„CAROL DAVILA" UNIVERSITY OF MEDICINE AND PHARMACY BUCHAREST

DOCTORAL SCHOOL

FIELD OF MEDICINE

C Peptide – Evolution and Clinical Correlations in Type 1 Diabetes Mellitus in Children and Teenagers

SUMMARY OF THE DOCTORATE THESIS

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Part I. Current Stage of Knowledge

Diabetes mellitus (DM) represents a major global public health problem, currently reaching alarming growth rates. At present, there are more than half a billion people living with this disease worldwide [0].

Type 1 DM is the most common form of DM in children and adolescents. This pathology occurs due to an absolute or significant insulin deficiency caused by the autoimmune destruction of β-pancreatic cells. The etiology has multiple factors, including genetic susceptibility and various environmental factors [2].

The number of children and adolescents suffering from DM increases alarmingly every year. In 2021, more than 1.2 million children and adolescents suffered from type 1 DM.

In Romania, according to the Romanian Childhood Diabetes Registry, in 2022 the incidence of type 1 DM among children and adolescents is of 13.8/100,000/year, increasing in the past few years. In 2019, the data report an incidence of 11.07/100,000/year, in 2021: 13.9/100,000/year.

In recent years, there has been increased interest in reexamining the natural history of pancreatic β-cell function. The subject of preserving the residual function of the β-pancreatic cells for as long as possible, of researching the factors that can influence the occurrence of partial remission and its extension seemed challenging to me, which is why I chose as a research topic the research of the clinical and biological parameters that set or not the "imprint", in maintaining endogenous insulin secretion. The residual function of pancreatic β-cells can be evaluated by measuring the level of C peptide C (CP), currently considered the most important marker for assessing endogenous insulin secretion in insulin-treated type 1 DM patients.

This research is structured in 3 parts:

1. A first study (l) was conducted on a group of 215 patients, in which I followed for 3 years the decline rate of serum CP and the correlations between it and various parameters from the time of diagnosis, aiming at identifying those parameters that influence the preservation or, on the contrary, the acceleration of the decline of the residual β-cellular secretory function.

2. The second study (II) included 291 patients in whom I analyzed both retrospectively and prospectively different clinical and biological factors both from the moment of onset and in evolution, in relation to CP secretion for at least 5 years.

3. The third study (III) was carried out on the same group of 291 patients and aimed at investigating the period of partial remission and at identifying the factors that could influence the occurrence and/or duration of this phase in the evolution of type 1 DM, in relation to the endogenous secretion of insulin quantified by serum CP dosing.

C Peptide ("*connecting"* peptide) is a polypeptide with the molecular formula $C_{112}H_{179}N_{35}O_{46}$, made up of 31 amino acids, which connects the A and B chains of insulin from the proinsulin molecule (insulin precursor). C Peptide was discovered in 1967, while researching on the process of insulin synthesis, by Steiner [0].

C-peptide (CP) has been known for several decades. However, over time there has been a lack of interest in the study of CP. Only in recent years has CP been accepted as a relevant outcome in studies aiming at preserving pancreatic β-cell function. Also, the persistence of CP correlates with a lower frequency of long-term complications in type 1 DM.

Experimental studies as well as recent clinical studies prove that CP is a biologically active product. Research has shown that CP binds specifically, with high affinity, to the membrane of certain cells (neuronal, endothelial, renal tubular, fibroblasts) through a G protein-coupled surface receptor, which causes the activation of intracellular Ca-dependent signaling pathways. Thus, CP seems to have a therapeutic potential in preventing some of the long-term complications of diabetes [132].

There is still no optimal standardization with regard to the measurement of CP, and for this reason it is necessary to carefully interpret its values, depending on the method of determination used. CP can be measured à jeun (fasting)/basally or stimulated, by several methods, in plasma or urine [152].

Currently, there are modern, ultrasensitive CP dosage methods that can detect extremely low values, down to 0.0015–0.0025 nmol/L [159].

Due to the autoimmune destruction of β-cells, in the first years, there is a progressive decrease in the number of insulin-producing cells in the natural evolution of type 1 DM. Clinical signs of type 1 DM appear when the mass of the pancreatic β-cells has decreased by approximately 70–80% [193].

Shortly after the onset, in the first year after the diagnosis, many patients go through a so-called "honeymoon", namely a transitory period of remission, in which the β-pancreatic

cells still secrete insulin. The most important factors that negatively influence the rate of partial remission in newly diagnosed patients with Type 1 DM are the antecedents of acute infection, the presence of ketoacidosis and young age at onset.

Based on the new staging of Type 1 DM, different immunotherapies have been developed to modify the evolution of Type 1 DM. Such interventions aim at stopping the autoimmune process and thus at preventing or delaying the onset of the symptomatic disease. Suppression of the autoimmune process would protect the residual pancreatic β-cell mass [248]. Teplizumab and ATG in a low dose could be successfully used to stop the autoimmune attack on pancreatic β-cells. However, after stopping the treatment, the remission disappears, making the immunomodulatory intervention necessary for an indefinite term [256].

