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NEW DATA – BASED INCLUDING ON ADDITIONAL MODERN RELATED BIOLOGIC PARAMETERS – REGARDING POSSIBLE RELATIONSHIPS BETWEEN BONES' METABOLIC STATUS AND FUNCTIONAL IMPAIRMENTS IN CHILDREN WITH CEREBRAL PALSY

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This study aimed, using additional biologic parameters, to obtain new data regarding possible changes in bone metabolism, mainly, considering the severe impairments of mobility in some cases of children with cerebral palsy. We analyzed 34 patients, aged between 4 and 11 years, divided into 2 main groups (cerebral palsy *vs*. scoliosis), the first, sub-divided into Ambulant/Non-Ambulant. The evaluating parameters used were: FIM scale, BMI, serum TSH, FT4, urinary calcium and phosphorus concentrations (24 hours urine collection); in addition, some newly accessed: Flanagan's QoL, PTH, 25-OH Vitamin D, Beta CrossLaps and Osteocalcin, serum values. There were no significant differences ($p > 0.05$) between all the groups/sub-groups, except for FIM and QoL. Cerebral palsy, including non-ambulatory, and scoliosis showed similar values for PTH, 25-OH Vitamin D and TSH; additionaly cerebral palsy group was similar to scoliosis group. In order to evaluate the parameters' contributivity and correlations, first PCA, then Spearman rank correlation coefficient matrix – Spearman's rho – were used. The values were close to 0 (*i.e.* showing non-correlated items) except for FIM and QoL, Osteocalcin and Beta CrossLaps, FIM and Osteocalcin. Processing of the available data, taking into account the coefficients above, indicate very few significant/highly significant correlations among parameters. Namely, values of Beta CrossLaps apparently correlate with PTH and FT4, which result should be confirmed by a larger sample study.

Key words: cerebral palsy, quality of life, biologic parameters.

INTRODUCTION

This study represents the continuation of a research on possible relationships between bones' metabolic status and functional impairments in children with cerebral palsy, using new biological parameters (PTH, 25-OH Vitamin D, Beta CrossLaps and Osteocalcin, serum values) and Visual Analogue Scale and Flanagan's Quality of Life Scale.

The term cerebral palsy represents a socio medical framework for children with special needs who have motor disorders[1]. It does not describe a single disease, but rather a multitude of disorders with different etiologies[1]. Sigmund Freud considered that cerebral palsy results from abnormal fetal development, before the concept to be accepted by the medical world (cited in Hotwani K, Sharma K).[2] The majority of cerebral palsies occur during prenatal, perinatal or early postnatal period.[2, 3]

The signs and symptoms are different depending on the child's maturation[3]. Currently, cerebral palsy is defined as a group of disorders of motor function and posture. It is permanent and is caused by abnormalities in the developing/immature brain or by non-progressive damage of it[1]. In addition to motor disorders, in the cerebral palsy we often meet disturbances of sensation, perception, cognition, communication and behavior, and secondary

muscle-skeletal problems[4]. The cerebral palsy in children often includes the risk of low bone mass[5].

The multitude of symptoms associated with cerebral palsy often causes social isolation, embarrassment both to the patient and the caregivers. The limitation of the independence followed by social isolation can cause depression. The presence of pain can also be a significant factor in psychological stress. The functional limitations are significantly correlated with the severity of cerebral palsy. Therefore, the functional status investigations should focus on mobility, self-care and daily activities.[6]

The severity of impairment generated by cerebral palsy has implications for the child at the play and action level, at social relationships with other children or adults. The data related to the quality of life of children with cerebral palsy and of their families has lead to better practice and improved therapeutic strategies.[7]

The quality of life refers to what offers safety and comfort for patients including with cerebral palsy.[8] Some studies that evaluated quality of life in children with cerebral palsy showed, somewhat surprising, that patients aged 8 to 12 years had a vision on the quality of their lives just as good as children who had no cerebral palsy.[1] In their families, the caregivers of children with cerebral palsy are facing many practical and emotional challenges.[9]

Our aim was to evaluate the possible changes in bone metabolism, mainly, considering the severe impairments of mobility in some cases of children with cerebral palsy.

