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Discrepancies in International Normalized Ratio Results between Instruments: A Model to Split the Variation into Subcomponents

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BACKGROUND: Observed differences between results obtained from comparison of instruments used to measure international normalized ratio (INR) have been higher than expected from the imprecision of the instruments. In this study the variation of these differences was divided into subcomponents, and each of the subcomponents was estimated.

METHODS: Blood samples were collected at 4 different patient visits from each of 36 outpatients who were receiving warfarin treatment and were included in the study. INR was determined on 1 laboratory instrument (STA Compact®) and 3 point-of-care instruments (Simple Simon®PT, CoaguChek®XS, and INRatio™). All 4 INR instruments were compared in pairs. Linear regression was used to correct for systematic deviations. The remaining variation of the differences was subdivided into between-subject, within-subject, and analytical variation in an ANOVA nested design.

RESULTS: The mean difference between instruments varied between 1.0% and 14.3%. Between-subject variation of the differences (expressed as CV) varied between 3.3% and 7.4%, whereas within-subject variation of the differences was approximately 5% for all 6 comparisons. The analytical imprecision of the differences varied between 3.8% and 8.6%.

CONCLUSIONS: The differences in INR between instruments were subdivided into calibration differences, between- and within-subject variation, and analytical imprecision. The magnitude of each subcomponent was estimated. Within results for individual patients the difference in INR between 2 instruments varied over time. The reasons for the between- and within-subject variations of the differences can probably be ascribed to different patient-specific effects in the patient plasma. To minimize this variation in a monitoring situation, each site and patient should use results from only 1 type of instrument.

Oral anticoagulant therapy with warfarin is increasingly used. Prothrombin time (PT),[4] expressed as international normalized ratio (INR), is the standard measurement procedure used to monitor the oral anticoagulant therapy *(1, 2)*. Individual responses to warfarin are variable, and frequent monitoring of INR is required to ensure that the anticoagulation effect remains within the narrow therapeutic range. INR may be measured by standard laboratory instruments or by point-of-care (POC) instruments.

Because measurements of INR are often performed in various locations by use of different types of instruments, it is important that the differences in results between instruments are minimized and that the causes of these differences are understood. Agreement in results between laboratory INR instruments and methods improved after the introduction of WHO guidelines that launched new calibration procedures *(2)*. Nevertheless, harmonization of results from different laboratories remains challenging *(3–5)*. Different types and sensitivities of thromboplastins *(2)* and interactions between thromboplastin and coagulation factors of individual patients may influence the accuracy of the instruments *(4, 6, 7)*. Furthermore, 2 different methods, the Quick *(8)* and the Owren *(9)*, are used in the operation of INR instruments, which may also cause discrepancies *(3)*. For POC instruments the calibration parameters are determined by the manufacturers *(10)*. Some POC instruments have been calibrated by using WHO guidelines *(11)* according to the procedure developed by Tripodi et al. *(12)*, but this process is not practicable outside specialized research

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[4] Nonstandard abbreviations: PT, prothrombin time; INR, international normalized ratio; POC, point-of-care; IRP, international reference preparation; ISI, International Sensitvity Index.

centers. The performance of POC instruments has mainly been examined by comparison of POC INR results with results from standard laboratory instruments *(13–16)* or against international reference preparations (IRP) (by use of the manual tilt-tube technique) *(11, 17, 18)*. The degree of deviation of the results between the instruments depends on the POC instruments compared. In Scandinavia a supplier-independent assessment of the analytical quality of the POC instruments is performed and results from these evaluations are used to formulate recommendations of what instruments to use, for example, in general practice *(19)*.

Differences in INR results between instruments are higher than would be expected from the imprecision of each of the instruments compared *(7, 20)*. The total uncertainty of INR results has been calculated *(7)* and 1 study has been performed to investigate differences in INR over time *(21)*. However, to our knowledge, no study has been performed to address the relative contributions of different factors to observed differences between the measurements.

In the present study 3 different POC instruments and a laboratory instrument for measuring INR were compared by using 4 different samples from 36 patients obtained at different time intervals. In this study we created a model in which the variations of the differences between 2 instruments were subdivided into calibration, between- and within-subject variation, and analytical imprecision components, and then we calculated the contribution of each of these components.

Materials and Methods

PATIENTS AND STUDY DESIGN

Consecutive patients who had undergone anticoagulant treatment with warfarin for more than 1 month and were undergoing their scheduled monitoring of INR at Haraldsplass Hospital (Bergen, Norway) were asked to participate in the study. The patients received written and oral information and gave written informed consent to participate. The study was approved by the ethics committee of West Norway. INR was analyzed on the laboratory instrument STA Compact (Diagnostica Stago) and on 3 POC instruments, Simple Simon PT (Zafena AB), CoaguChek XS (Roche Diagnostics Boehringer-Mannheim), and INRatio (Hemosense). When the patients attended their scheduled INR-monitoring appointments at Haraldsplass Hospital, INR was measured on the laboratory instrument and on the 3 POC instruments. This procedure was repeated for 3 additional monitoring visits (4 measurements on each instrument for each patient) that occurred at varying time intervals. Patients were recruited from May to November 2006. Fifty-seven patients were enrolled in the study. Sixteen patients were excluded from the study because they came for fewer than 4 visits, 4 patients because they withdrew their consent, and 1 patient because of problems with blood sampling. Thirty-six patients (20 males) completed the study with 4 samples each and were included in the initial calculations. Five of these patients had been on anticoagulant treatment for less than 3 months. Median age was 67.5 years (range 41–92 years). The median (10%–90% percentile) time span between the first and fourth visits was 7.4 (3.3–17.3) weeks. There was no significant difference in results between the patients who participated for fewer than 8 weeks and the patients who participated 8 weeks or longer.

