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Research article

Nationwide Hypophosphatemic Rickets Cohort Study

Runing Title: Nationwide Hypophosphatemic Rickets

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What is already known on this topic?
Hypophosphatemic rickets (HR) is a rare renal phosphate wasting disorder commonly related to X-linked form. There is no nationwide data on HR with initial and follow-up findings

What this study adds?
The age of diagnosis were similar in good and bad responder to conventional therapy. Good responders were better height SDS on admission. Higher treatment doses lead to nephrocalcinosis without any change in serum levels of phosphorus. The awareness about early diagnosis and complications should be improved.

Abstract:
Aim: Hypophosphatemic rickets (HR) is a rare renal phosphate wasting disorder commonly related to X-linked form, caused by *PHEX* mutations and its treatment and follow-up is challenging due to imperfect treatment options.
Here we presented nationwide data on HR with initial and follow-up data on the patients.
Results:
From 24 centers, 166 patients were included in the study data. Genetic analysis (n:75) showed PHEX mutation in 80% patients. The mean follow-up period of the patients was 6.7± 2.4 years. During first 3 year treatment (n:91), mild increase in phosphate , decrease in ALP and, elevation in PTH levels had been detected. The height SDS were -2.38, -2.77, -2.72, -2.47 at intitial, 1st, 2nd and 3rd year of treatment, respectively (p>0.05). In follow-up: 36% of the patients showed complete or significant improvement in leg deformities, and, these patients had similar phosphate levels at presentation with better levels in 1st and 2nd years of treatment, even the treatment doses of phosphate were similar. Furthermore, 27 patients developed nephrocalcinosis, the patients showed no difference in biochemical differences in presentation and follow-up, but 3rd year PTH was higher, however, higher treatment dose of phosphate and calcitriol has been detected in nephrocalcinosis group.
Conclusion: HR treatment and follow-up is challenging and our results showed higher treatment doses leading nephrocalcinosis without any change in serum levels, suggesting given higher doses lead higher phoshaturia probably through the stimulation FGF23. However, higher calcitriol doses could improve bone deformities. Safer and more efficacious therapies are needed.
Key words: hypophosphatemic rickets, *PHEX*, treatment

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Introduction

Hypophosphatemic rickets (HR) is a rare renal phosphate wasting disorder caused by several genetic mutations in factors leading to increase in FGF23 signalling or secretion , and in renal phosphate transporters (1). The most common inherited form of HR is X-linked hypophosphatemic rickets (XLH; OMIM: #307800), where inactivating mutations of the Phosphate Regulating Endopeptidase Homolog, X-Linked (*PHEX*, OMIM: #300550) gene lead to local and systemic effects (2). The incidence of XLH is 1/20 000 live births, and it accounts for approximately 80%of familial cases (3).

PHEX is predominantly expressed in osteoblasts and downregulation of PHEX increases serum levels of the phosphatonin, fibroblast growth factor 23 (FGF23). FGF23 has a central role in HR pathophysiology. Elevated levels of serum FGF23 increase urinary phosphate excretion by downregulating renal sodium-phosphate transporters, and decrease intestinal phosphate absorption by restricting active vitamin D synthesis (2). The other rare genetic disorders of excess FGF23 are autosomal dominant HR (caused by missense mutation in *FGF23*), autosomal recessive HR (type 1caused by mutations in the gene encoding dentin matrix protein (*DMP1*), type 2 caused by mutations in ectonucleotide pyrophosphate/phosphodiesterase 1 (*ENPP1*), type 3 caused by (*FAM20C*) (4).

Clinical presentation of HR include rickets, osteomalacia, short stature, leg deformities, dental abscesses, premature cranial synostosis, muscle weakness in children, and also pseudofractures, osteoarthritis, entesopathy in adults. The phenotype can be very widely, even in the same family and diagnosis can be delayed (5). In addition to the rarity and diagnostic difficulties, which has a significant impact on patient outcomes , treatment and follow-up of HR is very challenging.

Conventional treatment of HR includes a combination of oral phosphorus and active vitamin D. Unfortunately this therapy was unsuccessful in a significant proportion of patients in respect to healing of rickets and improvement in deformities, and can be associated with treatment related side effects (4,6).

