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# Studies on the Variation in Clinical Laboratory Data and Safety Evaluation of Pharmaceuticals

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The safety of pharmaceuticals has become increasingly important not only in daily medical treatment but also in clinical trials. Although clinical laboratory data are more objective than clinical symptoms, the determination as to whether they indicate abnormal variations depends largely upon the clinical judgment of physicians. The process of determination has not been sufficiently objectified. The present study investigated the indices of criteria for variations in clinical laboratory data obtained in clinical trials. Then, detection rates of abnormal variations were compared between our determination method that employs the reference change value (RCV) expressing the width of biological variation for each test component and conventional determination methods. The study also demonstrated that by combining standard values and the RCV for determination, abnormal variations were found at a rate greater than 50%. The method we propose was applied to the safety evaluation of pharmaceuticals. In clinical trials on the antiviral drug ribavirin administered alone, components of laboratory tests were selected that should be noted in studies on its effects. Expect for decreases in red blood cell counts and hemoglobin values, which are closely associated with anemic symptoms and well known to hepatologists, the increasing trend in platelet counts and decreasing trend in albumin were found to be laboratory test components that should be paid attention to, even though they may not be obvious.

**Key words**——clinical laboratory data; reference change value; safety evaluation; biological variation; laboratory test accuracy

## INTRODUCTION

The efficacy and safety of pharmaceuticals are evaluated based on the observation of symptoms and various clinical laboratory data in clinical trials. Safety is evaluated not only clinical trials but also in post-marketing surveillance (PMS). Blood and urine testing is widely used for basic safety evaluation, as it can be performed relatively easily regardless of the size of the medical facility. Test results can be expressed in objective numerical values, while the occurrence of symptoms cannot in most cases. If criteria for variations in laboratory data are established, abnormal clinical laboratory data can be supplemental and objective data that physicians can use to determine clinical importance. However, it is difficult to evaluate variations in clinical laboratory data uniformly, because physicians currently judge them differently. If the differences are resolved and Clinical Research Coordinator, pharmacists and medical technologists can make judgments independent of physicians, variations can be noted quickly and accurately without being overlooked during every aspect of clinical trials and daily medical treatment.

Simple evaluation methods are widely used in which deviations from standard values and variations that are markedly outside the standard values are used as indices of abnormal variations. In the cancer treatment, progression of grades in the Common Terminology Criteria for Adverse Events (CTCAE), Version 3[1] are used as the criteria for abnormal variations. The ''criteria for abnormal clinical laboratory data''[2] of the Japan Society of Chemotherapy are currently used as the most objective index of clinical laboratory data. However, the determination method generally used is a subjective method with which physicians judge data to be clinically significant variations. In PMS and daily medical treatment, the criteria for abnormal variations are rarely stated. The situation does not differ much from that in clinical trials. In most cases, as lone investigating physicians are responsible for making judgments on variations, the

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discrepancies among them are expected to be even greater than in clinical trials. The discrepancies in judgment can be adjusted when a third-party entity in clinical trials, such as the Efficacy and Safety Evaluation Committee, or Independent Data-Monitoring Committee[3] exists. Without a third party, judgment is inevitably dependent on the sole physician. Thus, it is necessary to establish criteria for the objective judgment of abnormal variations.

Our previous study reported biological variations in typical clinical laboratory data obtained from patients participating in PMS conducted at our institution. Biological variations among our patients correlated closely with the biological coefficient of variation presented by Ricos et al.[4] Their data[5] obtained in healthy European volunteers correlated closely with our data. Therefore, it was decided that their data could be used as data for our patients as biological coefficients of variation in clinical laboratory data. The reference change value (RCV) obtained using the biological coefficients of variation is used as the mean values in clinical trials. However, the RCV is not generally used in the practical operation of clinical trials.

The present study investigated differences between and points common to the criteria based on the RCV and conventional criteria (including the criteria of the Japan Society of Chemotherapy).

