The physiological effects of daily Cold-water immersion on 5-day tournament 1 performance in international standard youth field-hockey players 2 3 **Authors:** Malte Krueger^{1, 2}; Joseph T Costello³; Mirko Stenzel^{2, 4}; Joachim Mester²; 4 Patrick Wahl^{2, 5} 5 6 Institutions: 7 8 ¹ Institute of Cardiology and Sports Medicine - Department of Preventative and Rehabilitative Sports and Performance Medicine, German Sport University Cologne, 9 Cologne, Germany 10 ² The German Research Centre of Elite Sport, German Sport University Cologne, 11 Cologne, Germany 12 ³ Extreme Environments Laboratory, Department of Sport and Exercise Science, 13 University of Portsmouth, Portsmouth, UK 14 ⁴ German Hockey Federation, Mönchengladbach, Germany 15 ⁵ Institute of Cardiology and Sports Medicine – Department of Molecular and Cellular 16 17 Sports Medicine, German Sport University Cologne, Cologne, Germany 18 19 **Corresponding author** 20 21 Malte Krueger 22 German Sport University Cologne Institute of Cardiology and Sports Medicine, Depart. of Preventative and Rehabilitative 23 Sports and Performance Medicine 24 Am Sportpark Müngersdorf 6, 50933 Cologne, Germany 25 26 E-mail: malte.krueger@bayer04.de Phone: +49-(0)221-4982 6062 27 Abstract 28

PURPOSE: This study examined the effects of daily post-exercise cold-water immersion
(CWI) on match performance, perceptual recovery, and biomarkers of muscle damage
and metabolic load during a 5-day international tournament of elite youth field-hockey
players.

METHODS: The entire German under-18 national squad (n=18) were randomly assigned 33 to a daily CWI- (5-min at \sim 6°C; excluding the head; n=9) or passive recovery (CON; n=9) 34 intervention. Training- and match-performance was assessed using a GPS-tracking 35 system and perceived exertion (RPE). Daily ratings of delayed onset muscle soreness 36 (DOMS), perceived stress and recovery, quality of sleep, heart-rate recovery (HRR) and 37 serum creatinekinase (CK), lactate-dehydrogenase, and urea nitrogen were also 38 recorded. Repeated sprint ability (RSA) and counter-movement jump (CMJ) was carried 39 out on day 1 and 5. 40

RESULTS: There was no significant between intervention differences in time on pitch, total distance, velocity-zones and accelerometer-bases parameters during match performance (all p > 0.05). DOMS (p < 0.01), RPE (p < 0.01), and CK (p < 0.01) were significantly elevated over the course of the tournament; however, no between intervention effects were observed (all p > 0.05). Both groups were able to maintain RSA and CMJ (all p > 0.05).

47 CONCLUSION: In conclusion, daily post-exercise CWI did not improve match 48 performance, perceptual recovery, or biomarkers of muscle damage and metabolic load 49 in elite youth field-hockey players.

50 **Keywords:** Recovery, DOMS, GPS-tracking, Elite athletes, Biomarkers

51	Abbreviati	ons:
52	ACC	Accelerations
53	ANOVA	Analysis of variance
54	BUN	Blood urea nitrogen
55	СК	Creatinekinase
56	CMJ	Counter-movement jump
57	CON	Control
58	CWI	Cold-water immersion
59	DEC	Decelerations
60	DOMS	delayed onset muscle soreness
61	ExE	Explosive efforts
62	GPS	Global positioning system
63	HR _{ex}	Exercise heart rate
64	HRR	Heart rate recovery
65	HSD	High-speed distance
66	LDH	Lactate- dehydrogenase
67	RPE	Rating of perceived exertion
68	RSA	Repeated sprint ability
69	TD	Total distance
70	T _{skin}	Skin temperature
71	VZ	Velocity zone
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76	Introductio	on

In elite international field hockey tournaments, players are required to play and achieve 77 their best performance possible almost daily, with the first 3 matches often scheduled 78 within 4 days (Jennings et al. 2012). Repeating high-intensity exercise with limited time 79 for recovery may lead to accumulated fatigue and consequently, reduced performance 80 capacity (Spencer et al. 2005, Rowsell et al. 2011, Clarkson and Hubal 2002). Reductions 81 in exercise duration and intensity for 1-2 days, following high-intensity exercise as 82 recommended (Cheung et al. 2003), are not always possible due to the tournaments 83 schedule. Therefore, protocols to enhancing recovery or reducing fatigue between 84 85 matches are an important factor for success and the players' ability to maintain performance throughout the tournament. 86

