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The relation between umbilical cord coiling index and thyroid function disorders

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Several articles have demonstrated an association between specific pregnancy-related medical issues and the prevalence of the umbilical coiling index.
The objective: **to determine the potential impact of the aberrant coiling index of the human umbilical cord on thyroid function content and its potential deleterious effects on infant health.**
Materials and methods**. Umbilical cord samples were taken from 105 practically healthy pregnant women. Umbilical cords were collected immediately after delivery and stored in formalin (10%) based on the type of coil. Umbilical cords were divided into three categories of 35 samples for each experimental group – normocoiled, hypercoiled, and hypocoiled. This division was performed according to the umbilical cord index.**
Thyroid hormones (thyroid-stimulating hormone – TSH), triiodothyronine – T3 and thyroxine – T4) were determined using the enzyme immunoassay method in maternal venous blood serum and umbilical cord blood.
Results**. There was no significant difference in the concentration of TSH, T3 and T4 hormones in the blood serum of mothers between the three different study groups (P>0.05). There was also no statistically significant difference (P>0.05) when comparing the levels of TSH and T3 in the umbilical cord in all three study groups.**
Regarding concentration of T4, the data showed a significant difference in the obtained values (P<0.01) when comparing venous blood samples in normocoiled cords compared to hypercoiled and hypocoiled cords. In addition, comparison of thyroid hormone levels in maternal venous blood and umbilical cord blood demonstrated that in all study groups there was a significant difference (P<0.05) in TSH levels between maternal and umbilical cord blood serum.
The concentration of T3 did not differ significantly (P>0.05) in maternal blood serum compared to umbilical cord blood. In the statistical analysis, the levels of T4 in venous blood samples from normocoiled and hypocoiled umbilical cords differ significantly from the level of this hormone in the mother's blood (P<0.01, P<0.05). There was no significant difference in the concentration of T4 in hypercoiled umbilical cord blood samples (P>0.05) compared to maternal blood serum.
Conclusions**. The concentration of T4 increases in the fetal circulation with normocoiled umbilical cord, since this form of coiling is the best situation for proper feto-maternal exchange function of this hormone.**
Keywords: *umbilical coiling index, thyroid hormones, umbilical cord.*

Зв'язок між індексом звивистості пуповини та порушеннями функції щитоподібної залози

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Результати декількох досліджень свідчать про зв'язок між певними медичними патологіями, пов'язаними з вагітністю, та значенням індексу звивистості пуповини.
Мета дослідження: встановлення потенційного впливу аберантного індексу звивистості пуповини людини на функцію щитоподібної залози та його потенційного шкідливого впливу на здоров'я немовлят.
Матеріали та методи**.** Відібрано зразки пуповини у 105 практично здорових вагітних. Пуповини відбирали зразу після пологів та зберігали у формаліні (10%) залежно від типу звивистості. Проведено поділ пуповин на три категорії по 35 зразків для кожної дослідної групи – пуповини з нормальною звивистістю, гіперзвивисті та гіпозвивисті. Цей поділ проводили відповідно до індексу звивистості пуповини.
Гормони щитоподібної залози (тиреотропний гормон – ТТГ), трийодтиронін – Т3 та тироксин – Т4) визначали за допомогою імуноферментного методу у сироватці венозної крові матері та пуповини.
Результати**.** Достовірної різниці стосовно концентрації гормонів ТТГ, Т3 та Т4 у сироватці крові матерів між різними трьома групами дослідження не встановлено (P>0,05). Також не було статистично значущої різниці (P>0,05) при порівнянні рівнів ТТГ і Т3 у пуповині у всіх трьох групах дослідження.
Що стосується рівнів Т4, то дані свідчать про суттєву різницю в отриманих значеннях (P<0,01) під час порівняння зразків венозної крові у нормально звивистих пуповинах порівняно з гіперзвивистими та гіпозвивистими пуповинами. Крім того, порівняння рівнів гормонів щитоподібної залози у венозній крові матері та пуповині продемонструвало, що у всіх дослідних групах наявна достовірна різниця (P<0,05) у рівнях ТТГ між сироваткою крові матері та пуповини.
Концентрація Т3 суттєво не відрізнялась (P>0,05) у сироватці крові матері щодо крові пуповини. У статистичному аналізі рівні Т4 у зразках венозної крові з нормально звивистих і гіпозвивистих пуповин значно відрізняються від рівня даного гормону у крові матері (P<0,01, P<0,05). Не спостерігалося суттєвої різниці у концентрації Т4 у зразках крові гіперзвивистих пуповин (P>0,05) порівняно з сироваткою крові матері.
Висновки**.** Концентрація Т4 підвищується у кровообігу плода при нормальній звивистості пуповини, оскільки ця форма звивистості є найкращою ситуацією для належного фето-материнського обміну даного гормону.
Ключові слова: *індекс звивистості пуповини, гормони щитоподібної залози, пуповина.*

