Prediction of Proliferative Diabetic Retinopathy With Hemoglobin Level

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Objective: To investigate the role of hemoglobin (HGB) level in predicting proliferative diabetic retinopathy (PDR).

Methods: We assessed 426 individuals without PDR at baseline (213 men; 213 women) from the Pittsburgh Epidemiology of Diabetes Complications Study, an 18-year prospective cohort study of childhood-onset type 1 diabetes. Presence of PDR was determined by stereo fundus photography. Cox proportional hazards modeling with stepwise regression was used to determine the independent association of HGB level with PDR. Analyses were sex specific.

Results: There were 206 events. Although the incidence of PDR did not vary by sex (48% in both men and women), in men, HGB exhibited a positive linear relationship with 18-year incidence of PDR (hazard ratio, 1.33; 95% confidence interval, 1.10-1.60; $P=.003$), while in women, HGB level exhibited a quadratic relationship with PDR ($P<.001$). After multivariable adjustment for univariately significant covariates, HGB level remained significantly predictive of PDR in both men ($P=.004$) and women ($P=.04$).

Conclusion: Higher HGB level predicts the incidence of PDR in type 1 diabetes mellitus, though the association varies by sex, being linear and positive in men and quadratic in women.

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PROLIFERATIVE DIABETIC RETINOPATHY (PDR) is a leading cause of blindness in the United States and has been linked to retinal hypoxia and characterized by small blood vessel proliferation, retinal microaneurysms, and vitreous hemorrhaging.[1] Lower hematocrit (HCT) levels have been associated with the progression of diabetic kidney disease[2,3] and with the incidence of PDR,[4] suggesting that anemia may increase complication risk. In contrast, however, stabilization of retinopathy has also been reported in those with end-stage renal disease,[5,6] a condition associated with anemia. The association between hemoglobin (HGB) and retinopathy is likely, therefore, to be complex, particularly given the recent observation of unusually high HGB levels, reaching 18.8 g/dL, in study participants with nephropathic type 1 diabetes mellitus.[7]

Hemorheological factors are altered in diabetes, and there is some evidence that these disturbances may also be associated with diabetic retinopathy.[8] Increased HCT[8] and blood viscosity[9,10] have been observed cross-sectionally in those with diabetic retinopathy. Elevated erythropoietin has also been observed in the vitreous fluid of those with diabetic retinopathy and found to be an angiogenic promoter of PDR.[11]

Given these observations, we decided to investigate the association of HGB with the incidence of PDR in type 1 diabetes mellitus.

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METHODS

The Pittsburgh Epidemiology of Diabetes Complications (EDC) study is an 18-year prospective study based on a well-defined cohort with childhood-onset (age <17 years) type 1 diabetes living within 100 miles of the University of Pittsburgh at study baseline. The 658 participants (325 female, 333 male; 67% of all those eligible) were diagnosed between January 1, 1950, and May 30, 1980, at Children's Hospital of Pittsburgh, and first seen in EDC between 1986 and 1988, when the mean age and diabetes duration were 28 and 19 years, respectively. They were then seen biennially for 10 years, after which they were followed up by survey with examinations limited to certain subgroups. Between 2004 and 2007, however, an 18-year follow-up was conducted for all participants. The design and methods of the study have been previously described.[12]

Before attending each cycle of examinations, information was collected by questionnaire concerning demographic characteris-

tics, medical history, and health care behaviors, as previously described.[13] At each cycle, both a standardized medical history and clinical examination were performed by a trained internist to document complications of diabetes.

Fasting blood samples were assayed for lipids, lipoproteins, glycosylated hemoglobin (HbA_1), and HGB. Hemoglobin was measured using the Coulter Counter Model S-Plus IV (Coulter Electronics, Hialeah, Florida) automated blood cell counter. High-density lipoprotein cholesterol level was determined by a heparin and manganese procedure, a modification of the Lipid Research Clinics method.[14] Cholesterol was measured enzymatically. Glomerular filtration rate was estimated using the 4-variable Modification of Diet in Renal Disease formula.[15]

Stable HbA_1 was originally measured in saline-incubated samples by microcolumn cation exchange chromatography (Isolab, Akron, Ohio). On October 26, 1987, the method was changed to high-performance liquid chromatography (Diamat, Bio-Rad Laboratories, Hercules, California). The 2 methods were highly correlated ($r=0.95$; Diamat HbA_1, $-0.18+1.00$ Isolab HbA_1). After the 10-year follow-up, glycosylated hemoglobin A_{1c} (HbA_{1c}) was measured with the DCA 2000 analyzer (Bayer, Tarrytown, New York). The Diamat and DCA were also highly correlated ($r=0.95$; DCA $HbA_{1c}=-0.18+1.00$ Diamat HbA_1). The HbA_1 and HbA_{1c} values were converted to Diabetes Control and Complications Trial (DCCT)–aligned HbA_{1c} values using regression formulas derived from duplicate analyses (DCCT $HbA_{1c}=[0.83 \times$ EDC $HbA_1] + 0.14$; DCCT $HbA_{1c}=[$EDC $HbA_{1c} - 1.13]/0.81$).

