



# Automated in-toilet hydration sensor for urinalysis on participants during a four-day prolonged walking exercise event: Prototype validation study

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## ABSTRACT

Underhydration is a common problem in elderly, which can lead to serious health complications if left untreated. However, currently, there is no sufficient, non-obtrusive method to monitor underhydration status automatically. Therefore, a prototype was developed that automatically measures urine concentration during a toilet visit. This study validates this prototype for its capability to repeatedly and accurately measure the urine of 106 participants participating in a 4-day prolonged walking exercise event. The prototype contains 4 sensors that measure the color, conductivity, pH, and temperature of the urine sample while the participant uses the toilet. In total, the prototype analyzed 514 urine samples. In addition, the urine was automatically collected to measure the gold standard for urine concentration, urine specific gravity (USG), and osmolality. With a linear regression classification model and the USG reference, the data collected with the prototype was classified. The measured reference USG values range between 1.0025 and 1.0345. The prototype measurements strongly correlated to the USG reference, with an R-squared of 0.85 and a mean absolute error of 0.00215. As such, this study reports on a method that allows automated, repeated, and accurate urinalysis during a toilet visit. When used daily, this prototype offers potential for the non-obtrusive and time-effective underhydration estimation by monitoring urine.

## 1. Introduction

Underhydration is a state at which the water homeostatic mechanism has been activated due to low water intake, while the total body water remains constant [1]. It has been associated with serious complications such as metabolic and chronic diseases, and higher mortality rates, whereas by increasing the fluid intake the development of disorders like urinary tract infections and kidney stones can be prevented [2–6]. People particularly at risk for underhydration are elderly as with age the thirst stimulus decreases [7]. In a study from 2020, 95 % of US adults (age 51–70 years) who do not meet the hydration criteria (65 %) are underhydrated [4]. With age also comes a decrease in total body water and lower-functioning kidneys, which also puts elderly at higher risk for dehydration, defined as a condition that occurs when fluid losses exceed fluid intake, leading to a deficiency of water and other fluids necessary for normal bodily functions.

Therefore, the ESPEN (European Society for Clinical Nutrition and Metabolism) guideline considers all persons 65 years or older at risk for low-intake dehydration [8]. This guideline recommends 1.6 L/day for elder women and 2 L/day for elder men in conditions of moderate environmental temperature and moderate physical activity levels. Dehydration should be assessed by directly measuring serum or plasma osmolality, with a suggested threshold for dehydration of 300 mOsm/kg. However, no method for underhydration assessment or monitoring is mentioned. For the monitoring of underhydration, the use of urinary markers has been suggested in the literature [1,3,9–11]. The most widely used urinary markers are urine osmolality, urine specific gravity, and urine color [12–15]. Urine measurements can be considered noninvasive, making them practical for routine monitoring of hydration status. However, current urine testing still requires manual actions, such as collecting and assessing the urine sample, making daily monitoring unfeasible.

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To overcome this, a urine analysis prototype was developed that can automatically measure a urine sample and collect it without the need for any manual action. This study aimed to validate this prototype for its capability to estimate the underhydration status of participants during a four-day walking event, using urine osmolality and urine specific gravity as a reference. In addition, the possibility of using urine temperature to estimate core temperature was explored, as this may provide a non-invasive, easy way of estimating body temperature. The results of our study show that the measurements of our prototype strongly correlate to the USG reference, while measurements were performed automatically during a toilet visit. These findings support that this prototype could be used for non-obtrusive, time-effective, and accurate underhydration monitoring.

## 2. Materials and methods

### 2.1. Study design and population

This study took place during the Nijmegen Four Days Marches of 2023, which is a large walking event, during which participants walk 30, 40, or 50 km at a self-selected pace on 4 consecutive days. Before the walking event, participants volunteered to participate in this study and gave their written informed consent. This study was performed in compliance with the Declaration of Helsinki and was approved by the Medical Ethical Committee of the Radboud University Nijmegen Medical Centre (CMO registration number: 2007/148).

All measurements were taken on a study site located at the start and finish area of the walking event. At this location, two existing toilets were equipped with an in-house-built prototype, which automatically analyzes, and collects the urine of participants (Fig. S1). In the room next to those toilets, reference measurements on the collected urine samples were performed within 3 h after collection. Urine was analyzed at baseline (one or two days before the first walking day (D0)) and directly post-exercise on the 4 walking days (D1-D4). On D0, general demographic data of the participants was collected (Table 1). In addition to the urine measurements, 39 participants ingested a temperature capsule (e-Celsius, Bodycap, Caen, France) on the evening before D1 to measure their core temperature on the first and if possible, the second day of walking, depending on the gut transit time of the participant.

### 2.2. Prototype

The in-house-built prototype consists of a 'hydration sensor' and a 'collection device' for respectively automatic measurements and collection of the urine sample. Fig. 1 shows the prototype. Fig. S2 and S3 show respectively a schematic overview, and a block diagram of the whole setup.

#### 2.2.1. Hydration sensor

The hydration sensor consists of a sampler to guide the urine via gravity to the sensor chamber, which is positioned underneath the sampler. Fig. S4 shows an engineering drawing of the hydration sensor. The probe is placed inside the sensor chamber and is manually

**Table 1**  
Demographic data of the participants included in this study.

	Male	Female
Sex [n (%)]	74 (70)	32 (30)
Age [mean $\pm$ std]	69 $\pm$ 7	66 $\pm$ 6
BMI [median (IQR <sup>1</sup> )]	25.9 (23.9–28.0)	25.7 (22.5–27.0)
Walking distance [n]		
30 km	48	18
40 km	25	14
50 km	1	0

<sup>1</sup> IQR: Interquartile Range.

assembled with two in-house made sensors to measure color and conductivity, and two off-the-shelf sensors to measure pH (Micro pH Probe, AtlasScientific, USA) and temperature (MA100BF103A, Amphenol, USA). The probe is connected to a hardware box that hangs outside the toilet and is battery-powered. The probe is controlled with an in-house made PCB that measures the color, pH, and temperature with a sampling frequency of 1 Hz, and the conductivity with a sampling frequency of 64 Hz.

