The disease-originated examination of two common congenital abnormalities of the male sex organ (hypospadias and cryptorchidism)

PhD Thesis Summary

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INTRODUCTION:

In my dissertation I will search for the cause of the two congenital malformations, the hypospadias and the cryptorchidism.

Hypospadias is defined as the position of the urethral meatus is on the ventral side ectopically proximally from the tip of the glans, while of cryptorchidism means that the testes cannot be found in the scrotum at birth.

Covering a period of 50 years from 1962 to 2011 an increasing tendency of hypospadias and cryptorchidism can be observed in prevalence.

The phenomenon has been registered in several countries, including Hungary.

The aim of the study is to re-analyze the frequency of the two diseases at birth in Hungary and to explore the possible maternal risk factors, like the role in the development of both diseases, associated with acute and chronic maternal illnesses and medications during the pregnancy.

Therefore, the Hungarian Case-Control Surveillance of Congenital Abnormalities (HCCSCA) database has been recorded and processed with respect to the data on maternal socio-demographic status and the maternal risk factors from 1980 to 1996.

THE OBJECTIVE OF THE STUDY:

The examination of the hypospadias and the cryptorchidism, considering the following three pre-defined criteria:

a. Based on the varied manifestations of these birth defects (CA) the definition of hypospadias and cryptorchid cases is clinically important.
b. The analysis of the frequency of hypospadias and cryptorchidism cases at birth in Hungary, as in previous disclosures showed an upward trend.
c. The etiological research of the origin of these two CAs is to find out whether the development of these diseases are caused by the maternal illnesses during pregnancy or the related drug treatments, as prevention can only be hoped in the light of their aetiology

MATERIALS AND METHODS:

Hungarian Congenital Abnormality Registry:

The Hungarian Congenital Abnormality Registry (HCAR): began operations in 1970.

This was the time when they started processing the reported cases of CA back to 1962.

The notification of the CA's cases has been done from several sources up today:

1. Obstetric institutions
2. Paediatric institutions
3. Pathology institutions
4. Since 1984 the foetal abnormalities diagnostic recognized in the foetal centres have also been reported.

The tasks and objectives of the HCAR:

a. To define the prevalence of each reported CA.
b. Early detection of any time, and spatial accumulations, launching studies to explore the causes and the initiation of measures aimed at eliminating preventable causes.
c. Medical care support based on the expected CA case estimates.
d. Determining the public health significance of the different CA units.
e. Measuring ("monitoring") of developments in the new genetic diseases.
f. Utilizing the HCAR database for scientific research.

Hungarian Case-Control Surveillance of Congenital Abnormalities:

The HCAR proved to be suitable to meet the set out goals in 1969. However, when assessing accumulation of the cases it turned out that beside the registration the exploration of the causes is also necessary.

On 1st January 1980 the HCCSCA was established whose most important goal was to explore the causes of developmental disorders.

The primarily processed information contained the following: the chronic and the acute maternal illness during pregnancy, the complications during pregnancy, the mothers' medication during pregnancy, and the information on the parents 'social status.

The reported CA diagnoses to HCAR can be checked with the help of HCCSCA. The HCCSCA is perfect for the quality control of HCAR, and for improving the reliability of the data, as well.

The group of afflicted with HCCSCA CA was selected from the HCAR.

The cases could be born alive, stillborn or after fetal diagnosis fetus from abortion.

When choosing them, three criteria must be met:

1. Only those cases were put from the HCAR to the HCCSCA, whose birth or interruption of pregnancy was reported in less than three months time. In the study period 77% of the reported CA cases were transferred from the HCAR to the HCCSCA.

2. Three CA- diseases have not been transferred from the VRONY, which are common and the origin of the disease is quite well known, moreover, they have particular clinical relevance. Such is the dislocation predisposition based on Ortolani-positivity, the congenital inguinal hernia and the large haemangioma.

3. The multiple CA caused by gene mutations and chromosomal aberrations (except for Down's disease) were omitted from the HCCSCA database.

As mentioned, the Down syndrome has been moved from the HCAR into the HCCSCA, although this CA cannot be related to the after conception harmful effects on the foetus. But the group of Down's syndrome children can be an adequate reference group, as we can calculate with the same remembrance efficiency with the mothers because of the CA, as with the CA cases thus the non-CA controls' mothers' memory distortion can be estimated (so-called. recall bias).

