Intrahepatic cholestasis of pregnancy : a retrospective case-control study of perinatal outcome in the University Hospital of Split

Nunhofer, Verena

Master's thesis / Diplomski rad

2018

Degree Grantor / Ustanova koja je dodijelila akademski / stručni stupanj: **University of Split, School of Medicine / Sveučilište u Splitu, Medicinski fakultet**

Permanent link / Trajna poveznica: https://urn.nsk.hr/urn:nbn:hr:171:483003

Rights / Prava: In copyright / Zaštićeno autorskim pravom.

Download date / Datum preuzimanja: **2024-03-29**

SVEUČILIŠTE U SPLITU
MEDICINSKI FAKULTET
UNIVERSITAS STUDIORUM SPALATENSIS
FACULTAS MEDICA

Repository / Repozitorij:

MEFST Repository

zir.nsk.hr

UNIVERSITY OF SPLIT

dabar
DIGITALNI AKADEMSKI ARHIVI I REPOZITORIJI

UNIVERSITY OF SPLIT

SCHOOL OF MEDICINE

Verena Nunhofer

INTRAHEPATIC CHOLESTASIS OF PREGNANCY : A RETROSPECTIVE CASE-CONTROL STUDY OF PERINATAL OUTCOME IN THE UNIVERSITY HOSPITAL OF SPLIT

Diploma thesis

Academic year :

2017/2018

Mentor :

Assoc. Prof. Marko Vulić, MD, PhD

Split, September 2018

UNIVERSITY OF SPLIT

SCHOOL OF MEDICINE

Verena Nunhofer

INTRAHEPATIC CHOLESTASIS OF PREGNANCY : A RETROSPECTIVE CASE-CONTROL STUDY OF PERINATAL OUTCOME IN THE UNIVERSITY HOSPITAL OF SPLIT

Diploma thesis

Academic year :

2017/2018

Mentor :

Assoc. Prof. Marko Vulić, MD, PhD

Split, September 2018

TABLE OF CONTENTS

1. INTRODUCTION 1

1.1. Intrahepatic cholestasis of pregnancy 2

1.2. Etiology and pathogenesis 3

1.3. Diagnosis 5

1.4. Pharmacological management 8

1.5. Non-pharmacological management 10

1.6. After birth: follow-up and counseling 12

2. HYPOTHESIS 13

3. MATERIALS AND METHODS 14

4. RESULTS 16

5. DISCUSSION 21

6. CONCLUSIONS 31

7. REFERENCES 33

8. SUMMARY 38

9. CROATIAN SUMMARY 40

10. CURRICULUM VITAE 43

1. INTRODUCTION

1.1. Intrahepatic cholestasis of pregnancy

Intrahepatic cholestasis of pregnancy (ICP) is a unique hepatic disorder characterized by mild to severe pruritus and disturbed liver function tests. After viral hepatitis, it is the second most common cause of jaundice in pregnant women (1,2).

A wide variation of incidence in different regions globally has been observed, being the highest among pregnancies in Chile, Bolivia, and Scandinavian countries; however, a declining trend was noticed here, with more recent reports of prevalences between 1.4 and 4.0% in Chile and 0.1-1.5% in Scandinavia. Although there is no definitive explanation for this drop, the aforementioned differences may suggest a geographical and seasonal environmental influence (2,4). As outlined later, variations in the mother's serum levels of zinc, selenium, and copper, explainable with differences in dietary intake, seem to be noteworthy in this context (1).

Obstetric cholestasis typically appears during the late second or the third trimester. It is characterized by maternal signs and symptoms for which alternative causes could be ruled out: pruritus of varying severity without skin rash is considered the cardinal symptom, characteristically affecting the palms and the soles of the feet; patients may also suffer nausea, vomiting and upper abdominal pain. The diagnosis is established with serologically proven abnormal liver function tests and additionally or alternatively an increase in bile acid levels, the latter being the laboratory parameter with the highest importance during workup (1,3,4,5). The condition usually resolves within a short time after delivery, typically within a period of 48 hours (1,3).

Although the morbidity of mothers affected by ICP consists mainly of pruritus and an increased risk of postpartum bleeding, their prognosis is good. Fetal morbidity and mortality, however, appear to be markedly increased in pregnancies affected by ICP. A number of complications can arise as a result of ICP, including preterm delivery (spontaneous or iatrogenic), fetal distress, meconium staining of amniotic fluid and death in utero (1,3).

The aforementioned characteristics of ICP coupled with a remarkable prevelance makes the condition discussed a matter of clinical importance and relevant for current day research.

1.2. Etiology and pathogenesis

Even though the exact etiology for ICP is unknown, research sugests it to be multifactoral. This includes factors such as hormonal inbalance, genetics, environment and even inflammatory mechanisms appear to contribute to the pathogenesis of the condition investigated (1,6).

1.2.1 Hormonal Influence:

Among steroid hormones, increased plasma estrogen and progesterone are proposed to play a role in the development of ICP, which can be supported by multiple observations: The condition appears during the third trimester, is more prevalent in pregnancies with multiples and can feature complications similar to those in women using contraceptive medication with high estrogen content. One can assume that, because of those higher estrogen values, a vicious cycle of impaired sulfonation and transport of bile acids, resulting cholestasis and increased oxidative stress on hepatocytes emerges. Furthermore, a disturbance in the elimination of progesterone is discussed by some authors as an underlying pathophysiological mechanism. Explanations include errors in metabolic and secretory mechanisms with a possible interplay of overproduction of reduced hormone metabolites and malfunctioning or overloaded transport systems (1,2,6,7).

1.2.2 Genetic Influence:

The possibility of genetic predisposition to intrahepatic cholestasis of pregnancy is suggested by various factors, including clustering of that condition and other diseases of the biliary system in families, differences in prevalence according to ethnicity and geographical region and genetic mutations, even though it should be emphasized that the latter could be proven only in a small proportion of cases (1,2). The theory is that genetically determined changes in transport mechanisms of hepatocytes and cells of the biliary tract finally lead to their malfunction. In this context most commonly mentioned is the canalicular phospolipid export pump MDR3, coded in the gene ABCB4; due to mutations the pump seems to be expressed in reduced number in the cellular membrane which causes a lower concentration of phospholipids in bile (1,2,5,8). This phenomenon comes into the picture of several clinical conditions: ICP, progressive familial intrahepatic cholestasis (PFIC) type 3, juvenile cholelithiasis, neonatal and drug-induced cholestasis (8). Therapeutic relevance of this circumstance lies within the aspect that that the abnormally low expression of the MDR3 pump can be stimulated and enhanced with drugs to a certain degree (9). Also on examination

of placentas of affected pregnancies, an insufficiency in the expression of bile acid transporters was detected. Furthermore, based on placental genetic profiles of some cases of ICP, it can be assumed that also the immune system and the regulation of angiogenesis are relevant for the emergence of the pathology discussed here (1). Still, one should be aware of the fact that genetic mutations such as the ones just explained above can be proven only in a very low proportion of patients suffering obstetric cholestasis (2).

In addition, a significant incidence of ICP in hepatitis C – positive pregnant women has been described. However, also here the correlation between these two diseases is yet to be established (8).

1.2.3 Environmental Influence

Apart from the above mentioned and some other contributors, also factors from the environment seem to play a key role in the development of intrahepatic cholestasis of pregnancy, especially in women with hereditary predisposition. Studies have found that the disease seems to appear more commonly during the winter months. This has been attributed to the lower availability of selenium, a cofactor in metabolic processes in liver cells, in seasonable diet, next to the diminished dietary intake of zinc during this period of the year. In the same season-related context the connection of ICP with lower serum levels of 1,25-dihydroxy vitamin D seems explainable. Levels of copper, on the other hand, were found to be increased (1,10).

Not any less complex than the etiology of ICP are the pathophysiological circumstances of this disease which bare a risk for the fetus in addition to the mother.