The color sensor consists of an off-the-shelf LED emitting white light (158,301,240, Wurth Eletronik, Germany) and photodiode (TCS3472, Texas Advanced Optoelectronic Solutions Inc., USA) that are soldered on an in-house-built printed circuit board (PCB). The photodiode contains three filters measuring 'red', 'green', and 'blue' light. In the measurement chamber, opposite the color sensor, a white piece of Teflon of 5 mm thick is positioned at a distance of approximately 1.5 cm to ensure a strong reflection of the light. To prevent urine from damaging the electronics in the probe, the PCB with the LED and photodiode was covered with transparent epoxy (RESION UV Epoxy resin, Polyestershopp, the Netherlands).

The conductivity sensor consists of 2 stainless steel electrodes with a diameter of 2 mm that stick out of the probe by 2.5 mm and are located 5 mm apart. The conductance was controlled with a single-channel integrated front-end chip (Max30001, Analog Device, USA).

#### 2.2.2. Collection device

This study aimed to validate the urinalysis with the hydration sensor against reference measurements. As such, a collection device was added to the hydration sensor to automatically collect the urine that was measured with the hydration sensor. Nevertheless, this collection device is not needed for performing urinalysis with the hydration sensor and will not be needed if validation is not required.

The collection device is connected to the sensor chamber via a tube to transport the urine to the falcon tube underneath the collection device. For this, the collection device contains a peristaltic pump and a valve. As soon as the falcon tube is filled, the remaining urine is pumped back into the toilet bowl. The collection device is connected to a box standing next to the toilet that contains the hardware. Fig. S5 shows the state machine diagram of the collection device. In short, the collection device starts pumping the fluid after it is detected in the sampler (using the capacitance sensor in the sampler). In case the temperature of the fluid (measured by the temperature sensor in the sampler) is above 25 °C, the falcon tube is attached to the collection device (using a pressure switch), and not full (using a capacitance sensor) the valve allows the urine to be pumped to the falcon tube. As soon as the falcon tube is full, the valve transports the remaining urine back to the toilet. When the participant is finished and flushes the toilet, the temperature and capacitance sensor in the sampler detect flush water, which initiates the collection device to start pumping the flush water through the whole system to clean it.

#### 2.2.3. Data acquisition

For each toilet visit, first, the researcher scanned a barcode that consisted of the participant number and the measurement day and attached it to the falcon tube. Second, the falcon tube was placed in the collection device. This automatically turned on the sensors so that the hydration sensor started collecting data. Next, the participant entered the room and used the toilet. Participants were instructed to use the toilet seated and to only throw toilet paper in the toilet bowl and not in the sampler. In addition, participants were asked to flush the toilet afterwards. The hydration sensor automatically measured the urine, and the collection device automatically pumped the urine into the falcon tube. The device cleaned itself afterwards with the flush water. Finally, the researcher detached the falcon tube, filled with urine, from the collection device and brought it to the room next door for the reference measurements. Detaching the falcon tube from the collection device turned off the sensors of the hydration sensor and stopped the data



**Fig. 1.** The in-house-built prototype. This prototype consists of a hydration sensor and a collection device to automatically measure and collect the urine sample respectively. The probe is manually assembled with sensors to measure color, conductivity, pH, and temperature.

collection.

### 2.3. Reference measurements

Urine osmolality and urine specific gravity (USG) were used as references to assess the urine concentration. Both reference measurements were measured at the field location, within 3 h after collection. Urine osmolality was determined via freezing point depression osmometry using an Osmometer (Gonotex Osmomat 3000 Basic, Gonotec, Germany). The obtained urine osmolality value is the average of two measurements. In case the first two measurements differed more than 6 mOsm/kg an additional measurement was taken, and the largest outlier was excluded. Urine specific gravity (USG) was measured once using a refractometer (Atago 4410 PAL—10S, Atago, Japan).

Gastrointestinal temperature was assessed using an ingestible telemetric temperature capsule (eCelsius Medical system, BodyCAP, Caen, France), which is a reliable surrogate marker for core temperature [16]. Participants were instructed to ingest the temperature capsule on the evening before exercise day 1 (D1). Gastrointestinal temperature was measured continuously throughout exercise at 5 min intervals.

### 2.4. Data preprocessing

#### 2.4.1. Calibrations

The color sensor was calibrated during the study: for each urine sample, the output value of the urine measurement per individual filter (red, green, and blue) was divided by the output value of the flush water, measured after the urine sample. In case no flush measurement was obtained, the urine sample was excluded from further analysis.

For the remaining sensors, each probe was individually calibrated within one week before and within one week after the study to ensure a reliable calibration. The functionality of the conductivity and pH sensor was characterized by performing measurements using buffer solutions. For the conductivity sensor, 9 buffer solutions were measured in the range of 1.413 to 111.9 mS/cm using 3 buffer solutions (conductivity = 1.413, 14.13 & 111.9 mS/cm, Thermo Fisher Scientific, USA). The values of the mixed conductivity buffer solutions were measured with a conductivity reference device (HQ2100 Portable Multi-meter, Hach, the Netherlands). The conductivity buffer solutions were poured into the

measurements chamber and measured by the probe while gradually heating the solutions from lab temperature ( $\sim 22^\circ\text{C}$ ) to  $45^\circ\text{C}$ . Thereby, the effect of temperature fluctuations on conductivity values can be corrected. For the pH sensor, four buffer solutions (Orion pH buffers with pH = 1.68, 4.01, 7 & 10.01, Thermo Fisher Scientific, USA) at around  $37^\circ\text{C}$  were measured. The temperature sensor was calibrated against a reference thermometer (OM-HL-EH-TC, Omega, USA) using demi water that was gradually heated to  $45^\circ\text{C}$ .

#### 2.4.2. Timestamp annotation

Fig. 2 shows a typical example of a measurement of the hydration sensor. To subtract the right features from this dataset, an algorithm was used to annotate the timeframe of the urine and flush water measurements. For this, Python (Python Software Foundation. Python Language Reference, version 3.9. Available at <http://www.python.org>) was used.

