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Placental Growth Hormone Levels in Normal Pregnancy and in Pregnancies with Intrauterine Growth Retardation

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ABSTRACT. To assess the possible role of placental growth hormone (GH) in fetoplacental growth, we measured placental and pituitary GH (GHN) in maternal plasma by means of two RIA using two MAb (5B4 recognizing both placental GH and GHN, and K24 recognizing only GHN) during pregnancy. IGF-I also was measured by RIA in the same samples after extraction. A transverse study of 186 samples obtained between 8 wk of amenorrhea (WA) and term confirmed the reported rise in GH immunoreactivity with 5B4 after 24 to 25 WA from 12.3 ± 2.0 mU/L (mean ± SEM) to a plateau of 27.5 ± 3.4 mU/L at 34 to 35 WA together with the decrease in GHN to undetectable levels by 24 to 25 WA. IGF-I levels increased from 164.0 ± 44.6 μg/L at 24 to 25 WA to 331.6 ± 63.6 μg/L at term. A longitudinal study of 31 normal pregnant women confirmed this hormonal pattern and the reported placental GH plateau after 35 WA. A drastic decrease in placental GH was observed with the onset of labor (from 26.9 ± 2.1 to 2.7 ± 1.1 mU/L), whereas the decrease in IGF-I was not significant (from 212.9 ± 26.5 to 162.4 ± 16.9 μg/L). Interestingly, maternal plasma samples obtained after 31 WA until the initiation of labor in 22 cases of intrauterine growth retardation (six cases of toxemia, one chromosomal aberration, one maternofetal infection, 14 idiopathic) contained significantly lower amounts of placental GH (14.9 ± 1.6 mU/L *versus* 26.5 ± 1.2 mU/L in normal pregnancies; $p < 0.001$). Plasma IGF-I levels were also lower than normal (156.0 ± 25.5 μg/L *versus* 285.1 ± 40.8 μg/L; $p < 0.001$). These results suggest a relationship between placental GH levels in the maternal plasma and the development of the fetoplacental unit. (*Pediatr Res* 34: 439–442, 1993)

Abbreviations

GH, growth hormone
GHN, pituitary growth hormone
IUGR, intrauterine growth retardation
WA, weeks of amenorrhea

The placenta and the fetus form a functional unit during pregnancy. The correlation between placental size in late gestation and fetal growth is well established (1), but the placental-fetal relationship is highly complex. The human placenta recently has been shown to express the GH-V gene specifically, leading to the production of placental GH (2–4).

Placental GH differs from GHN by 13 amino acid residues, is more basic than GHN, and contains a unique N-linked glycosylation site at asparagine 140 (5–7). Placental GH is produced by the syncytiotrophoblast *in vivo* (8, 9) and *in vitro* (10), and we recently have demonstrated that this hormone is involved in autocrine mechanism in the syncytiotrophoblast (11).

Placental GH binds to a GH-binding protein (12) and can be biologically active mainly as a somatogen but also as a lactogen (13). Placental GH can be detected in the maternal blood and is distinguishable from GHN on the basis of its reactivity with two MAb (14). However, the precise function of placental GH is unknown.

IUGR for gestational age is a major clinical concern: An exponential relationship exists between the degree of growth retardation and perinatal morbidity and mortality. However, the endocrine factors accounting for this abnormal growth are poorly understood.

To assess the possible role of placental GH in fetoplacental growth, placental GH was measured in transverse and longitudinal studies during the course of normal pregnancies, together with maternal IGF-I. The levels were compared with those observed during fetal growth retardation of various origin diagnosed during the 3rd trimester of pregnancy.

MATERIALS AND METHODS

Subjects. Transverse study. Plasma samples were collected from healthy pregnant women from 8 WA to full term ($n = 186$) who were referred to the maternity unit of the Hôpital Saint-Vincent de Paul (Paris). The length of pregnancy was determined as: n wk of gestation = n − 2 WA. All these women gave birth to healthy infants with a normal birth weight (mean, 3299 ± 449 g) according to the Leroy-Lefort curve (15). The study was approved by the local ethics committee.