The use of time-motion analysis and GPS-tracking devices is widely spread to quantify 87 physical activity in team sports (Lythe and Kilding 2011, White and MacFarlane 2013, 88 White and MacFarlane 2015, Cummins et al. 2013). It has been well demonstrated that 89 decrements in total running distance (TD) and high-speed running distance (HSD) occur 90 throughout team-sport tournaments (e.g. soccer and rugby), potentially indicating 91 accumulated fatigue (Rowsell et al. 2011, Johnston et al. 2013). In elite field hockey, 92 findings are inconsistent as some studies reported reductions in TD, HSD and number of 93 sprints (Spencer et al. 2005, Ihsan et al. 2017), while another stated no changes 94 (Jennings et al. 2012). However, it is well established that there is a large variability in 95 physical match performance in elite teams, which is likely attributed to technical and 96 tactical requirements (Gregson et al. 2010). Thus, players may not be required to reach 97 their physiological limits and decrements in certain recovery parameters are not 98 necessarily associated with accumulated fatigue. 99

100 Therefore, various surrogate outcome measures are used to monitor fatigue throughout consecutive training and competition, such as counter-movement jumps (CMJ), repeated 101 sprint ability (RSA) or heart rate recovery tests (HRR) (Rowsell et al. 2009, Buchheit et 102 al. 2013, Thorpe et al. 2015). Moreover, perturbations of various biomarkers related to 103 exercise-induced muscle damage and inflammation are frequently assessed to quantify 104 105 the athletes load and recovery status during training and competition (Meyer and Meister 2011). Additionally, the players' internal load can be assessed using psychometric 106 questionnaires related to delayed onset muscle soreness (DOMS), freshness, quality of 107 108 sleep, fatigue and stress (Buchheit et al. 2013, Hitzschke et al. 2015, Williamson and Hoggart 2005). In order to get a better understanding of any potential impact of an 109 recovery-intervention, it has been recommended to use multiple parameters and 110 111 assessments to quantify performance, fatigue and recovery (Halson 2014, Kellmann et al. 2018). 112

Post-exercise cooling is a common recovery modality for prevention and treatment of 113 exercise-induced fatigue (Poppendieck et al. 2013, Hohenauer et al. 2015). Among the 114 most common methods are brief exposures to cold water (cold-water immersion, CWI, ~ 115 5-13°C for 5-20 min), extremely cold air (whole-body cryotherapy, WBC, ~ -110°C for 2 116 to 4 min) or extremely cold vaporized liquid nitrogen (partial- body cryotherapy, PBC, -117 110-195°C for 1-3 min) (Bleakley et al. 2012, Costello et al. 2015, Bouzigon et al. 2016, 118 Leeder et al. 2012). Despite its increasing popularity and application in sports, evidence 119 that cryotherapy improves recovery is equivocal (Murray and Cardinale 2015, 120 Poppendieck et al. 2013, Leeder et al. 2012). Several reviews of the literature suggest, 121 that by its primary effects of reducing tissue temperature, peripheral blood flow, and nerve 122

conductance velocity, cryotherapy attenuates the perception of exercise-induced muscle 123 damage and DOMS (Bleakley et al. 2012, Hohenauer et al. 2015, Ihsan et al. 2016). 124 However, concurrent findings are published regarding the effects of cryotherapy on 125 biomarkers of muscle damage and inflammation (Ascensao et al. 2011, Pournot et al. 126 2011, Ingram et al. 2009, Krueger et al. 2018). These disparate results might be related 127 to differences in methodology, such as cooling temperature, immersion depth, initial 128 exercise modality and time points of recovery parameter assessment. In addition, it has 129 been suggested that CWI is most effective after prolonged whole body endurance and/or 130 131 intermittent based exercise (Bleakley et al. 2012, Ihsan et al. 2016). However, most studies focused on single exercise bouts and single applications of CWI in recreational 132 trained participants, thus data of daily CWI applications following repetitive high-intensity 133 training and match performance in international level athletes is extremely limited. To our 134 knowledge, no study has investigated the effects of CWI compared to a control condition 135 on 1) match performance and 2) various recovery parameters during a tournament in elite 136 field hockey to date. 137

Accordingly, the aim of the present study was to investigate the effectiveness of daily post-exercise CWI on physiological and perceptual markers of fatigue and recovery and physical match-performance during a 5-day tournament in international standard youth field-hockey players. We hypothesised that 1) CWI would not improve performancerelated parameters or biomarkers of muscle damage or fatigue, and 2) that CWI would improve perceptual markers associated with fatigue and recovery.

145 Methods

146 **Participants**

147 The under-18 German national Squad in field hockey participated in the present study. The team won the European Championship in the in the year of the present studies 148 conduction. During the investigated tournament the national squad consisted of 18 149 150 healthy male field hockey players (mean \pm SD: age 16.6 \pm 0.6 yrs.; height 182.1 \pm 5.5; mass 73.8 ± 7.8 kg). All participants played field hockey for at least five years with three 151 152 or more training sessions per week and regular league matches in their respective home clubs. Additionally, they were familiar with different recovery methods like message or 153 CWI. Before the tournament, players were informed about the testing procedures and 154 155 potential risks and familiarized with the questionnaires used during the investigation. After that, all volunteers and their parents or legal guardians gave their written informed 156 consent to participate. The study, including all procedures was conducted in accordance 157 with the Declaration of Helsinki and approved by the ethical committee of the German 158 Sport University Cologne. 159

