MATURATION OF THE SECRETION OF THYROID HORMONE AND THYROID-STIMULATING HORMONE IN THE FETUS

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Abstract *Background.* Data on human fetal thyroid function have largely been derived from histologic studies or studies of cord-blood samples obtained at hysterotomy or delivery. These data may not represent true normal values. Cordocentesis (ultrasound-guided blood sampling from the umbilical cord) is a technique that allows investigation of physiologic processes in fetuses not under stress.

Methods. We measured serum thyroid-stimulating hormone, total and free thyroxine (T_4), total and free triiodothyronine (T_3), and thyroxine-binding globulin in blood samples from 62 fetuses. The samples were obtained by cordocentesis (n = 58) or cardiocentesis (n = 4) at 12 to 37 weeks of gestation. Maternal serum samples were obtained immediately before fetal blood sampling.

Results. Fetal serum thyroid-stimulating hormone, thyroxine-binding globulin, and total and free T_4 and T_3 concentrations increased significantly with the length of gestation ($P<0.001$). The only significant association among these variables, independent of the length of gestation, was between thyroid-stimulating hormone and free T_4 ($P<0.0001$). Maternal serum concentrations of these variables did not change during gestation, and there was no significant relation between fetal and maternal values. Most fetal serum concentrations of thyroid-stimulating hormone were higher, whereas most serum total and free T_3 concentrations were lower than the respective values for normal adults. The fetal serum total T_4, free T_4, and thyroxine-binding globulin values reached the level of the mean adult values at approximately 36 weeks of gestation.

Conclusions. The increases in fetal serum concentrations of thyroid-stimulating hormone, thyroxine-binding globulin, and total and free T_4 and T_3 during gestation reflect increasing maturation of the pituitary, thyroid, and liver. The finding of increasing fetal serum concentrations of thyroid-stimulating hormone in the presence of increasing thyroid hormone concentrations suggests that the sensitivity of the fetal pituitary gland to negative feedback is limited or is counterbalanced by increasing stimulation by thyrotropin-releasing hormone from the hypothalamus. (N Engl J Med 1991; 324:532-6.)

THYROID hormones have a vital role in fetal development in many species. The best studied are fetal rats and sheep, in which thyroid hormones have important physiologic functions, including stimulation of neural growth and pulmonary development.[1] What is known about pituitary and thyroid development in humans is based on histologic studies of abortuses or analysis of blood samples obtained in early pregnancy at hysterotomy for elective abortion or in later pregnancy at delivery.[2-5] However, the results derived from samples obtained at the time of hysterotomy or cesarean section may have been influenced by maternal fasting or transient hypotension, which could alter placental perfusion and therefore affect the supply of oxygen and nutrients to the fetus.[6] Furthermore, samples obtained after premature delivery may not be representative of normal prelabor values, since thyroid-stimulating hormone levels undergo marked and rapid changes in the immediate postnatal period. Indeed, the condition causing premature delivery itself could influence fetal serum thyroid-stimulating hormone and thyroid hormone levels, since it is unlikely that fetuses delivered before 37 weeks of gestation can truly be described as normal. Despite their limitations, such studies indicate that the thyroid gland begins to produce thyroxine at 10 to 12 weeks of gestation.[7,8] However, there is conflicting evidence about the time during gestation at which functional maturation of the fetal pituitary–thyroid axis is achieved. Some authors suggest that the secretion of thyroid-stimulating hormone is responsive to changes in serum free thyroxine (T_4) concentrations as early as 11 weeks of gestation[2]; others suggest that such maturation occurs largely during the last half of pregnancy.[3,4,9]

Recently, Ballabio et al. used cordocentesis — that is, ultrasound-guided blood sampling from the umbilical cord — to study thyroid function at 18 to 31 weeks of gestation.[10] However, only 23 fetuses were studied, 15 of whom were severely anemic as a result of red-cell isoimmunization. Nevertheless, the study demonstrated that fetal serum thyroid-stimulating hormone, total T_4, and free T_4 concentrations increased during gestation. Fetal serum thyroid-stimulating hormone levels were always higher and total T_4 levels always lower than adult values, whereas free T_4 values reached adult levels by 28 weeks of gestation. Ballabio et al. explained these findings by suggesting that the threshold for negative feedback from thyroid hormones to the pituitary is set at a higher level in fetal than in postnatal life.