BLOOD SAMPLES

Venous and capillary blood samples were collected at the same time from each patient. Venous blood, collected in 0.109 mol/L (3.2%) trisodium citrate, was used for duplicate measurements by the Simple Simon POC instrument within approximately 30 min, and always within 2 hours *(22)*. Platelet-poor plasma was obtained by centrifugation within 2 hours *(23)* for 15 min at 2500*g*, and INR was analyzed directly in duplicate with the STA Compact. All samples for measurements on the STA Compact and Simple Simon were collected and handled identically by the same trained personnel during the whole study and after international guidelines *(24)*. Capillary blood was used for duplicate measurements on the CoaguChek XS and the INRatio instruments (2 finger sticks). The finger stick and application of blood drop on the strips were performed according to laboratory procedure by the same trained personnel during the whole study. Two experienced biomedical laboratory scientists performed the testing on the Simple Simon and another 3 performed the testing on the CoaguChek XS and INRatio. The CV% was calculated for the 3 POC instruments for the results from each scientist, and no significant difference between their results was found ($P > 0.05$, F-test).

STA Compact

The STA Compact is an automated laboratory instrument. STA-SPA 50 (Diagnostica Stago) was used as a reagent. It is a combined rabbit brain thromboplastin with added bovine plasma (Owren-based method) *(9)*. Two different batches of the reagent were used in the study, with International Sensitvity Index (ISI) values of 1.00 and 1.02, respectively. The STA Compact was calibrated with 2 calibrators from EQUALIS (External Quality Assurance in Laboratory Medicine in Sweden), which are traceable to reference thromboplastin RBT/90 from WHO *(25)*. Two calibrators and a control from EQUALIS (MediRox AB) were analyzed as control samples on the STA Compact 3 times during

Table 1. Range of 4 INR measurements (mean of the duplicates) on the hospital INR instrument (STA Compact) for all 36 patients during the study period.

Patient no.	INR range	Patient no.	INR range	Patient no.	INR range
1	2.1–4.7	13	2.3–3.1	25	2.0–2.4
2	1.5–3.1	14	3.3–4.1	26	2.6–3.0
3	1.5–2.5	15	1.5–2.3	27	2.2–2.8
4	1.5–2.2	16	2.2–4.3	28	2.2–4.6
5	1.8–2.7	17	2.4–2.7	29	1.5–2.4
6	1.1–2.5	18	2.2–3.3	30	1.8–2.4
7	1.9–2.5	19	2.6–3.0	31	2.3–2.7
8	2.5–3.6	20	2.3–3.2	32	1.9–2.7
9	2.3–3.4	21	2.5–3.9	33	1.8–2.3
10	2.4–3.4	22	2.7–3.0	34	2.8–3.4
11	2.8–3.3	23	2.0–3.1	35	1.1–3.1
12	2.1–2.9	24	2.0–3.1	36	2.2–3.0

the study period. Results are available at www.skup.nu *(26)*. Daily internal QC was performed with Scandinorm and Scandipath (Diagnostica Stago) and no systematic deviation was found during the study period. The day-to-day imprecision CV was between 2% and 4%. The analytical imprecision based on duplicate measurements on patient samples was 1.6%. The ranges of INR measured on the STA Compact for all 36 patients during the study period are shown in Table 1.

Simple Simon PT

The Simple Simon instrument contains a reader, reagent, buffer, reagent tubes, pipettes, and pipette tips, all with the same lot number *(27)*. The reagent was a combined rabbit brain thromboplastin with added bovine plasma (Owren-based method) *(9)*. The ISI was 1.25. The Simple Simon was calibrated against instruments at Scandinavian hospital laboratories by use of patient samples *(27)*. We used 2 lot numbers in this study. We analyzed blood from the first 12 patients using lot G024MI. The results from these 12 patients were excluded, however, because the producer changed the calibration procedure and decided to withdraw the lot from the market *(26)*. Internal QC was performed regularly with normal control plasma and abnormal control plasma (MediRox AB). The controls were within the target interval and stable throughout the study period. The day-to-day imprecision CVs were 2.4% and 3.7% for normal control plasma and abnormal control plasma, respectively. The analytical imprecision calculated on the basis of duplicate measurements on patient samples was 3.6%.

