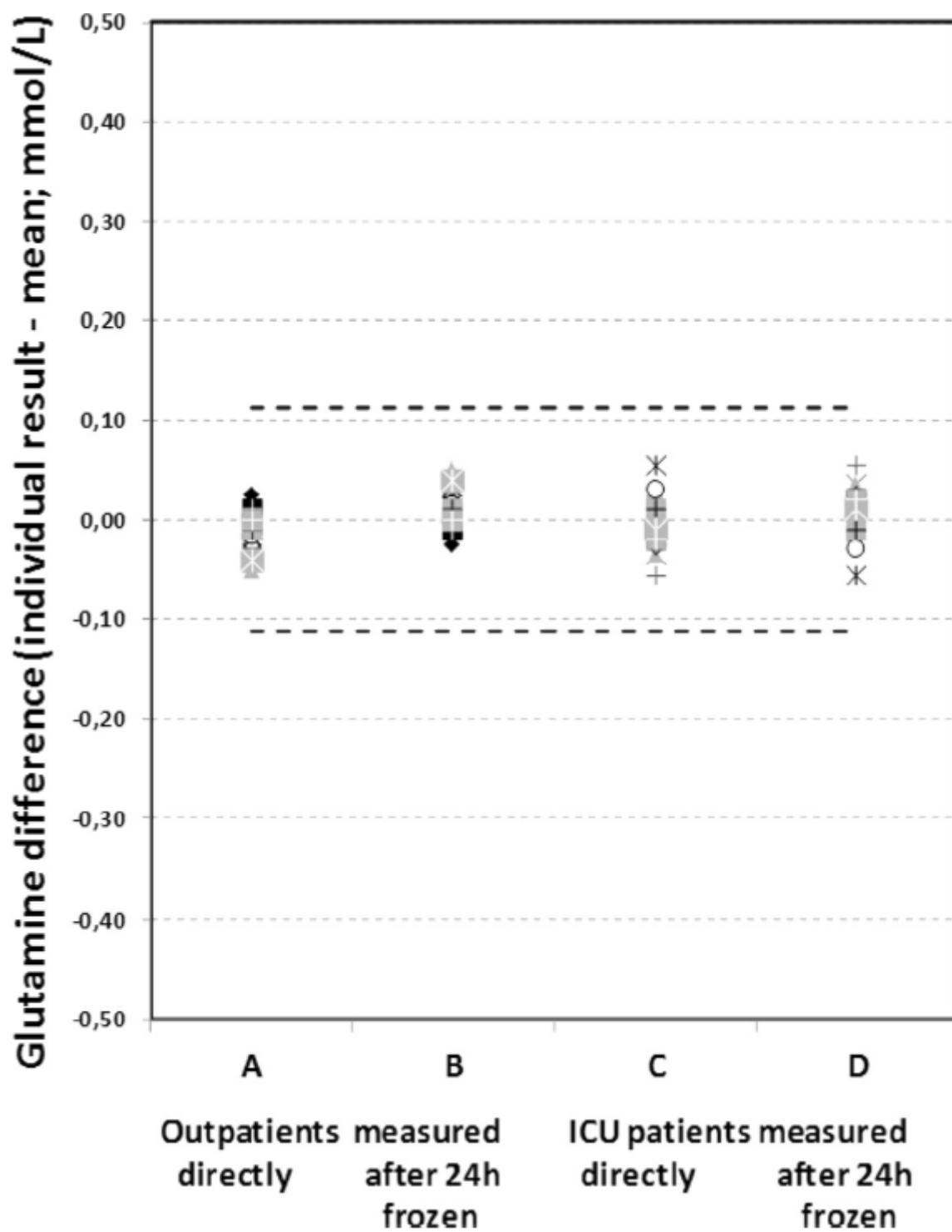


## Additional file 1

### Glutamine measurements

Measurement of glutamine was performed with the Bioprofile 100 plus analyser (Nova Biomedical U.K., Cheshire, UK). This analyser was originally intended for monitoring metabolites and nutrients in cell culture systems, and was used in this study to monitor glutamine in plasma samples (plasma separator tubes; Becton Dickinson B.V., Breda, the Netherlands). For pre-validation of the analyser, we used water-based controls from the supplier of this analyser, and the measurements appeared reproducible. Control measurements were as follows: level 1 mean ( $\pm$  standard deviation) was  $1.06 \pm 0.061$  mmol/l, the CV (coefficient of variation) was 5.7%, and  $n = 143$ ; level 2 mean was  $5.31 \pm 0.400$  mmol/l, the CV was 7.5%, and  $n = 152$ . Using home-made serum/plasma controls, we obtained the following results: heparin pool 1,  $0.558 \pm 0.056$  mmol/l, CV = 10.1%,  $n = 45$ ; serum pool,  $1.038 \pm 0.086$  mmol/l, CV = 8.3%,  $n = 45$ ; and heparin pool 2,  $5.279 \pm 0.259$  mmol/l, CV = 4.9%,  $n = 42$ . We