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Working Group for the Rational Use of Urinary Test Strips

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Protocol for the evaluation of reagent sets (Chemicals and Instruments) with special reference to "SPOT-tests"

The continuous development and marketing of new reagents and instruments makes it desirable to design a protocol which utilizes acceptable and well recognized methods. Such an approach is particularly important since many laboratories are faced with a demand to compare different reagent sets: It would be an advantage also if manufacturers would describe their products in these terms. The working group is in the process of finalazing a report on evaluation of test strips, highlighting also problems of discrimination values in binary and paucinary tests.
The following provisonal protocol has been designed to estimate imprecision and inaccuracy from patient samples as well as from control and reference materials. Also, the with-in, between-series and total variation should be estimated. Special methods are advised for the estimation of linearity, and the effects of lipemia and hemolysis. Special attention should be given to the effect of the sample matrix. Several recommendation for evaluation of methods have been published with details on statistical treatment of the results (1-3).

1. Imprecision and inaccuracy

1.1. PATIENT SAMPLES
About 36 samples should be determined in duplicates on three different occasions. The samples should also be analyzed with an agreeable, well controlled method which agrees as much as possible with a reference methodology. The patient samples should be of different concentrations.

1.1.1. Estimate the regression between the means of the duplicates and the values obtained by that used as a reference.

1.1.2. Estimate the standad deviation from the results of the duplicate analyses of three levels, subnormal, "decision interval" and increased values.

1.1.3. Identify and reexamine all outliners. Evaluate deviation in view of matrix on systemic effects.

1.2. REFERENCE MATERIAL
Use three levels of concentrations of a reference material which is as close as possible to human. The number of analyses on each level should be abut 25.

1.2.1. Estimate the mean, standard deviation and coefficient of variation for each reference material.

1.2.2. Use the reference material which is closest to the decision level and determine the concentrations in 15 different runs. Several runs may be carried out per day but they must then be initiated the normal way, including shut-down of the instrument, recalibration etc. In each run 5 values should be determined.

1.2.2.1. Calculate the average within-batch variation.

1.2.2.2. Calculate the between-batch variation.

1.2.2.3. Calculate the total variation.

2. **Testing other parameters of performance.**

2.1. HEMOLYSIS (addition of hemolysed erythrocytes).
A hemolysate is prepared from washed human erythrocytes which are frozen and thawed several times. A series of dilution is made from this concentrate, using 0.15 mol/L NaCl as diluent. Suitable concentrations are 320, 160, 80, 40, 20, 10 and 5 g/L. A serum sample is diluted 1:10 with the appropriate hemoglobin sample. These samples as well as a sample diluted with 0.15 mol/L NaCL 1:10 are determined in duplicates.

2.2. LIPEMIA (addition of fat emulsion, Intralipid^{R} is provisionally suggested).
The initial concentration of 100 g/L is diluted with 0.15 mol/L NaCl to concentrations of 100, 50, 25, 12 and 6 g/L. A serum sample is then diluted 1:10 with these dilutions of the fat emulsion, including a zero sample. Samples should be analysed in duplicates.

2.3. BILIRUBINEMIA.
A similar test of the influence of bilirubin should be carried out. A suitable test material is still to be found.

2.4. DRUGS.
Drugs interfering with the methods chemically should be systematically tested. The booklet "Drug interference and effects in Clinical Chemistry" (Apoteksbolaget, Stockholm, Sweden) should be consulted.

2.5. LINEARITY
The linearity of the method should be tested over as wide a range as possible. Select a high and a low concentration serum and mix them in a suitable manner to obtain at least five points. Plot the found vs. the calculated results, if possible describe the performance using regression analysis.

2.6. SAMPLE VOLUME.
Analytical results should be determined using from -10 to +10% of the recommended volume of sample.

2.7 ERYTHROCYTE, VOLUME FRACTION.
In case whole blood is recommended the effect of varying the EVF should be considered within the fysiological range.

It is understood that deviations from the outlined method could be necessary due to technical or other circumstances. Depending on the reagent set evaluated, other items might also be important to consider.

Several reagent sets (chemicals and/or instruments) are in the process of being tested in accordance with this protocol.

Extensive lists of further reference to the literature are fund in these papers.

1. IFCC Expert Group on Diagnostic Kits and Reagents.
   a) Recommendations for specifications on Labelling of Clinical Laboratory Materials.
   b) Guidelines for the Evaluation of Clinical Chemistry Kits in IFCC Recommendations and Related Documents Volume I, 1978-83 Walter De Gruyter 1984. ISBN 3-11-008766-9.
2. IFCC/WHO Principles and Recommendations of Evaluation of Diagnostic Reagent Sets used in Health Laboratories with limited resources. J Clin Chem Clin Biochem (1984) **22**; 817 with appendicies A-E (Glucose, Urea, Total Bilirubin, Total Protein and Albumin).
3. White GH and Fraser CG: The evaluation kit for clinical chemistry: a practical guide for the evaluation of methods, instruments and reagent kits. J Autom Chem (1984) **6**; 122.