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Prenatal Screening for Hemoglobinopathies. I. A Prospective Regional Trial

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Summary

Prenatal hemoglobinopathy screening was chosen as a model system for the study of patient receptivity to unsolicited genetic information. Providers of prenatal care in Rochester, NY, were offered free testing of all their prenatal patients and genetic counseling of women found positive. The 18,907 prenatal samples tested in a 5-year period represented 35.1% of the pregnancies in the Rochester metropolitan region. A hemoglobinopathy was found in 810 pregnancies (4.3%). Of the 21 different types of hemoglobinopathies detected, the most common were sickle cell trait (59%), hemoglobin C trait (19%), beta-thalassemia trait (11%), and hemoglobin E trait (5%). At the time of phlebotomy, 75% of the pregnancies were of less than 18 wk duration. Sixty-six percent of the pregnancies occurred in patients unaware of their diagnosis, and 80% occurred in patients unaware that they might be at risk for a child with a serious blood disorder. Of the 810 positive pregnancies, 551 (68%) occurred in patients who came for counseling. Of 453 women counseled during their first screened pregnancy, 390 (86%) said they wanted their partners tested and 254 (55%) had their partner tested. In the 77 pregnancies thus found to be at risk, the couple was too late for prenatal diagnosis in 12 cases, and the condition for which the fetus was at risk was too mild in 12 cases. Prenatal diagnosis was offered in the remaining 53 pregnancies and was accepted by 25 couples (47%). These results indicate that unselected patients in the primary care setting in this region, even though pregnant, are receptive to and utilize genetic information.

Introduction

Recombinant DNA technology has vastly enriched the diagnostic armamentarium of medical genetics. Yet there is little information as to whether the average citizen wants, can comprehend, or will use genetic information (Rowley 1984; Holtzman 1989). In most studies evaluating genetic counseling, individuals of a higher socioeconomic status have been overrepresented. Studies of the response to genetic screening for single-gene disorders have involved screening of only high-risk groups, e.g., blacks in the case of sickle cell trait (Whitten et al. 1981) and Ashkenazim in the case of Tay-Sachs trait (Childs et al. 1976).

Analysis of receptivity to genetic information is complex in the setting of retrospective counseling, i.e., the counseling of individuals following the diagnosis of an affected relative. In order to interpret receptivity to genetic information as a function of counselee characteristics, it is advantageous to study prospective counseling, i.e., the counseling of individuals without known affected relatives, such as those identified in carrier screening programs. The latter population is more homogeneous prior to counseling, because they have not been diversified by different experiences with the disease.

Hemoglobinopathies have advantages for pilot studies of genetic screening (Rowley et al. 1979). The carrier state is common. Carrier testing is widely available. Treatment of the disease states is unsatisfactory. Most important, prenatal diagnosis is available, originally by placental blood sampling (Kan et al. 1972; Chang et al. 1975) and more recently by DNA analysis of amniotic fluid cells, a safer and more accurate method (Kan et al. 1976; Kazazian et al. 1980; Chang

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and Kan 1981; Boehm et al. 1983; Orkin et al. 1983). In fact, the prenatal diagnosis of hemoglobinopathies was the first diagnostic application of recombinant-DNA technology.

If couples are detected as at risk only as a result of having an affected child, the incidence of the disease can be little influenced (Rowley et al. 1985). A more efficient screening approach is to test for carriers. If the development of safe, accurate prenatal diagnosis of hemoglobinopathies is to result in the avoidance of the unwanted birth of the first affected child, four intermediate steps are necessary. First, a prospective parent must be identified as a carrier through screening. Second, the carrier's partner must be tested and found positive. Third, the couple must learn of the risks and their options. Fourth, the couple must voluntarily choose prenatal diagnosis.

Pregnancy has both advantages and disadvantages as a time for carrier screening. Regarding advantages, first, the relevant mate is generally identifiable. Second, identification occurs close to the time for decision about prenatal diagnosis. Third, comprehensive screening of the reproductively active portion of the population is facilitated. Regarding disadvantages, first, pregnancy is too late for those who would have preferred to deal with their risk by avoiding conception. Second, for some women, the first prenatal visit is too late for the consideration of prenatal diagnosis. Third, our earlier studies (Lipkin et al. 1986) suggested that pregnant women may be more anxious than others identified and, as a result, not refer their partners for testing.

