patients with and without Parsabiv treatment

Table 4.7. descriptive statistics of serum sclerostin in patients with and without Parsabiv treatment

		бр	Средно аритметично	медиана	Стандартно отклонение	Стандартна грешка
Sclerostin value pmol/l	HD without Parsabiv treatment	32	131	124	19.3	3.41
	HD with Parsabiv treatment	27	251	256	39.7	7.64

The comparative analysis again revealed a statistically significant difference (p<0.001) (Table 4.8) (Figure 4.9).

Table 4.8. comparison of serum sclerostin values in HD patients with Parsabiv treatment and HD patients without Parsabiv treatment

							95% Confidence Interval	
		Statistic	df	p	Mean difference	SE difference	Lower	Upper
Sclerostin value pmol/l	Student's t	-15.1 [a]	57.0	<0.001	-120	7.93	-136	-104

[a] Levene's test is significant (p < .05), suggesting a violation of the assumption of equal variances

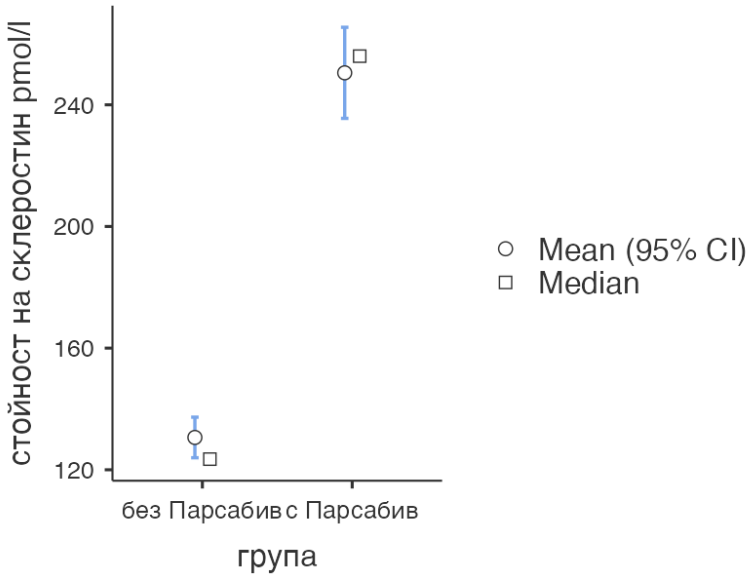


Fig.4.9. comparison of serum sclerostin values in HD patients with Parsabiv treatment and HD patients without Parsabiv treatment

From the results, it was found that etelcalcetide (Parsabiv) controlled secondary hyperparathyroidism and increased sclerostin levels in hemodialysis patients. (Fig.4.9.)

Task 5. To analyze and compare the effect of convective hemodialysis and hemodiafiltration on hyperphosphatemia in dialysis patients.

Fourteen patients (7 undergoing HD and 7 undergoing HDF) were studied for a period of 6 months in the Clinic of Nephrology and Dialysis at the University Hospital "St. Marina"- Varna, using a polysulfone capillary filter (1.6m2, 40 microm fiber inner diameter and 200 microm wall thickness for HD and HDF. Treatment time for all procedures was 4 hours, blood flow rate 300ml/min. Dialysis solutions and UF were the same for HD and HDF. Ca, P values were examined at 1st, 3rd and 6th month, iPTH values at 6th month.

Descriptive statistics were done with serum calcium values in HD group (Table 5.1) and HDF patients (Table 5.2).

Table 5.1 Descriptive characteristics of serum calcium values in HD patients

	N	Arithmetic mean	Standard deviation	Min	Max	ranks
Calcium 1m	7	2.5143	0.14920	2.25	2.70	2.57
Calcium 3m	7	2.4500	0.08660	2.30	2.50	1.71
Calcium 6m	7	2.4429	0.07868	2.30	2.50	1.71

Interpret the ranks from the nonparametric test, which shows that they are the same at month 3 and month 6. The test is statistically insignificant, no differences are found between periods and measurements (X2=4.571, p=0.102).

Table 5.2 Descriptive characteristics of serum calcium values in patients on HDF

	N	Arithmetic mean	Standard deviation	Min	Max	ranks
Calcium 1m	7	2.4214	0.15774	2.20	2.60	2.21
Calcium 3m	7	2.4143	0.13452	2.20	2.60	2.00
Calcium 6m	7	2.3886	0.11654	2.22	2.50	1.79

In patients on HDF, there was also no statistical significance in terms of serum calcium values at the time points of the studies (X2=1.20, p=0.549)

Graphical demonstration of serum calcium values by ranks at the three periods in HD patients (Fig. 5.3) and the HDF group (Fig. 5.4).

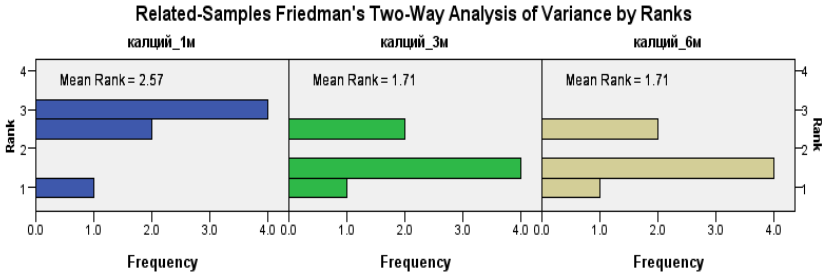


Fig. 5.3. comparison of calcium (Ca) values in the three periods in HD patients

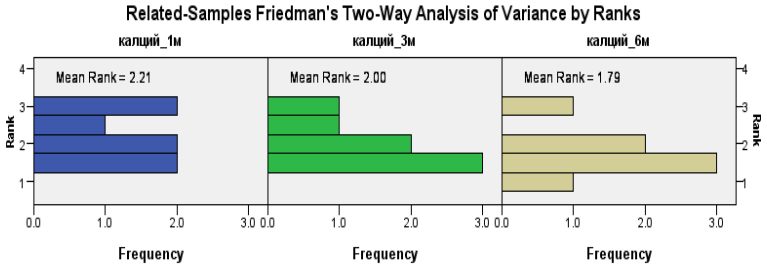


Fig. 5.4 Comparison of calcium (Ca) values by period in patients on HDF

Table 5.5. descriptive characteristics of serum phosphorus values in HD patients

	N	Arithmetic mean	Standard deviation	Min	Max	ranks
Phosphorus 1m	7	2.1000	0.11547	1.90	2.20	2.71
Phosphorus 3m	7	2.0429	0.12724	1.80	2.20	1.86
Phosphorus 6m	7	2.0143	0.12150	1.80	2.10	1.43

From the analysis, significant differences were found between periods and measurements (X2=9.333, p=0.09). Figure 5.6 illustrates the comparisons by ranks in the three periods.

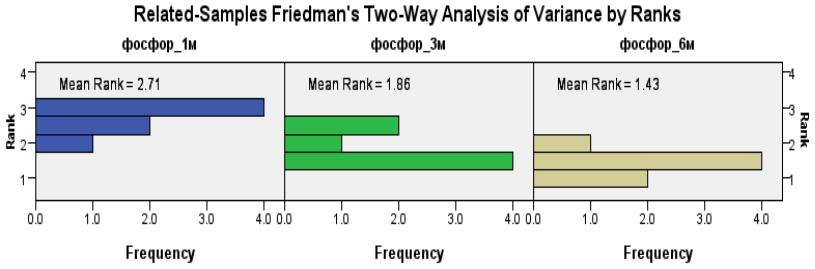
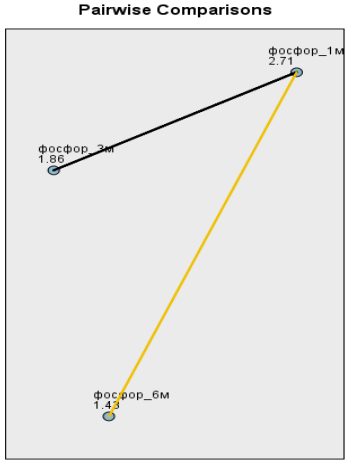


Fig. 5.6. comparison of phosphorus (P) values in the three periods in HD patients

Statistical significance regarding serum phosphorus values was found only between the 1st and 6th month (p=0.048). There was no statistically significant difference between the 1st and 3rd months and between the 3rd and 6th months.

Fig. 5.7 shows the specific differences in phosphorus values during the different periods (the statistically significant difference between 1st and 6th month is coloured yellow).



Sample1-Sample2	Test Statistic	Std. Error	Std. Test Statistic	Sig.	Adj.Sig.
фосфор_6м-фосфор_3м	.429	.535	.802	.423	1.000
фосфор_6м-фосфор_1м	1.286	.535	2.405	.016	.048
фосфор_3м-фосфор_1м	.857	.535	1.604	.109	.326

Fig. 5.7. differences in phosphorus (P) values during different periods in HD patients

In patients undergoing hemodiafiltration, there was a statistically significant difference in serum phosphorus in the periods studied (X2=14, p=0.001). There was a significant decrease in serum P as early as the 3rd month of HDF (Fig. 5.8).

