Evaluating and testing hydration status is increasingly requested by rehabilitation, sport, military and performance-related activities. Besides commonly used biochemical hydration assessment markers within blood and urine, which have their advantages and limitations in collection and evaluating hydration status, there are other potential markers present within saliva, sweat or tear. This literature review focuses on body fluids saliva, sweat and tear compared to blood and urine regarding practicality and hydration status influenced by fluid restriction and/or physical activity. The selected articles included healthy subjects, biochemical hydration assessment markers and a well-described (de)hydration procedure. The included studies (n = 16) revealed that the setting and the method of collecting respectively accessing body fluids are particularly important aspects to choose the optimal hydration marker. To obtain a sample of saliva is one of the simplest ways to collect body fluids. During exercise and heat exposures, saliva composition might be an effective index but seems to be highly variable. The collection of sweat is a more extensive and time-consuming technique making it more difficult to evaluate dehydration and to make a statement about the hydration status at a particular time. The collection procedure of tear fluid is easy to access and causes very little discomfort to the subject. Tear osmolarity increases with dehydration in parallel to alterations in plasma osmolality and urine-specific gravity. But at the individual level, its sensitivity has to be further determined.

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As blood sampling is often difficult to execute in the field, evaluating hydration status in these settings includes determination of urine osmolality (URosm), urine-specific gravity (URsg) and urine colour (URcol). URsg seems to be the most valid marker in the setting of dynamic (monitoring over time) dehydration assessment among these urinary markers. To be of practical use, measuring and evaluating hydration status should have the possibility to be used on a daily or even hourly basis. The ability to monitor hydration status has become increasingly studied within the rehabilitation, sport, military and performance-related activities. Besides BMC, BPosm and URsg seem to be the most effective markers to monitor hydration status among the aforementioned markers.

There might be other potential biochemical markers present within saliva, sweat and tears being noninvasive ‘freely accessible’ body fluids. These markers can offer further in-depth knowledge of an individual’s hydration status. On this basis, the interest of this review is to evaluate ‘freely accessible’ body fluids (saliva, sweat and tear) as hydration assessment markers compared to the aforementioned body fluids (blood: BPosm, BPNa+), BSosm, urine: URosm, URsg, URcol) during a well-described dehydration procedure. To obtain salivary concentration samples, the participants were asked to swallow followed by a period of 2 min of passive saliva collection. The collected saliva was then expelled (spit) into a polypropylene Falcon tube (for example, Voigt Global Distribution, Leicestershire, UK) which was placed under the tongue for 2 min. During this time, the participants were asked to remain still, and saliva samples were collected. Regarding saliva, Smith et al. measured SAosm after centrifugation by an eight-point color chart (8-Point Color Chart). An ion-selective electrode measures the potential of a specific ion in solution (mmol/l); BP[Na+]: 135–145 mmol/l (euyhydrated), 22,23

Specific gravity (g/ml) is the ratio of the density of a substance to the density of a reference substance and is measured by a refractometer: URwg: < 1.010 g/ml (euhydrated).13,14

The urine colour is measured by an 8-point color chart: URcol: 1 or 2 (euhydrated).14

RESULTS

In the Cochrane Library n = 388 studies were recorded: dehydration and body weight (279 hits), dehydration AND saliva (19 hits), dehydration AND sweat (75 hits), dehydration AND tear (15 hits), and dehydration AND axillary moisture (0 hits). In PubMed n = 312 studies were found. After excluding records by duplicates and by not relevant titles, abstracts and full texts, 15 records met our inclusion criteria and one record was included through links of related articles (references). In summary, a total of n = 16 studies was included in this review. Figure 1 presents the search strategy and selection process.

Dehydration procedures

In this section the dehydration procedures of the included studies of saliva (Table 1), sweat (Table 2) and tear are listed. To achieve an euhydrated state before testing all studies conducted a well-described hydration protocol. For a controlled dehydrated status, either a stationary cycle ergometer was used in an environmental chamber (sweat,19,39,40 and tear7,13) or a treadmill (sweat35,34,39,41 and sweat42). Passive dehydration was either used with fluid restriction42,43 or extracellular dehydration using a loop diuretic (Furosemide). For comparison reasons, the results stand for the control/placebo groups to evaluate (de)hydration status.