The aim of this study was to establish reference ranges for fetal serum thyroid hormone and thyroid-stimulating hormone concentrations and examine the interrelation of these hormones at 12 to 37 weeks of gestation in normal fetuses.

Methods

Blood was obtained by cordocentesis (17 to 37 weeks of gestation) or cardiocentesis (<14 weeks of gestation) from 62 fetuses.[11] Blood was collected from the antecubital fossa in 52 of the mothers immediately before fetal blood sampling. Cordocentesis was used to obtain blood from 58 fetuses for the following purposes: prenatal diagnosis of blood disorders such as hemophilia A (n = 9); fetal karyotyping in women of advanced age or with a low serum alpha-fetoprotein level when amniocyte culture had been unsuccessful

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or when results were needed rapidly (n = 14); karyotyping for fetal malformations, such as mild hydronephrosis, unilateral polycystic kidney, or congenital diaphragmatic hernia (n = 24); investigation of maternal primary toxoplasmosis (n = 2); and fetal blood typing of those at risk for red-cell isoimmunization, in which the fetal blood was found to be negative on Coombs' testing (n = 9). In all cases, the fetal abdominal circumference and blood gas values at the time of cordocentesis were within our reference ranges for the length of gestation and the fetal karyotype was normal. Furthermore, the fetuses did not have the blood disorder or infection for which they were tested. Blood was obtained from four fetuses by cardiocentesis immediately before the intracardiac injection of potassium chloride for feticide in multifetal pregnancies. Kleihauer–Betke testing confirmed that all samples contained only fetal blood. The project was approved by the hospital ethics committee, and informed consent was obtained from all the mothers.

For measurements of serum thyroid-stimulating hormone, thyroxine-binding globulin, and thyroid hormones, fetal (0.3 to 0.8 ml) and maternal (5 ml) blood samples were collected in plain tubes and centrifuged for 10 minutes at 2000 rpm; the serum was then collected and stored at −20°C. Thyroid-stimulating hormone was measured in all fetal serum samples, and total T_4, free T_4, total triiodothyronine (T_3), free T_3, and thyroxine-binding globulin were measured in most samples. Thyroid-stimulating hormone was measured by immunoradiometric assay (Celltech Diagnostics, Slough, United Kingdom). Total T_4 and T_3 were measured by solid-phase radioimmunoassays; free T_4 and T_3 were measured by solid-phase analogue radioimmunoassays (Diagnostic Products, Los Angeles). Thyroxine-binding globulin was also measured by radioimmunoassay (Behring, Marburg, Germany). The manufacturers of the kits used for the measurement of free T_4 and T_3 have demonstrated a lack of interference by thyroxine-binding globulin (up to 0.86 mmol per liter), albumin (up to 1.2 mmol per liter), or nonesterified fatty acids (up to 10 mmol per liter). The interassay and intraassay coefficients of variation for the thyroid-stimulating hormone, thyroxine-binding globulin, total T_4, free T_4, total T_3, and free T_3 assays were 4.5 and 3.9 percent, 4.5 and 3.4 percent, 4.9 and 4.1 percent, 6.5 and 4.4 percent, 6.6 and 5.4 percent, and 4.8 and 4.5 percent, respectively. The ranges in nonpregnant adults (mean ±2 SD) used for thyroid-stimulating hormone, thyroxine-binding globulin, total T_4, free T_4, total T_3, and free T_3 were those of the test manufacturers and were obtained from 347, 368, 335, 251, 335, and 405 normal subjects, respectively.