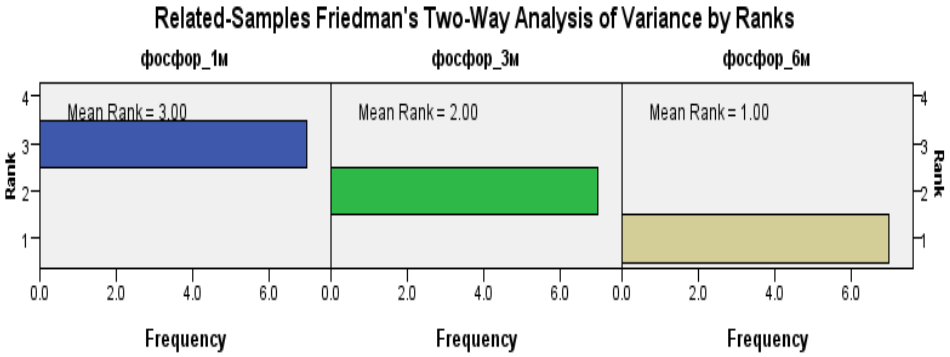
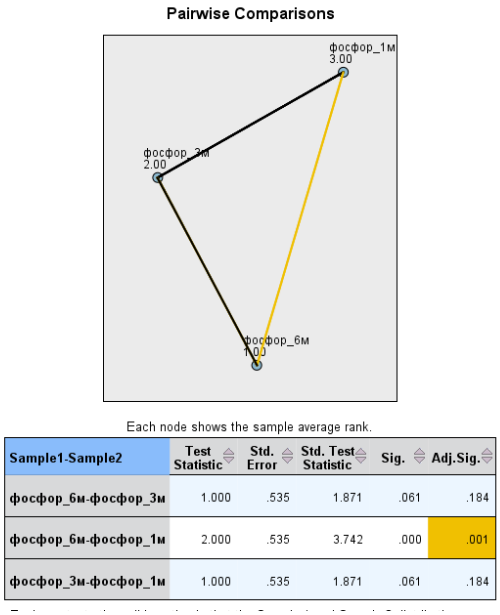


Fig. 5.8. comparison of phosphorus (P) values in the three periods in patients on HDF

Fig. 5.9 shows the specific differences of phosphorus values in the periods, with statistical significance between the 1st and 6th month.



Sample1-Sample2	Test Statistic	Std. Error	Std. Test Statistic	Sig.	Adj.Sig.
фосфор_6м-фосфор_3м	1.000	.535	1.871	.061	.184
фосфор_6м-фосфор_1м	2.000	.535	3.742	.000	.001
фосфор_3м-фосфор_1м	1.000	.535	1.871	.061	.184

Fig. 5.9. differences in phosphorus (P) values during different periods in patients on HDF

Table 5.10. descriptive characteristics of PTH values in HD patients

PTH	N	Arithmetic mean	Median	Standard deviation	Standard error
Baseline	7	1219	1050	587	222
6m	7	1107	1000	513	194

Table 5.11. descriptive characteristics of PTH values in HDF patients

	N	Arithmetic mean	Median	Standard deviation	Standard error
Baseline	7	1080	850	636	240
6m	7	781	590	556	210

Analysis of PTH values in patients undergoing HD did not demonstrate statistical significance between the two study periods (W=21, p=0.135). PTH values after the 6th month were similar to baseline, whereas serum PTH was statistically significantly different in patients on hemodiafiltration (W=28, p=0.016).

A comparative analysis of Ca, P and PTH values was done between the two groups of patients on HD and HDF.

There was no statistical significance when comparing calcium values between HD and HDF patients (X2=5.167, p=0.76). The results are described in Table 5.12 and Figure 5.13.

Table 5.12. descriptive characteristics of comparison of calcium (Ca) values in patients on HD and HDF

	N	Arithmetic mean	Standard deviation	Min	Max	ranks
Calcium 1m	14	2.4679	0.15518	2.20	2.70	2.39
Calcium 3m	14	2.4321	0.11026	2.20	2.60	1.86
Calcium 6m	14	2.4157	0.09959	2.22	2.50	1.75

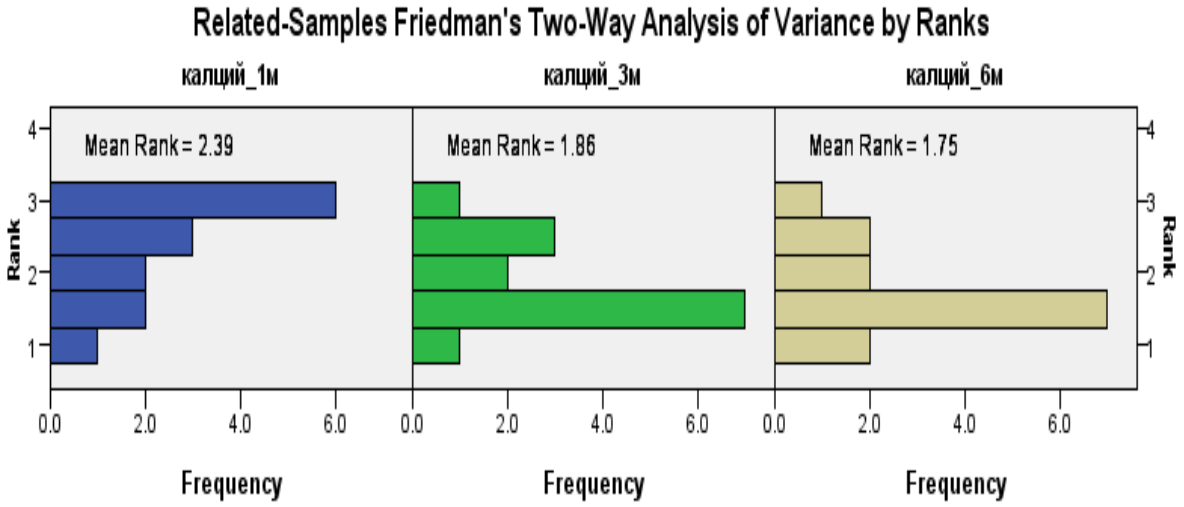


Fig 5.13. comparison of calcium (Ca) values in the three periods between patients on HD and HDF

Comparison of phosphorus values between the two groups showed an increase in phosphate clearance (Kd) (Fig. 5.14). Statistical significance was demonstrated between measurements in HD and HDF patients (X2=23.130, p=0.001) (Fig. 5.15).

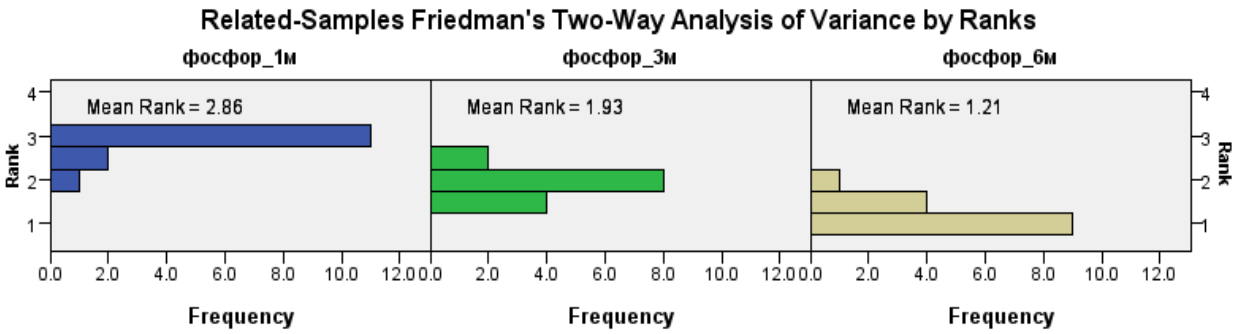
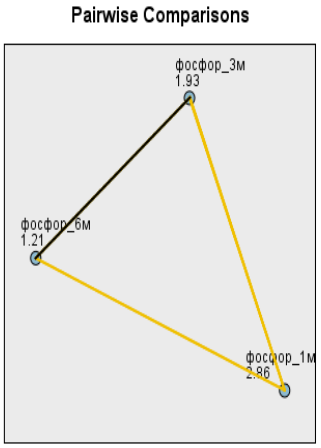


Fig 5.14. comparison of phosphorus (P) values in the three periods between patients on HD and HDF



Sample1-Sample2	Test Statistic	Std. Error	Std. Test Statistic	Sig.	Adj.Sig.
фосфор_6м-фосфор_3м	.714	.378	1.890	.059	.176
фосфор_6м-фосфор_1м	1.643	.378	4.347	.000	.000
фосфор_3м-фосфор_1м	.929	.378	2.457	.014	.042

Fig. 5.15. comparison of phosphorus (P) values during different periods in patients on HD and HDF

Significant differences were between 1st and 6th month (F=1.643, p=0.0001) and between 1st and 3rd month (F=0.929, p=0.042).

The proportion of patients reaching target phosphorus levels increased from 67% to 76% in patients on HDF and was stable in HD patients (63% and 63%); the difference between groups had statistical significance (P=0.04).

Phosphorus elimination was found to be better on HDF and may be considered as an additional treatment option for hyperphosphatemia in dialysis patients.

Regarding PTH values, Wilcoxson test revealed a statistical difference between the two groups after 6 months (W=36.50, P=0.038). (Tables 5.16 and 5.17).

Table 5.16. descriptive characteristics of comparison of parathyroid hormone (PTH) values in patients on HD and HDF

	group	N	Arithmetic mean	Standard deviation	Standart error
PTH - baseline	HD	7	1218.5714	586.86820	221.81533
	HDF	7	1080.0000	636.23895	240.47572
PTH 6m	HD	7	1107.1429	512.69596	193.78086
	HDF	7	781.4286	556.19027	210.22016

Table 5.17. comparison of parathyroid hormone (PTH) values in the three periods between patients on HD and HDF

	PTH Baseline	PTH 6m
Wilcoxon W	42.500	36.500
Z	-1.289	-2.047
p	.209[b]	0.038[b]

After 6 months, lower PTH values were observed in patients on HDF compared to patients on HD.