Measurement equipment and direct fluid collection of saliva, sweat and tear

For most of the included studies, the measurement equipment to evaluate osmolality and sodium concentration of saliva and sweat do not differ compared to the evaluation of blood and urine.19,20,28,29,34,36,37,39,40,42,44

Regarding saliva, Smith et al. measured SAosm was measured after centrifugation by a freezing-point depression osmometer (for example, Fiske Associates, Fiske Micro-Osmometer, Model 210/Fiske One-Ten Osmometer) and Taylor et al. measured SAosm was measured after centrifugation by an ion-selective electrode (Beckman Synchron E/T-ISE, Fullerton, CA, USA43,44). Direct collection of saliva samples is possible in two ways as described in the study by Ely et al. Saliva samples (expectorated29,38,44) were drawn by participants initially asked to swallow followed by a period of 2 min of passive saliva collection. The collected saliva was then expelled (spit) into a polypropylene Falcon tube (for example, Voigt Global Distribution, Inc., Lawrence, KS, USA). Saliva samples (salivette29,43,44) were asked to swallow before saliva collection. Saliva was collected with the use of a pre-weighted polyester salivette swab (for example, Sarstedt, Leics, UK) which was placed under the tongue for 2 min. During the collection period participants avoided any orofacial movements. To obtain salivary concentration samples, the participants were asked to accumulate saliva in their mouth (that is, passive

Saliva fluid. SAosm was measured after centrifugation by a freezing-point depression osmometer (for example, Fiske Associates, Fiske Micro-Osmometer, Model 210/Fiske One-Ten Osmometer) and Taylor et al. measured SAosm was measured after centrifugation by an ion-selective electrode (Beckman Synchron E/T-ISE, Fullerton, CA, USA). Direct collection of saliva samples is possible in two ways as described in the study by Ely et al. Saliva samples (expectorated29,38,44) were drawn by participants initially asked to swallow followed by a period of 2 min of passive saliva collection. The collected saliva was then expelled (spit) into a polypropylene Falcon tube (for example, Voigt Global Distribution, Inc., Lawrence, KS, USA). Saliva samples (salivette29,43,44) were asked to swallow before saliva collection. Saliva was collected with the use of a pre-weighted polyester salivette swab (for example, Sarstedt, Leics, UK) which was placed under the tongue for 2 min. During the collection period participants avoided any orofacial movements. To obtain salivary concentration samples, the participants were asked to accumulate saliva in their mouth (that is, passive

Measurement equipment and quantity estimates

For evaluating hydration status, osmolality (mOsmol/l) and osmolality (mOsmol/kg) are mainly measured by freezing-point depressors. The freezing points of solutions are lower with osmolality (mOsmol/l) are mainly measured by freezing-point (BPosm: 275–295 mOsmol/kg (euyhydrated), 20,23,34 BSosm: 282–295 mOsmol/kg (euyhydrated), 25 and URosm: < 700 mOsmol/kg (euhydrated).13,36

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drool technique) and finally expel around 1 ml of saliva into a Dixie cup.

Sweat can be directly collected via two different techniques: (1) absorbancy method (i.e. sweat patch collection); and (2) whole-body washdown technique.\(^ {34}\)

(1) For the absorbancy method, the patch location was first cleaned (alcohol and distilled water) and then dried (air). Afterwards sweat patches were used where they were needed (for example, side-by-side to the upper back, just below the shoulder blades, to the forearm, chest and mid-thigh on the right-hand side—the patches remained in place throughout the trial).\(^ {34,40}\) Sweat was measured in small batches by for example a Cobas C311 module (Roche Diagnostics, Basel, Switzerland) using the ion-selective electrode technique for SW\([Na^+]\) (Easylyte Plus, Medica Corporation, Bedford, MA, USA,\(^ {19}\) Beckman Synchron El-ISE, Fullerton, CA, USA,\(^ {34}\) Beckman Instruments Inc., AS 80 System, Galway, Ireland\(^ {39}\)) or flame photometry (Sherwood, Cambridge, UK\(^ {40}\)).

