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Single-Void Urine Selenium Level Expressed in Terms of Creatinine Content as an Effective and Convenient Indicator of Human Selenium Status

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Selenium was known only for its toxic effects long before it was recognized as an essential nutrient (STADTMAN 1977). The sole biologically active form of Se hitherto found in mammals is a Se-dependent enzyme glutathione peroxidase (abbreviated as GSH-Px) (ROTRUCK et al. 1973) although there are other enzymes to exhibit the GSH-Px activity independent of Se (LAWRENCE & BURK 1976). There are only narrow safety margins among the dietary levels at which Se can cause deficient, physiologic and toxic symptoms in animals. In addition, Se salts are the most toxic of all trace elements on a molar basis (VENUGOPAL & LUCKEY 1978). Therefore, effective and convenient means of assessing Se status are now needed for human health and disease (BURK 1976).

Selenium levels in 24 h urines expressed in ng per mL are used as an indicator of Se status. Selenium concentrations in single urine samples are of little use because they are subject to dietary intake and dilution effects. Twenty-four hour urines, however, are difficult to collect and preserve (THOMSON & ROBINSON 1980).

The author recently reported that Se level expressed in concentration per creatinine (abbreviated as CT) concentration, Se(ng/mg CT), is a better indicator of urinary Se than Se(ng/mL). In addition, single-void urine sample was adequate to estimate urinary Se level as a substitute for 24 h urine sample if Se(ng/mg CT) is employed as a Se level (HOJO 1981). These were concluded from variation in 24 h urine Se levels during 7 months and in single-void urine Se levels for 24 h in any one individual.

The purpose of this study is to show the effectiveness of single-void urine Se(ng/mg CT) as an indicator of human Se status by measuring the relationship of urinary Se(ng/mg CT) with erythrocyte Se and GSH-Px levels which are known as indexes of Se status (THOMSON & ROBINSON 1980).

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EXPERIMENTAL

Samples of single-void urine and venous blood were obtained from twenty healthy male undergraduates from 19 to 27 years old. Urine samples were analysed for Se and creatinine. Five parts of blood were collected in a heparinised syringe , placed in a tube containing 3 parts of dextran(6%) in saline(0.9%) solution, and allowed to sediment at 0° C for 1 h. The supernatant was discarded and the remaining cells were mixed with the same volume of saline solution and centrifuged at 1000 × g for 10 min at 0° C. The residue after removal of the supernatant was hemolysed with 5 parts of water, frozen, thawed and then diluted with 4 parts of water. The resulting solution was analysed for Se, GSH-Px and protein within 12 h after sample collection.

Selenium was determined by the fluorimetric method (WATKINSON 1966), protein by the method of Lowry et al. (1951) and creatinine by Folin-Wu method (BONSNES & TAUSSKY 1945). The Se-dependent GSH-Px activity was assayed with hydrogen peroxide as substrate (LEE et al. 1979) by the method of Hafeman et al. (1974). Organic hydroperoxides were not used because they are substrates for total GSH-Px activity, that is, the sum of Se-dependent and Se-independent GSH-Px activities (LEE et al. 1979). One GSH-Px activity unit was expressed as a decrease in the log([GSH] × 10000) of 0.001 per min after subtraction of the non-enzymatic rate of GSH destruction (HAFEMAN et al. 1974). The obtained data were statistically tested by the Student's t-test and simple linear regression analysis (SNEDECOR & COCHRAN 1967).

RESULTS AND DISCUSSION

Table 1 shows the average, range and coefficient of variation for the single-void urine Se levels expressed by Se(ng/mg CT) and Se(ng/mL), which are plotted in Figs. 1-3. Se(ng/mg CT) had much a narrower range and a lower coefficient of variation than Se(ng/mL). To estimate

TABLE 1. Single-void urine Se levels for 20 individuals.