Part II. Personal Contributions

C Peptide (CP) dosage is the method by which the residual function of pancreatic β-cells can be quantified. The DCCT (Diabetes Control and Complication Trial) reported that a stimulated CP value > 0.2 nmol/L (> 0.6 ng/ mL) is associated with a lower frequency of retinopathy, nephropathy and hypoglycemia [185,189].

The purpose of this study is to investigate the correlations that may exist between the value of serum CP and various clinical and laboratory parameters, as well as the clinical evolution and the identification of factors that can be considered predictive for the preservation of residual β-pancreatic function in children and adolescents with Type 1 DM registered in the records of the Diabetes Department of the Emergency Clinical Hospital for Children "M.S.Curie", Bucharest.

7. Study I - Analysis of factors influencing the evolution of C peptide in the first 3 years since the onset of type 1 DM

7.1. Working hypothesis and specific objectives

This study aims at investigating the correlations between serum CP levels and different clinical and laboratory factors in children and adolescents with type 1 DM during the first 3 years after the diagnosis.

7.2. Methodology of Research

A number of 215 patients who met the inclusion criteria (102 girls and 113 boys), aged between 0-15, were included in the study.

At the time of diagnosis, I assessed: the age, the onset manner in terms of symptom severity (without DKA (diabetic ketoacidosis), with mild DKA, with a moderate DKA or severe DKA), the association or not of an acute infectious disease, the value of serum CP a jeun, Hb A1c. DKA patients were divided into 3 groups according to the degree of acidosis: mild DKA (pH = 7.29-7.20), moderate DKA (pH = 7.19-7.11), severe DKA (pH ≤ 7.10). I followed the evolution of the CP decline rate in the first 3 years after diagnosis by determining the fasting CP (a jeun), annually, for 3 years.

Patients were thus divided into two groups according to CP value after 3 years of diabetes evolution: group 1 with low CP < 0.6 ng/mL, and group 2 with preserved insulin reserve, according to DCCT, where CP was ≥ 0.6 ng/ml.

7.3. Results and discussions

In the cohort of 215 children diagnosed with type 1 DM at a young age (average age 7.33±3.73 years old), I analyzed the factors that can influence the rate of CP decline in the first 3 years after the clinical onset of diabetes.

Table 7.1. The demographic and clinical characteristics of the subjects included in the study

Variable	**No. (%)**	**Group 1 (CP<0.6ng/mL) No.(%)**	**Group 2 (CPC≥0.6ng/mL) No.(%)**
Number of patients	215(100)	140(65%)	75(35%)
Age on diagnosis (yo)	7.33±3.73	6.03±3.54	9.76±2.75
Girls	102(100)	65(63.7)	37(36.3)
Boys	113(100)	75(66.4)	38(33.6)
Family history (type 1 DM/type 2 DM)	97(100)	62(64)	35(36)
Acute infectious disease	47(100)	31(65)	16(35)
HbA1c on setting the diagnosis (%)	11.47±2.21	11.24±2.14	11.91±2.27

Table 7.2. Values of the average CP at the onset and in evolution in the 2 groups

Variable	**No. (%)**	**Group 1 (CP<0.6ng/mL) No.(%)**	**Group 2 (CPC≥0.6ng/mL) No.(%)**
CP average at the onset (ng/mL)	0.79±0.52	0.55±0.36	1.11±0.59
Patients with CP < 0.6 ng/mL at the onset	39 (100)	37 (37)	2 (3.5)
Patients with CP ≥ 0.6 ng/mL at the onset	119 (100)	64 (63)	55 (96,5)
CP average at 1 yo (ng/mL)	0.71±0.54	0.45±0.31	1.21±0.55
CP average at 2 yo (ng/mL)	0.55±0.45	0.3±0.26	1±0.39
CP average at 3 yo (ng/mL)	0.46±0.43	0.21±0.22	0.94±0.31

An important factor for the loss of CP in the first 3 years is the age at the moment of diagnosis. Patients with preserved CP levels (group 2) were older at the onset (9.76±2.75 years compared to 6.03±3.54 years, in group 1) and presented no DKA more frequently (60% against 32%) than the group with a low CP (Group 1).