established that plasma glutamine levels were stable when plasma was separated from cells within 3 h after sampling and frozen until analysis (or measured within 3 h after plasma preparation) (Fig. 1). No significant differences in plasma glutamine levels were observed in outpatients (mean difference [direct measurement versus measurement after 24 h at  $-20^{\circ}\text{C}$ ]:  $-0.013$  mmol/L; 95% confidence interval:  $-0.035$  to  $-0.010$ ; two-sided  $P = 0.2675$ ,  $n = 20$ ) and in patients with sepsis (mean difference [direct measurement versus measurement after 24 h at  $-20^{\circ}\text{C}$ ]:  $-0.022$  mmol/L; 95% confidence interval:  $-0.039$  to  $-0.005$ ; two-sided  $P = 0.0137$ ,  $n = 20$ ).

In outpatients ( $n = 51$ ), we obtained a plasma glutamine reference interval, which ranged from 0.22–0.59 mmol/l (Fig. 2).



**Fig. 1** Stability of glutamine in plasma. Plasma was separated from cells within 3 h after collection, frozen immediately thereafter, and measured 24 h later.

Glutamine (mmol/L)

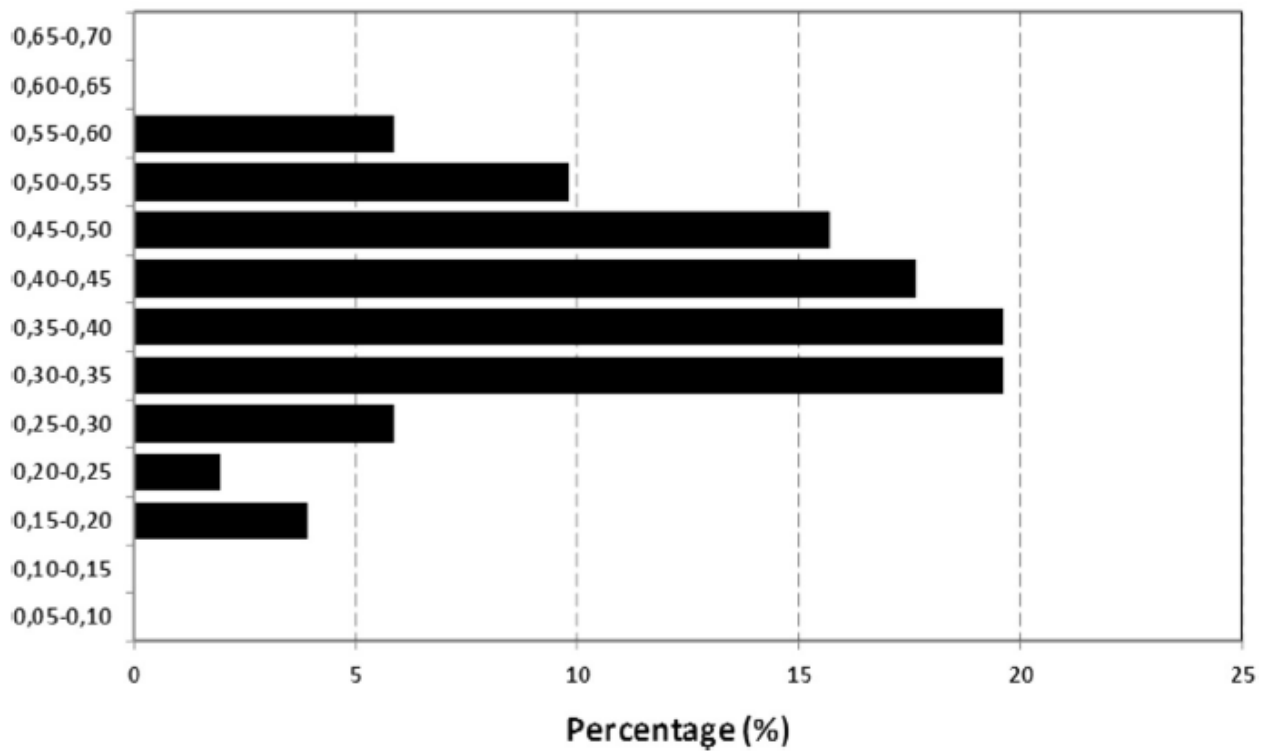


Fig. 2 Histogram of glutamine values found in 51 outpatients