(2) For the whole-body washdown technique, a kiddie pool placed in a fully enclosed walk-in tent was prepared in advance. After completion of the performance trial, each participant entered the plastic kiddie pool and approximately 1.5 l of distilled water was poured over the participant's head and body.\(^ {34}\) The participant was then asked to remove all clothing in the privacy of the tent and then pour the remaining amount of distilled water (3.78 l total) over his or her body. The removed clothes remained in the kiddie pool and to obtain water samples Eppendorf tubes were used.

Tear fluid was collected and analysed for TE\(_{\text{osm}}\) using a commercially available diagnostic device (TearLab Osmolarity System; TearLab, San Diego, CA, USA).\(^ {27,31}\) Participants blinked three times and squeezed their eyes shut. The released tear fluid from the lacrimal gland was immediately collected from the right eye using a handheld pen. A signal was transmitted when a sufficient volume (50 nl) was collected, which typically took < 5 s. Once docked on the TearLab platform, the outcome was presented within 10 s. BP\(_{\text{osm}}\) was measured by a freezing-point depression osmometer (Model 2020\(^ {27}/\)Model 330 MO,\(^ {31}\) Advanced Instruments).

**DISCUSSION**

The aim of the review was to evaluate ‘freely accessible’ and noninvasive body fluids (saliva, sweat and tear) compared to biochemical hydration assessment markers such as those within blood and urine during a well-described (de)hydration procedure influenced by fluid restriction and/or physical activity. First, the practical use of the different hydration assessment markers and, second, the results of saliva, sweat and tear body fluids are discussed regarding hydration status.