160 Experimental design

161 An overview of the experimental design is presented in figure 1.

Briefly, the tournament took place immediately after the last week of the regular season with players arriving from their respective home clubs in the morning of day 1. After anthropometric measurements and baseline assessment of questionnaires and biomarkers, the team completed a 15-min warm-up, followed by performance testing of CMJ and 6 x 30m RSA. Afterwards, the players were paired by the coaches according to

their playing position, level of experience and anthropometrics. Randomly (coin toss) one 167 player of each pair was assigned to the intervention (CWI) and control (CON) group, 168 respectively. In the afternoon of day 1 a friendly match against a local club was carried 169 out, consisting of 3 x 20 min. On day 2 tactical coaching and a 2 h – training session was 170 conducted. On days 3-5, the team participated in an international tournament with one 171 match per day. In the afternoon of day 5 assessment of CMJ and RSA performance was 172 repeated. Prior to the experiment, the coach had requested that all players would play the 173 same amount of time on the pitch and cover an similar distance. Interchanges were 174 175 conducted accordingly to achieve this during the matches.

176

*** Figure 1 near here****

177 Performance tests (RSA, CMJ)

178 CMJ-height was measured using the Optojump photoelectric cell system (Microgate, Bolzano, Italy) as previously described (Glatthorn et al. 2011). Players were advised to 179 place their hands at the hips to avoid additional arm swing. The best of three maximal 180 effort jumps was taken for statistical analysis. The RSA-test consisted of 6 maximal 30m 181 efforts, starting every 25 sec as recommended for field hockey players and described 182 elsewhere (Tanner and Gore 2013). Sprint time was assessed using a dual beam sports 183 timing system (Swiftperformance, Wacol, Australia). Gates were placed at the start, 10m, 184 20m and 30m. Total sprint time was calculated using the sum of all six sprints (total sprint 185 186 time). The test-retest coefficient of variation for youth male team-sport athletes tested in our laboratory is 2.8% for CMJ and 1.1% for RSA. 187

188 Physical Performance Monitoring during match and training

The physical activity of each match and training session was monitored using a portable 189 positional tracking device (10hz GPS and GLONASS) with integrated 3D-Accelerometer, 190 3D-Gyroscope and 3D-Magnetometer using a sampling rate of 100hz, respectively 191 (OptimEye S5, Catapult sports, Melbourne, Australia). Each player's individually assigned 192 device was worn at the upper back between the left and right scapulae in a custom-made 193 vest (Catapult sports). All devices were turned on at least 30 min before exercise for 194 satellite connection. During data collection the average number of satellites and horizontal 195 dilution of precision per unit (mean \pm SD) were 12.0 \pm 0.0 and 0.61 \pm 0.01, indicating an 196 197 almost ideal signal quality (Malone et al. 2017). During matches, the players' rotations were precisely documented. After recording, data was downloaded, analysed and 198 reported using the appending software (OpenField v1.17.0). Goalkeepers were excluded 199 200 from statistical analysis. Bench time of the field players was excluded and values calculated for time on pitch only, as described previously and recommended for time-201 depended variables in field hockey (White and MacFarlane 2013, Ihsan et al. 2017). 202 Thresholds for six velocity zones (VZ) were matched with previously published 203 investigations in field hockey (VZ1, < 6 km \cdot h⁻¹; VZ2, 6 – 11 km \cdot h⁻¹; VZ3, 11 – 15 km \cdot h⁻¹; 204 VZ4, 15 – 19 km·h⁻¹; VZ5, 19 – 23 km·h⁻¹; VZ6, > 23 km·h⁻¹) (White and MacFarlane 205 2015, White and MacFarlane 2013, Lythe and Kilding 2011). The time spend in each VZ 206 was calculated as percentage of time on pitch. HSD was measured for speeds above 19 207 km·h⁻¹. The software's inertial movement analysis was used to detect accelerations 208 (ACC) and decelerations (DEC) above 2.5 m·s⁻² and explosive efforts (ExE), defined as 209 the sum of accelerations, decelerations, changes of direction to the left and right above 210 3.5 m·s⁻². TD, HSD, ACC, DEC and ExE were normalised to one minute for comparison. 211

30 min after the end of each exercise, players were asked to rate their perceived exertion of the previous session (RPE_{session}) on a 1 - 10 Borg scale (Foster et al. 2001).

214 Cold-water immersion and passive recovery

15 minutes after each match or training on days 1 - 4 the CWI-group fully immersed 215 themselves into an ice-water barrel for 5 minutes, with only the head out of the water. The 216 barrels were prepared with crushed ice to a water temperature between 5 and 8° C (6.4 ± 217 0.8°C). During immersion, the temperature was frequently controlled and ice added if 218 necessary. Players were instructed to slowly move their arms and legs to avoid the 219 formation of warmer water layers around the body. Similar protocols have been used 220 extensively elsewhere (Ingram et al. 2009, Rowsell et al. 2009, Costello et al. 2012). 221 222 During the same time, the CON-group remained passive and seated at room temperature. Afterwards, the whole team continued with the schedule of the tournament together. 223

224 Mean Skin temperature

Skin temperature was assessed by means of a thermoimaging camera (Testo 880, Testo GmbH, Vienna, Austria) as described elsewhere (Moreira et al. 2017). Photos were taken of the trunk, thigh and lower arm before and after CWI and analysed using the appending software (IRsoft Version 3.1 SP3, Testo GmbH, Vienna, Austria). Mean skin temperature (T_{skin}) was calculated using the Burton (Burton 1935) equation: $T_{skin} = 0.5 T_{trunk} + 0.36$ $T_{thigh} + 0.14 T_{arm}$.