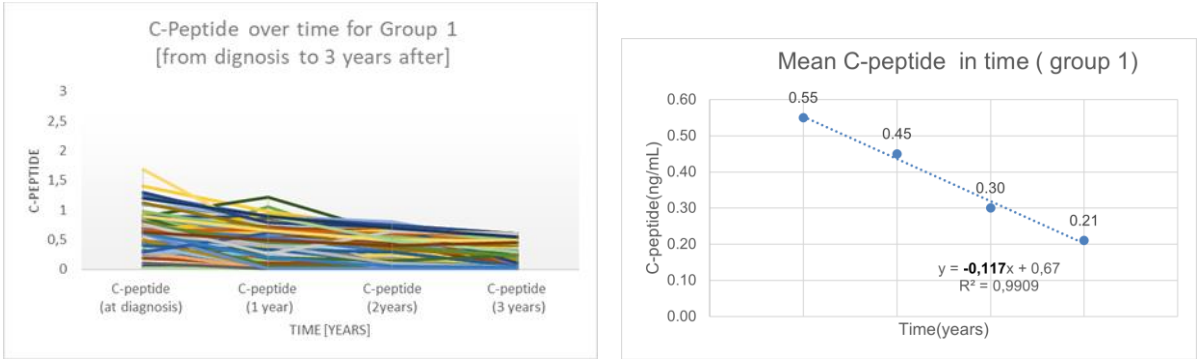
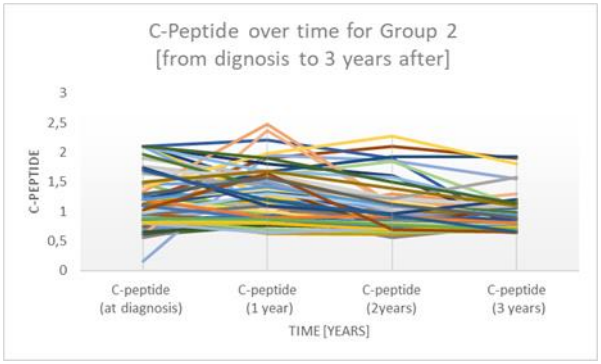


Fig. 7.4. Rate of CP decrease in the first 3 years of evolution in group 1



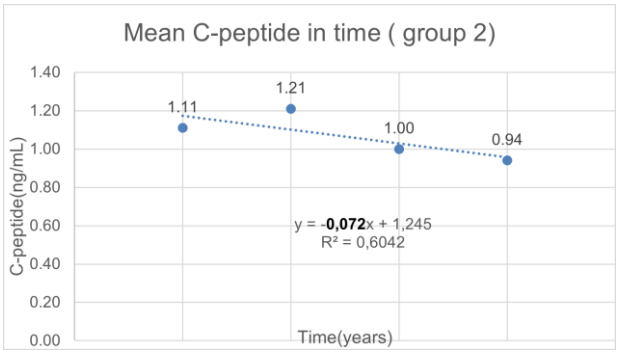


Fig. 7.5. Rate of CP decrease in the first 3 years of evolution in group 2

The CP decrease rate was calculated, and for patients in group 1 was: 0.117 ng/mL per year (Fig. 7.4.), and for those in group 2, with preserved insulin reserve, CP remained at values ≥ 0.6 ng/mL throughout the 3 years. (Fig. 7.5.). The decrease rate was lower: 0.072 ng/mL per year.

8. Study II - Research on the evolution of CP in relation to various clinical and biological parameters after 5 years of evolution of type 1 DM

8.1. Working hypothesis and specific objectives

Identification of possible clinical, biological and evolutionary factors that could be associated with the preservation of a reserve of endogenous β-cell secretion 5 years after the setting of the diagnosis of type 1 DM in children and adolescents.

8.2. Research methodology

The study included a number of 291 children with type 1 DM, whose CP value was evaluated in 2019, with the purpose of observing its correlation with the duration of diabetes.

In order to identify other correlations between CP and other clinical and biological parameters, serum CP values were followed in evolution for 5 years.

I systematized CP values as follows: CP ≥ 0.6 ng/mL (group A), CP > 0.01-0.6 ng/mL (group B), CP ≤ 0.01 ng/mL (group C), and patients were grouped according to CP value in 2019 into three groups: A, B and C.

8.3. Results and discussions

The loss of endogenous insulin secretion is the main characteristic in type 1 DM, a deficit that sets in over time, starting even before the time of diagnosis, but also in the following period [276].

8.3.1. The duration of type 1 DM and the evolution of CP

The longer the duration of type 1 DM, the lower the CP value, so that, after 9 years of evolution, CP becomes undetectable.

