External Quality Assessment of HbA$_{1c}$ for Point of Care Testing

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ABSTRACT

Objectives. To evaluate the long term total imprecision of HbA$_{1c}$ testing within the county of Uppsala in relation to the Swedish analytical goal of coefficient of variation (CV) <3% for HbA$_{1c}$ and to study the cost of an external quality assurance program for point-of-care HbA$_{1c}$ The county uses Bayer DCA 2000™ for point-of care HbA$_{1c}$ testing currently having 23 of these instruments.

Methods. Method imprecision was assessed by analysis of patient samples performed as split samples during a 3 year period (2002-2004) as part of the quality assurance program for point-of-care HbA$_{1c}$ testing. The samples were first analysed on a Bayer DCA 2000™ and the samples were then sent to the centralised laboratory for reanalysis with an HPLC system (Variant II™, Biorad). The testing was performed approximately 8 times per year with each instrument.

Results. The median CV between the HPLC method and the point-of-care instruments for each unit was slightly higher than 3%.

Conclusion. The DCA 2000™ systems have an acceptable imprecision and agreement with the central laboratory. The test results show acceptable agreements within the county regardless where the patient is tested. The cost of the external quality assurance program is calculated to be approximately SEK 1340 (Euro 150) per instrument.

INTRODUCTION

Contemporary lifestyles and increased life expectancies, have contributed to a dramatic rise in the prevalence of diabetes mellitus during the last decades [1]. In 1995, the worldwide prevalence of diabetes was approximately 4.0% in adults and this figure is expected to increase to 5.5% by the year 2025 [2]. Diabetes is linked to several serious health problems, including cardiovascular disease and kidney disease and cause diabet-

ic microangiopathy in the kidneys, nerves and eyes. Good glycaemic control reduces the risk of development and progression of late diabetes complications [3-5]. Glycated haemoglobin [HbA_{1c}] is widely accepted as the best marker for long-term glycaemic balance in diabetic patients and the treatment targeted based on the HbA_{1c} results [6-8]. In Sweden HbA_{1c} values are also used as indicators of the quality of diabetes care when comparing different care regions [9]. To obtain a good glycaemic control it is essential that the patient and physician work together and that the patient get regular feedback on the treatment results. The feedback is considered to be more effective if the test results are available during the consultation. This requires that the patient either provides blood samples prior to the consultation or that the unit has a capability to perform rapid testing of relevant markers. This has led to the development of point-of-care testing POCT instruments for measuring glucose, HbA_{1c} and urine albumin excretion. HbA_{1c} testing is presently performed both at central laboratories and as POCT. Over the last decade several HbA_{1c} methods have been developed for POCT settings. The methods provide rapid test results that are greatly appreciated by both clinicians and patients. The availability of the test results and the discussion of test results during the consultation is believed to increase patient compliance to the treatment plan. As diabetes patients are visiting both hospital and primary care units in some areas as part of a "shared-care" program it is important that the test result is the same regardless of where the HbA_{1c} test is performed. There is a Swedish goal of 3% for total coefficient of variation (CV) for HbA_{1c} (10). These recommendations are set in relation to a very low intraindividual CV (<2%).

There is a limited patient transfer between primary care centers while there is a considerable movement between primary care and the hospital. It is thus important to be aware of the total CV within the county council area. As the clinician often does not distinguish between HbA_{1c} results performed with the centralised method and the POCT method, it is important that both methods provide similar test results.

The aim of this work was to evaluate the analytical performance of the HbA_{1c} methods used within the county of Uppsala and to calculate the cost for this quality control program.