231 Biomarkers

Every morning 300 µL of capillary blood was collected from the earlobe of each athlete, 232 transferred in a heparinized centrifuge tube, centrifuged for 2 minutes and immediately 233 analysed for serum levels in Creatinkinase (CK), Lactate-Dehydrogenase (LDH) and 234 Blood Urea Nitrogen (BUN) using a mobile Point-of-care device (Spotchem EZ SP-4430, 235 Axonlab, Baden-Daettwil, Germany). Serum levels were determined by optical 236 237 measurement of reflection intensity (at 550 or 610 nm) for a reagent colour reaction on three single reagent strips. The reagent strip is composed of a multi-layered test field 238 containing reagents necessary to generate a colour that is quantified by reflectance 239 240 spectrophotometry. The precision (CV %) of consecutive replicate measurements was 3.8% for CK (94.7 U·L⁻¹), 3.1 % for BUN (6.1 mmol·L⁻¹) and 3.7% for LDH (322.9 U·L⁻¹). 241

242 **Psychological questionnaires, quality of sleep and Pain scale**

Every morning before breakfast, the players answered a questionnaire assessing their acute state of recovery and stress. To avoid misunderstanding and translational problems a questionnaire in German language was chosen, that has already been validated in field hockey (Hitzschke et al. 2015). The items of the "Short Recovery and Stress Scale for Sport" were rated on a scale from 0 ("not at all") to 6 ("absolutely"). Furthermore, players were asked to rate the quality of last nights' sleep on a scale from 1 ("very good") to 6 ("very bad").

Moreover, DOMS was measured every morning and evening, using a 100 mm visual analog scale, ranging from 0 ("no soreness") to 100 ("severe soreness") (Williamson and Hoggart 2005). The players rated their individual DOMS after slowly sitting down and standing up from a chair (knee angle approximately 90°).

254 Heart rate recovery test

A heart rate recovery test was included into the warm-up session before each match or 255 training. On an indoor pitch (length: 50m) the entire team ran for 5 min at a constant speed 256 of 9 km/h, controlled by a digital metronome, followed by sitting in a relaxed position for 257 another 5 min. Heart rate was recorded continuously using a mobile team system 258 (Acentas GmbH, Hörgertshausen, Germany). Exercise heart rate (HR_{ex}), defined as the 259 average heart rate during the last 30 seconds and heart rate recovery (HRR), defined as 260 the difference between HRex and heart rate after one minute of recovery were calculated 261 as described elsewhere (Buchheit 2014, Borresen and Lambert 2007). HRex was 262 263 expressed as percentage of the players' individual maximal HR, HRR was expressed as percentage of HR_{ex}. 264

265 Statistical analysis

All statistical tests were carried out using the Statistica software package for Windows® 266 (version 13.0, StatSoft Inc., Tulsa, OK, U.S.A). The distribution of data was assessed 267 using descriptive methods (skewness, outliers, and distribution plots) and inferential 268 statistics (Shapiro-Wilk test). Normal distribution was observed for all parameters except 269 CK. Normally distributed data is presented as mean values ± standard deviations (SD). 270 and mean differences (MD) with 95% confidence intervals for performance tests and 271 distribution of velocity zones. Data deviating from normal distribution is presented as 272 median and inner-quartile range (Q₂₅ and Q₇₅). A two way (treatment [CWI, CON] * time 273 [Days 1-5]) repeated-measures analysis of variance (ANOVA) was applied to compare 274 all biomarkers (CK, LDH, BUN), Questionnaires (perceived recovery, perceived stress, 275

quality of sleep, DOMS), heart rate recovery tests (HR_{ex}, HRR), performance tests (RSA, CMJ) and tracking data (time on pitch, total distance, normalised distance, HSD, ACC, DEC and ExE). If main effects for time, intervention or interaction were identified, Bonferroni post-hoc analysis was applied where appropriate. Student paired t-tests were used for analysis of percentage time spend in each velocity zone during tournament matches (days 3-5) as well as the effect of CWI on T_{skin} (pre vs. post). Statistical significance was accepted at p < 0.05.