The proper goal of a hemoglobinopathy screening program is not the reduction of the incidence of the disease in question; that would involve directive counseling. Instead, the proper goal is to allow the couple at risk to make decisions based on all the relevant information and on their own reproductive goals.

Therefore, in order to address the general question, What is the receptivity of the general public to the diagnostic information which can be provided through recombinant DNA methods? we have asked a more specific question, Should pregnant women be routinely screened so that couples at risk can be identified and offered prenatal diagnosis? To answer this, we enlisted providers of prenatal care in Rochester, NY, screened all bloods drawn at the first prenatal visit for hemoglobinopathies, and offered genetic counseling to women found to be positive. We have asked four questions. First, does the positive woman make a special visit to receive an explanation? Second, after learning the alternatives, does she want her partner tested? Third, does the partner actually come for testing? Fourth, do couples found to be at risk in this way choose prenatal diagnosis? The present report presents the design of the study and the overall results. A second paper in this issue presents the findings regarding learning in response to genetic counseling. A third paper in this issue analyzes counseling response in terms of the Health Belief Model.

Methods

We approached those providers of prenatal care having the larger numbers of patients in the Rochester metropolitan region and asked them to help us in a study of their patients' receptivity to genetic information. We offered to collect from their offices samples on all their prenatal patients, to test them, and to provide genetic counseling to all women found positive. We also offered to test the partners of all positive women and to provide counseling for all couples found to be at risk for a symptomatic child. We agreed to provide these services at no cost to the patient or to the provider; we promised to pay the cost of any DNA analyses performed but not the cost of the amniocentesis. We requested a tube of anticoagulated blood when blood was drawn for other purposes, usually at the first prenatal visit. The provider had the option of asking the patient for consent to be screened. However, only one of the 19 centers chose to do this and then only for the first several years; the other providers felt that they had their patients' implicit consent for relevant diagnostic blood tests.

Table 1 lists the providers of prenatal care in the Rochester area who participated in the study; all the neighborhood health centers and all but one of the hospitals with prenatal clinics participated. Participating providers included neighborhood health centers (e.g., Rochester Health Network), a large health maintenance organization (Genesee Valley Group Health), hospital prenatal clinics, a family medicine program, and nine private practices. Obstetricians, family practitioners, and family medicine physicians were all represented, as were both fee-for-service and prepaid plans.

Figure 1 presents a flow diagram of the project. On each sample was performed a red-cell mean-corpuscular-volume (MCV) determination by a Coulter counter model S (Coulter Electronics, Hialeah, FL) and alkaline hemoglobin electrophoresis (Schneider and Schmidt 1975). If the MCV was 80 fl or less,

Table 1

Providers of Prenatal Care in Study

Provider	Specialty[a]	Method of Payment[b]	% of Patients Who Were Black	Total No. of Samples[c]	Total No. of Positives
Brown Square Health Center	GP	P	33	634	28
Genesee Health Service	OB	P	50	2,086	113
Jordan Health Center	OB	P	65	1,739	159
Northeast Health Center	OB	P	30	1,134	48
St. Mary's Hospital	OB	P	50	1,488	59
Woodward Health Center	GP	P	75	661	41
Genesee Valley Group Health	OB	P	10	2,441	52
Rochester General Hospital Clinic	OB	F	65	876	43
Strong Memorial Hospital Clinic	OB	F	20	2,109	166
Family Medicine Program	FP	F	10	1,143	41
Private practice	OB	F	5–80	4,596	60
Total				18,907	810

[a] GP = general practitioner; FP = family practitioner; OB = obstetrician.
[b] P = prepaid plan; F = fee for service.
[c] Total number of prenatal blood samples provided for hemoglobinopathy detection.

hemoglobin A_2 was quantitated by DEAE-cellulose chromatography (Pearson et al. 1973; Huisman et al. 1979). A value of 3.5% or greater was considered diagnostic of beta-thalassemia trait. Hemoglobins of abnormal mobility were identified by solubility tests, acid-citrate agar gel electrophoresis, or globin chain electrophoresis (Schneider and Schmidt 1975). If a hemoglobinopathy was detected, the physician was contacted for permission to contact the patient. The patient was telephoned if possible or, if not, sent a certified return-receipt letter, as well as a letter by regular mail, which stated the patient's diagnosis, provided reassurance that trait would not affect her health, and alerted her to the risk of a serious inherited red-blood-cell disorder in her fetus should her partner also be a carrier. The patient was invited to come for an explanation of the significance of this finding for her fetus.