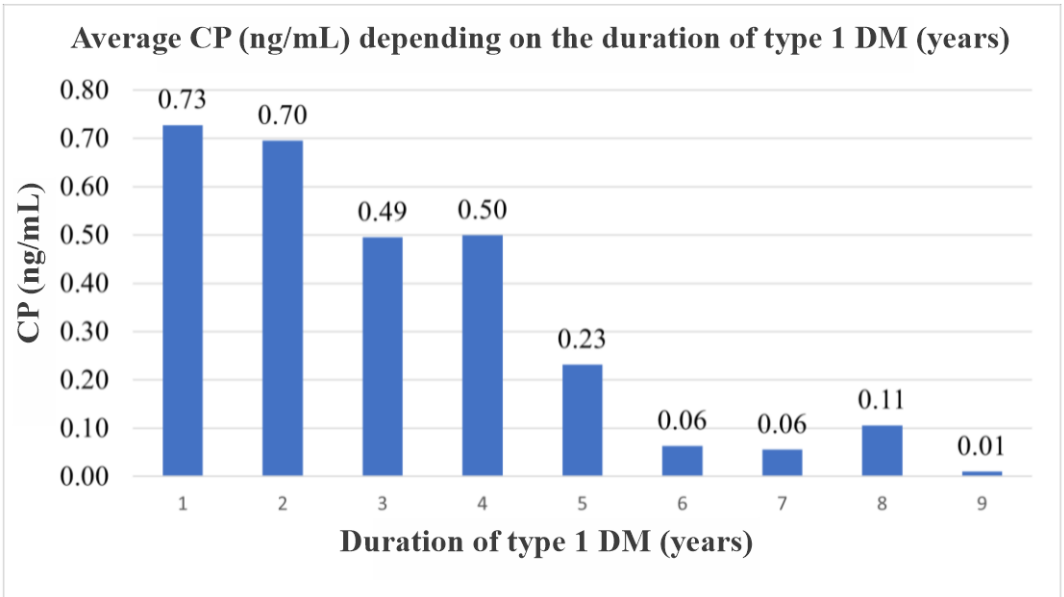


Fig. 8.2. Values of average CP (ng/mL) depending on the duration of type 1 DM (years)

The evolution of the CP average is marked by a constant decrease during the first 5 years of evolution. On average, the CP value after 3 years decreases by 55% compared to the onset, and after 5 years of evolution, it is 74% lower compared to the value on the setting of the diagnosis.

Evolution of average CP after 1 year	Evolution of average CP after 2 years	Evolution of average CP after 3 years	Evolution of average CP after 4 years	Evolution of average CP after 5 years
-23%	-41%	-55%	-67%	-74%

The duration of DM is significant in determining the value of CP. When the duration of DM increases, the value of CP decreases. As the duration of diabetes increases by one year, the value of CP decreases by approximately 0.06 ng/ml.

8.3.2. Age at onset and evolution of CP

I assessed fasting CP levels, age at onset, as well as value of CP in evolution, annually, for 5 years, to look for possible clinical-evolutionary correlations regarding the preservation of residual insulin function, assessed by serum CP levels and the age of the patients at the

onset. I divided the subjects included in the study into three groups, according to the age at diagnosis, namely: group 0: 0-4.9 years, group 1: 5-9.9 years and group 2: 10 -13 years.

In each age group, I investigated CP at the time of diagnosis and its evolution over 5 years (Fig. 8.18.)

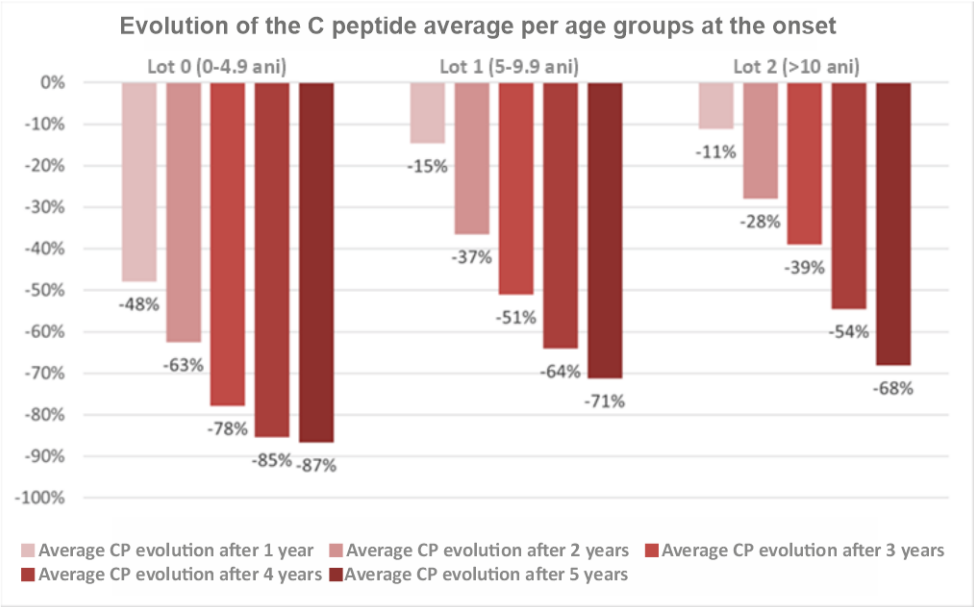


Fig. 8. 18. Evolution of CP average by age groups for 5 years

Age at onset is significant in determining CP value (p-value = 0.01, $p < 0.05$). The model indicates that, as the age at diagnosis increases by one year, the CP value after one year increases by approximately 0.04 ng/ml (with a probability of 18%). When the age at the time of diagnosis is bigger, the CP values remain at high values.

8.3.3. HbA1c and CP evolution

CP decrease rate in patients with HbA1c ≥ 10% at disease onset is more accelerated (0.126 ng/mL/year) compared to the CP decrease rate in patients whose HbA1c at the time of diagnosis was < 10% (0.107 ng/ mL/year). The correlation coefficient in both situations was significant (98%).

I followed the correlation between the HbA1c value and serum CP over time, in patients with HbA1c < 7%, compared to those where HbA1c was ≥ 7%. I grouped those with good metabolic control, with HbA1c < 7%, and respectively those with HbA1c ≥ 7%.