Regression analysis was used to examine any relation between measured variables and gestational age. Data or residuals from linear regression were tested for normality. For the measurements that were not distributed normally, the distribution was made to conform to a gaussian curve by logarithmic transformation. For those that changed significantly with the length of gestation, the regression lines were used to calculate the adjusted means and residual standard deviations. To determine the reference ranges during gestation in the original units, the limits of the calculated reference range in logarithms were subjected to antilogarithmic transformation.

Results

Fetal serum concentrations of thyroid-stimulating hormone (Fig. 1), total and free T_4 (Fig. 2), total and free T_3 (Fig. 3), and thyroxine-binding globulin (Fig. 4) increased progressively during gestation, and the associations between each of them and the length of gestation were significant. Although there were also significant associations among these variables (data not shown), multiple regression analysis demonstrated that the only significant association independent of the length of gestation was that between serum thyroid-stimulating hormone and free T_4 (free T_4 = −1.0873 + 0.768 [thyroid-stimulating hormone] + 0.042 week of gestation; R = 0.896, P<0.0001).

There were no changes in maternal hormone or thyroxine-binding globulin concentrations as a function of the length of gestation (Fig. 1 through 4). Likewise, there were no significant associations between the values in maternal serum and those in fetal serum.

The vertical lines at the right side of each figure are the mean (±2 SD) serum concentrations in normal nonpregnant adults. Most fetal serum thyroid-stimu-

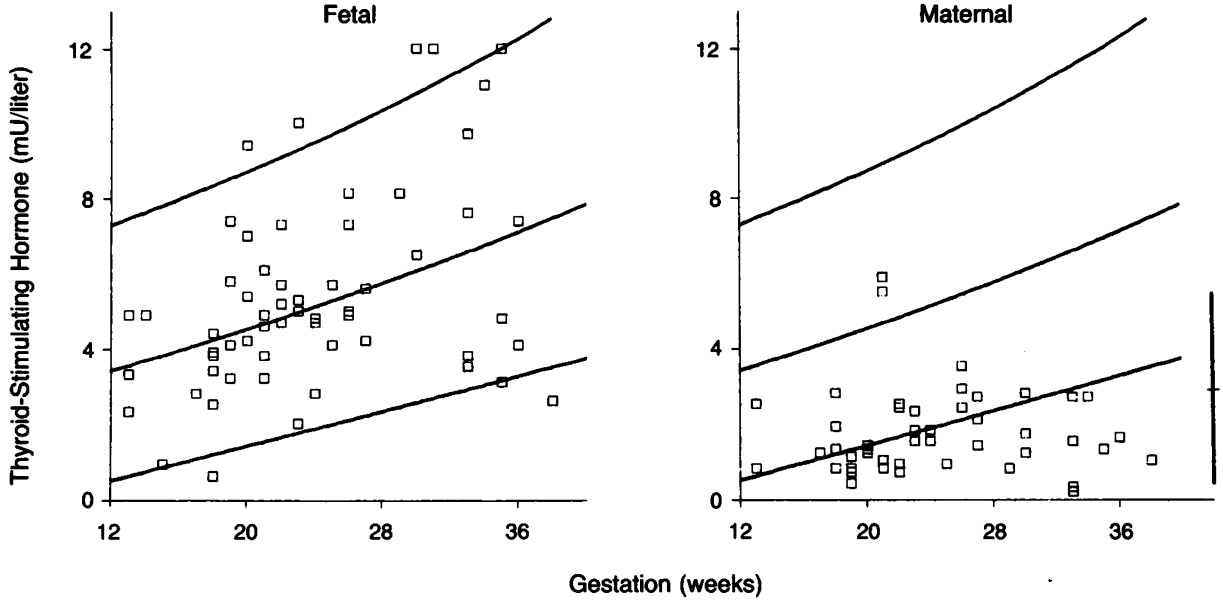

Figure 1. Individual Fetal and Maternal Serum Thyroid-Stimulating Hormone Concentrations Plotted as a Function of Length of Gestation.