Information about the clinical, laboratory, genetic and follow-up characteristics of HR patients is very scarce for our population, only few small series have been reported (7,8).

We aimed to present a nationwide data on HR with initial and follow-up data on the patients resented to the pediatric endocrinology clinics before the age of 18 years.

Cases and Methods

In this study, we cross-sectionally analyzed the data of 166 children and adolescents with HR who were being followed in 24 centers in Turkey. A nation-wide web based CEDD-NET Data System (http://cedd.saglik-network.org/) was used for data collection between December 2016 and April 2018. A form including clinical, genetic, laboratory and follow-up information of the patients was uploaded to the website and filled by the patient's doctors. Study approval was given by the Ankara University Ethics Committee (Approval number: 06-229-16).

The patients aged between 0 to18 years were included in to the study and patients with calciopenic rickets (related to vitamine D deficiency or hypocalcemia, vitamine D dependent rickets ect) were excluded.

The following data on the patients' admission clinical and laboratory characteristics were collected: birth weight, age at diagnosis, age of first symptoms, positive family history, the time of starting walk, height, weight, Height SDS, limb deformities (genu varum, genuu valgum, etc.) with incondyler and intermalleolar distance, other skeletal deformities (cranial, thoracal) and craniosynostosis, dental abscess, serum calcium, phosphorus, ALP, PTH, 25OHD3 levels, tubular phosphate reabsorption, Urinary Ca/Cr, radiological findings. The researchers were also asked to enter other clinical features that were not included in the questionnaire form to the system. ALP SD of patients were calculated according to reference data (9).

The questionnaire form also included the genetic test results of the patient. Genetic analysis of PHEX, and other genes causing HR were asked to enter to the system.

If there was a specific diagnosis causing hypophosphatemia like tubulopathy or McCune Albright syndrome, they were asked to indicate them.

The participating centers were asked to enter to the system if there were any other known pathological laboratory or clinical findings.

The researchers were also asked to enter to the system the treatment doses of phosphate and calcitiriol, and any other treatments. The yearly heights and improvement of deformities of patients who were given conventional treatment were recorded. The compliance of the treatment were evaluated by asking if there are missing of planned visit and/or not giving recommended dose of therapy by parents.

Follow-up patients were grouped as good responder (complete or significant improvement of deformities with either clinically and radiologically) or bad responder (no improvement or worsening of deformities).

The follow-up form also included complications (nephrocalcinosis, hyperparathyroidism, hypertension, dental abscess, cranial synostosis, enteshopathy etc.) developing during the follow-up. Entering additional information not included in the questionnaire form was optional.

Statistical analyses were performed by using SPSS for Windows V. 22.0 statistical software. Frequencies and percentages represented the descriptive statistics for categorical variables, and mean ± standard deviation values, median (min-max) when required were used for continuous variables. Student's t, chi-square, Fisher exact tests, repeated measures ANOVA for normally distributed continuous variables and Friedman ANOVA as nonparametric test were used. Post hoc multiple comparison test was also used.

Statistically significance was held as $p<0.05$.

Results

From 24 centers, 166 patients (98 females, 68 males; 18 pubertal, 148 prepubertal) were included in the study data. The mean age of diagnosis and was 5.1 ± 3.7 years. The mean time beginning of symptoms and start of walking were 1.89 ± 1.96 years and 15.5 ± 3.82 months respectively (Table 1). Almost half of patients (n: 80, 48.2%) had history affected at least one family member.

Clinical and Laboratory Characteristics on Admission:

The mean height SDS was -2.43 ± 1.35. The most frequent reported clinical findings were lower limb deformities (genu varum 80.1 % (n: 133), genu valgum 7.8% (n:13). Bone pain 16.8% (n:28), widening of wrist 31% (n:51), rachitic rosary/thoracal abnomalities 8.4% (n:14) and frontal bossing 7.2% (n:12). The late walking, lordosis, congenital hip dysplasia were among the reported findings. Seven (4.2%) asymptomatic patients were detected due to the positive family history. No craniosynostosis was reported in our patient cohort.

