Bull. Environ. Contam. Toxicol. (1992) 49:504–508

Environmental Contamination and Toxicology

Stable Blood Cell Counts after One-Week Storage at Room Temperature

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It is quite a regular practice in clinical medicine that blood samples should be subjected to hematological examinations as soon after sampling as possible and that the results should be evaluated at the earliest convenience. Whereas this is also true in occupational and environmental medicine in principle, the practice may be hindered by various study conditions. For example, it would be inevitable to spend more than a day for transportation of the samples, in case the site of blood sampling is very distant from a hematological laboratory. The critical question then is how stable the hematological parameters are when kept under what conditions. Despite remarkable progress in blood conservation technique for transfusion (Grode et al. 1985, Snyder et al. 1985), the questions regarding stability of hematological findings generally remain unsolved, in contrast to serum biochemistry for which effective storage conditions are more or less standardized (Gemba 1985, Kanno 1989). The present examination was initiated to find answers to this practical problem.

MATERIALS AND METHODS

About 28 ml blood samples were drawn from cubital vein of each of 20 healthy volunteers (15 men and 5 women at the ages of 20 to 39 years; Table 1), and immediately divided into 2-ml portions in blood sample containers [Vacutainer^R from Becton Dickinson, Rutherford, NJ 07070, U.S.A.; 3 mg EDTA di-potassium salt (Kanno 1989) is present in each container]. Seven samples out of 14 from each person (or 140 samples in total) were kept at 4°C in a refrigerator, whereas remaining seven (or 140 in total) were left at room temperature (i.e., about 20-25°C). The refrigerated samples were kept at room temperature for 30 min before subjected to the study.

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Table 1. Hematology parameters of the samples at time 0

Parameter	(Unit)	Mean	SD	CV(%)	Min. - Max.
RBC counts	($x10^4$ cells/mm^3)	477	43.8	9.2	405 - 573
Hb conc.	(g/100 ml)	15.1	1.76	11.7	11.6 - 18.3
Ht values	(%)	44.2	4.73	10.7	35.9 - 54.2
WBC counts	(100 cells/mm^3)	64.9	19.84	31.5	39.0 - 12.5
PLT counts	($x10^3$ cells/mm^3)	24.1	5.06	21.0	16.1 - 32.8

n=20

Five hematological parameters of red blood cell (RBC) counts, hemoglobin (Hb) concentrations, hematocrit (Ht) values, white blood cell (WBC) counts and platelet (PLT) counts were studied taking advantage of an automated multiple hematological counter (Sysmex E-3000, Toa Medical Electronics, Kobe, Japan); RBC, WBC and PLT were counted by the electric resistance detection method, whereas Ht was measured after the theory that the pulse height due to voltage changes (produced by the blood cells passing through an aperture of a detection unit of the equipment) is proportional to cell volume (Toa Medical Electronics 1991. Hb was measured by the oxyhemoglogin method at the wavelength of 535 nm. The samples stored either in a refrigerator or at room temperature were subjected to the measurements at 0 (i.e., immediately after blood sampling), 24, 48, 72, 96, 120 and 168 hours after initiation of the storage and the values were expressed in percent taking 0-time value of each subjects as 100. Statistical differences in means were examined by *t*-test.

The mean, SD and the minimum and the maximum of 0-time values for the five parameters are summarized in Table 1. Assuming normal distributions for the five measures, means of each study items are shown as a function of storage duration in Fig. 1, together with standard deviations which are in some cases too small to be presented.

There were no changes in RBC counts and Hb concentrations throughout the study period of 168 hours regardless of storage at room temperature or in a refrigerator. In the case of Ht values, there was a gradual increase as a function of storage duration when kept at room temperature, and the increase was already statistically significant ($p<0.05$) in 24 hours. Although the changes in Ht were smaller when refrigerated, it became significant ($p<0.05$) in 120 hours. No significant changes were observed in WBC counts either when kept refrigerated or not, although