Item	Se(ng/mg creatinine)	Se(ng/mL)
Mean ± SD	43.4 ± 9.0	104.7 ± 51.7
Range	27 - 64	20 - 233
Coefficient of variation (%)	20.7	49.4

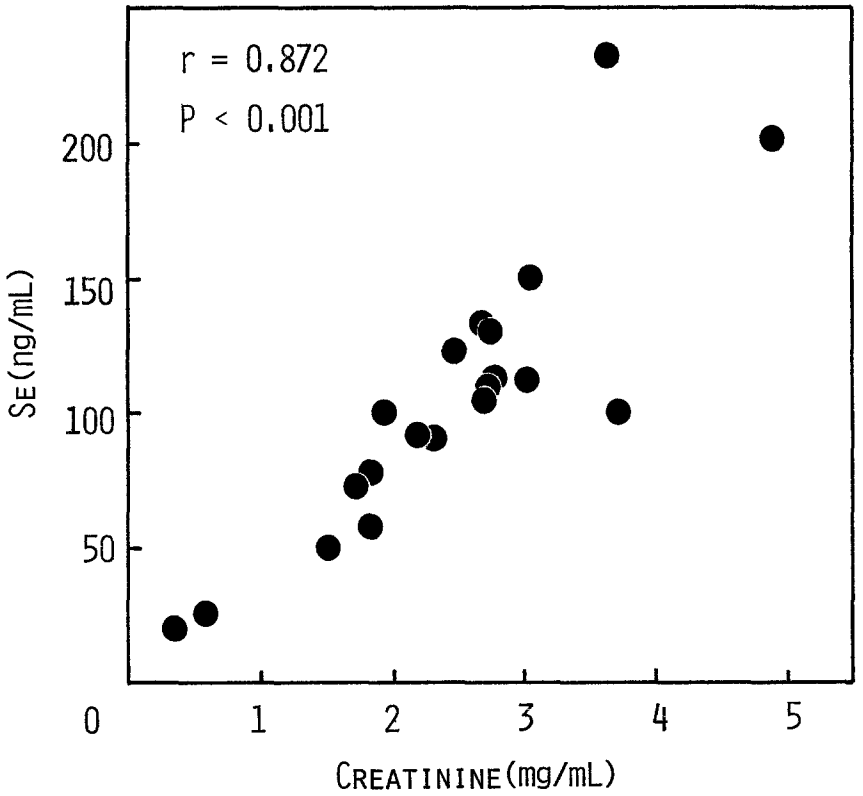

Fig. 1. Se concentration plotted against creatinine concentration in single-void urines from 20 individuals.

urinary excretion of chemical substances without influence of dilution of urine, expression of urinary level of substances in concentration per CT concentration has been used recently (HUNTER et al. 1972, JAFFÉ et al. 1972, VANDERLINDE et al. 1979). The author reported the effectiveness of this expression for Se levels in 24 h urines and single-void urines obtained from any one individual (HOJO 1981). The similar effectiveness of this expression was indicated for Se levels in single-void urines obtained from different persons in the present study.

Fig. 1 gives a highly significant correlation between Se and CT concentrations in single-void urines obtained from different individuals. This is perhaps responsible for lower degree of variation in Se(ng/mg CT) compared with that in Se(ng/mL) as shown previously in my report (HOJO 1981).

Fig. 2 shows that significant correlation coefficient of erythrocyte Se(ng/mg protein) level with single-void urine Se(ng/mg CT) level is larger, though not significantly, than that with urine Se(ng/mL) level.