The counseling provided to those who responded is described in detail in the following paper in this issue. The principal message of the counseling session was that, if the patient wished to know whether her baby was truly at risk for the disease in question, the baby's father would also have to be tested. His blood could be drawn either at the medical center or by the patient's or her partner's physician for free testing in our laboratory. It was emphasized that, if prenatal diagnosis were to be considered, it could not be performed after 20 wk of pregnancy. Following counseling, we asked the patient whether she intended to refer the baby's father for testing. If the patient expressed an intent to ask her partner to be tested but did not contact us in the next 7 d, we contacted her to confirm her intent.

Results and Discussion

The overall results of the project are presented in figure 2.

Diagnoses Made

The 18,907 prenatal samples received represented approximately 35% of the pregnancies in the Rochester metropolitan area (Monroe County) occurring during the 5-year period of the study. The diagnoses made on these samples are shown in table 2. Of the 22 diagnoses made, the most common were sickle cell trait (59%), hemoglobin C trait (19%), beta-thalassemia trait (11%), and hemoglobin E trait (5%). It is noteworthy that many of those identified were not of the racial or ethnic group commonly associated with that condition. Specifically, 7% of the sickle trait patients were not black and 22% of the beta-thalassemia trait patients were not Mediterranean, black, or Asian. This finding is an argument in favor of the prenatal screening of all women rather than only of high-risk groups. Four hemoglobinopathies that are usually symptomatic—namely, sickle cell anemia, hemoglobin SC disease, sickle beta-thalassemia, and hemoglobin H disease—were each diagnosed three times. Of these 12 patients, 11 were unaware of their risk of having a child with a hemoglobinopathy. Of

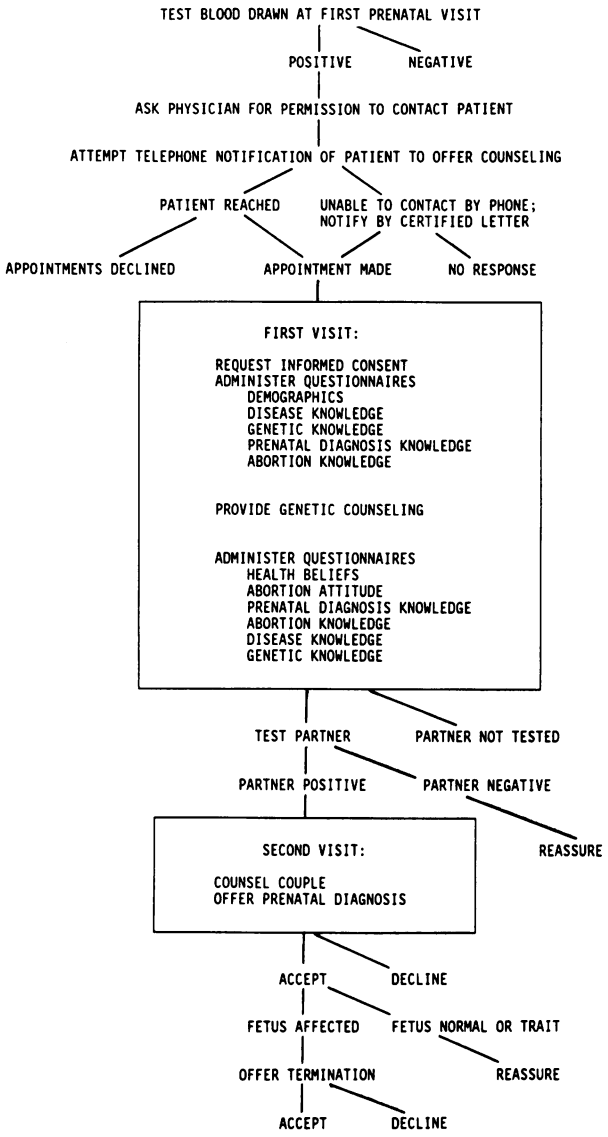

Figure 1 Flow diagram of project to determine receptivity to prenatal hemoglobinopathy screening.

the patients with hemoglobin SC disease and sickle beta-thalassemia, two of each type were asymptomatic and unaware of their diagnosis. Of the hemoglobin H disease patients, one was asymptomatic and all three were unaware of their diagnosis.

