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Diurnal Variations in Serum and Urine Markers of Type I and Type III Collagen Turnover in Children, Ole D. Wolthers,1, 2 Carsten Heuck,2 and 
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New serum and urine markers of type I and type III collagen turnover have recently been introduced in chil
dren. These markers include the formation markers of type I collagen turnover, serum N-terminal (PINP) and C-terminal (PICP) propeptides of type I procollagen, and serum N-terminal propeptide of type III procollagen (PIIINP), as well as the resorption markers serum cross-linked C-terminal telopeptide of type I collagen (ICTP) and urine cross-linked N-telopeptides of type I collagen (Ntx) and deoxypyridinoline (DPD) (1–5). The aim of the 
study was to assess diurnal variations in serum PINP, PICP, ICTP, PIIINP, and urine DPD and Ntx in children.

Two boys and five girls 10.4–14.4 years (mean, 12.2 years) were studied. One boy and three girls were in puberty. Height SD scores varied from −2.5 to 2.2 (mean, 0.6) and weight SD scores from −1.4 to 1.0 (mean, −0.4). The study was approved by the local ethics committee, and informed consent was obtained from all children and their parents.

The first sample was urine collected from 2400 to 0800 on the morning of the day of investigation. Thereafter, 
urine was collected in 4-h intervals until 2400 and in another 8-h interval from 2400 to 0800. Blood samples were taken at 0900 and every 2 h thereafter until 0700 the following morning. The samples were centrifuged at 3000g for 10 min within 1 h after they were collected. After centrifugation, the samples were stored at −80 °C and batch-assayed at the completion of the study.

Each child received breakfast at 0815, lunch at 1300, an ice cream at 1630, and dinner at 1900. Sleep was permitted from 2400 to 0730.

Serum concentrations of PICP, PINP, ICTP, and PIIINP were determined by specific RIAs based on human anti
gen (Orion Diagnostica) (1–3). Intra- and interassay variat
ions were 3.5–3.9% and 4.1–7.2%, respectively. Urine 
DPD was measured by a solid-phase chemiluminescent 
enzyme immunoassay on an automated instrument (Im
mulite Pyrilinks-D; Diagnostica Products Corporation) (4). Urine Ntx was measured by the Osteomark immuno
assay (Ostex) (5). Intra- and interassay variations were 8% 
and 9% for the DPD assay, respectively, and 8% and 12% 
for the Ntx assay, respectively.

Data are described as percentages of the overall day mean ± SE of the mean in the 24-h profile. To evaluate the 
24-h profiles one-way ANOVA for repeated measures 
was performed followed by the Student-Newman-Keuls method for all pairwise multiple comparisons. The 
5% level of significance was used.

PICP and ICTP were relatively low during the day (Fig. 1) with increased PICP concentrations from 0100 to 0500 
(P = 0.006; F = 5.0) and ICTP concentrations from 0100 to 0700 (P = 0.002; F = 6.2). Peak concentrations of PICP 
(mean ± SE) occurred at 0500 [342.0 µg/L (91.6 µg/L)] and trough concentrations at 1100 [286.0 µg/L (46.4 µg/L)]; P = 0.01. Peak concentrations of ICTP were detected at 0700 [12.1 µg/L (1.4 µg/L)] and trough concentrations at 2100 [10.3 µg/L (1.3 µg/L); P = 0.02]. No significant variations in PINP (F = 2.1; P = 0.17) or 
PIIINP (F = 2.1; P = 0.15) were detected.

A significant diurnal variation in urine DPD (F = 15.1; 
P < 0.001) and Ntx (F = 8.2; P < 0.001) was found. Peak 
concentrations of DPD occurred in urine collected at 
0800–1200 [19.7 nmol/mmol (1.6 nmol/mmol)] and trough concentrations in urine collected at 2000–2400 
[12.4 nmol/mmol (1.0 nmol/mmol); P <0.01]. DPD in the 
urine collected at 0800–1200 and two samples collected at 2400–0800 did not vary, whereas DPD concentrations in each of these periods were higher than in the samples collected at 1200–1600, 1600–2000, and 2000–2400 (P
Peak concentrations of Ntx were found during the second 2400–0800 sampling [1323.3 (134.4) nmol/mmol] and trough concentrations during the 2000–2400 time period [812.1 (98.8) nmol/mmol; P < 0.01]. Ntx in both samples collected at 2400–0800 were higher than in the samples collected at 1200–1600, 1600–2000, and 2000–2400 (P < 0.05 for all comparisons).

Considering the marked diurnal variations in PICP and ICTP, it may be difficult to interpret the finding of no rhythmicity in PINP concentrations. There are no available data in adults or in children for comparison of the finding. Perhaps differences in assay sensitivity and metabolism of the propeptides may play a role, necessitating a larger study population than the present to detect possible variations in PINP. Our observation of no diurnal variation in PIIINP concentrations, however, is in accord with a previous report in children (1).

Using an immunoassay, a study of a group of healthy girls, 10–14 years of age, revealed nocturnal peak concentrations of DPD at 0300 and trough concentrations at 1300 (6). The findings are in accord with observations of adults based on immunoassay (7) and HPLC (8) for assessment of total DPD. Furthermore, our results obtained by an immunoassay that specifically measures the free fraction of DPD are consistent with the findings of Robins et al. (4). One study using HPLC, however, found a statistically significant circadian variation only in total, not in free, DPD (9), thus supporting suggestions that in adults, diurnal variations in free DPD may be less marked than in total DPD. Whether this may reflect that the ratio between free and total DPD may vary between children and adults is unclear. Furthermore, the present finding of diurnal variations in Ntx corrected for creatinine is in accord with studies in adults (10). However, when Ntx was not corrected for creatinine in adults, a circadian variation could not be detected. To what extent diurnal variations in DPD and Ntx in children may depend on variations in creatinine needs further study.

References


Diagnostic Accuracies for Celiac Disease of Four Tissue Transglutaminase Autoantibody Tests Using Human Antigen, Silvia Martini, Roberto Pagni, and Carla Sategna-Guidetti (1 Dipartimento di Medicina Interna, Università di Torino, 10126 Torino, Italy; 2 Unità Operativa Autonoma Laboratorio Analisi Chimico-Cliniche, Azienda Ospedaliera San Giovanni Battista, Corso Bramante, 88, 10126 Torino, Italy; * author for correspondence: fax 39-011-676052, e-mail gmengozzi@molinette.piemonte.it)

Celiac disease (CD), a genetic, immunologically mediated small bowel enteropathy that causes malabsorption, is one of the more common disorders in Western countries and is frequently underdiagnosed because of its protean presentations (1). Early diagnosis and treatment with a gluten-free diet may reduce the risk for nutritional (2),