Hypopituitarism due to pituitary adenomas, traumatic brain injury and stroke

Ph.D. Thesis

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1 Introduction

1.1 Hypopituitarism: definition, epidemiology and etiology

Hypopituitarism first described in 1914 by Simmonds, results from the complete or partial dysfunction of the anterior and/or posterior pituitary gland.

The prevalence in adulthood is 45/100 000, average incidence is 4/100 000/year. Hypopituitarism is a potentially life threatening condition, which increases mortality in the long term, too.

Pituitary dysfunction can result from congenital abnormalities and acquired diseases of the hypothalamo-hypophyseal structures, or from pituitary stalk lesions. A population based study assessing the prevalence and incidence of hypopituitarism revealed, that pituitary tumors and perisellar masses were responsible for its development in 61 % and 9 % of the cases, respectively. Non-tumor origin was detected in 30 % of the patients, while idiopathic pituitary disease was diagnosed in 11 %, probably as the result of previously not documented TBI, genetic disorders, or empty sella. The prevalence of pituitary tumors is higher than previously assumed, a meta-analysis found it to be 16.5%.

Recently it has been discovered that traumatic brain injury (TBI) and subarachnoid hemorrhage (SAH) induced pituitary dysfunction is common and mostly underdiagnosed. Some degree of hypopituitarism was found in 35 % of TBI patients, and in 48 % of SAH patients, growth hormone and gonadotropin deficiencies being the most frequent. Post TBI pituitary dysfunction probably attributes to the impaired recovery and cognitive deficits of these patients. It has been well documented that perisellar irradiation leads to hypopituitarism in the long term. Agha et al. found, that in 41 % of patients with a medical history of radiation therapy for adult non-pituitary brain tumors developed some degree of pituitary dysfunction later. Data revealing impaired pituitary function in 19 % of ischemic stroke patients and in 38 % of patients with prior surgery for non-pituitary brain tumors were more surprising.

2 Aims

The objective of the thesis was

1. to analyze the prevalence of hypopituitarism in a large cohort of Hungarian patients with pituitary adenoma
2. to find risk factors for the development of hypopituitarism in patients treated with pituitary adenomas
3. to assess the long-term prevalence of hypopituitarism after TBI in a large group of patients
4. to find possible risk factors/predictors of hypopituitarism in patients who suffered severe/moderate head trauma
5. to evaluate the possible role of early clinical parameters (on-admission laboratory and ICU monitored parameters) of severe brain trauma patients in the development of endocrine deficits
6. to determine the prevalence of impaired GH secretion in patients after stroke
7. to find the most effective diagnostic tool to verify GH deficiency in post-stroke patients by comparing different GH stimulatory tests

3 Hypopituitarism in patients with pituitary adenomas

3.1 Patients and methods

This retrospective study was based on the data of 224 patients (113 women and 111 men, average age at time of diagnosis: 43 years, min.: 16 years, max.: 80 years), treated at the endocrine clinic of the 1st Department of Internal Medicine, University of Pécs, with pituitary adenomas between 1972 and 2011. Different treatment modalities, their effectiveness and side effects were evaluated. Patients' data were analyzed in terms of gender, age, adenoma size, tissue types, therapeutic approaches (drugs, surgery, and irradiation) and side effects. Data assessment was done by Windows Excel program, for the statistical analysis Student's t-test, chi-square test and ANOVA were used.

3.2 Results and Discussion

Hypopituitarism, the most frequent complication of pituitary adenomas results from either surgery or irradiation or from the adenoma itself compressing normal pituitary

tissue. In 115 patients of the studied 224, different severity of pituitary insufficiency developed during the follow-up period. Mostly non-functioning adenomas were responsible for the pituitary dysfunction. In addition, this type of tumor tended to result in more severe pituitary insufficiency, with multiple hormonal dysfunctions. Due to irradiation, 86.3 % of the patients developed hypopituitarism in the long-term; almost two thirds of them needed treatment for severe hypopituitarism. Pituitary adenoma apoplexy resulted in hypopituitarism in all cases.

Figure 1. Distribution of pituitary deficiency by the severity (N=115)

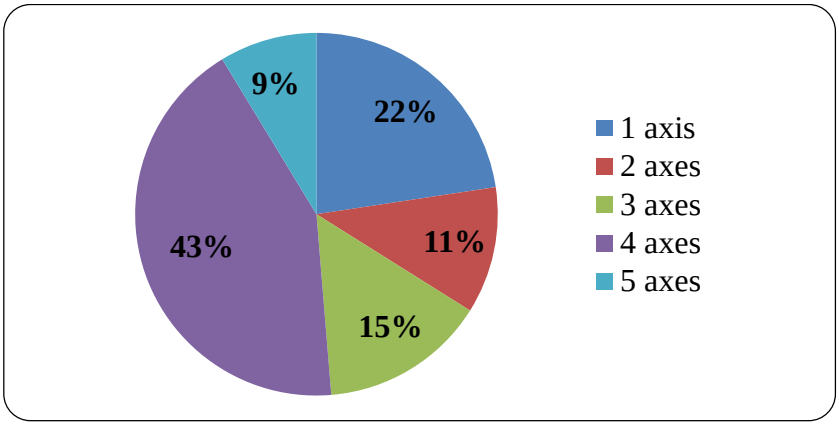
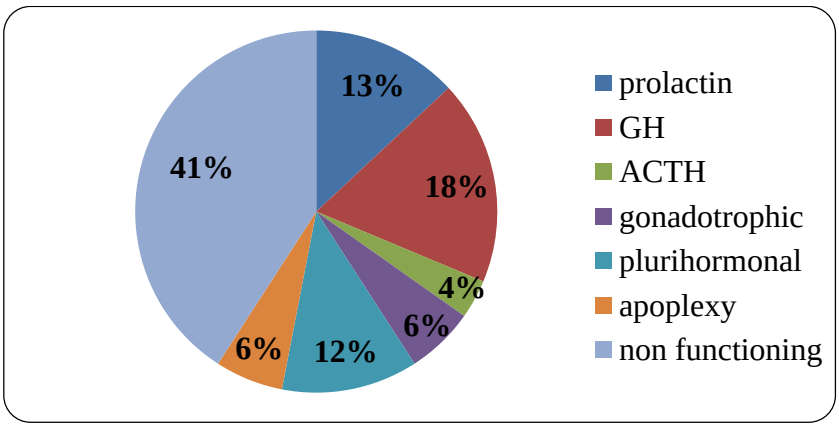


