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# The influence of low and high daily fluid intake on the cortisol awakening response

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## Abstract

The cortisol awakening response (CAR), the sharp rise in cortisol level upon awakening, represents a unique aspect of hypothalamic pituitary adrenal (HPA) axis activity, combining features of circadian regulation and reactivity to awakening. Studies highlight that the CAR is influenced by state-like factors (e.g., subjective stress and poor sleep) and relates to health and ageing[(1)]. Fluid regulation and the CAR share a common pathway, whereby hydration directly influences the secretion of arginine-vasopressin (AVP), which in-turn modulates HPA-axis activity and cortisol release. Studies have observed higher circulating and saliva cortisol in healthy, low drinking adults presenting with suboptimal hydration (e.g., elevated urine osmolality, UOsm); however, cortisol was only assessed at a single timepoint[(2,3)]. The study aim was to assess the influence of a change in daily fluid intake on the dynamic CAR.

From a sample of 71 eligible healthy adults (25 females, 46 males), 16 low drinkers (1315 ± 401 ml/day) and 16 high drinkers (4372 ± 1220 ml/day) were identified by adopting daily fluid intake thresholds from a matched UK population [(4)]. In pairs, comprising a low and high drinker, participants underwent a 7-day habitual phase followed by a 7-day intervention phase. During the intervention phase low drinkers increased (+1953 ± 430 ml/day) and high drinkers reduced daily fluid intake (-3168 ± 1379 ml/day), adjusting only water intake. Saliva samples were collected at 0, 15, 30, and 45 minutes after awakening on the last two mornings of each phase, with compliance to sample timing (± 5 minutes) monitored as recommended [(1)]. Saliva cortisol was assessed by ELISA and the CAR was reported as the area under the curve with respect to increase ($AUC_I$) [(1)]. Urine samples were collected the day before and the day of CAR sample collection (16:00-20:00) to assess UOsm.

Linear mixed model revealed a group (low drinkers, high drinkers)*phase (habitual drinking, intervention drinking) interaction for hydration ($P < 0.01$), whereby UOsm changed significantly in both low drinkers (mean ± SD: 582 ± 213 to 363 ± 200 mOsm/kg) and high drinkers (265 ± 153 to 612 ± 189 mOsm/kg), indicating the success of the intervention. However, no group*phase interaction was observed for the CAR ($P = 0.9$). The CAR was similar before and after the intervention in both low ($AUC_I$: 15 ± 15 *vs.* 16 ± 8) and high drinkers ($AUC_I$: 11 ± 14 *vs.* 12 ± 11), indicating that changing fluid intake did not affect the CAR. These findings remained when observations were restricted to participants who accurately complied to the CAR sampling protocol ($N = 19$).

These results suggest that the cortisol awakening response, which combines elements of circadian regulation and reactivity to awakening, is not influenced by daily fluid intake.

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