In addition, the present study investigated variation in each component of general hematological and biochemical tests conducted as part of the safety evaluation of pharmaceuticals in 24 patients who received ribavirin alone 4 weeks prior to interferon (IFN) $\alpha$-2b in a clinical study.[6] By combining the criteria using biological coefficients of variation and conventional criteria, important components of the safety control of pharmaceuticals were identified and test components that should be noted were selected.

## ETHICS

Clinical data used in comparisons of criteria of variations in clinical laboratory data were not obtained in clinical trials but from patient data in daily medical treatment which were extracted based on the prescribed standard. Thus, the data, which corresponded to epidemiological data without factitious intervention, were handled following the ''Ethical Guidelines of Epidemiological Studies''[7] and others[8] and based on the Privacy Information Protection Law. Source data, which may be connected to privacy information, were not used. Statistical data alone were used instead.

For the evaluation of abnormal variations in pharmaceuticals, among clinical laboratory data of patients who participated in clinical trials implemented after approval by the institutional review board and sufficient informed consent were obtained, clinical laboratory data concerning safety alone were used.

## METHODS

**1. Investigation of Criteria for Abnormal Variations in Clinical Laboratory Data Obtained in Clinical Trials** The investigation included 72 protocols of clinical trials and post marketing clinical studies implemented at our institution in the 2-year period from January 2003 to December 2004, which stated expressly the ''handling of abnormal variations in clinical laboratory data.''

**2. Confirmation of the Accuracy of Laboratory Tests** Clinical laboratory tests performed at our institution use an automatic Hemanalyzer Celdyne 400 (Abbott Japan) for the measurement of hemacytes, Hitachi autoanalyzer 7700 (Hitachi) for analysis of serum, and Nesscol (Azwell) for control of various components. Standard deviation is calculated by performing multiple measurements (repeated measurements of the same specimen) when the use of the control specimen began (January 2002) to obtain simultaneous reproducibility data. Thereafter, the control specimen was measured at specified times 5 times on weekdays and 3 times on weekends (measured once) to confirm that no deviation exceeded 3 SD, when the range of 2 SD calculated based on multiple measurements was defined as standard. The reason why 2 SD was defined as the standard was that when the standard value of each component was established, the tolerance range was 2 SD.[9] When data deviated from the standard values, measurements was performed again. When there was still deviation, the measuring apparatus was stopped and the inside cleaned before measurements resumed.

After it was confirmed that successive data were within the range of 2 SD (daily reproducibility), which were obtained in single measurements every January and February from 2002 when the use of the control specimen began to 2005 when the present study was conducted, the coefficient of variation obtained in

multiple measurements was defined as the coefficient of variation at the time of analysis ($CV_A$). To unify the external environmental conditions, January and February, the months when the present study was conducted, were selected. Although the $CV_A$ should be calculated based on multiple measurements at the time of every measurement, it is impossible for business reasons. Accordingly, the coefficient of variation of initial multiple measurements was used as the representative after it had been confirmed in daily accuracy control that numerical values were stable. In the present study, the correlation between daily reproducibility and simultaneous reproducibility was confirmed in 14 measurement components, red blood cell (RBC), hemoglobin (HGB), white blood cell (WBC), platelet count (PLT), aspartate aminotransferase (AST), alanine amiotransferase (ALT), glucose (GLU), creatinine (CRE), $\gamma$-glutamic pyruvic transferase ($\gamma$-GPT), albumin (ALB), alkaline phosphatase (ALP), total bilirubin (T-Bil), uric acid (UA) and amylase (AMY).

**3. Selection of Patients and Determination of Abnormal Variation in Each Test Component** Patients who met the following 4 criteria were selected for the study of the variation in clinical laboratory data:

1) In patients from whom blood samples were collected early in the morning to avoid daily variation.

2) Patients who were available during 3 consecutive months (December 2004 to February 2005) to avoid seasonal variation.

3) Patients who could be confirmed to be hospitalized for 14 to 21 days, based on average hospital stay. Blood samples were collected before and after this period. Blood samples were not collected during the period, as there were large dispersions among patients.