Blood pressure was measured by a random-zero sphygmomanometer according to a standardized protocol[16] after a 5-minute rest period. Blood pressure levels were analyzed using the mean of the second and third readings.

Retinopathy was assessed in 3 standardized visual fields. Stereoscopic fundus photographs were taken in fields 1, 2, and 4 with a Zeiss camera (Carl Zeiss, Oberkochen, Germany), and were read by the Fundus Photography Reading Center, University of Wisconsin, Madison. Gradings were classified by the modified Arlie House System, with PDR defined as a grade of 60 or higher in at least 1 eye. Individuals without gradable fundus photographs were graded according to their medical history data, with confirmation by their ophthalmologist if required. Those with panretinal photocoagulation scars and a grade of less than 60 were recorded as having PDR if the medical history indicated that the laser therapy was for PDR. Two-step progression, an increase of at least 2 steps on the modified Arlie House System,[17] and clinically significant macula edema were also identified.[18]

Nephropathy status was determined based on consistent results from at least 2 of 3 timed urine collections (24-hour, overnight, and timed samples during examination) and urine albumin excretion rate. The adequacy of urine collection was determined by whether the total amount of creatinine excreted was appropriate for an individual's age, based on body size and sex. Urinary albumin was determined immunonephelometrically.[19] All procedures were approved by the institutional review board of the University of Pittsburgh.

Continuous data were compared between groups by the *t* test, with serum creatinine, triglycerides, high-density lipoprotein cholesterol, and non–high-density lipoprotein cholesterol being log transformed prior to testing. Categorical data were compared between groups by χ^2 tests. Cox proportional hazards modeling with forward selection was used to determine the association of baseline HGB with the 18-year incidence of PDR. The functional form of HGB was assessed by plotting a restricted cubic spline transformation for HGB against the log hazard ratio separately for men and women using Harrell's[20] SAS macro, %PSPLINET. A Wald test was used to test the assumption of a linear relationship between HGB and the log hazard ratio. Owing to the sex differences in HGB distribution, analyses were conducted sex specifically. Censoring occurred at the time of event or at last follow-up.

RESULTS

Hemoglobin data was available for 652 study participants; the HGB levels ranged from 9.2 to 20.0 g/dL (to convert to grams per liter, multiply by 10), with a median (interquartile range) of 16.2 (15.4-16.9) g/dL in men and 14.1 (13.5-15.0) g/dL in women. Two hundred two participants had PDR at baseline and were excluded from further analyses. Of the remaining 450, 24 provided no further PDR information and complete follow-up to 18 years was available for 78% of those still alive, follow-up being censored at last observation for the remainder. Baseline characteristics of the 426 study participants free of PDR at baseline are described in **Table 1** and **Table 2** for men and women, respectively.

The cumulative incidence of PDR was 48% after 18 years. The median time to follow-up was 14.6 years. In men, there was no suggestion of any deviation from a linear relationship between HGB and the log hazard ratio (Wald test of linearity $P=.15$); therefore, HGB was entered into Cox models as a continuous linear covariate. In women, however, the Wald test showed a significant deviation from linearity ($P=.02$) and a graph of the restricted cubic spline suggested a quadratic relationship between HGB and the log hazard ratio (**Figure**); therefore, HGB was entered into Cox models as a quadratic term. Univariately, HGB showed a significant linear association with the risk of developing PDR in men ($P=.003$) and a significant nonlinear association in women ($P=.008$, quadratic term).

In multivariable analyses allowing for univariately significant covariates, HGB remained significantly predictive of PDR in men (hazard ratio,1.29; 95% confidence interval, 1.08-1.54; $P=.004$) (**Table 3**). In women, HGB as a quadratic term also remained significantly predictive of PDR ($P=.04$). Other significant multivariable predictors included HbA_{1c} and diastolic blood pressure in both sexes, albumin excretion rate in men, and systolic blood pressure and fibrinogen in women. Finally, the association of HGB with other components (markers) of retinopathy was also explored. A significant prediction of both macular edema ($P<.001$ and $P=.03$ for men and women, respectively) and 2-step progression ($P=.03$ and $P<.001$ for men and women, respectively) was observed.