The umbilical cord is an essential and fundamental part that communicates the mother and her fetus together [1], by which the fetus obtains crucial fluids, oxygen, and nutrition from the placenta [2]. This importance is described by Ian Donald when he assumed that "The baby's life hangs by a cord" [3]. Accordingly, the abnormalities of the umbilical cord are always accompanied by mutable results of the mother and her fetus; these aberrations are linked to almost 22.5 percent of intrauterine deaths of fetuses [4]. The thyroid gland is the initial endocrine gland to undergo development in the human embryo; the initial stage of this developmental process involves the thickening of the floor of the rudimentary pharynx, specifically occurring between the diverging aorta, around 22 days after conception [5].

Before that and during the early stages of pregnancy, the embryo relies totally on the supply of thyroid hormone from the mother [6]. During the initial trimester, the syncytiotrophoblast cells of the blastocyst emit elevated quantities of human chorionic gonadotropin (HCG). This hormone serves as a signal to the corpus luteum, prompting it to sustain progesterone synthesis. The presence of progesterone is crucial for the preservation of the pregnancy. HCG has structural similarities to thyroid-stimulating hormone (TSH) and consequently exerts thyrotropic effects.

This results in an elevation of free thyroxine (FT4) and free triiodothyronine (FT3) levels during the first trimester of pregnancy, accompanied by a concomitant reduction of circulating TSH. It is suggested that the embryo/fetus utilizes the control of the maternal endocrine system to ensure sufficient substrate levels for its growth. This process occurs when the fetal thyroid gland does not yet possess the ability to synthesize thyroid hormones [7, 8]. The role of the thyroid gland is evident in secretes triiodothyronine (T3) and Thyroxine (T4) hormones [9, 10], which may be identified in the fetal bloodstream during the early stages of pregnancy.

These hormones significantly affect the fetus's development, metabolism, and maturation across several species, including human neonates. The bioavailability of substances in fetal plasma and tissues is subject to developmental regulation. It exhibits variations between species, gestational age, nutrition, oxygen availability, and the endocrine environment throughout intrauterine development [11].

Many researches have shown a correlation between some pregnancy-related medical conditions and the occurrence of umbilical coiling index. A prior investigation has established that neonates born to females with gestational diabetes are more likely to possess either hypercoiled or non-coiled umbilical blood vessels [12]. Dogne Nalini *et al*. found a notable statistical correlation between medical issues and the umbilical cord coiling index. The researchers concluded a robust link between thyroid illness and improper coiling of the umbilical cord, particularly in hypothyroidism with a hypocoiled umbilical cord [13].

The purpose of this study is to investigate the potential impact of the aberrant coiling index of the human umbilical cord on thyroid function content and its potential deleterious effects on infant health.

MATERIALS AND METHODS

This study was conducted at the Department of Obstetrics and Gynecology in Al-Emamin Al-Kadhimain Medical City during the period from January 2023 to June 2023. The study samples were collected from 105 healthy pregnant women; the pediatrician confirmed a standard APGAR score before collecting the umbilical cord samples. The umbilical cord samples were collected immediately after the labor process and kept in formalin (10%) for histological and immunohistochemical study, according to the coil type of the cord.

The total number of samples was 105; the samples were divided equally into three major categories (35 samples of normocoiled, 35 samples of hypercoiled, and 35 samples of hypocoiled). Additionally, the process of obtaining blood samples consists of two steps. The first step is executed before labor, which involves collecting blood samples from the mothers. The second step is executed after birth and before the separation of the umbilical cord from the baby's side, which consists of collecting approximately 3–5 ml of umbilical cord blood.

These blood samples were used for serology tests of thyroid hormones analyses. All newborn infants had a comprehensive medical assessment conducted by a specialized physician to verify the overall state of the baby's health and well-being depending majorly on the Apgar score. The Apgar score assesses an infant's state immediately following delivery. It assesses a neonate's condition at 1 minute and 5 minutes postpartum. The score serves as a rapid means for physicians to evaluate the well-being of newborns and provide guidance to midwives, physicians, and nurses regarding the necessity of prompt intervention or observation for a baby.