4) Patients on to the same ward (for diseases and blood collection methods to be uniform).

The above conditions were established to unify physical conditions such as the process of measurement and environmental conditions. The drugs used and treatment were not taken into consideration, because the objective of the study was to collect patient data to determine physiological variation, not to determine effects of specific drugs.

The following procedures for the determination of abnormal variations were implemented to investigate differences: judgment method A, judgment method B, and judgment method C (C–1 and C–2).

**Judgment Method A**: Values deviating from standard values were determined to be abnormal variations. When values within standard values change to become out side the standard values and values outside the standard values worsen, the changes are defined as abnormal variations.

**Judgment Method B**: Abnormal variations were determined following the criteria of the Japan Society of Chemotherapy.

1) Variations with relatively high coefficients of variation

- Normal value Rabnormal value: Changes of 120 % or greater relative to the upper limit of normal values are defined as abnormal variations.
- Abnormal value R abnormal value: Changes of 200% or greater relative to the first value are defined as abnormal variations.

2) Variations other than 1)

- Normal value R abnormal value: Changes of 20 % or greater relative to the first value are defined as abnormal variations.
- Abnormal value R abnormal value: Changes of 200% or greater relative to the first value are defined as abnormal variations.

**Judgment Method C**: Abnormal variations were determined by taking into consideration the range of biological variation in each test component.

When values exceeded the RCV indicating the range of biological variations in each test component, they were defined as abnormal variations. The equation below (*) was used to calculate the RCV between 2 points, that is, before and after changes.[10]

$$RCV = 2^{1/2} \times Z \times (CV_A^2 + CV_I^2)^{1/2} \quad \cdots\cdots\cdots\cdots (*)$$

where $CV_A$ is the measured variation in each test component (test accuracy), $CV_I$ is the biological variation in each test component (in Ricos et al.'s data) and Z is the Z-score (standard normal deviates).

Z was either ''1.96 that gives a significant RCV with 95% probability''[10] or ''2.58 that gives a markedly significant RCV with 99% probability.''[10] Determination using the former was called judgment method C–1 and the latter judgment method C–2.

In addition, in judgment method A, which is highly generalized, and judgment method C–1, which has a broad detection range of variation, variation was determined by combining the two ($A \cup C$–1) or using the common parts of the two ($A \cap C$–1).

**4. Safety Evaluation of a Drug (Ribavirin)** Our proposed method was applied to the safety evaluation of ribavirin. The clinical study was conducted in the Department of Digestive Medicine of our Institution[6] among patients who received the antiviral agent. Data from 24 patients who received ribavirin (4-week antecedent administration) were examined for abnormal variations in the 13 components of RBC, HGB, WBC, PLT, AST, ALT, CRE, $\gamma$-GPT, ALB, ALP, T-Bil, UA and AMY. Data were compared using the paired $t$-test ($p<0.05$ was considered to represent a significant difference clinically.) and the judgment methods described above. Components that should be noted in studies of the drug effects on clinical laboratory data concerning the safety of ribavirin were selected and classified.

## RESULTS

**1. Investigation of Determination Criteria for Abnormal Variations in Clinical Laboratory Data Obtained in Clinical Trials** The following 72 protocols were investigated and classified into 3 groups based on criteria 1), 2) and 3) below, and in the note to Fig. 1:

1) Determination criteria (following completely or partially the definition of the Japan Society of Chemotherapy) were clearly established: 6 protocols.

2) Brief criteria (values outside standard values were defined as ''abnormal'', and normal R abnormal and abnormal R worsened abnormal were defined as abnormal variation: 27 protocols (including 5 protocols in which values progressing one rank in the NCI-CTCAE grades were defined as abnormal variations).

3) No objective determination criteria were established: 39 protocols.

The investigation found that criteria common to all 72 protocols were ''determination by physicians'', such as ''abnormal variations that were judged to be clinically significant'', and the final determination was entrusted to individual physicians.