CoaguChek XS

The CoaguChek XS is a small hand-held meter with disposable test strips. The reactive components on the test strips are recombinant thromboplastin reagent, with an ISI of 1.0, and a peptide substrate *(28)*. CoaguChek XS uses a modified version of the Quick method *(8)*, because the end point is thrombin generation and not fibrin. The test strips are calibrated against the WHO IRP human recombinant reference thromboplastin rTF/95 and certified reference material 149S *(2, 28)*. There are no liquid controls for the CoaguChek XS. The on-board single-channel strip-control system checks reagent integrity of each test strip. However, a control designed for CoaguChek XS Plus was tested regularly 32 times on the CoaguChek XS during the study period and gave reproducible results. The day-to-day imprecision CV was 3.4%. The analytical imprecision based on duplicate measurements on patient samples was 3.2%. One patient (no. 24) stated that he had systemic lupus erythematosus. Blood samples from this patient showed lupus-antibody–sensitive activated partial thromboplastin time that was prolonged and activities of coagulation factors V, VII, and X that were within reference intervals, indicating that the patient had antiphospholipid antibody syndrome *(29)*. A precaution is given in the instrument manual that antiphospholipid antibodies may lead to falsely high INRs, and therefore this patient was excluded from the calculation results from the with CoaguChek XS.

INRatio PROTHROMBIN TIME MONITORING SYSTEM

The INRatio is a small hand-held meter that uses single disposable test strips. The reactive component on the test strip is recombinant thromboplastin reagent (Dade-Behring) (i.e., Quick method) *(8)* with an ISI of 1.0. The test strips are calibrated against the WHO IRP rTF/95 (2). The meter detects clot end points by measuring changes in the electrical impedance of a fresh capillary blood sample. Each test strip incorporates 2 control channels that automatically test a low and high control each time a sample is tested. If the control results are out of range a warning will appear on the display, and no INR results will be reported *(14)*. Consequently, no additional liquid internal QC was performed during the study. The analytical imprecision based on duplicate measurements on patient samples was 7.9%.

MODEL FOR INSTRUMENT COMPARISON AND STATISTICAL ANALYSIS

Statistical analysis and other calculations were performed with Microsoft Office Excel 2003 and SPSS 15.00 for Windows.

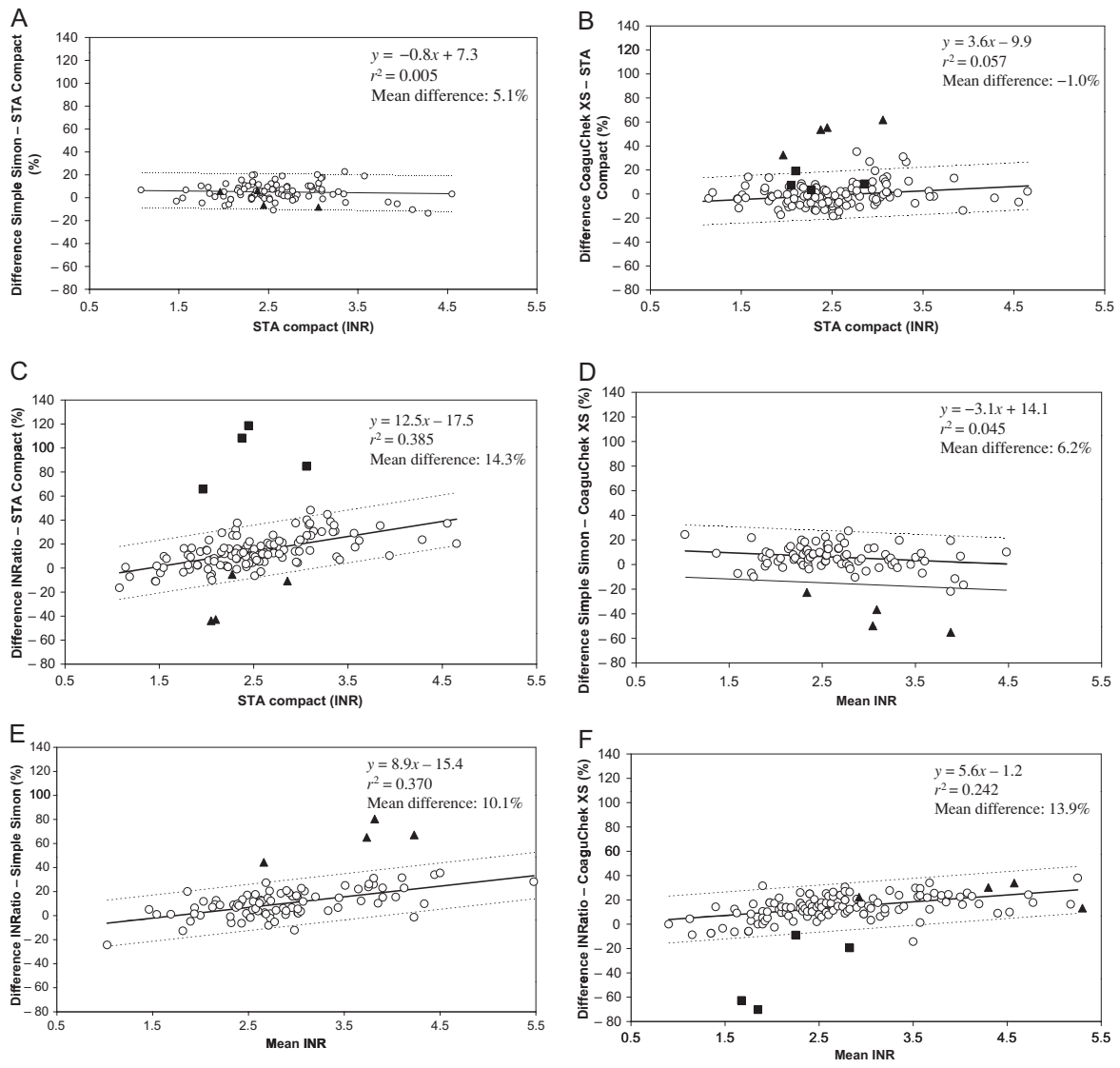

Fig. 1. Difference plots before correction for systematic differences between the instruments.