Task 6. To analyze the survival and quality of life in patients on dialysis treatment in terms of biochemical markers of BMD-CKD

A comparative analysis was conducted at the Clinic of Nephrology and Dialysis of the University Hospital "St. Marina" - Varna to investigate the factors influencing health-related quality of life (HRQoL) in patients on dialysis treatment.

Patients on dialysis with more than 1 year of HD treatment were studied. Quality of life was assessed using questionnaires.

Included 86 patients on HD with serum intact parathyroid hormone level was 153.55 +/- 823.5 pg/ml; serum calcium concentration was 2.46 +/-0.39 mmol/l, serum phosphorus concentration was 1.99 +/-0.61mmol/l. The age-related quality of life score of CKD G5 was 63.23 +/-11.45 and the KDQoL-SF-36 score was 55.74 +/-19.38.

Patients were divided into three groups according to serum Ca levels (Ca <2.2 mmol/l; Ca 2.2-2.55 mmol/l and Ca >2.55 mmol/l); serum P concentrations (P <1.2 mmol/l; P 1.2-1.8 mmol/l and P >1.8 mmol/l) and PTH levels (iPTH 150-300pg/ml; iPTH 300-600 pg/ml and iPTH above 600 pg/ml).

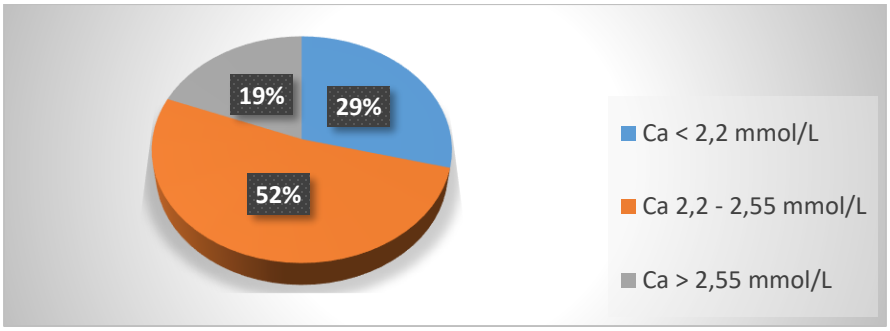


Fig. 6.1. Distribution of patients according to serum calcium values

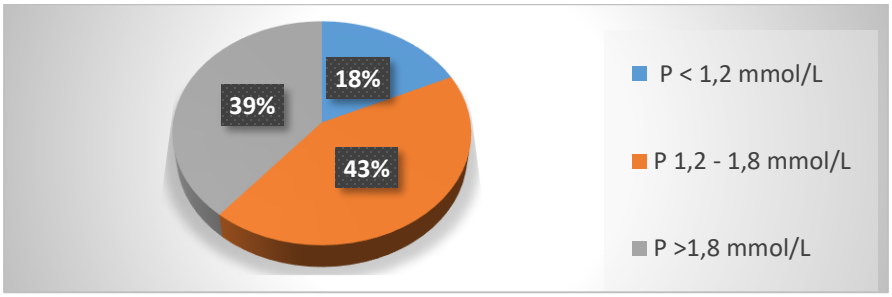


Fig. 6.2. Distribution of patients according to serum phosphorus values

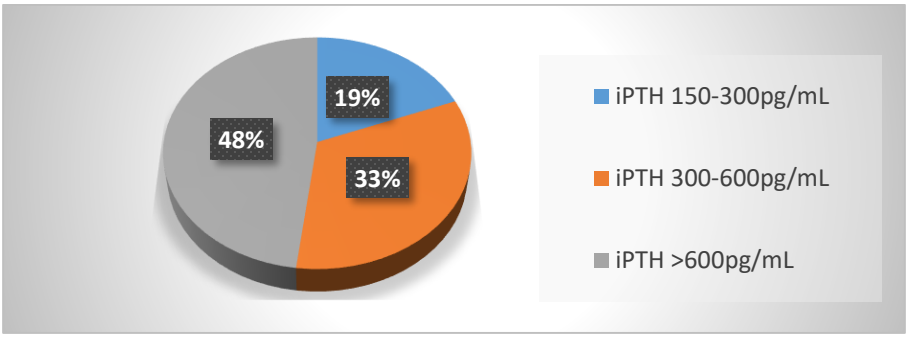


Fig. 6.3. Distribution of patients according to iPTH values

The following figures demonstrate the distributions of patients' specific responses from the survey questions.

Fig. 6.4, Fig. 6.5 and Fig. 6.6 summarise the impact of the haemodialysis procedure on physical activity and endurance in relation to Ca, P and iPTH values.

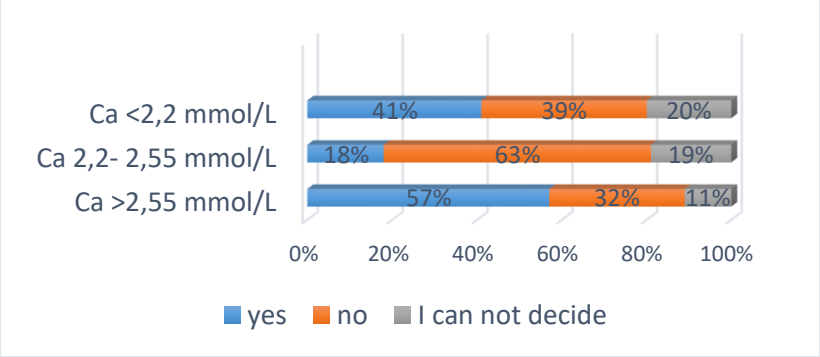


Fig 6.4. assessment of the effect of hemodialysis on physical activity and endurance according to serum calcium values

Statistically significant value (p=0.039) was observed in HD patients with serum Ca concentration above 2.55mmol/l.

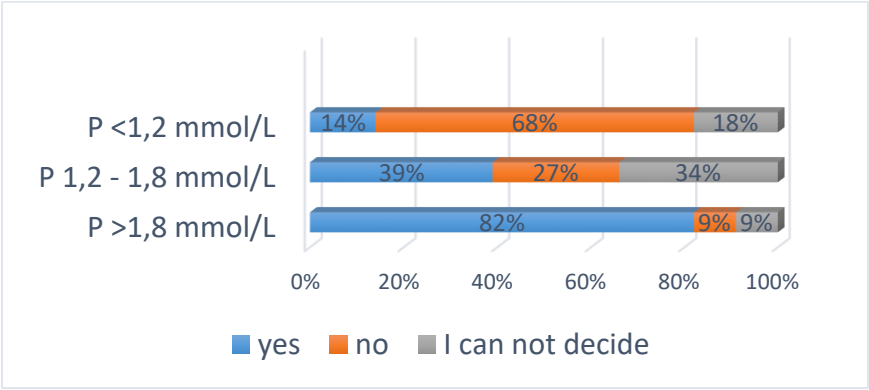


Fig. 6.5. assessment of the effect of haemodialysis on physical activity and endurance according to serum phosphorus values

The effect is significantly more pronounced in HD patients with serum phosphorus values above 1.8 mmol/l.

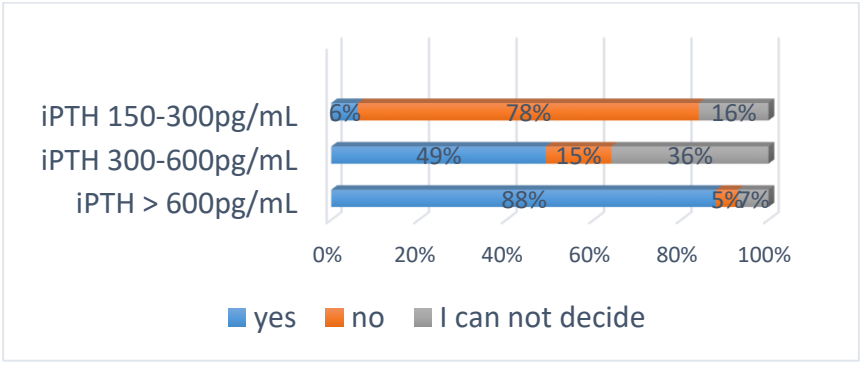


Fig. 6.6. assessment of the impact of haemodialysis on physical activity and endurance according to iPTH values

In more than half of patients with PTH values > 600pg/ml, HD affected physical activity and endurance, which affected their quality of life (p=0.022).

The effect of pain on the work environment and quality of life in dialysis patients was examined according to Ca, P and iPTH values (Fig. 6.7; Fig. 6.8 and Fig. 6.9).