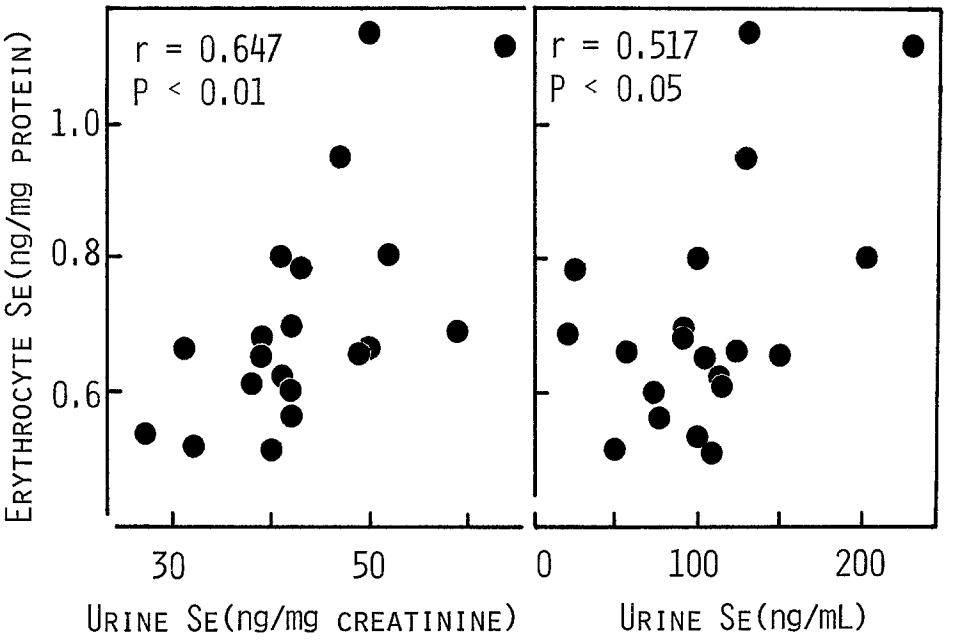

Fig. 2. Erythrocyte Se level plotted against single-void urine Se levels in ng per mg creatinine and in ng per mL for 20 individuals.

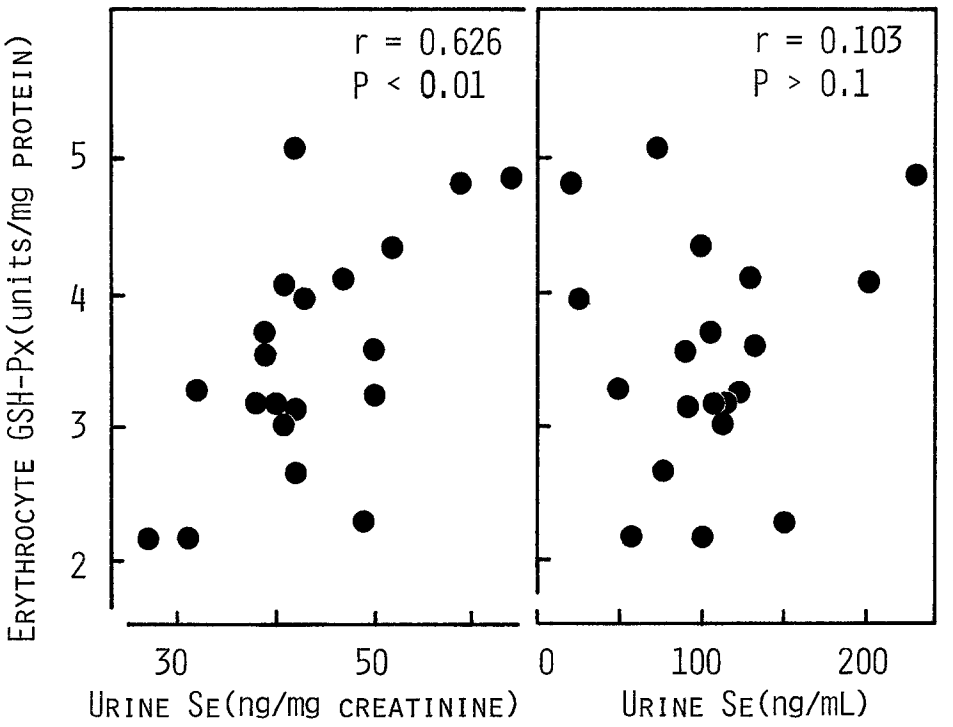

Fig. 3. Erythrocyte GSH-Px activity level plotted against single-void urine Se levels in ng per mg creatinine and in ng per mL for 20 individuals.

Erythrocyte GSH-Px(units/mg protein) level correlated significantly with single-void urine Se(ng/mg CT) level, but not with urine Se(ng/mL) level as shown in Fig. 3.

These results suggest that single-void urine Se(ng/mg CT) is not only a better expression of Se level in single-void urine, but also a more effective indicator of Se status in humans in comparison with Se(ng/mL). It is because both erythrocyte Se and GSH-Px levels are known to be useful indexes of Se status(THOMSON & ROBINSON 1980). Single-void urine sample is more convenient of collection and preservation than other body fluid samples hitherto used to assess Se status, that is, 24 h urine, blood(whole blood, erythrocyte and plasma), hair and nails (BURK 1976). Therefore, single-void urine Se(ng/mg CT) can be used as effective and convenient indicator of human Se status. This can be supported in part by my recent report (HOJO 1981).

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