Comparison of Simple Simon and STA Compact (A), CoaguChek XS and STA Compact (B), and INRatio and STA Compact (C). Differences in percentages are plotted as a function of data from the STA Compact. Comparison of Simple Simon and CoaguChek XS (D), INRatio and Simple Simon (E), and INRatio and CoaguChek XS (F). Differences in percentages are plotted as a function of the mean of the results from the 2 instruments. The number of patients is given in Table 2.

Regression lines (—), equation for the regression lines ($y = ax + b$), coefficient of determination (r^2), mean difference, and 95% tolerance intervals (- - -) are indicated (calculated after exclusion of patients 12 and 24). (B, C, F), Results for patient 12 are shown (■); (A–F), results from patient 24 are shown (▲).

DIFFERENCE PLOTS

All 4 INR instruments were compared in pairs. Difference plots (in percent) showing the percentage difference between results from the POC instrument minus the hospital instrument (STA Compact) (Fig. 1, A–C) were drawn with percentage difference as a function of the hospital method, because this method was chosen as the designated comparison method and the analytical imprecision was small compared to the POC instrument (see above). No regression to the mean was found. The corresponding difference plots for the remaining 3 comparisons (Fig. 1, D–F) have the mean of

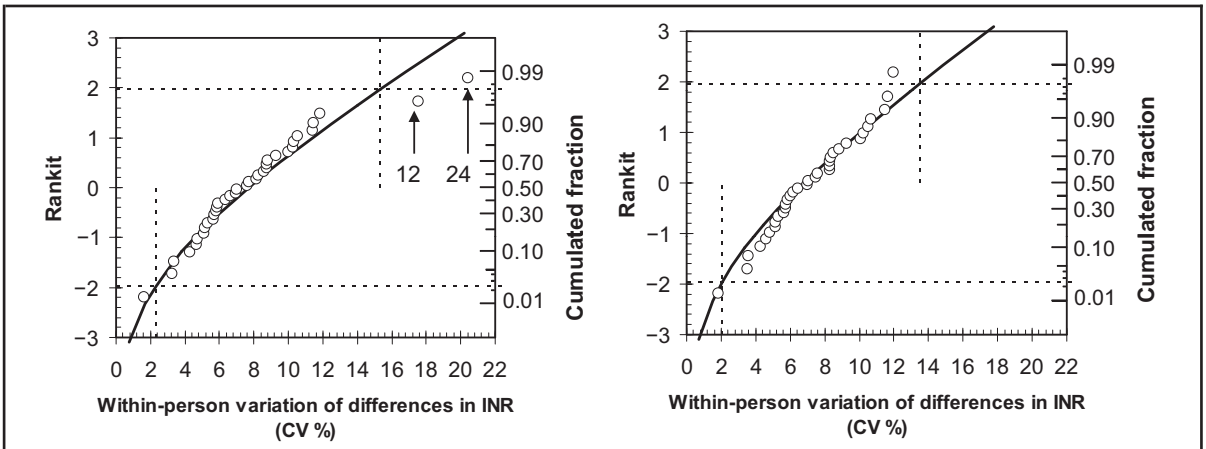

Fig. 2. Variance homogeneity plots.

Variance homogeneity plots for $CV_{within\text{-}person}$ variation after correction for systematic differences between the instruments. The plots show the accumulated ranked fractions as a function of $CV_{within\text{-}person}$ variation of differences in INR between INRatio and STA Compact before (left panel) and after (right panel) exclusion of patients 12 and 24. The $CV_{within\text{-}person}$ values for patients 12 and 24 are indicated. The solid curves indicate the expected distribution of homogeneous measured CV values ($CV_{pooled\ within\text{-}person} * \sqrt{chi^2/df}$); 95% CIs (- - -) are indicated.

results from both instruments as the abscissa *(30)* to avoid possible effects of regression to the mean.

CORRECTION FOR SYSTEMATIC DIFFERENCES

In the difference plots at all 4 time points, data for patient no. 12 deviated when the results from the INRatio were compared with those from the STA Compact and CoaguChek XS (this patient's samples were not analyzed on the Simple Simon) (Fig. 1, C and F). Data for patient no. 24 deviated when results from the CoaguChek XS were compared with those from the STA Compact (Fig. 1B), when results from the INRatio were compared with those from the STA Compact and Simple Simon (Fig. 1, C and E), and when results from the Simple Simon were compared with those from the CoaguChek XS (Fig. 1D). Percentage differences between the means of duplicate measurements of INR on the 2 compared instruments were calculated for each sampling time for each patient after exclusion of patients 12 and 24. The formula for each linear regression line (Fig. 1) was used to recalculate the results in percent, and these were then called "corrected results" because they were then corrected for systematic differences.

