Maternal thyroxine and 3,5,3'-tri-iodothyronine kinetics in near-term pregnant rats at two different levels of hypothyroidism

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Abstract

Thyroid hormones are extremely important for development of the fetal central nervous system. Thyroidectomy results in severe hypothyroidism. In this study two levels of maternal hypothyroidism were reached by administration of different amounts of thyroxine (T_4) and 3,5,3'-tri-iodothyronine (T_3) to thyroidectomized pregnant rats. We examined the production, distribution and transport of T_4 and T_3 by performing a kinetic experiment (three-compartment analysis) with intact and thyroidectomized near-term pregnant rats which received either very low (Tx + lowTH) or normal (Tx + TH) doses of T_4 and T_3 . Despite administration of normal doses of thyroid hormones, plasma TSH was still elevated in the Tx + TH rats, meaning that these rats were still mildly hypothyroid. The Tx + lowTH rats were markedly hypothyroid, the plasma T_4 and T_3 levels being very low. In the mildly hypothyroid rats the transport of T_4 from plasma to the fast pool and *vice versa* was decreased compared with intact near-term pregnant rats. This could imply that much less T_4 is transported to the feto-placental compartment. Liver type I deiodinase was decreased, resulting in lowered plasma T_3 values. In the markedly hypothyroid rats all pools and rates of transport of T_4 and T_3 were greatly decreased. In conclusion, even mild hypothyroidism, despite normal plasma T_4 values, results in significant changes, especially in maternal T_4 transport. We suggest that even mild maternal hypothyroidism will have a negative effect on the availability of maternal T_4 for fetuses.

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Introduction

Thyroid hormones are extremely important for fetal development, especially of the central nervous system. In man, the children of mothers who were hypothyroid during pregnancy show a high incidence of behavioral and neurological disorders (1).

Rat studies have demonstrated that maternal hypothyroidism during pregnancy has irreversible effects on brain development, as indicated by a decrease in the activity of various cell marker enzymes in the brain of the progeny (2). The influence of maternal hypothyroidism on fetal development is particularly dramatic during the first half of the gestational period (3). However, even after the fetal thyroid starts to function, on day 17.5–18 of gestation, maternal thyroid hormones contribute to the fetal iodothyronine pool (4). The effects of maternal thyroidectomy, resulting in severe hypothyroidism, on fetal development and thyroid hormone levels have been studied extensively by the Madrid group (5–7). They demonstrated a decrease in the number of fetuses and the fetal body weight of thyroidectomized dams (5–7). Maternal plasma thyroxine (T_4) and 3,5,3'-tri-iodothyronine (T_3) levels decreased markedly, while thyrotropin

(TSH) was highly elevated in thyroidectomized rats. T_4 and T_3 concentrations were decreased in both liver and brain tissue (6, 8, 9). The concentrations of T_4 and T_3 in embryonic tissues from thyroidectomized dams were undetectable before the onset of fetal thyroid function (5). At the end of gestation, on day 21, fetal plasma TSH was elevated (3) and the plasma T_4 and T_3 levels of the fetuses of thyroidectomized dams were similar (7) or even slightly elevated (6, 9) compared with those of intact dams. Total thyroidal T_4 and T_3 levels in fetuses from thyroidectomized dams had decreased markedly by the end of gestation (7). The fetal liver T_4 concentration was decreased, whereas such a change was not found for hepatic T_3 . The deiodinase type I (ID-I) activity in fetal liver had decreased by 50% (7). No effect of maternal thyroidectomy on T_4 and T_3 concentrations in fetal brain on day 20 was found; in contrast, an increase in the cerebral T_4 and T_3 concentrations on day 21 was reported (5, 6).

Euthyroidism can be restored in thyroidectomized rats by administration of both T_4 and T_3 (10). If the amount of thyroid hormones administered is less than the amount normally produced, the animal will still be hypothyroid. In the present study two levels of thyroid hormone status were achieved by the administration of

different amounts of T_4 and T_3 to thyroidectomized pregnant rats.