MATERIALS AND METHODS

Study population
In the county of Uppsala health care region (all primary care units and hospitals within the Uppsala county council area), primary and secondary care units are using DCA 2000™ for POCT analysis of HbA_{1c}. All instruments are required to participate in a quality assurance program in order to monitor the quality of the instruments. The quality control consists of three parts: An instrument control with an optical test cassette is performed once a month. Internal quality controls are performed once a week with one normal and one abnormal control. External quality controls are performed as split samples once a month with interruptions for summer and major holidays. Usually, approximately 8 external controls are run per instru-

ment and year. The external quality assurance program is performed with a split sample technique utilizing whole blood from diabetes patients. The samples are collected in vacutainer tubes containing EDTA (Becton Dickinson, Franklin Lakes, NJ, USA). The DCA 2000™ users perform a routine patient analysis and then the sample is sent to the central laboratory for HbA_{1c} analysis with an HPLC technique. Both results are reported to the person responsible for supervising the quality assurance program.

DCA2000™

DCA2000™ (Bayer, Tarrytown, NY, USA) measures HbA_{1c} based on specific inhibition of latex immunoagglutination. Special single use cartridges containing the reagents are used. No manual haemolysis step is required. 1 µL blood is collected in the cartridge and the unit is then inserted into the analyzer. The measurement is then performed automatically and the instrument measures the concentration of HbA_{1c} and total hemoglobin and the ratio is presented as $\%HbA_{1c}$. The procedure is fully automated and the test time is approximately 6 min. At the beginning of 2002 there were 11 DCA 2000™ instruments in the program. The number had increased to 23 at the end of 2004.

Variant II™

Variant II™ (BioRad Laboratories, Hercules, CA, USA) was the HbA_{1c} method used at the central laboratory. 5 µL are mixed with 1 µL diluent and the samples are loaded into an autoinjector. The samples are then injected into the HPLC system. The haemoglobin is separated and the HbA_{1c} fraction is calculated as $\%HbA_{1c}$ in relation to total haemoglobin.

The Variant II™ method is validated by participation in the Swedish national external quality assurance organisation (Equalis, Uppsala Sweden) external quality assurance programs for HbA_{1c} and filter paper-HbA_{1c}.

Statistical calculations

All calculations were performed with Microsoft Excel and Maple (Microsoft Corporation, Seattle, WA, USA). The test results from the DCA 2000™ and Variant II™ were handled as duplicate analyses when the CV was calculated. All samples analyzed as part of the quality assurance program during the same week were used to calculate a median CV. The CV is calculated without considering whether the difference between the two instruments were positive or negative.

Cost evaluation

The economical calculations are based upon interviews of three randomly selected laboratory technicians performing HbA_{1c} assays at primary health care centers and the laboratory technician responsible for the quality assurance program. Each technician estimated the time used for the external quality assurance testing. The labor cost for a laboratory technician, including overhead, was estimated at SEK 5 per

min. As the initial DCA 2000™ test was performed as part of the normal patient care, no additional reagent cost was included in the calculation. The samples were sent with the regular test sample transports, thus there was no extra cost for the transport of the samples. The cost charged by the centralized laboratory for HbA_{1c} was SEK 52/sample which was included in the cost estimate. There was also a labor cost for handling the test result at the primary care centre and for the laboratory technician responsible for the program for assembling the test results for each quality assurance round.

RESULTS

Total CV for all instruments

The mean CV for the units involved in the external quality assurance program during 2002-2004 is presented in Fig. 1. During 2002 the mean CV varied slightly around the 3% level from one test occasion to another. During early fall, 2003 an increased CV was observed. There was no noticeable effect on patient mean values during this time period. The mean CV decreased during 2004 and was again varying around the 3% threshold. Median CV for the whole study period was slightly higher than 3%. When comparing CV for different HbA1c intervals (<6, 6-8 and >8) no significant differences were noted.

Figure 1. Mean coefficients of variation (CV) of all units on each test occasion during 2002-2004. The CV for each unit was calculated based on split samples analysed locally (DCA 2000 TM) and at the central lab (Variant II TM). The dotted line denotes the Swedish goal of 3% for total coefficient of variation.