A combination of distinct factors seems to lead to hostile conditions for the fetus alongside other complications. According to literature, increased flow of bile acids from the maternal to the fetal compartment coupled with the limited capacity of the fetus to eliminate those and functional changes in placental tissue coming down to the level of cell signaling play a role (1,11). The consequently abundantly incurred bile acids result in multi-system toxic effects: They are, on one hand, already in low concentrations hepatotoxic in the fetal compartment and induce apoptosis in the fetal liver (1,10,11). On the other hand, these bile acids have a cardiotoxic effect on the fetus and can therefore cause dysrhythmias and even sudden intrauterine death (7). Apart from the fetal myocard, the increased flow of bile acids through the placental vasculature exposes also this tissue to oxidative stress. Accordingly it was observed that bile acids have a vasoconstrictive effect on chorionic veins in vitro as well

as umbilical cord vessels. Those two mechanisms affecting heart and placenta can provide an explanation for acute fetal anoxia and consequently enhanced peristalsis, meconium staining, the inhalation of the same and resulting neonatal death (1,7,11,12). Moreover, even though the exact chain of events of preterm labor requires further exploration, also this pregnancy complication might be explainable with results from in-vitro research: It has been shown that elevated bile acid levels stimulate myometrial contractions and enhance the activity of oxytocin, probably by inducing increased expression of oxytocin receptors in human myometrium. Additionally, it is assumed that a modified and enhanced prostaglandin synthesis and secretion is a contributing factor to premature labor (1,6,8,12).

Although in literature emphasis traditionally has been given to the risk on the fetus while stating that there is no serious danger resulting from ICP for the mothers, the potential maternal risk factors pre – and postpartum should not be neglected. One such example is the risk of hemorrhagic complications such as postpartal bleeding resulting from a deficiency in vitamin K, increasing fetal as well as maternal mortality (1,10). Furthermore to mention is the fact that women affected by cholestasis of pregnancy have an increased risk for the development of other diseases of the hepatobiliary system such as hepatocellular carcinoma. Also a more frequent appearance of cardiovascular conditions has been described, as well as of diseases mediated by the immune system such as Crohn's disease and diabetes (10).

1.3. Diagnosis

The diagnostic workup of intrahepatic cholestasis of pregnancy is based on a suspect clinical presentation and supported by various laboratory tests. According to diverse current guidelines such as the Clinical Guideline used in South Australia or the Management Guideline provided by NHS Worcestershire Acute Hospitals in England, the way to a diagnosis generally includes the following:

In every pregnant woman presenting with pruritus, serum bile acid measurements and liver function tests are undertaken. In case of abnormalities in these values, a differential diagnostic evaluation for viral infections, autoantibodies including those against smooth muscle or mitochondria, and coagulation studies need to be performed. Alongside these laboratory tests abdominal ultrasound studies to examine the hepatobiliary system is

appropriate especially where abdominal complaints or unclear laboratory results are present. The diagnosis is finally obtained on the basis of laboratory work (3,4,13).

To date there is not a single and independent diagnostic test for ICP (4). Therefore the diagnosis has to be made on grounds of the synopsis of different laboratory parameters and after exclusion of alternative causes for the clinical and laboratory abnormalities. It should be pointed out that reference values specific for pregnancy need to be applied. However, also here a consensus about aspects such as acceptable ranges of values and circumstances for blood sampling are missing (4,14).

As already mentioned, the laboratory test of bile acids in the serum has a high sensitivity and therefore the best diagnostic value, especially in patients with pruritus but still normal transaminases (13). Their elevation can also markedly aid distinction from other pregnancy-specific liver diseases, for example, hyperemisis gravidarum, HELLP ("hemolysis", "elevated liver enzymes", "low platelets") syndrome or fatty liver of pregnancy, in which this parameter stays within normal limits. (5) Regarding bile acids, some studies attribute high significance especially to an increased ratio of cholic acid and chenodeoxycholic acid (CA/CDA), however, other papers negate a diagnostic advantage (2,13,14). A variation of limit values is used and debated; this is accredited to a number of factors such as fasting state during blood drawing, the technique used in the laboratory and the gestational age at the time of diagnosis (4,10,12,15). Bile acid values from 10 to 15 µmol/L are considered diagnostic (2,3,10,12). Values of 40 or also 50 to 70 µmol/L are associated with an increased fetal risk. Therefore serum bile acid values can be viewed as diagnostic as well as prognostic (5,10,14).

Next to the above mentioned, transaminases can provide valuable clues in diagnostic processes regarding ICP. Accordingly, the alanine aminotransferase (ALT) value is regarded as very sensitive, and its elevation indicating cell damage in liver tissue should be alarming and lead to further investigations (13). However, one needs to consider that this parameter is within normal limits in a third of all cases (14). Consequently, in literature an increased value is at times not considered diagnostic (4).

The same is true for alkaline phosphatase (ALP) which rises in case of pregnancy cholestasis, but as well only with little diagnostic importance. The reason for this is that this enzyme is also produced in placental and bone tissue; therefore, an increase in serum concentration does not necessarily need to indicate a disease (13,14,15).

Literature shows a great disagreement about whether an increase in γ-glutamyl transferase (γ-GT) in ICP is characteristic for the condition at all. Unchanged values are being reported, but also slight or severe elevation (2,5,14,16,17). Relevance lies in the fact that a higher plasma value is suggestive for MDR3-mutations, which, however, is currently not tested routinely (13).

Only in 15 – 20 % of all cases of ICP, bilirubin in serum is elevated. However, it can become apparent in urine (13).

The missing consensus regarding reference values contributes to the fact that intrahepatic cholestasis of pregnancy still is a diagnosis of exclusion (14).

Generally it must not be forgotten that the condition may not immediately become evident in abnormal laboratory values as some pregnant women may suffer the characteristic itching but still have normal liver function test results. In these cases a repeat measurement is required after one or two weeks (4,15).

The differential diagnosis is broad-ranging and includes diagnoses that can be specific for pregnancy or exist independently from it (8). Accordingly, by anamnesis one needs to exclude causes like medicaments or alcohol abuse. Additionally, serological tests should be performed to rule out hepatitis caused by agents such as hepatitis virus A, B and C or also as consequence of infections with cytomegalovirus (CMV) or Epstein-Barr virus (EBV). Also other, genetically caused hepatopathies can present with a similar clinical picture, suspicion in those cases is raised by icterus and / or pruritus as adverse reaction to oral contraceptives or as a problem in childhood. Also autoimmune conditions affecting the liver should be taken into consideration, especially when the family history of the patient is positive. Other reasons for pruritus can range from polymorphic eruption of pregnancy to dermatologically alternatively explainable eczemas, atopies or pemphigoid (4,8,15). In early or otherwise atypical cases, pre-eclampsia and fatty liver of pregnancy as reasons for diminished liver function should be investigated, as mentioned earlier (15).

1.4. Pharmacological management

ICP is a condition that requires management. The aim is to alleviate maternal symptoms and to reduce the risk to the fetus as much as possible, pharmacologically mainly by means of the reduction of cholestasis and of bile acids in serum (1,18).

Currently considered the first line treatment option is ursodeoxycholic acid (UDCA), a physiological component of human biliary secretions (18). There are several theories about the mechanism of action of UDCA which, besides other aspects, concern the reparation or correction of bile acid transport and consequently the enhancement of transplacental excretion of bile acids, their reduction in maternal serum and amniotic fluid (1,18). After all, those changes may be protective for hepatocytes, bile ducts and the fetal heart. Also, they can contribute to an improved perinatal outcome, not only by their protective effect itself, but also by a consequently longer maintenance of the pregnancy (1,18). However, there is no consensus in current research about this improvement (19). A Cochrane review from 2013 stated that by usage of UDCA pruritus could be mildly diminished. However, in comparison with a placebo group, no statistically significant difference regarding the benefit for fetal distress could be observed (20). There are different dosage recommendations. Generally, 2 x 500 mg per day or 10, 15 or 20 mg/kg/d are prescribed, and an effect in low as well as in higher standard doses is reported (1,18). This was noted on symptomatic as well as on biochemical levels already after two weeks of therapy (21). In general, UDCA has a favorable profile of side effects for mother and fetus, and the use in the third trimester of pregnancy is safe (1). Only mild diarrhea was reported as a potential adverse drug reaction among mothers treated with it (13). After follow-up checks on the children of mothers whose ICP was treated with UDCA that lasted up to 12 years, no negative consequences on longer term regarding growth and development or diseases could be observed (22). Despite all the mentioned advantages and the medication's popularity, it has to be noted though that its use in ICP is still off-label: although its use has been approved for a number of cholestatic hepatopathies, this approval has not been obtained yet with pregnancy cholestasis as an indication (13).