Laboratory features were consistent with HR with hypophosphatemia, normocalcemia, high ALP, normal/normal- high PTH, low TPR (Table 1). 25-OH Vitamin D levels were 35.97±15.61 ng/ml (n: 139). Low 25-OH Vitamin D levels (< 20 ng/dl) was detected in 27 patients. High dose vitamin D ingestion was also reported in 16 cases due to misdiagnosis of nutritional rickets before the admission to pediatric endocrinology clinics.

Etiology of HR:

Seven patients in our cohort had specific diagnosis from additional clinical and laboratory findings as cystinosis in three, tyrosinemia in two, Dent diseases with *CLCN5* mutation in one and McCune Albright syndrome in one patient.

Genetic analysis had been performed in 75 of 159 patients, and 65 of them showed a genetic mutation: *PHEX* mutation in 60 (80%), *DMP1* mutation in 3 (4%), *SLC34A3* mutation in 2 (2.6%) patients and no mutation had been detected in 10 patients who were screened for *PHEX* gene by sequencing (Figure 1).

Treatment and follow-up:

Almost all patients were treated with oral phosphate and vitamin D (calcitriol) supplements. Patients with *SLC34A3* mutation were treated only phosphate replacement. The mean follow-up period of the patients was 6.67± 2.3 years. First 3 years treatment response were evaluated for 91 patients which completed the 3 years follow-up. Although an increasing trend in serum phosphate and PTH levels were seen, no statistical significant differences from initial to 1st, 2nd and 3rd year of follow-up had been detected (p: 0563 and 0.796 respectively). A decrease in ALP and ALP SD were evident (Table 2). The height SDS were -2.38, -2.77, -2.72, -2.47 at initial, 1st, 2nd and 3rd year of treatment, respectively (p: 0.570).

In follow-up, 36% of 159 HR patients without specific etiology (such a cystinosis) showed complete or significant improvement in leg deformities. Improvement of leg deformities were evaluated by clinically and raiologically by each center. These patients had similar ages at the time of diagnosis (4.39 vs 5.3 years, p: 0,12), however, had better height SDS (-2,07 vs -2.57, p: 0.039), worse tubular phosphate resorption (70% vs 77%, p: 0.46), and worse ALP SDS at presentation (p:0.03). When we evaluate the following years, both groups had similar phosphate levels at presentation with better levels in 1st and 2nd years of treatment, even the treatment doses of phosphate were similar. However, significantly higher calcitriol treatment doses in 1st and 3rd years were found in improved group (Table 3 and Table 4).

Complications during follow-up:

When the complications of treatment were evaluated, 27 out of 159 patients (17 %) developed nephrocalcinosis (NC) on follow-up. The patients who developed NC showed no difference in biochemical differences at presentation and follow-up, however, their PTH levels at 3rd year was higher (145 vs. 78 pg/ml, p:0.002), and, they had higher treatment dose of phosphate and calcitriol ($p<0.05$) (Table 5 and Table 6).

Additionally, osteotomy was required in 15 subjects (9.4%), while dental abscess in 14 (8.4%) subjects, parathyroid hyperplasia developed in 4 subjects (2%), hypertension in 1 subject (<1%), and depression in 1 subject (<1%) among 159 cases.

Growth hormone therapy were given to 10 patients. Although duration and dose of treatment were variable, the patients treated with GH in addition to conventional therapy had similar height SDS before and after GH treatment (-3.47 and -3.3 respectively with delta height SDS of 0.173).

DISCUSSION:

Characteristics on Admission:

Here, we first described large series of HP patient with clinical, laboratory and follow-up and etiological characteristics in our country. The mean age of diagnosis was almost three years later from the mean time beginning of symptoms as being 5.1 years. In a Norvegian series, the age of diagnosis was 2.1 years (10). Early diagnosis is very important for treatment response and healing especially and, if the treatment started <1 year of age, height SDS become better (10,11).

Among the clinical findings, short stature and lower limb deformities were the most striking features. In our cases the mean height SDS was -2.41 at the admission. Disproportioned short stature is a definitive features of HR and is primarily related to the reduced growth of long bones and deformities (2,11). It was known that there is a great variability of height among individuals, and adults with XLH have a significantly reduced final height up to 20 cm (-1.9 height SDS) (12). Almost half of cases (80/166) had a HR diagnosis at least one individual in family. Despite positive family history, diagnosis was late in our series.