The aim of our study was to examine the effects of a mild and a marked hypothyroid state on maternal thyroid hormone metabolism in the near-term pregnant rat. T_4 and T_3 metabolism, distribution, and transport were examined by performing kinetic studies using the three-compartmental model of distribution and metabolism developed by DiStefano (11, 12). In this model three compartments can be distinguished: (i) the plasma; (ii) tissues that exchange T_4 and T_3 with plasma at a fast rate, the fast pool; and (iii) tissues which exchange at a slow rate, the slow pool. Liver and kidney are considered to be the main components of the fast pool, whereas skin, muscle and brain belong to the slow pool (11, 12). Previously we suggested that in normal near-term pregnant rats T_4 is transported very rapidly from plasma to the fetoplacental compartment (13). In the present study we compared the transport of maternal thyroid hormones to the fetoplacental compartment during different levels of hypothyroidism. The mildly hypothyroid rats are of special interest; will there already be an effect on maternal thyroid hormone metabolism, and is the maternal contribution of thyroid hormones to the fetal compartment affected?

Materials and methods

Animals and diet

Female Wistar rats (CPB/WU, Iffa Credo, Brussels, Belgium) weighing 150 g were rendered hypothyroid by means of ^{131}I thyroid ablation (a single intraperitoneal injection of 15 MBq (0.4 mCi) Na^{131}I). One week before and one week after radiothyroidectomy the rats received iodine-free American Institute of Nutrition (AIN) food (14). Subsequently iodide-rich AIN food (40 μg KI per g food) was given for 5 days to wash the ^{131}I out of the animals' system. During the rest of the experiment the animals received AIN food (dry powder mixed with distilled water (60% dry weight:40% water)) containing 55 ng iodide/g food.

Intact, age-matched controls also consumed AIN food with 55 ng iodide/g food.

Hypothyroidism was demonstrated by undetectable plasma T_4 levels six weeks after radiation. No sooner than two months after radiothyroidectomy the rats were mated. Until mating the rats received 0.25 μg T_3 daily. Mating was confirmed by the presence of sperm in a vaginal lavage the following morning, called day 0 of gestation.

Design of the study

Three groups of rats were used: (i) intact: intact, pregnant euthyroid rats; (ii) Tx + TH: pregnant radiothyroidectomized (Tx) rats on high T_4 and T_3 (0.25 μg T_3 and 2.5 μg T_4 per day added to the AIN diet);

(iii) Tx + lowTH: pregnant radiothyroidectomized rats on low T_4 and T_3 (0.01 μg T_3 and 0.1 μg T_4 per day added to the AIN diet).

T_4 and T_3 were purchased from Sigma (St Louis, MO, USA).

Kinetic experiments were performed on day 19 of gestation. Twenty-four hours after the kinetic experiment the animals were killed; maternal blood, liver, brain and pituitary were collected.

The experiments were approved by the University Committee on Animal Care and Use of the Agricultural University of Wageningen.

Kinetic and analytical protocols

^{125}I - T_4 and ^{131}I - T_3 (specific activity 80 and 130 MBq/ μg (2200 and 3500 $\mu\text{Ci}/\mu\text{g}$) respectively) were prepared in our laboratory (15, 16). Na^{125}I and Na^{131}I were obtained from Amersham (Arlington Heights, IL, USA); L- T_3 and 3,5-di-iodothyronine, the respective substrates for labeling, were purchased from Sigma. Purity of the tracers was assessed by means of HPLC.

Via a cannula inserted into the right jugular vein (17) the rats received a 400 μl bolus injection of a solution of 3700 Bq (10 μCi) ^{125}I - T_4 and 3700 Bq (10 μCi) ^{131}I - T_3 in saline containing heparin (0.3 U/ml; Organon, Tilburg, The Netherlands), ticarcillin (0.4 mg/ml; Ticarpen, Beecham S.A., Heppignies, Belgium) and 5% normal rat serum. Ticarcillin was added to solutions containing labeled iodothyronines to prevent eventual bacterial infections and reduce artefactual deiodination, as described by Van Doorn *et al.* (18).

Blood samples of 0.2 ml were taken at 1, 4.5, 10, 23, 44, 115, 202 and 315 min; 0.4 ml samples were drawn at 465, 600, 900, 1200 and 1440 min, being the optimal time schedule according to DiStefano *et al.* (11, 12).

Fifty to one hundred microliters plasma (in duplicate) were used to assess the total ^{131}I and ^{125}I activities. The plasma samples were extracted with ethanol/ammonia (25%)(197:3 v/v) containing 0.1 mmol/l propylthiouracil (PTU). The dried extracts were dissolved in 0.1 ml 0.2 mol/l ammonia containing carrier T_4 , T_3 and KI (1 mg/10 ml) and subjected to HPLC to separate the iodothyronines, according to the method described by Schröder-van der Elst and van der Heide (19).