To every case in the HCCSCA two controls were matched with the help of the Census Institute of the Central Statistical Office, who had no CA, who were the same sex with the case, were born in the same week and their parents lived in the same region.

Between 1986 and 1992, to each CA case three controls were selected as with the increasing of the number of controls improves the reliability of the results and the statistical strength of the study.

However, after 1992 there was no financial support for the third-control applications.

A letter was sent to each mother; it was the description of the research, the essence and the importance of it which was accompanied by a printed questionnaire and a form proving their support and voluntary cooperation and a list of common maternal diseases and drugs (asking them to read before completing the survey) as well as some brochures related to the relevant CA.

The brochure included the supposed aetiology of the CA of mother's child occurring, the necessary treatment and rehabilitation procedures, as well as the possibilities of prevention for the next pregnancy.

The exposure data necessary for the exploration of the potential teratogenic effects in the HCCSCA database came from three sources:

a. Prospective data and medical records: The cases' and controls' parents were asked to send all the original medical documentation, or the copy concerning the pregnancy, the childbirth and the newborn (pregnancy care book, discharge report etc). The original documents were returned within 4 weeks.
b. Retrospective information from the mother: The mother were asked to fill in a structured, printed questionnaire concerning the months of the pregnancy, which included the maternal characteristics (age, marital status, occupation), the family history, the complications of the pregnancy, the maternal diseases, the drugs and vitamins taken by the mother (eg. Folic acid). All mothers were asked to read the attached list of medicines and disease before completing the questionnaire in order to refresh their memory. 3.9 ± 2.2, 5.2 ± 2.9 end 3.5 ± 1.2 months passed in the case group in the control group and in the Down syndrome group between the end of the mother's pregnancy and the return of the "information package".
c. Information obtained from the families who did not answer, but they were visited later.

   In the case of children born with CA unresponsive in letter, but their addresses were known, regional district nurses visited them and helped in filling out the questionnaire.

   In addition, they checked the pregnancy care book, the childbirth discharge report and other documentation of medical data and recorded the necessary information.

   The district nurses visited only 0.4% of the non-answering controls' parents, because the Ethics Committee has not contributed to visiting all the non-answering mothers of the controls, as the Committee considered it disturbing for the non CA children's family.

   Thus in the two validation studies only 200 and 600 control mothers were visited to obtain the necessary information.

In 1997 the HCCSCA system of data collection changed therefore in my dissertation I have only analyzed the data between 1980 and 1996.

For the period between 1980 and 1996, the HCCSCA contains 22,843 cases of CA, 38,151 population controls and 834 cases of Down's syndrome, as malformative control.

Between 1980 and 1996 the necessary data were available in 96.3% of the cases of CA (84.4% replied by letter, 11.9% were personally visited by the district nurses).

82.6% of the controls respondents answered by mail, and the nurses visited just only the 0.4%.

95.0% of the Down syndrome controls could the necessary data be obtained, namely 84.0% in response and 11.0% by family visits.

98.4% of the cases' and the Down's syndrome controls' mothers contributed to the register of their child's name and address.

The names and addresses of the children whose mothers did not permit the data record were removed from the register. The name and address of the controls were not recorded.

Introducing the Analyzed Exposures:

The examination of the following exposures was one of our objectives:

a. the analysis of the acute maternal diseases in the critical period of the two examined CA,
b. the investigation of chronic maternal illnesses,
c. the analysis of teratogenicity of the taken/given medications to the mothers in the critical period of the two examined CA,
d. the effect of pregnancy vitamins (especially folic acid) on the emergence of different CAs,
e. the processing of variables referring to the parents' social situation: the mother's age, marital status, occupation.

The Main Principles of Data Analysis

When analyzing the CAs the epidemiological principles must be taken into account, and all efforts should be done in order to limit interferences. This was the main ambition when planning the study, and also in the statistical analysis. Before the analysis we had to find an appropriate solution to the following problems:

The Main Principles of Data Analysis

When analyzing the CAs the epidemiological principles must be taken into account and all efforts should be done in order to limit interferences. This was the main ambition when planning the study, and also in the statistical analysis. Before the analysis we had to find an appropriate solution to the following problems:

a. How to select the critical period: Before starting the test, it is very important to take into consideration that the different CAs emerged in different periods. Thus first and foremost the exposures should be examined in the "critical" period.

b. The analysis and management of the types of so-called distorting effects:

The distortions are systematic errors of the estimated parameters, which can be classified as follows: sample selection bias (selection bias), distortion of information (information bias) and disruptive effect (confounding).