The sloping lines are the mean and 5th and 95th percentile values for fetal serum thyroid-stimulating hormone (r = 0.415, n = 62, P<0.001). The maternal serum concentrations of thyroid-stimulating hormone did not change significantly with the length of gestation (r = −0.021, n = 46). The vertical line on the right is the normal range in nonpregnant adults (mean, 2.9 mU per liter; range, 0.4 to 5.5).

lating hormone values were higher and most fetal serum total and free T_3 values were lower than the adult values. The fetal serum total T_4, free T_4, and thyroxine-binding globulin values reached the level of the mean adult values at approximately 36 weeks of gestation.

Discussion

Maternal serum levels of thyroid hormones are higher than those in nonpregnant adults. The increases in serum total T_4 and T_3 concentrations are primarily a consequence of an estrogen-induced elevation of serum thyroxine-binding globulin levels. Thyroid stimulation due to the weak thyroid-stimulating hormone–like activity of human chorionic gonadotropin contributes to the increase in total T_4 and T_3 concentrations and accounts for the slight increase in free T_4 and T_3 concentrations.[12-14] Although in this study the maternal levels of thyroid-stimulating hormone did not change significantly with the length of gestation, in a larger series Fung et al. reported that the levels increased with increasing lengths of gestation.[15] The absence of a significant correlation between fetal and maternal thyroid hormone and thyroid-stimulating hormone levels is compatible with the results of previous studies of cord-blood and maternal serum thyroid hormone levels at delivery.[1]

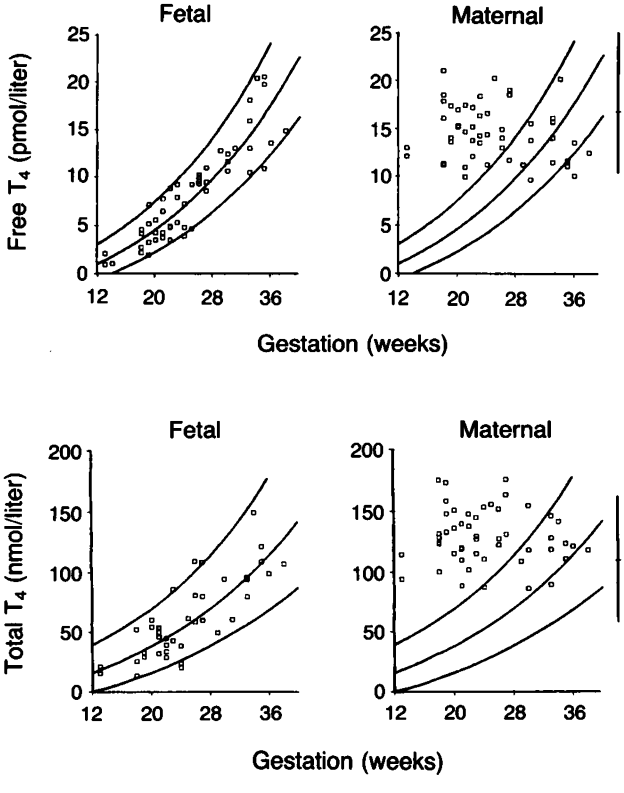

Figure 2. Individual Fetal and Maternal Serum Free and Total T_4 Concentrations Plotted as a Function of Length of Gestation.

The curved lines are the mean and 5th and 95th percentile values for fetal serum free T_4 ($r = 0.894$, $n = 56$, $P<0.0001$) and total T_4 ($r = 0.804$, $n = 41$, $P<0.0001$). The maternal serum free T_4 ($r = -0.022$, $n = 52$) and total T_4 ($r = 0.015$, $n = 50$) concentrations did not change significantly with the length of gestation. The vertical lines on the right are the normal ranges in nonpregnant adults (free T_4: mean, 18 pmol per liter; range, 10.3 to 25.7; total T_4: mean, 109 nmol per liter; range, 58 to 161).