First, initial timeframes (vertical dashed lines in Fig. 2) were annotated using specific characteristics, such as a sharp increase or decrease and value, of the conductivity and temperature sensor data. These initial timeframes marked the time at which the urine (orange) or flush (gray) entered and left the sensor chamber. As such, these timeframes included a period at the beginning and end at which the measurement chamber was not completely filled. A completely filled measurement chamber is needed to extract a reliable feature. As such, second, the final timeframes (solid vertical lines in Fig. 2) were derived from the initial timeframes. Starting from the initial timestamp at which the fluid enters the chamber, the algorithm searches for the maximal timeframe at which the color sensor deviates less than 1 % of the maximum value that can be obtained (this is the value measured in demi water). This low deviation in color indicates that the chamber is completely filled.

#### 2.4.3. Feature selection

Using the final timeframes, 6 features were obtained from the hydration sensor: conductivity, temperature, pH, and the red, green, and blue color content from the color sensor. Since the conductivity sensor required some settling time, only the mean and standard deviation of the last second of the timeframe were used. For pH, the mean and standard deviation over the whole timeframe were used as features. The temperature feature was obtained by taking the maximum temperature over the whole timeframe. For color, the mean of the urine measurement was

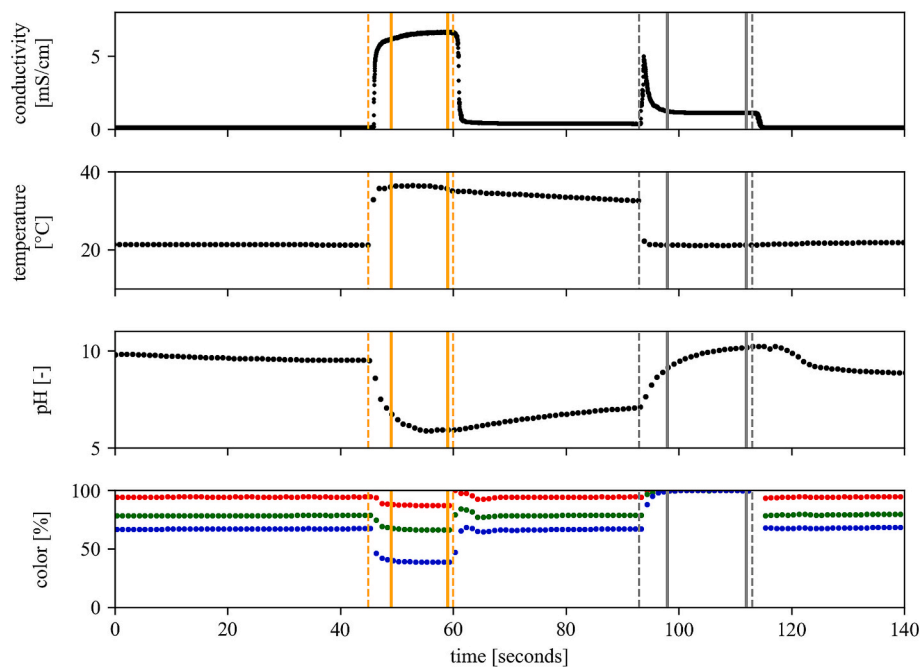


Fig. 2. Typical example of a calibrated measurement as obtained by the hydration sensor. From top to bottom the conductivity, temperature, pH, and 3 color filters (red, green, and blue). The vertical dotted and solid lines represent respectively the initial and final timeframes of the urine stream (orange) and flush (gray). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

divided by the mean of the flush water. In the rare case that the participant did not flush the toilet, no color measurement could be obtained.

## 2.5. Data analysis

All data analysis was performed using Python.

### 2.5.1. Classification model for underhydration estimation

The 6 features obtained from the hydration sensor measurements were used to estimate the (under)hydration status, using USG or osmolality as a reference. Since USG is the frequently used standard in field tests for measuring hydration status, and Cheuvront et al. showed that USG shows low inter-individual variability when compared with urine osmolality, USG is used as the main reference in this study [17].

Before building a classification model, first, feature selection was performed. The relation between a feature and the reference is expected to be linear and was therefore analyzed using the Spearman's rank correlation coefficient ( $\rho$ ). An absolute  $\rho$  of 1 indicates a perfect

monotonic correlation, whereas a  $\rho$  of 0 indicates no correlation. In addition, a  $P$  value is calculated for the  $\rho$ . A  $P$  value lower than 0.05 is considered statistically significant. Features that did not contribute to the estimation of the hydration status, i.e. have a  $|\rho| < 0.5$ , were excluded from further analysis. Second, a standard normal variate (SNV) correction was applied to each individual feature measured with a specific probe [18]. This is needed to correct for the manual making of the probes, which can result in measurement variations that are not related to the properties of the measured urine sample. With SNV, the mean and the standard deviation of a series of measurements is set to respectively zero and one. Finally, the Variance Inflation Factor (VIF) was used to examine the multicollinearity of the features. Features were removed until the VIF factor was lower than 10 for all features.

With the remaining SNV normalized features a linear regression classification model was built using 5-fold cross-validation. In this cross-validation, one of the 5 measurement days (D0-D4) was used as a test set and the remaining days were used as a training set. The performance of the model was given as the mean and standard deviation and the median and interquartile range over the 5 folds using the R-squared ( $R^2$ ) and

Table 2

Overview of obtained data during the study.

	D0	D1	D2	D3	D4
Toilet visits <sup>1</sup>	105	105	103	101	100
Available hydration sensor measurements <sup>2</sup>	101	104	103	67	100
Correct urine measurements	57 %	74 %	72 %	82 %	84 %
Correct flush measurements	90 %	93 %	97 %	92 %	94 %
Useful data per hydration sensor <sup>3</sup>					
Probe 1	28	41	39	42	9
Probe 2	17	28	32	7	47
Probe 3	n.a.	n.a.	n.a.	n.a.	21
Available ingestible capsule temperature matched with urine measurement	n.a.	18	9	n.a.	n.a.

