Rapid HbA_{1c} Testing in a Community Setting

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OBJECTIVE — To determine whether the DCA 2000 analyzer provides valid and reliable HbA_{1c} results when used under field conditions and operated by nonmedical personnel. This study was part of a community diabetes education program, the Native American Diabetes Project, in which HbA_{1c} was measured as an indicator of average glycemic control.

RESEARCH DESIGN AND METHODS — Two study samples were taken, the first in the spring of 1994 and the second in the spring of 1995. Seven community members in 1994 and six new community members in 1995 were trained over 2 days, using standard protocol, to operate the DCA 2000 HbA_{1c} analyzer and to collect two capillary blood samples from participants in the Native American Diabetes Project. Duplicate DCA 2000 HbA_{1c} measurements performed by the community workers were compared with measurements from a high-performance liquid chromatography (HPLC) system. Validity and reliability measures were calculated.

RESULTS — Of the participants, 43 were studied in 1994 and 14 in 1995. Comparison of the mean DCA 2000 results with those of HPLC showed high validity, with the absolute relative difference between the mean DCA 2000 and the external reference of HPLC (| mean DCA 2000 - HPLC | /HPLC) as 4.0 and 2.0% for 1994 and 1995, respectively. The Pearson correlation coefficients (r) between these two measures were 0.968 and 0.996 for 1994 and 1995, respectively. While the 1994 data appeared to have less validity for values >10%, they included only one value with a 60-min warm-up of the DCA analyzer. The 1995 data, all collected after a 60-min warm-up, had good correlation throughout the range of values. The within-run reliability was excellent, with an intraclass correlation coefficient of reliability of 0.959 and 0.975 for paired samples, for 1994 and 1995 respectively. The mean coefficient of variation for these paired measures was 3.0% in 1994 and 2.8% in 1995. Both validity and reliability were improved by changing the warm-up period of the DCA 2000 analyzer from 5 to 60 min. All correlation coefficients were statistically significant ($P < 0.0001$).

CONCLUSIONS — The DCA 2000 gave valid and reliable HbA_{1c} results when operated in a community setting by nonmedical personnel. Extending the warm-up period of the device to 60 min slightly improved the validity and reliability of the test.

The present study was conducted as part of the Native American Diabetes Project, a 4-year diabetes community intervention in eight Native American communities. The Native American Diabetes Project was designed to improve glycemic control and diabetes-related morbidity, using a community-based curriculum to increase exercise and healthful dietary behavior. HbA_{1c} was selected as the outcome measure for the present study because it has been established as a measure of average glucose and is now routinely used to monitor glycemic control in individuals with diabetes (1). Furthermore, the landmark Diabetes Control and Complications Trial (DCCT), using HbA_{1c} as a measure of glycemic control, provided evidence linking glycemic control to the development and progression of diabetic complications (2,3). The DCA 2000 HbA_{1c} analyzer (Bayer Diagnostics, Tarrytown, NY) was selected to measure HbA_{1c} in the three field sites for its portability, rapid on-site results, and ease of use. Because the analyzer had not been studied in a community setting with nonmedical operators, the purpose of the present study was to determine validity (accuracy) and reliability (precision) of the results in this setting and from year to year.

RESEARCH DESIGN AND METHODS

Setting and target population

The study setting was three field sites that served eight Native American communities. Six bilingual community workers (two for each site) were recruited in 1994 from the communities participating in the study, and six new bilingual community workers were hired in 1995. One person, also a community member, remained with the project in the position of field coordinator for the 2 years of the study. Three of the community workers had some medical background before this study, two as tribal health workers and the other as an emergency medical technician. They received ~6 h of training at the University of New Mexico over 2 days by a sales representative who used an expanded version of the manufacture's standard protocol. A return demonstration and verbal questioning ensured proficiency in the operation of the analyzer and in the collection of blood samples. The field workers then returned to small offices in or adjacent to the communities in the study. They had phone contact with study coordinators, and research personnel from the University of New Mexico visited the sites weekly. Study participants were community members with di-