The endogenous concentrations of T_4 and T_3 in plasma were measured by means of a specific rat RIA (20) of samples taken via the cannula directly before the kinetic experiments were performed.

Plasma TSH was measured by the specific RIA developed for the rat by the National Institute of Diabetes and Digestive and Kidney Diseases of the National Institute of Health (USA). Reference preparation RP-2 was used as a standard.

Calculations

The percentage doses of ^{131}I - T_3 and ^{125}I - T_4 per ml plasma were calculated from the total radioactivities

and the [¹³¹I]T₃ and [¹²⁵I]T₄ distributions on the HPLC chromatogram. The percentage doses per ml, together with the plasma volume at t(0) (11), were fitted individually to sums of $n = 1$ to 3 exponential:

$$Y(t) = A1 \exp(\lambda_1 * t) + A2 \exp(\lambda_2 * t) + A3 \exp(\lambda_3 * t)$$

using the program DIMSUM, whereby A1 is expressed in % dose/ml, λ_1 in min⁻¹ (21).

To calculate the parameters of production, distribution and metabolism, the program MAMPOOL (22) was used. The sum of the three exponential functions was fitted (by weighted least squares regression analysis) to the data collected for each individual rat and, together with the plasma T₄ and T₃ levels, substituted into the three-compartment model, according to the kinetic T₄ and T₃ studies of DiStefano *et al.* (11, 12).

All data are expressed as means $\pm$ S.E.M. Data were analyzed using the Statistical Package for Social Sciences (SPSS) (23). All data were subjected to one-way analysis of variance, and statistical differences between groups were determined by means of the modified least significant difference method.

Analytical determinations

Mitochondrial fractions from liver and brain were obtained by differential centrifugation, according to the separation scheme described by van Doorn *et al.* (18).

Liver and brain mitochondrial alpha-glycerophosphate dehydrogenase (α -GPD; EC 1.1.2.1) was measured by means of the method described by Garrib and McMurray (24).

The determination of type I deiodinase (ID-I) activity in liver homogenates was performed by the method described by Janssen *et al.* (25).

Protein was determined by the bicinchoninic acid (BCA) method (Pierce Europe, Oud Beijerland, The Netherlands) using bovine serum albumin as standard.

Results

Body weight and plasma hormones

Table 1 shows the body weight (BW) and plasma thyroid hormone levels. At the start of pregnancy all rats had a similar BW. Hypothyroidism affects the BW of the near-term pregnant rat. Tx + lowTH rats had a significantly lower BW. The BW of the fetuses of Tx + lowTH mothers was also decreased. In this group the number of viable fetuses was decreased, while more dead and resorbed fetuses were found.

Plasma T₄ was markedly decreased in the Tx + lowTH group, while in the Tx+TH group a normal T₄ level was reached.

Plasma T₃ was decreased in both Tx groups; no difference was found between the Tx + lowTH and Tx + TH groups.

Plasma TSH was highly elevated in both Tx groups. In Tx + lowTH rats the plasma TSH level was higher than in Tx + TH rats.

T₄ kinetics

The disappearance curves for T₄, based on the mean A and λ values for the three groups, are shown in Fig. 1. The slope of the regression lines for plasma was decreased in Tx + lowTH rats, meaning that T₄ disappeared from plasma more slowly in this group. The fractional turnover rates for all three pools were decreased (data not shown). As a result the transport between plasma and the fast pool was 10% of that for intact controls in Tx+TH rats and only 1% in the Tx + lowTH group.

For T₄, distribution volumes, pool sizes, and transport rates are summarized in Table 2. The plasma clearance rate was decreased in both Tx groups, but no additional decrease was found in Tx + lowTH rats. The calculated rate of production of T₄ in the Tx + TH group was similar to that found in intact rats. The low 'T₄ production' in the Tx + lowTH group resulted in an equivalent decrease in the amount of T₄ in all three pools. The distribution volumes of T₄ remained unchanged.

The rate of transport of T₄ from plasma to the fast pool and *vice versa* was markedly decreased in both Tx groups; an additional decrease was found in Tx + lowTH rats. The transport of T₄ between plasma and the slow pool was only decreased in the Tx + lowTH group. It also appeared that in these rats an increasing fraction of T₄ was transported unidirectionally from plasma to the slow tissue pool.

The transit times in all three pools were increased in both Tx groups, but no significant change in the total residence time was found (results not shown).

