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# A MULTIDISCIPLINARY APPROACH TO OVERREACHING DETECTION IN ENDURANCE TRAINED ATHLETES

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1           **A MULTIDISCIPLINARY APPROACH TO OVERREACHING DETECTION IN**  
2                                   **ENDURANCE TRAINED ATHLETES**

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4  
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15 **Running title:** Multifactorial analysis for overreaching detection

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2

3 The experiments were conducted 32 in the laboratory of the National Institute of Sport, Expertise and

4 Performance, Paris, France

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10 *Collection of the data*

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21 All authors approved the final version of the manuscript.

1 **ABSTRACT**

2

3 In sport, high training load required to reach peak performance push human adaptation to their limits.  
4 In that process, athletes may experience general fatigue, impaired performance and may be identified  
5 as overreached (OR). When this state lasts for several months, an overtraining syndrome is diagnosed  
6 (OT). Until now, no variable *per se* can detect OR, a requirement to prevent the transition from OR to  
7 OT. It encouraged us to further investigate OR using a multivariate approach including physiological,  
8 biomechanical, cognitive and perceptive monitoring. Twenty-four highly trained triathletes were  
9 separated into an overload group and a normo-trained group (NT) during three weeks of training.  
10 Given the decrement of their running performance, eleven triathletes were diagnosed as OR after this  
11 period. A discriminant analysis showed that the changes of eight parameters measured during a  
12 maximal incremental test could explain 98.2% of the OR state (lactataemia, heart rate, biomechanical  
13 parameters and effort perception). Variations in heart rate and lactataemia were the two most  
14 discriminating factors. When the multifactorial analysis was restricted to these variables, the  
15 classification score reached 89.5%. Catecholamines and creatine kinase concentrations at rest did not  
16 change significantly in both groups. Running pattern was preserved and cognitive performance  
17 decrement was observed only at exhaustion in OR subjects. This study showed that monitoring various  
18 variables is required to prevent the transition between NT and OR. It emphasized that an OR index,  
19 which combines heart rate and blood lactate concentration changes after a strenuous training period,  
20 could be helpful to routinely detect OR.

## 1 INTRODUCTION

2

3

4 Increases in training and volume are typically undertaken by athletes in an attempt to enhance  
5 physical performance. High training loads (i.e. increased training volume and intensity) can place  
6 significant stress on the athlete's cognitive and physiological systems and if not matched by  
7 appropriate rest/recovery can lead to maladaptation, leading to increased fatigue and reduced  
8 performance (30, 41). When athletes require several days or weeks to recover physical performance,  
9 they are diagnosed as being overreached (OR) (30). Common symptoms reported with OR include  
10 general fatigue, sleep disorders, decreased appetite, loss of body weight, anxiety, reduce motivation,  
11 lack of concentration and variation of mood (18). In severe cases of maladaptive training, known as  
12 overtraining (OT), athletes may have reduced performance capacity either with or without these  
13 clinical symptoms that remain for several months or years. This most severe form of training  
14 maladaptation presents a serious threat for athletic performance and health. The currently accepted  
15 method for diagnosing OR/OT is to monitor performance after completion of a resting period of  
16 several days or weeks (18). Nevertheless, this method is frequently rejected by coaches and athletes  
17 because it may endanger the training continuum and it could lead to potential detraining. It is therefore  
18 important to identify early markers of OR/OT to limit the occurrence of these training maladaptation  
19 forms in population at risk.