After 5 years of evolution of type 1 DM, it is noticed that almost 95% (no. = 74) of the patients with unfavorable glycemic control, have low serum CP concentrations < 0.6 ng/mL.

In the group of those with optimal metabolic control (HbA1c<7%), a higher share of those with preserved CP reserve is noticed (65.9%).

8.3.4. DKA at the onset and CP evolution

The group of patients who experienced a clinical onset with a more severe symptomatology (DKA) and the group of patients who did not present initial DKA were compared by analyzing the following variables: gender, environment of origin, age at onset, presence or absence of an acute infectious disease at the time of establishing the diagnosis, as well as the average values of C peptide in evolution after 1 year, after 3 years and after 5 years (Table 8.2)

Table 8.2. Comparison between patients with DKA present at onset and those with DKA absent in terms of several variables

Distribution of patients depending on onset DKA		DKA	non-DKA	p-value*
Gender	Female	58%	42%	0.99674
	Male	58%	42%	
Environment of origin	Rural	64%	36%	0.34634
	Urban	57%	43%	
Agee on onset	Group 0-5 years	67%	33%	0.00052
	Group 5-10 years	62%	38%	
	Group >10 years	39%	61%	
Acute infection on onset	Yes	69%	31%	0.02422
	No	54%	46%	

*p-value associated to the value of the Chi-square statistic test

Analyzing the previous table, it can be stated that there are associations between DKA at onset and age at onset, infection at onset (p value<0.05).

Table 8.3. The evolution of CP values over time depending on the presence or absence of onset DKA

Distribution of patients depending on onset DKA		**DKA**	**Non-DKA**	**p-value***
Value of C Peptide after 1 year	**< 0.6 ng/ mL**	67%	33%	0.00438
	≥ 0.6 ng/ mL	48%	52%	
Value of C Peptide after 3 years	**< 0.6 ng/ mL**	68%	32%	0.00076
	≥ 0.6 ng/ mL	45%	55%	
Value of C Peptide after 5 years	**< 0.6 ng/ mL**	64%	36%	0.01188
	≥ 0.6 ng/ mL	38%	62%	

CP values after 1 year of evolution are lower in patients with DKA at onset (67% of patients with CP < 0.6 ng/mL had DKA at onset). Average CP after 3 years of evolution is < 0.6 ng/mL in 68% of those with DKA at onset. And after 5 years, 64% of those with low CP come from those with DKA at onset.

9. Study III - Partial remission and correlations with CP

9.1. Working hypothesis and specific objectives

In this study, my goal is to analyze the correlations between the factors that could be considered predictive for the occurrence of the remission period and the serum CP level, as a marker of the endogenous insulin reserve.

9.2. Research methodology

In this study I used the definition of the remission period proposed by the ISPAD 2022 guideline [2,332], namely: low insulin requirement < 0.5 U/kg/day, HbA1c < 7%, duration of diabetes greater than 4 weeks, the absence of clinical symptoms.

The 291 children and adolescents included in this study were divided into two distinct groups, depending on the presence or absence of the remission period, namely: 1) the R group, of those who had remission and 2) the non-R group, of patients who did not experience the remission phase. A number of 189 patients showed partial remission (64.9%), and were included in the R group, while the remaining 102 (35.1%) did not have partial remission and were included in the non-R group.

9.3. Results and discussions

A significantly higher CP average is observed in the group of those who showed partial remission compared to those who did not, in the evolution of type 1 DM in the patients of the studied group. Thus, 1 year after the diagnosis, the average CP in those who showed remission is 0.86 ng/mL. As the duration of Type 1 DM increases, there is an obvious tendency of decrease of the average CP, progressively every year, so that, after 5 years of evolution, it is 0.35 ng/mL in children with remission compared to an average CP of only 0.07ng/mL in those who did not experience partial remission (Fig. 9.3.)