Longitudinal study. Blood samples were taken every 4 to 6 wk between 15 and 40 WA from 31 healthy pregnant women. In 12 cases, blood was obtained 2 to 4 d before delivery and at the onset of labor (contractions monitored on a tocometer and dilating the cervix). Blood was collected in EDTA vacutainers and immediately centrifuged; plasma was stored at −20°C until assay.

IUGR group. These patients ($n = 22$) were selected on the basis of clinical findings (small uterine height) confirmed by echography (<3rd percentile on Lubchenko curves) during the 3rd trimester of pregnancy (range, 33 to 39 WA). The routine

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etiologic study included morphologic echography and Doppler ultrasound studies and tests for maternal hypertension and infection. Fetal karyotyping was carried out in two cases with echographic abnormalities.

The birth weight of all these infants was below the 3rd percentile on the Leroy-Lefort chart (15). Among them 11 had an height below the 3rd percentile and the others had an height below the 25th percentile. The head circumference of these infants ranged from the 3rd to the 30th percentile on the Leroy-Lefort chart (15). The placental weights were correlated to the birth weights. Toxemia, defined as the association of maternal hypertension, lower-limb edema, and proteinuria, was found in six cases. Maternofetal cytomegalovirus infection was detectable in one case, and fetal malformation (trisomia 18) was found in another case. The 14 remaining cases were considered idiopathic.

Hormone assays. GH was measured as previously described (10) by means of two RIA using two MAb (5B4 and K24) raised against purified hGH. The immunochemistry of the MAb and the characteristics of the relevant RIA have been reported elsewhere (14). Briefly, the affinity constants of the binding reaction with 22-kD hGH were 5×10^9 M^{-1} and 1.02×10^{11} M^{-1} for K24 and 5B4, respectively. The detection limits of the assays were 1 mU/L (K24 RIA) and 0.5 mU/L (5B4 RIA). MAb 5B4 is directed toward an N-terminal epitope and recognizes all known placental GH and GHN variants. MAb K24 reacts with a more internal epitope, recognizing the 22-kD GHN variant but not the placental one. Placental GH thus can be distinguished from GHN by its lack of reactivity with K24. Cross-reactivity with human chorionic somatomammotropin is <0.005% in both systems.

IGF-I was measured after acidic extraction of the plasma by means of RIA with a specific monoclonal anti-IGF-I antibody (1 IU corresponds to 192 μg/L) (16).

Data analyses. Data are expressed as mean ± SEM. Significant differences were identified with *t* test at a threshold of $p < 0.05$.

RESULTS

As shown in Figure 1*A*, the transversal study of 186 individual samples obtained from 8 to 10 WA to full term confirmed the reported rise in GH immunoreactivity with MAb 5B4 after 24 to 25 WA (14) from 12.3 ± 2.0 mU/L to a plateau of 27.5 ± 3.4 mU/L at 34 to 35 WA and the corresponding decrease in GH immunoreactivity with K24 from 2.7 ± 1.1 mU/L to below the detection limit by 24 to 25 WA. After 24 to 25 WA, GH immunoreactivity measured with the 5B4 thus corresponded to mainly placental GH. IGF-I levels did not vary significantly during the first weeks of gestation but increased progressively from 164.0 ± 44.6 μg/L at 24 to 25 WA to 331.6 ± 63.6 μg/L at term (Fig. 1*B*). There was a weak correlation between individual IGF-I values and 5B4-immunoreactive GH from 26 wk of gestation to term ($p < 0.04$). Mean values of placental GH and IGF-I levels at each stage of pregnancy were significantly correlated ($p < 0.01$). Figure 2*A* and *B* shows two representative examples among 31 of GH immunoreactivity with 5B4 and K24 in the longitudinal study. The shaded area of the figure represents the mean ± SD of plasma GH during normal pregnancy from the cross-sectional study. These two examples were chosen to show the great variability of 5B4 immunoreactivity profile from one normal pregnant woman to another. However, when this longitudinal study in 31 women was analyzed in a transversal way, it confirmed the hormone pattern observed in the transverse study, as well as the plateau of placental GH after 34 WA (Fig. 2*C*).