283

284 **Results**

285 Tracking data

All GPS and accelerometer-based parameters are presented in Table 1 and 2 (means \pm SD). All parameters (time on pitch, TD, normalised distance, normalised HSD, normalised DEC) except normalised ACC (p = 0.21) and normalised ExE (p = 0.62) significantly increased from day 1 to day 5 (p < 0.01). However, this is most likely related to the extended training duration on day 2. There were no significant intervention or interaction effects (all p > 0.05) (Table 1). During tournament matches there were also no differences (all p > 0.05) between CWI and CON in time spent in each velocity zone (Table 2).

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*** Table 1 and 2 near here****

294 **Performance tests**

There were no time, interaction or intervention effects (all p > 0.05) in RSA total sprint time (mean ± SD; mean difference [95% CI]) from day 1 (CWI: 26.23 ± 1.06 s, CON: 26.05 ± 0.69 s; MD: 0.18 [-0.78 – 1.14]) to day 5 (CWI: 26.37 ± 1.06 s, CON: 26.34 ± 0.73 s; MD: 0.04 [-0.86 – 0.92]). The same results (all p > 0.05) were observed in CMJ with similarly no differences from day 1 (CON: 42.06-± 3.48 cm, CWI: 41.37 ± 5.31 cm; MD: 0.69 [-3.71 – 5.09])to day 5 (CON: 41.82 ± 3.64 cm, CWI: 41.59 ± 4.61 cm; MD: 0.23 [-3.84 – 4.30]), respectively.

302 Heart rate recovery / Questionnaires

The results for the daily measurements of the heart rate recovery and questionnaires are detailed in Table 3. Throughout the course of the tournament RPE_{session} and perceived stress increased, while perceived recovery, HR_{ex} and HRR decreased (all p < 0.05). However, no significant intervention or interaction effects were observed in heart rate recovery- or in questionnaire data (all p > 0.05). DOMS (Figure 2) increased over time (p < 0.01), but no intervention (p = 0.09) and interaction (p = 0.41) effect was evident.

309 *** Table 3 and Figure 2 near here****

310 Biomarkers

Serum biomarkers data for CK, LDH and BUN is presented in Figure 3. CK increased over time (p < 0.01), but no other significant differences were observed (all p > 0.05).

313 *** Figure 3 near here****

314 CWI - Mean skin temperature

T_{skin} was significantly reduced (p < 0.01; d = 11.5) from 29.8 \pm 1.3 °C to 14.9 \pm 1.3 °C immediately after each application of CWI.

317

318 Discussion

319 The present study is the first to delineate the effects of daily post exercise CWI on performance capacity, biomarkers of muscle damage and metabolic load, fatigue, and 320 recovery during a 5-day junior elite field hockey tournament. The main findings of the 321 present study are: (1) in support of our hypothesis, daily CWI did not alter performance 322 parameters during consecutive matches, (2) regardless of recovery intervention, elite 323 youth field hockey players were able to maintain match-to-match performance, (3) 324 exercise-induced alterations in biomarkers were not attenuated by CWI, and (4) contrary 325 to our second hypothesis, CWI did not improve perceptual recovery. Collectively, these 326 327 data suggest that daily post exercise CWI has no beneficial effects during a 5-day 328 tournament and therefore its usage is not recommended in international standard male junior field hockey players 329

It has previously been reported that CWI might be able to reduce fatigue associated 330 331 decrements in physical performance (i.e. TD and HSD) during consecutive tournament soccer matches in junior males (Rowsell et al. 2011). The findings of the current study 332 suggest that these benefits may not be extended to field hockey (Table 1 and 2), as elite 333 youth athletes were able to maintain their physical match performance even without the 334 use of CWI. These contrasting findings might be explained by the large differences in 335 336 running characteristics observed between both sports. Total duration is less in field hockey (~ 30 min) and unlimited interchanges allow individual recovery breaks and higher 337 intensities on the pitch (Spencer et al. 2005). However, the present data suggests that 338

the repeated high intensity exercise throughout the tournament did not lead to reduced 339 match running performance or exercise intensity. Distribution of percentage durations per 340 velocity zones was constant over the course of the tournament and significant time effects 341 in time on pitch, TD and normalised distance, HSD and DEC were caused by day 2 342 training session. These results corroborate the findings of Jennings et al. (Jennings et al. 343 2012) and Ihsan et al (Ihsan et al. 2017), who also reported that elite hockey players were 344 able to maintain exercise intensity when playing six matches in nine days. Contrary, 345 Spencer and colleagues (Spencer et al. 2005) described a decrease in jogging and 346 347 increase in walking duration in three subsequent field-hockey matches within four days. It must be acknowledged that match performance variables may not reflect the true status 348 of recovery, as tactical requirements may result in high between match variability 349 (Gregson et al. 2010). Regardless, it appears likely that young elite field hockey players 350 are able to maintain their physical performance throughout consecutive matches. 351

This is further supported by the results of the jump and repeated sprint performance 352 outcomes (CMJ and RSA). These tests have previously been used to monitor fatigue and 353 354 recovery state in team sports, as alterations are associated with training and match performance (Thorpe et al. 2015, Buchheit et al. 2013, Claudino et al. 2017, Ingram et al. 355 2009). In contrast to observations in other team sport tournaments (Rowsell et al. 2009, 356 Montgomery et al. 2008), performance in CMJ remained consistent from day 1 to day 5 357 and RSA was only reduced by ~1%, with no differences between CWI and CON-group. 358 These results indicate that the 5-day tournament did not induce accumulated 359 neuromuscular fatigue and CWI had no impact on athletic performance. 360