Practical use of body fluid hydration assessment markers

The measurement equipment for the evaluation of the hydration status change did not differ substantially between the included studies. This means that the underlying methods to evaluate osmolarity, osmomiality\(^ {20}\) and sodium concentration\(^ {22}\) of body fluids were comparable. In this regard, not only the assessment technique itself but the procedure of collecting body fluids has a fundamental impact on the use of the biochemical hydration assessment markers. Collecting blood is an invasive procedure that makes it often difficult to execute in the field. Furthermore,
| Study            | Dehydration procedure                  | Monitoring time (body mass) | Urine  | Abs  | Rel | Blood  | Abs  | Rel | Saliva  | Abs  | Rel |
|------------------|---------------------------------------|----------------------------|--------|------|-----|--------|------|-----|---------|------|-----|
| Cheuvront et al. | Cycling/running (indoor) and fluid restriction | Baseline (78.2) | UR\text{\textsubscript{osm}}: 614 | —     | —   | BP\text{\textsubscript{osm}}: 292 | —    | —   | SA\text{\textsubscript{osm}}: 71 | —    | —   |
|                  |                                        | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.018 | UR\text{\textsubscript{col}}: 3.8 | —     | —   | BP\text{\textsubscript{Na\textsuperscript{+}}}: 143 | —    | —   | SA\text{\textsubscript{Na\textsuperscript{+}}}: 18 | —    | —   |
|                  |                                        | BMC 2.5% | UR\text{\textsubscript{osm}}: 1018 | 404   | 65.8 | BP\text{\textsubscript{osm}}: 301 | 9    | 3.1 | SA\text{\textsubscript{osm}}: 86 | 15   | 21.1 |
| Ely et al.       | Cycling/running (indoor) and fluid restriction | Baseline (83.8) | UR\text{\textsubscript{osm}}: 600 | —     | —   | BP\text{\textsubscript{osm}}: 291 | —    | —   | SA\text{\textsubscript{osm}}: 58 | —    | —   |
|                  |                                        | BMC 4% | UR\text{\textsubscript{osm}}: 1.018 | 0.01  | 1    | BP\text{\textsubscript{osm}}: 303 | 12   | 4.1 | SA\text{\textsubscript{osm}}: 66 | 38   | 65.5 |
| Ely et al.       | Pharmaceutical (furosemide) and fluid restriction | Baseline (79.1) | UR\text{\textsubscript{osm}}: 1.015 | —     | —   | BP\text{\textsubscript{osm}}: 289 | —    | —   | SA\text{\textsubscript{osm}}: 66 | 38   | 65.5 |
| Hew-Butler et al.| Running (indoor) and fluid restriction  | Baseline (79.1) | UR\text{\textsubscript{osm}}: 600 | —     | —   | BP\text{\textsubscript{osm}}: 289 | —    | —   | SA\text{\textsubscript{osm}}: 66 | 38   | 65.5 |
|                  |                                        | UR\text{\textsubscript{Na\textsuperscript{+}}}: 110 | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.028 | 0.006 | 0.6 | BP\text{\textsubscript{Na\textsuperscript{+}}}: 292 | 3    | 1   | SA\text{\textsubscript{Na\textsuperscript{+}}}: 77 | 9    | 13.2 |
| Muñoz et al.     | Cycling (indoor) and fluid restriction  | Baseline (79.1) | UR\text{\textsubscript{osm}}: 610 | —     | —   | BP\text{\textsubscript{osm}}: 110 | —    | —   | SA\text{\textsubscript{osm}}: 64 | —    | —   |
|                  |                                        | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.012 | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.018 | 0.006 | 0.6 | BP\text{\textsubscript{Na\textsuperscript{+}}}: 110 | —    | —   | SA\text{\textsubscript{Na\textsuperscript{+}}}: 64 | —    | —   |
| Oliver et al.    | Walking (indoor) and fluid restriction  | Baseline (74.7) | UR\text{\textsubscript{osm}}: 640 | 40    | 6.7  | BP\text{\textsubscript{osm}}: 294 | 5    | 1.7 | SA\text{\textsubscript{Na\textsuperscript{+}}}: 20 | 2    | 11.1 |
| Perrier et al.   | Fluid restriction                      | Baseline (74.7) | UR\text{\textsubscript{osm}}: 610 | 40    | 6.7  | BP\text{\textsubscript{osm}}: 294 | 5    | 1.7 | SA\text{\textsubscript{Na\textsuperscript{+}}}: 20 | 2    | 11.1 |
|                  |                                        | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.012 | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.018 | 0.006 | 0.6 | BP\text{\textsubscript{Na\textsuperscript{+}}}: 110 | —    | —   | SA\text{\textsubscript{Na\textsuperscript{+}}}: 64 | —    | —   |
| Pross et al.     | Fluid restriction                      | Baseline (74.7) | UR\text{\textsubscript{osm}}: 640 | 40    | 6.7  | BP\text{\textsubscript{osm}}: 294 | 5    | 1.7 | SA\text{\textsubscript{Na\textsuperscript{+}}}: 20 | 2    | 11.1 |
|                  |                                        | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.012 | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.018 | 0.006 | 0.6 | BP\text{\textsubscript{Na\textsuperscript{+}}}: 110 | —    | —   | SA\text{\textsubscript{Na\textsuperscript{+}}}: 64 | —    | —   |
| Smith et al.     | Running (indoor) and fluid restriction  | Baseline (81.7) | UR\text{\textsubscript{osm}}: 640 | 40    | 6.7  | BP\text{\textsubscript{osm}}: 294 | 5    | 1.7 | SA\text{\textsubscript{Na\textsuperscript{+}}}: 20 | 2    | 11.1 |
|                  |                                        | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.012 | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.018 | 0.006 | 0.6 | BP\text{\textsubscript{Na\textsuperscript{+}}}: 110 | —    | —   | SA\text{\textsubscript{Na\textsuperscript{+}}}: 64 | —    | —   |
| Taylor et al.    | Cycling (indoor) and fluid restriction  | Baseline, trial 1 (79.1) | UR\text{\textsubscript{osm}}: 640 | 40    | 6.7  | BP\text{\textsubscript{osm}}: 294 | 5    | 1.7 | SA\text{\textsubscript{Na\textsuperscript{+}}}: 20 | 2    | 11.1 |
|                  |                                        | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.012 | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.018 | 0.006 | 0.6 | BP\text{\textsubscript{Na\textsuperscript{+}}}: 110 | —    | —   | SA\text{\textsubscript{Na\textsuperscript{+}}}: 64 | —    | —   |
| Walsh et al.     | Cycling (indoor) and fluid restriction  | Baseline (73.9) | UR\text{\textsubscript{osm}}: 610 | 40    | 6.7  | BP\text{\textsubscript{osm}}: 294 | 5    | 1.7 | SA\text{\textsubscript{Na\textsuperscript{+}}}: 20 | 2    | 11.1 |
|                  |                                        | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.012 | UR\text{\textsubscript{Na\textsuperscript{+}}}: 1.018 | 0.006 | 0.6 | BP\text{\textsubscript{Na\textsuperscript{+}}}: 110 | —    | —   | SA\text{\textsubscript{Na\textsuperscript{+}}}: 64 | —    | —   |