Figure 2. Distribution of pituitary deficiency according to the hormone production of adenomas



4 Hypopituitarism after traumatic brain injury

4.1 Patients and methods

Patients available for endocrine follow-up suffered TBI between 2003 and 2013. Data were collected regarding the type and severity of brain injury, on endocrine function, clinical and radiological parameters using the joint database of the Department of Neurosurgery and the 1st Department of Internal Medicine, Endocrine Division, University of Pecs, Hungary. Endocrine evaluation was either part of the routine neurosurgical follow-up, or patients were asked by letter to participate in the endocrine screening. Of the 86 survivals of 413 severe head trauma patients treated at this center during a 10-year long period, 76 had endocrine test results. Fifty of the 392 moderate head trauma patients treated between 2007- 2012 answered to the invitation to participate in our study.

Post-TBI pituitary functions were evaluated in 126 patients: 103 men and 23 women. Nine patients were younger than 18 years at the time of brain trauma, the youngest being 11, and the oldest patient 89 years old. Their mean age at the time of brain injury was 42.4 years (men: 42.3 years, women: 43.0 years, NS).

The severity of brain injury was determined according to the most severe Glasgow Coma Scale (GCS) score during neurosurgical hospitalization and intensive care. This classification was chosen because on-admission high GCS scores deteriorate significantly in many patients, representing more severe brain injury. Based on this, patients were divided into a severe (lowest GCS score $\leq$8) and a moderate (GCS score 9-12) head trauma group.

According to this classification, 76 patients had severe, and 50 patients had moderately severe brain injury. Neurosurgical intervention has been performed in 68 subjects also including external ventricular drainage (EVD) in 38 patients. In 25 cases, exclusively EVD was applied. Intensive care without surgical intervention was sufficient in 33 patients.

In order to determine possible risk factors for post TBI hypopituitarism, CT and/or MRI findings during the acute phase

were also assessed. Primarily focal brain injury was present in 87 cases, while 39 patients suffered predominantly diffuse brain injury. The leading diagnoses according to the imaging procedures were subdural hemorrhage (SDH): 37 patients, intracranial hemorrhage (ICH): 27 patients, SDH+ICH: 12 cases, epidural hemorrhage (EDH): 16 patients, diffuse injury (DIFF): 34 patients. In 22 cases, base skull fracture was also present.

The first endocrine evaluation after TBI varied between 1 month and 5.75 years (average 2.0 years). Multiple blood tests were performed in 82 patients; their average endocrine follow-up period was 3 years. The mean$\pm$SD of follow-up time after TBI was 3.98$\pm$2.54 years.

To consider the differences in data quality, we divided our patients into three groups according to the completeness of endocrine data. Group A (n: 44): subjects with single basal hormone (free FT4, TSH, testosterone, LH, FSH, ACTH, cortisol, GH, IGF1, prolactin) results, group B (n: 48): subjects with stimulation tests for GH and/or ACTH axes in addition to basal hormone measurements, group C (n: 34): subjects with multiple basal hormone tests. In optimal circumstances, stimulation tests would have been done more frequently but patients in group A were lost for follow-up. If other pituitary failure was evident from basal hormone results, one stimulation test was used to diagnose GHD (ITT in 25 cases, glucagon test in 14 cases and arginine test in 1 case). Two GH stimulation tests were required in eight patients as GH production was the only affected pituitary axis: in five patients arginine and glucagon tests, in two cases ITT and glucagon and in one case ITT and arginine tests were done.

Table 1. Definitions of hormonal dysfunctions used in the study

Thyroid-stimulating hormone (TSH) deficiency	free thyroxine <12 pmol/L and TSH ≤2.5 U/L
Adrenocorticotropic hormone (ACTH) deficiency	basal cortisol <100 nmol/L or peak cortisol <500 nmol/L in stimulation tests (insulin tolerance test or glucagon test)
Luteinizing hormone (LH)/follicle stimulating hormone (FSH) deficiency in men	testosterone level <9.9 nmol/L and LH <= 8.6 U/L and/or FSH <= 12.4 U/L
LH/ FSH deficiency in women <50 years of age	amenorrhea and/or LH <= 1.7 U/L and FSH <= 1.5 U/L
LH/ FSH deficiency in women >50 years of age	LH <= 7.7 U/L and/or FSH: <= 15 U/L
Growth hormone deficiency (GHD)	peak GH below the cut-off value in the stimulation tests (ITT/glucagon tests: peak GH < 3 ng/ml, arginine test: peak GH <4-11 ng/ml depending on the BMI)
Growth hormone insufficiency (GHI)	insulin-like growth factor-1 (IGF-I) level below the age-and sex specific reference value (IGF-I SDS < -2.00) and stimulation test is not possible or peak GH levels between 3 and 10 ng/ml in ITT/glucagon tests

The use of the GHI category is rather controversial, as cut-off value for GHD (3 ng/ml) is arbitrary and a number of studies suggested the use of higher cut-off values based on ROC analysis (for ITT it was found being 5.62 ng/ml). As many other biological parameters, the impairment of GH secretion forms a continuous variable. Patients with borderline response in the stimulation tests may have symptoms of GHD. In our GHI patient population (N=31) multiple hormone deficits were detected in 14 cases. The IGF-I SDS values were significantly lower (mean±SD: -2.96±1.72 ng/ml, p=0.000), than in patients without impaired GH secretion (mean±SD: -1.14±1.55 ng/ml) and was not different from the GHD group (mean±SD: -2.50±1.48 ng/ml, p=0.35). Stimulation tests were possible in 13 subjects and the mean GH peak was 7.04 ng/ml (min: 3.36 ng/ml, max: 9.4 ng/ml). All the available data were evaluated individually when patients were classified to this category.