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CV, coefficient of variation; HPLC, high-performance liquid chromatography.

abetes who were recruited by the community workers through letters, phone calls, and in-person visits. The participants came to one of the three field offices for an interview and clinical measures as part of baseline data collected for the Native American Diabetes Project. The first 47 participants (15, 15, and 17 from each site) who were recruited for the project participated in the 1st year of the HbA_{1c} validity and reliability study. During the 2nd year, 5 participants from each site were selected (15 total), who were expected to have a range of HbA_{1c} results based on their HbA_{1c} values from 1994. The DCA 2000 records values above 14 as >14; therefore, 4 participants in 1994 and 1 participant in 1995 with a DCA 2000 result of >14 were excluded from our analysis, leaving 43 and 14 participants for 1994 and 1995, respectively.

Table 1—*Measures of validity and reliability by year and warm-up period*

	Validity (mean DCA vs. HPLC)		Reliability (DCA 1 and DCA 2)			
	ARD %	*r*	r_I	*r*	CV (%)	MAD
Overall results 1994	4.0 ± 3.7 (43)	0.968	0.959	0.959	3.0	0.1 (0–2.3)
5-min warm-up	4.9 ± 4.4 (22)	0.962	0.948	0.950	3.4	0.1 (0–2.3)
60-min warm-up	3.1 ± 3.3 (21)	0.971	0.969	0.977	2.6	0.1 (0–0.8)
Overall results 1995	2.0 ± 1.2 (14)	0.996	0.975	0.974	2.8	0.1 (0–0.9)

Data are means ± SD (*n*) or median (range), unless otherwise indicated. ARD, absolute relative difference; *r*, Pearson correlation coefficient; r_I, intraclass correlation coefficient of reliability; CV, mean within-run coefficient of variation; MAD, median absolute difference (range) between 1st and 2nd DCA 2000 measurements as absolute percentage of HbA_{1c}. For all correlation coefficients, $P < 0.0001$.

Blood sample collection

Blood samples were obtained by fingerstick using a glycolet device (Bayer Diagnostics, Elkhart, IN) after sterilizing the finger with an alcohol swab. The first sample was collected directly into the 1-μl DCA 2000 capillary tube, which was then snapped into the reagent cartridge and immediately placed into the DCA analyzer for analysis. The second fingerstick sample consisted of 75 μl of blood collected into a heparinized plastic microtainer tube (Becton Dickinson, Rutherford, NJ) and a repeat DCA 2000 capillary tube sample, collected and analyzed as above. The microtainer tube was refrigerated for up to 7 days before being shipped at 4°C to the University of Missouri for analysis by high-performance liquid chromatography (HPLC).

HbA_{1c} methods

The DCA 2000 analyzer, which relies on an immunochemical technique for the measurement of HbA_{1c}, was used throughout the study following the manufacturer's protocol. The method has been previously described and tested in clinical settings (4,5). The instrument is small (10.7 × 9.4 × 9.4 in, 11 lb) and uses a reagent cartridge that contains all the necessary reagents required to perform the HbA_{1c} assay. The assay uses a 1-μl capillary blood sample and reports results in 9 min as a percentage of HbA_{1c}. The DCA 2000 records values above 14 as >14. One reagent lot was used throughout the 1st year of the study; a second reagent lot was used during the 2nd year. During the 1st year, the warm-up period for the DCA 2000 instrument was changed from 5 to 60 min. A 60-min warm-up period was used during the 2nd year. Two levels (normal and abnormal) of the manufacturer's lyophilized quality control specimens were analyzed with each run; a different lot of each control was used for each year of the study. Since no accepted standard exists and a "true" HbA_{1c} concentration cannot therefore be determined, all data were compared with an established reference that has been shown to be consistent over time and has an established relationship to mean blood glucose (2). The Primus CLC330 Analyzer (Primus, Kansas City, MO) was used to measure HbA_{1c} in all of the samples from the present study. The HPLC system at the University of Missouri was calibrated by a reference method and was monitored to ensure long-term reproducibility (6,7). In this way, consistency of results during the study (between lots of the reagent and years of the study) could be verified.