HOMOGENEITY

A presupposition for calculating the within-subject variation for the differences between the instruments is the assumption of variance homogeneity. With the use of the corrected results, SDs of the 4 differences (1 for each sampling time) for each of the 6 comparisons were then calculated. Because these differences are percentages of measured INR, the calculated SDs are equivalent to CVs. Note that the $CV_{within\text{-}person}$ here includes the analytical variance. The ranked variances should follow a χ^2 distribution, which was tested by Bartlett's test *(31)*. Statistical significance was set to <0.05, which was also illustrated in rankit plots with $CV_{within\text{-}person}$ as the abscissa and ranked cumulated fractions as the ordinate. The ranked cumulated fractions were compared to the curve for $CV_{pooled\text{-}within\text{-}person}\ \% * \sqrt{chi^2/df}$, for which the degrees of freedom (df) $= 4 - 1$ (Fig. 2). Data for patients 12 and 24 deviated with respect to within-person variation of the differences in the rankit plots. After we excluded these patients all comparisons showed variance homogeneity. This is illustrated in Fig. 2 for INRatio vs STA Compact before (Fig. 2 left panel) and after exclusion of patients 12 and 24 (Fig. 2 right panel).

BETWEEN- AND WITHIN-SUBJECT VARIATION AND ANALYTICAL IMPRECISION

The corrected results (percent differences in duplicate) could then be used for calculation of total, between-subject, within-subject, and analytical variation for the differences according to an unbalanced 2-fold nested random ANOVA model *(32)*. For details see the Appendix in the Data Supplement that accompanies the online version of this article at http://www.clinchem.org/content/vol56/issue10. CIs were calculated according to Burdick and Graybill *(32)*.

Table 2. Mean systematic difference before correction and total variation, between- and within-subject variation, and analytical imprecision of the differences between instruments after correction for systematic differences.[a]

Instruments compared	n[b]	Mean systematic difference, % (95% CI)	Total CV, % (95% CI)	Between-subject CV, % (95% CI)	Within-subject CV, % (95% CI)	Analytical imprecision CV, % (95% CI)
Simple Simon and STA Compact	23	5.1 (3.8 to 6.5)	7.9 (7.1 to 9.4)	3.3 (0.9 to 5.7)	5.9 (4.8 to 7.3)	4.1 (3.6 to 4.8)
CoaguChek XS and STA Compact	34	−1.0 (−2.4 to 0.4)	9.6 (8.8 to 12.3)	7.4 (6.1 to 10.6)	4.8 (4.1 to 6.0)	3.8 (3.4 to 4.3)
INRatio and STA Compact	34	14.3 (12.3 to 16.2)	11.8 (10.7 to 13.7)	6.7 (4.9 to 9.4)	4.6 (1.8 to 6.5)	8.6 (7.6 to 9.7)
Simple Simon and CoaguChek XS	23	6.2 (4.4 to 7.9)	10.0 (9.1 to 13.7)	7.3 (5.9 to 11.8)	4.9 (3.7 to 6.4)	4.9 (4.3 to 5.8)
INRatio and Simple Simon	23	10.1 (8.1 to 12.1)	9.8 (8.8 to 12.2)	5.8 (4.0 to 9.1)	4.4 (2.5 to 6.3)	6.6 (5.7 to 7.7)
INRatio and CoaguChek XS	34	13.9 (12.3 to 15.4)	10.1 (9.5 to 12.4)	5.5 (4.4 to 8.8)	5.5 (4.2 to 7.2)	6.4 (5.7 to 7.3)

[a] For details about the ANOVA model see the Appendix in the online Data Supplement. For calculations of CIs see Burdick and Graybill *(32)*.
[b] Number of patients compared (4 sampling times per patient). Patients 12 and 24 were excluded.

Between-run analytical variation will be a part of the calculated within- and between-subject variation. Between-run variation can be estimated by comparing total analytical imprecision (given for STA Compact, Simple Simon, and CoaguChek XS above) with within-run analytical imprecision (see above) for each instrument. It can bee seen that there are only minor differences. Thus, we assumed that the between-run variation had only a minor impact on the within- and between-subject variation.

OTHER STATISTICS

Outliers were excluded according to the method of Burnett *(33)* before correction for the systematic differences. For duplicate measurements on the 3 POC instruments and the hospital instrument there were no outliers. For the differences in INR when we compared 2 instruments, the following samples were excluded: first visit and first duplicate for the second visit for patient 14 when we compared the CoaguChek XS and STA Compact, and both duplicates for the first visit for patient 14 when we compared the INRatio and Simple Simon.

Results

DIFFERENCE PLOTS

Thirty-six patients completed the study with 4 sampling times on the hospital instrument STA Compact and the 3 POC instruments (Simple Simon [patients 13–36], CoaguChek XS, and INRatio). All 4 INR instruments were compared in pairs, and the mean differences in results between the instruments varied over time (Fig. 1). The mean differences in INR varied between 1.0% and 14.3% (Table 2 and Fig. 1). In addition, data from patients 12 and 24, who were excluded from the calculations because they caused variance inhomogeneity (Fig. 2), deviated considerably from the data from other patients in some of the comparisons (Fig. 1).