T₃ kinetics

The kinetic parameters for T₃ are significantly different from intact rats only in the Tx + lowTH group. The fractional rate of turnover of T₃ in plasma and the transport rates from plasma to the tissue pools were decreased (data not shown).

Most changes found in the distribution, metabolism and transport of T₃ (Table 3) were similar for the Tx + TH and Tx + lowTH groups. The plasma clearance rate for T₃ was decreased. The plasma appearance rate and the production rate for T₃ were diminished in all Tx rats. The total distribution volume as well as the distribution volume of the tissue pools was reduced only in the Tx + lowTH group. In all pools the amount of T₃ had decreased in both Tx groups. The transport of T₃ in all directions was reduced.

No significant changes, except an increase in the transit time for plasma (0.6 ± 0.06 min and 1.2 ± 0.15 min for intact and Tx + lowTH rats respectively), were found for the transit times and the total residence time of T₃.

Table 1 Body weight and plasma thyroid hormone concentrations in the intact, Tx + TH and Tx + lowTH groups. Values are means $\pm$ S.E.M.

	Intact group (n = 8)	Tx + TH group (n = 7)	Tx + lowTH group (n = 8)
Body wt on day 0 (g)	226 $\pm$ 3	229 $\pm$ 4	228 $\pm$ 5
Body wt on day 19 (g)	299 $\pm$ 7	300 $\pm$ 3	252 $\pm$ 5*#
No. of fetuses	11 $\pm$ 1	9 $\pm$ 1	7 $\pm$ 2
Fetal body wt (g)	4.24 $\pm$ 0.07	4.14 $\pm$ 0.06	3.38 $\pm$ 0.19*#
T ₄ (nmol/l)	23.6 $\pm$ 1.6	30.6 $\pm$ 3.1	4.0 $\pm$ 0.9*#
T ₃ (nmol/l)	0.50 $\pm$ 0.06	0.28 $\pm$ 0.05 [†]	0.21 $\pm$ 0.01*
TSH (ng/ml)	0.56 $\pm$ 0.08	4.57 $\pm$ 0.61 [†]	7.83 $\pm$ 0.32*#

$P < 0.05$: [†] Tx + TH vs intact, * Tx + lowTH vs intact, # Tx + lowTH vs Tx + TH.

Enzyme activities

The α -GPD activities in liver and brain are shown in Fig. 2. Hepatic α -GPD activity was reduced in the Tx + TH group; an even stronger decrease was found for the Tx + lowTH group. No effect of hypothyroidism on α -GPD activity in the brain could be demonstrated.

ID-I activity (in mOd/min/mg protein) was diminished in all Tx rats: 267 $\pm$ 13, 147 $\pm$ 16, and 73 $\pm$ 8 for intact, Tx + TH and Tx + lowTH rats respectively. Thus the strongest reduction was found in the Tx + lowTH rats.

Discussion

Thyroidectomy resulted in a lower increase in body weight during pregnancy. This is due not only to the diminished weight of the mother, but also to reduction of the fetal compartment. Treatment of the Tx animals with T₄ and T₃ (Tx + TH) normalized the body weights of mother and fetus.

Since the plasma thyroid hormone and TSH levels were not normal, the Tx + TH group was certainly not euthyroid; we describe them as mildly hypothyroid. Tx animals which received a very low dose of T₄ and T₃ (Tx + lowTH) were markedly hypothyroid, the levels of plasma T₄ and T₃ being very low.

Hypothyroidism is known to influence thyroid hormone metabolism. In this study we concentrated on the effects of hypothyroidism on maternal thyroid hormone metabolism. The most important information about the effects of hypothyroidism on the availability of maternal thyroid hormones for fetuses will be obtained from the mildly hypothyroid group; does this state already have an effect on maternal–fetal transport?

Despite normal plasma T₄ values in the mildly hypothyroid group, the transport of T₄ from plasma to the fast pool, compared with intact near-term pregnant rats, was already strongly reduced; in markedly hypothyroid rats almost no T₄ was transported to the fast pool. Previously we suggested that in pregnant rats the fast pool consists of the liver, the kidney (11, 12), and the feto–placental compartment. In intact rats pregnancy results in a pronounced increase in the transport of T₄ from plasma to the fast pool (13).