Statistical methods

Validity of the DCA 2000 analyzer, relative to the HPLC method, was determined by two methods: *1*) the mean absolute relative difference, which is the mean of the absolute value of the difference between the reference value and the observed mean of the two DCA 2000 samples, divided by the reference value (| mean DCA 2000 - HPLC | /HPLC) (8), and *2*) the Pearson correlation coefficient. The reliability was determined by calculating *1*) the intraclass correlation coefficient of

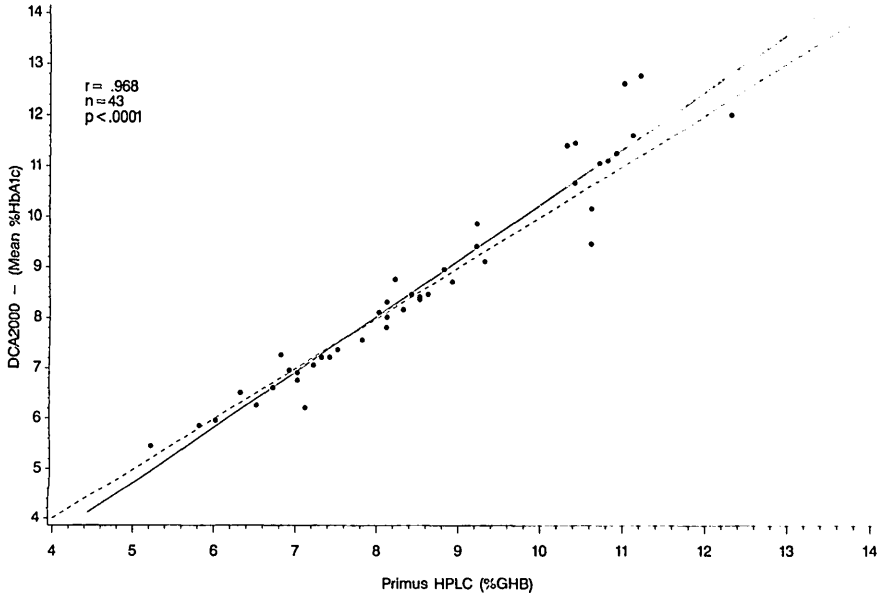

Figure 1—*Validation of DCA 2000 method; 1994 HbA_{1c} values.* ——, *linear regression;* - - - -, y = x.

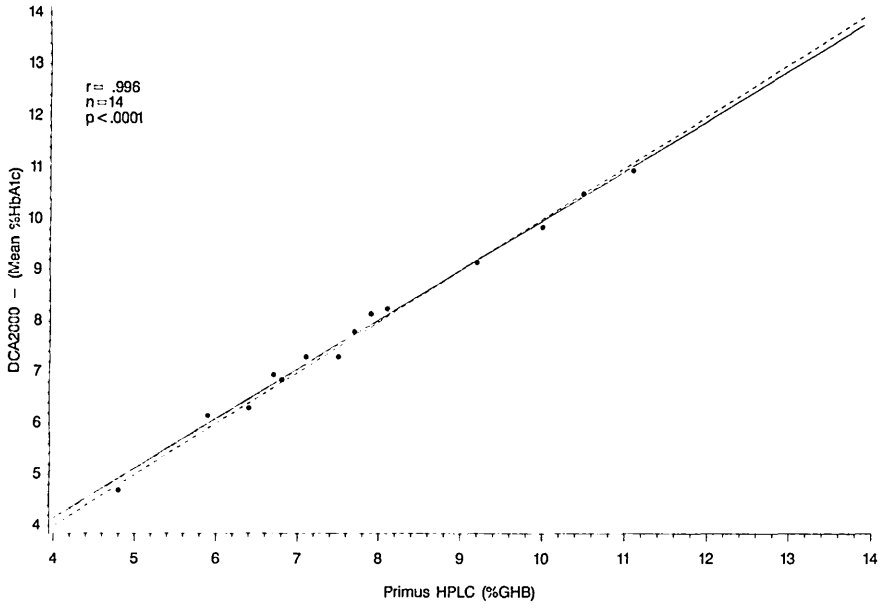

Figure 2—*Validation of DCA 2000 method; 1995 HbA_{1c} values.* ——, *linear regression;* - - - -, y = x.