Patients identified as having hemoglobinopathies had a mean ± SD age of 23.2 ± 5.1 years. Seventy-five percent had a gestational age of less than 18 wk. Only 34% had previously been aware that they had a hemoglobinopathy.

Counselee Characteristics

Of the 810 positive pregnancies, 551 (68%) occurred in women who, as a result of both identification of a hemoglobinopathy and the offer of information, came for an explanation of their diagnosis. Of these 551 pregnancies, only 463 occurred in women to whom we provided genetic counseling during that pregnancy; 85 pregnancies occurred in women we had counseled during a previous pregnancy, and three pregnancies occurred in women we had counseled as a result of the birth of a child identified, by state-mandated sickle cell screening of newborns, as having a hemoglobinopathy.

Characteristics of patients coming for counseling are shown in table 3. Sixty-six percent did not know that they had a hemoglobinopathy, and 80% did not know that they might be at risk for a child with an inherited blood disease. Therefore 14% recalled having been told they had a hemoglobinopathy but did not understand its reproductive significance. Fifty percent were not living with the father of the fetus, and 62% were single. Sixty-seven percent had not planned the pregnancy, although 97% now wanted the pregnancy. Seventy-six percent did not want another pregnancy or were undecided. A noteworthy finding was that 75% were no more than 18 wk pregnant when tested, early enough for them to consider prenatal diagnosis if they were eligible for it.

Patients Not Counseled

Two hundred fifty-nine pregnancies occurred in women with hemoglobinopathies who, for various reasons, were not counseled by us. In 104 pregnancies, the women did not respond to a certified letter, despite the fact that the letter had been received (as demonstrated by a returned receipt). In 61 pregnancies, the women failed to keep two appointments. In 29 pregnancies, the women failed to respond to the offer of counseling, in most cases because they felt they already had adequate information. Twenty-four pregnancies represented repeat diagnoses in women who had failed to respond to the offer of counseling in previous pregnancies. Twenty-three pregnancies ended between identification and the scheduled appointments—namely, 11 in spontaneous abortion, four in therapeutic abortion, and eight in delivery. Eleven pregnancies were identified at 18 wk or later during the first few months of the study, before we had decided to inform women of their carrier status even if they were too late for prenatal diagnosis. Three patients were informed by their providers. Two patients moved and were unlo-