4.2. Statistical analysis

Statistical analyses were performed using the SPSS 22.0 software (SPSS, Inc., Chicago, IL, USA). Descriptive statistics of ratio scaled variables are expressed as mean ± standard deviation (SD). Relationships between binomial variables were tested using Chi-square and Fischer's exact tests as appropriate. Ratio scaled variables of subgroups were compared using Student's t-test. Relationships between ratio scaled variables were evaluated with bivariate correlation. To identify the determinants of pituitary failure,

multiple and single hormone deficiencies and new hormonal disturbances, binary logistic regression analysis using backward method was performed. Values of $P < 0.05$ were considered statistically significant.

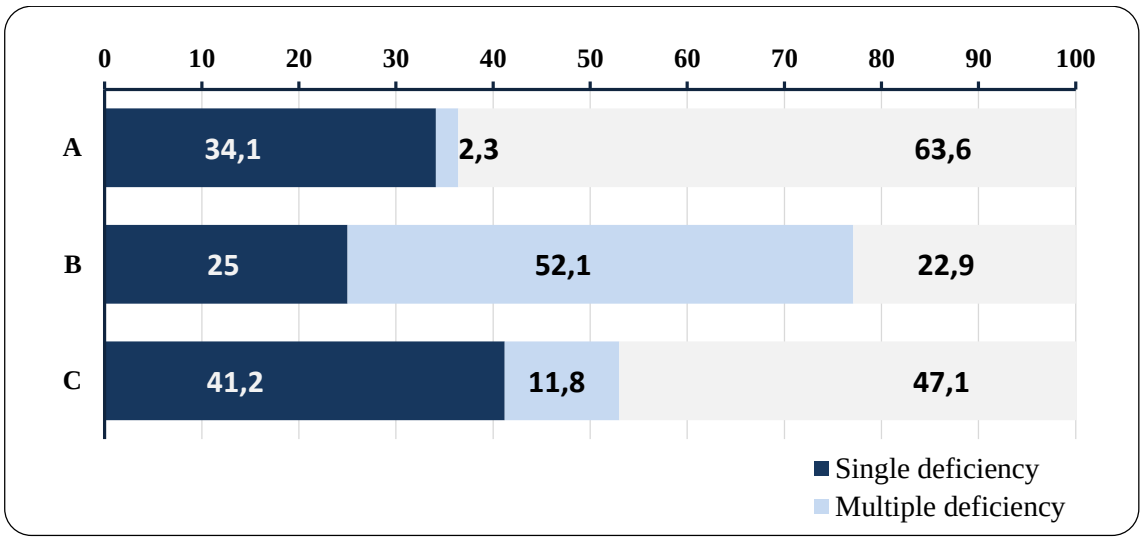
4.3. Results: prevalence of pituitary dysfunction

The prevalence of any major anterior pituitary hormone deficiency among the 126 patients was 57.1%. GHD/GHI was the most frequent (39.7%) abnormality, followed by secondary hypogonadism (23.0%), while secondary hypothyroidism and ACTH deficiency were diagnosed in 16.7 and 10.3% of all TBI patients, respectively. Of the investigated men 28.2% exhibited secondary hypogonadism but no affected women was detected.

In 56.9% of the cases with hormone deficiency, only one pituitary axis was impaired. Two patients developed complete anterior pituitary insufficiency, in which all four hormone axes were affected. Not just the GHD/GHI occurred as isolated deficiency, 20 other isolated hormone failures (9 TSH, 9 FSH/LH and 2 ACTH) were detected.

Multiple pituitary dysfunctions were found most frequently (52.1%) in those patients who had stimulation tests, too (group B), while single deficiency was diagnosed in patients with only basal endocrine evaluations (group A: 34.1%, group C: 41.2 %).

Figure 3. Prevalence of single and multiple pituitary deficiencies in % by different definitions



A: patients with single basal hormone measurement, B: patients with basal hormone results and stimulation tests, C: patients with multiple basal hormone measurements

Although a selection bias definitely affected the comparisons of these groups, since stimulation tests were more frequently done in patients with abnormal basal hormone measurements, basal hormone measurements have been enough to assess the thyroid and gonadal axes (stimulation tests would be required only for the evaluation of GH and ACTH productions). No statistically significant association has been established between the type of brain injury and pituitary malfunction.

Of the 82 patients with multiple endocrine evaluations, 31.7 % presented changes in major hormonal deficiencies during the follow-up period. Sixteen patients had new hormone deficiencies in the course of an average follow up period of 44 months (GHD/GHI: 3/5, LH/FSH: 9, ACTH: 5, TSH: 2), while 10 subjects' hormone deficiencies resolved during the average follow up period of 52 months (GHI: 1, LH/FSH: 4, TSH: 4, ACTH: 1).