COMMENT

The cumulative incidence of PDR in this population was 48% after 18 years. The EDC incidence rates are similar to those observed in the DCCT/Epidemiology of Diabetes Interventions and Complications conventional treatment group of similar diabetes duration (49.7% at 30 years

Table 1. Baseline Characteristics of Men by Subsequent Proliferative Diabetic Retinopathy Status

Characteristics	Mean (SD) PDR (n=103)	Mean (SD) No PDR (n=110)	*P* Value
Age, y	25.1 (6.4)	24.2 (7.8)	.35
Diabetes duration, y	16.8 (5.7)	16.1 (6.7)	.43
Hemoglobin, g/dL	16.5 (1.2)	16.1 (1.2)	.009
White blood cells, No. $\times 10^3/mm^2$	6.2 (1.6)	6.2 (1.8)	.92
Body mass index[a]	23.7 (2.9)	22.9 (3.4)	.05
Overt nephropathy, No. (%)	36 (39.1)	14 (12.8)	<.001
Median (IQR) estimated GFR (MDRD), mL/min/1.73 m^2[b]	107.1 (92.2-146.2)	107.1 (88.7-139.1)	.70
Median (IQR) albumin excretion rate, µg/min[b]	14.8 (9.0-65.2)	8.6 (5.7-21.6)	<.001
Total cholesterol, mg/dL[b]	186.1 (37.7)	174.4 (33.0)	.02
Non-HDL cholesterol, mg/dL[b]	136.9 (37.0)	124.1 (33.6)	.009
HDL cholesterol, mg/dL	49.2 (9.0)	50.2 (10.0)	.42
Fibrinogen, mg/dL	262.8 (80.2)	266.2 (81.6)	.76
Systolic blood pressure, mm Hg	114.8 (13.6)	111.3 (12.5)	.05
Diastolic blood pressure, mm Hg	74.4 (9.5)	71.0 (9.9)	.01
HbA_{1c}, %	9.0 (1.5)	8.5 (1.5)	.009
Total insulin, injections/d	2 (1-2)	2 (1-2)	.15
Current smoker, %	22 (21.8)	19 (19.0)	.62

Abbreviations: GFR (MDRD), estimated glomerular filtration rate (Modification of Diet in Renal Disease formula); HbA_{1c}, glycosylated hemoglobin A_{1c}; HDL, high-density lipoprotein; IQR, interquartile range; PDR, proliferative diabetic retinopathy.

SI conversion factors: To convert mean hemoglobin concentration to grams per liter, multiply by 10; cholesterol to millimoles per liter, 0.0259; fibrinogen to micromoles per liter, 0.0294; HbA_{1c} to proportion of total hemoglobin, 0.01.

[a] Calculated as weight in kilograms divided by height in meters squared.

[b] Natural logarithmically transformed before analysis.

Table 2. Baseline Characteristics of Women by Subsequent Proliferative Diabetic Retinopathy Status

Characteristic	Mean (SD) PDR (n=103)	Mean (SD) No PDR (n=110)	*P* Value
Age, y	26.0 (7.1)	25.7 (8.5)	.76
Diabetes duration, y	17.0 (6.8)	17.1 (7.5)	.97
Hemoglobin, g/dL	14.4 (1.2)	14.2 (1.0)	.24
White blood cells, No. $\times 10^3/mm^2$	6.8 (1.9)	6.2 (1.8)	.03
Body mass index[a]	23.3 (3.1)	23.2 (3.5)	.70
Overt nephropathy, No. (%)	33 (35.5)	15 (14.0)	<.001
Median (IQR) estimated GFR (MDRD), mL/min/1.73 m^2[b]	105.2 (86.0-134.3)	104.2 (87.6-129.4)	.92
Median (IQR) albumin excretion rate, µg/min[b]	12.9 (7.7-71.6)	9.0 (5.6-19.4)	.002
Total cholesterol, mg/dL[b]	195.6 (41.1)	178.6 (32.0)	<.001
Non-HDL cholesterol, mg/dL	136.1 (41.3)	117.8 (30.5)	<.001
HDL cholesterol, mg/dL	59.5 (13.7)	60.8 (11.8)	.48
Fibrinogen, mg/dL	310.7 (90.5)	270.6 (60.8)	<.001
Systolic blood pressure, mm Hg	109.9 (13.2)	105.3 (10.1)	.004
Diastolic blood pressure, mm Hg	70.3 (10.7)	67.7 (7.9)	.05
HbA_{1c}, %	9.2 (1.7)	8.4 (1.3)	<.001
Total insulin, injections/d	2 (1-2)	2 (1-2)	.17
Current smoker, No. (%)	18 (18.4)	19 (17.4)	.86

Abbreviations: GFR (MDRD), estimated glomerular filtration rate (Modification of Diet in Renal Disease formula); HbA_{1c}, glycosylated hemoglobin A_{1c}; HDL, high-density lipoprotein; IQR, interquartile range; PDR, proliferative diabetic retinopathy.

SI conversion factors: To convert hemoglobin concentration to grams per liter, multiply by 10; cholesterol to millimoles per liter, 0.0259; fibrinogen to micromoles per liter, 0.0294; HbA_{1c} to proportion of total hemoglobin, 0.01.

[a] Calculated as weight in kilograms divided by height in meters squared.