The sample selection bias can occur because of the selection of subjects and their different participation rates may.

The relationship between the examined exposure and the CA may be different among participants and those who were left out from the study, and this can lead to the distortion of the results. In the data which were the base of the research, the participation rate differs between the cases and the controls, as while the not responding CA cases were visited by regional district nurses, this recording was only possible in 800 control families.

One of the Hungarian validity studies showed that concerning the most commonly used drugs there was no great difference between the responding and the non-responding controls.

In case the classification of the exposure or the outcome is wrong, it can lead to a systematically under- or overestimation of the results, giving an exposure bias at the end.

There are several options are available to prevent a recall bias. We can use only prospective data or we can make an abnormal control group.

In the HCCSCA memory distortion was minimized with the use of prospective data and / or an abnormal control group.

The distractions can change the size of the relationship between the exposure and the outcome in relation to the true value.

The third requirement is that there cannot be intermediate factors, or more precisely, the relationship chain between the exposure and outcome may not include it.

The disturbing effects can be blocked out already in the planning phase, for example by fitting the controls, by restrictions or by randomization.

In our analysis we used stratified analysis, or multiple (multivariate) regression models to block out the impact of the known compounding factors, which resulted a corrected estimate.

The selection of the reference group:

The decision essential of the modern epidemiological studies is the choosing of the appropriate reference group.

When examining the casual relationships of CA we can choose from several options.

One option is choosing a "healthy" sample - born without CA - (healthy controls), which represents the population from which our cases come from. The greatest difficulty for this reference group is the distortion of their memoirs.

Another option can be a Down's syndrome affected abnormal group as a reference. Here the memory distortion makes no difference between the case and the control groups, and the genetic origin of the Down's syndrome is proven.

In case the research focuses on the examination of a specific disorder, the memory distortion can be reduced with choosing a control group that includes all the other CAs beside the examined disorders.

In the required analyses a healthy control group and a Down syndrome in control group (abnormal control) were used.

Raw and adjusted odds ratios:

In the data processing it is very important to filter out the possible disruptive effects and one of the opportunities is the use of multiple regression models. Crude estimates do not give exact values, because many other variables can affect the test exposure - outcome relationship, thus the correction is a need. The logistic regression model used in the research of the different causes of disease provides this opportunity. This allows for an adjusted /

standardized (adjusted) value instead of raw (crude) risk, which already eliminates the effects of the confounding factors. In analyzing the exposures always the adjusted odds ratio was estimated, taking into account the variables that might be confounding factors in that particular case.

Matched (conditional) or non-matching (unconditional) controls:

When matching the cases with the controls we select the data (for example, the same age, gender, etc.) on the basis of variables in a predefined thoughtful manner. When performing the alignment attention should be paid to the matching variables that they may not be the risk factors to be tested, but they should be the output risk factors. The goal of matching is the handling of the disturbing factors. Namely, in the course of this method the risk assessment is carried out according to the layers of the matching of the variables separately, thus we can filter out the disturbing effects.

In any case the alignment should be retained in the model, as it is does not result a biased estimate.

However, if the output variables are rare, and the number of exposed is small, it is conceivable that the aligned model is not applicable.

The non-aligned model should be selected instead while the effect of the disruptive effect of the alignment variables should be taken into consideration, as we did in the HCCSCA as well.

The problem of the multiple comparisons (multiple comparisons):

The researchers' opinion is different concerning the importance of the problem of multiple comparisons.

In case we examine more multiple outcomes or more exposure analysis at the same time we should be aware of the distortions resulting from multiple comparisons.

If we accept a value of p 0.05 level as the lowest limit of significance, the amount of 0.05 concerning the development CA out of 20 exposures can be the result of coincidence.

To eliminate these biases of the results there are several options.

One of the simplest and most frequently used method is the Bonferroni correction, in which the p-value is decreased to transfer the limit from the used p = 0.05 to p = 0.05/n (where *n stands for* the number of examined comparisons).

The examining of gene-environment interactions and other forms of interaction:

The analysis of the CAs covered the examination of the genes in the past decades, so when examining the causes of the disease it is very important to consider the effects on gene-environment interactions.

In our study, however, only the parents' and siblings' data were available concerning the examined CAs, and because of the relatively low recurrence risk these data did not allow to take the effects of genetic factors into account.