Significant reductions in HRex and HRR were observed in the present study in the CWI-361 and CON-groups, with the lowest values recorded on day 3 and 4. This is counterintuitive, 362 as lower HRex and HRR (when expressed as %HRex) is typically associated with improved 363 fitness and reduced fatigue (Daanen et al. 2012, Buchheit et al. 2013). Comparable data 364 have been explained as demonstrating symptoms of overreaching (Borresen and 365 366 Lambert 2007); however, this is very unlikely in the present study as physical performance was maintained throughout the tournament. Further, these international standard players 367 were highly trained prior to the start of the tournament. CWI has been proposed to 368 369 increase parasympathetic activity along with cardiac output (Ihsan et al. 2016), consequently lower HRex and HRR could have been expected in the CWI-group. 370 However, CWI did not induce changes in exercise- induced alterations of heart-rate 371 derived variables compared to CON. 372

Post-exercise ratings of perceived exertion (RPEsession) increased from day 1 to 4 in both 373 groups and slightly dropped on day 5 (Table 3). As performance values and exercise 374 intensity were on the same level throughout tournament matches, this increase might be 375 an indicator for increasing perceptual fatigue as shown during a 5-day soccer tournament 376 as well (Rowsell et al. 2009). DOMS also accumulated throughout the course of the 377 tournament. Days 3-5 showed significantly elevated levels compared to baseline, 378 however, there were no intervention or interaction effects in this field based analysis 379 (Figure 2). Even though some studies (Pournot et al. 2011, Corbett et al. 2012) reported 380 no effects of CWI on DOMS as well, several empirical laboratory and field based studies 381 (Ascensao et al. 2011, Bailey et al. 2007, Rowsell et al. 2009, Ingram et al. 2009) and 382 reviews (Ihsan et al. 2016, Hohenauer et al. 2015, Bleakley et al. 2012, Leeder et al. 383

2012) suggest that post exercise CWI is effective in reducing DOMS. These findings may 384 be explained by the training status of the international level athletes, which one would 385 expect to result in relatively limited DOMS observed in the current study. Furthermore, 386 DOMS typically peaks ~48 hrs after exercise (Goodall and Howatson 2008), however the 387 team was required to daily, thus the time course for peak DOMS of a single exercise is 388 389 difficult to identify. Moreover, as exercise itself has been shown to reduce DOMS (Corbett et al. 2012), it is plausible that the exercise may have led to an attenuation of DOMS in 390 the CON- group. Perceived stress and recovery remained similar throughout the 391 392 tournament and only increased from day 4 to day 5. As the final match decided about winning the tournament, these time effects are most likely related to this. The exercise 393 itself and the recovery modality had no influence on the players' ratings of perceived 394 stress, recovery, or quality of sleep (Table 3). 395

Findings regarding the effects of CWI on exercise- induced alterations of biomarkers 396 associated with muscle damage and inflammation are contradictory in the literature [for 397 review see (Leeder et al. 2012, Bleakley et al. 2012, Hohenauer et al. 2015, Ihsan et al. 398 2016)]. For example, several studies have demonstrated decreased levels of CK, 399 Myoglobin or C-reactive Protein compared to thermoneutral or contrast water immersion 400 and passive control (Ascensao et al. 2011, Pournot et al. 2011, Bailey et al. 2007) while 401 others reported no change (Ingram et al. 2009, Pointon et al. 2012, Corbett et al. 2012). 402 In line with the current investigation Rowsell at al. (Rowsell et al. 2009) found no effect 403 on CK-clearance following the use of CWI after 5-day soccer tournament. These finding 404 corroborates the current results as CK increased from day 1 to 2 and remained elevated 405 throughout the course of the tournament, indicating high muscular load in both groups; 406

407 yet, CWI did not ameliorate the elevation in CK. However, no changes in LDH or BUN 408 were observed (Figure 3). Similar differences in CK and LDH responses to exercise have 409 been reported before and are potentially attributed to the different structural areas where 410 they are sequestered within the muscle sarcomere (Pournot et al. 2011). Nevertheless, it 411 could be argued that exercise intensity over the course of the five days was not severe 412 enough to induce a higher protein turnover rate, which would have been indicated by 413 elevated levels in BUN (Meyer and Meister 2011).