At diagnosis, one patient which diagnosed as Mc cune Albright syndrome, two patients with *PHEX* mutation, and two asymptomatic patients which were screened because of affected siblings have normal tubuler phosphate reabsorption. Also 3 patients have normal ALP levels despite hypophosphatemia and low TPR on admission. Patients characteristics, mistake for sampling, or some methodological problems could lead the normal laboratory results. During follow-up, for all of them precise diagnosis of HR were made.

Before diagnosis most of the cases were given therapeutic doses of vitamin D, assumed that they have calciopenic rickets. So the percentage of vitamin D deficiency (below 20 ng/dl) was 19.4% (27/139) in our series at the time of diagnosis.. Serum PTH levels were very helpful for diagnosis of HR. While calciopenic rickets showed increased level of PTH, patients with HR had normal or upper normal levels of PTH.

Etiology of HR:

Seven reported cases with HR had specific diagnosis with tubulopathy (cystinosis, Dent disease, tyrosinemia) or McCune Albright syndrome. Genetic analysis was performed in 75 of 159 cases. Results showed that the most frequent reason of HR is XLH with 80 % of patient had *PHEX* mutation. Interestingly, this frequency was similar to other reported series in literature which disease-causing genetic variants were identified (3,7,8,13). In fact, we were expecting higher frequency of autosomal recessive forms of HR in our population due to high consanguineous marriage rates, so far, *PHEX* mutation is the most prominent form of HR regardless of the population and consanguinity rates all over the word. PHEX protein is a member of the neutral endopeptidase family of zinc metalloproteinases and predominantly expressed in osteocytes and osteoblasts (3,5). It emolliates the inhibitory effect of "matrix extracellular phosphoglycoprotein" (MEPE) proteins on bone mineralization. PHEX composed a trimeric complex with dentin matrix protein -1 (DMP1) and α5β3-integrin, and then restricts FGF23 expression. While MEPE derived "acidic serine and aspartate rich motif" (ASARM) peptides inhibit these trimeric complex. So inactivating *PHEX* mutations lead to increase both ASARM peptides, and serum FGF23 levels (3, 14). All of these caused FGF23 related phosphaturia, hypophosphatemia, short stature, bone deformities as in our cases.
Loss of function mutation in *PHEX* causes X-linked dominant HR. Males to females ratio was reported as equal (5). In our cases with PHEX mutation male to femal ratio was 21/32 (0.65).
In our series, autosomal recessive HR type 1 was detected in three cases from one family which had *DMP1* mutation. Clinical and laboratory finding of those cases were reported previously (15) and which were similar those seen in patients with XLH. *DMP1* mutation cause to increased FGF23 production and clinical, laboratory and radiological findings could be expected to resembling those in XLH (3).
Two cases had *SLC34A3* mutations that lead to hereditary hypophosphatemic rickets with hypercalciuria. These disease is characterized by hypophosphatemia and rickets due to renal phosphorus wasting (3,8). Because of serum 1.25(OH)2D is high and FGF23 and PTH are reduced in those cases, secondary hypercalcemia and medullary calcinosis, also urolithiasis could be occurred (3,8).
In 10 cases which *PHEX* mutation were negative, *FGF23* had been also analyzed in 6 of them and found as negative. Other mutations leading HR and deletions and duplication which cannot be detected in sanger sequencing should be considered in cases with negative *PHEX* mutations.

Treatment results:

The conventional treatment of HR includes active vitamin D analogues and phosphate supplementations. The recommended doses of calcitirol 0.5-1.5 mcg/day or 20-30 ng/kg/day, and phosphorus 20-60 mg/kg/day (16,17). In our series the doses of calcitriol and phosporus were appropriate for recommendations but compliance of patients to treatment was the issue and which is almost always poor due to the frequent dosing of drugs (4-6 times of phosphate and 1-2 times of calcitriol) and bitter taste of phosphate solutions.. Optimal dose of treatment can varies from patient to patient. Usually higher doses were needed during rapid growth phases and at initiation of treatment (15).
After 3rd year of conventional treatment, patients showed no improvement of height SDS. This could be partly related to the late diagnosis of our patients.
It is known that 25 to 40% of the patients with HR who are close follow-up and compliant to treatment have growth retardation. Almost 2/3 of our HR cases do not show good response to conventional treatment. Although the earlier diagnosis and immediate treatment can be important prognostic factor on height improvement, we could not see any statistically differences between the groups for the age of diagnosis, when we separated the patients as good responder and bad responder.
The most striking feature was better height SDS in good responder than bad responder on presentation. This finding may indicate bad responder could be more severely affected than good responder in respect to bone pathology. Additionally, our cases with good responder and bad responder have similar serum Calcium and PTH levels. Good responders had higher serum phosphate levels despite having similar doses of P replacement, higher calcitriol doses were given during follow-up,
Our study show that conventional treatment could not lead an additional growth promoting effect in all HR patients, but this treatment seems to stop deteriorations of growth. The effects of conventional therapy on growth were reported in small cases. In a study, 13 cases showed height increment from -1.58 to -1.25 after two years conventional therapy. In untreated historical controls (n:16) height SDS was reported as -2.02±1.30 (18). In another study, cases treated with vitamin D and phosphate replacement showed improvement in lineer growth, as height SDS increased from -2.89 to -1.98 (19).
In general, bone deformities and abnormal growth of skeleton could not be completely heal despite early and optimal doses of phosphorus and calcitriol. In fact unsuccessful therapy of HR lead to search other treatment options. Growth hormone treatment is one of them. In our series, growth hormone therapy was given in 10 patients. But these patients showed similar height increment to patients whom were given only conventional treatment. Growth hormone treatment in HR were reported usually limited pilot studies, which suggest a beneficial effect. While one randomized study showed significant improvement of height SDS in eight severely short XLH children treated with rGH for 3 years, follow-up study showed that this treatment did not significantly increase adult height (20,21).
During follow-up, corrective osteotomy was required in 15 (9.4%) among the 159 subjects.
Development of progressive bone deformities in HR could lead to progressive gait disturbance, functional impairment, and severe arthritis. In such conditions surgical treatment could help improve patients quality of life (22). Each patient should be evaualted carrefully for requirment of corrective surgery.

Complications during follow-up:

The compliance of treatment is a difficult issue in HR since phosphate needs 4-6 daily doses a calcitriol needs 1-2 daily doses.
During conventional treatment of HR, several complications can be occured such as nephrocalcinosis, dental abscesses, enteshopathy, craniosynostosis etc. Among these, nephrocalcinosis is more frequently reported (1-4,17).
We detected 17 % of all patients with hereditary hypophosphatemia had nephrocalcinosis, as being relatively low when compared to other series. Nephrocalcinosis has been reported in 22% to 100% of XLH patients who are under conventional

therapy (1-4). But those were small series and diagnosis was earlier than ours. So far, we speculated that earlier therapy would lead NC more frequently.

Nephrocalcinosis usually developed after conventional treatment of HR, and accepted as a treatment complication. It is known that higher doses of phosphate replacement caused to NC and hyperparathyroidism. Higher doses of phosphate replacement may increase of FGF23, and posphaturia will be increased (13). Similarly, our cases with nephrocalcinosis were treated with higher doses of phosphate than cases without nephrocalcinosis. In addition to higher dose of phosphate replacement and calcitriol may lead to hypercalcemia and have additional effect on nephrocalcinosis. In our cases higher doses of calcitriol at initial therapy seems to have an additional effect on NC development. The reason for high dose therapy were not recorded in all cases. However, some physicians might have thought an insufficient response to the treatment and aimed at the serum phosphate levels to keep in the reference values. In fact, the primary aim in conventional HR therapy should keep not serum P but Ca, ALP, PTH, and urinary Ca excretion in normal limits.

Although higher doses of phosphate and calcitriol were given, in patients with nephrocalcinosis growth was not affected. While dental abscess were recognized in 14 cases, the other complications were infrequently reported, as parathyroid hyperplasia in 4 subjects (2%), hypertension in 1 subject (<1%), and no neurological complications including craniosynostosis. The awareness about complications of HR should be improved among medical profesionals.