Interestingly, there was a drastic decrease in placental GH at the onset of labor (from 26.9 ± 2.1 mU/L to 2.7 ± 1.1 mU/L). In contrast, IGF-I levels did not vary significantly (212.9 ± 26.5 to 162.4 ± 16.9 μg/L).

Given this drop in placental GH with the onset of labor, the placental GH pattern in the women with IUGR was studied during the 3rd trimester of pregnancy but some time before labor. As shown in Figure 3, there was a significant decrease in placental GH (5B4 immunoreactivity) levels after 33 WA ($p < 0.001$) relative to normal pregnancies (14.9 ± 1.6 mU/L *versus* 26.5 ± 1.2 mU/L). IGF-I levels also were significantly decreased in women with IUGR (mean ± SEM; 156.0 ± 25.5 μg/L) relative to normal pregnancies (285.1 ± 40.8 μg/L). According to the IUGR etiologies, there was no significant difference between placental GH and IGF-I maternal levels in IUGR associated with maternal toxemia ($n = 6$) and IUGR of unknown causes (idiopathic IUGR, $n = 14$).

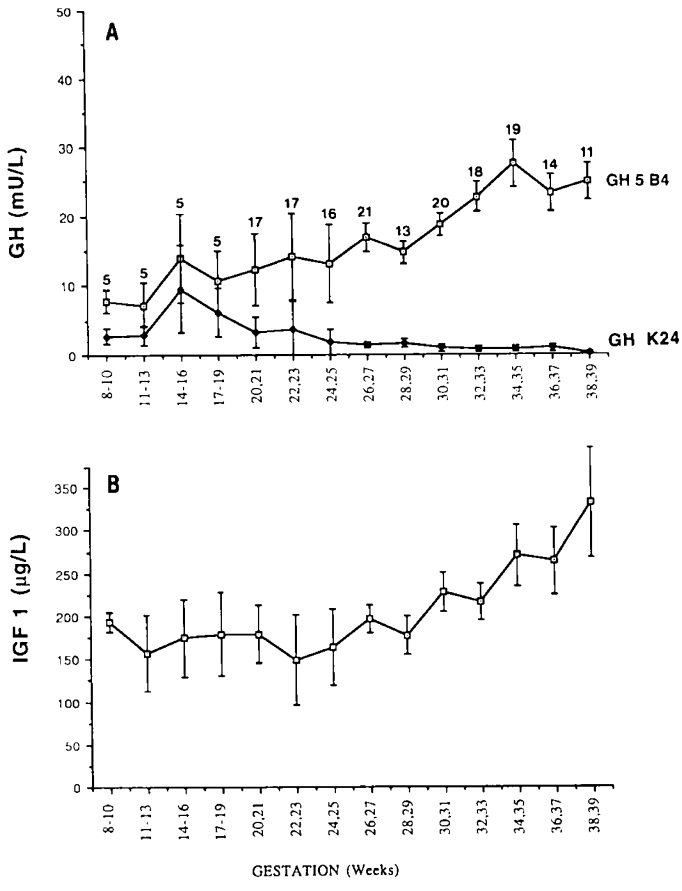

Fig. 1. Transverse study of maternal plasma GH (*A*) and IGF-I (*B*) levels during pregnancy ($n = 186$). Each point represents the mean ± SEM of values from individual samples obtained in pregnant women at the indicated periods of pregnancy expressed in WA. Each period consists of 3 wk until the 20th wk and then consists of 2 wk thereafter. Number of individual assays in GH and IGF-I for each gestational stage is indicated in *panel A* on *top of the vertical bars.*

DISCUSSION

In the transverse study, we observed a progressive increase in placental GH levels in the maternal plasma during pregnancy, and a corresponding decrease in GHN to undetectable levels. The increase in placental GH paralleled that in IGF-I. There were large individual differences in the pattern of placental GH in the longitudinal study, but both GH and IGF-I were significantly reduced in the maternal plasma in the cases of IUGR.