<sup>1</sup> D0 & D1: 1 participant didn't come to the measurement location. D2-D4: participants stopped the walking event.

<sup>2</sup> D0, D1 & D3: respectively 4, 1, and 34 visits were not stored.

<sup>3</sup> 3 probes were used in this study. 2 for each toilet and 1 to replace probe 1. The data is considered useful if: 1) Both urine and flush measurements are correct, 2) both reference measurements are available, 3) outliers are removed (4× reference measurements correlation was off, 4× conductivity measurement of hydration sensor was 0).

mean absolute error (MAE).

### 2.5.2. Core temperature estimation

Urine temperature measured with the probe was compared to temperature measurement using the temperature capsule at the time the maximum urine temperature was measured. Since a monotonic correlation is expected, the relation is accessed by the  $\rho$ .

## 3. Results

### 3.1. Obtained data

Table 2 gives an overview of the obtained data during the study. In total, 106 participants were included in the study. On D0, one participant forgot to go to the toilet, and throughout the event, 6 participants stopped. For almost all toilet visits, a measurement was taken using the hydration sensor. However, on D0, D1, and D3 respectively 4, 1, and 34 measurements were not stored correctly. This error was fixed by restarting the system. Of the available hydration sensor measurements, on average 94 % of the flush measurements were correct, which means that it was possible to correctly annotate a timestamp for the flush. Missing flush measurements were related to a short flush or not flushing the toilet at all. On average, in 75 % of the available hydration sensor measurements, a correct urine measurement was obtained. This number increased throughout the measurement days.

In total, 3 probes (the part of the hydration sensor that contains the 4 sensors) were used during the study. Two for each toilet and one extra to replace probe 1 on D4 because the pH sensor of probe 1 broke during the trial. Both core temperature and urine temperature, respectively measured with the temperature capsule and the hydration sensor, could be obtained from 27 toilet visits. Of the 39 participants that took an ingestible temperature capsule, in 32 cases there was temperature data at the time of the urine measurement. In 5 rare cases, the maximum recorded urine temperature was however not within the annotated urine timeframe. These measurements were removed from further analysis.

### 3.2. Classification results for underhydration estimation

Fig. 3a shows the positive linear correlation between the reference methods urine specific gravity (USG) and osmolality ( $\rho = 0.95$ ,  $P <$

0.001). This correlation was independent of measurement day, sex, and walking distance (for all variables, the  $\rho$  is between 0.91 and 0.99). The distribution of USG between the walking days (D1–4) is similar, as shown in Fig. 3b. On the day before the walking event (D0), slightly lower USG levels were observed compared to the post-exercise values.

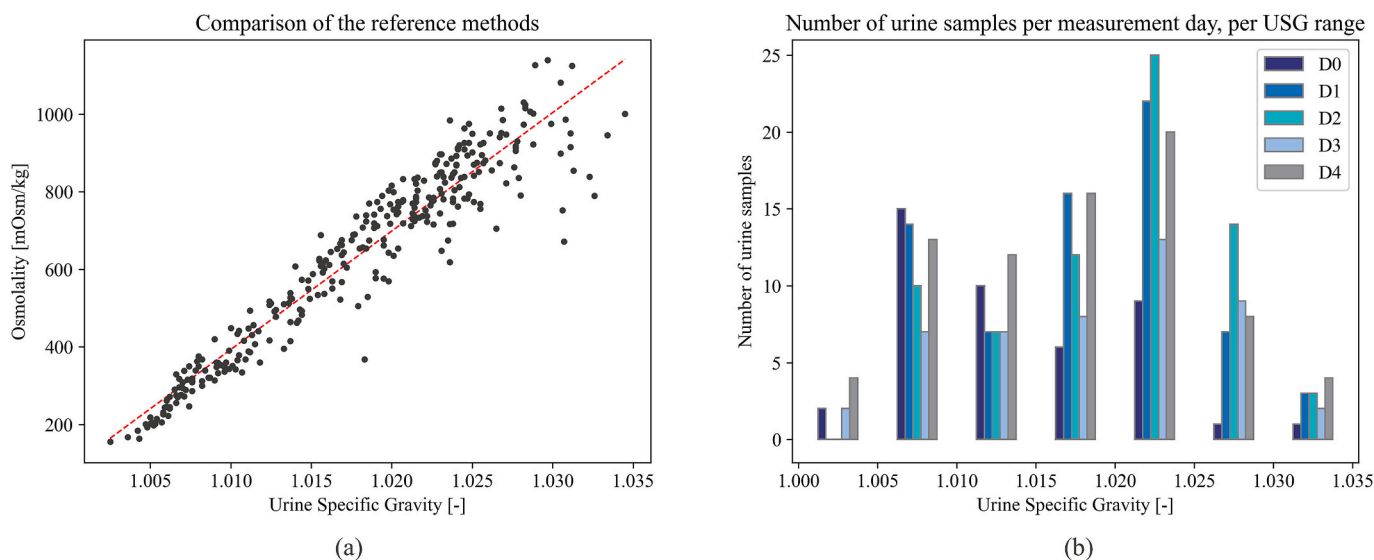
Fig. 4 shows the linear correlation of the 6 features measured with the hydration sensor to USG. Because the pH sensor of probe 1 broke during the trial, these measurements were excluded from analysis and are not shown. Both temperature and pH do not correlate with USG, with a  $\rho$  of  $-0.11$  and  $-0.02$  respectively, and are excluded from further analysis in estimating the hydration status. The remaining features, conductivity, and the 3 color filters have a high correlation with USG. However, a clear dependence on the probe used is observed. To correct for this, SNV normalization was applied to each individual feature for each probe (Fig. 5). As can be seen, for each feature, the  $\rho$  per probe remains the same but the combined  $\rho$  over all probes increases. Using the VIF score, a high multicollinearity (defined as VIF higher than 10) between the color features was observed. To increase the reliability of the regression model and prevent overfitting of the model, only one color filter was included in the regression model. This resulted in 7 regression models, of which the performance on the test sets is shown in Table 3. The performance on the test sets using the median and interquartile range, and the performance of the training sets are given in Table S1 and Table S2, respectively.