Conventional treatment is aimed to compensate for renal phosphate wasting and to counter 1,25 OHD deficiency (4), which is commonly related to excess level of FGF23 in HR. Nowadays, some strategies that manipulate FGF23 signaling have been described. Burosumab, a monoclonal antibody directed at FGF23, is one of them (23). In children with HR, treatment with burosumab had been reported to improve renal tubular phosphate reabsorbsion, serum phosphore levels, linear growth, and severity of rickets (24). Burosumab is expensive, and long-term outcomes are not yet available.

Conventional therapy is still the first line therapy for hypophosphatemic rickets. When patients were correctly treated with conventional therapy, NC would be lower, and important part of them have good response.

There are some limitations of study. Despite all data of cases wanted to enter to the web system, some findings could have not been reported. Because the radiological findings were evaluated in each center, follow-up characteristics were mainly based to healing of skeletal deformities. Unfortunately genetic diagnosis could not be done in all cases. Almost half of all cases could be analysed, and definitive PHEX mutation found 80% of genetically analysed but 36% of all cases.

As conclusion; Age of diagnosis of HR was late in our series despite having positive family history, and the most frequent reason is *PHEX* mutation. HR treatment and follow-up is challenging and our results showed higher treatment doses leading NC without any change in serum levels, suggesting given higher doses lead higher phoshaturia probably through the stimulation FGF23. However, higher calcitriol doses could improve bone deformities.

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Table 1: Clinical and laboratory characteristics of all cases on admission

Parameter	Mean±SD	Median (min-max)
Age of presentation*	5.1±3.7 years	3,6 (0,12-16)
Age of first symptoms (years)	1.89±1.96	1.8 (0.5-9)
Height SDS	-2.43 ±1.35	-2.6 (-5.52-1.18)
BMI*	17.7±2.62	17.6 (11.3-28)
Calcium (mg/dl)	9,5±0.46	9,6 (8.2-10.9)
Phosphate (mg/dl)*	2,51±0.49	2,5 (1.1-5)
ALP (U/L)*	738±481	607 (143-3200)
ALP SD*	1,08±2.73	0.48 (-3.9- 12.5)
Tubuler phosphate reabsorption (%)*	73.2±16.2	79 (16-98)
PTH (pg/ml)*	66.8±45.37	57.1 (2.3-254)

Table 2: Laboratory evaluation of 91 cases for 3 years follow-up

N: 91	On Admission	1. year	2. year	3. year	P value
Calcium (mg/dl)	9,48±0.51 9.5 (8.2-10.9)	9,45±0,51 9.4 (7.4-10.3)	9,56±0,49 9.6 (8.4-10.7)	9,58±0,46 9.6 (8-10.9)	0,062
Phosphate (mg/dl)*	2,58±0,55 2.5 (1.1-5)	2,76±0,62 2.7 (1.53-5.54)	2,83±0,71 2.7 (1.6-5.4)	2,85±0,73 2.83 (1.69-4.7)	0.563
ALP (U/L)*	786±522 624 (45-3200)	627±449 510 (173-2957)	561±319 450 (169-1650)	546±327 458 (142-25349	<0.001
ALP SD*	1.08±2.73 0.80 (-3.9-12.5)	0.33±2,47 -0.04 (-4.1-8.8)	0.12 ± 2 -0.4 (-3.3-5.1)	0.03±2.09 -0.19 (-3.9-5.99)	<0.001
PTH (pg/ml)*	68,4±48 60 (5-254)	85,9±77 75.6 (4.6-537)	80,68±65.9 66.5 (3.4-419)	93,15±99 64.4 (5.2-574)	0.796

Table 3: Laboratory characteristics of good responder and bad responder groups for 3 years follow-up

Table 4: Doses of treatment and growth characteristics of good responder and bad responder groups for 3 years follow-up