Abbreviations: Abs (absolute difference to baseline); BMC (body mass change, percentage change); BP\text{\textsubscript{osm}}/BS\text{\textsubscript{osm}} (blood plasma/serum osmolality, mOsmol/kg); BP\text{\textsubscript{Na\textsuperscript{+}}}/BS\text{\textsubscript{Na\textsuperscript{+}}} (blood plasma/serum sodium concentration, mmol/l); Rel (relative difference to baseline); SA\text{\textsubscript{osm}} (saliva osmolality, mOsmol/kg, euhydrated: 83 mOsmol/kg); SA\text{\textsubscript{Na\textsuperscript{+}}} (saliva sodium concentration, mmol/l); UR\text{\textsubscript{osm}} (urine osmolality, mOsmol/kg); UR\text{\textsubscript{Na\textsuperscript{+}}} (urine sodium concentration, mmol/l); UR\text{\textsubscript{col}} (urine colour, units); values estimated out of figures°; italic = athletes/soldiers.
BP_{osm} and BS_{osm} are tightly regulated in the brain. They are not good indices of the hydration status across days but across hours, because the kidneys constantly attempt to bring tonicity back below 296 mOsmol/kg.45

Urinary collection is a noninvasive method. A limiting factor during the dehydration process is the availability of urine (for example, bladder voiding is not always feasible). Measuring UR_{ig} with a refractometer is less subjective than UR_{col} as well as simple to use.14 Although UR_{osm}, UR_{ig}, and UR_{col} have been suggested for screening older adults for dehydration, their diagnostic accuracy is too marginal to be beneficial.46 In addition, a dehydration procedure for older adults is not reasonable. Compared to blood and urine, saliva samples can always be directly collected and are always available. But saliva markers seem to be highly variable between subjects (see below). Furthermore, there are two possible techniques to collect sweat—the absorbancy method (that is, sweat patch collection) or the whole-body washdown technique.34 Compared to the collection of the other body fluids both techniques provide no baseline measurement. The collection of sweat is time-consuming and the sweating protocol from plasma through acinar cells. The ECF sodium concentration increases and this is reflected in an increase in BP_{osm} during hypertonic-hypovolemia dehydration what might be linked with the secretion of more concentrated saliva with a decrease in TBW.36 SA_{osm} has been shown to increase with progressive dehydration,29,36,38 fluid deprivation and restriction.29,43 Furthermore, to record alterations during hypertonic-hypovolemia dehydration, SA_{osm} might be as sensitive as UR_{osm}. Given that a fluid intake of 1.0 l per day seems to be insufficient to compensate water losses during the day,49 it was assumed that there would be differences between the low and high fluid intake regarding SA_{osm}.42 However, no differences in SA_{osm} were reported by Perrier et al.42 In this regard SA_{osm} was highly variable between participants as also shown in prior studies.28,36

During active heat exposure in the study by Muñoz et al.,37 BS_{osm} and SA_{osm} were the most effective hydration assessment markers (that is, high specificity and sensitivity). Further, for single measurements, BS_{osm} and SA_{osm} propose good usability during high temperature and exercise. For measurements over time BS_{osm}, UR_{ig} and BMc seem to be the most valid hydration assessment markers. In this regard, Cheuvront et al.28 suggest that BP_{osm}, UR_{ig} and BMc are appropriate markers during dynamic (monitoring over time) dehydration but only BP_{osm} (not SA_{osm}) as useful marker for static (one time) dehydration assessment. There were weak significant correlations reported between SA_{Na^+} and BP_{Na^+} (r = 0.45). Thus, the use of saliva provides limited support as a potential substitute for reporting changes in BP_{Na^+} in real time36 during exercise,37 probably because of reduced parasympathetic stimuli that alter secretion rates of saliva.37 However, the difference between BP_{Na^+} and SA_{Na^+} was approximately sevenfold.