[b] Natural logarithmically transformed before analysis.

of diabetes duration[21]) and the Wisconsin Epidemiological Study of Diabetic Retinopathy population (37% at approximately 26.6 years of diabetes duration[18]). Although much attention has been given in recent years to anemia and low levels of HGB in diabetes, particularly as it relates to kidney disease,[22-28] this is the first study, to our knowledge, to show high HGB levels to be predictive of the long-term incidence of PDR. We have also shown that the relationship of HGB with PDR varies by sex, with HGB demonstrating a linear relationship in men, but a nonlinear quadratic (P=.008) relationship in women. These results, if confirmed, may have important clinical relevance, as they may both identify new pathogenetic pathways to PDR and influence clinical treatment. There are several potential explanations for these findings.

First, the HGB levels in this population were relatively high, ranging from 9.2 to 20.0 g/dL. We have observed that HGB levels in our population, including the 120 participants with PDR and 228 with overt nephropathy at baseline, were approximately 1 g/dL higher in both men and women compared with the National Health and Nutrition Examination Survey III white population limited to the same age range.[29] Higher HGB levels in type 1 diabetes were also noted by Pichler et al,[30] who observed a higher (0.8 g/dL) difference in HGB levels between children and adolescents with type 1 diabetes compared with controls. Factors related to increased HGB such as testosterone, hypoxia, growth factors, and viscosity may responsible for the PDR associated with increased HGB in this population.

Androgens, in particular testosterone, are a known stimulant of erythropoiesis,[31] and testosterone has been fairly consistently shown to be elevated in type 1 diabetes. Haffner et al[32] found higher testosterone levels and lower sex hormone-binding globulin[33] in male participants with type 1 diabetes and PDR compared with those without PDR. Chaurasia et al[34] also found higher testosterone levels in those with PDR. Factors associated with the anabolic effects of androgens such as erythropoietin stimulation may thus accelerate progression to PDR.

Erythropoietin is a glycoprotein produced by the peritubular fibroblasts of the kidney, and its production is primarily determined by tissue hypoxia in those with

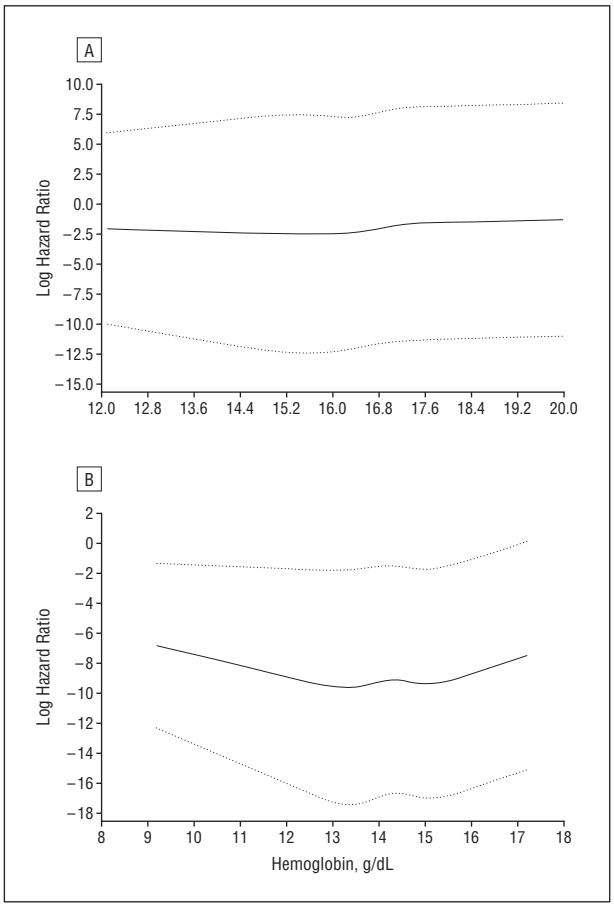

Figure. Restricted cubic spline of hemoglobin and 95% confidence interval by the log hazard ratio of proliferative diabetic retinopathy in men (A) and women (B). To convert hemoglobin concentration to grams per liter, multiply by 10.

Table 3. Multivariable Prediction of the 18-Year Incidence of Proliferative Diabetic Retinopathy in Type 1 Diabetes Mellitus[a]

Variable	HR (95% CI) Men (n=213, 103 events)	HR (95% CI) Women (n=213, 103 events)
HGB, g/dL	1.29 (1.08-1.54)	NA
HGB quadratic, g/dL	NA	1.10 (1.00-1.20)
HbA_{1c}, %	1.30 (1.14-1.49)	1.32 (1.16-1.50)
Albumin excretion rate, μ/min[b]	1.34 (1.20-1.50)	NS
DBP, mm Hg	1.03 (1.00-1.05)	2.28 (1.02-5.07)
SBP, mm Hg	NS	1.03 (1.01-1.05)
Fibrinogen, mg/dL	NS	1.00 (1.00-1.01)

Abbreviations: CI, confidence interval; DBP, diastolic blood pressure; HbA_{1c}, glycosylated hemoglobin A1c; HGB, hemoglobin; HR, hazard ratio; NA, not applicable; NS, not selected; SBP, systolic blood pressure.