It is well established that IGF-I levels increase gradually in pregnant women, but the factors involved are not fully understood (17, 18). Maternal IGF-I is mainly synthesized by the liver (19), but perfusion studies of the placenta *in vitro* have suggested that some placental IGF-I is secreted into the maternal circulation (20). Interestingly, maternal IGF-I levels are not under the control of pituitary GH during pregnancy as shown by studies of acromegalic women in whom, despite the apparent stability

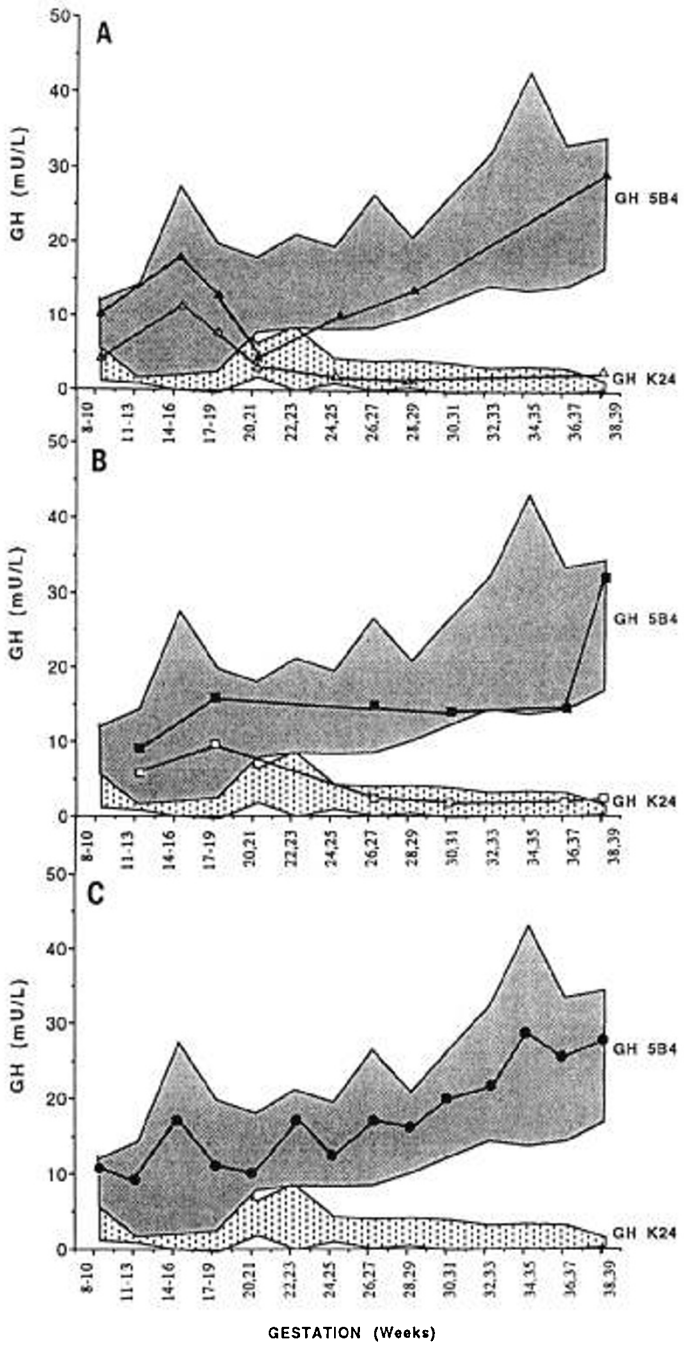

Fig. 2. *A* and *B*, Two representative examples of individual variations in plasma GH levels in pregnant women. *C*, Mean values of plasma GH levels obtained from the longitudinal study in 31 pregnant women. *Shaded areas* show means ± SD of plasma GH values during normal pregnancy obtained from the cross-sectional study. Gestational age is expressed in WA (see legend of Fig. 1).