MATERIAL AND METHODS

The study was conducted at National Clinic Center for Neuropsychomotor Rehabilitation in Children ”Dr. Nicolae Robănescu”, and included 34 patients, aged between 4 and 11 years, divided into 2 groups (the first, sub-divided into 2 sub-groups):

– 13 ambulant patients with cerebral palsy (group A in what follows),

– 15 non-ambulant (NA group) patients with cerebral palsy

– 6 patients with scoliosis (Sc) – apparently without changes in the indicators of bone metabolism and functionality regarding ambulation (group Sc).

The evaluating parameters used were the “classical” ones: Functional Independence Measure (FIM) scale, Body Mass Index (BMI), serum Thyroid Stimulating Hormone (TSH), Free Thyroxin (FT4), urinary calcium and phosphorus concentrations (24 hours urine collection); and some newly accessed: Visual Analogue Scale (VAS, *i.e.* the four-point categorical verbal rating scale – VRS), (Flanagan’s) Quality of Life Scale (QoL), Parathyroid hormone (PTH), 25-OH Vitamin D, Beta CrossLaps and Osteocalcin, serum values.

PTH is secreted by the parathyroid glands and has a polypeptide structure[10] with a complex actions on bone, kidney and intestinal level. By its effects on 1.25 $(OH)_2$ D synthesis, leads to increased concentration of calcium.[10,11]. The primary function of PTH is to maintain ionic calcium concentration in the extracellular fluid[11] increasing osteoclastic activity in bone.[12]

In addition, PTH stimulates the mineral bone resorption and increase the flux of the calcium from the bones to blood, decrease renal clearance of calcium and increase efficiency of calcium absorption in the intestine.[11]

The tendency to hypocalcaemia (given by an insufficient calcium intake) is compensated by an increased secretion of PTH[11]. The excessive action of the PTH on renal reabsorption of phosphorus leads to lower phosphorus in the blood.[11]

In the liver, the Vitamin D undergoes the first hydroxylation in order to form 25-OH Vitamin D (calcidiol). This is a biologically metabolite with a limited activity. 25-OH Vitamin D is the most relevant indicator of overall Vitamin D status; its mild to moderate deficiency can cause osteoporosis and secondary hyperparathyroidism.[13]

The low level of 25-OH Vitamin D can be caused by: inadequate food intake, deficiency of Vitamin D absorption in the intestine, insufficient exposure to sunlight, anticonvulsant drugs. The use of medicines that antagonize the action of Vitamin D lead also to resistance towards its.[11]

Vitamin D and 25-OH Vitamin D serum levels may suffer seasonal variations (in winter are lower than in the summer because of reduced exposure to UV radiation and food intake).[11]

Osteocalcin is the major non-collagenous protein of the bone, produced only by osteoblasts; it is a marker of bone formation and is involved in its mineralization. It is specific to bone and dentin, and

for its synthesis are required the presence of Vitamin D 3 and Vitamin K. The most of it (80%) is a component of the bone matrix.[14]

Increased values of Osteocalcin in adult appear in hyperparathyroidism, fractures, and prolonged bed rest. Decreased values for adult are met in hypoparathyroidism.[14]

In Romania there are no related references values in children.

Beta CrossLaps is a specific marker of bone resorption which appears in the early stages of type I collagen degradation. Its dosage is recommended in osteoporosis and for predicting fracture risk.[15]

There are no references values available for Romanian children.

In foreign literature, there have not been established the normal values for people under 18 years, but increased serum concentration levels are reported in adults with increased bone resorption.[16]

About FIM scale we have written elsewere.[17]

VAS (*i.e.* VRS), commonly used in clinical practice, allows for classification of pain, as:

– None VRS = 0,
– Mild VRS 1–3,
– Moderate VRS 4–6,
– Severe VRS 7–10.[18, 19]

Flanagan's QoL measurement for the patients with cerebral palsy gives us data to determine the impact of the health care when cure is not possible.[20]

Initially the scale had 15 items but it was added a new item "Independence, doing for yourself" after a qualitative study which indicated validity in chronic illness groups.[21]

The adapted scale has 16 items and 6 domains as follows: "physical and material well-being; relationships with other people; social, community, and civic activities; personal development and fulfillment; recreation; independence".[20]

The Board of Ethics approved this study (467/ January 18, 2016).