4.4. Risk factors associated with hypopituitarism

Concerning the possible risk factors for the development of post-traumatic pituitary dysfunction, the prevalence of hormone deficiencies was analyzed in relation to age, gender, GCS scores, injury types, basal skull fracture, ventricular drain insertion and requirement for neurosurgery. GHD+GHI were more frequent in patients with severe brain injury, ventricular drain insertion and neurosurgery (OR: 2.70, 2.58 and 3.53). (*Table 2.)*

Table 2. P values of relationships between hormonal failures and anthropometric parameters, type of trauma, interventions. Significant correlations are enhanced.

Hormone deficiency	**severity of TBI[a]**	**type of TBI[a]**	**ventricular drain[a]**	**surgery[a]**	**basal skull fracture[a]**	**age[b]**	**gender[a]**
GHI+GHD	**0.011**	0.171	**0.011**	**0.001**	0.074	0.313	0.052
GHD	0.784	**0.037**	0.803	**0.001**	0.835	0.320	0.112
FSH/LH	0.051	0.655	0.138	0.155	0.602	0.751	**0.004**
TSH	0.103	0.196	0.232	0.079	0.294	0.753	0.918
ACTH	0.615	0.211	0.380	0.563	0.835	0.872	0.635
Multiple	**0.036**	0.301	0.094	**0.004**	0.218	0.713	0.060
All	**0.002**	0.617	**0.012**	0.063	0.223	0.131	0.143

a: Chi-square test, b:Student t-test

GHD was more prevalent after focal injury (OR: 4.49) and markedly associated to surgical intervention (OR: 9.33). Male gender predisposed to FSH/LH deficit (OR: 9.01). Multiple hormone deficiencies correlated to the severity of TBI (OR: 2.66) and neurosurgery (OR: 3.72). All hormonal disturbances were more prevalent after severe head trauma (OR: 3.25) and ventricular drain insertion (OR: 2.52).

The aforementioned factors were included in a backward binary logistic regression model to test for independent determinants of hypopituitarism. The individual and combined hormone deficiencies and the changes during follow-up time were analyzed separately. Hormonal disturbances detected at the first investigation were determined by the severity of trauma and by focal injury. Later, only the severity of TBI remained an independent predictor. None of the investigated factors related to the development of new hormonal failures. Multiple hormonal deficiencies, GHD+GHI and GHD were all influenced by the requirement of surgical intervention. GHD+GHI subgroup was associated to ventricular drain insertion, too. Independent predictors were not identified for the evolution of FSH/LH, TSH and ACTH deficiency.

4.5. Discussion, conclusion

In summary, our data confirm hypopituitarism being common due to TBI, especially in severe cases. It seems that neurosurgical intervention is an independent risk factor. Acute circumstances can influence the development of early pituitary dysfunctions, but they are usually not predictive for the evolving long-term hormonal disturbances, since pituitary failure may be a dynamic condition in these patients. Our knowledge about the pathomechanism of pituitary damage and the way of regeneration is still incomplete. Periodic evaluations of endocrine function after the first post-injury year may be necessary in a selected subgroup - especially after severe head injury, requirement for neurosurgical interventions, incriminating clinical signs - since pituitary function may change in a considerable proportion of these patients in long term.

5 Can early clinical parameters predict post-traumatic pituitary dysfunction in severe traumatic brain injury?

5.1 Methods and Materials

Data were collected in a prospective fashion regarding the type of brain injury, on endocrine dysfunction, clinical, laboratory and intensive care unit (ICU) monitored parameters using the joint database of the Department of Neurosurgery and the Ist Department of Internal Medicine, Endocrine Division, University of Pecs, Hungary. Patients available for endocrine follow-up suffered TBI between 2003 and 2013. Endocrine evaluation was part of the routine neurosurgical follow-up. During this 10 year-long period, of the 413 consecutive severe TBI patients 86 survived. Endocrine screening was performed in 76 patients, but only 63 injured's on-admission clinical parameters and ICU monitored data were available to statistic evaluation. The mean age of our patients at the time of TBI was 37.5±17.0 years and they were predominantly males (82.5%). The mean on admission Glasgow Coma Scale (GCS) and GCS motor scores were 6.7±2.8 and 3.7±1.5, respectively. The predominant injury type, affecting 47.6 % of the patients was road traffic accident (RTA), 13 patients suffered multitrauma. CT scans were evaluated according to the Marshall CT classification system. Diffuse brain damage defined by lack of focal lesions was seen in 33.3 % of the patients, subdural hematoma was the second most frequent finding, affecting 18 injured. Half of the studied patients (50.8 %) had skull fractures, too. The average days spent in the Intensive Care Unit (ICU) was 13.8±9.7 days, 61.9 % of the investigated TBI patients required neurosurgical intervention and 87.3 % had ventriculostomy. The median time of the first endocrine evaluation after TBI was 1.1 year. Multiple endocrine evaluations were performed in 48 patients; their average endocrine follow-up period was 3.9 years. *Table 3*.

The definitions of hormonal dysfunctions used in the study are presented in *Table 1*, in section 4.1. Endocrine evaluations were done on a controlled basis.

Statistical analyses were performed using the IBM SPSS Statistics 23 software (IBM Corporation, Armonk, NY, USA). In addition to the descriptive statistics for the identification of the determinants of pituitary failure, multiple and single hormone deficiencies and new hormonal disturbances, binary logistic regression analysis was performed. Values of $p < 0.05$ were considered statistically significant.