reliability (9), 2) the Pearson correlation coefficient, and 3) the coefficient of variation (CV) (8). Reliability and validity measures were computed for the overall 1994 data, the two separate warm-up periods in 1994, and the overall 1995 data.

RESULTS— Study participants had HbA_{1c} values ranging from 4.6 to 13.0% from the DCA 2000 method and 4.8 to 12.5% from the HPLC method. Measures of validity and reliability are presented in Table 1. Mean DCA 2000 results compared with those of the HPLC showed high validity during 1994 and 1995, with a mean absolute relative difference between DCA 2000 measurements and HPLC measurements of 4.0% (overall) in 1994 and 2.0% in 1995. The Pearson correlation coefficients for the 2 years, 1994 (all data) and 1995, were 0.968 and 0.996 (Figs. 1 and 2), respectively. While there appeared to be less validity for values >10% in the 1994 data, the data included only one value with a 60-min warm-up. The 1995 data, which were all collected after a 60-min warm-up, had good correlation throughout the range of values. Within-run reliability was excellent in both years, with intraclass correlation coefficients of reliability of 0.959 in 1994 and 0.975 in 1995, and the mean within-run CV was <3.4% for all analyses (Table 1). The median absolute difference between the first and second DCA 2000 measurements was 0.1% (Table 1). Based on analysis of the quality-control specimens, the between-run CVs were 3.2% (mean normal control = 4.7%) and 4.1% (mean abnormal control = 9.7%) for 1994 and 3.2% (mean normal control = 4.8%) and 2.8% (mean abnormal control = 10.9%) for 1995. All correlation coefficients were statistically significant ($P < 0.0001$). To determine the validity of the test using only one DCA 2000 measurement rather than the mean of two measures, the first DCA 2000 measurement was compared with the HPLC measurement. The mean absolute relative difference was 4.1% in 1994 and 1.2% in 1995, and the Pearson correlation coefficients were 0.974 in 1994 and 0.993 in 1995, indicating excellent validity for a single DCA 2000 result. During the 1st year, the results were reviewed halfway through the study, and the warm-up period was increased from 5 to 60 min. The longer warm-up period increased both the validity and reliability measurements in the second half of the study in 1994 (Table 1). Consequently, throughout the 2nd year, a warm-up period of 60 min was used, which gave clear improvements in results.

CONCLUSIONS— The DCA 2000 analyzer was used successfully in rural field offices by nonmedical personnel who were supervised by telephone and weekly on-site visits. The method was well accepted by the research staff, field workers, and study participants. This particular method had specific advantages for use in field studies: *1*) the field staff required minimal training, which suggests that the instrument is useful in situations where laboratory personnel are not available on-site, 2) the analyzer is small and convenient for an office setting, 3) only a minute amount of blood (1 μl) is required for the test and may be acquired from a fingerstick rather than a venipuncture, and *4*) a 9-min analysis time allows prompt feedback of the results for the patient. Furthermore, the consistent relationship between the DCA 2000 analyzer and a reference method over the first 2 years of the Native American Diabetes Project is a good indication of the validity of this method for other long-term diabetes studies. Lastly, a 60-min warm-up period for the analyzer was associated with improved validity and reliability (accuracy and precision), compared with the results obtained using a 5-min warm-up. The improved results may have been due to a more stable optimal temperature condition for the instrument with a longer warm-up period. A limitation of this method is the inability to record values >14%. This requires researchers to report median rather than mean values, and it does not allow the clinician to see values >14%. We believe this easy-to-use portable analyzer has advantages that outweigh these disadvantages in rural community settings.

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