Dr. Virginia Apgar, an anesthesiologist at Columbia University, created it in 1952 [14]. The adverse and abnormal delivery outcomes that have been linked to postnatal hypercoiling frequently coincide with those observed in cases of hypocoiling. These outcomes include a poorer APGAR score at 5 minutes and a non-reassuring fetal state [15, 16]. Similar to hypercoiling, hypocoiling has also been linked to abnormal and adverse birth outcomes. According to prior data from many researchers, the occurrence rate of hypocoiled cords in unselected singleton pregnancies varies between 7.5% and 16.0% [17].

Postnatal hypocoiling has been linked to many atypical birth outcomes, including spontaneous preterm delivery, lower Apgar score at 5 minutes, and non-reassuring fetal condition [18]. This study is a trial to explore the effect of umbilical cord twisting on overall baby health, including thyroid hormone levels, and how the baby adjusts these levels to normal even in abnormal umbilical cord patterns. The reason for collecting the mothers' serum was to confirm the maternal thyroid gland's normality and eliminate the mothers' cause if the umbilical serum thyroid tests were abnormal.

Determining the umbilical cord coiling index necessitates considering two fundamental principles pertaining to the dimensions of the umbilical cord. This measurement should be conducted promptly following the birth of the neonate. The first principle involves the measurement of the umbilical cord's length, which can be accomplished us-

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ing a steel measuring tape. The second principle quantifies the number of coils within the umbilical cord [19, 20]. The umbilical cord coiling index can easily be estimated by dividing the total number of coils by the total length of the umbilical cord as the equation below:

UCI = Number of coils/
Length of umbilical cord in Centimeters.

Thyroid function tests were done by using AIA-360 immunoassay analyzer manufacturing by TOSOH Bioscience company, Japan.

The ST AIA-Pack TSH is a two-site immunoenzymometric assay performed entirely in the ST AIA-Pack TSH test cups. TSH present in the test sample is bound with monoclonal antibody immobilized on magnetic beads and monoclonal antibody conjugated with bovine alkaline phosphatase in the test cups. The magnetic beads are washed to remove any non-bound enzyme-labelled monoclonal antibodies and then incubated with a fluorogenic substrate, 4-methylumbelliteryl phosphate (4MUP). The amount of enzyme conjugated with monoclonal antibody that binds to the beads is directly proportional to the TSH concentration in the test sample. A standard curve is constructed, and unknow sample concentration are calculated using this curve.

Moreover, the analysis principle of ST AIA-PACK T3 involves a competitive enzyme immunoassay performed entirely within the ST AIA-PACK TT3 test cartridges. In this assay, triiodothyronine (T3) competes with enzyme-labeled T3 for a limited number of binding sites on T3-specific antibodies attached to magnetic beads. After washing away unbound enzyme-labeled T3, the beads are incubated with a fluorogenic substrate (4MUP). The amount of enzyme labeled T3 bound to the beads is inversely proportional to the T3 concentration in the sample. Using known standard concentration ranges, a standard curve is generated, and the unknown T3 concentration in the sample is calculated based on this curve. Likewise, the test principle for T4 utilizes a competitive enzyme immunoassay. In this assay:

- Thyroxine (T4) present in the sample competes with enzymatically labeled T4 for binding sites on T4-specific antibodies immobilized on magnetic beads.
- After washing away unbound labeled T4, the beads are incubated with a fluorogenic substrate (4MUP).
- The amount of enzymatically labeled T4 bound to the beads is inversely proportional to the T4 concentration in the sample.

This technology provides test results of exceptional quality promptly. All participants agreed to provide the investigator with the specimens. The ethics committee of College of Science, Mustansiriyah University approved this work. Additionally, the article costs were borne by the author and his colleagues themselves with no external funding sources.

The obtained data were subjected to analysis of variance (ANOVA) test to compare the means of various groups with each other. Results were expressed in descriptive analysis. LSD test was used to calculate the significant differences between tested means. The indication of no significant differences between tested mean ($P \leq 0.05$) was considered statically significant while values of ($P \geq 0.05$) were considered statistically nonsignificant. The statistical analysis was carried out by SPSS version 20.

RESULTS AND DISCUSSION

The thyroid function test (including the concentration of TSH, T3, and T4) was evaluated for the venous blood obtained from the pregnant mothers and the umbilical cord venous blood obtained from all three groups (Normocoiled, Hypercoiled and hypocoiled), the venous blood samples obtained immediately after labor, and before delivery of the placenta. There was no statistical difference ($P>0.05$) detected when comparing the results of this biochemical analysis of these hormones in the blood of the pregnant mothers of the three groups as indicated in tables 1, 2 and 3.