Cholestyramine belongs to the group of anion exchange resins and acts by binding bile salts, diminishing their presence in the enterohepatic circulatory system (1,13,20). Acting in this fashion, it would also bind UDCA and impair its action. Therefore, those drugs should be administered separately from each other if one decides for the simultaneous usage of both (13). Cholestyramine is shown to improve the mother's morbidity, however, not the

biochemical test results or the fetal situation (1,18). According to the aforementioned Cochrane review, sufficient evidence for its usefulness is missing (20). Furthermore, the relief of the patient's symptoms might be only of short duration and their recurrence after about a week after treatment initiation has been reported (18). The use of this substance is considered in very severe cases (17). Cholestyramine does not only bind and eliminate UDCA, it also complexes with fat-soluble vitamins and lessens their enteral absorption (18,23). In this context, especially the importance of vitamin K is emphasized in literature sources, but also the other liposoluble vitamins A, D, and E and a sufficient maternal dietary intake or supplementation of these should be considered (23,24). In case of a prolonged prothrombin time, vitamin K should be administered in a daily oral dose of 10 mg (4). For the reason explained previously, it is recommended to avoid the use of cholestyramine if possible (18).

S-Adenosyl-L-methionine (SAMe) is a substance changing the composition and liquidity of cell membranes and enhancing the metabolism and elimination of hormone metabolites (1). The benefit of its usage, however, is questionable, and there is not enough evidence for its medicative effect: Not only were the results compared with placebo conflicting, also an increased incidence of preterm delivery was reported after usage of SAMe as a monotherapy which is the reason to advise against this way of application (18,20).

Phenobarbital is used in case of ICP to improve pruritus and is effective in it in 50% of all cases (1). This improvement is suggested to be caused by the medication's effect on the central nervous system (18). However, no improvement could be seen in laboratory studies, and also adverse effects such as malformations of the fetus, hypoventilation or diminished intelligence must be pointed out (1,18).

For the treatment of itching, rifampin and antihistamines are in use as well. Because of its sedating effect, the latter ones proved to be helpful especially against nocturnal pruritus (1).

Also dexamethasone in high doses a 12 mg per day is thought to be helpful in the management of ICP. However, solid evidence for this is lacking, and the drug seems to be not very effective in lowering serum levels of bilirubin and bile acids and inefficient in the therapy of pruritus (1,20). Not only the insignificant effect is a point to consider, but also that dexamethasone has important side effects such as insulin resistance in the treated mother (20). Even though this drug is known to lower the incidence of respiratory distress in neonates by

enhancing lung maturation in utero, on the other hand, it is still suggested by at least one source to not use it, or at least not as a monotherapy (20).

In cases not responding to pharmacological treatment, the application of plasmapheresis for the removal of bile acids from the serum or phototherapy with ultraviolet B (UVB) radiation have been reported to be a beneficial treatment option (25,26).

1.5. Non-pharmacological management

Also non-pharmacological management in terms of surveillance and finally the timing of delivery are aspects that are approached with several distinct concepts (13). Generally, active and expectant management are distinguished, and both concepts have advantages and disadvantages.

Regarding surveillance during the time of pregnancy it is worth mentioning that different techniques are used, however, not in a provably effective way. A precise prediction or even prevention of stillbirth seem not to be possible to date (15,27). Surveillance got intensified since many practitioners seem to prefer expectant management, keeping in mind though that an early onset or detection, respectively, seems to be associated with a higher risk for the fetus (13,19).

There is disagreement about several aspects of care for pregnant women affected by intrahepatic cholestasis, starting with the seemingly simple question of how often tests and examinations should be performed. Trying to find an answer to this question, one should keep in mind that death was not the result of a chronically developing problem but of a sudden event (2). Weekly controls are suggested, next to the mother's own continuous monitoring of aspects that also lie within her own perception such as fetal movements, their decreasing intensity or absence (1,4,13,15). This expectant management might take place in an outpatient scenario, as suggested by the South Australian Perinatal Practice Guidelines, as long as serum bile acids are below 40 µmol/l and ALT below 200 units per liter (4). In fact, as mentioned earlier, bile acids are not only of importance for diagnosis but also during surveillance when it is about to decide how to continue the management of the patient (2).

From the 30th week of gestation onwards, next to weekly non-stress tests, assessment of amniotic fluid volume, Doppler sonography of the umbilical arteries and a growth scan are

suggested for surveillance (1). It is notable that the recommendation of the last mentioned measure is in contradiction to the fact that fetal death happens as a sudden event, as explained earlier (2). Other sources propose weekly cardiotocography, furthermore a weekly control of laboratory values including coagulation studies such as prothrombin time (1,13). As a part of active management, amniocentesis in the 36th gestational week for the control for meconium and signs of fetal lung maturity are advised next to close and attentive monitoring, before labor should be induced in week 37 (2). The just described active way of management is supposed to make it possible to continue a pregnancy under close surveillance to the latest possible point in time (29). However, some authors emphasize that there is no indication for the endorsement of active management of pregnancies complicated by obstetric cholestasis (30).

After all, birth is talked about as the only method that can prevent fetal mortality definitely (27). When timing birth, for each patient the risk that early delivery carries needs to be traded off individually against the dangers of continuing the pregnancy (1,2,13).

There are various opinions about the best point in time to induce labor. Some experts suggest this for as early as week 36, or, in case ICP is diagnosed later, immediately at that point, and justify this with the apparently then lowest risk of fetal, neonatal and infant mortality; at the same time though they point to the morbidities of premature infants of this gestational age (27).

Often, the 37th week of gestation is suggested as the most favorable moment for delivery to prevent a stillbirth afterwards (15,31). However, this point in time might not be optimal for twin pregnancies (7). Reference is made to biochemical tests as crucial for the decision to induce birth: In case of bad or worsening laboratory values, or more specifically, in case of bile acid values of 40 µmol/l or higher, induction of labor should be considered (3,32). For the period of the 37th or also 38th week of pregnancy as optimal time window for induction of labor in case of ICP solid evidence is lacking (1,10). Nevertheless, some specialists prefer week 38 due to the aforementioned unpredictability of stillbirth after this period (4,27). In case of favorable and stable laboratory results, also the planning of birth only in week 39 is mentioned in literature (3).

Summarizing, one can note that also for timing of labor in case of obstetric cholestasis there is no consensus and accordingly also no clear guideline (2).

1.6. After birth: follow-up and counseling

The circumstance that obstetric cholestasis is a diagnosis confirmed only after birth makes following up the mother for a couple of weeks reasonable (3).

After one or two days, itching should disappear, jaundice after approximately one week, and also bile acid and liver function test values should be within normal ranges within a week to ten days postpartally (3,4). Normalization of liver values may take six to twelve weeks though; if they are too high after this period of six weeks or longer, the patient should be referred to a gastroenterologist (3). This is because such test results may indicate an underlying liver disease (4). For this case, a screening for liver cancer and other diseases such as autoimmune disorders is recommended (33).

Furthermore, counseling regarding a couple of points concerning the time after a pregnancy complicated by ICP appears appropriate. For example, affected women should be informed that, even though pruritus disappeared after pregnancy, it might return in future pregnancies (34). Information about probability of this to happen varies from source to source from 40 or 45 up to 60, 70 or even 90%, respectively (2,3,4.8). Also, skin itching might appear cyclically around the time of ovulation (2). Regarding contraception, women should be advised that oral combined contraceptives can be used as soon as liver test results normalized after pregnancy, however, with the risk of pruritus and elevation of liver enzymes. (10) Knowing of the risk of contraceptive-induced cholestasis, this medication should be discontinued as soon as symptoms occur (8,13). There is no contraindication for breastfeeding after suffering intrahepatic cholestasis during pregnancy (13).

Even though it is claimed that there are no serious long-term sequelae for the mother nor the child (4), it was shown that changes in metabolism of children of affected mothers can occur, becoming evident in the alternations in blood lipid profiles or increase in body mass index (BMI) (33). Also an increased risk for cancer of the hepatobiliary system, cardiovascular and autoimmune disorders in affected mothers was noticed, as well as a connection to other, hereditary diseases of the liver and bile tract, next to a higher risk of the development of obstetric cholestasis in female offspring of affected women (2,4,33).

2. HYPOTHESIS

There is a significant difference in epidemiology and perinatal outcome between pregnancies with cholestasis and healthy pregnancies. We assume a worse perinatal outcome in the case group. In the case group we expect a higher age of mothers, higher BMI of mothers, shorter pregnancy duration, more Cesarean sections, a lower Apgar score, a lower umbilical cord blood pH and a lower weight, length and size for gestational age of the fetus in comparison to healthy pregnancies.

3. MATERIALS AND METHODS

We conducted a retrospective case-control study and used data collected on women who gave birth between 1st of January and 31st of December 2016 in the University Hospital of Split (KBC Split) and their newborns. The information about cases and controls were obtained from handwritten birth protocols in the delivery room and patient files archived in the Department of Gynecology and Obstetrics and transferred anonymized to an Excel table.