**Sweat and hydration status**

In a hot environment or during exercise, body temperature is controlled by the evaporation of sweat. The deficit in electrolytes can be preserved by means of sodium reabsorption from the duct of sweat glands. Evidence supporting blood osmolality as a hydration assessment marker usually comes from studies that integrate a sweat-loss model of hypertonic-hypovolemia in young, fit and healthy individuals. In this regard, blood osmolality is unsuitable to detect isotonic-hypovolemia often following from illness and medications (for example, diuretics) in a clinical setting.47

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**Table 2. General characteristics of the included studies**

| Study                        | Dehydration procedure | Monitoring time (body mass) | Urine | Abs | Rel | Blood | Abs | Rel | Sweat |
|------------------------------|-----------------------|-----------------------------|-------|-----|-----|-------|-----|-----|-------|
| Hamouti et al.19             | Cycling (indoor) and fluid restriction | Baseline | UR_{osm}: 570 | —   | —   | BS_{osm}: 282 | —   | —   |       |
|                              |                       | BMc 2% | UR_{osm}: 760 | 190  | 33.3 | BS_{osm}: 292 | 10  | 103.5 | SW_{Na^+}: 65 |
|                              |                       | BMc 1.3% | UR_{osm}: 640 | 40  | 6.7 | BP_{osm}: 294 | 5   | 1.7  | SW_{Na^+}: 80 |
|                              |                       | BMc 1.7% | UR_{osm}: 640 | 40  | 6.7 | BP_{osm}: 296 | 9   | 3.1  | SW_{Na^+}: 80 |
| Hew-Butler et al.34          | Running (indoor) and fluid restriction | Baseline | UR_{osm}: 600 | 110 | —   | BP_{osm}: 289 | —   | —   |       |
|                              |                       | BMc 1.3% | UR_{osm}: 640 | 40  | 6.7 | BP_{osm}: 294 | 5   | 1.7  | SW_{Na^+}: 80 |
|                              |                       | BMc 1.7% | UR_{osm}: 640 | 40  | 6.7 | BP_{osm}: 296 | 9   | 3.1  | SW_{Na^+}: 80 |
| Morgan et al.40              | Cycling (indoor) and fluid restriction | Baseline (78.7) | UR_{osm}: 60 | 50  | 45.5 | BS_{osm}: 287 | —   | —   |       |
|                              |                       | Post 60 min | —   | —   | BS_{osm}: 292 | 5   | 1.7  |       |
|                              |                       | Post 120 min | —   | —   | BS_{osm}: 295 | 8   | 2.8  | SW_{osm}: 172 |
| Walsh et al.39               | Cycling (indoor) and fluid restriction | BMc: 1.8% | UR_{osm}: 9.9 | 60  | —   | BS_{osm}: 142 | 2   | 1.4  | SW_{Na^+}: 91.1 |

**Abbreviations:** Abs (absolute difference to baseline); BMc (body mass change, percentage change); BP_{osm}/BS_{osm} (blood plasma/serum osmolality, mOsmol/kg); BP_{Na^+}/BS_{Na^+} (blood plasma/serum sodium concentration, mmol/l); Rel (relative difference to baseline); SW_{osm} (sweat osmolality, mOsmol/kg), SW_{Na^+} (sweat sodium concentration, mmol/l); UR_{osm} (urine osmolality, mOsmol/kg); UR_{Na^+} (urine sodium concentration, mmol/l); UR_{ig} (urine-specific gravity, g/ml); values estimated out of figures; italic = athletes/soldiers.
exercise-induced dehydration, the calculated ECF volume by augmented activity of the sympathetic nervous system. During SW[Na+] were higher in a dehydrated state.40 When the subjects et al. proposed that differences between dehydration and euhydration resulting from the greater ECF[Na+] could not solely account for ~ 10 mmol/l. Therefore, a simple displacement into primary sweat, 3 mmol/l, which was lower than that found in sweat, captured electrolytes in clothes and/or collection area, or contamination from other solutions.