Table 3. Descriptive statistic characteristics of the enrolled 63 sTBI subjects

	n	63	
Demographic characteristics	Age (mean±SD)	37.5±17.0 y	
	Gender	Female:	11 (17.5%)
		Male:	52 (82.5%)
On admission parameters	GCS on admission (mean±SD)	6.7±2.8	
	GCS motor score (mean±SD)	3.7±1.5	
	Mechanism	RTA:	30 (47.6%)
		Fall:	18 (28.6%)
		Other/unknown:	15 (23.8%)
	Multitrauma	13 (20.6%)	
	Main diagnosis/type of intracranial lesion	SDH:	18 (28.6%)
		EDH:	11 (17.5%)
		ICH:	10 (15.9%)
		Diffuse:	21 (33.3%)
		Other/complex:	3 (4.8%)
	Skull fracture	32 (50.8%)	
	Reaction of pupils	Both:	29 (46.0%)
		One:	5 (7.9%)
		None:	21 (33.3%)
		Unknown:	8 (12.7%)
	Coagulopathy	29 (46.0%)	
	1st blood glucose (mean±SD)	7.5±2.3 mmol/L	
	1st blood Hgb (mean±SD)	124.7±17.3 g/L	
	1st ICP (mean±SD)	8.0±9.9 Hgmm	
	1st MABP (mean±SD)	88.7±15.3 Hgmm	
Parameters of prolonged treatment and ICU monitored data	Ventriculostomy	55 (87.3%)	
	Surgical intervention	39 (61.9%)	
	Days spent on ICU (mean±SD)	13.8±9.7	
	Systematic and/or CSF infection	31 (49.2%)	
	ICP>20 Hgmm	12.7±15.5%	
	CPP<60 Hgmm	9.4±12.9%	
Endocrine alterations (revealed on follow up visits)	GH axis	32 (50.8%)	
	Gonadal axis	15 (23.8%)	
	Thyroid axis	14 (22.2%)	
	Adrenal axis	6 (9.5%)	
	Sum	43 (68.3%)	

5.2 Results

Post-traumatic hypopituitarism (PTH) was diagnosed during long-term endocrine follow up in 68.3 % of the 63 studied severe TBI patients. The growth hormone deficiency and insufficiency (GHD+GHI) were the most frequently affected pituitary axis, present in all together 50.8 % of the cases. (GHD: 11.1 %, GHI: 39.7 %) Central hypogonadism affected 23.8 % of the male patients; hypothyroidism and secondary adrenal failure were found in 22.2 % and 9.5 % of the investigated population, respectively. Isolated hormone deficiency was found in 25 cases: GH: 16, LH/FSH: 3, TSH: 5, ACTH: 1. Two hormonal axes were affected in 13 patients: GH+LH/FSH: 6, GH+TSH: 3, GH+ACTH: 2, LH/FSH+ACTH: 1, TSH+ACTH: 1. The combination of GH, LH/FSH and TSH deficiency was detected in 4 subjects and one patient suffered from complete adenohypophysis failure. Early onset (within 1 year of the brain trauma) PTH was found in 24 patients (38.1%). Binary logistic regression was performed to find a possible connection between on-admission and ICU monitored clinical parameters and the development of different pituitary hormone deficiencies. No significant predictive parameter was found in the analysis. When studying the same clinical parameters in connection with early and late (defined as onset of more than 1 year post-injury) PTH, we found significant correlations between early endocrine dysfunctions and surgical intervention (OR: 4.64) and subdural hematoma (OR: 12). In opposite, development of late onset hypopituitarism was less prevalent after road traffic accident (OR: 0.22). *Table 4.*

Table 4. Connections between endocrine alterations, early and late onset PTH and TBI parameters (bold characters and asterisks sign the significant results)