Identified as a case of ICP and therefore included into the case group was every woman who presented with a clinical picture matching the diagnosis of ICP and whose serum transaminases were elevated to double of normal. Exclusion criteria were aneuploidy, malformations of the child and multiple pregnancy. Since such cases were not found within the group of women with obstetric cholestasis, effectively none of the patients were excluded.

Included into the control group via a matching process was every birth consecutive to a case of ICP with parity identical to that case. The data investigated included the mothers' age in years, maternal BMI, parity, duration of pregnancy in completed weeks, fetal weight in grams, fetal length in centimeters, Apgar score, umbilical cord blood pH, delivery mode and size for gestational age.

Within the given period, we identified 28 cases of intrahepatic cholestasis of pregnancy and obtained an equal number of controls. However, documentation was partly incomplete in four patients of the control group and four patients in the case group so that in analysis of some variables this number differed: In the case group, we obtained umbilical cord blood pH values in only 23 cases and the mother's BMI in 27 cases. In the control group, the maternal BMI was documented 27 times, the pH value 26 times, and the newborn's size for gestational age 27 times.

For statistical testing we used the program Statistica 12 (StatSoft, USA). Student's t-test was used to test the difference between two numeric variables with normal distribution. Normality was tested using Kolmogorov-Smirnov test.

Tests on categorical variables were χ^2 test and, in case data sets did not meet the prerequisites for it, Fisher's exact test. To examine the difference in two proportions, two proportion Z-test was used. P-values of less than 0.05 were considered statistically significant.

4. RESULTS

Out of 4266 births that took place between the 1st of January 2016 and the 31st of December 2016 in the University Hospital of Split, 56 were included in our study.

Parity was equally distributed in test and control groups of equal sizes (28 women each) as we used this parameter for matching of the two. As shown in Figure 1, in both groups, the documented birth was the first for 11 patients, for 12 patients each it was the second, for 4 patients each it was the fourth, and for 1 patient each it was the fifth time giving birth.

Further epidemiological and clinical characteristics observed in both groups are given in Table 1. In the control group, the mean of the mother's age was 30.43 years with an average deviation from the mean of 4.66 years. The mean BMI calculated in these mothers was 23.53 with an average deviation of 4.99. In the case group, the mean of the mother's age was 30.25 years with an average deviation from the mean of 4.07 years. The mean BMI found in this group was 22.86 with a deviation of 2.58 on average. Values that were normally distributed had an empirical p-value of more than 0.05 each. Evaluations of the difference in values of case and control group gave a p-value of greater than 0.05 and therefore did not indicate statistical significance.

Table 1. Epidemological and clinical characteristics of pregnant women with and without intrahepatic cholestasis

	ICP[a] (N=28)	**Control group (N=28)**	***P****
Mothers' age (years)	30.25±4.07	30.43±4.66	0.879
Maternal BMI[b] (kg/m^2)	22.86±2.58	23.53±4.99	0.535

*Student's t-test
[a] Intrahepatic cholestasis of pregnancy
[b] Body mass index

As aspects of perinatal outcome examined, in the case group, average fetal weight was 3493.57 g with an average deviation from the mean of 551.75 grams. Average fetal length was 50.39 cm with an average deviation from the mean of 1.97 cm. The average pH of

umbilical cord blood was 7.32 with an average deviation from the mean of 0.09. Values that were normally distributed had an empirical p-value of more than 0.05 each. In the control group average fetal weight was 3373.21 g with an average deviation from the mean of 517.02 grams. Average fetal length was 49.75 cm with an average deviation from the mean of 2.01 cm. The average pH of umbilical cord blood was 7.34 with an average deviation from the mean of 0.07. Values that were normally distributed each had an empirical p-value of more than 0.05. Since the p-value at the evaluation of differences was greater than 0.05, no statistical significance could be observed here. The just described findings are shown in Table 2.

Table 2. Perinatal outcome of pregnancies with and without intrahepatic cholestasis

	ICP[a] (N=28)	**Control (N=28)**	***P****
Fetal weight (g)	3493.57±551.75	3373.21±517.02	0.403
Fetal lenght (cm)	50.39±1.97	49.75±2.01	0.232
pH	7.32±0.09	7.34±0.07	0.431

*Student's t-test
[a] Intrahepatic cholestasis of pregnancy

It could be observed that in the case group fewer patients (25 = 89.29%) were pregnant for 37 weeks or longer compared to the control group (27 = 96.43%). Two of the pregnancies in the case group (7.14%) lasted between 34 and $36^{+6/7}$ weeks, compared to none in the control group. One pregnancy on the case group (3.57%) and none in the control group was between 32 and $33^{+6/7}$ weeks long. The control group contained one pregnancy that lasted between 28 and $31^{+6/7}$ weeks (3.57%), in contrary to the control group which contained no pregnancy of such duration. After creating, two groups regarding pregnancy duration, one including pregnancies of 37 and more weeks, and the second one including those that lasted shorter than 37 weeks and testing independence using Fisher exact test, we obtained an

empirical p-value of 0.118 from which we could conclude that differences in pregnancy duration of cases and controls were not significant.

Examining Apgar scores in both groups, all 28 newborns (100%) in the control group obtained one between 8 and 10. However, in the case group this was the case only in 26 (92.86%) of all deliveries, where two cases were observed with a score in the range between 4 and 7 (7.14 %). Independence was tested using Fisher exact test, and we obtained a p-value of 0,245 which led us to the conclusion that the difference in the prevalence of Apgar scores among case and control patients was not of statistical significance.

22 pregnant women with cholestasis (78.57%) had a vaginal delivery in contrast to 18 (64.29%) in the control group. A Cesarean section was performed in 6 women in the case group (21.43%) and 10 women in the control group (35.71%). We used χ^2 test to examine whether there is a correlation between the condition of intrahepatic cholestasis and mode of delivery. Based on the obtained empirical χ^2 value of 1.411 with degree of freedom of 1 and a p-value of 0.235 we can conclude that the type of delivery is not significantly affected by the presence or absence of ICP.

Children whose size was appropriate for gestational age (AGA) were given birth to by 23 women (85.2%) of the control group and 25 women (89.29%) with cholestasis. 2 children (7.4%) each were considered small for gestational age (SGA) or large for gestational age (LGA), respectively, in the control group. In the case group, 10.71% of born children were LGA and none were SGA. Respective p-values over 0.05 indicated statistical insignificance of the just mentioned differences in proportions.

Table 3. Perinatal outcome of pregnancies with and without intrahepatic cholestasis

	ICP, n (%)	**Control, n (%)**	***P***
Pregnancy duration (completed weeks)			0.118 *
<37	3 (10.71)	1 (3.57)	
< 28	0	0	
28-31$^{6/7}$	0	1 (3.57)	
32-33$^{6/7}$	1 (3.57)	0	
34-36$^{6/7}$	2 (7.14)	0	
37 ≤	25 (89.29)	27 (96.43)	
Apgar score			0.118 *
4-7	2 (7.14)	0	
8-10	26 (92.86)	28 (100.0)	
Delivery mode			0.235 †
Vaginal	22 (78.57)	18 (64.29)	
CS[a]	6 (21.43)	10 (35.71)	
Size for gestational age			
SGA[b]	0	2 (7.4)	0.142 ‡
AGA[c]	25 (89.29)	23 (85.2)	0.648 ‡
LGA[d]	3 (10.71)	2 (7.4)	0.670 ‡

*Fisher's exact test
† χ^2-test
‡ Two proportion Z-test
[a] Cesarean section
[b] Small for gestational age
[c] Appropriate for gestational age
[d] Large for gestational age

5. DISCUSSION

While affected mothers traditionally are considered to have a good prognosis, fetal morbidity and mortality appear to be considerably increased in pregnancies complicated by intrahepatic cholestasis (1,3). This and the fact that medical research still has not come to satisfactory understanding of the disease, make the condition discussed here a matter of clinical and scientific interest. Various theories about the pathophysiology of ICP exist, as outlined earlier. All these points may contribute to the circumstance that to date there are several approaches to and no consensus among national and regional guidelines concerning the management of pregnancies affected by cholestasis. Consequently, a review and evaluation of pregnancy outcomes in the University Hospital of Split with the local current practice seemed to be beneficial; identifying differences in cases of cholestasis compared to unaffected control pregnancies could give important hints and could support or point to a need of changing today's approach and management of pregnant women with ICP.