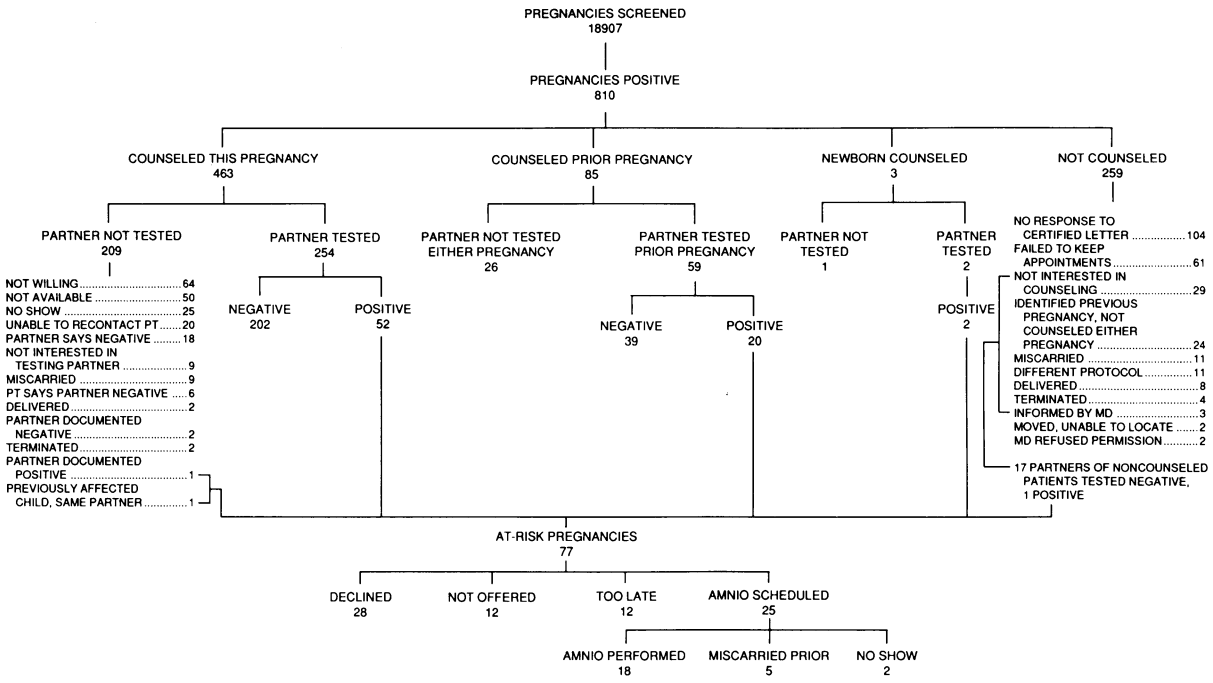

Figure 2 Outcomes of hemoglobinopathy screening

Table 2

Diagnoses Made

Diagnosis	No. of Pregnancies	Ethnic or Racial Group
Sickle cell trait	474	black (439), Hispanic (26), Italian (6), Indian (2), and other white (1)
Hb C trait	150	black (145), Hispanic (4), and other white (1)
Beta-thalassemia trait	92	black (35), Italian (32), other white (20), Vietnamese (3), Laotian (1), and Indian (1)
Hb E trait	37	Laotian (21), Cambodian (14), and Vietnamese (2)
Hb D or G trait	17	black (11) and white (6)
Homozygous Hb E	6	Laotian (3) and Cambodian (3)
Delta/beta-thalassemia trait	6	Italian (3) and black (3)
Fast Hb	3	white (2) and black (1)
Sickle/beta-thalassemia	3	black
Hb SC disease	3	black
Hb E trait/alpha-thalassemia trait	3	Laotian (2) and Cambodian (1)
Hb H disease	3	Laotian
Alpha-thalassemia trait	3	Laotian
Hb O trait	2	black
Sickle cell anemia	2	black
Hb C/HPFH[a]	1	black
Hb I or J trait	1	black
Hb S/HPFH[a]	1	black
Hemoglobin Richmond trait	1	black
Hb C trait/Hb G trait	1	black
Hb C/beta-thalassemia	1	black
Total	810	

[a] HPFH = hereditary persistence of fetal hemoglobin trait.

Table 3

Characteristics of Patients Coming for Counseling (*N* = 463)

Characteristic or Question	Value
Age	22.9 (±5.0[SD]) years
Education (years)	11.3 (±2.5 [SD]) years (range 0–18)
Employment	No 61.7%, yes 38.3%
Religion	Protestant 54%, none 24%, Catholic 14%, other 8%
Marital Status	Single 62%; married 31%; divorced, separated, or widowed 7%
Gestational age when tested	75% before 18 wk
Have you ever been told you have a hemoglobinopathy?	No 66%, yes 34%
Have you ever been told that you may be at risk for a child with an inherited red blood cell disease?	No 80%, yes 20%
Are you living with the father of the baby?	Yes 50%, no 50%
Was this pregnancy planned?	No 67%, yes 33%
Do you want this pregnancy?	Yes 97%, no 3%
Do you want additional pregnancies?	Undecided 39%, no 37%, yes 24%

catable. Finally, in two cases the physician refused us permission to contact the patient.

Partner Testing

At the end of the counseling session the patients were asked whether they intended to ask the baby's father to be tested; 86% said they did. In 254 cases (55%), the partner was tested. In the other 209 pregnancies there were various reasons why the partner was not tested. Sixty-four partners were not willing. Fifty patients could not locate their partners. Twenty-five partners made appointments but did not keep them. In twenty cases efforts to recontact the patient failed. In eighteen cases, although the partner claimed to be negative, documentation was not available. In nine cases the patient was not interested in having her partner tested. Nine patients miscarried before the partner could be tested. Six patients claimed that their partners were negative, but this could not be documented. In two other such cases it was documented that the partner was negative. Two pregnancies were spontaneously delivered soon after the patient's carrier diagnosis. Two women had terminations unrelated to the patient's carrier diagnosis. The partner was known to be positive in one case through documentation of a previously positive test and in another case by having had an affected child.