RESULTS AND DISCUSSION

We applied Principal Component Analysis (PCA)[22] procedure taking into account data from all patients, on the variables: FIM, QoL, BMI, (serum values of) TSH, FT4, PTH, 25-OH Vitamin D, Osteocalcin, Beta CrossLaps and respectively, urinary calcium and phosphorus, concentrations.
(VRS was eliminated because being a scale encompassing very few items, within statistical analysis, the data it produced could not be assimilated to thorough quantitative information.)
PCA identified 3 main components which were responsible for 58.31% of data variability (Tables 1 and 2).

In Table 2 are given details of these three components.

Table 1

Main components responsible for data distribution

Total Variance Explained

Component	Initial Eigenvalues			Extraction Sums of Squared Loadings		
	Total	% of Variance	Cumulative %	Total	% of Variance	Cumulative %
1	3.248	29.525	29.525	3.248	29.525	29.525
2	1.908	17.344	46.869	1.908	17.344	46.869
3	1.258	11.440	58.309	1.258	11.440	58.309
4	.993	9.026	67.335			
5	.901	8.189	75.524			
6	.787	7.158	82.682			
7	.580	5.277	87.959			
8	.515	4.686	92.645			
9	.322	2.931	95.576			
10	.305	2.773	98.349			
11	.182	1.651	100.000			

Extraction Method: Principal Component Analysis.

Table 2

Extracted components

Component Matrix[a]

	Component		
	1	2	3
BMI	.516	.299	-.048
FIM	.597	.494	-.257
TSH	.293	-.035	.545
F T 4	-.577	.494	-.082
Urinary Ca	.470	.336	.429
Urinary Ph	.556	.116	**.643**
PTH	.636	-.425	.021
25-OH Vit.D	-.159	.582	.034
Osteocalcin	**.818**	-.016	-.392
Beta crosslaps	.658	-.416	-.290
QoL	.382	**.714**	-.220

Extraction Method: Principal Component Analysis.

a. 3 components extracted.

As it can be seen in Table 2, the three main components could be represented by serum Osteocalcin, QoL and respectively, urinary phosphorus.

The data can be used to represent the 11 variables through dots in principal plane, as in:

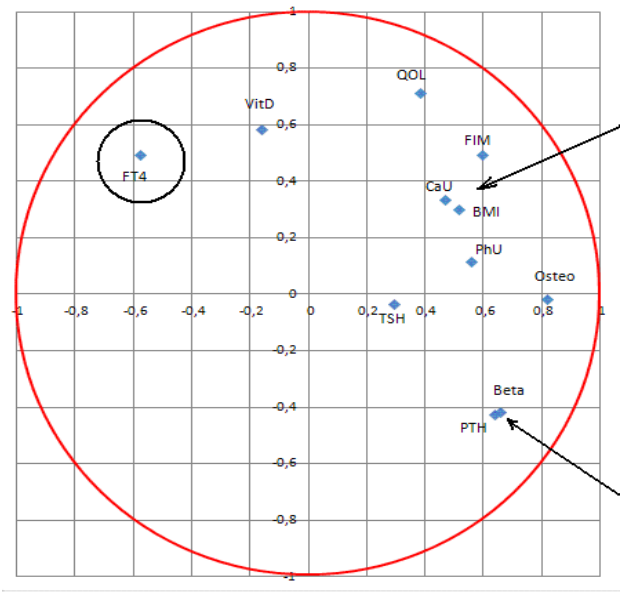

Figure 1. Variables in principal axes plane (component 2 *vs.* component 1).

In the figure above, it can be noticed an apparently very strong correlation between PTH and Beta CrossLaps (apparently redundant?). However, to state that they are "correlated" we should take into account also the third component, and this one is not confirming this! It can be noticed, in the Figure 1 above, an apparently strong enough correlation between urinary calcium and BMI (this is also unconfirmed by the third component!) and also an independence of FT4 of the other variables.

PCA produce also the scores of each patient in these three components, which will help us perform a diagram to "see" how all three groups are overlapping.