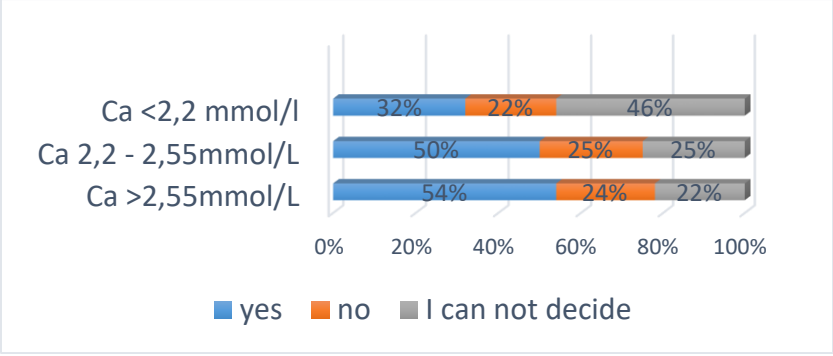


Fig.6.7. Assessment of the impact of pain on work environment and quality of life according to serum calcium values

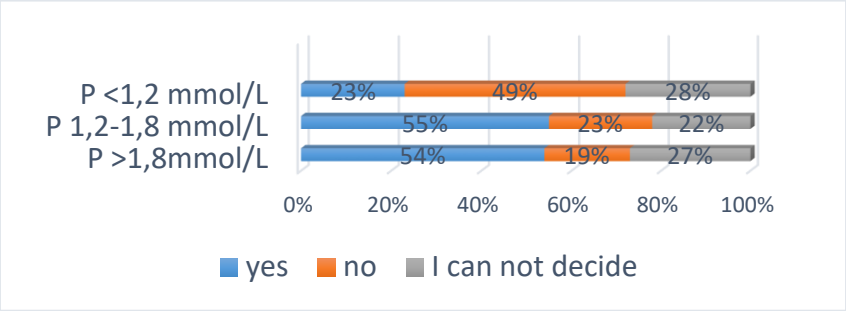


Fig.6.8. Assessment of the impact of pain on work environment and quality of life according to serum phosphorus values

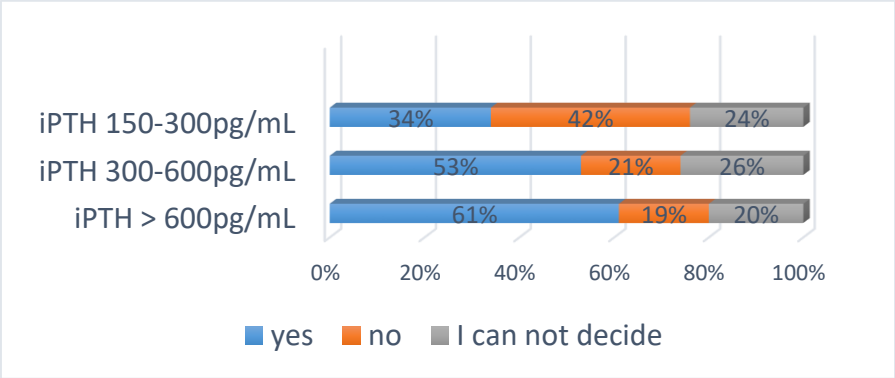


Fig.6.9. Assessment of the impact of pain on the working environment and quality of life according to iPTH values

Pain syndrome was found to affect the work environment in more than 50% of patients with hypercalcemia, hyperphosphatemia, and high iPTH values. Statistical significance was found with respect to their quality of life (p=0.025).

A pooled assessment of the impact of HD on the mental health and emotional stability of the patients was analyzed according to their Ca, P and iPTH values.

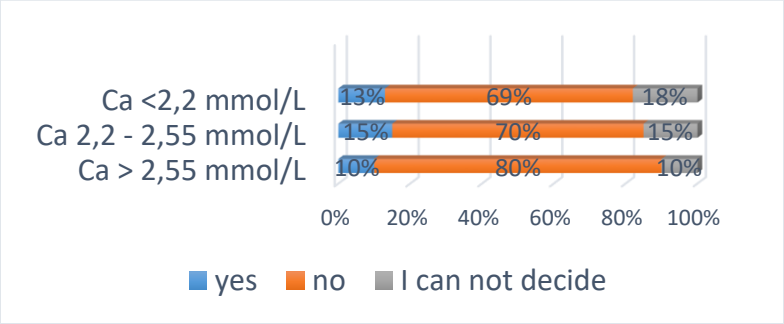


Fig 6.10. assessment of the impact of HD on mental health and emotional stability according to serum calcium concentrations

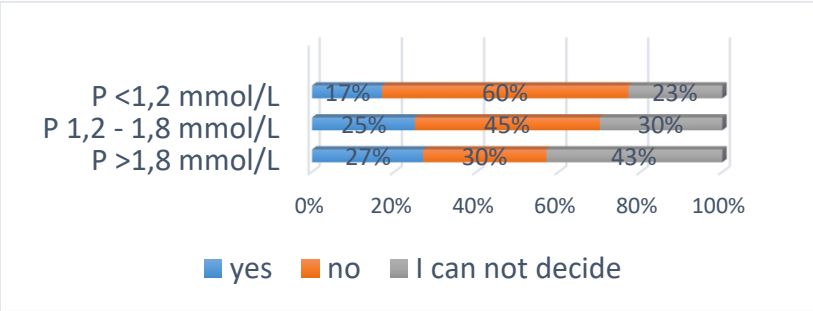


Fig 6.11 assessment of the impact of HD on mental health and emotional stability according to serum phophorus concentrations

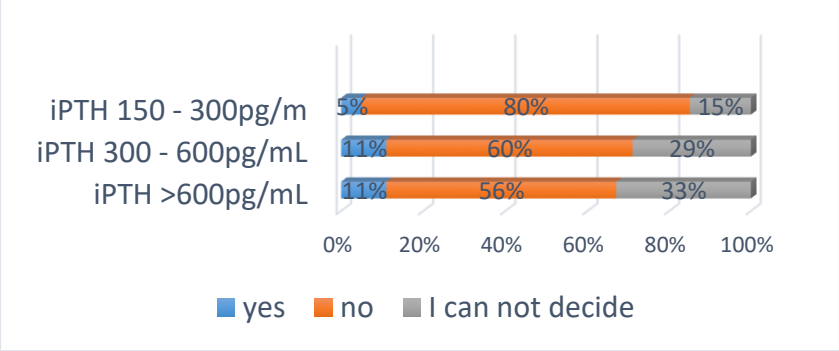


Fig 6.12. assessment of the impact of HD on mental health and emotional stability according to iPTH values

With regard to the assessment of the impact of hemodialysis on the mental health and emotional stability of patients, no statistically significant difference was found.

The results of the comparative analysis of patients' responses on their assessment of the impact of HD on their perception of general health according to Ca, P and iPTH values are illustrated in Figures 6.13; 6.14 and 6.15.

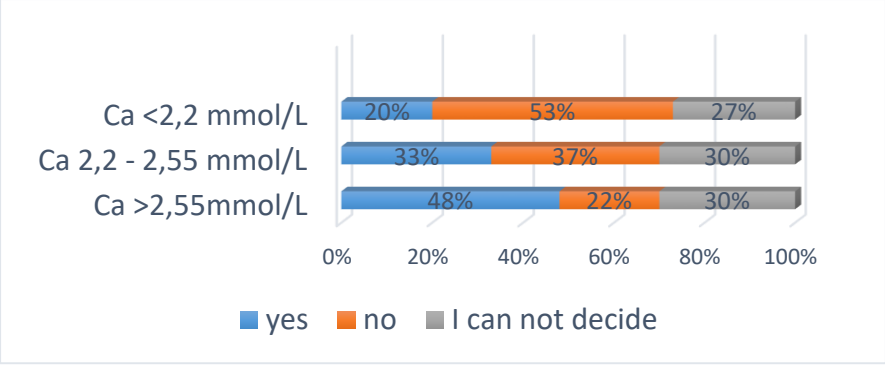


Fig. 6.13. assessment of the impact of HD on the perception of general health according to serum calcium values

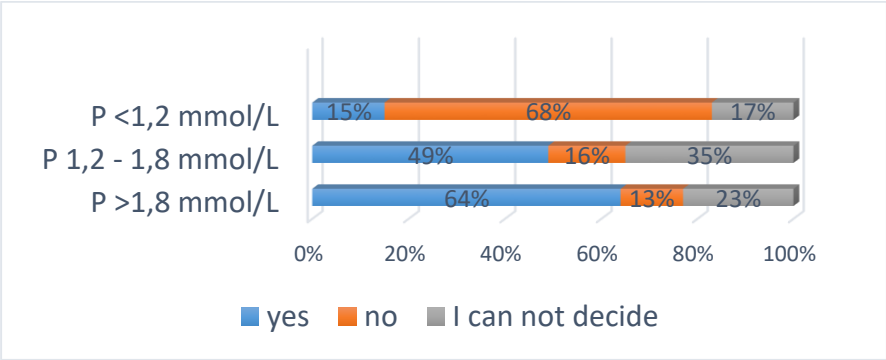


Fig. 6.14. assessment of the impact of HD on the perception of general health according to serum phosphorus values

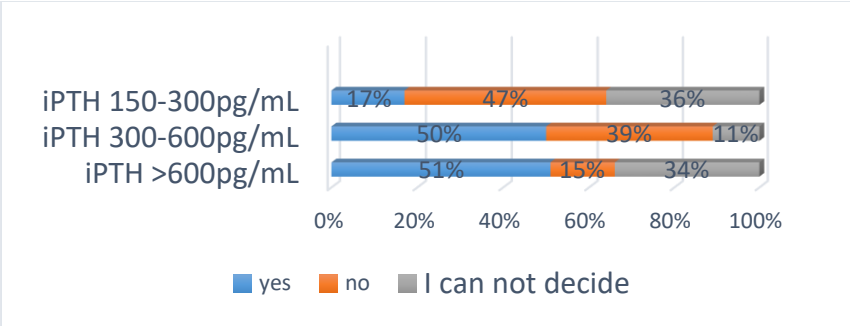


Fig. 6.15. assessment of the impact of HD on the perception of general health according to iPTH values

A significant proportion of patients in the high iPTH, Ca and P groups reported that HD and its complications had an impact on their perception of general health (p=0.003).

The highest HRQoL was reported in patients with PTH concentration 150 to 300pg/ml, Ca 2.10-2.55mmol/l and serum P concentration <1.2mmol/l

Satisfactory ratings of the degree of impact of hemodialysis treatment on emotional stability and mental health were prevalent. HD mostly had a moderate impact on patients' physical activity as well as their perception of general health.

V. DISCUSSION

Chronic kidney disease (CKD) is a global health problem affecting 5- 10% of the world's population, with the majority of these patients at increased risk of developing bone and mineral metabolism disorders. Affected patients present with symptoms such as: bone pain, muscle-tendon rupture, pruritus and high incidence of fractures. Subsequently, evidence suggests that patients are also predisposed to cardiovascular calcification with associated high morbidity and mortality.