The best classification result was obtained by combining the green filter of the color sensor with the conductivity sensor. With an  $R^2$  of 0.85, 85 % of the variance in USG can be explained by these two features. Fig. 6 shows the classification result of this green filter and conductivity in estimating the USG. From the Bland-Altman plot, the bias (mean difference) is  $<0.00001$  with a 95 % limits of agreement of 0.00550.

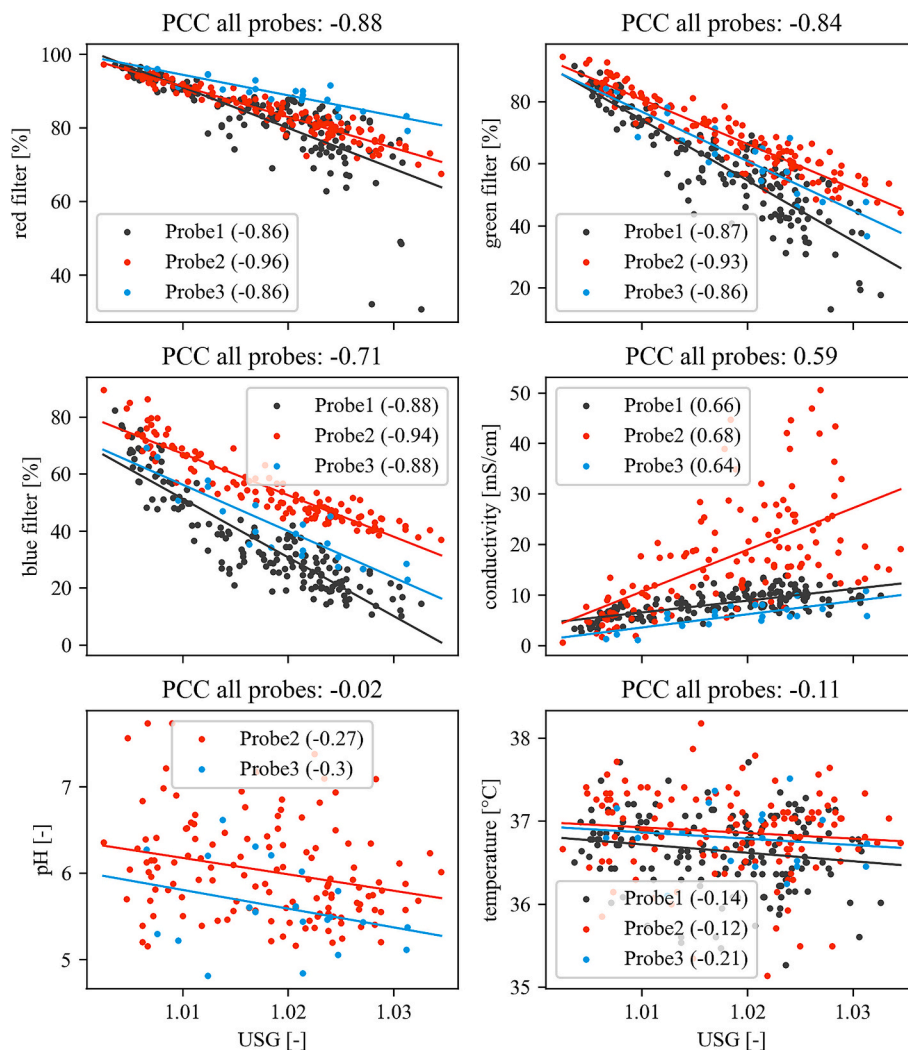
When using urinary osmolality as the reference method, the performance of the hydration sensor using the green filter and conductivity is slightly lower with an  $R^2$  of  $0.76 \pm 0.10$  and a mean average error of  $111 \pm 12$  mOsm/kg. Fig. S6 shows its correlation and Bland-Altman plot.

### 3.3. Core temperature estimation

The linear correlation and Bland-Altman plot of the temperature measured with the probe and temperature capsule are given in Fig. 7. There is a linear correlation between the two methods with a  $\rho$  of 0.62 ( $P$



**Fig. 3.** Reference measurements. (a) Correlation between the two reference methods: urine specific gravity (USG) and osmolality. Each point represents a urine sample. The red line shows the linear correlation between the two methods with a Spearman correlation coefficient of 0.95. The standard deviation of osmolality is plotted but smaller than the size of the plotted points. (b) The distribution of USG for each measurement day. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)



**Fig. 4.** Correlation of the calibrated features measured with the hydration sensor. For each feature, the  $\rho$  per probe (in the legend) and for all probes combined (in the title) are given. Because the pH sensor of probe 1 broke during the trial, its measurements are not shown. Besides the pH ( $P = 0.690$ ) and temperature ( $P = 0.046$ ), all correlations were significantly different with  $P < 0.001$ .

$< 0.001$ ). On average, urine temperature was  $0.82$  °C lower than core temperature, with a 95 % limits of agreement of  $0.58$  °C.