CORRECTION FOR SYSTEMATIC DIFFERENCES

Both intercepts and slopes for the regression lines used for correction for systematic differences were dependent on the instruments compared (Fig. 1). Thus, the degrees of correction varied, but after correction, the total variation CVs for the pairwise differences between instruments were about 10% except for the CV between the STA compact and Simple Simon, which was 7.9%.

VARIANCE HOMOGENEITY

All combinations of instruments showed variance homogeneity for the differences between instruments after exclusion of patients 12 and 24 (illustrated for INRatio vs STA Compact in Fig. 2).

ESTIMATED BETWEEN- AND WITHIN-SUBJECT VARIATIONS AND ANALYTICAL IMPRECISION

After correction for systematic differences and exclusion of outliers, the between- and within-subject variation and analytical imprecision subcomponents of the differences were estimated for all comparisons (Table 2). The mean between-subject variation subcomponent of the difference depended on which instruments were compared, and when this result was expressed as the CV it varied between 3.3% (Simple Simon vs STA Compact) and 7.4% (CoaguChek XS vs STA Compact). However, the mean within-subject variation subcomponent of the difference was comparable for all

6 comparisons with a CV of about 5% (Table 2). Exclusion of the 5 patients who had been on anticoagulant treatment for less than 3 months had no demonstrable effect on the within-subject variation (overlapping CIs) (see online Supplemental Table 1). The analytical imprecision subcomponent of the differences expressed as CV varied between 3.8% (CoaguChek XS vs STA Compact) and 8.6% (INRatio vs STA Compact).

Discussion

Discrepancies in INR results between POC instruments and laboratory instruments and between different POC instruments are well known and have been described *(13–15, 34)*. In the present study, in which 4 different INR instruments were compared at 4 different time points in the same patients, we found that the discrepancies between instruments varied over time, and we developed a model to split the variation into its subcomponents. In this model the variation of the differences between instruments was divided into calibration effects and between- and within-subject variation as well as analytical imprecision. To our knowledge no study has been performed previously to address the relative contribution of these components to differences between instruments.

SYSTEMATIC DIFFERENCES

Difference plots revealed discrepancies between the 4 INR instruments in pairwise comparisons (Fig. 1), and the first step in the model was to calculate the variations caused by differences in calibration (Table 2). The magnitude of the calibration differences depended on the instruments compared. All instruments used in this study were calibrated against an IRP or methods traceable to an IRP in accordance with WHO guidelines *(2)*. However, it has been shown that the calibration procedure does not necessarily harmonize with the INR results *(3, 5)*. Differences in calibration were taken into account by correction of all results by use of linear regression. When significant systematic differences exist, instruments should not be used interchangeably for management of oral anticoagulant therapy. Correction of the results for systematic differences may improve the consistency of results obtained when instruments are used interchangeably but, as shown below, comparison of results from different instruments is still expected to reveal a considerable amount of random variation. The next step in the model was to examine the variance homogeneity and to divide the remaining variation into between-subject, within-subject, and analytical components.

HOMOGENEITY

Difference plots in percent (Fig. 1) and variance homogeneity plots for the differences between the instruments (illustrated for INRatio vs STA Compact in Fig. 2) revealed that the patients belonged to a homogenous population, after data from 2 patients were excluded (patient 12 and 24) (Fig. 2, right panel). For patient 12 we found no explanation for the deviations. Patient 24 had systemic lupus erythematosus, and it has been reported that in patients with lupus anticoagulants a discrepancy in INR may occur with some thromboplastins *(35)* and methods (Quick vs Owren) *(36)*.

BETWEEN- AND WITHIN-SUBJECT VARIATION AND ANALYTICAL IMPRECISION

The between-subject variation of the differences depended on which instruments were compared (Table 2). If all patients had the same constant variation, this finding would have been a part of the systematic deviation between the instruments. INR is influenced by multiple factors in the blood sample from the patients *(4, 6, 7)*, including the level of different coagulation factors and their interaction with thromboplastin reagents that have different composition and sensitivity *(2, 6)*. These factors can cause patient-specific effects that cannot be eliminated when different INR systems are compared *(4, 7, 13)*. The least variation was seen when the 2 Owren-based methods, STA Compact and Simple Simon, were compared (3.3%), and the low variation can be explained by a high dilution of plasma in both instruments, the addition of factor V and fibrinogen to reagents as well as the use of a similar rabbit brain thromboplastin. For the other instrument combinations the between-subject variation of the differences was higher and ranged between 5.5% and 7.4% (Table 2). CoaguChek XS and INRatio measure thrombin and fibrin end products, respectively, and this method difference may also cause higher between-subject variation for the differences between these 2 instruments. CoaguChek XS and INRatio use human recombinant thromboplastins, which can vary in composition *(4)*. Thromboplastins used by the different instruments in this study had nearly the same ISI values [close to 1 as recommended *(37)*]. However, it has been shown that the discrepancy in INR is not necessarily related to the ISI value *(3, 4, 38)*. After correction for systematic differences, clinicians could expect larger variation in the differences between a POC instrument and a laboratory instrument between different subjects than within the same subject. To minimize this variation, POC instruments having the lowest between-subject variation for the differences between a POC instrument and a hospital instrument should be used.