**2. Confirmation of the Accuracy of Laboratory Tests** Table 1 shows data from multiple measurements (measurements repeated 20 times) performed using control specimens. Table 2 shows data from successive measurements performed every January and February from 2002 to 2005. The data were from patient blood samples collected between 06：00 and 08：00. Therefore, data of control specimens were those measured once at 07：00. These two tables demonstrate that all values in Table 2 are within the range of 2 SD shown in Table 1 (multiple measurements). Thus, the coefficient of variation obtained in multiple measurements of control specimens was designated as $CV_A$ used at our institution.

**3. Selection of Patients and Differences in Determination of Abnormal Variation in Each Test Component** During the 2-month period of January and February 2005, 103 patients met 4 criteria described in the Methods. The primary disease was lung cancer. There were 85 males and 18 females with

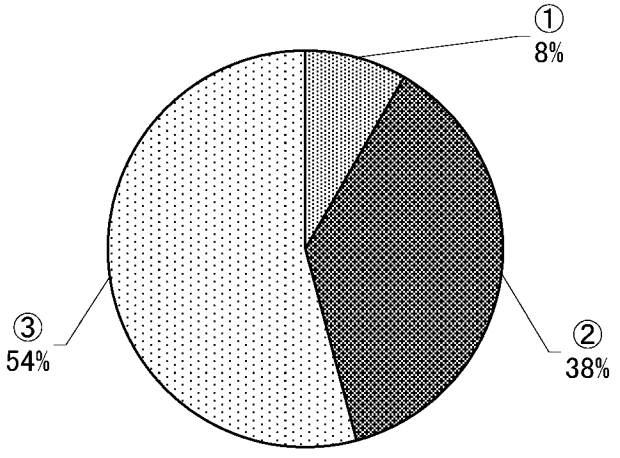


Fig. 1. Establishing of Criteria for Variations

Notes: 1) n=72, 2) ① shows ''objective criteria are established'', 3) ② shows ''brief objective criteria are established'', 4) ③ shows ''no objective criteria are established''.

Table 1. Simultaneous Reproducibility of Test Components

| | Unit | Mean | SD | CV(%) |
|---|---|---|---|---|
| RBC | $\times10^4/\mu l$ | 454 | 5 | 1.1 |
| HGB | g/dL | 13.5 | 0.07 | 0.5 |
| WBC | $/\mu l$ | 7460 | 120 | 1.6 |
| PLT | $\times10^4/\mu l$ | 23.1 | 0.67 | 2.9 |
| AST | IU/l | 27.8 | 0.71 | 2.6 |
| ALT | IU/l | 13.4 | 0.68 | 5.1 |
| GLU | mg/dl | 92.1 | 0.79 | 0.9 |
| CRE | mg/dl | 0.99 | 0.01 | 1.0 |
| $\gamma$-GTP | IU/l | 58.6 | 1.19 | 2.0 |
| ALB | g/dl | 4.10 | 0.02 | 0.5 |
| ALP | IU/l | 141.2 | 1.01 | 0.7 |
| T-Bil | mg/dl | 0.38 | 0.01 | 2.6 |
| UA | mg/dl | 3.65 | 0.05 | 1.4 |
| AMY | IU/l | 239 | 2.05 | 0.9 |

Notes: 1) Multiple measurements 20 times, 2) Use of control of each component, 3) Application from CV (%) to $CV_A$.