The statistical analysis of the data:

The statistical analysis of the data has mainly done by the help of the SAS 9.1 (SAS Institute, Cary North Carolina, USA) program package.

We used the PROC LOGISTIC, and the PROC PHREG procedures to calculate the risk estimates.

First, the acute and chronic maternal disease incidence was calculated in the case and control groups particularly specifying the acute diseases in the critical period of the relevant CA.

Comparison of acute and chronic maternal illnesses incidence of CA and control groups multiple logistic regression model was maternal age, sequence of birth, mother's occupation and marital status adjusted odds ratios (abbreviation: OR) and 95% confidence intervals (used abbreviation: CI) can help.

We calculated the adjusted OR and the 95% CI for the maternal age, the order of birth, the mother's occupation, sometimes the taken folic acid / multivitamin.

The effect of medicines taken by the mother in the tested critical period of CA was examined by creating frequency boards.

Subsequently, we estimated the raw, then the adjusted OR and the 95% CI in logistic regression models.

To carry out the statistical analysis, we assessed those maternal diseases in our study that were available in sufficient numbers in the database. The results were considered significant in the case of $p < 0.01$.

RESULTS:

A. Hypospadias

1. The frequency of isolate hypospadias (IHS) cases at birth in the HCAR in the 1970s, 1980s, 1990s, 2000s was 1.59, 2.24, 2.07, 2.53 per 1000 births, while the average was 2.04 / 1000. All these data vividly demonstrate the frequency dependence of the IHS on the diagnosis practices and / or the reporting discipline.

However, if we want to verify the authenticity of the changes or just their artefacts' nature, we need to know the IHS 'real' frequency.

In a previous study10203 new-borns were examined on the first day of their life by a specialist obstetrician and neonatologist and 44 new-borns were diagnosed with IHS.

We accepted this 4.31 / 1000 (95% CI: 2.51 to 8.82 / 1000) as a reference value.

From that perspective the IHS increase in frequency detected after the 1970 up to 2005 can only be explained by the more complete notifications, that is it does not seem to be a true increase of incidence.

In contrast, the frequencies between 2005 and 2010 significantly exceeded the reference value, so it seems a real accumulation of cases.

However, the IHS frequency value can be dependent on the extent to which the minor anomaly computer glandular or the coronal cases not requiring medical treatment are taken into account.

Between 1997 and 2004, out of 1685 IHS cases of the HCAR in 605 (35.9%) was the IHS type known, and within that 242 (40.0%) had coronal IHS type.

Between 2005 and 2010, 1693 IHS cases were registered in the HCAR in, 533 (31.5%) were specified and 288 (54.0%) of them were coronal type.

So we can assume that the growth rate of IHS cannot be considered real between 2005 and 2010. That is, it cannot be traced back to etiological causes, but merely because of the increased attention the IHS cases, which required medical care, were announced at a higher rate.

In Hungary the IHS cases registered in the HCAR have not been assessed since the 1980s.

The data of the past 50 years offer a unique opportunity for the analysis of IHS frequencies nationwide or population-wide.

According to this I can state that the growth of the earlier detected IHS frequency in Hungary can be related to the wholeness of registration and not to the enforcement of a new disease.

The registered significant increase in the incidence between 2005 and 2010 is due to the increased registration of the glandular coronal cases of minor anomaly which do not require medical treatment.

But all these assumptions should be checked on the basis of the new data of the HCAR.

We evaluated 3038 IHS cases, 24 814 population boy controls and 11 096 malformative (affected by other isolated CA) controls in detail being registered in the HCCSCA.

2. The cases of IHS are more common in twins (1.6% vs. 1.1%; OR, 95% CI: 1.56, 1.15 to 2.12), preterm infants (10.0% vs. 8.4%; OR 95 % CI: 1.22, 1.07 to 1.38), but mainly in low birth weight neonates (13.0% vs. 5.0%; OR, 95% CI: 2.85, 2.53 to 3.22), if we the compare the IHS cases' and population controls' mothers.

In the background of the high frequency low-weight new-borns there can be the foetuses' retardation in growth in the womb.

Thus, the formation of IHS is associated with the foetuses' intrauterine retardation of development.

The question is whether it may also be a common cause or a consequence to these two phenomena.

The monthly frequency of IHS cases confirmed the previously found seasonality of births, and it can refer to a relationship between the regulating hormones involved in the gender differentiation and the length of daylight.