The present study has limitations that warrant mention. Firstly, we acknowledge the small 414 sample size of n=18. However, the entire national squad participated in the study; 415 416 therefore, this sample represent the largest number of international level athletes we were able to recruit. Secondly, as indicated by elevated levels in CK, DOMS and stress at 417 baseline, the athletes did not commence the tournament in a completely 'rested' state. 418 419 This is not surprising and reflects the intense schedule of elite junior team sport athletes that participate in tournaments or international competitions alongside, or immediately 420 after, their regular season. Thirdly, performance data (i.e. CMJ and RSA) was assessed 421 after the athletes arrived on site (circa 10 am) on day 1 and on day 5 three hours following 422 the final match of the tournament (circa 4 pm). We acknowledge that the data may be 423 reflective of this; however, again this represents the challenge of conducting applied 424 research on international athletes and working within their time schedules. However, both 425 the CWI and the control groups were randomised and we do not believe this invalidates 426 our findings regarding the effectiveness, or lack thereof, of the CWI intervention. Finally, 427 we acknowledge that additional follow up data after the tournament (i.e. day 6 and 7) 428

would have added additional insights, but this was not feasible as the athletes returnedhome, all across the country, after the tournament.

431

432 Conclusion

433 Despite elevated levels of CK, RPE and DOMS, several parameters (i.e. LDH, BUN, perceived recovery and stress, quality of sleep) were not altered over the course of the 434 5-day tournament. This suggests that there was an absence of severe cumulative fatigue 435 and/or the international standard athletes were sufficiently capable of completing and 436 437 recovering from the intensive training load, as indicated by maintained physical performance. The results of the present study also indicate that CWI had no beneficial 438 effects on physical performance, perceptual wellbeing, or biomarkers associated with 439 440 muscle damage compared to passive recovery. In conclusion, the application of daily CWI appears to be superfluous and we do not recommend its use in elite junior field-hockey 441 players during a 5-day tournament. 442

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448

449 **Disclosure statement**

450 The authors confirm there are no conflicts of interest.

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459	FIGURE CAPTIONS
460	Fig. 1 Schematic presentation of the study design. CWI: Cold-water immersion; RSA:
461	Repeated sprint ability; CMJ: Counter movement jump; DOMS: rating of delayed onset
462	muscle soreness
463	Fig. 2 Perceived ratings (mean \pm SD) of delayed onset muscle soreness (DOMS) in the
464	morning (pre) and evening (post) of each day (Days 1-5). * P < 0.05 time effects compared
465	to baseline (day 1 pre), for both interventions (cold-water immersion [CWI] and control
466	[CON]) combined
467	Fig. 3 Serum concentrations of Creatinkinase (CK) (median ± inner quartile range),
468	Lactate Dehydrogenase (LDH) and Blood Urea Nitrogen (BUN) (mean ± SD) in the

469	morning of each da	v (Da	vs 1-5), * P < 0.05 time effects compared to baseline	(dav	1).	. for
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both interventions (cold-water immersion [CWI] and control [CON]) combined

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Variable	Intervention	Day 1 – FM	Day 2 – TR	Day 3 – TM 1	Day 4 – TM 2	Day 5 – TM 3	p-value			
							[time]	[int.]	[time*int.]	
Time on pitch	CWI	46.6 ± 10.4	120.0 ± 0.0	53.5 ± 6.0	46.5 ± 11.1	51.0 ± 5.4	< 0.01	0.87	0.61	
[min]	CON	42.3 ± 5.4	120.0 ± 0.0	51.5 ± 6.4	47.6 ± 8.7	54.2 ± 8.9				
Total distance	CWI	5487 ± 1013	6883 ± 690	6193 ± 800	5614 ± 1408	5765 ± 614	< 0.01	0.81	0.70	
[m]	CON	5146 ± 697	6990 ± 394	5940 ± 956	5337 ± 976	6185 ± 1082	0.01	0.01	0170	
Normalised distance	CWI	119.3 ± 11.0	57.4 ± 5.7	115.8 ± 7.2	120.5 ± 6.2	113.3 ± 7.0	< 0.01	0.82	0.24	
[m∙min⁻¹]	CON	121.9 ± 7.6	58.2 ± 3.3	115.3 ± 12.8	112.8 ± 11.8	114.0 ± 10.6	0.01			
Normalised HSD	CWI	8.86 ± 4.63	2.16 ± 1.02	7.61 ± 3.81	10.02 ± 4.63	8.40 ± 2.60	< 0.01	0.65	0.83	
[n·min⁻¹]	CON	10.32 ± 2.14	2.38 ± 0.74	8.56 ± 2.27	10.43 ± 3.14	7.60 ± 1.79	< 0.01			
Normalised ACC	CWI	0.29 ± 0.09	0.22 ± 0.06	0.25 ± 0.07	0.25 ± 0.13	0.18 ± 0.05	0.21	0.10	0.54	
[n∙min⁻¹]	CON	0.19 ± 0.06	0.17 ± 0.07	0.13 ± 0.07	0.13 ± 0.07	0.23 ± 0.08	0.21	0.110		
Normalised DEC	CWI	0.35 ± 0.17	0.14 ± 0.08	0.33 ± 0.16	0.38 ± 0.10	0.30 ± 0.12	< 0.01	0.63	0.42	
[n∙min⁻¹]	CON	0.28 ± 0.12	0.18 ± 0.08	0.31 ± 0.11	0.31 ± 0.10	0.32 ± 0.09	0.01	0.00	0112	
Normalised ExE	CWI	0.42 ± 0.12	0.35 ± 0.08	0.46 ± 0.15	0.37 ± 0.09	0.32 ± 0.08	0.62	0 19	0.52	
[n∙min⁻¹]	CON	0.29 ± 0.08	0.33 ± 0.08	0.32 ± 0.12	0.41 ± 0.17	0.39 ± 0.10	0.02	0.17	0.32	