Table 2. Daily Reproducibility of Test Components

| | Unit | Year | Mean | SD | CV(%) |
|---|---|---|---|---|---|
| RBC | $\times 10^4/\mu l$ | 2002 | 455 | 6 | 1.32 |
| | | 2003 | 460 | 3 | 0.65 |
| | | 2004 | 449 | 4 | 0.89 |
| | | 2005 | 452 | 7 | 1.55 |
| HGB | g/dl | 2002 | 13.5 | 0.10 | 0.74 |
| | | 2003 | 13.5 | 0.07 | 0.52 |
| | | 2004 | 13.4 | 0.04 | 0.30 |
| | | 2005 | 13.6 | 0.03 | 0.22 |
| WBC | $/\mu l$ | 2002 | 7490 | 158 | 2.11 |
| | | 2003 | 7410 | 152 | 2.05 |
| | | 2004 | 7350 | 110 | 1.50 |
| | | 2005 | 7420 | 112 | 1.51 |
| PLT | $\times 10^4/\mu l$ | 2002 | 23.4 | 0.78 | 3.33 |
| | | 2003 | 23.1 | 0.76 | 3.29 |
| | | 2004 | 23.2 | 0.62 | 2.67 |
| | | 2005 | 22.9 | 0.73 | 3.19 |
| AST | IU/l | 2002 | 27.5 | 0.71 | 2.58 |
| | | 2003 | 27.8 | 0.63 | 2.27 |
| | | 2004 | 27.3 | 0.79 | 2.89 |
| | | 2005 | 27.1 | 0.61 | 2.25 |
| ALT | IU/l | 2002 | 13.2 | 0.66 | 5.00 |
| | | 2003 | 13.4 | 0.78 | 5.82 |
| | | 2004 | 13.8 | 0.85 | 6.16 |
| | | 2005 | 13.6 | 0.97 | 7.13 |
| GLU | mg/dl | 2002 | 92.2 | 0.81 | 0.88 |
| | | 2003 | 92.6 | 0.72 | 0.78 |
| | | 2004 | 91.8 | 0.79 | 0.86 |
| | | 2005 | 92.8 | 0.71 | 0.75 |
| CRE | mg/dl | 2002 | 0.98 | 0.01 | 1.02 |
| | | 2003 | 0.99 | 0.01 | 1.01 |
| | | 2004 | 0.99 | 0.01 | 1.01 |
| | | 2005 | 0.98 | 0.01 | 1.02 |
| $\gamma$-GTP | IU/l | 2002 | 58.8 | 1.21 | 2.06 |
| | | 2003 | 59.1 | 1.08 | 1.83 |
| | | 2004 | 58.2 | 1.05 | 1.80 |
| | | 2005 | 58.3 | 1.17 | 2.01 |
| ALB | g/dl | 2002 | 4.12 | 0.02 | 0.49 |
| | | 2003 | 4.08 | 0.02 | 0.49 |
| | | 2004 | 4.09 | 0.02 | 0.49 |
| | | 2005 | 4.10 | 0.03 | 0.73 |
| ALP | IU/l | 2002 | 141.3 | 1.02 | 0.72 |
| | | 2003 | 141.9 | 1.05 | 0.74 |
| | | 2004 | 141.6 | 0.99 | 0.70 |
| | | 2005 | 141.1 | 1.07 | 0.76 |
| T-Bil | mg/dl | 2002 | 0.38 | 0.01 | 2.63 |
| | | 2003 | 0.38 | 0.01 | 2.63 |
| | | 2004 | 0.39 | 0.01 | 2.56 |
| | | 2005 | 0.38 | 0.01 | 2.63 |
| UA | mg/dl | 2002 | 3.64 | 0.10 | 2.75 |
| | | 2003 | 3.66 | 0.09 | 2.46 |
| | | 2004 | 3.61 | 0.06 | 1.66 |
| | | 2005 | 3.63 | 0.07 | 1.93 |
| AMY | IU/l | 2002 | 241 | 1.98 | 0.82 |
| | | 2003 | 240 | 2.01 | 0.84 |
| | | 2004 | 237 | 2.02 | 0.85 |
| | | 2005 | 238 | 1.96 | 0.82 |

Notes: 1) Investigation period was January and February each year (from 2002 to 2005), 2) Consecutive single measurements, 3) CV, coefficient of variation (unit: %), 4) Mean and SD of RBC, 100 times the nosocomial value (nosocomial nation: $\times 10^6/\mu l$), 5) Mean and SD of WBC, 1000 times the nosocomial value (Nosocomial nation: $\times 10^3/\mu l$).

a mean age of 62.5(10.5 years.