Table 3

Scores of patients in the three principal components

No.	group	Comp 1	Comp 2	Comp 3
1	A	0.192	0.518	1.014
2	A	–0.431	0.023	1.151
3	NA	–1.337	1.212	1.102
4	NA	0.006	0.491	–0.150
5	NA	–0.034	0.897	–0.576
6	A	–0.808	–0.752	0.306
7	NA	–0.538	–0.867	1.842
8	Sc	3.410	–0.630	0.738
9	NA	0.257	–2.455	–0.578
10	Sc	–0.038	0.309	1.288
11	NA	0.450	–0.868	1.072
12	A	2.435	0.906	0.790
13	NA	–0.207	–0.110	1.385
14	NA	0.693	–1.238	–2.106
15	Sc	0.911	0.621	–0.830
16	NA	–1.988	–0.078	1.433
17	NA	–0.878	–1.715	–0.469
18	Sc	–0.962	0.502	–2.006
19	A	–0.026	0.704	–0.073
20	NA	–0.760	–1.712	1.022
21	A	0.854	0.917	0.974
22	A	–0.043	–1.049	–0.655
23	A	–0.791	0.253	–1.147
24	Sc	–1.072	0.490	–0.737
25	A	0.370	–0.176	–1.055
26	A	0.618	–0.992	–0.728
27	Sc	–0.235	2.612	–0.698
28	NA	–0.244	0.656	–0.147
29	NA	0.518	–0.119	0.101
30	NA	–0.404	0.381	–0.078
31	A	0.223	1.055	–0.306
32	A	–0.763	–0.241	–0.468
33	A	0.661	0.614	–0.441
34	NA	–0.056	–0.178	–0.988

A = ambulant,
NA = non-ambulant,
Sc = scoliosis.

The representation above does not show an obvious, sensitive separation, between groups. To study potential correlations between variables,

besides the charts obtained using the multivariate PCA method, we could use a bivariate method: the calculation of a correlation coefficient. We choose the Spearman one because we are not sure of the (quasi)normality of the data.

First, considering all our 34 patients, the Spearman correlation coefficients[23] are given in the following table.

The coefficients which are star-marked, respectively double star-marked in this table indicate strong, significant respectively highly significant correlations. There are 5 significant and 3 highly significant.

In general, the rest of the values in the table above are close to 0 (*i.e.* showing non-correlated items) except for a few. Namely, the highly significant are as follows:

1) FIM *versus* QoL (rho = 0.601, $p < 0.001$),

2) Osteocalcin *versus* Beta CrossLaps (rho = 0.61, $p < 0.001$), and

3) FIM *versus* Osteocalcin (rho = 0.505, $p = 0.002$).

Table 4

Correlations between variables (and their significances)

Correlations

			BMI	FIM	TSH	F T 4	Urinary Ca	Urinary Ph	PTH	25-OH Vit.D	Osteocalcin	Beta crosslaps	QoL
Spearman's rho	BMI	Correlation Coefficient	1,000	,174	,107	,011	,050	,193	,093	,182	,356*	,223	,239
		Sig. (2-tailed)	.	,324	,546	,950	,778	,275	,599	,302	,039	,205	,173
	FIM	Correlation Coefficient	,174	1,000	,105	-,056	,263	,120	,096	-,082	,505**	,098	,601**
		Sig. (2-tailed)	,324	.	,555	,753	,133	,500	,591	,643	,002	,583	,000
	TSH	Correlation Coefficient	,107	,105	1,000	-,102	,116	,245	,238	,023	,105	,166	-,113
		Sig. (2-tailed)	,546	,555	.	,565	,515	,163	,175	,896	,555	,348	,526
	F T 4	Correlation Coefficient	,011	-,056	-,102	1,000	-,112	-,186	-,279	,276	-,276	-,405*	,123
		Sig. (2-tailed)	,950	,753	,565	.	,528	,293	,110	,114	,115	,018	,487
	Urinary Ca	Correlation Coefficient	,050	,263	,116	-,112	1,000	,379*	,015	,050	,104	,238	,223
		Sig. (2-tailed)	,778	,133	,515	,528	.	,027	,935	,780	,559	,176	,205
	Urinary Ph	Correlation Coefficient	,193	,120	,245	-,186	,379*	1,000	,155	,030	,081	,127	,104
		Sig. (2-tailed)	,275	,500	,163	,293	,027	.	,381	,867	,650	,474	,560
	PTH	Correlation Coefficient	,093	,096	,238	-,279	,015	,155	1,000	-,097	,365*	,411*	-,144
		Sig. (2-tailed)	,599	,591	,175	,110	,935	,381	.	,587	,034	,016	,418
	25-OH Vit.D	Correlation Coefficient	,182	-,082	,023	,276	,050	,030	-,097	1,000	-,036	-,303	,064
		Sig. (2-tailed)	,302	,643	,896	,114	,780	,867	,587	.	,839	,081	,720
	Osteocalcin	Correlation Coefficient	,356*	,505**	,105	-,276	,104	,081	,365*	-,036	1,000	,584**	,250
		Sig. (2-tailed)	,039	,002	,555	,115	,559	,650	,034	,839	.	,000	,154
	Beta crosslaps	Correlation Coefficient	,223	,098	,166	-,405*	,238	,127	,411*	-,303	,584**	1,000	-,094
		Sig. (2-tailed)	,205	,583	,348	,018	,176	,474	,016	,081	,000	.	,596
	QoL	Correlation Coefficient	,239	,601**	-,113	,123	,223	,104	-,144	,064	,250	-,094	1,000
		Sig. (2-tailed)	,173	,000	,526	,487	,205	,560	,418	,720	,154	,596	.