		GH axis OR [95%CI]	Gonadal axis OR [95%CI]	Adrenal axis OR [95%CI]	Thyroid axis OR [95%CI]	AH OR [95%CI]	Early onset PTH OR [95%CI]	Late onset PTH OR [95%CI]
Age		0.98 [0.95; 1.01]	1.02 [0.98; 1.05]	0.95 [0.89; 1.01]	1.00 [0.96; 1.03]	0.98 [0.95; 1.01]	1.00 [0.97; 1.04]	1.03 [0.99; 1.07]
Gender		0.77 [0.21; 2.85]	*Not calculable*	2.67 [0.42; 16.80]	0.30 [0.04; 2.57]	0.49 [0.13; 1.84]	2.88 [0.63; 13.22]	1.07 [0.25; 4.60]
GCS on admission		0.95 [0.79; 1.15]	1.02 [0.82; 1.26]	0.89 [0.62; 1.29]	0.89 [0.69; 1.15]	1.08 [0.87; 1.34]	1.01 [0.81; 1.26]	1.01 [0.81; 1.26]
GCS motor score		0.89 [0.61; 1.31]	0.83 [0.52; 1.32]	0.84 [0.47; 1.50]	0.65 [0.40; 1.05]	0.86 [0.57; 1.31]	0.72 [0.44; 1.15]	0.88 [0.56; 1.39]
Mechanism	**RTA/Others**	0.93 [0.34; 2.59]	0.63 [0.17; 2.27]	*Not calculable*	0.53 [0.15; 1.85]	0.66 [0.22; 1.98]	0.46 [0.14; 1.52]	**0.22 [0.06; 0.81]***
	Fall/Others	0.69 [0.23; 2.11]	0.71 [0.17; 3.01]	*Not calculable*	1.02 [0.27; 3.86]	0.65 [0.20; 2.08]	1.20 [0.32; 4.51]	2.13 [0.53; 8.45]
Multitrauma		1.73 [0.50; 6.04]	1.58 [0.41; 6.11]	2.09 [0.34; 12.91]	1.06 [0.25; 4.55]	1.72 [0.42; 7.08]	3.58 [0.80; 16.05]	0.49 [0.12; 2.01]
Main diagnosis/type of intracranial lesion	**SDH/Others**	1.31 [0.44; 3.92]	0.88 [0.24; 3.25]	0.47 [0.06; 4.34]	1.54 [0.44; 5.45]	1.93 [0.54; 6.86]	**12.00 [2.27; 63.56]***	0.98 [0.27; 3.52]
	EDH/Others	3.11 [0.74; 13.06]	0.67 [0.13; 3.49]	*Not calculable*	1.40 [0.32; 6.17]	1.30 [0.30; 5.51]	0.87 [0.17; 4.38]	6.30 [0.70; 57.07]
	ICH/Others	0.36 [0.08; 1.52]	1.46 [0.33; 6.55]	*Not calculable*	1.64 [0.36; 7.38]	0.65 [0.16; 2.62]	0.42 [0.07; 2.42]	0.28 [0.05; 1.64]
	Diffuse/Others	0.83 [0.29; 2.36]	0.66 [0.18; 2.41]	2.17 [0.40; 11.80]	0.26 [0.05; 1.31]	0.65 [0.22; 1.97]	0.44 [0.13; 1.47]	0.77 [0.24; 2.47]
Skull fracture		0.50 [0.17; 1.53]	0.40 [0.11; 1.47]	0.42 [0.06; 2.77]	0.79 [0.21; 2.98]	0.38 [0.11; 1.28]	0.34 [0.10; 1.22]	0.49 [0.14; 1.72]
Reaction of pupils		0.92 [0.52; 1.61]	1.01 [0.52; 1.96]	0.83 [0.34; 2.03]	1.03 [0.52; 2.04]	1.03 [0.56; 1.91]	0.78 [0.41; 1.48]	1.26 [0.66; 2.40]
Coagulopathy		0.78 [0.29; 2.11]	0.70 [0.21; 2.27]	6.67 [0.73; 60.85]	0.56 [0.16; 1.90]	0.97 [0.33; 2.85]	0.32 [0.10; 1.06]	0.64 [0.20; 2.05]
1st blood glucose		1.19 [0.92; 1.53]	1.13 [0.88; 1.45]	0.98 [0.68; 1.43]	1.14 [0.89; 1.47]	1.03 [0.79; 1.35]	1.13 [0.86; 1.48]	0.99 [0.76; 1.28]
1st blood Hgb		1.01 [0.98; 1.04]	1.00 [0.96; 1.03]	0.97 [0.93; 1.03]	1.01 [0.98; 1.05]	1.00 [0.97; 1.03]	0.97 [0.93; 1.00]	1.02 [0.98; 1.05]
1st ICP		1.03 [0.97; 1.10]	1.04 [0.97; 1.11]	0.89 [0.77; 1.03]	0.99 [0.92; 1.06]	1.04 [0.97; 1.12]	1.05 [0.97; 1.14]	0.94 [0.87; 1.02]
1st MABP		1.01 [0.98; 1.05]	1.00 [0.96; 1.04]	0.99 [0.93; 1.05]	0.98 [0.94; 1.02]	0.99 [0.95; 1.03]	0.99 [0.95; 1.03]	1.00 [0.97; 1.04]
Ventriculostomy		3.60 [0.67; 19.43]	0.93 [0.17; 5.17]	*Not calculable*	0.84 [0.15; 4.69]	1.34 [0.29; 6.27]	0.83 [0.15; 4.58]	2.67 [0.44; 16.20]
Surgical intervention		1.81 [0.65; 5.07]	3.11 [0.78; 12.46]	0.58 [0.11; 3.16]	2.75 [0.68; 11.11]	2.07 [0.70; 6.12]	**4.64 [1.31; 16.42]***	1.89 [0.59; 6.04]
Days spent on ICU		0.98 [0.93; 1.03]	1.04 [0.98; 1.11]	1.02 [0.94; 1.10]	0.94 [0.86; 1.02]	0.99 [0.94; 1.05]	0.98 [0.92; 1.05]	0.96 [0.90; 1.03]
Systematic and/or CSF inf.		1.38 [0.51; 3.71]	1.77 [0.55; 5.76]	2.22 [0.38; 13.12]	1.51 [0.46; 4.99]	1.28 [0.44; 3.71]	1.50 [0.47; 4.79]	0.47 [0.14; 1.52]
ICP>20 Hgmm		0.46 [0.01; 18.54]	15.46 [0.26; 907.22]	0.21 [0.00; 262.33]	1.60 [0.02; 117.22]	3.30 [0.03; 338.72]	0.99 [0.00; 264.66]	0.08 [0.00; 24.57]
CPP<60 Hgmm		3.99 [0.04; 411.92]	0.87 [0.00; 178.58]	0.03 [0.00; 526.17]	1.79 [0.01; 332.01]	96.28 [0.07; 126389.55]	148.28 [0.13; 168447.58]	1.35 [0.00; 1185.19]

5.3 Conclusions

Although neurosurgical interventions and the presence of subdural hematomas were associated with a higher incidence of early onset PTH, our results indicate that the broad spectrum of investigated clinical and laboratory parameters were not predictive to identify high-risk patients for endocrine dysfunctions. This may show that not just the injury itself but also the regeneration process and other individual variables are important in determining the endocrine outcome. Our results support the absolute necessity of regular endocrine screening during the follow-up of severe TBI survivors.