A further limitation of the collection of sweat is the missing baseline value. To compare a dehydrated with an euhydrated status, Morgan et al.49 tested participants ingesting either no fluid (dehydration) or a 20 mmol/l sodium chloride solution (euhydration) during exercise. They showed that dehydration caused an increase in SW[Na+] with regard to an euhydrated state. It is proposed that differences between dehydration and euhydration in ECF[Na+], acute aldosterone and sympathetic nervous activity could cause the changed sweat composition. Both BS[Na+] and SW[Na+] were higher in a dehydrated state.40 When the subjects were dehydrated due to higher BS[Na+]31, it is predicted by Morgan et al.40 that an increased sodium concentration would have appeared in the primary sweat. However, the difference between dehydration and euhydration for BS[Na+]31 was approximately 3 mmol/l, which was lower than that found in sweat, which was ~ 10 mmol/l. Therefore, a simple displacement into primary sweat, resulting from the greater ECF[Na+]31 could not solely account for the higher SW[Na+]. A second possible explanation for the increase in SW[Na+] during dehydration could have been an influence of elevated aldosterone on the secretory coil. A third explanation for the increase in SW[Na+] could also have been an augmented activity of the sympathetic nervous system. During exercise-induced dehydration, the calculated ECF volume by Hamouti et al.39 declined progressively from exercise-baseline value likely due to water losses through sweating while BSosmol increased. Furthermore, SW[Na+] losses, as a result of a higher SW [Na+] concentration, can significantly affect post-race BS[Na+] concentration in long-runners,50 and SW[Na+] did not reflect the same pattern as UR[Na+].34

Tear fluid status

Tear fluid is a complex solution intended to sustain the surface of the eye.51 The lacrimal gland secretes tear fluid composed mainly of water and electrolytes, and human tears have been disclosed to be isotonic with plasma.52 TEosm increased with dehydration and tracked changes in BPosmol and URsg, and therefore it might offer a new hydration assessment technique in rehabilitation, sport, military and performance-related activities. TEosm can record alterations in hydration status due to water consumption during progressive rehydration following exercise as well as differentiate between dehydration (2–3% BMc) and euhydration during exercise.27 It seems that BPosmol and TEosm have the strongest correlation over the widely used hydration assessments (eg. BMc, URsg).

It is suggested that a TEosm value > 309 mOsmol/l reflects dehydration.31 This value was not reached in the two included studies but increases in BPosmol during exercise-evoked dehydration27,31 and subsequent overnight fluid restriction31 were represented in increases in TEosm. The data indicate that TEosm was ~ 5–10 mOsmol higher than the respective BPosmol (Table 3). The BPosmol cutoff for minimal dehydration is 295 mOsmol/kg.14 Compared with the TEosm values, 301 mOsmol/l could be the cutoff value for a minimally dehydrated status. It has to be taken into account that TEosm was measured as osmolarity (number of osmoles of solute per liter of solution) and BPosmol was measured as osmolality (number of osmoles of solute per kilogram of solvent). Furthermore, in the response of TEosm to changes in hydration status, there are large differences among subjects limiting its validity and usefulness at the individual level. The potential usefulness of TEosm to estimate hydration status at the individual level has to be further determined as well as how its validity and reliability are impacted by field conditions.27 Nevertheless, it can be suggested that TEosm has utility as a marker of hydration status (strong correlation between TEosm and BP posmol: r = 0.93). The correlation between TEosm and BPosmol was even stronger than that between Ur and BPosmol (r = 0.72).31

Dehydration and hyperthermia

Trangmar and González-Alonso6 showed that progressive exercise-induced dehydration, with concomitant hyperthermia, can be associated with impaired perfusion to tissues and organs. In most included studies, the combination of exercise-induced dehydration and heat stress was presented, which makes it difficult to separate the effects of dehydration and hyperthermia in each compartment.53 It is well known that hyperthermia negatively influences endurance performance,24 but the effect on short-term high-intensity performance is still unclear.55 Thermoregulatory functions depend on sufficient body water. Consequently, losses in TBW can challenge the thermoregulatory system. A deficit of TBW with a BMc of ≥ 2% (dehydration) is the threshold for measurably altered thermoregulation.56

Recent evidence further complicates the assessment of hydration status, in that different hydration assessment markers may validly identify dehydration in one circumstance but not another.27,47