Eighty-five pregnancies were repeat pregnancies in women counseled by us in an earlier pregnancy. In 59 (69%) of these, the partner had been tested in the index pregnancy; in the remaining 26 (31%), the partner was not tested in either pregnancy.

Finally, in three cases the mother had been counseled by us as a result of having a newborn with a hemoglobinopathy. In two of these cases, the partner was tested and was positive.

Prenatal Diagnosis

Of the 77 at-risk pregnancies there were 12 cases in which the couple was too late to be offered prenatal diagnosis. In another 12 cases, the condition for which the fetus was at risk (homozygosity for either hemoglobin C or hemoglobin E) was too mild to justify the offer. In the remaining 53 pregnancies, the couples were offered prenatal diagnosis; the offer was accepted in 25 cases and declined in 28 cases, giving an acceptance rate of 47%. Of the 25 aminocenteses scheduled, 18 were performed and seven were not; in five of these seven cases the patient had a spontaneous abortion in the interval, in the other two cases the patient did not keep the amniocentesis appointment.

The amniocenteses performed are summarized in table 4. Of eight fetuses at risk for sickle cell anemia, one was found to be affected and seven were found to have trait or to be normal. Of six fetuses at risk for hemoglobin SC disease, two were found to be affected and four were found to have the trait or to be normal. Two Southeast Asian couples were at risk for a fetus with hemoglobin H disease with hemoglobin E trait. One fetus was found to have hemoglobin E trait and 2-alpha gene deletion thalassemia, and the other was found to be normal. Finally, two fetuses were at risk for hemoglobin H disease; one was found to have 2-alpha-gene deletion thalassemia, and the other was found to have a 50% risk of hemoglobin H disease, rather than the 25% risk which could be estimated

Table 4

Amniocenteses Performed

Condition for Which Fetus Was at Risk (*N*)	Diagnosis Made (*N*)	Pregnancy Outcome
Sickle cell anemia (8)	AS (5)	Continued
	AA (2)	Continued
	SS (1)	Continued
Hemoglobin SC disease (6)	AA (2)	Continued
	AS (2)	Continued
	SC (2)	Continued
Hb H disease/hemoglobin E trait (2)	Normal (1)	Continued
	AE/2-alpha-gene deletion thalassemia (1)	Continued
Hb H disease (2)	2-alpha-gene deletion thalassemia (1)	Continued
	Hb H disease (1)	Terminated

without prenatal diagnosis. Only in the last case was the pregnancy terminated. The patient herself had hemoglobin H disease, felt significantly disabled, and did not want to risk having a child with the same condition.

Other Benefits of Carrier Detection

Selective termination is not the only benefit of prenatal diagnosis of genetic disease. Learning the diagnosis during pregnancy can help a couple prepare for parenting a child with special needs. Furthermore, carrier identification may be useful for future reproductive planning, even though some may not apply the information to a pregnancy in progress. No doubt it would be preferable for a woman to know her carrier status prior to her first pregnancy. However, many women do not consult a physician until they are already pregnant. Thus, the main argument for pregnancy as a time for screening is a pragmatic one.

Informed Consent

As previously mentioned, the providers decided whether to ask their patients for informed consent prior to screening them. Most providers decided not to. Nevertheless most patients who were found to be carriers felt it had been appropriate for their doctor to test them. In fact, at counseling many with previous children asked, "Why didn't my doctor test for this condition in my first pregnancy?" An additional defense of their physician's decision to screen without explicit consent is the fact that, whereas detection of the common abnormal hemoglobins is accurate, screening methods for beta-thalassemia trait fail to detect all trait individuals. Further, one might argue that informed consent (and, especially, informed refusal) requires as much time as the counseling of a known positive. Many primary care providers felt that such time was better spent with patients detected to be carriers.

Since most of our patients were not asked for consent, the screened population represented an essentially unselected one. An unselected population was advantageous for research purposes because, if only patients interested in receiving information had been tested, any conclusion about patient receptivity might be falsely optimistic.

Patient Receptivity

The patient response observed in this program seems to us an encouraging one. Many patients who came for counseling had never been to the medical center before, and many of these were adolescents or belonged to minority groups.