3. The average age of IHS cases' mothers at childbirth was lower than the population controls' mothers (25.2 ± 5.2 vs. 25.4 ± 4.9; p = 0.035), primarily due to a higher proportion of the younger (under 20 years) age group (11.4% vs. 8.6%). This is principally because of the ratio of women giving birth for the first time was greater with the mothers of IHS cases (50.5% vs. 47.9%).

The increased frequency of IHS cases from the first pregnancies may be related to a higher-level of these pregnants' oestrogen.

However, among the mothers of IHS cases the ratio of mothers with three or more births was also higher (16.2% vs. 14.6%).

During the mothers' previous pregnancies of IHS case the spontaneous abortion of did not occur more frequently.

More mothers of IHS cases were not married than the controls' mothers, and their occupational dispersion referred to a lower social status.

While the ratio of intellectuals and administrative workers is lower (31.8% vs. 38.8%), the rate of semi-skilled, temporary workers, and housewives was much higher (35.3% vs. 27.9%). So the mothers of IHS cases were in a more disadvantageous social position and this may be related to their above-mentioned younger age and the early first and three or more births.

4. Int the pregnancies of the IHS cases ending with birth among the complications of the pregnancy the pre-eclampsia / eclampsia and the gestational diabetes was more common, while in the early stage of the pregnancy the severe sickness, nausea and vomiting which required treatment was less frequent.

It is possible that the preeclampsia also plays a role not only in the retardation of weight gain of the IHS cases, but also in the aetiology of IHS, similarly to the gestational diabetes.

In contrast, it is proven that the sickness, the severe nausea and the vomiting in a need of treatment, is the indicator of a strong placenta and thus it indirectly protects against some CAs.

Therefore this rare complication of pregnancy may be related to a higher risk of the IHS, as well.

5. In the studied pregnancies the acute infectious diseases were not more common in the mothers of IHS cases than in the mothers of the population controls.

6. In turn, two chronic diseases could be associated to a higher risk of IHS.

One of them was the epilepsy (OR, 95% CI: 1.97, 1.07 to 3.61), but two additional explanations are needed to the evaluation of this mother's disease.

On the one hand the comparison of the IHS cases and with the malformative controls did not show such association.

Namely the epilepsy and the related treatment may cause other CAs too; therefore the effect causing IHS is not specific.

On the other hand, this association basically caused the antiepileptic valproate, which may be related to the agonism of the gonadotropin-releasing hormone of this product.

At the same time, antiepileptics cause specific CA syndromes in general, though it seems that they can cause isolated valproate CAs, like the IHS.

We could find a link between the mothers' medically registered cervical erosions of the IHS cases and their sons' increased risk of IHS when we compared them with the population control mothers (OR, 95% CI: 4.09, 1.84 to 9.12).

With this the results of an earlier Hungarian study was confirmed.

This association cannot be explained with the drug treatments related to the cervical erosion, but it may be related to these patients' higher oestrogen levels, which can play a role in aetiology of both this disease and the IHS.

When comparing the IHS cases with the mothers of the malformative controls the IHS risk of cervical erosions only approached the significance level (OR, 95% CI: 1.9, 0,9-4.4).

7. When examining the teratogenic effect of medicines we confirmed mild human teratogenic effect of the antifungicin oral nystatin treatment in the critical period of IHS (OR, 95% CI: 3.2, 1.6 to 6.2).

The lynestrenol (Orgametril) treatment showed a strong association with the risk of the IHS (OR, 95% CI: 26.7, 8.7 to 81.8), although it was only based on the exposure of this kind of 13 pregnants.

The Limovan^{R} (ethinyl estradiol + ethisterone) (OR, 95% CI: 3.5, 1.6-7.7) and Limovanil^{R} (estradiol + progesterone) (OR, 95% CI: 3.7, 1.3 to 10.7) also showed association with the higher risk of IHS.

However, the dose-response relationship is important; as we found the large dose of Limovan to be teratogenic in this study, while the low-dose ethynilestradiol in the birth control pills we did not.

8. Within the medicine we achieved the most important result with the examination of protective agents.

Namely, these did not show association with the risk of IHS except for one, including the folic acid containing multivitamins.

The exception was the folic acid. The high dose of folic acid use (its mean estimated daily dose was 5.7 mg) was less common in the mothers of IHS cases during the pregnancy than in the controls (OR 95% CI: 0.79, 0,73-0.85).