*. Correlation is significant at the 0.05 level (2-tailed).

**. Correlation is significant at the 0.01 level (2-tailed).

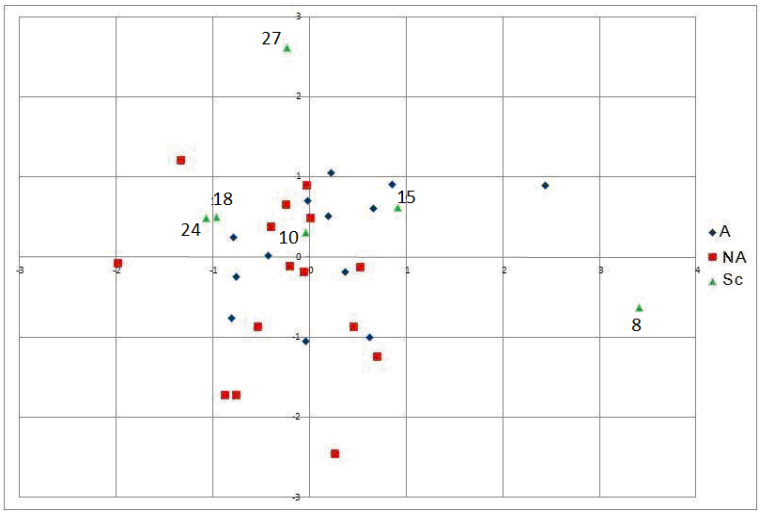

Figure 2. Representing patients with dots in the principal plane (indicating the group by colors).
(In the diagram each patient with scoliosis is identified by its number).

Additionally, serum values of Beta CrossLaps are significantly correlated with FT4 and PTH. Regarding the relations between the three groups (identified by A, NA and Sc) of patients, taking into account all the 11 variables above, and given the small number of patients in each group, the calculation of the correlation coefficients gives more plausible results than the PCA. Only non-parametric methods been available: Kruskal-Wallis, median tests (for the 3 groups A, NA, Sc) and Mann-Whitney U test (when only 2 groups: A+NA, Sc were involved). These methods show that, in general, there is no significant difference ($p > 0.05$) between the groups; the only exceptions were:

1) in the case of FIM (A *versus* NA, $p < 0.001$; A+NA *versus* Sc, $p = 0.001$; and NA *versus* Sc, $p < 0.001$), and

2) in the case of QoL (A+NA *versus* Sc, $p = 0.006$; NA *versus* Sc, $p = 0.005$).

In addition, A+NA and Sc showed similar (serum) values for PTH ($p > 0.95$), for 25-OH Vitamin D ($p > 0.99$) and for TSH ($p > 0.98$).

Study limitation: because most of the cerebral palsy non-ambulant children in our study needed indwelling catheter for prelevating a 24 hours urine sample, the parents/caregivers refused to enroll in the study in an amount of about 40%.

CONCLUSIONS

All the data collected and processed by now indicate very few significant strong correlations between the considered variables: serum values of Beta CrossLaps correlate with the ones of PTH, FT4 and Osteocalcin. Are they are redundant?

On the other hand, the groups appear to be similar for all variables, except for FIM and QoL (which is to be expected), confirming some of our previous conclusions. [17]

Because of small number of patients, it is objectively difficult at this stage to assess the normality of population's data for the evaluated groups; in this respect, a larger number of patients is needed to obtain stronger related evidences – including through the possibility of using parametric tests and more statistical methods.

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