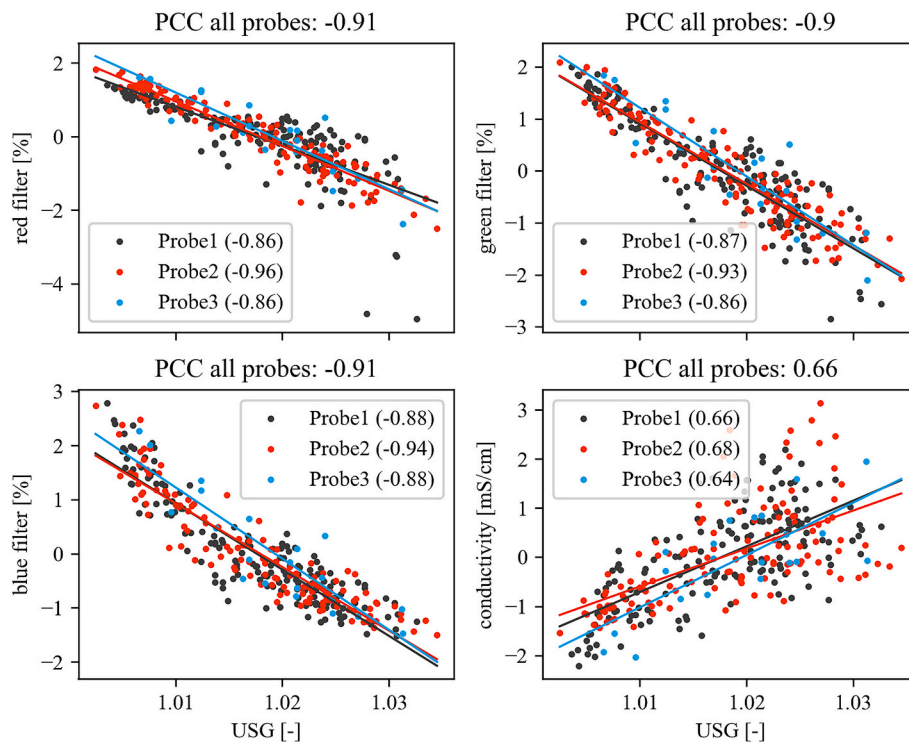
#### 4. Discussion

This study aimed to validate a urine analysis hydration prototype that was developed to automatically assess underhydration status during a toilet visit. In total, 106 participants were measured on 5 days during a large four-days walking event, resulting in more than 500 measurements. We showed that this prototype can estimate underhydration status of the participants with an  $R^2$  of 0.85, using urine specific gravity as a reference. To the best of our knowledge, this is the first report on an accurate method for underhydration assessment by which urine is automatically measured when participants go to the toilet. These results offer great potential for the repeated, non-obtrusive, and automated monitoring of underhydration status. Underhydration is a common problem in elderly but due to the lack of non-obtrusive and easy methods, the current practice in care homes in the Netherlands is to not monitor underhydration by default. Wilson et al. showed that in many residents in two care homes in West London were at risk of chronic underhydration by consuming less than 1.5 L of fluid daily [19]. One of their strategies to overcome this is to identify and respond when hydration needs are not met. As such, one of our envisioned use cases is the

at-home monitoring of elderly so that underhydration is detected and treated at an early stage.

In this study, we were able to automatically to analyze the urine in 75 % of 514 toilet visits during a multiple-day experiment in field settings. The fact that we were not able to obtain good quality measurements for all toilet visits is partly related to the study setup in which participants were asked to visit the toilet when they arrived at the measurement location, even if they did not feel the urge to urinate. This effect is particularly prominent at the first measurement day when participants did not yet consider the planned toilet visit. For the envisioned use case of monitoring elderly in care homes, we do not foresee a problem here as we expect elderly to only go to the toilet when they feel an urge.

An annotation and classification algorithm was developed that can automatically analyze the measured data. This would allow real-time feedback about someone's hydration status in the future. In comparison with current urine-testing practices, which require manual actions like collecting and assessing the urine sample [12–15], our proposed solution is time-effective and does not require any manual action from a healthcare professional or the end user. The urine sample is measured when someone goes to the toilet without the need to collect a sample, and with the developed annotation and classification algorithm, underhydration status can be automatically assessed.



**Fig. 5.** Correlation of the features measured with the hydration sensor after SNV normalization. For each feature, the  $\rho$  per probe (in the legend) and for all probes combined (in the title) are given. All correlations were significantly different with  $P < 0.001$ .

**Table 3**

Performance of the linear regression model on the test set, using USG as reference, per (combination) of feature(s) used for the model.

	R-squared	MAE <sup>1</sup>
	mean $\pm$ standard deviation	mean $\pm$ standard deviation
Green + Conductivity	0.85 $\pm$ 0.03	0.00215 $\pm$ 0.00013
Blue + Conductivity	0.83 $\pm$ 0.02	0.00232 $\pm$ 0.00016
Red + Conductivity	0.81 $\pm$ 0.03	0.00242 $\pm$ 0.00014
Blue	0.80 $\pm$ 0.03	0.00258 $\pm$ 0.00021
Green	0.80 $\pm$ 0.03	0.00262 $\pm$ 0.00013
Red	0.72 $\pm$ 0.03	0.00298 $\pm$ 0.00019
Conductivity	0.35 $\pm$ 0.08	0.00483 $\pm$ 0.00040

<sup>1</sup> MAE: mean absolute error.