Considering only the medically documented folic acid taking in the critical period, the protective effect (OR, 95% CI: 0.36, 0.32 to 0.41) of the IHS became even more apparent.

Earlier only in the so called splitting (schisis) CAs, like the neural tube closure CAs was the protective effect of folic acid proven, and it was published in the cases of facial clefts, too; although probably only by using larger doses of folic acid.

The IHS is the closure of the urethral plate CA, too; therefore, presumably the protective effect of the folic acid may also refer to certain common origin in the case of these CAs.

Anyway, as far as I know, this would be the first and foremost possibility to prevent the IHS cases.

9. The IHS is of multifactorial origin, the polygenic genetic predisposition can hardly be reduced; moreover we can expect an increase due to the v-selective impact of the recently introduced high-performance treatments (artificial sperm entry, in vitro fertilization).

Therefore it would be very important to know the polygamous disposition provoking harmful external factors as by curbing them the incidence of IHS cases may be reduced.

Moreover, we can even offer protection against the development tendency by the suppression of the polygamous disposition maybe by some conscious influence like a higher dose of folic acid treatment.

B. Cryptorchidism

1. The increasing frequency of the isolated undescended testis (IUT) cases was detected in the 1970s and the 2000s on the bases of the data of HCAR.

We carried out a critical assessment of the data over the past 50 years (between 1962 and 2011) and on this basis it cannot be stated that in Hungary over the the IUT live birth rate increased in the past decade.

Because if we compare the IUT frequency values to the real frequency signal of reference value (6.86 ± 0.62 / 1000) in the HCAR, we can see that the registered rate was significantly lower.

The previously reported rate increases can be traced back to the more complete notifications.

We evaluated in detail 2052 isolated real ("true"), UT (ITUT) cases recorded in the HCCSCA material, 24 814 population boy controls and 12 082 malformative (others hit by isolated CAs) controls.

I would like to put forward that the most important findings of our investigation is that the need of the separation of the IUT at birth and the posnatalis ITUT cases diagnosed 3 months after birth has been established because of their fundamentally different disease origins.

In a significant proportion of IUT cases at birth the completion of the down sprain of the testicles beginning in the utero can be expected, actually it is about a minor anomaly which does not require medical treatment.

Their typical occurrence can be observed in preterm infants, so it is usually detected in prematurely born twins more frequently. In contrast, the ITUT is a real CA, which is characterized by the retardation of intrauterine weight growth.

2. In the ITUT cases the ratio of premature infants (12.5% vs 8.4%; OR, 95% CI: 1.56, 1.36 to 1.80) and low-weight (LBW low birthweight =) (12.5% vs 5.0%; OR, 95% CI: 2.72, 2.36 to 3.14) was 1.5 times and 2.5 timeshigher than population control values reflecting the total population values. The born to time, but weighing small neonates (small for gestational age = SGA) frequency (6.0% vs. 1.9%) was 3.2 times more common in the ITUT cases.

The frequency of the ITUT twin cases was slightly higher (1.5% vs. 1.1%; 0.39, 0.95 to 2.03) but did not reach the level of significance, compared to the IUT cases, where a significant surplus of twins was noticed.

The birth of ITUT cases showed no seasonality; compared with the IUT cases previously found typical monthly changes of frequency.

Comparing the bilateral cases with the unilateral ones, these CAs are more severe manifestations, which appear in the birth details, as well.

3. The mothers' average age of ITUT cases at birth did not differ from the age of the mothers' of the population controls. Yet the rate The ITUT cases of birth mothers of the average age of the population did not differ from controls mothers value. Yet among the ITUT cases the mothers had a higher rate of those under 20 years (10.7% vs. 8.6%) and people over 30 years (21.5% vs. 18.9%). among the mothers of the ITUT cases under 20 (10.7% vs. 8.6%) and over 30 (21.5% vs. 18.9%) was higher.

The average order of chilf birth was higher with the mothers of ITUT cases (1.9 ± 1.2 vs. 1.7 ± 0.9, $p < 0.0001$), as the proportion of those who gave birth to three or more children was higher with the mothers of ITUT cases than with the population controls (21.6% vs. 14.6%)

The proportion of unmarried women was higher among mothers of ITUT cases.

The rate of intellectuals and administrative employees was lower (29.5%) among the mothers of the ITUT cases, than with mothers of the population controls (38.8%).

In contrast, proportion of the semi-skilled, unskilled workers and the housewives was much higher among them (39.0%) than in the reference group (27.9%).