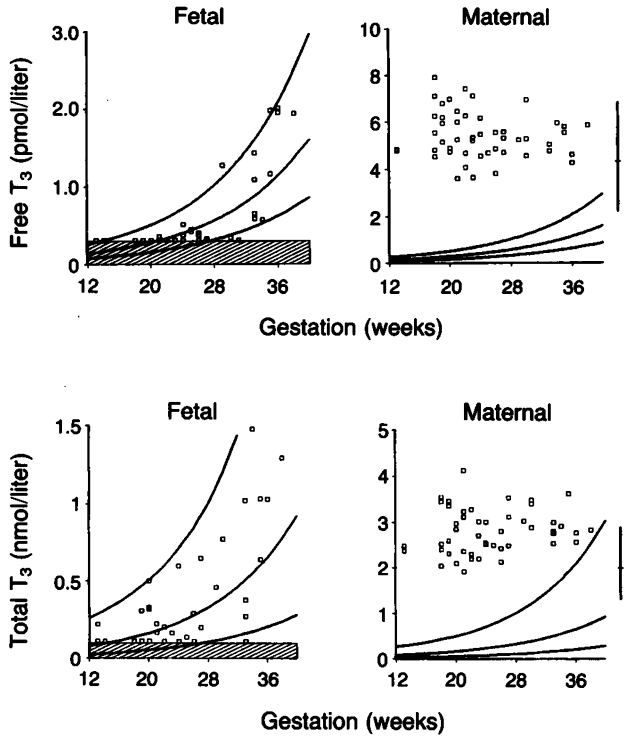

Figure 3. Individual Fetal and Maternal Serum Free and Total T_3 Concentrations Plotted as a Function of Length of Gestation.

The curved lines are the mean and 5th and 95th percentile values for fetal serum free T_3 ($r = 0.833$, $n = 49$, $P<0.0001$) and total T_3 ($r = 0.667$, $n = 39$, $P<0.0001$). The maternal serum free T_3 ($r = -0.176$, $n = 52$) and total T_3 ($r = 0.173$, $n = 52$) concentrations did not change significantly with the length of gestation. The vertical lines on the right are the normal ranges in nonpregnant adults (free T_3: mean, 4.5 pmol per liter; range, 2.2 to 6.8; total T_3: mean, 2.1 nmol per liter; range, 1.3 to 2.9). Hatched areas represent the lower limits of sensitivity of the assays.

The increase in the fetal serum thyroid hormone concentrations during gestation presumably reflects the increasing stimulation and maturation of the fetal thyroid gland and increasing serum thyroxine-binding globulin concentrations. However, although fetal serum total and free T_4 concentrations reached adult levels by 36 weeks of gestation, fetal serum total and free T_3 concentrations were always less than half the respective maternal concentrations. Since the chief source of T_3 is peripheral conversion of T_4, these findings suggest that during intrauterine life the processes required for this conversion either are immature or lack the necessary stimulus for their activation. In vitro studies in sheep have demonstrated that during labor and in the neonatal period there is a dramatic increase in the capacity for hepatic conversion of T_4 to T_3.[16] Similarly, Fisher et al. found a 10-fold increase in the ratio of serum T_3 to T_4 from 30 weeks of gestation to 1 month after birth in humans.[1] An alternative explanation for the low fetal T_3 concentrations could be rapid deiodination by the placenta.

Fetal serum concentrations of thyroid-stimulating hormone increased significantly during gestation and