SI conversion factors: To convert hemoglobin concentration to grams per liter, multiply by 10; fibrinogen to micromoles per liter, 0.0294; HbA_{1c} to proportion of total hemoglobin, 0.01.

[a] n=426. The following variables were also made available for multivariable analyses in the stepwise selection model: diabetes duration, body mass index, non-HDL cholesterol, white blood cell count, fibrinogen, and hypertension medication.

[b] Natural logarithmically transformed before analysis.

normal renal function.[35] Erythropoietin stimulates angiogenesis[11] and has been found to be associated with the retinopathy of prematurity.[36,37] In diabetes, Watanabe et al[11] found both erythropoietin and vascular endothelial growth factor to be independently associated with PDR cross-sectionally, while blocking of erythropoietin was found to inhibit retinal neovascularization.[38] In preterm infants, Romagnoli et al[36] observed that infants treated with recombinant human erythropoietin to reduce the need for blood transfusions were at an increased risk of retinopathy of prematurity, with no change in the need for transfusion. Brown et al[37] also noted an increased risk of prematurity in preterm infants given recombinant erythropoietin. In diabetic patients receiving hemodialysis, Diskin et al[39] observed that recombinant erythropoietin dose per week and HCT were positively associated with deterioration of retinopathy. Finally, recent trials have also observed an increase in adverse events, ie, a deterioration of kidney disease and more cardiovascular events and mortality, associated with the correction of anemia by erythropoietin therapy in those with kidney disease.[40,41] Because these target treatment HGB levels were relatively low, this seems to suggest that growth factors stimulated by erythropoietin are more likely to be associated with adverse outcomes rather than high HGB per se.

Another potential related mechanism is that the HGB association with PDR reflects increased erythropoietin production stimulated by general hypoxia,[11] which may contribute to the microvascular disease seen in both the retina and the kidneys in type 1 diabetes mellitus. Some recent studies have shown increased levels of anemia in type 1 diabetes to be associated with complications of diabetes, including retinopathy, renal impairment, and macrovascular disease. However, these patients were from tertiary care clinics and were older[23,28] than our participants. In contrast, other literature suggests increased blood viscosity in diabetes,[10,42-44] which may lead to tissue ischemia.[8] Vekasi et al[8] found higher levels of HCT, plasma fibrinogen, and higher plasma and whole blood viscosity in those with diabetic retinopathy compared with nondiabetic controls. Blood viscosity is directly related to HCT.[10] As higher HCT and blood viscosity have also been observed in those with retinal vein occlusion,[45] and the HGB levels associated with PDR in our population were quite high, it is quite possible that our PDR association may be partly due to increased viscosity and associated sludging. Unfortunately, direct viscosity measures are not available.

The sex differences, eg, HGB demonstrating a nonlinear relationship with PDR in women and a linear relationship in men, are of interest. However, this is likely to just reflect the small sample sizes at the lower ends of the distribution in men (n=5) at levels (≤13.4 g/dL) where an increased risk was seen in women (n=46). A U-shaped relationship is thus likely to be true, and many U-shaped relationships exist between risk states and outcomes (eg, weight and mortality), with different pathological pathways operating at each end of the spectrum. Interestingly, a very recent study of type 1 diabetes found a genetic variant promoter of the *EPO* gene to be associated with both PDR and end-stage renal disease.[46] An-

other limitation of our study was that we could not adequately stratify by renal disease status, a complication associated with both HGB levels and diabetic retinopathy, as only 14% (n=56) of those free of PDR at baseline had overt nephropathy. However, we did try to account for kidney disease by controlling for albumin excretion rate, which, while predictive of PDR, did not account for HGB's association with PDR incidence.

In conclusion, these data suggest that high HGB levels may be associated with increased risk of PDR. Furthermore, these PDR findings are complimented by observations that HGB also predicts 2-step progression and macula edema. Increased testosterone in type 1 diabetes, growth factors, viscosity, and/or compensation for generalized ischemia/hypoxia may account for the association of high HGB levels with PDR. Taken as a whole, these results suggest that further evaluation of the potential adverse role of high HGB in those with type 1 diabetes is warranted.