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Economical calculation

The DCA 2000™ assay was a routine test and thus no time was attributed to the quality assurance program. The time used to send the samples to the centralised laboratory and handle the test result was estimated to be 5 min per sample (SEK 25). Eight samples per year and 23 instruments add up to SEK 4600 per year. The list price for the centralised test is SEK 52 per sample that represents a cost of SEK 9568 per year. This covers all the costs for the centralised HbA_{1c} test. The time spent at the primary care units for handling the test results from the centralized HbA_{1c} test was also estimated to be 5 min per sample (SEK 4600). There is also a cost for the laboratory technician responsible for monitoring of the program and providing feed back which is estimated to be approximately 2400 min per year (SEK 12000). Thus, the total yearly cost for the quality assurance program is approximately SEK 30800 (Table 1). SEK 30800 corresponds to approximately Euro 3380.

	Per sample	Total cost/year
Variant II	52	9500
Transport	0	0
Personnel cost		
Handling of sample	25	4600
Handling of Variant II test result	25	4600
Compilation of results		12000
Total yearly expense		30768
Yearly cost per instrument		1340

Table 1. Expenses for the external quality assurance program for POCT HbA_{1c}. The calculation is based on the number of DCA 2000™ instruments at the end of 2004 (n=23) and an average of eight tests per year. Prices are calculated in Swedish kronor (SEK).

DISCUSSION

Glycated proteins are formed posttranslationally due to the slow nonenzymatic reaction between protein amino groups and the aldehyde group of glucose [11-13]. The synthesis of glycated haemoglobin is a function of the concentration of glucose to which the erythrocytes are exposed and the exposure time [14,15]. Usually HbA_{1c} is an index of mean glycaemia during the preceding 120 days. However, conditions that shorten the erythrocyte survival also lower HbA_{1c} regardless of the assay method as the exposure time is reduced [16,17].

In comparison with commercially available quality assurance programs the split-

sample technique makes it easier to focus on samples with HbA_{1c} levels in the range of the patient samples. A quality assurance program often utilises samples from healthy donors with HbA_{1c} within the reference range. A low CV in the normal (non-diabetic) reference range is of less clinical importance than a low CV in the 6-8% range.

We have calculated the total cost for the program. The use of split-sample technique with patient samples reduces quality assurance cost in relation to commercially available quality assurance programs. The net costs for the county is somewhat lower than our figures as the Variant II costs is based on the list price for the test. Thus, the price also includes costs that are volume independent (e.g. instrument investment and service) which is a cost that is the same regardless of the quality assurance program. Also, the units can to some extent choose when to run the tests, depending on the day-to-day workload, thus utilising staff more effectively. The median total CV for the HbA_{1c} testing that a single patient will encounter when moving between primary and secondary care is close to 3%. There were occasional samples that differed more than 7% but these differences were not repeated at the next sampling time. Thus, these erroneous results do not seem to be due to systematic problems. The increased CV in the early autumn 2003 could be due to both increased variability of the DCA 2000™ instruments and the Variant II™ instrument. There were 5 new units that started performing POCT testing for HbA_{1c} and they had higher CV than the rest of the units during the first test rounds. During 2004 the same units had improved their CVs not being different from the rest of the DCA 2000™ instruments. There is no sign of obvious drift in patient results and the controls fell within the predefined limits during this period for the Variant II' instrument which could be an indication that there were no problems with the instrument. However, during the time period with increased CV there were also an increased variability in the internal controls with a variability of up to 7% for the high control while during other time periods the control had been much more stable. Thus, it seemed to have been a problem with the Variant II™ instrument during this period that was not detected by the patient values. Accordingly, the increased CV during this time period was probably due to a combination of the introduction of new instruments and increased variability of the Variant II instrument reflected by the results of the internal control.

We conclude that the median interassay CV in our health care region with a mixture of HPLC and immunological methods for HbA_{1c} fulfils the recommendation of a CV < 5% [18] and is close to 3%. The split-sample technique is a cost-effective way of performing external quality control of HbA_{1c} testing with a high proportion of the samples in the diabetic range.

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