Figure 1 T₄ disappearance curves obtained with a three exponential model. (A) Plot of $\log y(t)$ (in % dose/ml) against time, with the least squares regression line on the final straight part of the curve, giving estimates of coefficient A3 and exponent λ_3 . (B) Plot of $\log [y(t) - A_3 \exp(\lambda_3 t)]$ against time, with the least squares regression line on the curve, giving estimates of coefficient A2 and exponent λ_2 . (C) Plot of $\log [y(t) - A_3 \exp(\lambda_3 t) - A_2 \exp(\lambda_2 t)]$ against time, giving estimates of coefficient A1 and exponent λ_1 . (+) Intact; (▲) Tx + TH; (●) Tx + lowTH groups.

Table 2 Parameters of distribution, volumes, pool sizes, and rates of transport of T₄ (expressed per 100 g body weight) in the intact, Tx + TH and Tx + lowTH groups. Values are means ± S.E.M.

	Intact group	Tx + TH group	Tx + lowTH group
PCR (ml/h)	1.64 ± 0.04	1.19 ± 0.07 [†]	1.26 ± 0.09*
PR (pmol/h)	38.5 ± 2.5	35.6 ± 2.7	4.7 ± 1.0* [#]
Vtotal (ml)	17.6 ± 1.0	16.6 ± 0.8	17.4 ± 2.4
Vp (ml)	4.9 ± 0.1	5.0 ± 0.1	4.8 ± 0.2
V2 (ml)	3.8 ± 0.4	3.8 ± 0.7	2.4 ± 0.5
V3 (ml)	8.9 ± 0.6	7.8 ± 0.3	10.2 ± 2.6
Qtotal (pmol)	437 ± 32	519 ± 78	64 ± 14* [#]
Qp (pmol)	116 ± 6	156 ± 20	19 ± 5* [#]
Q2 (pmol)	88 ± 9	122 ± 33	9 ± 3* [#]
Q3 (pmol)	207 ± 17	239 ± 30	36 ± 9* [#]
TRpf (pmol/h)	2542 ± 390	794 ± 177 [†]	33 ± 8*
TRfp (pmol/h)	2523 ± 389	776 ± 177 [†]	31 ± 8*
DRfo (pmol/h)	19.3 ± 1.2	17.8 ± 1.3	2.4 ± 0.5* [#]
TRps (pmol/h)	195 ± 36	143 ± 43	16 ± 4* [#]
TRsp (pmol/h)	176 ± 35	125 ± 43	13 ± 4*
DRso (pmol/h)	19.3 ± 1.2	17.8 ± 1.3	2.4 ± 0.5* [#]
% PF	92.9 ± 0.8	85.0 ± 2.3	67.2 ± 10.0*
% PS	7.1 ± 0.8	15.0 ± 2.3	32.8 ± 10.0*

PCR, plasma clearance rate; PR, production rate; V, plasma equivalent distribution volume; Q, pool quantity; TRpf, TRfp, TRps, and TRsp, rates of transport between plasma and the fast and slow pools respectively, in each direction; DRfo and DRso, disposal rate from fast and slow pool respectively; % PF and % PS, fraction of T₄ in plasma transported unidirectionally to the fast and slow tissue pools respectively; p, plasma; 2, fast pool; 3, slow pool.

P < 0.05: [†] Tx + TH vs intact, * Tx + lowTH vs intact, [#] Tx + lowTH vs Tx + TH.

It is unlikely that the total decrease in the transport of T₄ to the fast pool in mildly hypothyroid rats can be explained solely by a decrease in the transport of T₄ to the liver and kidney. This implies, therefore, that even in a mildly hypothyroid situation the transport of maternal

T₄ to the placenta can also be diminished. This is very important, because T₄ plays a major role in the development of the fetal central nervous system. Only maternal T₄ is able to mitigate fetal cerebral T₄ and T₃ deficiency before the fetal thyroid starts to function (8).

Table 3 Parameters of distribution, volumes, pool sizes, and rates of transport of T₃ (expressed per 100 g body weight) in the intact, Tx + TH and Tx + lowTH groups. Values are means ± S.E.M.