of GHN levels, serum IGF-I levels increase during pregnancy (21).

The physiologic role of placental GH is unknown but a direct action on fetal growth seems unlikely because placental GH is not detected in the fetal circulation (22). However, placental GH could indirectly modulate fetal growth either by interfering with the maternal metabolism, *e.g.* by increasing IGF-I levels, or by modulating placental development. Indeed, during the last trimester of pregnancy, fetal growth is normally constrained by maternal factors (23, 24). In addition, we recently have shown that placental GH is subject to autocrine control within the syncytiotrophoblast (10), which produces placental GH and expresses GH receptors (11).

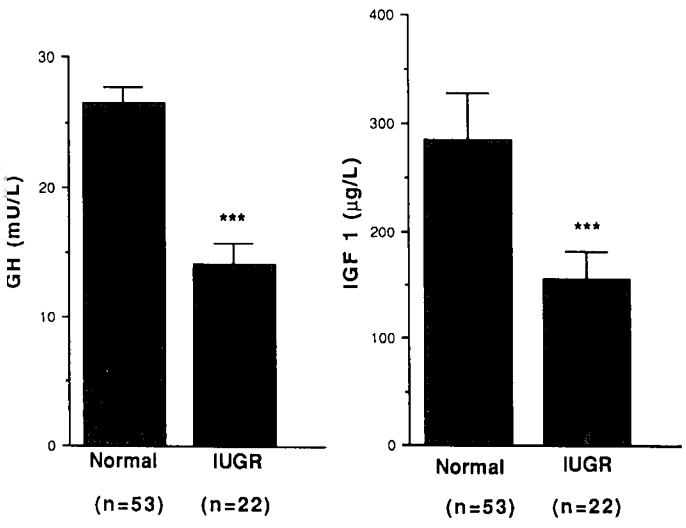

Fig. 3. Maternal plasma placental GH (5B4 immunoreactivity) and IGF-I levels in normal ($n = 53$) and IUGR ($n = 22$) pregnancies between 33 WA and term. *Vertical bars* represent the mean ± SEM. ***, Significantly different from normal values ($p < 0.001$).

Placental GH levels fell drastically with the onset of labor, probably due to the decrease in uteroplacental blood flow (25, 26). In addition, a release of proteases by the placenta cannot be ruled out because placental GH has a short half-life (14). In contrast, the maternal level of IGF-I was not significantly modified by the onset of labor. This difference may be due to the longer half-life of IGF-I or to the fact that not all maternal IGF-I is produced by the placenta (23). A larger study would be necessary to confirm these results and to exclude a significant decrease of IGF-I during labor, obscured in this study by the large variations of IGF-I levels in the controls.

We found a significant decrease in placental GH levels in the women with intrauterine growth retarded fetus. After 26 wk of gestation (28 WA), *i.e.* during the last trimester of pregnancy when most cases of IUGR are diagnosed, maternal circulating GH derives essentially from the placenta and is significantly reduced in cases of IUGR relative to normal pregnancies. No difference was found in the levels of placental GH in terms of etiology of IUGR. This suggests that placental GH levels reflect placental biologic activity and might be useful in assessing the chronic fetal distress associated with abnormal placental structure and/or function. Further studies are necessary to determine whether placental GH levels in the maternal plasma of pregnant women might be of value for the diagnosis and the follow-up of IUGR.

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