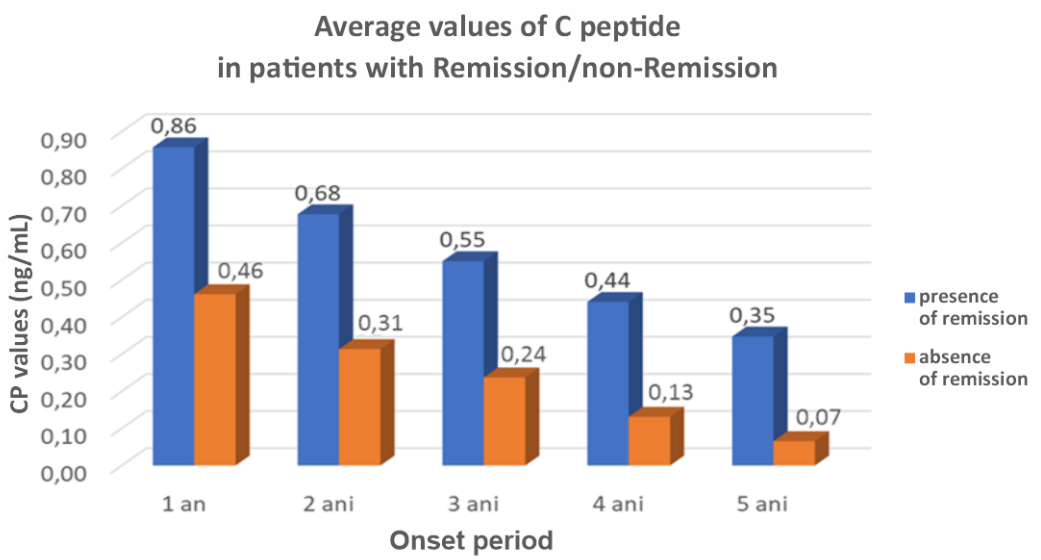


Fig. 9.3. Evolution in time of CP values in patients with and without remission

In Fig. 9.4. one may notice how the evolution of the average annual CP values are closely correlated with the presence or absence of remission. More precisely, the evolution of average CP values is much smoother in the case of those who have shown remission, while it is much more aggressive for non-remission ones.

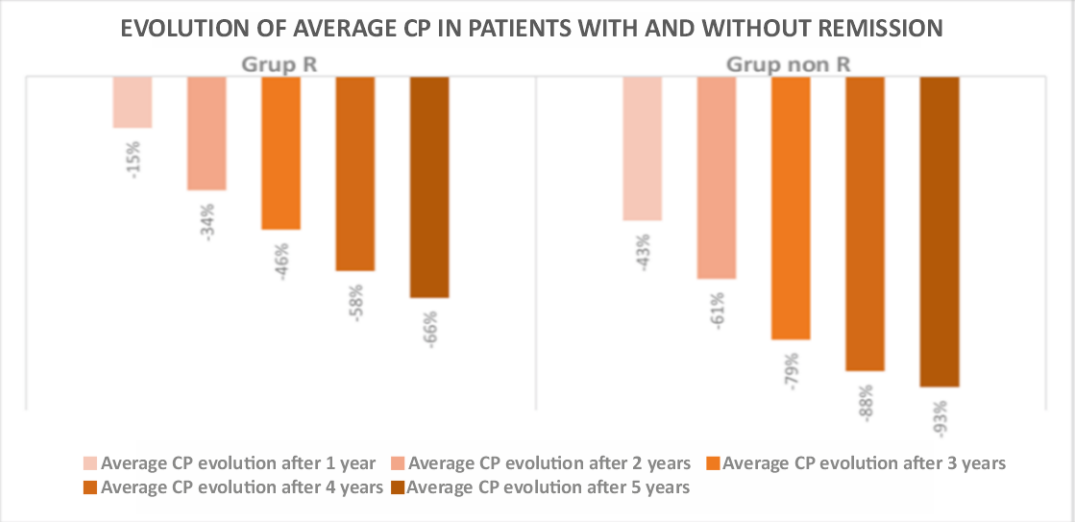


Fig. 9.4. Evolution in time of CP values in the two groups: with remission and without remission

In the group of those who showed a period of remission, after 1 year of evolution, there is a decrease in the average CP by 15% from the onset value, while 1 year after the evolution in the group of patients without remission, the decrease in the average CP is 43 % of the initial value. After 5 years of evolution, the CP average drops by 66% from the onset value, in the group of patients with remission compared to a 93% decrease, shown by the non-remission group.

I also evaluated the possible correlations between the onset manner and the occurrence of the remission period, but also the influence of the degree of severity of the onset symptoms on the duration of remission. I divided the group of 291 patients into 4 groups according to the onset manner: without DKA, with mild DKA, with moderate DKA and, respectively, with severe DKA.

In Fig. 9.14., the distribution of cases with remission, respectively, without remission, depending on the category of DKA, is noted. It can be stated that the two parameters: the occurrence of remission and DKA, are in close correlation (p-value = 0.0047, $p < 0.05$).

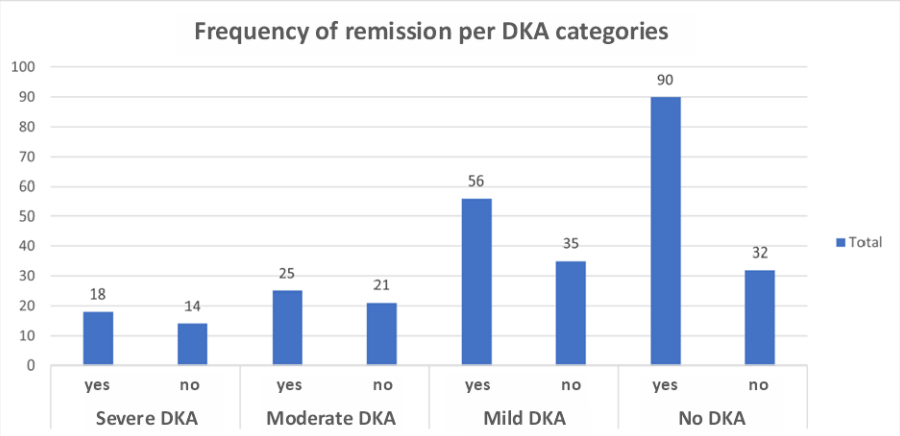


Fig. 9.14. The frequency of remission according to the presence of DKA at the onset and the severity of DKA

The graph below shows the distribution of cases with remission by age groups: group 0 = 0-4.9 yo, group 1 = 5-9.9 yo and group 3 = 10-18 yo.