The three types of decisions women made are summarized in table 5. When told they had an abnormal blood test, women came for an explanation in 68%

Table 5

Summary of Decisions Made by Patients

Decision	Accepted/Offered
If tested positive, came for counseling	551/810 (68%)
If counseled, had partner tested	315/551 (57%)
If offered prenatal diagnosis, accepted	25/53 (47%)

of cases. When counseled, women had their partners tested in 57% of cases. Finally, when offered prenatal diagnosis, couples accepted the offer in 47% of cases. These results indicate that, despite pregnancy, patients are highly receptive to genetic information. This is especially remarkable since the decision as to whether to ask patients for consent to be carrier tested was in the hands of the prenatal care providers and since many providers did not ask patients for consent. Thus our data is of special interest because it describes significant receptivity to genetic screening in an essentially unselected population.

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References

Boehm CD, Antonarakis SE, Phillips JA, Stretten G, Kazazian HH Jr (1983) Prenatal diagnosis using DNA polymorphisms: report on 95 pregnancies at risk for sickle-cell disease or β-thalassemia. N Engl J Med 308:1054–1058

Chang H, Modell CB, Alter BP, Dickinson MJ, Frigoletto FD, Huehns ER, Nathan DG (1975) Expression of the β-thalassemia gene in the first trimester fetus. Proc Natl Acad Sci USA 72:3633–3637

Chang JC, Kan YW (1981) Antenatal diagnosis of sickle cell anemia by direct analysis of the sickle mutation. Lancet 2:1127–1129

Childs B, Gordis L, Kaback MM, Kazazian HH Jr (1976) Tay-Sachs screening: motives for participating and knowledge of genetics and probability. Am J Hum Genet 28:537–549

Holtzman NA (1989) Proceed with caution: predicting genetic risks in the recombinant DNA era. Johns Hopkins University Press, Baltimore

Huisman TH, Schroeder WA, Brodie AN, Mayson SM, Jakway J (1979) Microchromatography of hemoglobin. III. A simplified procedure for the determination of hemoglobin A_2. J Lab Clin Med 86:700–702

Kan YW, Dozy AM, Alter BP, Frigoletto FD, Nathan DG (1972) Detection of the sickle gene in the human fetus: potential for intrauterine diagnosis. N Engl J Med 287: 1–5

Kan YW, Golbus MS, Dozy AM (1976) Prenatal diagnosis of β-thalassemia. N Engl J Med 295:1165–1167

Kazazian HH Jr, Phillips JA, Boehm CD, Vik TA, Mahoney MJ, Ritchey AK (1980) Prenatal diagnosis of β-thalassemia by amniocentesis: linkage analysis using multiple polymorphic restriction endonuclease sites. Blood 56: 926–930

Lipkin M Jr, Fisher L, Rowley PT, Loader S, Iker HP (1986) Genetic counseling in a primary care setting: a study of the effectiveness of screening and counseling for beta-thalassemia trait. Ann Intern Med 105:115–123

Orkin SH, Markham AF, Kazazian HH Jr (1983) Direct detection of the common Mediterranean β-thalassemia gene with synthetic DNA probes. J Clin Invest 71:775–779

Pearson HA, O'Brien RT, McIntosh S (1973) Screening for thalassemia trait by electronic measurement of mean corpuscular volume. N Engl J Med 288:351–353

Rowley PT (1984) Genetic screening: marvel or menace? Science 225:138–144

Rowley PT, Fisher L, Lipkin M Jr (1979) Screening and genetic counseling for beta-thalassemia trait in a population unselected for interest: effects on knowledge and mood. Am J Hum Genet 31:718–730

Rowley PT, Loader S, Walden ME (1985) Toward providing parents the option of avoiding the birth of the first child with Cooley's anemia: response to hemoglobinopathy screening and counseling during pregnancy. Ann NY Acad Sci 445:408–416

Schneider RG, Schmidt RM (1975) Electrophoretic screening for abnormal hemoglobins. In: Schmidt RM (ed) Abnormal haemoglobins and thalassemia: diagnostic aspects. Academic Press, New York, pp 39–63

Whitten CF, Thomas JF, Nishura EN (1981) Sickle cell trait counseling-evaluation of counselees and counselors. Am J Hum Genet 33:802–816