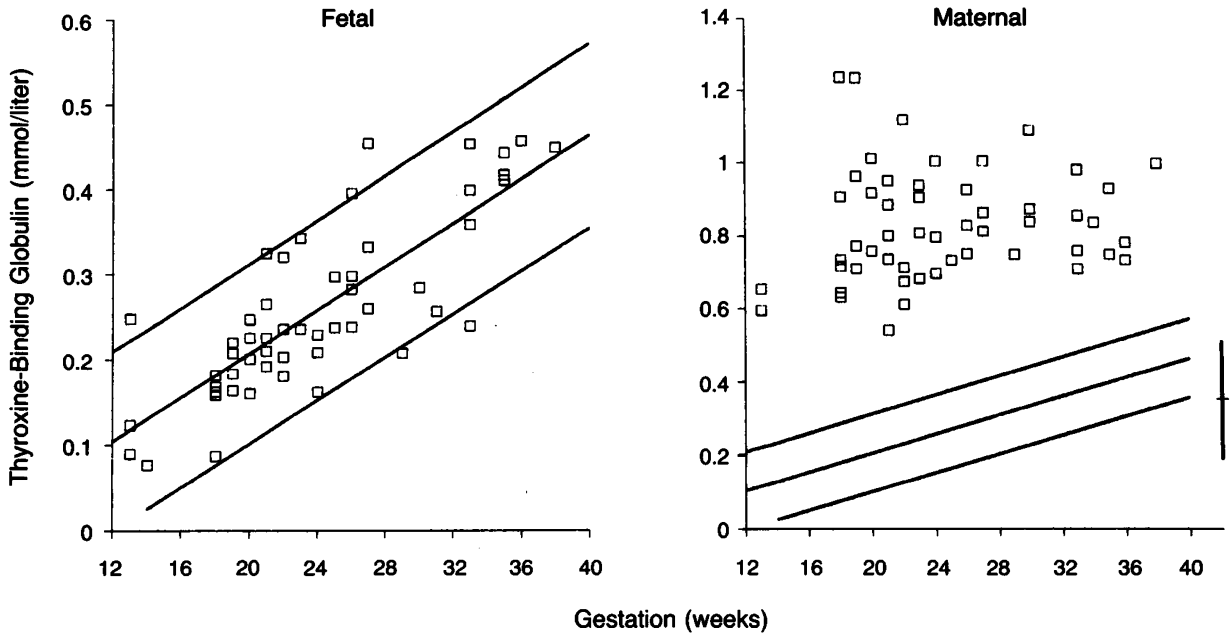

Figure 4. Individual Fetal and Maternal Serum Thyroxine-Binding Globulin Concentrations Plotted as a Function of Length of Gestation. The sloping lines are the mean and 5th and 95th percentile values for fetal serum thyroxine-binding globulin ($r = 0.805$, $n = 52$, $P<0.0001$). The maternal serum concentrations of thyroxine-binding globulin did not change significantly with the length of gestation ($r = 0.13$, $n = 52$). The vertical line on the right is the normal range in nonpregnant adults (mean, 0.35 μmol per liter; range, 0.19 to 0.51).

were always higher than maternal levels. After birth the chief determinant of negative feedback is T_3, produced largely by intrapituitary local monodeiodination of T_4.[17] In this study, there was a positive association between serum free T_4 and thyroid-stimulating hormone that was independent of the length of gestation, and thyroid-stimulating hormone levels continued to rise even in the third trimester, when adult concentrations of total and free T_4 were reached. These findings suggest that the fetal pituitary is relatively insensitive to negative feedback from T_4 or that the inhibitory action of T_4 on the secretion of thyroid-stimulating hormone is counterbalanced by increasing secretion of thyrotropin-releasing hormone from the hypothalamus. Alternatively, circulating T_3 has a more important role in feedback in the fetus than in the adult; since serum total and free T_3 concentrations are much lower before than after birth, the threshold for negative feedback is never reached in utero.

Fetal serum concentrations of thyroxine-binding globulin increased with increasing lengths of gestation and reached adult levels during the third trimester. This increase presumably reflects the functional maturation of the fetal liver and its increasing capacity to manufacture proteins; fetal albumin concentrations also increase with gestation.[18]

Our results support the findings of Greenberg et al. that serum concentrations of thyroid-stimulating hormone and total and free T_4 increase between 11 and 24 weeks of gestation. However, adult levels were not reached by 16 to 20 weeks of gestation, as they reported.[2] Similarly, although our results are in general agreement with those of Fisher et al., we did not confirm their finding that fetal serum concentrations of thyroid-stimulating hormone rise between 12 and 24 weeks of gestation but not thereafter.[1] Our results agree most closely with those of Ballabio et al.,[10] who also obtained blood by cordocentesis and who similarly demonstrated pituitary resistance to increasing serum thyroid hormone concentrations.