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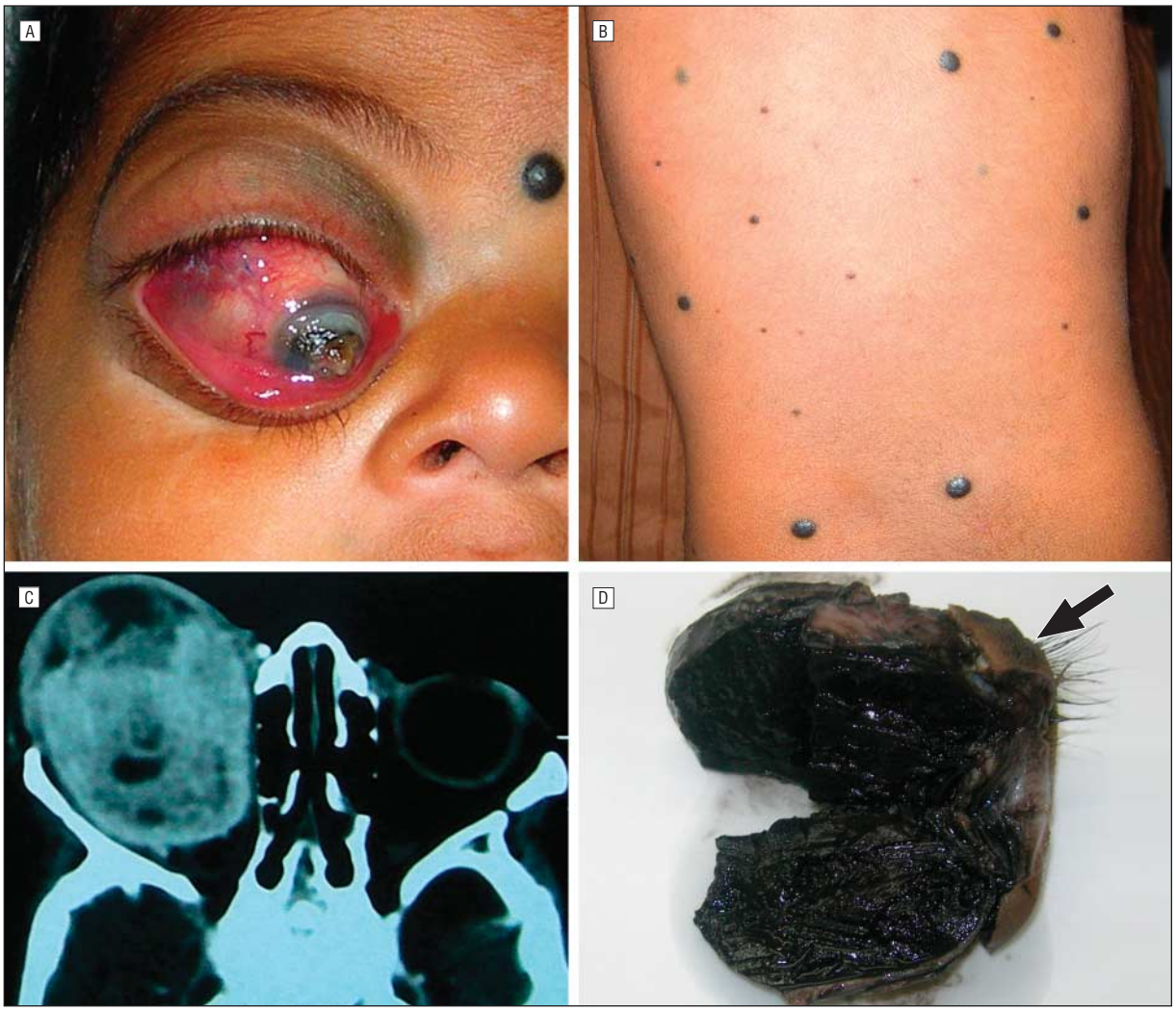
REFERENCES