6 Evaluation of growth hormone secretion after stroke

6.1 Patients and methods

Seventeen patients were included in the study (12 males; mean ± SD age: 60.5 ± 9.8 years;median (interquartile ranges) body mass index, 26.0 (23.6-29.7) kg/m2). All subjects had a previous cerebrovascular stroke that was ischemic type in 16 cases and hemorrhagic in a single patient (Pt# 7). The mean interval between the stroke and endocrine evaluation was 19.6 (± 7.3) months. All participants were ambulatory and in fair general condition. Applying the National Institutes of Health Stroke Scale (NIHSS) for the estimation of post-stroke state severity, at the time of endocrine evaluation all the patients had a low score (<8). Blood samples were collected after overnight fasting. Basal morning free thyroxin, TSH, LH, FSH, testosterone in men, cortisol, ACTH, GH, IGF1 and prolactin levels were determined. Glucagon test was carried out according to the standard procedure. The applied cut-off value of peak GH response was 3 μg/L. GHRH was given as bolus intravenous injection and was followed by an infusion of 0.5 g/kg L-arginine monohydrochloride (maximum dose 30 g) as a 10% solution (30g/300 mL) in normal saline over 30 min. Blood was taken for GH measurement at + 30, 60, 90, 120 and 150 min after start of arginine infusion. For screening, both glucagon and low dose (0.15 μg/kg) GHRH-arginine (ldGHRH-A) stimulation tests were carried out in consecutive days. If maximal GH values were less, than (i) 3 μg/L in the glucagon test and/or (ii) 11.0, 8.0, or 4.0 μg/L in the ldGHRH-A test, according to BMI <25; 25-30; >30 kg/m2, respectively, a high dose (1 μg/kg or maximum dose of 100 μg) GHRH-arginine (hdGHRH-A) stimulation, as a standard confirmatory test was carried out, as

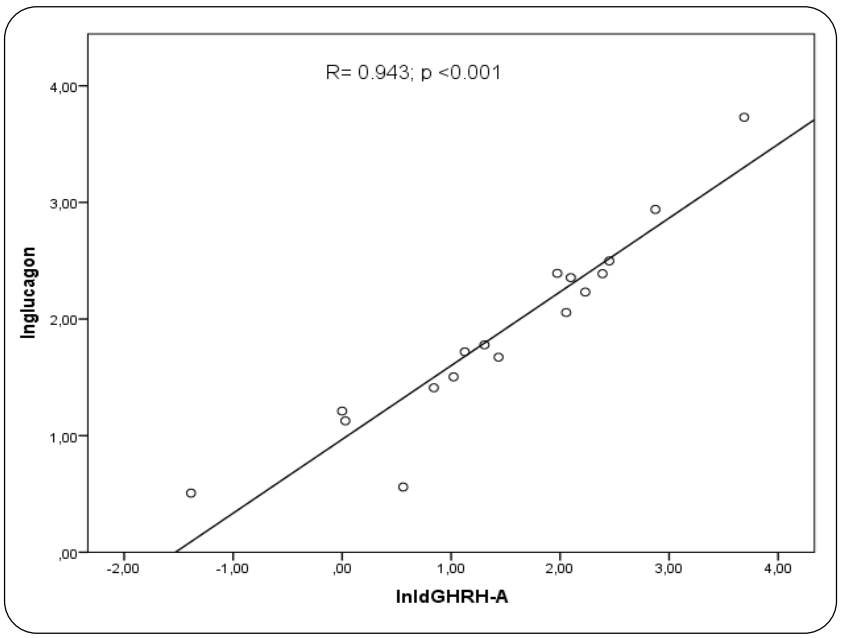
well, except one patient who refused the test (Pt# 12). Various peak GH criteria were tested in GHRH-A tests to determine impaired growth hormone response. First, BMI dependent peak GH cut-off values were analyzed, then the same results were investigated applying the universal 4.1μg/L cut-off value determined by the Endocrine Society Clinical Practice Guideline.

Statistical analyses were performed using version 22.0 of SPSS (SPSS, Inc., Chicago, IL, USA). Normality of distribution of data was tested by Kolmogorov-Smirnov test. Non-normally distributed parameters were transformed logarithmically to correct their skewed distributions. Correlations between continuous variables were assessed by calculation of linear regression using Pearson's test. Data were expressed as means ± S.D. in case of normal distribution, and median and interquartiles in case of non-normal distribution. Values of $P < 0.05$ were considered statistically significant.

6.2 Results

Impaired GH secretion could be demonstrated in 6/17 cases (35.3%) using the glucagon test. Peak GH values following glucagon stimulation test (median 3.97 μg/L, interquartile range 2.32/9.02) did not differ significantly from ldGHRH-A results (3.49 μg/L (2.73/7.93), p=0.654), and they showed excellent correlation to each other (r=0.943; p<0.001) *Figure 4.*

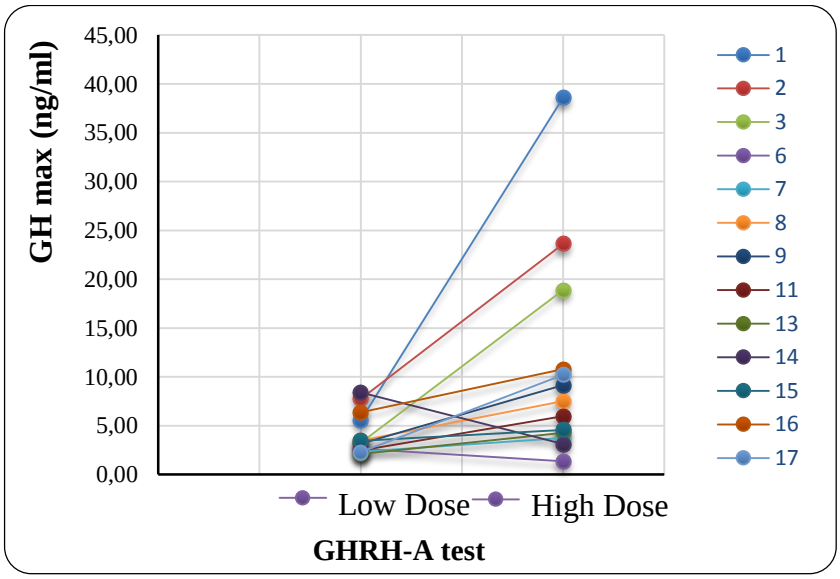
Figure 4. Correlation of maximal GH responses in glucagon and low dose(ld) GHRH-A tests. GH values did not show a normal distribution and were logarithmically transformed.