The within-subject variation of the differences was similar and was about 5% for all comparisons (Table 2), which indicates that the variation within each sub-

ject was independent of the instruments compared. Thus, for each patient the difference in INR results between 2 instruments varied similarly over time. In agreement with this theory are data reported by Tripodi et al. *(21)*, who found that the differences between a POC system and a laboratory method varied over time for 4 different patients on anticoagulant treatment. An explanation for this within-subject variation could be that there are patient-specific effects that vary over time and have similar effects across different measurement procedures, in contrast to the between-subject patient-specific effects. Therefore, even without analytical variation, the differences in results from the same patient between 2 analyzers will vary up to ±10%. Because it is imperative to drive out as much analytical and interinstrument variation as possible to reduce the likelihood of unnecessary dosage adjustments in oral anticoagulant treatment, the same instrument should be used for each individual patient.

Discrepancies between the 4 INR instruments might theoretically also be caused by various preanalytical factors associated with venous (STA Compact and Simple Simon) and capillary (CoaguChek XS and INRatio) sampling, which could affect within-subject variation. However, because all steps have been performed according to established procedures and guidelines by the same trained personnel and because this variation was in the same magnitude for all comparisons (Table 2), we assume that the impact of these effects was negligible.

Analytical imprecision of the differences depended on the instruments compared. Therefore, if either instrument has significant imprecision, caution should be exercised when the instruments are used interchangeably.

Conclusions

We have created a model that subdivides into subcomponents the variation of the differences between 2 instruments used to measure INR and calculated the contribution of each of these components. In addition to systematic differences between instruments, there are random between- and within-patient specific effects as well as analytical imprecision that influence the differences in results when 2 instruments are compared. To minimize this variation in a monitoring situation, each site should use results from only 1 type of instrument for each patient. Additional studies are needed to fully elucidate the between- and within-subject variation associated with the differences observed between instruments.

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References

1. WHO Expert Committee on Biological Standardization. Requirements for thromboplastins and plasmas used to control oral anticoagulant therapy. World Health Organ Tech Rep Ser 1983;687:81–105.
2. WHO Expert Committee on Biological Standardization. Guidelines for thromboplastins and plasma used to control oral anticoagulant therapy. World Health Organ Tech Rep Ser 1999;889:64–93.
3. Horsti J, Uppa H, Vilpo JA. Poor agreement among prothrombin time international normalized ratio methods: comparison of seven commercial reagents. Clin Chem 2005;51:553–60.
4. Jackson CM, Esnouf MP, Lindahl TL. A critical evaluation of the prothrombin time for monitoring oral anticoagulant therapy. Pathophysiol Haemost Thromb 2003;33:43–51.
5. Attermann J. Inaccuracy of international normalized ratio (INR) measurements and suggestions for improved WHO guidelines for calibration of reference preparations; a statistical study. J Thromb Haemost 2003;1:537–44.
6. Testa S, Morstabilini G, Fattorini A, Galli L, Denti N, D'Angelo A. Discrepant sensitivity of thromboplastin reagents to clotting factor levels explored by the prothrombin time in patients on stable oral anticoagulant treatment: impact on the international normalized ratio system. Haematologica 2002;87:1265–73.
7. van den Besselaar AMHP. Precision and accuracy of the international normalized ratio in oral anticoagulant control. Haemostasis 1996;26(suppl 4):248–65.
8. Quick AJ. The prothrombin in hemophilia and obstructive jaundice. J Biol Chem 1935;109:73–84.
9. Owren PA. Thrombotest: a new method for controlling anticoagulant therapy. Lancet 1959;2:754–8.
10. Tripodi A, Chantarangkul V, Mannucci P. Near-patient testing devices to monitor oral anticoagulant therapy. Br J Haematol 2001;113:847–52.
11. Poller L, Keown M, Chauhan N, van den Besselaar AMHP, Tripodi A, Shiach C, et al. European Concerted Action on Anticoagulation. Correction of displayed international normalized ratio on two point-of-care test whole-blood prothrombin time monitors (CoaguChek Mini and TAS PT-NC) by independent international sensitivity index calibration. Br J Haematol 2003;122:944–9.
12. Tripodi A, Arbini AA, Chantarangkul V, Bettega D, Mannucci PM. Are capillary whole blood coagulation monitors suitable for the control of oral anticoagulant treatment by the international normalized ratio? Thromb Haemost 1993;70:921–4.
13. Jonsson M, Hillarp A, Svensson P. Comparison between CoaguChek S- and Owren-type prothrombin time assay for monitoring anticoagulant therapy. Thromb Res 2004;114:83–9.
14. Taborski U, Braun SL, Voller H. Analytical performance of the new coagulation monitoring system INRatio for the determination of INR compared with the coagulation monitor Coaguchek S and an established laboratory method. J Thromb Thrombolysis 2004;18:103–7.