	Phosphate level (mg/dl)			**Calcium level (mg/dl)**			**ALP SDS**			**PTH level**		
	Good responder	**Bad responder**	***P values***	**Good responder**	**Bad responder**	***P values***	**Good responder**	**Bad responder**	***P values***	**Good responder**	**Bad responder**	***P values***
At Diagnosis	**2,53±0.52**	**2,6±0,58**	*0,24*	**9,52±0.56**	**9,46±0,47**	***0,58***	**2.1±3.81**	**0.95±2,02**	*0,01*	**68.35±41.5**	**68,6±44**	*0,47*
1. Year	**2,95±0.79**	**2,63±0,46**	*0,01*	**9,4±0,55**	**9,48±0,48**	***0,52***	**1.1±3,06**	**-0,16±1,86**	*0,009*	**91±101**	**82,35±58,5**	*0,38*
2. Year	**3,02±0.85**	**2,72±0,58**	***0,044***	**9,54±0,51**	**9,57±0,47**	***0,73***	**0.80±2,3**	**-0.3±1,67**	*0,014*	**73.3±50.2**	**84,79±73,3**	*0,18*
3. Year	**2,94±0,77**	**2,73±0,61**	*0,08*	**9,63±0,51**	**9,55±0,43**	***0,45***	**0.35±2,46**	**-0,14±1,85**	*0,28*	**109.6±139**	**83,66±65,8**	*0,12*

	Phosphate dose (mg/kg)			**Calcitriol dose (ng/kg)**			**Height SDS**		
	Good responder	**Bad responder**	***P values***	**Good responder**	**Bad responder**	***P values***	**Good responder**	**Bad responder**	***P values***

At Diagnosis	**61,6±42,7**	**60,8±38,4**	***0,765***	**34,33±24,6**	**26,9±12,2**	***0,129***	**-2,07±1.01**	**-2,61±1,4**	***0,039***
1. Year	**66,25±38,14**	**62,1±42,2**	***0,268***	**33,43±19,1**	**24,9±13,22**	***0,031***	**-2,07±0.87**	**-2,67±1,37**	***0,021***
2. Year	**58,72±28,11**	**56,5±33,7**	***0,298***	**29,21±14**	**26,3±11,62**	***0,366***	**-2,07±0.86**	**-2,71±1,25**	***0,012***
3. Year	**66,34±31,8**	**54,69±32,3**	***0,079***	**26,9±10,61**	**22,06±11,3**	***0,04***	**-1,94±0.89**	**-2,8±1,28**	***0,002***

Table 5: **Laboratory characteristics of patients according to development of nephrocalcinosis**

Table 6: Treatment characteristics of patients according to development of nephrocalcinosis

	Phosphate level (mg/dl)			Calcium level (mg/dl)			ALP SDS			PTH level		
	NC (+)	NC (-)	*P*	NC (+)	NC (-)	*P*	NC (+)	NC (-)	*P*	NC (+)	NC (-)	*P*
At diagnosis	2,54±0,76	2,59±0,52	*0,209*	9,44±0,49	9,5±0,51	*0,25*	1,88±2,6	1,28±2,9	*0,29*	50,5±24,8	72,7±50,8	*0,06*
1. Year	2,95±0.82	2,71±0,56	*0,114*	9,32±0,65	9,48±0,46	*0,11*	0,44±1,74	0,31±2,6	*0,4*	114,7±154	79,57±48	*0,069*
2. Year	2,99±0,97	2,81±0,64	*0,24*	9,54±9,46	9,56±0,5	*0,42*	0,13±1,7	0,11±2,08	*0,44*	85,6±102	79,6±56	*0,36*
3. Year	2,92±0,85	2,82±0,64	*0,36*	9,5±0,41	9,6±0,47	*0,21*	0,31±1,94	-0,02±2,1	*0,37*	154,5±171	78,28±66	*0,002*

	Phosphate dose (mg/kg)	Calcitriol dose (ng/kg)

	NC (+)	NC (-)	*P values*	NC (+)	NC (-)	*P values*
Begining of treatment	89.9±46.4	55,95±32.4	*0,003*	62,37±26.7	27,92±11.5	*0,006*
1. Year	74,31±37.1	60,61±42.7	*0,13*	34,9±12.4	26,4±10.5	*0,04*
2. Year	71,93±38.3	54,69±28.8	*0,033*	26,43±11.8	27,17±12.6	*0,48*
3. Year	69,4±29.7	56,76±33.3	*0,096*	18,66±10.4	25,16±11.8	*0,035*

Figure 1: The results of genetic analysis of patients