	Intact group	Tx + TH group	Tx + lowTH group
PCR (ml/h)	25.0 ± 2.4	17.6 ± 1.3 [†]	16.8 ± 1.9*
PAR (pmol/h)	12.2 ± 1.2	4.9 ± 0.9 [†]	3.6 ± 0.5*
PR (pmol/h)	13.5 ± 1.1	5.5 ± 1.0 [†]	4.2 ± 0.8*
Vtotal (ml)	201 ± 20	173 ± 13	138 ± 7*
Vp (ml)	4.8 ± 0.1	5.4 ± 0.6	5.6 ± 0.5*
V2 (ml)	28.3 ± 3.0	28.1 ± 2.7	17.3 ± 1.3* [#]
V3 (ml)	200 ± 37	139 ± 11	115 ± 7*
Qtotal (pmol)	115 ± 19	49 ± 9 [†]	29 ± 2*
Qp (pmol)	2.4 ± 0.3	1.5 ± 0.2 [†]	1.1 ± 0.1*
Q2 (pmol)	13.8 ± 1.3	7.9 ± 1.3 [†]	3.6 ± 0.3* [#]
Q3 (pmol)	99 ± 19	44 ± 12 [†]	24 ± 2*
TRpf (pmol/h)	205 ± 27	115 ± 21 [†]	50 ± 6*
TRfp (pmol/h)	199 ± 27	112 ± 20 [†]	48 ± 6*
DRfo (pmol/h)	6.1 ± 0.6	2.5 ± 0.5 [†]	1.8 ± 0.3*
TRps (pmol/h)	53.3 ± 13.4	16.5 ± 2.6 [†]	10.3 ± 0.8*
TRsp (pmol/h)	54.9 ± 13.8	16.9 ± 2.7 [†]	10.6 ± 0.9*
DRso (pmol/h)	7.4 ± 0.6	3.0 ± 0.6 [†]	2.5 ± 0.5*
% PF	80.5 ± 3.4	86.7 ± 1.5	81.8 ± 2.2
% PS	19.5 ± 3.4	13.3 ± 1.5	18.2 ± 2.2

PCR, plasma clearance rate; PAR, plasma appearance rate; PR, production rate; V, plasma equivalent distribution volume; Q, pool quantity; TRpf, TRfp, TRps and TRsp, rates of transport between plasma and the fast and slow pools respectively, in each direction; DRfo and DRso, disposal rate from fast and slow pool respectively; % PF and % PS, fraction of T₃ in plasma transported unidirectionally to the fast and slow tissue pools respectively; p, plasma; 2, fast pool; 3, slow pool.

P < 0.05: [†] Tx + TH vs intact, * Tx + lowTH vs intact, [#] Tx + lowTH vs Tx + TH.

Figure 2 α -GPD in maternal liver and maternal brain in the three groups studied (intact, Tx + TH, Tx + lowTH). $P < 0.05$: [&]Tx + TH vs intact; ^{*}Tx + lowTH vs intact; [#]Tx + lowTH vs Tx + TH.

The plasma T_3 level in the mildly hypothyroid group was not normal. The amount of T_3 added was less than the amount of T_3 produced daily in intact rats ($0.25 \mu\text{g}$ vs $0.60 \mu\text{g}$ per day). However, since the plasma T_4 level was normal, it was to be expected that T_3 would also be produced locally. T_3 produced locally in the liver contributes in particular to the plasma T_3 level (18). However, in the hypothyroid rat the local production of T_3 is decreased in the liver (18). This was also demonstrated by the decrease in ID-I activity in the liver; this can possibly be explained by the regulation of hepatic ID-I activity by T_3 (26). Therefore, even in a mildly hypothyroid condition with normal plasma T_4 levels, the T_3 level in the liver will be reduced; this hypothesis is supported by the decrease in hepatic α -GPD activity.

For non-pregnant thyroidectomized rats receiving an infusion of T_4 and T_3 it has been reported that in the event of normal plasma T_4 levels and a reduced plasma T_3 , hepatic ID-I and cerebral ID-II activities will be normal (10). However, despite the administration of T_4 in an amount which equals the amount normally

produced, local T_3 production was decreased in our near-term pregnant rats. It appears, therefore, that in near-term pregnant rats the effect of mild hypothyroidism is stronger than in non-pregnant rats.

Marked hypothyroidism in both non-pregnant and near-term pregnant rats results in highly reduced pools and diminished rates of transport of T_4 and T_3 .

In conclusion, even mild hypothyroidism results in significant alterations in maternal thyroid hormone metabolism, despite normal plasma T_4 levels. In particular the transport of T_4 from plasma to the fast pool will be reduced. We speculate that this results in a decrease in the availability of maternal T_4 for the fetuses. This could affect the development of the fetal brain, which depends mainly on maternal T_4 for normal T_3 values.

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