Likewise, the analysis of the biochemical data of thyroid test done for the serum samples obtained from the umbilical cords of the three groups indicates no statistically significant difference ($P>0.05$) when comparing the level of TSH and T3 as shown in tables 4 and 5. The data obtained from measuring the T4 level in all three groups of umbilical cords showed a significant difference in the values obtained ($P<0.01$) when comparing the venous blood samples of normocoiled cords with those of hypercoiled and hypocoiled cords as it revealed in table 6.

The comparative analysis of the level of thyroid hormones in the venous blood of the mothers and the umbilical cords obtained from each of them showed the following:

1. A statistically significant difference ($P<0.05$) in TSH levels between mother and cord serum was observed in all the study groups, as indicated in table 7.

2. The T3 level did not exhibit any statistically significant difference ($P>0.05$) when comparing the levels in serum taken from the mother/cord, as indicated in table 8.

Table 1

TSH levels in serum of the mothers

TSH/MOTHWER	Normocoiled	Hypercoiled	Hypocoiled	P value
Number of samples	35	35	35	
Mean	2.69	2.91	2.44	Non-Significant differences
Std. Error of Mean	0.27	0.33	0.33	
Median	2.82	3.20	2.52	
Std. Deviation	1.05	1.22	1.29	
Minimum	1.28	0.86	0.45	
Maximum	4.16	4.86	4.27	
The unit of measurement of TSH: μIU/mL.				

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Table 2

T3 levels in serum of the mothers

T3/MOTHERS	Normocoiled	Hypercoiled	Hypocoiled	P value
Number of samples	35	35	35	
Mean	2.12	2.29	2.54	Non-Significant differences
Std. Error of Mean	0.19	0.25	0.19	
Median	1.98	2.36	2.84	
Std. Deviation	0.75	0.93	0.72	
Minimum	1.11	0.77	1.31	
Maximum	3.28	3.57	3.28	
The unit of measurement of T3: nmol/L.				

Table 3

T4 levels in serum of the mothers

T4/MOTHERS	Normocoiled	Hypercoiled	Hypocoiled	P value
Number of samples	35	35	35	
Mean	99.50	108.16	114.90	Non-Significant differences
Std. Error of Mean	6.70	7.35	7.47	
Median	97.05	110.00	114.33	
Std. Deviation	25.95	27.51	28.94	
Minimum	59.16	64.37	76.42	
Maximum	144.26	142.92	182.14	
The unit of measurement of TSH: nmol/L.				

Table 4

TSH levels in serum of the umbilical cord

TSH/umbilical cord	Normocoiled	Hypercoiled	Hypocoiled	P value
Number of samples	35	35	35	
Mean	4.69	4.29	5.59	Non-Significant differences
Std. Error of Mean	0.35	0.42	0.49	
Median	4.24	3.88	5.35	
Std. Deviation	1.37	1.63	1.91	
Minimum	3.01	2.19	3.17	
Maximum	7.72	7.55	10.60	
The unit of measurement of TSH: μIU/mL.				

Table 5

T3 levels in serum of the umbilical cord

T3/umbilical cord	Normocoiled	Hypercoiled	Hypocoiled	P value
Number of samples	35	35	35	
Mean	1.76	2.17	1.85	Non-Significant differences
Std. Error of Mean	0.17	0.22	0.18	
Median	1.78	2.25	1.95	
Std. Deviation	0.66	0.87	0.70	
Minimum	0.81	0.82	0.67	
Maximum	2.83	3.29	3.13	
The unit of measurement of T3: nmol/L.				

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Table 6

T4 levels in serum of the umbilical cord

T4/umbilical cord	Normocoiled	Hypercoiled	Hypocoiled	P value
Number of samples	35	35	35	
Mean	146.80	112.48	127.04	0.01 when comparing T4 levels of normocoiled cords with those of hypercoiled and hypocoiled cords
Std. Error of Mean	10.20	7.69	10.66	
Median	148.80	111.80	139.91	
Std. Deviation	39.49	29.79	41.27	
Minimum	87.30	57.97	68.77	
Maximum	212.40	163.29	218.90	
The unit of measurement of T4: nmol/L.				