Table 1 Comparison of external load parameters (mean ± SD) between recovery modalities cold-water immersion (CWI) and control (CON)

624 FM: friendly match; TR: training; TM: tournament match; HSD: high speed distance; ACC: accelerations; DEC: decelerations; ExE: explosive efforts;

625 Int.: intervention

Table 2 Comparison of percentage duration [mean ± SD; MD and 95% CI] spend in

each velocity zone during tournament matches 1-3 combined

Velocity zone	CWI	CON	MD [95% CI]	p- value
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$< 6 \text{ km} \cdot \text{h}^{-1}$	54.8 ± 3.7	56.4 ± 6.0	1.6 [-6.5–3.3]	0.34
$6 - 11 \text{ km} \cdot \text{h}^{-1}$	24.4 ± 2.1	23.3 ± 2.8	1.1 [-1.3-3.5]	0.18
$11 - 15 \text{ km} \cdot \text{h}^{-1}$	12.9 ± 1.8	12.2 ± 2.6	0.7 [-1.5-2.9]	0.30
$15 - 19 \text{ km} \cdot \text{h}^{-1}$	5.4 ± 1.2	5.5 ± 1.5	0.1 [-1.4-1.2]	0.77
$19 - 23 \text{ km} \cdot \text{h}^{-1}$	1.9 ± 0.7	1.9 ± 0.5	0.0 [-0.6-0.6]	0.82
$> 23 \text{ km} \cdot \text{h}^{-1}$	0.5 ± 0.4	0.5 ± 0.3	0.0 [-0.3-0.3]	0.49

626 CWI: cold water immersion; CON: control

Variable	Intervention	Day 1 – FM	Day 2 – TR	Day 3 – TM 1	Day 4 – TM 2	Day 5 – TM 3	[time]	p-value [int.]	[time*int.]
RPE _{session}	CWI	4.9 ± 0.6	5.1 ± 0.7	5.7 ± 1.3	6.8 ± 1.7	6.0 ± 1.3	< 0.01	0.58	0.81
[AU]	CON	5.2 ± 1.0	5.7 ± 0.7	5.7 ± 0.7	6.7 ± 1.2	6.3 ± 1.2	< 0.01	0.58	0.81
Quality of sleep	CWI	2.6 ± 1.2	2.6 ± 1.0	2.7 ± 0.8	2.3 ± 0.9	2.7 ± 1.1	0.38	0.67	0.81
[AU]	CON	2.8 ± 1.2	2.2 ± 0.6	3.1 ± 1.3	2.3 ± 0.8	2.9 ± 0.7			
Perceived stress	CWI	2.6 ± 0.7	2.1 ± 0.3	2.1 ± 0.4	2.2 ± 1.0	2.8 ± 1.2	0.02	0.87	0.38
[AU]	CON	2.2 ± 0.7	2.5 ± 0.6	2.3 ± 1.0	2.0 ± 0.9	2.8 ± 0.7			
Perceived recovery	CWI	3.6 ± 0.5	3.6 ± 0.5	4.0 ± 0.6	3.9 ± 0.9	3.1 ± 1.0	< 0.01	0.82	0.85
[AU]	CON	3.7 ± 0.8	3.6 ± 0.7	3.7 ± 0.8	3.9 ±0.8	3.1 ± 0.6			
	CWI	77.5 ± 5.0	79.5 ± 4.0	73.6 ± 4.4 *	73.2 ± 4.9 *	76.0 ± 3.2	< 0.01	0.67	0.69
[70TIKmax]	CON	77.2 ± 3.9	80.6 ± 4.3	75.8 ± 4.1	73.9 ± 4.8	76.2 ± 3.3			
HRR [%HR _{ex}]	CWI	67.4 ± 3.2	65.4 ± 4.0	62.8 ± 6.3 *	60.3 ± 6.1 *	62.8 ± 5.0	< 0.01	0.36	0.26
	CON	70.1 ± 3.7	64.7 ± 3.8	62.4 ± 6.0	63.8 ± 6.5	66.4 ± 5.3			

Table 3 Comparison of perceptual and HR-based parameters (mean ± SD) between recovery modalities cold-water immersion (CWI) and control (CON)

628 FM: friendly match; TR: training; TM: tournament match; RPE: rating perceived excertion; HR_{ex}: exercise heart rate; HRmax: maximum heart rate;

- 629 HRR: heart rate recovery; Int.: intervention; * indicates significant time effects (p < 0.05) compared to Day 1





🗐 Questionnaire

Biomarkers

CA. Heart rate recovery



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