1. Dintenfass L. Blood viscosity factors in severe nondiabetic and diabetic retinopathy. *Biorheology*. 1977;14(4):151-157.
2. Mohanram A, Zhang Z, Shahinfar S, Keane W, Brenner BM, Toto RD. Anaemia and end-stage renal disease in patients with type 2 diabetes and nephropathy. *Kidney Int*. 2004;66(3):1131-1138.
3. Conway B, Fried L, Costacou T, Orchard T. Hematocrit predicts end stage renal disease (ESRD) and non-ESRD mortality in type 1 diabetes with overt nephropathy. *Diabetes*. 2005;54(S1):A551.
4. Davis MD, Fisher M, Gangnon R, et al. Risk factors for high-risk proliferative diabetic retinopathy and severe visual loss: Early Treatment Diabetic Retinopathy Study report #18. *Invest Ophthalmol Vis Sci*. 1998;39(2):233-252.
5. Yoshimoto M, Matsumoto S. Changes in diabetic retinopathy and visual acuity in patients with end-stage diabetic nephropathy after the introduction of hemodialysis [in Japanese]. *Nippon Ganka Gakkai Zasshi*. 2006;110(4):271-275.
6. Watanabe Y, Yuzawa Y, Mizumoto D, et al. Long-term follow-up study of 268 diabetic patients undergoing haemodialysis, with special attention to visual acuity and heterogeneity. *Nephrol Dial Transplant*. 1993;8(8):725-734.
7. Conway B, Fried L, Orchard T. Hemoglobin and overt nephropathy complications in type 1 diabetes. *Ann Epidemiol*. 2008;18(2):147-155.
8. Vekasi J, Marton Zs, Kesmarky G, Cser A, Russai A, Horvath B. Hemorheological alterations in patients with diabetic retinopathy. *Clin Hemorheol Microcirc*. 2001;24(1):59-64.
9. Trope GE, Lowe G, McArdle B, et al. Abnormal blood viscosity and haemostasis in long standing retinal vein occlusion. *Br J Ophthalmol*. 1983;67(3):137-142.
10. Piermarocchi S, Segato T, Bertoja H, et al. Branch retinal vein occlusion: the pathogenic role of blood viscosity. *Ann Ophthalmol*. 1990;22(8):303-311.
11. Watanabe D, Suzuma K, Matsui S, et al. Erythropoietin as a retinal angiogenic factor in proliferative diabetic retinopathy. *N Engl J Med*. 2005;353(8):782-792.
12. Orchard TJ, Dorman JS, Masser RE, et al. Prevalence of complications in IDDM by sex and duration: Pittsburgh Epidemiology of Diabetes Complications Study II. *Diabetes*. 1990;39(9):1116-1124.
13. Orchard TJ, Dorman JS, Maser RE, et al. Factors associated with avoidance of severe complications after 25 yr of IDDM: Pittsburgh Epidemiology of Diabetes Complications Study I. *Diabetes Care*. 1990;13(7):741-747.
14. National Institute of Health, Department of Health, Education and Welfare. 1978 Lipid Research Clinics Program. Washington, DC: US Government Printing Office; 1975. NIH publication 75-628.
15. Levey AS, Bosch J, Lewis B, Greene T, Rogers N, Roth D; Modification of Diet in Renal Disease Study Group. A more accurate method to estimate glomerular filtration rate from serum creatinine: a new prediction equation. *Ann Intern Med*. 1999;130(6):461-470.
16. Hypertension Detection and Follow-up Program Cooperative Group. The hypertension detection and follow-up program. *Prev Med*. 1976;5(2):207-215.
17. Lloyd CE, Klein R, Maser RE, Kuller LH, Becker DJ, Orchard TJ. The progression of retinopathy over 2 years: the Pittsburgh Epidemiology of Diabetes Complications (EDC) Study. *J Diabetes Complications*. 1995;9(3):140-148.
18. Klein R, Klein B, Moss S, Cruickshanks K. The Wisconsin Epidemiologic Study of Diabetic Retinopathy XVII: the 14-year incidence and progression of diabetic retinopathy and associated risk factors in type 1 diabetes. *Opthalmol*. 1998; 105:1801-1815.
19. Ellis D, Buffone G. A new approach to the evaluation of proteinuric states. *Clin Chem*. 1977;23(4):660-670.
20. Harrell F. Survrisk.pak with %SPLINET MACRO. http://biostat.mc.vanderbilt.edu/twiki/pub/Main/SasMacros/survrisk.txt. Accessed July 16, 2008.
21. Nathan DM, Zinman B, Cleary PA, Backlund J-YC, Miller R, Orchard TJ. Modern-day clinical course of type 1 diabetes mellitus: the DCCT/EDIC experience 1983-2008. *Diabetologia*. 2008;51(suppl 1):S50 Abstract 106.
22. Craig KJ, Williams J, Riley S, et al. Anaemia and diabetes in the absence of nephropathy. *Diabetes Care*. 2005;28(5):1118-1123.
23. Thomas M, MacIsaac R, Tsalamandris C, et al. Anemia in patients with type 1 diabetes. *J Clin Endocrinol Metab*. 2004;89(9):4359-4363.
24. Thomas MC, MacIsaac RJ, Tsalamandris C, Power D, Jerums G. Unrecognized anemia in patients with diabetes: a cross-sectional survey. *Diabetes Care*. 2003; 26(4):1164-1169.
25. Thomas S, Rampersad M. Anaemia in diabetes. *Acta Diabetol*. 2004;41(suppl 1) :S13-S17.
26. Cusick M, Chew E, Hoogwerf B, et al; Early Treatment Diabetic Retinopathy Study Research Group. Risk factors for renal replacement therapy in the Early Treatment Diabetic Retinopathy Study (ETDRS): Early Treatment Diabetic Retinopathy Study Report No. 26. *Kidney Int*. 2004;66(3):1173-1179.
27. Bosman D, Winkler A, Marsden J, Macdougall I, Watkins P. Erythropoietin deficiency anaemia is common in early diabetic nephropathy. *Diabetes Care*. 2001; 24(3):495-499.
28. Fischer H, Schroeter W, Nauck M. Anemia in patients with type 1 and type 2 diabetes. *Diabetes*. 2005;54(suppl 1):A832.
29. Orchard T, Conway B. Are hemoglobin levels elevated in type 1 diabetes? Presented at: The 43rd Annual Meeting of the European Diabetes Epidemiology Group of the EASD; April 5-8, 2008; Lo-skolen Elsinore, Denmark.
30. Pichler G, Urlesberger B, Jirak P, et al. Reduced forearm blood flow in children and adolescents with type 1 diabetes (measured by near-infrared spectroscopy). *Diabetes Care*. 2004;27(8):1942-1946.
31. Krantz S, Jacobson L. *Erythropoietin and the Regulation of Erythropoiesis*. Chicago, IL: The University of Chicago Press; 1970.
32. Haffner SM, Klein R, Dunn JF, Moss SE, Klein BE. Increased testosterone in type 1 diabetic subjects with severe retinopathy. *Opthalmol*. 1990;97(10):1270-1274.
33. Haffner SM, Klein R, Dunn JE, Klein BE. Sex hormones and the incidence of severe retinopathy in male subjects with type 1 diabetes. *Opthalmol*. 1993;100 (12):1782-1786.
34. Chaurasia RK, Singh R, Agrawal JK, Maurya OP. Sex hormones and diabetic retinopathy. *Ann Ophthalmol*. 1993;25(6):227-230.
35. Al-Khoury S, Afzali B, Thomas S. Diabetes, kidney disease, and anaemia: time to tackle a troublesome triad? *Int J Clin Pract*. 2007;61(2):281-289.
36. Romagnoli C, Zecca E, Gallini F, Girlando P, Zuppa A. Do recombinant human erythropoietin and iron supplementation increase the risk of retinopathy of prematurity? *Eur J Pediatr*. 2000;159(8):627-628.
37. Brown MA, Baron AE, France EK, Hamman RF. Association between higher cumulative doses of recombinant erythropoietin and risk for retinopathy of prematurity. *J AAPOS*. 2006;10(2):143-149.
38. Takagi H, Watanabe D, Suzuma K, et al. Novel role of erythropoietin in proliferative diabetic retinopathy. *Diabetes Res Clin Pract*. 2007;77(suppl 1):S62-S64.
39. Diskin CJ, Stokes TJ, Dansby LM, Radcliff L, Carter TB. A hypothesis: can erythropoietin administration affect the severity of retinopathy in diabetic patients with renal failure? *Am J Med Sci*. 2007;334(4):260-264.
40. Drüeke T, Locatelli F, Clyne N, et al; CREATE Investigators. Normalization of hemoglobin level in patients with chronic kidney disease and anemia. *N Engl J Med*. 2006;355(20):2071-2084.
41. Singh AK, Szczech L, Tang KL, et al; CHOIR Investigators. Correction of anemia with epoetin alfa in chronic kidney disease. *N Engl J Med*. 2006;355(20):2085-2098.
42. Turczyński B, Michalska-Małecka K, Słowińska L, Szczesny S, Romaniuk W. Cor-