Thus, it can be stated that remission is more frequent among older patients (group 2 = 10-18 yo).

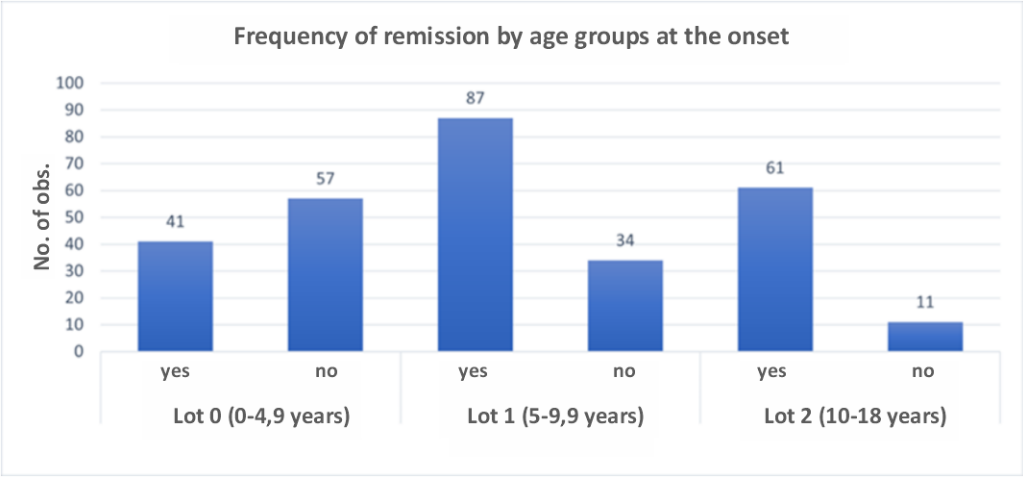


Fig. 9.15. Frequency of remission according to age at onset

10. Conclusions and personal contributions

Personal contributions

The presented study supports what is already known about the decline of the secretory function of pancreatic β-cells in the evolution of type 1 DM and brings data about the natural evolution of C-peptide in a significant group of children and adolescents from the pediatric population in Romania. The research included not only a representative group of patients, but also the entire pediatric age range: 0-18 years.

The studies aiming at researching the preservation of C peptide in patients with type 1 DM were carried out in the vast majority over a short period of time, of 1-2 years, proposing to study the period of partial remission. The study presented herein was carried out over a period of 5 years, bringing important data from this point of view as well.

Another significant aspect in this study is the representative number of patients aged 0-5, considering that there are few studies in the specialized literature that include patients in this age group.

Conclusions

The current research brings forth the following findings:

- The urban environment of origin recorded the highest prevalence (84.5%), the average onset age of type 1 DM was of approximately 7 years. The age at which the onset of type 1 DM occurred for the largest number of patients was 0-1 years (no = 35), representing 12% of all the subjects included in the study.

- In the group of 291 patients included in the study, the majority was represented by male patients: 155 (53%) patients, and 136 (47%) of the subjects were female.

- In terms of family history of type 1 diabetes and type 2 diabetes, it was present in the case of 46 patients (16%) for type 1 DM and respectively 85 patients (29%) for type 2 DM. It should be emphasized that in most of the patients included in the study, 160 (55%) did not have a family history of diabetes.

- Older age (9.76±3.54 years) at diagnosis increases the probability of maintaining residual insulin secretion reflected in CP levels ≥ 0.6 ng/mL after 3 years of evolution, which can be considered a protective factor for a slower decline of CP secretion. Younger age (6.03±3.54 years) at onset was correlated with CP levels < 0.6 ng/mL after 3 years of evolution, data that also agree with other published studies [184,241,285,290,305].

- Age at onset is significant in determining CP value (p-value of statistical test is < 0.01). With the increase in age at the time of diagnosis by 1 year, the CP value at one year increases by approximately 0.04 ng/mL.

- Among patients with DKA, their majority, respectively 31% (no = 91) presented mild forms, 11% of them (no = 32), had an onset with moderate DKA, and in 16% (no = 46), the onset was with severe DKA.

- Younger patients are more prone to DKA at onset: 67% of patients aged 0-5 and respectively, 62% of those aged 5-10 had DKA at onset, while only 39% of the patients aged: >10 had DKA at onset (p=0.00052, p<0.05).

- Patients with severe symptoms (DKA) or acute infection at the onset are at a higher risk to experience a faster CP decline. An onset infection is more frequent (69%) among patients with DKA at the time of diagnosis (p = 0.02422, p<0.05).

- From the onset, a correlation is noted between the CP serum level and the degree of DKA severity, in the sense that, the more severe the DKA, the lower the CP serum concentration at the onset.