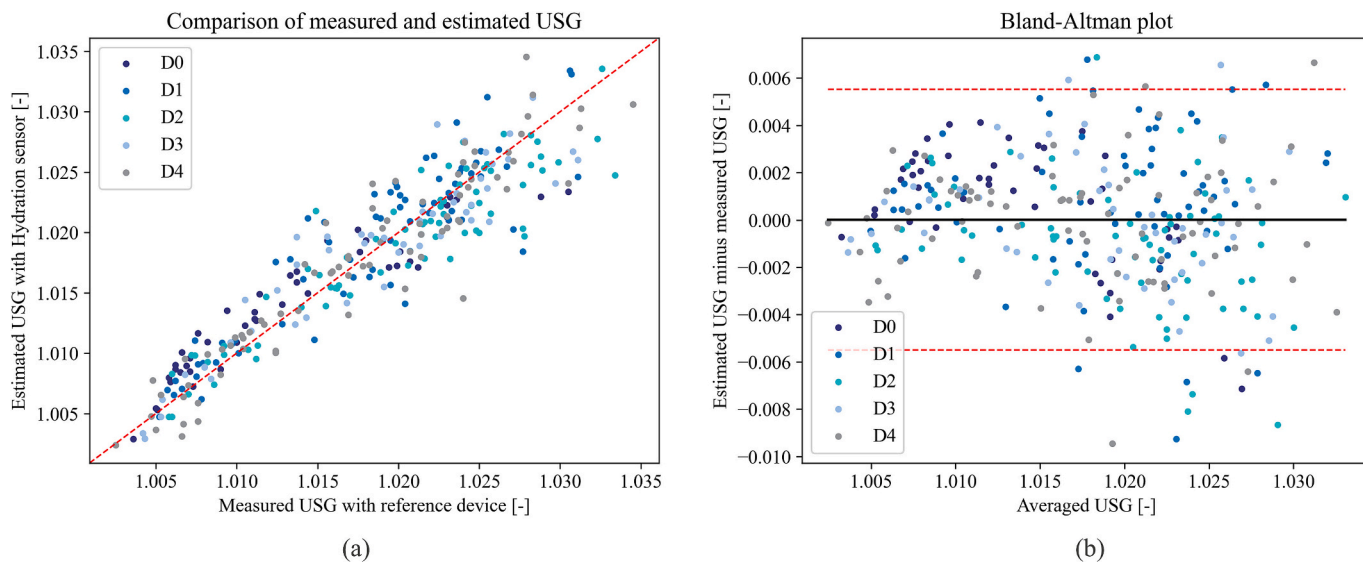
As a reference for the hydration sensor, both osmolality and urine specific gravity were used. They show a high linear correlation that was not affected by the measurement day, sex, or walking distance. Other reported urinary reference methods, such as assessing urine color with a color chart, or measuring the hydration status with a dipstick, were not used because of their low reported performance [20–22]. Of osmolality and USG, osmolality values can vary widely across cultures, which may be related to differences in diets such as sodium intake [23]. USG is the frequently used standard in field tests and shows low inter-individual variability for measuring hydration status [13]. As such, we focused on using USG as a reference. We showed that the hydration sensor could estimate the USG with an R-squared of 0.85 and a mean absolute error of 0.00215, in a USG range between 1.0025 and 1.0345. This covers the whole range described by Casa et al. and is similar to other USG values reported in the literature [24,25].

There are a few articles that report on an automated method to assess underhydration status by measuring urine [26,27]. However, these devices were tested by pouring collected urine samples into the devices instead of automatically measuring urine when participants went to the toilet. Bender et al. developed an automated urinalysis device that can be installed in a urinal and tested it on collected urine samples of 151

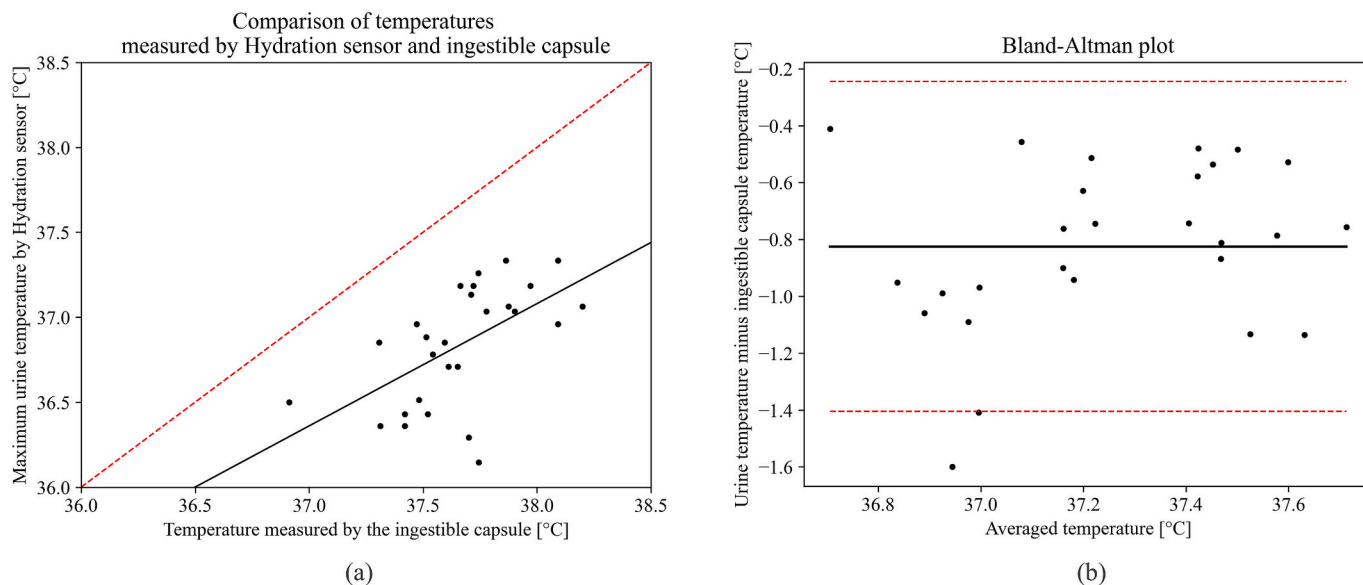
male athletes [26]. They reported a mean absolute error of  $0.0029 \pm 0.0021$  (in this study  $0.00215 \pm 0.00013$ ). In another study by Walawender et al., a novel mobile technology application to measure the light penetrance of a collected urine sample to measure the urine concentration was developed [27]. This was tested on 25 randomly selected urine samples from children, and they showed an R-squared of 0.59 in comparison with urine osmolality (0.76 in this study).

Which performance is clinically needed to monitor and detect dehydration is not clearly defined. Casa et al. defined 4 stages of (de) hydration, from ‘well hydrated’ to ‘serious dehydration’, in steps of 0.010 USG [24]. However, these fixed cutoffs may not be generally applicable as they depend on body size and composition. As such, Wilson proposes to use a personalized approach for defining someone’s hydration status, especially for people in the upper quintiles of body mass index and lean body mass [28]. By using our prototype, underhydration status can be personally assessed each time someone goes to the toilet. Thereby, a trend can be measured over a long time, which allows for a personalized analysis of the variation of hydration status. As such, instead of using predefined general cutoffs, feedback about someone’s underhydration status can be individualized.