The test components studied in all study subjects were RBC, HGB, WBC, PLT, AST, ALT, GLU and CRE. Table 3 shows variation detection rates for each component using judgment methods A, B, C−1, C−2, A or C−1 ($A \cup C-1$) and A and C−1 ($A \cap C-1$). The mean detection rates of components were nearly the same in (1) A and (3) C−1. However, (6) A and C−1 indicated that between 41% (14.2/34.3) and 45% (14.2/31.6) were common to the two, but the remaining 50% or more were not. As (2) B is a severely restricted version of (1) and (4) C−2 is that of (3), the former are completely included in the latter. In conclusion, (5) A or C−1 was consistently high in all components and the most valid index of an objective judgment.

**4. Safety Evaluation of Ribavirin** 24 patients with chronic hepatitis C (age: 56.0(9.9 years) who received ribavirin for 4 weeks prior to IFN $\alpha$-2b as instructed in the package insert were enrolled. Table 4 shows variations in clinical laboratory data confirmed before and after the 4-week administration of ribavirin alone and compared in the paired $t$-test ($p<0.05$). It was determined that PLT, T-Bil and UA increased significantly and RBC, HGB, AST and ALB decreased significantly. ALT, $\gamma$-GTP and ALP were considered to shown the decreasing tendency and WBC, CRE and AMY showed no significant changes.

Using judgment method A or C−1 ($A \cup C-1$), abnormal variation was determined, and test components were classified based on the determination (Table 5). The $CV_1$ used in Table 5 was obtained from the data of Ricos et al. (RBC : 3.2, HGB : 2.8, WBC : 10.9, PLT : 9.1, AST : 11.9, ALT : 24.3, CRE : 4.3, $\gamma$-GPT : 13.8, ALB : 3.1, ALP : 6.4, T-Bil : 25.6, UA : 8.6, AMY : 9.5) and $CV_A$ was calculated using initial simultaneous reproducibility as explained in Table 2.

## DISCUSSION

The relationship between the variation in clinical laboratory data and standard values can be classified into the following 4 categories: 1) values outside the standard values change to become outside standard values; 2) values outside standard values change to other values outside the standard values (values outside the standard values move either further from or closer to the standard values); 3) values outside the lower limit of the standard value change to outside

Table 3. Variation Detection Rates

| Judgment method | RBC | HGB | WBC | PLT | AST | ALT | GLU | CRE | Average | Variance |
|---|---|---|---|---|---|---|---|---|---|---|
| ①A | 57.3 | 52.1 | 30.1 | 31.1 | 15.5 | 21.4 | 19.8 | 25.2 | 31.6 | 15.2 |
| ②B | 1.0 | 1.0 | 15.6 | 14.6 | 7.8 | 7.8 | 10.9 | 7.8 | 8.3 | 5.4 |
| ③C-1 | 33.0 | 30.2 | 52.4 | 35.0 | 38.8 | 26.2 | 27.4 | 31.1 | 34.3 | 8.3 |
| ④C-2 | 22.3 | 20.2 | 39.8 | 25.2 | 30.1 | 23.3 | 21.5 | 22.3 | 25.6 | 6.5 |
| ⑤A∪C-1 | 70.9 | 65.1 | 62.1 | 54.4 | 42.7 | 37.9 | 33.6 | 46.6 | 51.7 | 13.6 |
| ⑥A∩C-1 | 19.4 | 17.2 | 20.4 | 11.7 | 11.6 | 9.7 | 13.6 | 9.7 | 14.2 | 4.3 |

Notes: 1) $n=103$, 2) Unit is %, 3) ①A shows detection rate of A alone, 4) ②B shows detection rate of B alone, 5) ③C-1 shows detection rate of C-1 alone, 6) ④C-2 shows detection rate of C-2 alone, 7) ⑤A∪C-1 shows detection rate of A or C-1, 8) ⑥A∩C-1 shows detection rate of A and C-1.