In Hungary, most of the housewives belong to the lower social classes. All these figures refer to a lower socioeconomic status of the mothers of ITUT cases.

According to this we can establish that the ITUT cases are those pregnant women's children who live in difficult social conditions with higher birth order.

4. The complications of pregnancy ended in birth were not more common in the ITUT cases than the ones found in the IHS.

5. The acute infectious diseases were not more common in the mothers' pregnancies of ITUT cases than in the cases of the population control group.

6. However there was a chronic disease, which showed association with a higher risk of ITUT. The preconceptional endometriosis was more common in the mothers of ITUT cases than in the population controls (1.9% vs 0.8%; OR, 95% CI: 2.42, 1.71 to 3.42).

This association essentially remained when comparing the malformative controls.

Therefore it seems to be a specific effect.

The role of the oestrogens is important in the development of both the endometriosis and the ITUT, so in their etiology the common cause has to be searched.

7. The investigation of drugs indicated of a possible etiological role of a hormonal preparation in the origin of the ITUT disease.

The dihydrogesterone treatment during early pregnancy increased the risk of ITUT in the boys in almost 3 fold (OR, 95% CI: 2.75 (1.04 to 7.28) comparing to the population controls.

This association was confirmed when compared with the malformative controls (OR, 95% CI: 2.46, 1.10 to 6.98).

Several authors reported about the role of the synthetic progesterone derivatives in the origin of the male genital CAs.

However, due to dihydrogesterone ITUT risk it did not arise.

When evaluating this potential etiology, it should be noted that although the hormone treatment was medically documented in the pregnancy care book, this association was based on only five pregnants.

8. Taking high doses of folic acid during the pregnancy reduced the incidence of the ITUT cases comparing to the population controls, especially if we evaluated the medically documented takings in the critical period of the ITUT (15.3% vs. 32.7%; OR, 95% CI: 0, 46, 0.30 to 0.62).

However this protective effect was not detected when comparing the malformative controls. So we cannot speak about a specific ITUT protective effect.

9. The ITUT multifactorial originated polygenic genetic predisposition can hardly be reduced because of the kontraszelektiv effect of the recently introduced high-performance treatments (artificial sperm entry, in vitro fertilization); we can rather expect an increase of it. That is why it is very important to know the susceptibility of the polygon-provoking harmful external factors, as by curbing them the incidence of IHS the cases would be reduce. Therefore, the uses of sexual hormone preparations deserve consideration.

It would be very important to strengthen the preventing effect of the the higher dose folic acid treatment against the potential ITUT, as this would require a conscious application of these protective agents at the end of the pregnancy, too.

SUMMARY:

1. Hypospadias

It was found that:

- In Hungary, the prevalence of real IHS at birth did not increase in the examined 50-year period based on a critical evaluation of IHS cases registered in the HCAR.
- The IHS is more prevalent in children with mothers giving birth for the first time, with twin births, with premature infants, and in addition, especially with low-birth-weight children, and it can be explained the restricted fetal intrauterine growth.
- With mothers of IHS cases the gestational diabetes and the preeclampsia incidence was demonstrably higher
- During pregnancy, severe nausea and vomiting was lower with the mothers of registered IHS cases.
- There is a greater risk of HIS development when it is detected during the critical period of maternal cervical erosion, as well as with treatment of oral valproate, nystatin, linestrenol, ethynilestradiol and ethynilestradiol.
- There is correlation between the higher dose folic acid and the decrease of the risk of IHS. However, this analyzis is based on the observation data, thus it needs to be strengthened.
- The maternal Socio-demographic variability of the IHS birth does not affect significantly its prevalence.

2. Cryptorchidism

It was found that:

- In Hungary, the birth prevalence of IUT showed no real growth in the studied 50-year period (1962-2011). The growth registered in the VRONY difference is a significant difference in the frequency values, which can be explained with the changes in obligation of notification.
- Among the under 20 and over 30 mothers the rate of the ITUT cases was higher
- The average birth order of mothers of the ITUT cases was higher, the proportion of unmarried women was higher among mothers of the ITUT cases,
- The socially disadvantaged mothers of the ITUT occurrence more frequently.
- There is a possible correlation of the ITUT with the maternal endometriosis dihydrogesteron treatment during the pregnancy. This statement is yet to be confirmed
- The frequency of the acute infectious diseases during pregnancy with mothers of the ITUT cases was not greater comparing to the control group.