Table 7

TSH levels in serum of the mothers and umbilical cord

TSH/umbilical cord	Normocoiled	Hypercoiled	Hypocoiled
Number of samples	35	35	35
Mean	4.69	4.29	5.59
Std. Deviation	1.37	1.63	1.91
TSH/MOTHWER	Normocolied	Hypercoiled	Hypocoiled
Mean	2.69	2.91	2.44
Std. Deviation	1.05	1.22	1.29
p value	0.05	0.05	0.05
The unit of measurement of TSH: μIU/mL.			

Table 8

T3 levels in serum of the mothers and umbilical cord

T3/umbilical cord	Normocoiled	Hypercoiled	Hypocoiled
Number of samples	35	35	35
Mean	1.76	2.17	1.85
Std. Deviation	0.66	0.87	0.70
T3/MOTHERS	Normocoiled	Hypercoiled	Hypocoiled
Mean	2.12	2.29	2.54
Std. Deviation	0.75	0.93	0.72
p value	Non-Significant differences	Non-Significant differences	Non-Significant differences
The unit of measurement of T3: nmol/L.			

3. The data in table 9 showed diversity when comparing the T4 level in venous blood samples obtained from the normocoiled and hypocoiled cords with that of the mothers. A significant difference ($P<0.01$, $P<0.05$) was observed in the statistical analysis of these data. However, no statistical difference was found when comparing the T4 concentration in the venous blood samples obtained from the hypercoiled cords ($P>0.05$) with the mothers.

The results showed no statistically significant variations among the maternal serum in the biochemical analysis of these hormones in association with the variable coiling indices. The test of T3 and TSH hormones done for the serum of umbilical vessels of the three groups also showed no statistically significant variations. However, the test of T4 in the umbilical showed significant increment in normocoiled cords compared to hypercoiled and hypocoiled cords.

The previous articles reported that T4 hormone is important for the intrauterine normal development of the brain [21]. This T4 hormone reaches the fetus in sufficient amount from his mother [22]. These fact leads to suggestions that T4 increased in the fetal circulation in association with normocoiling as this pattern of coiling is the best situation for the proper feto-maternal exchange function, accordingly, T4 level in the fetal blood of the normocoiled cord is derived from both fetal thyroid gland and T4 transported from the mother via the placenta.

Therefore, in spite of the structural modulations occurring in the hypocoiled and hypercoiled cords, still the

Table 9

T4 levels in serum of the mothers and umbilical cord

T4/umbilical cord	Normocoiled	Hypercoiled	Hypocoiled
Number of samples	35	35	35
Mean	146.80	112.48	127.04
Std. Deviation	39.49	29.79	41.27
TR/MOTHERS	Normocoiled	Hypercoiled	Hypocoiled
Mean	99.50	108.16	114.90
Std. Deviation	25.95	27.51	28.94
p value	0.01	Non-Significant differences	0.05
The unit of measurement of T4: nmol/L.			

transport of the T4 hormone from the mother to the fetus is defected, and T4 level in the hypocoiled and hypercoiled umbilical cords blood is mainly representing the level secreted from the fetal thyroid gland with less contribution of this hormone from the maternal circulation. The results of this study showed that the statistically significant difference of the umbilical blood level of T3-TSH in comparison to T4 in the normocoiled cord was not associated with any adverse perinatal outcome. This result supported by a conclusion suggested previously that the

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extent of fetal consequences associated with thyroid hormones dysfunctions is unknown [23, 24].

According to above formulated conclusion about the statistical analysis of umbilical cord serum T4 hormone, the results of this study suggested that TSH and T3 hormonal levels in the umbilical cord blood is related to the fetal thyroid gland function and are not derived from the maternal circulation vial the feto-placental exchange function. This suggestion supported by the reported results previously documented that the maternal TSH is not readily crossing the placenta to reach the fetus [25].

The alterations in thyroid hormones in the blood of the fetus or the newborns reported to be associated with defected brain development [26], these reports mandated future studies to investigate the prenatal histological and histochemical changed of the brain in association with variable coiling indices.

CONCLUSIONS

The results from mother's serum shows that there was no statistical difference detected when comparing these hormones of the study groups (normocoiled , hypercoiled and hypocoiled).As for the cord serum the results of the T3 and TSH levels show no statistically significant difference in normcoiled , hypercoiled and hypocoiled groups. Finally, T4 levels were higher in the venous blood samples of normocoiled cords when compared with those of hypercoiled and hypocoiled cords.

Conflict of interest. The authors declare that there are no conflicts of interest regarding the publication of this paper. All authors concur that no financial, personal, or professional affiliations could be construed as influencing the research presented.

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