For this study we identified all 28 documented cases in the birth registers of the University Hospital of Split in 2016 (1.1. -31.12.) and compared next to epidemiological data about the mothers (age, parity, BMI) information about the perinatal outcome (pregnancy duration, delivery more, Apgar score of the first minute and umbilical cord blood pH, fetal weight and length and size for gestational age) with those of a control group of equal size. Doing so, we could not find any statistically significant differences in the parameters that were analyzed and will be discussed separately below:

According to various sources such as the ACG Clinical Guideline referring to liver disorders in pregnancy, higher age is counted as a risk factor for ICP, which made it important to us to examine this parameter in our patient group (31). Doing so in our study, we found that women in the case group were marginally younger than women of the healthy control group (on average 30.25 vs. 30.43 years), however, this finding turned out to be of no statistical significance ($p>0.05$). A similar but also insignificant observation was made by researchers in Saudi Arabia who found women with ICP to be at the average age of 29.18 years compared to the 29.86 years of unaffected mothers (35). These results differ from other studies which suggest a link between the age of the mother and prevalence of ICP (8,10,31). In a study focusing on twin pregnancies, for example, mothers with cholestasis turned out to be slightly older (7). A different paper published by authors from Turkey which compared milder and more severe cases of cholestasis according to their outcome even found a direct correlation of higher age and greater severity of the disease ($p<0.05$) (36).

Since age is listed as a risk factor for the development of obstetric cholestasis, as explained above, and regardless of our results obtained from a rather small patient group, it might be advisable to still take it into account and treat older mothers with increased vigilance (10,28,31).

As we used parity as the characteristic to match controls to cases, we could not make an observation about the difference in this variable between diseased and healthy pregnant women, unfortunately. The Clinical Guideline for management of obstetric cholestasis released by the Government of South Australia states that its distribution among women who have not been pregnant before and multigravidae is generally equal (4). On average, women with and without diagnosed ICP who gave birth in KBC Split had two children. In other studies mothers with cholestasis were mostly primiparae (2,7,19). Otherwise cases of ICP were found to have a lower parity compared to the control group; however, this was not a significant observation (35).

Since a high body weight is commonly known to be a risk factor for many adverse health events, among which metabolic diseases and cancers that can as well affect the liver are listed, it was of interest to us to investigate also this parameter in the context of ICP (40). In our study the body mass index before pregnancy of mothers suffering cholestasis turned out to be insignificantly lower than the BMI of mothers that were not affected by this condition. Contradicting this, the results of a different study published in 2017 showed that more severe cases of obstetric cholestasis tended to have a higher BMI before pregnancy (12). However, our findings are in concordance with the results of the study on twin pregnancies in which women with ICP appeared to have a significantly lower pre-pregnancy BMI (22.9±2.9 compared to 23.6±3.2; p=0.017) (7). Likewise, the research conducted in Saudi Arabia mentioned previously states state the observation of an insignificantly higher body mass index in the control group (p=0.34). It is interesting to notice that in their study case as well as control group were, according to World Health Organization (WHO) – definitions, with average BMIs of 29.55 and 30.23 overweight or obese, respectively (35,40). Consequently one might wonder how much of a role in the development of ICP a too high pre-pregnancy BMI plays at all.

Still, dyslipidemia, a common finding in individuals with a too high BMI, i.e. a BMI equal to or greater than 25, has been observed in the context of ICP (11,40). Furthermore, as explained earlier, increased estrogen levels might play a role in the pathophysiology of the disease (1,2). These two aspects support the idea that a high body mass index, which per

definition by the WHO corresponds to a great body fat percentage, in mothers might be a factor favoring the development of obstetric cholestasis (40).

Shortened pregnancy duration is known to be linked to fetal risks and complications. Consequently, the circumstance that preterm birth has been linked to ICP was our motivation to conduct statistical analysis regarding this aspect among the patients of our groups. (1,3) However, our results show no significant difference between women with and without the diagnosis of ICP in the average duration of pregnancy or, more specifically, the incidence of preterm birth which is defined as a delivery before the completion of the 37th week of pregnancy (37). We could observe an insignificantly elevated rate of preterm birth in the case group ($p=0.118$).

Still, the observation of a more common occurrence of preterm delivery was made in many other studies: an article published in 1994 reports a significant ($p<0.05$) "2.8-fold increase in the incidence of premature delivery (<37 weeks)" and also points out that this observation includes a finding of three times as many spontaneous deliveries compared to the healthy control group (38). Also other authors who compared cases of severe cholestasis with healthy pregnancies found that affected women gave birth significantly earlier than women without ICP (25% vs. 6.5%, $p<0.001$); however, in their study sample most preterm births were iatrogenic (39). Iatrogenicity of preterm birth was not examined in our study. Matching these findings, researchers from Istanbul conclude that the level of serum bile acids is in correlation with premature birth (6). Also the figures of the study from 2017 that was referred to earlier show that mothers affected by ICP gave birth earlier with increasing severity of their disease according to their bile acid levels ($p = 0.002$) (12).

These results are in direct contrast to the findings of other research papers such as of a study conducted in Istanbul, Turkey, which could not correlate the degree of derangement of laboratory values at diagnosis of ICP (serum TBA, AST, ALT, GGT and AP) with the gestational age at delivery, however, with a p-value greater than 0.05 this finding was statistically insignificant (19). As well opposing our results but also with a p-value indicating insignificancy, the authors from Saudi Arabia reported a slightly higher incidence of preterm birth in their case group (35).

Remarkable for the studies compared above are the varying and also contradicting reports of the incidence of deliveries before week 37, especially iatrogenic ones. This circumstance might reflect the missing consensus among professionals regarding active or

expectant management strategies and the recommendations in the many different guidelines regarding timing of a possible induction of labor in case of cholestasis of pregnancy that was discussed earlier.

Obstetric situations can quickly become acute and borderline and therefore require the appropriate choice of delivery mode. C-section is often the method of choice in critical situations and valued by many obstetricians because as surgical intervention it makes birth a more controllable process (41). Given the risks that a pregnancy complicated with ICP carries, a higher proportion of Cesarean sections would be expectable in the case group. However, an insignificantly higher proportion of Cesarean sections as opposed to vaginal deliveries in the control group compared to the case group could be observed in the control group in our study ($p=0.235$). This distribution stands in contrast to the findings of other studies. In one the authors report Cesarean section as mode of delivery in 25.9% of their ICP group and 16.9% ($p<0.05$) and explain this higher proportion of C-sections in this group with an insignificantly increased rate of elective procedures (37). Likewise, the incidence of Cesarean deliveries elsewhere was reported to be higher in their case group (19.7%) compared to their control group (14.5%), however with a higher percentage of emergency rather than elective procedures in both groups ($p=0.15$) (35). Also figures published in 2014 showed proportionally more Cesarean sections in the cholestasis group (25%) compared to healthy pregnancies (23%), however, p-values higher than 0.05 indicate no statistical significance (39).

Whether C-sections were elective, as it is not rarely the case in Western medical practice, or necessary procedures in our study population was not subject to our evaluation (41).

Apgar score has established itself as an important step in the standard protocol of the first evaluation of the newborn and makes it possible to get an impression of the neonate's physical condition seconds after delivery which made it a valuable component of our analysis of perinatal outcome (37). All newborns born to mothers of our control group had Apgar scores of the 1st minute between 8 and 10, and in the case group this score was reached by 26 out of 28 neonates. According to the scoring concept proposed by Virginia Apgar in 1953, children with this score are considered to be in "good condition" (42). The difference in this subjectively obtained indicator of perinatal outcome measured in our study turned out to be insignificant.

Also, other sources do not mention a significant difference in Apgar scores between neonates born to affected and unaffected mothers, respectively. However, a comparison between our and their percentages might not be very sound. The reason for this is that their definition of a satisfying score for the newborn seems to be a bit more permissive with a score of only lower than 7 being used to describe a problematic perinatal outcome: One research group reported a score < 7 in the first minute in 7.8% of their cases compared to 7.2% in their control group (37). In the population studied with focus on twin pregnancies, after five minutes, 2.4% of newborns of mothers suffering ICP had an Apgar score below 7, compared to 2.0% in the healthy control group (p=0.565) (7). Other authors who studied a group of ICP patients and put them into groups according to whether there were adverse perinatal outcomes (Group I) or not (Group II) reported insignificantly better Apgar scores after one minute in their second group (8.27±0.9 compared to 7.94±1.1 in Group I) (36). Overall, no significant correlation between Apgar score and the presence or absence of cholestasis could be made.