Patients who suffer from ESKD have reduced renal function which alters calcium, phosphorus and vitamin D metabolism. These alterations often lead to SHPT, which is characterized by elevated PTH levels and is often associated with parathyroid hyperplasia (Cunningham, et al. 2011; Goodman and Quarles 2008; Joy, et al. 2007; Ruda, et al. 2004). This chronic and progressive disease develops early in the course of CKD, worsens as renal function declines, and affects most patients with advanced CKD. SHPT may manifest earliest in stage 3 CKD when the glomerular filtration rate (GFR) falls below 60 mL/min/1.73 m2 (Gal Moscovici and Sprague 2006; Joy, et al. 2007; Urena-Torres, et al. 2013). ESKD (CKD G5) is defined as the need for renal replacement therapy (dialysis or kidney transplant) where residual renal function is non-existent or found to be too low (GFR < 15 mL/min/1.73 m2) (Arneson, et al. 2013; Joy, et al. 2007; Nuijten, et al. 2009). In patients with CKD and SHPT, accompanying metabolic disturbances in calcium and phosphorus homeostasis can lead to pathological changes in bone tissue and vessels that may increase the risk of bone fractures and cardiovascular events (Moe and Drueke 2003). High levels of PTH, calcium, and phosphorus are associated with an increase in mortality rates in patients with SHPT on dialysis (Natoli, et al. 2013). (72); (75); (98); (100); (108); (137); (147); (149); (168); (188);

Serum P levels above 3.5 mg/dL, in pre-dialysis patients, are known to be associated with increased mortality. In CKD 5D, findings from observational studies show different values associated with risk of cardiovascular complications or death. However, an analysis of a cohort of 40,000 prevalent HD patients has demonstrated that the risk of death increases when phosphorus is above 5.0 mg/dL (Block GA et al. 2004). Thus, evidence suggests that serum phosphorus levels within the normal range are associated with better outcomes. Studies have shown that the serum phosphorus concentration remains within the normal range until the GFR drops to 20 to 30 mL/min. There is still a need for intervention studies that could more accurately identify optimal phosphorus levels in patients with CKD. (31); (99);

SHPT can lead to hyperparathyroid bone disease, a common cause of bone and joint pain in dialysis patients, decreased bone mass, and increased risk of bone fractures (Goodman and Quarles 2008; Saliba and El-Haddad 2009). The risk of hip fractures is approximately 3 to 4 times higher in the ESKD population than in the general mixed case population. Evidence suggests that severe uncontrolled SHPT contributes to a higher risk of fracture in ESKD patients: in the DOPPS study, PTH levels >900 pg/mL were associated with a 72% higher risk of fracture compared with PTH in the controlled range of 150 to 300 pg/mL (Jadoul, et al. 2006). A higher risk of fracture has also been reported in patients with elevated alkaline phosphatase, an important marker of bone condition associated with SHPT (Blayney, et al. 2008). Evidence is more limited in patients whose disease is less severe, as in these patients elevated PTH levels are only a moderate predictor of fracture risk or have no association with it at all. Disturbances in

calcium and phosphorus metabolism in SHPT are also thought to contribute to soft tissue and vascular calcification (Goodman and Quarles 2008), with a 50% to 80% prevalence of calcification in dialysis patients (Floege and Ketteler 2004). Vascular calcification, is considered a risk factor for the increased incidence of CV events and mortality in dialysis patients (Block, et al. 2017). Several large observational studies and a recent meta-analysis have consistently reported the association of high PTH, calcium, and phosphate levels with mortality (Block, et al. 2004a; Fernandez-Martin, et al. 2015; Floege, et al. 2011; Natoli, et al. 2013). The study conducted by Block et al. (Block, et al. 2004a) also reported a higher incidence of all-cause hospitalizations and hospitalizations related to CV events in association with elevated PTH and phosphate levels. Similarly, an analysis of DOPPS data demonstrated an increased risk of all-cause mortality and CV event-related mortality, as well as all-cause hospitalization and CV event-related hospitalization, in patients with elevated PTH levels (Tentori, et al. 2015). (29); (30); (55); (65); (66); (67); (68); (75); (78); (79); (95); (147); (184);

In addition, the COSMOS (Current Symptom Management of Secondary Hyperparathyroidism: A Multicenter Observational Study) (Fernandez-Martin, et al. 2015) also demonstrated that changes in biochemical parameters to control the SHPT are associated with improved survival. In a study conducted by Danese et al (Danese, et al. 2008), simultaneous control of PTH, calcium, and phosphate was associated with improved survival compared with single control of one or two of these parameters; similarly, long-term consistent control of these biomarkers was associated with improved survival compared with episodic control. Results from a recent large observational study categorising patients into phenotypes based on PTH, calcium and phosphate levels demonstrated that phenotypes with elevated PTH and phosphate levels and phenotypes with elevated PTH and calcium levels were at higher risk of death or hospitalisation related to a cardiovascular event compared with phenotypes where all three parameters were within target values. (53); (66);

A randomized clinical trial revealed a significant reduction in serum phosphate and a nonsignificant reduction in serum FGF-23 level, and worsening of coronary calcification scores in patients who received phosphate-lowering therapy. The updated KDIGO recommendation that prevention of hyperphosphatemia in patients with CKD stages G3a to G5D may be more important than treatment or normalization of phosphate levels. (88);

Graciolli FG et al. (2017) assume that some factors secreted by osteocytes play an important role in the pathophysiology of the complex disease, chronic kidney disease and impaired bone mineral metabolism. The authors investigated bone expression of some proteins in patients with chronic kidney disease at stages G2-3, as well as G4 and G5 on dialysis treatment, and in healthy subjects and analyzed comparatively the levels of various markers of bone remodeling in specific bone biopsy findings. As chronic kidney disease progressed, there was a decrease in serum calcium concentration and an increase in levels of phosphorus, alkaline phosphatase, fibroblast growth factor-23, parathyroid hormone, and osteoprotegerin. There is a gradual increase in bone resorption associated with decreased bone matrix formation and impaired bone mineralization. Bone expression of sclerostin and parathyroid hormone receptor-1 increases during the early stages, whereas that of phosphorus, alkaline phosphatase, fibroblast and phosphorylated β-catenin levels increases during the late stages of chronic kidney disease. Sclerostin and fibroblast growth

factor-23 have different immunohistochemical localization, indicating that they are secreted by different osteocytes. A positive correlation between serum concentrations and bone expression of fibroblast growth factor-23 was demonstrated. (47); (169);

Disorders of bone and mineral metabolism in patients with chronic kidney disease have been described in detail (D. Ionova, 2018), as well as in patients on hemodialysis treatment and after kidney transplantation (D. Ionova, 2015). New developments in calcium and phosphorus metabolism (D. Ionova, 2018) and its disorders in chronic kidney diseases (D. Ionova and E. Kumchev, 2015) are systematized. Some basic metabolic disorders in patients with chronic kidney diseases are presented (D. Ionova, 2014). New data on parathyroid hormone, C-reactive protein and the occurrence of vascular calcifications in patients with chronic kidney disease undergoing dialysis treatment are discussed. (4); (5); (6);

Prevention of hyperphosphatemia includes dietary phosphate restriction, use of phosphate-lowering agents, and dialysis for CKD patients with stage G5-D.

Three approaches that collectively work to control the 3 key laboratory values in BMD-CKD include diet, dialysis, and treatment with medications (phosphorus-binding agents, vitamin D analogues, and/or calcimimetics. These are referred to as the "3D" of hyperphosphatemia correction, diet, dialysis, and drugs (Diet, Dialysis, Drugs).

The dietary source of phosphate should be considered while making dietary recommendations. This is necessary because intestinal absorptive capacity varies for different sources of phosphate. The intestinal absorption rate of inorganic phosphate such as in supplements and beverages is between 80% and 100%, whereas that of plant-based phosphate such as nuts is between 20% and 40%.

Phosphorus-binding medications are usually administered with meals to limit phosphate absorption from the intestine by forming a non-absorbable complex with phosphate. The three main classes of phosphorus-binding medications are aluminum-based: Ca-based phosphates and non-Ca-based phosphate binders. Prolonged use of aluminum-based phosphorus-binding medications is limited by associated side effects, such as aluminum-induced osteomalacia and encephalopathy. The choice between the use of either calcium-containing or calcium-free phosphate binders should be guided by the patients' serum calcium and PTH levels. Excessive use of calcium phosphate binders has been associated with deleterious effects particularly in non-dialysis patients; for example, a study that compared calcium phosphate binders with a non-calcium-based phosphate binder (sevelamer) in patients on maintenance hemodialysis showed that coronary artery calcification occurred more rapidly in patients on calcium-containing, phosphate-binding medications. In addition to reducing calcium, sevelamer has been associated with lowering cholesterol and uric acid levels and has anti-inflammatory effects. In another study, the mortality outcome between patients on calcium phosphate binders and sevelamer was comparable. (92); (145); (146); (180); (185);

Phosphate removal by extracorporeal treatment is dependent on the type of dialysis and the duration of the dialysis session.

For a dialysis session duration of 4 hours at a frequency of 3 times per week, approximately 2.3 - 2.6 g of phosphorus per week is removed. If the session length is increased to 8 hrs 3 times per week (as in night dialysis), phosphate removal increases to 3.0 -3.6 g per week.

We found specific dynamic changes in serum concentrations of the modern and traditional markers of bone mineral metabolism included in our original constellation in patients with advanced chronic kidney disease. When comparing between the two groups - pre-dialysis and dialysis patients in terms of calcium, phosphorus and parathormone values, the group of patients on HD had higher phosphorus and parathormone values, and lower calcium values compared to the control group - pre-dialysis patients, which is consistent with modern findings.