Table 4. Various Changes in Clinical Laboratory Data of Pre- and Post-Preceded Ribavirin Medication

| | | | Average | Variance | SD | $p$-value | Significance |
|---|---|---|---|---|---|---|---|
| RBC | $\times 10^4/\mu l$ | Pre | 456.04 | 1865.35 | 43.19 | 0.00001 | ↓↓ |
| | | Post | 393.00 | 3202.96 | 56.59 | | |
| HGB | g/dl | Pre | 14.35 | 1.03 | 1.02 | <0.00001 | ↓↓ |
| | | Post | 12.50 | 1.65 | 1.28 | | |
| WBC | $/\mu l$ | Pre | 4875.00 | 2161087 | 1470 | 0.2126 | — |
| | | Post | 4646.83 | 1483460 | 1218 | | |
| PLT | $\times 10^4/\mu l$ | Pre | 14.89 | 30.32 | 5.50 | 0.0003 | ↑↑ |
| | | Post | 17.41 | 34.51 | 5.87 | | |
| AST | IU/l | Pre | 70.71 | 1067.35 | 32.67 | 0.0015 | ↓↓ |
| | | Post | 49.42 | 388.78 | 19.72 | | |
| ALT | IU/l | Pre | 91.17 | 2325.19 | 48.22 | 0.0052 | ↓ |
| | | Post | 63.54 | 1167.48 | 34.17 | | |
| CRE | mg/dl | Pre | 0.69 | 0.01 | 0.10 | 0.0639 | — |
| | | Post | 0.72 | 0.01 | 0.12 | | |
| $\gamma$-GTP | IU/l | Pre | 81.17 | 2491.79 | 49.92 | 0.0084 | ↓ |
| | | Post | 67.28 | 3486.92 | 59.05 | | |
| ALB | g/dl | Pre | 4.20 | 0.13 | 0.36 | 0.0004 | ↓↓ |
| | | Post | 3.91 | 0.12 | 0.34 | | |
| ALP | IU/l | Pre | 301.58 | 9710.34 | 98.54 | 0.0070 | ↓ |
| | | Post | 253.79 | 6519.82 | 80.75 | | |
| T-Bill | mg/dl | Pre | 0.70 | 0.05 | 0.23 | 0.0002 | ↑↑ |
| | | Post | 1.06 | 0.18 | 0.42 | | |
| UA | mg/dl | Pre | 5.63 | 1.53 | 1.24 | 0.0016 | ↑↑ |
| | | Post | 6.33 | 1.75 | 1.32 | | |
| AMY | IU/l | Pre | 146.06 | 3695.30 | 60.79 | 0.7714 | — |
| | | Post | 144.75 | 3082.60 | 55.52 | | |

Notes: 1) $n=24$, 2) $p$-value was calculated on two sides, 3) ↑↑, Significantly increased with $p \leqq 0.0025$ (consideration of multiplicity), 4) ↓↓, Significantly decreased with $p \leqq 0.0025$ (consideration of multiplicity), 5) ↑, Increasing tendency with $p \leqq 0.05$, 6) ↓, Decreasing tendency with $p \leqq 0.05$, 7) —, No Change.

the upper limit of the standard values or the reverse; and 4) values change within the standard values.

While above 3) is clearly an abnormal variation, the definition of abnormal variation differs in cases 1), 2) and 4) depending upon how the standard is established. In judgment method A, which is generally used, variation under 1), one of 2), or values outside the standard values that move further from the stan-

Table 5. Variation Judgment Classification of Test Components with Ribavirin

| Significant difference | Detection rate of abnormal variation/Test components | |
|---|---|---|
| Present | 50% or higher | RBC GOT ALB HGB |
| | less than 50% | PLT T-Bil UA |
| Absent | 50% or higher | ALP |
| | less than 50% | WBC CRE AMY γ-GTP GPT |

Notes: 1) $n=24$, 2) Detection rate was calculated by the number of abnormal variations $/n(=24)\times 100$, 3) Abnormal variation was judged using method A or C-1.