As an objective numerical value, umbilical cord pH is a parameter that is recommended to obtain during the assessment of newborns at least after high-risk births. Since it is still not ideally reflecting the degree of asphyxia in the fetus, its appreciation in combination with the Apgar score appears reasonable (43). Reference ranges among different researchers and authors for the umbilical cord blood pH vary greatly, and the precise definition of significant acidemia of the fetus remains debatable (44). Overall, professionals seem to agree on a pH < 7.0 to be associated with severe consequences for the newborn if the child does not appear "vigorous" (43,45).

In our study, with average pH values of 7.34 and 7.32 in the control and the case group, respectively, we could not see a significant difference in this parameter. Generally and according to teaching literature, those values > 7.30 can be considered "normal" (41).

In accordance with the suggestion to perform umbilical cord blood pH measurement in births that seem to carry a risk, authors of an article mentioned earlier obtained the value in 21 newborns of the group with ICP and eight of the control group with the suspicion of distress to the fetus during the delivery process, i.e. the indication was given more often in the case group. They could detect asphyxia in a higher proportion of suspicious children of the case group (12 out of 21 cases, or 57%) than in the control group (3 out of 8, or 37.5%) with average values of 7.08 and 7.07, respectively (p-value was missing) that can be categorized as advanced acidosis (35,41).

Other studies rarely considered taking this variable into account to describe perinatal outcome, it does not seem to be performed as a standard test. A reason for this can be that this test, as all laboratory procedures, takes time and is an extra expense. Furthermore it has been described that the umbilical cord pH value has a lot of confounding factors including the timing of sampling and delivery mode (43). Besides, the Apgar score per se as a well-approved routine measure seems to give a sufficiently solid estimation of the newborn's wellbeing.

Birth weight, especially set in relation to gestational age, can be an important indicator concerning fetal risk, morbidity and mortality. Especially a low birth weight or small size for gestational age, respectively, is often the consequence of placental malfunction (37). Since, according to current research, also the placenta might be affected in ICP, it seemed reasonable to include data on fetal growth (weight, length and size for gestational age) as descriptor of perinatal outcome in our analysis (1). Doing so, we found an insignificantly higher birth weight in children born to mothers with obstetric cholestasis compared to healthy mothers. Furthermore, newborns of the case group were a bit longer; however, that difference had no statistical significance either. Our results regarding fetal weight are contrasting those of other research papers: authors of one article could observe the birth weight neonates of mothers affected by ICP to be lower (on average 3049.5g) than in their control group (3357.5g) and with a p-value < 0,001 their result was significant (39). Also in concordance with this, other researchers measured significantly lower weights in their case group (2360±425g) compared to patients of their control group (2459±446g) in their study on twin pregnancies (7).

Fetal length was not measured in other studies.

Newborns of the case group of our study were either of average size or in three cases large for gestational age, compared to the control group who gave birth mostly to average sized children, or, in two cases each, to children that were small or large for their gestational age. This determination was based on population-adjusted diagrams visualizing percentiles as body weight per weeks of gestation. However, the noted differences turned out as of no statistical significance.

A similar trend becomes apparent in the results of researchers who compared the birth weight centiles of their groups with and without cholestasis and detected significant differences. In the mean the neonates of the case group were around the 47.6th centile of size for gestational age and the ones of the control group were around the 40.8th ($p<0,001$). By

more detailed investigation of SGA (<10th centile) and LGA (>90th centile), they could detect a significant higher proportion of SGA deliveries in healthy mothers (16% vs. 11% in cases, p=0.007 after comparison adjusted for confounding factors). Also incidence of births to LGA children was, like in our population, higher in the case group, however insignificantly as a p-value of 0.44 showed (39).

Still, the above mentioned observations of mothers with ICP birthing slightly larger neonates are unexpected, looking at the results of other studies.

In their study involving twin pregnancies, the authors report with 9.9% a significantly higher incidence of newborns that were LGA in their healthy control group, compared to their case group where they obtained this finding in 5.9% (p=0.049) so that they were even referring to "a protective effect on the incidence of LGA". Their comparison of the incidence of SGA was not significant (p=0.247) (7). A different paper as well reported a significant correlation of the incidence of fetal growth restriction (p<0.01) with higher levels of total bile acids and an earlier diagnosis of cholestasis of pregnancy (19). On the other hand, other studies such as a recent one mentioned earlier could not observe such a pattern concerning bile acids and SGA births (12).

The missing congruence of results regarding the association of intrahepatic cholestasis of pregnancy and size for gestational age raises questions about placental involvement in chronically evolving problems during ICP as opposed to acute events such as sudden intrauterine death where placental involvement has been scientifically proposed. It may also motivate to investigate further variables that can potentially affect fetal growth during ICP, such as the mother's stress level, diet or metabolic profile.

A rather small set of data was subjected to statistical testing. Since all cases that we found have been integrated to come to one result per variable, it is not surprising that single exceptional findings could not come to attention but might still deserve it.

Concerning the distribution of suboptimal findings in the different parameters examined (specifically, a short pregnancy duration, low pH, and SGA / LGA), it is noticeable that they did not appear combined in the healthy control group but only single.

This is different in the case group where they can indeed be observed in combination, namely in younger primigravid women that were admitted mostly between September and

November 2016. This matches several epidemiological factors that have been linked to cholestasis of pregnancy in studies and literature:

- One child was born after $36^{5/7}$ weeks and showed an Apgar score of 6 with a pH of 7.211.
- One child was born after $36^{5/7}$ weeks with an umbilical cord blood pH of 7.231.
- One child was born after $32^{6/7}$ weeks with a weight of 1600g, still AGA, the pH value was not recorded.
- Two children had acidotic pH values of 7.190 and 7.273, respectively, and were LGA.

Even though the overall statistical picture is satisfying since the outcome in pregnancies complicated by ICP seems to be as good as the outcome of normal and healthy pregnancies in the University Hospital of Split, these specific findings encourage to continue taking the diagnosis of cholestasis in pregnancy especially serious and handle identified cases with care.

Strengths that can be attributed to our study are the following: The women in the case and control groups were of very similar epidemiological background which gave the observation of perinatal outcome itself a sort of neutral basis. Also, the umbilical cord pH that was mostly missing as a parameter in comparable studies had been obtained for almost all pregnancies.

Weaknesses of our study include a rather small sample size. This can be explained with that we obtained our data observing a period of time that is rather short for a disease with such a low incidence. Besides, during the referring time interval laboratory tests different from the ones that are common today were used for diagnosis; therefore, one may assume that with other tests and diagnostic criteria maybe more cases would have been identified. Furthermore, medical documentation sometimes shows deficits so that not only data in the birth protocols are missing occasionally but also patient histories seem to have some gaps. This human error may possibly also be transferred to the process of data collection which was conducted by a single person.

In the future it would be good to have a larger patient sample as well as to carry out the search over a longer period of time. To be as efficient as possible, obtaining data from other hospitals in Croatia would allow a larger sample size for the creation of a bigger population-based picture regarding ICP.

Also, by adapting laboratory diagnostic methods in the University Hospital of Split, for example the introduction of measurement of serum bile acids indicated by a suspicious clinical picture, more cases could be detected that might not come to attention with current procedures used in this hospital.

To gain more insight into the effectiveness of the management strategies of ICP in Split, future studies might follow patients regarding the treatment they received and other related aspects. Generally, further clarification of the etiology and pathophysiology of obstetric cholestasis is needed. With a deeper understanding, the treatment approach could be shifted from an empiric and experience-based to a more standardized and objective one so that a greater conformity and uniformity of guidelines for the management of ICP could be achieved.

6. CONCLUSIONS

1. There is no significant difference in epidemiology and perinatal outcome between pregnancies with and without ICP in our study population.

2. It is necessary to achieve more precise and more standardized diagnostic tools and methods for obstetric cholestasis in clinical settings as an important step towards a consensus regarding the management of patients with ICP.

7. REFERENCES

1. Ozkan S, Ceylan Y, Ozkan OV, Yildirim S. Review of a challenging clinical issue: Intrahepatic cholestasis of pregnancy. World J Gastroenterol. 2015;21(23):7134-41.

2. Diken Z, Usta IM, Nassar AH. A Clinical Approach to Intrahepatic Cholestasis of Pregnancy. Am J Perinatol. 2014;31:1-8.