During the second trimester of pregnancy, it is likely that the increase in fetal serum concentrations of thyroid-stimulating hormone, thyroid hormones, and thyroxine-binding globulin reflects the independent maturation of the pituitary, thyroid, and liver, respectively. With longer gestation there is an increase in serum thyroid-hormone levels, reflecting functional maturation of the thyroid gland and increasing serum thyroxine-binding globulin concentrations. Despite this increase, the serum total and free T_3 concentrations were always lower before birth than after birth, and production of thyroid-stimulating hormone was not inhibited.

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SPECIAL ARTICLE

BOARD-CERTIFIED PHYSICIANS IN THE UNITED STATES, 1971–1986

FRANCIS D. MOORE, M.D., AND CEDRIC PRIEBE, M.D.

Abstract *Background.* This is our third report covering the census of U.S. physicians over a 15-year period. The present report updates the information for 1980 to 1986.

Methods. Most of our data are based on published information from the Association of American Medical Colleges, the Educational Council for Foreign Medical Graduates, the American Board of Medical Specialties, and the National Resident Matching Program. Data on board-certified physicians were obtained from the Division of Survey and Data Resources of the American Medical Association and are not published elsewhere.

Results. After a steep rise in the 1970s, the annual number of physicians receiving licenses increased at a slower rate. The numbers of new board diplomas in medicine and primary care continued to grow. In other non-surgical clinical specialties there was less growth, and in certain fields of surgery the numbers declined. The board-certified percentage of all practitioners increased slightly (74 to 79 percent). About 14 to 16 percent of all active physicians are still in their residency and fellowship years. The percentage of all practitioners under the age of 35 who are women has increased from 8.4 percent in 1967 to 25.2 percent in 1986. The enrollment of some residency programs is currently more than 50 percent women.

Conclusions. The work force of physicians did not grow as rapidly in the 1980s as in the 1970s. This nonlinearity of growth and massive changes in the epidemiology and treatment of disease render predictions about the need for or the numbers of physicians a decade hence unreliable. (N Engl J Med 1991; 324:536-43.)

ON two previous occasions we presented the number and population ratios of board-certified physicians and surgeons in practice in the United States, comparing them over time with the total number of physicians in active practice beyond residency according to their declared fields of specialization. These data included information on residents in training and the age and sex of practitioners. Our purpose here is to update this information for the period from 1980 to 1986, adding a more detailed treatment of the number of women in the various specialties and age groups of medicine. Following on our reports of 1975[1] and 1981,[2] the information now covers a total of 15 years (1971 through 1986).

The standard method used throughout the study was based on collaboration with, or use of the resources provided by, five national agencies: the Association of American Medical Colleges (AAMC), the Educational Council for Foreign Medical Graduates (ECFMG), the American Board of Medical Specialties (ABMS), the National Resident Matching Program (NRMP), and the Division of Survey and Data Resources of the American Medical Association (AMA Department of Data Release Services). The "Masterfile" data from the AMA Department of Data Release Services (the AMA Masterfile unit) on board-certified physicians and surgeons in practice, classified according to their board certifications and corrected for certification by more than one board, are not published elsewhere. The data on physicians in practice were abstracted from *Physician Characteristics and Distribution,* published annually by the AMA. Osteopaths were not included in our study. Information on recent trends in women entering the work force of physicians included data on first-year residencies through 1989.

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METHODS

Data on the population of the United States (excluding the territories) were based on information from the Bureau of the Census, with standard interpolations. The numbers of U.S. medical graduates and holders of first state licenses were based on data published by the AAMC; data on graduates of foreign medical schools were obtained from the ECFMG. The annual number of diplomas (certificates) awarded by the American boards was obtained from the standard publications of the ABMS.[3-6]

The number of physicians in practice in the United States who held diplomas from the 23 specialty boards was taken from tabula-