- The presence of DKA at the onset of type 1 DM negatively impacts the preservation of pancreatic β-cell function over time. Consequently, an effective

prevention of DKA at the time when the diagnosis of this disease is established could provide long-lasting benefits.

- C-peptide values 1 year after the evolution are lower in patients with onset DKA (67% of patients with CP < 0.6 ng/mL had DKA at onset). Average CP after 3 years of evolution is < 0.6 ng/mL in 68% of those with DKA at onset. And after 5 years, 64% of those with low CP come from those with onset DKA.

- Patients with CP values ≥ 0.6 ng/mL at the time of diagnosis keep values > 0.6ng/mL even after 3 years of diabetes evolution. CP decline in this category of patients was slower (decline rate = 0.072 ng/mL, R^2= 0.6042) compared to patients who, at the onset, had a low CP level < 0.6ng/mL (decrease rate = 0.117 ng/mL, R^2=0.9909).

- The duration of DM is significant in determining CP value ($p<0.05$). There is an inversely proportional relation between the duration of Type 1 DM and the CP value. As the duration of diabetes increases by one year, the CP value decreases by approximately 0.06 ng/ml (with a probability of 27%).

- On average, the C peptide value after 3 years of evolution is 55% lower than the onset value, and after 5 years of evolution, it is 74% lower than the initial value.

- After 5 years of evolution: 23% of the patients included in the study presented CP values ≥ 0.6 ng/mL, 19% of them had CP=0.01-0.6 ng/mL and 58% of them showed CP levels < 0.01 ng/mL.

- The longer the duration of Type 1 DM, the lower the CP value, so that, after 9 years of evolution, CP becomes undetectable (< 0.01 ng/mL).

- One may notice that the decrease in CP values is on average slower in patients aged > 10 (it decreased by 68% after 5 years of evolution), constantly accelerating the younger the age. In the age group of 0-4.9, the average CP decreased by 87% after 5 years of evolution and in those aged between 5-9.9, the decrease in CP was at 5 years by 71%.

- The CP decrease rate in patients with HbA1c ≥ 10% at the onset of the disease is more accelerated (0.126 ng/mL/year, R^2= 0.9852) compared to the CP decrease rate in patients in whom HbA1c at the time of diagnosis was < 10% (0.107 ng/mL/year, R^2=0.9874). The correlation coefficient in both situations was statistically significant (98%).

- The association of another autoimmune pathology at the time of diagnosis of type 1 DM does not influence the decline of residual β-cell secretory function.

- After 3 years of evolution, in the group of patients with poor metabolic control (HbA1c ≥ 7%), there is a significantly higher proportion (84.6%) of patients with CP < 0.6 ng/ml compared to those who keep a reserve of endogenous insulin (15.4%), and after 5 years, the proportion of those with glycemic imbalance and low CP level < 0.6 ng/mL increases to 95%.

- Glycemic control, assessed by the HbA1c value, improves, as the CP serum concentration increases (the proportion of patients with HbA1c < 7% increases). This tendency is maintained regardless of the duration of evolution of type 1 DM.

- The prevalence of remission was of 64.9% in the children and adolescents included in the presented study, in accordance with the data from other published studies, that reveal a prevalence between 11% and 90% [333-335].

- There are significant variations in the occurrence of partial remission depending on the age group. Patients aged 5 to 9.9 have the highest rate of partial remission (55.6%), while patients aged 0 to 4.9 and those older than 10 show lower rates of partial remission: 21.7%, respectively 22.8%.

- Patients who experienced partial remission registered higher average CP values at onset (CP=0.94 ng/mL) compared to those who did not experience remission (average CP= 0.61ng/mL).

- Patients who show remission have optimal CP levels ≥ 0.6 ng/mL and improved glycemic control in the first 3-5 years.

- After 5 years of evolution, the CP average decreases by 66% from the onset value, in the group of those with remission, compared to a 93% decrease, shown by the group of those without remission.

- The presence of DKA at the time of diagnosis was associated with a lower frequency of remission ($p = 0.047$, $p < 0.05$). 74% of the patients without DKA at the onset experienced partial remission, while only 58% of those with DKA experienced a period of remission.

Research perspectives

This study reveals the need to carry out future research that would:

• Take into account periods longer than 5 years of monitoring the presence of residual β-pancreatic function, so that the data provided are more significant.

• Offer the possibility of continuing the study to patients from pediatric clinics to adult clinics as well, where they arrive after reaching the age of 18.

• Study the influence of puberty on the evolution of endogenous insulin secretion in patients diagnosed with type 1 DM.

• Centralize data from several regional pediatric diabetes centers in the country.

• It would be interesting to analyze the degree of loss of β-cell secretion depending on the recurrence of DKA during evolution, which could further damage the residual insulin secretion.

• It is advisable to start working with IDAA1c (HbA1c adjusted according to insulin dose) to define the remission phase of type 1 DM. IDAA1c correlates very well with CP level, being also recommended by ISPAD.

• There should be the possibility to carry out genetic tests (HLA study to establish the genetic subtypes of type 1 DM) which would bring added value to research in the field of type 1 DM

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