relations between the severity of retinopathy in diabetic patients and whole blood and plasma viscosity. *Clin Hemorheol Microcirc*. 2003;29(2):129-137.
43. Little HL, Sacks AH. Role of abnormal blood rheology in the pathogenesis of diabetic retinopathy. *Trans Sect Ophthalmol Am Acad Ophthalmol Otolaryngol*. 1977; 83(3 pt 1):522-534.
44. McMillan DE. Plasma protein changes, blood viscosity, and diabetic microangiopathy. *Diabetes*. 1976;25(suppl 2):858-864.
45. Lip PL, Blann AD, Jones AF, Lip GY. Abnormalities in haemorheological factors and lipoprotein (a) in retinal vascular occlusion: implications for increased vascular risk. *Eye*. 1998;12(pt 2):245-251.
46. Tong Z, Yang Z, Patel S, et al; Genetics of Diabetes and Diabetic Complication Study Group. Promoter polymorphism of the erythropoietin gene in severe diabetic eye and kidney complications. *Proc Natl Acad Sci U S A*. 2008;105(19): 6998-7003.

Archives *Web Quiz Winner*

Congratulations to the winner of our June quiz, Sachindra Laishram, MD, Consultant Cornea & Glaucoma Services, Shija Eye Care Foundation, Langol, Manipur, India. The correct answer to our June challenge was congenital melanocytoma. For a complete discussion of this case, see the Letters: Research Letters section in the July *Archives* (Bajaj MS, Khuraijam N, Sen S, Pushker N. Congenital melanocytoma manifesting as proptosis with multiple cutaneous melanocytic nevi and oculodermal melanosis. *Arch Ophthalmol*. 2009;127[7]:937-939).



Be sure to visit the *Archives of Ophthalmology* Web site (http://www.archophthalmol.com) and try your hand at our Clinical Challenge Interactive Quiz. We invite visitors to make a diagnosis based on selected information from a case report or other feature scheduled to be published in the following month's print edition of the *Archives*. The first visitor to e-mail our Web editors with the correct answer will be recognized in the print journal and on our Web site and will also be able to choose one of the following books published by AMA Press: *Clinical Eye Atlas, Clinical Retina*, or *Users' Guides to the Medical Literature*.