3. NHS Worcestershire Acute Hospitals, Worcestershire, England, Cholestasis in Pregnancy – Management guideline WHAT-OBS008 [Internet]. Available from: http://www2.worcsacute.nhs.uk/healthprofessionals/clinical-guidelines/?assetdet906991=10961&p=13. [updated 2015 July 03; cited 2018 April 3]

4. South Australian Maternal & Nenonatal Community of Practice, Government of South Australia, South Australia, Clinical guideline on the topic of Obstetric Cholestasis [Internet]. Available from : http://www.sahealth.sa.gov.au/wps/wcm/connect/f91fbf004ee530b2a5ebadd150ce4f37/obstetric+cholestasis_27042016.pdf?MOD=AJPERES&CACHEID=ROOTWORKSPACE-f91fbf004ee530b2a5ebadd150ce4f37-m08kAos.[updated 2016 April 19; cited 2018 April 3]

5. Trauner M, Fickert P, Pertl B. Schwangerschaftsspezifische Lebererkrankungen. Dtsch Arztebl. 2004;101:A3416-25.

6. Pata Ö, Vardareli E, Özcan A, Sertester M, Ünsal I, Saruç M et al. Intrahepatic cholestasis of pregnancy: Correlation of preterm delivery with bile acids. Turk J Gastroenterol. 2011;22(6):602-5.

7. Liu X, Landon MB, Chen Y, Cheng W. Perinatal outcomes with intrahepatic cholestasis of pregnancy in twin pregnancies. J Maternal Fetal Neonatal Med. 2016;29(13):2176-81.

8. Floreani A, Gervasi MT. New Insights on Intrahepatic Cholestasis of Pregnancy. Clin Liver Dis. 2016;20(1):177-89.

9. Böcker W, Denk H, Heitz PU, Moch H. Pathologie. 4th ed. München: Elsevier Urban & Fischer; 2008. p. 781-83.

10. Marschall H-U. Management of intrahepatic cholestasis of pregnancy. Expert Rev Gastroenterol Hepatol. 2015;9(10):1273-9.

11. Dixon PH, Williamson C. The pathophysiology of intrahepatic cholestasis of pregnancy. Clin Res Hepatol Gastroenterol. 2016;40(2):141-53.

12. Herrera CA, Manuck TA, Stoddard GJ, Varner MW, Esplin S, Clark EAS et al. Perinatal outcomes associated with intrahepatic cholestasis of pregnancy. J Matern Fetal Neonatal Med. 2018;31(14):1913-20.

13. Lammert F, Marschall H-U, Glantz A, Matern S. Intrahepatic cholestasis of pregnancy: molecular pathogenesis, diagnosis and management. J Hepatol. 2000;33(6):1012-21.

14. Kondrackiene J, Zalinkevicius R, Sumskiene J, Gintautas V, Kupcinskas L. Sensitivity and Specificity of Biochemical Tests for Diagnosis of Intrahepatic Cholestasis of Pregnancy. Ann Hepatol. 2017;16(4):569-73.

15. Royal College of Obstetricians & Gynecologists, United Kingdom, Obstetric Cholestasis, Green-top Guideline No. 43 [Internet]. Available from https://www.rcog.org.uk/globalassets/documents/guidelines/gtg_43.pdf. [updated 2011 May 19; cited 2018 April 3]

16. Schneider H, Husslein PW, Schneider K-TM. Die Geburtshilfe. 3rd ed. Heidelberg Berlin: Springer-Verlag; 2006. p. 284.

17. Stauber M, Weyerstahl T. Duale Reihe Gynäkologie und Geburtshilfe. 3rd ed. Stuttgart: Georg Thieme Verlag KG; 2007. p. 557.

18. Azzaroli F, Turco L, Lisotti A, Calvanese C, Mazzella G. The Parmacological Management of Intrahepatic Cholestasis of Pregnancy. Curr Clin Pharmacol. 2011;6(1):12-7.

19. Madazli R, Yuksel MA, Oncul M, Tuten A, Gurlap O, Aydin B. Pregnancy outcomes and prognostic factors in patients with intrahepatic cholestasis of pregnancy. J Obstet Gynaecol. 2015;35(4):358-61.

20. Gurung V, Stokes M, Middleton P, Milan SJ, Hague W, Thornton JG. Interventions for treating cholestasis in pregnancy (Review). Cochrane Database Syst Rev. 2013(6):CD000493.

21. Joutsiniemi T, Timonen S, Leino R, Paolo P, Ekbald U. Ursodeoxycholic acid in the treatment of intrahepatic cholestasis of pregnancy: a randomized controlled trial. Arch Gynecol Obstet. 2014;289(3):541-7.

22. Joutsiniemi T, Timonen S, Linden M, Suvitie P, Ekblad U. Intrahepatic cholestasis of pregnancy: observational study of the treatment with low-dose ursodeoxycholic acid. BMC Gastroenterol. 2015;15:92.

23. Bühling KJ, Friedmann W. Intensivkurs Gynäkologie und Geburtshilfe. 2nd ed. München Jena: Elsevier Urban & Fischer; 2009. p. 147.

24. Gerhard I. Geburtshilfe integrativ – Konventionelle und komplementäre Therapie. München: Elsevier Urban & Fischer; 2005. p. 227.

25. Covach AJ, Rose WN. Intrahepatic Cholestasis of Pregnancy Refractory to Multiple Medical Therapies and Plasmapheresis. Am J Perinatol Rep. 2017;7:e223-5.

26. Sävervall C, Sand FL, Thomsen SF. Dermatological Diseases Associated with Pregnancy: Pemphigoid Gestationis, Polymorphic Eruption of Pregnancy, Intrahepatic Cholestasis of Pregnancy, and Atopic Eruption of Pregnancy. Dermatol Res Pract. 2015;2015:979635.

27. Puljic A, Kim E, Page J, Esakoff T, Shaffer B, LaCoursiere DY et al. The risk of infant and fetal death by each additional week of expectant management in intrahepatic cholestasis of pregnancy by gestational age. Am J Obstet Gynecol. 2015;212:e1-5.

28. Kia L, Rinella ME. Interpretation and Management of Hepatic Abnormalities in Pregnancy. Clin Gastroenterol Hepatol. 2013;11(11):1392-8.

29. Jain R, Suri V, Chopra S, Chawla YK, Kohli KK. Obstetric cholestasis: outcome with active management. J Obstet Gynaecol Res. 2013;39(5):953-9.

30. Henderson CE, Shah RR, Gottimukkala S, Ferreira KK, Hamaoui A, Mercado R. Primum non nocere: how active management became modus operandi for intrahepatic cholestasis of pregnancy. Am J Obstet Gynecol. 2014;211(3):189-96.

31. Tran TT, Ahn J, Reau NS. ACC Clinical Guidline: Liver Disease and Pregnancy. Am J Gastroenterol. 2016;111:176-94.

32. Kong X, Kong Y, Zhang F, Wang T, Yan J. Evaluating the effectiveness and safety of ursodeoxycholic acid in treatment of intrahepatic cholestasis of pregnancy. Medicine (Baltimore). 2016;95(40):e4949.

33. Erlinger S. Intrahepatic cholestasis of pregnancy: A risk factor for cancer, autoimmune and cardiovascular diseases?. Clin Res Hepatol Gastroenterol. 2016;40(2):139-40.

34. Kamimura K, Abe H, Kawai H, Kamimura H, Kobayashi Y, Nomoto M et al. Advances in understanding and treating liver diseases during pregnancy: A review. World J Gastroenterol. 2015;21(17):5183-90.

35. Al Shobaili HA, Hamed HO, Al Robaee A, Alzolibani AA, Amin AF, Ahmad SR. Obstetrical and fetal outcomes of a new management strategy in patients with intrahepatic cholestasis of pregnancy. Arch Gynecol Obstet. 2011;283:1219-25.

36. Ekiz A, Kaya B, Avci ME, Polat I, Dikmen S, Yildirim G. Alanine amonotransferase as a predictor of adverse perinatal outcomes in women with intrahepatic cholestasis of pregnancy. Pak J Med Sci. 2016;32(2):418-22.

37. Reece EA, Barbieri RL. Obstetrics and Gynecology: The Essentials of Clinical Care. Stuttgart New York: Georg Thieme Verlag; 2010. p. 179.

38. Rioseco AJ, Ivankovic MB, Manzur A, Hamed F, Kato SR, Parer JT et al. Intrahepatic cholestasis of pregnancy: A retrospective case-control study of perinatal outcome. Am J Obstet Gynecol. 1994;170(3):890-5.