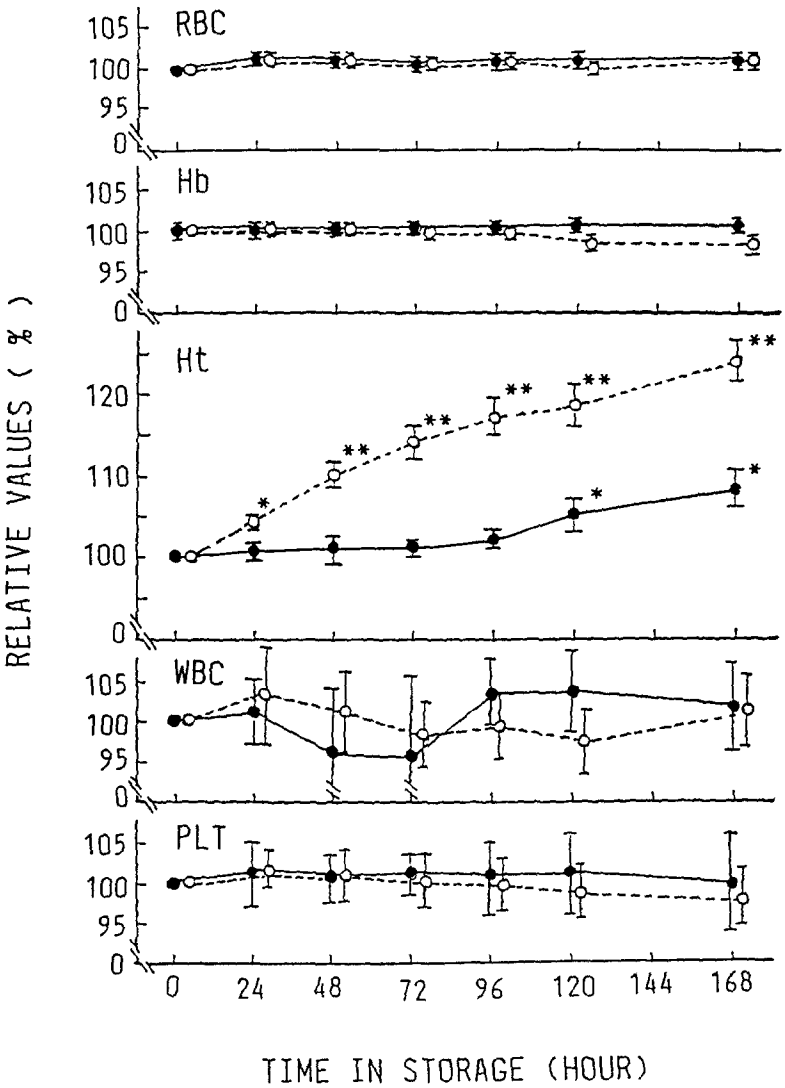

Figure 1. Possible changes in hematological parameters as a function of time in storage. Five hematological parameters of RBC (red blood cell) counts, Hb (hemoglobin) concentrations, Ht (hematocrit) values, WBC (white blood cell) counts and platelet (PLT) counts were studied. Open and solid circles show values when stored at room temperature and at 4°C, respectively. The relative changes were calculated taking individual 0-time values as 100(%). The symbols indicate arithmetic means and arrows standard deviations of 20 determinations. In some instances, the variation was too small to be shown in the figure. The asterisks indicate significant difference (** for $p<0.01$, * for $p<0.05$) from the corresponding 0-time values.

the individual variation was much larger (up to 6.2%) than that for RBC counts (less than 1.5%). PLT counts also stayed almost unchanged during the 168 hour period under 2 conditions of storage, i.e., at room temperature and in a refrigerator. The reason for a transient but wide variation especially at the 24th hour of the experiment remains unknown.

The present study indicates that there are no significant changes in RBC counts, Hb concentrations, WBC counts and PLT counts at least for a week after sampling when kept refrigerated or even at room temperature (Fig. 1). The observation is in a general agreement with the findings by Hamilton and Davidson (1973) on stability of RBC, Hb and WBC for 3 days and by Cohle et al. (1981) on stability of PLT in addition to RBC, Hb and WBC over a 5 day period. Despite rather wide fluctuation in PLT counts during the storage period as expressed by relatively large standard deviations, this may not cause serious problems in evaluation because the coefficient of variation for PLT counts among the healthy population is wide (Table 1).

Regarding the relative instability of Ht values, it is known that RBC needs ATP to keep an original diconcave discoid shape, and the RBC shape will change from discocytes to echinocytes and then to echinospherocytes as a function of ATP consumption during storage (Nishiguchi et al. 1980). Thus, apparent increases in Ht values (Fig. 1) may be attributable at least in part to the theoretical limitation that the same constant is applied in calculating Ht value from pulse height in voltage change regardless of the morphological changes during the storage. In fact, Ht values as determined by the conventional centrifuge method showed less increase than the value by the electronic method when kept at room temperature up to 3 days (Hamaguchi et al. 1984). Nevertheless, the values remain unchanged for 96 hours when kept at 4°C in the present examination.

The present observation implies in practice that the 5 hematological parameters (i.e., RBC counts, Hb concentrations, Ht values, WBC counts and PLT counts) can be taken as reliable when blood samples drawn late in the Friday afternoon are analyzed on Monday morning as far as the samples are kept refrigerated over a week-end. It is further possible to state that RBC counts, Hb concentrations, WBC counts and PLT counts in blood samples brought to a laboratory within a week are also allowed to be considered reliable. Although the present results are obtained with blood containers of one commercial brand, the observation suggests the possibilities that such may also be the cases with other commercial products.

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Received February 21, 1992; accepted April 10, 1992.