Limitations

When interpreting data, one should be aware of the relative small number of studies. Although there are many studies about (de) hydration, some aspects differ substantially such as dehydration

| Study | Dehydration procedure | Monitoring time (body mass) | Urine | Abs | Rel | Blood | Abs | Rel | Tear | Abs | Rel |
|-------|-----------------------|-----------------------------|-------|-----|-----|-------|-----|-----|------|-----|-----|
| Fortes et al.31 | Cycling (indoor) and fluid restriction | Baseline (68.1) | URsg°: 1.006 | — | — | BP osmol: 288 | — | — | TE osmol: 293 | — | — |
| BMc 1% | URsg°: 1.008 | 0.002 | BP osmol: 289 | 1 | 0.3 | BP osmol°: 299 | 6 | 2 |
| BMc 2% | URsg°: 1.017 | 0.011 | BP osmol: 292 | 3 | 1.4 | BP osmol°: 300 | 7 | 2.4 |
| BMc 3% | URsg°: 1.021 | 0.015 | BP osmol: 297 | 9 | 3.1 | BP osmol°: 305 | 12 | 4.1 |
| BMc (overnight) 3.5% | URsg°: 1.026 | 0.02 | BP osmol: 297 | 9 | 3.1 | BP osmol°: 304 | 11 | 3.8 |
| Ungaro et al.27 | Cycling (indoor) and fluid restriction | Baseline (75.7) | URsg°: 1.006 | — | — | BP osmol: 292 | — | — | BP osmol°: 296 | — | — |
| BMc 1% | URsg°: 1.012 | 0.006 | BP osmol: 293 | 1 | 0.3 | BP osmol°: 299 | 3 | 1 |
| BMc 2% | URsg°: 1.020 | 0.014 | BP osmol: 295 | 3 | 1 | BP osmol°: 301 | 5 | 1.7 |
| BMc 3% | URsg°: 1.021 | 0.015 | BP osmol: 297 | 5 | 1.7 | BP osmol°: 302 | 6 | 2 |

Abbreviations: Abs (absolute difference to baseline); BMc (body mass change, percentage change); BP osmol (blood plasma osmolality, mOsmol/kg); Rel (relative difference to baseline); TE osmol (tear osmolarity, mOsmol/l, euhydrated: < 310 mOsmol/l); URsg (urine-specific gravity, g/ml); values estimated out of figures°; italic = athletes.
procedures and used hydration assessment markers. First, to achieve a euhydration state before testing, all studies had to conduct a well-described hydration protocol and afterwards a well-described dehydration procedure (influenced by fluid restriction and/or physical activity). This limited the number of studies with tear fluid for example. Second, this review has focused on ‘freely accessible’ and direct evaluation of body fluids saliva, sweat and tear. Thus, saliva, sweat and tear could be directly compared to the other ‘standardized’ biochemical hydration assessment markers (see above) regarding osmosmolarity, osmomality and sodium concentration of body fluids. In particular, sweat as a hydration assessment marker was often indirectly evaluated.

CONCLUSION

In summary, the setting and the method of collecting respectively the body fluids (for example, blood) obtaining a sample of saliva is one of the simplest ways to collect body fluids. During exercise and heat exposures, saliva might be an effective index to evaluate hydration status but seems to be highly variable and should be carefully used as a substitute marker of other biochemical hydration assessment markers. The lack of a baseline measurement and the time-consuming collection of sweat makes it more difficult to evaluate dehydration and to make a statement about the hydration status at a particular time. The collection procedure of tears shows little discomfort to the participants and is easy to access. \( \text{TE}_{\text{osm}} \) can evaluate changes in hydration status and increase with dehydration and recorded changes in \( \text{BP}_{\text{osm}} \) with comparable utility to \( \text{UR}_{\text{cp}} \). But with only two included studies, it has to be further determined whether \( \text{TE}_{\text{osm}} \) is sensitive enough to evaluate dehydration at the individual level as its validity and reliability.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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AUTHOR CONTRIBUTIONS

Conceived and designed review: MV, RS, TV, EH, RC; analysed the data: MV, RS, TV, EH, MP, RC; wrote the paper: MV, RS, TV, EH, PC, FP, RC.

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