dard values, and 3) are considered to be abnormal variations. In judgment method B, which uses the numerical criteria of the Japan Society of Chemotherapy, the definition of abnormal variation is even more restricted. In judgment method C, the determination criteria of which is the coefficient of variation of each test component, abnormal variation is determined solely according to the RCV without referring to the standard value used in cases 1), 2), 3) and 4). Originally, confirmation of variation and determination of whether clinical laboratory data are within the standard values are based on the relationship between biological variation and group variation, or that of two with different fields. Therefore, it is inevitable that there are different conclusions. Although it may be convenient to process the two simultaneously, there will be partial distortion. In judgment methods A and B, physicians determine abnormal variation based on standard values, and changes in numerical values are used solely for reference. Conversely, in judgment method C, determination of abnormal variation is based on changes in biological variation and accordingly, standard values are considered to be group variation and not taken into consideration. In actual clinical practice, although it is standard values for group determination which often are preferred, determination for individual patients is also an important element for objective evaluation. When judgment method C was divided into C−1 and C−2, the determination rate in C−1 was found to be similar to that in judgment method A, as shown in Table 3.

We propose an ideal procedure in which judgment method A is used for initial determination, followed by judgment method C−1. By introducing judgment method A + C−1, the detection capacity of abnormal variation can be expected to be approximately 50%. As the detection capacity of the part common to A and C−1 is approximately 15%, there is approximately a 35% increase in the detection capacity when the two methods are used together. The 35% indicates the difference in determination between judgment method A and judgment method C−1. If the study is conducted focusing on the difference, the portion determined by physicians can be reduced dramatically and the portion of objective determination will be increased. The basis of clinical trials is to collect all types of information. To allow physicians to make all medical determinations as has been done in the past places a burden on physicians. By establishing a decision flow in which nearly half of the determination is carried out objectively by medical personnel other than physicians, including pharmacists, medical technologists and nurses (CRCs in clinical trials), and then the final medical determination for individual patients is made by physicians, the objectivity of determination of variation and the efficiency of performance will improve.

The determination criteria for abnormal variation presented by the Japan Society of Chemotherapy have a tendency toward unification (there are only 2 classifications of variations with relatively high coefficients of variation and others). Although they are easy to use, the process of objective determination will be clear if determination is made based on each test component to the extent possible.

One of the advantages of judgment method C is that it takes the accuracy of tests into consideration. If the foundation on which numerical values are produced is shaky, delicate differences and variations will be difficult to detect. Therefore, this is an important consideration.

The effects of pharmaceuticals on clinical laboratory data in safety studies can be evaluated with a significant difference in numerical values and the range of variation. Using the paired t-test to determine significant differences and the above-mentioned determination method (judgment method A+C−1), test components that should be noted become objectively clear. In the case of ribavirin, test components could be classified into 4 types based on the "presence or absence of significant difference" and the "detection rates of variation" (Table 5). Variation in RBC, HGB and AST were well known. However, PLT, T-Bil and UA were found to be components that pharmacists and medical representatives had to call to

physicians' attention. CRE, AMY and WBC could be said to be components that had no effects on safety evaluation.

While the ALT concentration is the highest in the liver, and it can be an important index in hepatitis, the *p* value of the difference before and after ribavirin treatment was greater than initially anticipated and there was no biological variation indicating abnormal variation. It can be predicted that it will be difficult for ALT alone to predict suppression of hepatitis.

## CONCLUSION

Abnormal variation in clinical laboratory data is generally investigated using standard values as the principal medium, as in judgment method A. The same method will continue to be used in the future. The introduction of the RCV, which originally was the criterion for the degree of variation, as the criterion for abnormal variation leads to objectification of determination. Thus, its introduction is important. While it is natural to pay attention to test components with significant differences in changes in values, components without apparent significant differences also should be noted if changes in values are determined to be great. However, it is unnecessary to pay the same degree of attention to components with neither significant differences nor changes as to the other important components. It may be possible for pharmacists and clinical laboratory technicians to process such test components without always consulting with physicians. This will be one factor in the rationalization of clinical trials.

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