If BMI dependent cut-off values were applied, 12/17 cases (70.6%) exhibited iGH-R in the ldGHRH-A test. If the universal 4.1 μg/L cut-off value was used in the ldGHRH-A test, the rate of GH deficient patients was 10/17 (58.8%).

The results of glucagon and ldGHRH-A tests were concordant only in 52.9% of the patients, regardless of the cut-off values applied. Stimulated individual maximal GH values in the ldGHRH-A and hdGHRH-A tests are shown in *Figure 5*.

Figure 5. Stimulated individual maximal GH values in the low and high dose GHRH-A tests



The median peak GH levels of hdGHRH-A test was significantly higher as compared to ldGHRH-A ones: 6.75 (3.88/10.95) vs. 3.49 (2.73/7.93) (interquartile values) (p=0.01). The hdGHRH test detected iGH-R in six cases using the BMI-based and in three patients using the 4.1 μg/L cut-off values.

Interestingly, the peak GH values in hdGHRH-A test did not correlate to maximum GH values either in ldGHRH-A or glucagon tests. The outcome of hdGHRH-A test was concordant with the results of glucagon test in 50.0% (BMI-matched cut-off) and 58.3% (universal cut-off) of the patients. The rate of concordance between the ldGHRH-A and hdGHRH-A was even worse, 41.7% with both BMI-based and universal cut-off values. At least one stimulation test detected iGH-R in 13/17 patients (76.5%). The positivity of two GH stimulation tests is required for the diagnosis of GHD in cases where no other pituitary deficiencies are found.

The highest prevalence of GHD was 29.4% as detected by the combination of glucagon+ldGHRH-A and ldGHRH-A+hdGHRH-A tests using BMI based cut-off values. Other combination of GH stimulation tests or cut-off values showed lower prevalence of GH deficiency. All the three tests were positive only in 2/17 cases (11.8%). IGF-I levels were below the age-adjusted mean values except four cases (IGF-I mean±SD: 135.4±63.2 ng/mL, SDS mean±SD: -0.8±1.3). However, no correlations were found between IGF-I and peak GH levels reached in any stimulation tests. Two of the four patients with IGF-I above average were found GH deficient in the glucagon test and all of them in the ldGHRH-A test.

6.3 Conclusion

In conclusion, presence of iGH-R is common in post-stroke patients. However, the assessment of its exact prevalence is highly influenced by the chosen stimulation test. Widespread discrepancies occurred in the results of the available tests. Moreover, cut-off values of GHRH-A tests may also essentially modify the interpretations. Based on our data, since no clear hierarchy among the tests can be established, none of the tests can be regarded as a gold standard for the diagnosis of GHD in stroke patients. Further studies are warranted to help the diagnosis and to establish the potential benefits of GH treatment in this special group of patients.

7 Summary of new scientific results

1. In 115 patients of the studied 224 with pituitary adenomas, different severity of pituitary insufficiency developed during the follow-up period. In most of the cases, non-functioning adenomas were responsible for the pituitary hypofunction. In addition, this type of tumor tended to result in more severe pituitary insufficiency, with multiple hormonal dysfunctions. Due to irradiation, 86.3 % of the patients developed hypopituitarism in the long-term, almost two thirds of the irradiated patients needed treatment for severe hypopituitarism. Pituitary adenoma apoplexy resulted in hypopituitarism in all cases.

2. The prevalence of any major anterior pituitary hormone deficiency among the 126 patients, who suffered severe or moderate TBI, was 57.1%. In 56.9% of the TBI cases with hormone deficiency, only one pituitary axis was affected. Multiple pituitary dysfunction was found most frequently (52.1%) in those patients who had stimulation tests, too (group B), while single deficiency was diagnosed in patients with basal endocrine evaluations (34.1% in group A, 41.2 % in group C).

   No statistically significant association has been established between the type of injury and pituitary malfunction.

   Of the 82 patients with multiple endocrine evaluations, 31.7 % presented changes in major hormonal deficiencies during the follow-up period.

   GHD+GHI were more frequent in patients with severe brain injury, ventricular drain insertion and neurosurgery. GHD was more prevalent after focal injury and markedly associated to surgical intervention (OR: 9.33). Male gender predisposed to FSH/LH deficit. Multiple hormone deficiencies correlated to the severity of TBI and neurosurgery. All hormonal disturbances were more prevalent after severe head trauma and ventricular drain insertion. Multiple hormonal deficiencies, GHD+GHI and GHD were all influenced by the requirement of surgical intervention, GHD+GHI subgroup was associated to ventricular drain insertion, too.

   No independent predictors were identified for the evolution of FSH/LH, TSH and ACTH deficiency.

3. The broad spectrum of investigated early and on admission clinical and laboratory parameters of severe brain trauma patients were not predictive to identify high-risk patients for endocrine dysfunctions. Our results support the absolute necessity of regular endocrine screening during the follow-up of severe TBI survivors.

4. Presence of iGH-R is common in post-stroke patients. However, the assessment of its exact prevalence is highly influenced by the chosen stimulation test. Widespread discrepancies occurred in the results of the available tests. Moreover, cut-off values of GHRH-A tests may also essentially modify the interpretations. Based on our data, since no clear hierarchy among the tests can be established, none of the tests can be regarded as a gold standard for the diagnosis of GHD in stroke patients. Further studies are warranted to help the diagnosis and to establish the potential benefits of GH treatment in this special group of patients.

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