Etelcalcetide directly interferes with the pathophysiology of SHPT by increasing the sensitivity of the calcium-sensitive receptor on the parathyroid glands to extracellular calcium. The results of two placebo-controlled trials and one trial comparing etelcalcetide with an oral calcimimetic indicate that etelcalcetide-treated patients can achieve clinically meaningful and sustained reductions in PTH, calcium, and phosphorus levels (Amgen 2014a; Amgen 2014b; Amgen 2015b). In addition, the intravenous (i.v.) route of administration of etelcalcetide contributes to better adherence to treatment, and may address some of the nonclinical reasons-patients may not adhere to or may discontinue treatment for SHPT (Amgen data on file 2015). Administration of etelcalcetide by MS may facilitate improved control of biochemical markers of SHPT in a broader spectrum of patients with SHPT. Thus, etelcalcetide may provide a treatment that addresses an unmet medical need of patients with CKD and secondary hyperparathyroidism. (13); (14); (15);

Block et al. conducted a phase 2, 3 parallel, randomized, placebo-controlled, double-blind, 26-week trial to evaluate the efficacy and safety of etelcalcetide in 1023 patients with CKD and secondary hyperparathyroidism on hemodialysis. The baseline characteristics in both studies were balanced. The primary efficacy end point was the proportion of patients achieving greater than a 30% reduction from baseline mean PTH concentrations during the evaluation phase (weeks 20 to 27). The secondary efficacy endpoint is the proportion of patients who achieve a mean PTH of 300 pg/mL or less. Patients were administered etelcalcetide or placebo three times weekly after hemodialysis. The mean age of patients was 58.2 years with a standard deviation of 14.4, and 60.4% of patients were male. A notable difference between the two trials was that laboratory data at pre-dialysis and post-dialysis, and electrocardiograms were performed in trial 2, whereas in trial 1 only pre-dialysis measurements were made. In less than 12.8% of patients, low baseline dialysate calcium concentrations were used (6.1% vs. 10.1%). The starting dose of etelcalcetide was 5.0 mg and could be increased by 2.5 mg or 5.0 mg during weeks 5, 9, 13, and 17 (maximum dose, 15 mg). Study drug is temporarily discontinued if the patient's PTH level is less than 100 pg/mL. The dose shall not be increased if the patient's PTH concentration is 300 pg/mL or less, serum calcium is less than 8.3 mg/dL, the patient is symptomatic with hypocalcemia, or at the discretion of the investigator. (77);

In study - 1, 254 patients were randomized to etelcalcetide, and 254 patients received placebo. 74% of patients randomized to etelcalcetide achieved the primary end point versus 8.3% of placebo-treated patients ($P<0.001$). Patients randomized to etelcalcetide were more likely to

achieve a PTH level of 300 pg/mL or less (49.6% for etelcalcetide versus 5.1% for placebo; P<0.001). The difference between patients achieving the secondary efficacy end point was significant (P<0.001).

In study - 2, 255 patients were randomized to etelcalcetide, and 260 were randomized to receive placebo. Of the patients randomized to etelcalcetide, 75.3% met the primary end point, whereas 9.6% met the primary end point for placebo (P<0.001). Patients randomized to etelcalcetide achieved a PTH level of 300 pg/mL, 53.3% versus 4.6% of patients on placebo.

Patients treated with etelcalcetide also showed decreases in serum intact fibroblast growth factor-23, bone-specific alkaline phosphatase, and collagen type-1 C-telopeptide. However, patients treated with etelcalcetide were more likely to experience muscle cramps, nausea, and vomiting than patients treated with placebo.

Based on the findings of these two trials, etelcalcetide demonstrated a beneficial effect compared to placebo on achieving a statistically significant decrease in PTH of over 30% in less than six weeks, along with a reduction in markers of bone function. The use of etelcalcide is generally safe and well tolerated.

We found that more than 50% of patients on etelcalcetide (Parsabiv) achieved more than a 30% reduction in PTH in the first trimester of treatment, and after 12 months more than 70% of patients achieved more than a 60% reduction in PTH.

In Table 7, we demonstrate the effect of different drug classes on the main markers of BMD-CKD:

Table 7. effect of drugs on markers of BMD-CKD

Class medication	Calcium	Phosphorus	Parathormone
Phosphorus-Binding	- / ↑	↓	↓
Vitamin D analogues	↑	↑	↓
Calcimimetics	↓	↓	↓

An additional phase 3 trial conducted by Block et al. is unique in that it represents a comparison between the relative efficacy and safety of intravenous calcimimetic etelcalcetide and oral calcimimetic cinacalcet. This study was a double-blind, randomized, active clinical trial that was conducted at 164 sites in the United States, Canada, Europe, Russia, and New Zealand over 26 weeks. Patients were randomized to i.v. etelcalcetide and oral placebo (n=340) or i.v. placebo and oral cinacalcet (n=343). The mean age of the enrolled patients was 54.7 years, and 56.2% were male. While the oral drug was administered daily, the i.v. drug was administered three times a week after hemodialysis. The primary efficacy endpoint was the noninferiority of etelcalcetide in achieving more than a 30% reduction from baseline in mean pre-dialysis PTH concentrations

during weeks 20 to 27. The secondary endpoints of this study were superiority in achieving biochemical endpoints greater than 50% and greater than 30% reduction in PTH and reporting only nausea and vomiting.

The results of this study concluded that etelcalcetide was non-inferior and superior to cinacalcet in achieving the primary endpoint. The difference in the proportion of etelcalcetide-treated patients achieving more than a 30% reduction in PTH concentrations compared with patients receiving cinacalcet was -10.5% (95% confidence interval, -17.5% to -3.5%; P for noninferiority, <0.001; P for superiority, 0.004). Of 178 patients randomized to etelcalzethide, 52.4% achieved greater than 50% reduction in PTH concentrations (vs 40.2% with sinacalcet; P<0.001). The most common adverse event was a decreased serum calcium value, which occurred in 68.9% of patients receiving etelcalcetide and 59.8% of patients receiving cinacalcet. Overall, etelcalcetide had minimal side effects, and the adverse effect profile consisted of a decrease in blood calcium, nausea, and vomiting. (77); (78);

Two, phase 3, randomized double-blind placebo-controlled clinical trials were conducted in parallel (Block, et al. 2017b). The primary objective of these studies was to demonstrate the superiority of etelcalcetide over placebo in terms of lowering PTH levels by >30% from baseline during the efficacy evaluation period (weeks 20 - 27). Patients were randomized 1:1 to etelcalcetide or placebo. Both groups were eligible to receive standard therapy (active vitamin D, phosphate binders, and calcium supplements) as needed. Secondary efficacy endpoints were examined for significance only if the primary endpoint was significant ($P < 0.05$). The efficacy analysis was based on the entire analysis population, which included all randomized patients. (79);

The two placebo-controlled trials were identical in design except that assessments of electrocardiograms (ECGs), laboratory and pharmacokinetic data before and after dialysis were performed in study 20120229, whereas only assessments before dialysis were performed in study 20120330.

An active-controlled, randomized, double-blind, double-masked phase 3 trial was conducted to compare the efficacy and safety of etelcalcetide with cinacalcet (Block, et al. 2017a). The primary objective of the study was to show that treatment with etelcalcetide was non-inferior to treatment with cinacalcet in lowering PTH levels by >30% from baseline during POE (weeks 20 - 27). If non-inferiority is demonstrated, the study could proceed with a sequential study to investigate whether etelcalcetide treatment is superior to cinacalcet treatment as defined by the three key secondary endpoints:

- decrease in PTH > 50% from baseline during POE
- decrease in PTH > 30% from baseline during POE
- average number of days of nausea and vomiting per week during the first 8 weeks

Patients were randomized 1:1 to receive etelcalcetide. In terms of superiority testing, the study had greater than 90% to detect statistically significant differences for each of the three key secondary endpoints, assuming a 5% significance level, two-tailed testing, and response rates of:

•60% etelcalcetide and 45% cinacalcet for >50% reduction in PTH

• 68% etelcalcetide and 57% cinacalcet for >30% reduction in PTH

• 0.1 etelcalcetide and 0.57 cinacalcet for mean number of days of vomiting or nausea per week (an overall standard deviation of 1.48 is allowed). (58); (59); (78); (141);