39. Geenes V, Chappell LC, Seed PT, Steer PJ, Knight M, Williamson C. Association of Severe Intrahepatic Cholestasis of Pregnancy With Adverse Pregnancy Outcomes: A Prospective Population-Based Case-Control Study. Hepatology. 2014;59(4):1482–91.

40. WHO fact sheet "Obesity and overweight" [Internet]. Available from http://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight. [updated 2017 October 18; cited on 2018 May 10]

41. Mändle C, Opitz-Kreuter S. Das Hebammenbuch: Lehrbuch der praktischen Geburtshilfe. 5th ed. Stuttgart New York: Schattauer GmbH; 2007.

42. Apgar V. A proposal for a New Method of Evaluation of the Newborn Infant. Curr Res Anesth Analg. 1953;32(4):260-7.

43. Armstrong L, Stenson BJ. Use of umbilical cord blood gas analysis in the assessment of the newborn. Arch Dis Child Fetal Neonatal Ed. 2007; 92(6): F430–4

44. Kliegman RM, Stanton BF, St Geme JW, Schor NF. Nelson Textbook of Pediatrics. 20th ed. Philadelphia PA: Elsevier; 2016. p. 811.

45. Helwig JI, Parter JT, Kilpatrick SJ, Laros RK. Umbilical cord blood acid-base state : What is normal? Am J Obstet Gynecol. 1996;174(6):1807-12.

8. SUMMARY

Objective: To determine whether there is a difference in the perinatal outcome in pregnancies complicated by intrahepatic cholestasis compared to unaffected pregnancies.

Materials and methods: Out of 2466 births that took place in the University Hospital of Split in 2016, 28 identified pregnancies affected by obstetric cholestasis were compared to an equally sized control group regarding maternal epidemiology and perinatal outcome. Included into the control group was every birth recorded consecutive to a case of with parity equal to that case. Investigated parameters were the mothers' age, maternal BMI, pregnancy duration, fetal weight, fetal length, Apgar score, umbilical cord blood pH, delivery mode and size for gestational age.

Results: There was no statistically significant difference in the observed variables in pregnancies affected and not affected by intrahepatic cholestasis. The average maternal age of the case group was 30.25±4.07 years, compared to 30.43±4.66 years in the control group (p=0.879). Mothers with ICP had an average BMI of 22.86±2.58, those without had one of 23.53±4.99 (p=0.535). Pregnancy lasted 37 weeks or longer in 89.29% of cases of ICP, shorter than that in 10.71%, compared to the control group where 96% of pregnancies lasted at least 37 weeks and 3.57% lasted shorter (p=0.118). Average fetal weight in the case group was 3493.57±551.75g and 3373.21±517.02g in the control group (p=0.403). Fetal length on average was 50.39±1.97cm in the case group and 49.75±2.01cm in the control group (p=0.232). 92.86% of babies of mothers with ICP had an Apgar score between 8 and 10 and 7.14% had one between 4 and 7, while after healthy pregnancies all neonates achieved a score between 8 and 10 (p=0.118). The average umbilical cord blood pH in newborns of the ICP group was 7.32±0.09 and 7.34±0.07 in the control group (p=0.431). 78.57% of women of the case group had a vaginal delivery and 21.43% underwent a Cesarean section, while in the control group 64.29% gave birth vaginally and 35.71% per C-section (0.235). No newborn of the ICP group and 7.41% of the ones born to healthy group were SGA (p=0.142), 89.29% in the case group and 85.19% in the control group were AGA (p=0.648), and 10.71% of neonates in the group with cholestasis were LGA compared to 7.41% in the group without cholestasis (p=0.670).

Conclusion: There is no difference in perinatal outcome between normal pregnancies and pregnancies complicated by intrahepatic cholestasis in the University Hospital of Split.

9. CROATIAN SUMMARY

Naslov: Intrahepatična kolestaza trudnoće: Retrospektivno istraživanje slučajeva i kontrola perinatalnog ishoda u KBC-u Split

Cilj: Odrediti da li postoji razlika u perinatalnom ishodu u trudnoćama sa intrahepatičnom kolestazom u usporedbi sa trudnoćama van utjecaja intrahepatične kolestaze.

Materijali i metode: Od 2466 porođaja koji su se dogodili u KBC-u Split u 2016. godini, 28 identificiranih trudnoća pod utjecajem intrahepatične kolestaze uspoređene su sa kontrolnom skupinom iste veličine u vezi epidemioloških karakteristika majki i perinatalnog ishoda. U kontrolnoj skupini je uključen svaki porod dokumentiran uzastopno slučaju intrahepatične kolestaze sa paritetom jednakim tom slučaju. Istraženi parametri su bili dob majke, BMI majke, trajanje trudnoće, porodna težina, porodna duljina, Apgar zbroj, pH krvi pupačne vrpce, način rođenja i trofičnost djece.

Rezultati: Nije bilo značajnih razlika u istraženim varijablama u trudnoćama sa intrahepatičnom kolestazom u usporedbi sa trudnoćama bez intrahepatične kolestaze. Prosječna dob majka u ispitivanoj skupini je bio 30,25±4,07 godina, u usporedbi sa 30,43 ±4,66 godina u kontrolnoj skupini (p=0,879). Majke sa kolestazom su imali prosječan BMI od 22,86±2,58, one bez kolestaze od 23,53±4,99 (p=0,535).Trudnoća je trajala 37 tjedana ili duže u 89,29% slučaja kolestaze, kraće od toga u 10,71%, u usporedbi sa kontrolnom skupinom u kojoj 96% trudnoća je trajalo barem 37 tjedana i 3,57% je trajalo kraće (p=0,118). Prosječna porodna težina u ispitivanoj skupini je bila 3493,57±551,75g i 3373,21±517,02g je bila u kontrolnoj skupini (p=0,403). Porodna duljina u prosjeku je bila 50,39±1,97cm u ispitivanoj skupini i 49,75±2,01cm u kontrolnoj skupini (p=0,232). 92,86% novorođenčadi majki sa intrahepatičnom kolestazom je imalo Apgar zbroj od 8 do 10 i 7,14% od 4 do 7, ali poslije zdravih trudnoća sva novorođenčad je postigla zbroj između 8 i 10 (p=0,118). Prosječan pH krvi pupačne vrpce u novorođenčadi od skupine sa kolestazom je bio 7,32±0,09, i 7,34±0,07 je bio u kontrolnoj skupini (p=0,431). 78,57% žena u ispitivanoj skupini su rodile vaginalno i 21,43% su se podvrgnule carskom rezu, dok u kontrolnoj skupini 64,29% su imale vaginalni porod i 35,71% carski rez (p=0,235). Nijedno novorođenče iz skupine sa kolestazom i 7,41% onih rođenih u zdravoj skupini nije bilo hipotrofično (p=0,142), 89,29% u ispitivanoj skupini i

85,19% u kontrolnoj skupini su bili eutrofični (p=0,648), i 10,71% novorođenčadi u skupini sa kolestazom su bili hipertrofični u usporedbi sa 7,41% u skupini bez kolestaze (p=0,670).

Zaključak: Ne postoji razlika perinatalnog ishoda između normalnih trudnoća i trudnoća kompliciranih sa intrahepatičnom kolestazom u KBC-u Split.

10. CURRICULUM VITAE

Personal information :

Name and surname :	Verena Nunhofer
Address :	Spinčićeva 33a, 21000 Split
Telephone number :	+385 95 7226779
E-mail address :	verena_nunhofer@yahoo.de
Date and place of birth :	31st August 1992, Fürth, Germany
Nationality :	German

Education :

2001 – 2010 :	Melanchthon-Gymnasium Nürnberg
WS 2010/11 :	Friedrich-Alexander-Universität Erlangen-Nürnberg, BA studies in Latin and Ancient Greek
2012 :	University of California Berkeley Extension, distance course in Human Physiology
2012 – 2018 :	University of Split School of Medicine, Medical Studies in English

Clinical traineeships :

08.2015 :	Klinikum Heidenheim, Heidenheim / Brenz (DE) : Cardiology
08.2016 :	Klinikum Nürnberg Süd, Nürnberg (DE) : Neurology
08.2017 :	Klinikum Nürnberg Süd, Nürnberg (DE) : Anesthesia
01.2018 :	Klinikum am Europakanal, Erlangen (DE) : Psychiatry

Other activities :

2006 – 2008 :	Workers' Welfare Association Nürnberg, voluntary tutoring of pupils
2008 – 2012 :	Private tutoring of pupils
2011 – 2012 :	Internistische Fachklinik Dr. Steger, Nürnberg, Federal Voluntary Service at the dialysis center