15. Shermock KM, Bragg L, Connor JT, Fink J, Mazzoli G, Kottke-Marchant K. Differences in warfarin dosing decisions based on international normalized ratio measurements with two point-of-care testing devices and a reference laboratory measurement. Pharmacotherapy 2002;22:1397–404.
16. Bereznicki L, Jackson S, Peterson G, Jeffrey E, Marsden K, Jupe D. Accuracy and clinical utility of the CoaguChek XS portable international normalised ratio monitor in a pilot study of warfarin home-monitoring. J Clin Pathol 2006;60:311–4.
17. Plesch W, van den Besselaar AMHP. Validation of the international normalized ratio (INR) in a new point-of-care system designed for home monitoring of oral anticoagulation therapy. Int J Lab Hematol 2009;31:20–5.
18. van den Besselaar AMHP. A comparison of INRs determined with a whole blood prothrombin time device and two international reference preparations for thromboplastin. Thromb Haemost 2000; 84:410–2.
19. Scandinavian evaluation of laboratory equipment for primary health care (SKUP). http://www.skup.nu (Accessed September 2010).
20. Attermann J, Andersen N, Korsgaard H, Maegaard M, Hasenkam J. Patient-specific errors in agreement between international normalized ratios measured by a whole blood coagulometer and by a routine plasma-based method. Haematologica 2004;89:504–5.
21. Tripodi A, Breukink-Engbers W, van den Besselaar AMHP. Oral anticoagulant monitoring by laboratory or near-patient testing: what a clinician should be aware of. Semin Vasc Med 2003; 3:243–54.
22. van Geest-Daalderop JHH, Mulder AB, Boonman-de Winter LJM, Hoekstra MMCL, van den Besselaar AMHP. Preanalytical variables and off-site blood collection: influences on the results of the prothrombin time/international normalized ratio test and implications for monitoring of oral anticoagulant therapy. Clin Chem 2005;51:561–8.
23. Salvagno GL, Lippi G, Montagnana M, Franchini M, Poli G, Guidi GC. Influence of temperature and time before centrifugation of specimens for routine coagulation testing. Int J Lab Hematol 2009; 31:462–7.
24. NCCLS. Collection, transport and processing of blood specimens for coagulation testing and general performance of coagulation assays; approved guideline. NCCLS document H21–A4. Wayne (PA): NCCLS; 2003.
25. Lindahl TL, Egberg N, Hillarp A, Odegaard OR, Edlund B, Svensson J, et al. INR calibration of Owren-type prothrombin time based on the relationship between PT% and INR utilizing normal plasma samples. Thromb Haemost 2004;91: 1223–31.
26. Scandinavian evaluation of laboratory equipment for primary health care (SKUP). Simple Simon® PT: a system for measurement of prothrombin time [P—PT (INR)] manufactured by Zafena AB, Sweden: report from an evaluation in a hospital laboratory organised by SKUP. http://www.skup.nu (Accessed September 2010). SKUP/2007/57*; Simple Simon PT.
27. Zafena. Simple Simon® PT product documentation. http://www.zafena.se/English/SimpleSimon (Accessed February 2010).
28. Leichsenring I, Plesch W, Unkrig V, Kitchen S, Kitchen D, Maclean R, et al. Multicentre ISI assignment and calibration of the INR measuring range of a new point-of-care system designed for home monitoring of oral anticoagulation therapy. Thromb Haemost 2007;97:856–61.
29. Brandt JT, Barna LK, Triplett DA. Laboratory identification of lupus anticoagulants: results of the Second International Workshop for Identification of Lupus Anticoagulants. On behalf of the Subcommittee on Lupus Anticoagulants/Antiphospholipid Antibodies of the ISTH. Thromb Haemost 1995;74:1597–603.
30. Bland JM, Altman DG. Statistical methods for assessing agreement between two methods of clinical measurement. Lancet 1986;1:307–10.
31. Aarsand AK, Petersen PH, Sandberg S. Estimation and application of biological variation of urinary delta-aminolevulinic acid and porphobilinogen in healthy individuals and in patients with acute intermittent porphyria. Clin Chem 2006;52: 650–6.
32. Burdick RK, Graybill FA. Confidence intervals on variance components. New York: Marcel Dekker; 1992. Statistics: textbooks and monographs, vol 127.
33. Burnett RW. Accurate estimation of standard deviations for quantitative methods used in clinical chemistry. Clin Chem 1975;21:1935–8.
34. Gosselin R, Owings JT, White RH, Hutchinson R, Branch J, Mahackian K, et al. A comparison of point-of-care instruments designed for monitoring oral anticoagulation with standard laboratory methods. Thromb Haemost 2000;83:698–703.
35. Moll S, Ortel TL. Monitoring warfarin therapy in patients with lupus anticoagulants. Ann Intern Med 1997;127:177–85.
36. Della Valle P, Crippa L, Garlando A, Pattarini E, Safa O, Vigano D'Angelo S, et al. Interference of lupus anticoagulants in prothrombin time assays: implications for selection of adequate methods to optimize the management of thrombosis in the antiphospholipid-antibody syndrome. Haematologica 1999;84:1065–74.
37. Fairweather RB, Ansell J, van den Besselaar AMHP, Brandt JT, Bussey H, Poller L, et al. College of American Pathologists Conference XXXI on laboratory monitoring of anticoagulant therapy: laboratory monitoring of oral anticoagulant therapy. Arch Pathol Lab Med 1998;122: 768–81.
38. Lind SE, Pearce LA, Feinberg WM, Bovill EG. Clinically significant differences in the International Normalized Ratio measured with reagents of different sensitivities. SPAF Investigators. Stroke prevention in atrial fibrillation. Blood Coagul Fibrinolysis 1999;10:215–27.