Besides estimating underhydration status, we explored the possibility of estimating core temperature with urine temperature as measured by our prototype. We showed a linear correlation between urine temperature and core temperature, with a mean difference of  $0.82 \text{ }^\circ\text{C}$ . This difference was not affected by core temperature. As expected, urine temperature was lower than core temperature because the urine cools down after leaving the human body and when it is collected by the prototype in the toilet. This difference might be corrected for by modeling this temperature decrease. However, this requires more samples than the number of samples in this study. The 95 % limits of agreement of the temperature difference was low ( $0.57 \text{ }^\circ\text{C}$ ) in comparison with other thermometry techniques that allow for regular self-assessment of core temperature [29,30]. Ekers et al. show that tympanic measurements show the closest agreement with the pulmonary artery standard with a mean difference in temperature of  $-0.20 \text{ }^\circ\text{C}$  and a



**Fig. 6.** Classification results using the green filter of the color sensor and conductivity. (a) In the correlation plot ( $\rho = 0.92, P < 0.001$ ) each point represents a urine sample. The red dotted line represents the  $x = y$  line. (b) The Bland-Altman plot ( $\rho = -0.19, P = 0.001$ ) shows that the error increases with USG. Here the solid black and dashed red lines indicate respectively the mean ( $< 0.00001$ ) and the 95 % limits of agreement (0.00550) difference in USG. For 20 samples the estimated USG was 0.0055 USG higher ( $n = 8$ ) or lower ( $n = 12$ ) than the real USG. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)



**Fig. 7.** Temperature measurements. (a) Correlation between urine temperature measured with the hydration sensor, and core temperature measured with the ingestible. The black line represents the linear correlation ( $\rho = 0.62, P < 0.001$ ), and the dotted red line represents the  $x = y$  line. (b) The Bland Altman plot of the two measured temperatures ( $\rho = 0.23, P = 0.26$ ). Here the solid black and dashed red lines indicated respectively the mean ( $-0.82 \text{ }^\circ\text{C}$ ) and the 95 % limits of agreement ( $0.58 \text{ }^\circ\text{C}$ ) difference in temperature. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

95 % limits of agreement of  $-0.92$  to  $0.52 \text{ }^\circ\text{C}$  [29]. Even though the number of samples in this study is low, and no hard conclusions can be drawn from the data presented in this study, our results indicate that urine temperature might be a promising measure to approximate core temperature. To strengthen this indication, a larger study should be conducted that also allows for the correction of the inevitable temperature decrease of the urine after leaving the body.

A strength of this study was that we were able to validate our prototype on a large number of participants for multiple days. Besides validating the performance of the prototype in estimating under-hydration status, this allowed us to test the intensive usage of the

prototype being about 50 toilet visits in 3 h per day. However, some limitations should be considered. First, the hydration sensors were prototypes and manually made. This resulted in small differences between the 3 probes that were used in the study, which affected the measurements. As such, the same urine sample would give slightly different measurements when measured with a different probe. We compensated for this by using standard normal variate normalization. By automating the manufacturing process, the next version could be made in a more reproducible manner, preventing the need for this preprocessing step in the future. Second, even though the prototypes measured over 500 toilet visits within a week, the prototype was not

validated to automatically measure the hydration status over a longer period of time. For a validation like this, a long-term study is needed. In such a study, the potential accumulation of urine sediment on the sensors, which did not emerge in this short-term study, should be accessed over a longer period of time. Finally, in this study, participants of a large walking event were measured, which makes the setup of this study not entirely representative of the envisioned use case of monitoring elderly at home. Even though our study population was on average older than 65 years, the participants were physically more active as they walked 30 to 50 km for four days in a row. Although we have no reason to assume that the accuracy of the prototype, as reported in this study, would differ when measuring elderly at home, a study to validate this should first be conducted.

Future directions of research will first include the development of an improved version of the prototype used in this study. Such a next version would need fewer sensors to be able to estimate the hydration status than the sensors in the current design. In this study, we showed that by only using conductivity and the green content of the color sensor, a high correlation with USG could be obtained. As such, the pH sensor and red and blue color content of the color sensor would not be needed. The temperature sensor, however, is still necessary for the calibration of the conductivity sensor. Besides reducing the number of sensors, the manufacturing process should be automated to prevent any inter-probe variations. Second, the improved version of the prototype should be further validated for our final use case, the repeated underhydration status monitoring at home. To allow this, a study should be conducted in which the toilet visits of participants are measured over a longer time, preferably more than one month. Such a study could also be used to research where trends in underhydration coexist with clinical dehydration, as assessed with serum or plasma osmolality. Finally, in this study, no feedback is given to the participants about their hydration status as this study aimed to validate the prototype. Nevertheless, with the models developed this would be possible and future research should focus on how to provide this feedback. Potentially, the current models can even be extended by personalized models that can predict the development of hydration status and give personalized feedback.

In summary, we showed that our hydration sensor could accurately estimate underhydration status of participants of a large four-day walking event, using urine specific gravity as a reference. The measurements were performed automatically, without any manual action from the participants needed, and a model has been developed that would allow real-time feedback. These findings support that this prototype could be used for non-obtrusive, time-effective, and accurate underhydration monitoring. The next step is to validate an improved version of this prototype in the proposed setting, being the monitoring of underhydration status in elderly over a long period in their home situation.

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### CRediT authorship contribution statement

**Esther Kho:** Writing – review & editing, Writing – original draft, Validation, Methodology, Investigation, Formal analysis, Data curation, Conceptualization. **Eva C. Wentink:** Writing – review & editing, Validation, Methodology, Investigation, Formal analysis, Conceptualization. **Vera D.M. Verbiest:** Writing – review & editing, Validation, Investigation, Formal analysis. **David T. Young:** Writing – review & editing, Validation, Methodology, Conceptualization. **Maria T.E. Hopman:** Writing – review & editing, Project administration, Conceptualization. **Coen C.W.G. Bongers:** Writing – review & editing, Project administration, Conceptualization.

### Declaration of competing interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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### Appendix A. Supplementary data

Supplementary data to this article can be found online at <https://doi.org/10.1016/j.sbsr.2025.100763>.

### Data availability

Data will be made available on request.

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