The analysis of the primary point is based on the Mantel-Haenszel method, with missing data explained using the null of uninformation. The pre-specified method of secondary endpoints at >30% and >50% reduction in PTH was imputation of treatment-naive patients. The efficacy analysis was based on the entire analysis population, which included all randomized patients.
The main clinical evidence that supports the use of etelcalcetide for the treatment of SHPT comes from the 3 RCTs (two placebo-controlled trials and a parallel clinical trial that compared etelcalcetide and cinacalcet) (Block, et al. 2017a; Block, et al. 2017b). The results of these trials suggest that patients treated with etelcalcetide can achieve clinically meaningful and sustained reductions in levels of the key biomarkers of SHPT: PTH, calcium, and phosphorus. (56); (141);
Studies of etelcalcetide have been performed for the treatment of SHPT in adult patients with CKD on hemodialysis. Available treatment options for SHPT used in this patient population (phosphate binders, vitamin D, and cinacalcet) do not always provide adequate control of PTH, calcium, and phosphorus levels, and are often associated with poor adherence and high rates of treatment discontinuation (Cozzolino, et al. 2015; Kilpatrick, et al. 2011; Park, et al. 2014; Reams, et al. 2015b). Etelcalcetide is a calcimimetic that can achieve clinically significant and sustained decreases in PTH, calcium, and phosphorus levels. In addition to this, the i.v. route of administration of etelcalcetide is also favoured with some of the non-clinical reasons for patients not adhering to or discontinuing treatments of SHPT. Thus, etelcalcetide may provide a treatment that addresses an unmet medical need of patients with CKD and SHPT. (50); (151); (161); (162);
The molecular size of sclerostin is approximately 22.5 kDa, and most of the sclerostin is probably filtered through glomeruli and reabsorbed by renal tubular cells in a normal kidney. B. Pietrzyk, K et al. 2019 first reported two enzyme-linked immunoassays, one to measure serum sclerostin levels and the other to measure plasma sclerostin levels, and the concentrations of sclerostin in serum and plasma were different when determined by the two methods. A comparative study of the two assays showed that plasma sclerostin levels were 30% higher than serum sclerostin levels, and that the coefficients of variation were less than 10% and less than 20%, respectively. (35); (123);
There is controversy about the mechanism involved in elevated serum sclerostin levels in patients with CKD. For example, Cejka et al. reported that renal elimination of sclerostin increased independent of the decline in renal function and urinary sclerostin excretion increased with decreasing GFR. Furthermore, increased extraskeletal production of sclerostin may be one reason for its high serum levels, whereas P. Kuczera et al. reported that iPTH levels did not increase in older individuals despite their high serum sclerostin levels. Circulating sclerostin levels have been found to be elevated in several cohorts of patients with CKD. Cejka et al. were the first to report finding elevated serum sclerostin levels in a cross-sectional study of dialysis patients, and their finding has been validated by other studies in patients with ESKD. Pelletier et al. reported that the higher serum sclerostin levels started at CKD stage 3a. However, the extent to which serum sclerostin levels reflect changes in expression versus accumulation in individuals with impaired renal function is not fully understood. A previous study examining local sclerostin expression across stages of CKD revealed that the highest osteocyte expression occurred in the

initial stages of the disease. Although, this study examined the number of sclerostin-positive osteocytes rather than absolute protein levels, the resulting data suggest that the accumulation of sclerostin in serum is due at least in part to increased osteocyte production. Furthermore, the rapid recovery of serum sclerostin to the normal range suggests that decreased renal clearance may also be responsible for the accumulation in the advanced stages. (39); (40); (153); (158);
A. Bouquegneau et al. 2020 reported finding that plasma sclerostin levels in hemodialysis patients were positively associated with phosphate levels and negatively associated with PTH levels. More recently, evidence of elevated serum sclerostin levels, and that serum sclerostin is closely related to serum phosphate and FGF-23 levels and vitamin D treatment in patients undergoing hemodialysis with low serum PTH levels, has been reported. Further study is needed to determine whether these relationships between serum sclerostin and PTH and FGF-23 levels are present in dialysis patients with spontaneously low PTH levels who are not treated with vitamin D. (25);

The role of sclerostin in BMD-CKD is an area of active research with conflicting results evaluating the association between serum sclerostin levels with vascular calcification, cardiovascular and all-cause mortality. Some authors report a positive association between sclerostin levels and all-cause mortality, whereas others report the opposite. The relationship between sclerostin levels and mortality in CKD patients awaits further elucidation.

Treatment of SHPT with calcimimetics reduces calcium, phosphate, and FGF-23 levels in patients with CKD, but the effect of these agents in sclerostin concentration remains to be elucidated.

The reasons for the increased cardiovascular risk associated with kidney disease reside in part in the chronic kidney disease-bone mineral disorder syndrome. Three cardiovascular risk factors [hyperphosphatemia, vascular calcification, and elevated fibroblast growth factor-23] have been identified within CKD-BMD in recent decades. Furthermore, sclerostin has recently presented as a novel biomarker for bone and vascular disease. This 22-kDa glycoprotein, secreted mainly by osteocytes, is a soluble inhibitor of the Wnt signaling pathway, which plays a major role in bone turnover. Higher levels of sclerostin have been reported in patients with CKD, and levels decrease during dialysis. Sclerostin is associated with vascular calcification and cardiovascular risk in CKD, although the data are still controversial. (200);

We found that patients undergoing extracorporeal treatment had up to 3 times higher serum sclerostin values compared to the control group – pre-dialysis patients. We also investigated the use of i.v. etelcalcetide in patients on HD treatment and evaluated the effect of Parsabiv treatment on sclerostin levels. The treatment of SHPT with etelcalcetide (Parsabiv) increased serum sclerostin concentration in patients undergoing hemodialysis.

Much recent evidence suggests that sclerostin also plays a role in uremic and non-uremic cardiovascular calcification processes: according to previous studies, both calcific aortic stenosis and calciphylaxis go together with de novo local sclerostin synthesis in vascular tissue and interstitium. Most notably, high levels of circulating sclerostin correlate positively with the extent of valvular calcification as assessed by quantitative computed tomography. Within the complex spectrum of BMD-CKD, dysregulation of sclerostin is a signalling early event.

Viaene et al. (2020) measured sclerostin levels in 100 dialysis patients from a single center who were followed for a median of 637 days. They included patients at baseline, with a median duration of hemodialysis of 40 months (16-69 months). Viaene et al. defined their cohort according to median sclerostin level and found a significant survival benefit for patients above the median after adjusting for age and sex (HR 0.33, 95% CI: 0.15-0.73, P = 0.006). However, within a fully adjusted model including bone alkaline phosphatase, the correlation between HD duration and sclerostin was not statistically significant. In contrast, the NECOSAD study showed an independent association between sclerostin and outcome, even after adjustment for AP. The reasons for these discrepancies between the two cohorts remain speculative. Problems and differences between sclerostin data may be involved. Cohort characteristics as well as characteristics of the sclerostin data may also contribute to the fact that the current NECOSAD cohort was unable to reproduce the differences between males and females as previously described. However, the key message is comparable in both studies, i.e the higher the circulating sclerostin - the better the outcome. (189);

Over the past few decades, hemodialysis (HD) has been the main renal replacement therapy for patients with end-stage kidney disease (ESKD). Although, extracorporeal treatment has made great scientific and technological advances, the morbidity and mortality of ESKD patients are higher than those in the general population. Cardiovascular events remain a leading cause of mortality in patients with end-stage kidney disease. The causes of high cardiovascular mortality are not yet fully understood, one of them is probably related to uremic toxicity. Therefore, dialysis modality may achieve higher elimination in uremic toxins administered in clinical practice.

Hemodiafiltration (HDF) is a dialysis modality using a kind of "high flux" hemodialysis water filter combined with a large number of plasma technology to increase convective transport for the removal of uremic toxins. Online hemodiafiltration (online HDF) is a new hemodialysis technique combining convection and diffusion and thus enabling the purification of large molecules.

Technological developments in membranes, instrumentation and dialysis solutions have contributed to making hemodiafiltration a safe and effective technique. Accurate volumetric ultrafiltration control systems in dialysis machines reduce the risk for fluid balance errors and allow for safe and effective online HDF. In fact, modern dialysis machines are equipped with specific balancing systems to control fluid reinfusion and ultrafiltration simultaneously.

Several studies have addressed the advantage of HDF compared to convective HD. The Convective Transport Study (CONTRAST) and the French Convective versus Hemodialysis in Elderly (FRENCHIE) trials, showed the effect of HDF treatment on serum phosphate concentration. Other studies have also shown that HDF is superior to convective HD in reducing inflammation, preventing protein loss, and reducing episodes of intradialytic hypotension. Several studies have shown beneficial effects on patient survival. The Dialysis Outcomes Study and practice patterns reported for the first time a 35% lower mortality when conducting higher-volume HDF compared with "low-flux" HD. Two European randomized control trials, CONTRAST and a Turkish study, demonstrated reduced mortality with high convection volumes

(>22 L/session) in their analyses. In the Estudio de Supervivencia de Hemodiafiltración Online trial, the primary analysis also demonstrated a 30% lower risk of both all-cause and cardiovascular mortality in patients with HDF than in those with "high flux" HD. (108); (120); (144); (166);

A literature review reveals that there are few long-term follow-up observational studies comparing online HDF with "high flux" HD in broad-spectrum large molecule removal. One small study observed 31 patients undergoing online HDF over a 4-year follow-up period. It aimed to compare the evolution of the following aspects before and after starting online HDF: dialysis dose, medium/large molecule clearance, inflammation, nutrition, Ca-P metabolism, anemia, and intradialytic complications. Online HDF increased Kt/V to 31.0% ($p>0.001$) and decreased post-dialysis beta-2-microglobulin to 66.4% ($p>0.001$). The other parameters analyzed did not vary significantly. During online HDF, episodes of intradialytic hypotension decreased by 45% compared with conventional hemodialysis and no complication was found. (114);

The CONTRAST study showed that pre-dialysis serum phosphate levels decreased by 6% and the proportion of patients achieving target serum phosphorus levels increased from 64% to 74%. Alternatively, the Turkish OL-HDF (Oket et al. 2013) and ESHOL studies revealed that there was no variation in serum phosphate levels. The differences between these studies and their inconclusive results can be rationalized on the fact that hyperphosphatemia is inadequately controlled, although, dialysis appears adequate and within reference values based on urea kinetics (Kt/V). Removal of phosphate depends on residual renal function levels and the use of specific medications for chronic kidney disease-mineral bone disorder, which includes calcimimetics, vitamin D analogues, and phosphate binders. Dialysis is only one element among many complex interactions. Therefore, HDF is an inappropriate choice if the only aim is to enhance clearance of small molecules such as phosphate. (37); (38); (94);

HDF offers greater clearance levels for other molecules, including complement factor D (a proinflammatory mediator), leptin, fibroblast growth factor-23 (associated with vascular calcification and metabolic bone disorder), several cytokines, glycated end product precursors, and circulating advanced glycation end products.

We found that there was an improvement of phosphate elimination in patients on online HDF. The proportion of patients reaching the phosphate treatment goal increased from 67% to 76% in patients on HDF and was stable in HD patients (63% and 63%).

Phosphate levels decreased from 2.40 +/- 0.10 (SE) mg/dL at baseline to 1.2 +/- 0.10 mg/dL after 6 months in patients undergoing HDF ($p<0.001$) and were stable in hemodialysis patients (2.50 +/- 0.10 mg/dL at baseline and 2.10 +/- 0.10 mg/dL after 6 months; ($p=0.048$).

Chronic kidney disease-bone mineral disorder (CKD-BMD) is a complication of chronic kidney disease (CKD) that seriously affects the prognosis of patients on hemodialysis, remarkably increasing the relative risks of all-cause mortality and cardiovascular mortality in these patients. It is also considered an important factor affecting HRQoL. Based on cross-sectional studies of hemodialysis patients, serum phosphorus (P) and intact parathyroid hormone (iPTH) values that are too high or too low are associated with low HRQoL. However, previous studies conducted

on this topic have mainly focused on the effects of individual CKD-BMD biochemical parameters on HRQoL, but these studies have not examined the combination of serum phosphorus, serum calcium, and iPTH values in assessing their correlation with HRQoL. There is a close relationship and interaction among these three factors.

We performed an analysis to examine factors influencing health-related quality of life (HRQoL) in CKD-BMD patients on hemodialysis treatment. This analysis aimed to observe the relationships between different combinations of serum Ca, P, and iPTH levels and HRQoL in patients on extracorporeal treatment and to explore the associated factors affecting HRQoL in these patients.

We used the KDQoL-SF-36 examination and assessment questionnaire after modification. This instrument has a number of advantages - validated in English, with easy iterpretation, the possibility of calculating two summary indicators of physical and mental health, comparing the obtained data with other populations, etc. (3);

The questionnaire includes 8 scales assessing different aspects of health: 1) physical activity; 2) physical endurance; 3) emotional stability; 4) social activity 5) mental health; 6) bodily pain; 7) vitality (energy/fatigue); 8) perception of general health. Responses included yes/no/cannot judge.

The questionnaire is self-administered, which takes 5-10minutes. When the card is completed, the SF-36 questions are scored, using a developed criteria to calculate the importance of individual responses, using a scoring system. The points given on the individual scale are used primarily for preliminary and indicative assessment. This assessment is carried out on the main eight aspects of the questionnaire and is an absolute assessment of the level of quality of life obtained by converting qualitative attributes into an individual rating scale of a certain dimensionality.

We found that bone mineral disorders significantly affected HRQoL. Correction of abnormal values of serum phosphorus, serum calcium and iPTH is of great importance to improve the quality of life in patients with CKD-CKD on dialysis treatment.

The widespread use of this questionnaire is related not only to its universal applicability to different disease states, but also to the common perception of different aspects of health, in general, physical, mental and social.

VI. CONCLUSION

SHPT is a chronic disease that develops early in CKD when decreased kidney function alters calcium, phosphorus and vitamin D levels. SHPT worsens with worsening renal function and is prevalent in the dialysis population, characterized by elevated parathyroid hormone levels. Abnormal values in calcium and phosphorus metabolism are common and metabolic bone disease develops frequently in patients with chronic renal failure. Effective clinical management includes measures to control phosphorus retention and prevent hyperphosphatemia, to maintain serum calcium concentrations within the normal range, and to prevent excess secretion of parathyroid hormone by using vitamin D analogues. Despite the importance of controlling phosphorus retention and preventing hyperphosphatemia in patients with CKD, current management strategies are often inadequate, particularly in those involving diets containing amounts of protein. Calcium-free phosphate-binding agents and novel vitamin D analogues and calcimimetic compounds offer new therapeutic alternatives for the management of BMD-CKD. Integrating these medications into existing treatment regimens may provide safer and more effective methods of controlling secondary hyperparathyroidism and renal bone disease while limiting the risks of vascular calcification in patients with CKD.

Phosphorus control is complex but important to the overall health and well-being of CKD patients, and understanding why and how phosphorus should be controlled is important for the entire healthcare team. One in 3 patients do not have phosphorus values below 5.5 mg/dL, and two in 3 patients have not reached target phosphorus levels as recommended by the most recent KDIGO guidelines. This indicates that it is time to reevaluate the approach to phosphorus management in ESKD patients. In the integrated approach, 3D - Diet, Dialysis, and Drugs (DDD) is used to simultaneously manage not only phosphorus but all 3 key laboratory values (calcium, phosphorus, and PTH).

Etelcalcetide is a novel intravenous calcimimetic drug that has shown better control of biochemical parameters compared with placebo and cinacalcet-based regimens for the treatment of SHPT in CKD patients on hemodialysis. It has an overall good tolerability and adverse event profile that is consistent with pre-existing comorbidities associated with SHPT and with the mechanism of action of calcimimetics. This favourable benefit/risk profile, combined with the ease of intravenous administration at the end of dialysis (giving professionals flexibility and control over administration) means that etelcalcetide represents a significant advance over existing therapies. Parsabiv (etelcalcetide) represents another option for controlling elevated parathyroid hormone levels in the treatment of CKD-BMD in hemodialysis patients. The availability of a parenteral formulation offsets the need to administer another oral medication for these patients, and post-dialysis seance administration ensures compliance. It has demonstrated comparable efficacy to cinacalcet in clinical trials, with reduced potential for drug-drug interactions and a favorable safety profile

The current treatment paradigm consists of a multifaceted, integrative approach to the control of hyperphosphatemia that includes serial studies of calcium, phosphorus, and PTH and must be accompanied by an understanding of the relationships between these markers, their uptake and

release from the gut and bone, and fluctuations with disease progression and treatment. When taken together, these factors should facilitate optimal patient management.

Online HDF is the better choice for patients in whom we need to increase replacement therapy, such as patients with large body surface area, patients with long-standing HD, and for whom we wish to prevent amyloidosis. Online HDF is safe and better tolerated than conventional hemodialysis.

Measuring and assessing quality of life in patients with CKD G5 allows for a more complete understanding of their specific needs and increased effectiveness of clinical management. This is a potential opportunity to improve the quality and outcome of health care and should be widely applied in Bulgarian medical practice, especially in the assessment of the quality of care and health management.

VII. FINDINGS

- It has been proven that, the group of HD patients had higher phosphorus and parathormone values and lower calcium values compared to the control group - pre-dialysis patients.
- Treatment with calcium carbonate and a combination of sevelamer and cinacalcet has been found to show a significant difference in hyperphosphatemia and SHPT.
- The present study proved for the first time in Bulgaria that more than half of the patients on HD treatment and etelcalcetide (Parsabiv) used, achieved more than 30% decrease in PTH in the first trimester, and more than 60% in the first year of treatment.
- It has been found that patients undergoing extracorporeal treatment had up to 3 times higher serum sclerostin values compared to the control group, pre-dialysis patients.
- In patients undergoing HDF, phosphorus elimination increased from 67% to 76%, whereas it was constant in patients on convection dialysis, indicating that HDF is a modality that can be considered as an additional treatment option for hyperphosphatemia in dialysis patients.
- Bone mineral disorders have been found to significantly affect quality of life in patients with CKD. Correction of abnormal values of serum phosphorus, serum calcium and iPTH are of determinant importance to improve quality of life in CKD-CKD patients on dialysis treatment.

VIII. CONTRIBUTIONS

Theoretical contributions:

1. For the first time in Bulgaria the serum biomarker Sclerostin has been studied and interpreted in patients with CKD - dialysis and pre-dialysis stage, which has a significant diagnostic - prognostic significance.
2. The correlation between etelcalcetide (Parsabiv) and elevated concentrations of the serum biomarker Sclerostin in patients undergoing hemodialysis treatment has been demonstrated for the first time in our country.
3. The high efficacy and comparative safety of Sevelamer and Cinacalcet in monitoring of hyperphosphatemia in patients with chronic kidney disease and impaired bone mineral metabolism have been established.
4. Demonstrate the advantage of a contemporary modality, hemodiafiltration, in patients with hyperphosphatemia for the prevention and treatment of SHPT.
5. Bone-mineral disorders are found to significantly affect physical activity, emotional and mental stability in patients, as well as their perception of general health.

Practical contributions:

1. The serum biomarker Sclerostin studied for the first time in our country in patients with chronic kidney disease allows to detect and interpret significant disturbances in bone-mineral metabolism and prevention of cardiovascular diseases.
2. Evaluated the use and benefits of intravenous Etelcalcetide and its effect on serum Sclerostin and PTH levels in patients on hemodialysis with SHPT.
3. Measuring and assessing quality of life in patients with CKD and hyperphosphatemia allows for a more complete understanding of their specific needs and increased effectiveness of clinical management.

IX. PUBLICATIONS RELATED TO THE THESIS

1. **С.Атанасова** - Нарушения в метаболизма на калция и фосфора при болни с хронични бъбречни заболявания - сп. Нефрология, диализа и трансплантация година 25, брой 4, 2019 стр. 18-24

2. **Снежана Атанасова** - Parsabiv (Etelcalcetide) - алостеричен модулатор на калциево - чувствителния рецептор при пациенти на хемодиализа с ВхПТ - сп. Актуална нефрология брой 1, том 14,2020 стр.9-12

3**.** Д. Ненова, **С.Атанасова**, С.Стайкова - Анализ върху постигнатото качество на живот при пациенти с краен стадий на ХБЗ, провеждащи online-хемодиалифтрация - списание „ Актуална Нефрология „ бр.1 том15 2021г стр – 39-44