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Many physiological variables have been recorded to detect OR and OT. One of the most reported physiological measures in endurance athletes has been a right shift in the lactate curve (4, 16, 22, 28, 39, 44). However, it has not been reported by all investigators (10, 26). Similarly, decreased nocturnal urinary catecholamine excretion has been associated with OT in endurance athletes and interpreted as lowered intrinsic sympathetic activity (25, 29). Nevertheless, a reduced intrinsic sympathetic activity has not been observed in all studies investigating OR/OT (19, 44, 46). A decrease in the ratio between the hormones testosterone or free testosterone and cortisol has also been proposed as a physiological marker of "anabolic-catabolic balance", a putative tool in the diagnosis of OT (1). Again, not all studies have observed changes in these variables with OR/OT (25, 29, 43, 46), and therefore, they are not considered as a good independent measure of maladaptive training (18). Finally, changes in heart rate (HR) at rest, and during both submaximal and maximal exercise have been reported to be associated with OR in various sports (9, 10, 19, 22, 26, 39). However, a recent meta-analysis examining the effect of overload training on resting, submaximal and maximal exercise HR and heart rate variability demonstrated that the small to moderate changes in these variables limits their clinical usefulness as idiosyncratic markers of OR and OT (5). Altogether then, the lack of consensus amongst research suggests that independent physiological markers may have limited practical usefulness if used as early warning markers of OR/OT.

38 In that context, there has been increasing interest in the application of cognitive tests as early  
39 warning measures of both OR and OT athletes (12, 13, 21, 31, 32). Nederhof et al. (32), reported that  
40 executive functions can be influenced by training tolerance and suggested that alterations in these  
41 functions may be an early indicator of maladaptive physical training. This hypothesis was  
42 strengthened by three studies that reported small increases in response time and increased number of  
43 mistakes in Stroop test at rest in OR and OT athletes (12, 13, 21). It remained that large inter-  
44 individual variability in the results of the cognitive tests limited their usefulness to assess a state of  
45 OR, especially when used alone. Also, cognitive performances had been assessed at rest and not  
46 during exercise, which could be a more suited measure to detect maladaptation in athletes.

47

48 In summary, investigations into early warning markers of OR / OT was still elusive and  
49 idiosyncratic physiological, biomechanical and cognitive variables that could identify OR remained to  
50 be found (18, 35, 45). It led us to propose a multivariate approach to identify athletes at risk of  
51 OR/OT. In order to test that hypothesis, we simultaneously monitored physiological, cognitive and  
52 biomechanical parameters at rest and during exercise in athletes progressively driven to OR by a  
53 prolonged period of overload training. We chose triathletes because they often undertake heavy loads  
54 during training and therefore have been reported to be at risk of OR and OT.

55

## 56 **Methods**

57

### 58 *Ethical approval*

59

60 Twenty-four well-trained triathletes volunteered to participate in this study. All subjects had  
61 competed in triathlons for at least 2 years and were training a minimum of 6 times per week. The  
62 experimental design of the study was approved by the Ethical Committee of Saint-Germain-en-Laye  
63 (acceptance no. 10054) and was done in accordance with the Declaration of Helsinki. Prior to  
64 participation in the investigation, subjects underwent medical assessment. After comprehensive verbal  
65 and written explanations of the study, all subjects gave their written informed consent.

66

67 The subjects were randomly assigned to either the experimental group (intensified training  
68 (IT) group) or the control group (normal training group, NT) according to a matched group  
69 experimental design based on maximal oxygen uptake ( $\dot{V}O_{2max}$ ) and maximal aerobic speed (MAS).  
70 Subjects' characteristics are presented in Table 1.

71

### 72 *Experimental protocol*

73

74 The protocol is illustrated in Figure 1. The investigation was conducted in September/October  
75 at the end of the competitive triathlon season to ensure a high fitness level for all participants. The  
76 training of each triathlete was monitored for a period of 7 weeks in total, which was divided into three  
77 distinct phases. The two first phases were similar for both IT and NT groups. The first phase (I)  
78 consisted of 3 weeks during which the subjects completed their usual amount and type of training  
79 (classic training). The second phase (II) consisted of one week of moderate training during which the  
80 subjects were asked to divide their normal training week by a half (recovery week). During the third  
81 period (III), the IT group completed a 3-week intensified program designed to deliberately overreach  
82 the triathletes; the duration of each training sessions of the classic training period was increased by  
83 40%. The NT group reproduced its classic training program during the same period. Throughout the  
84 entire experiment, the same sport scientist coached all triathletes. Training schedule was controlled to  
85 remain similar during each week of phase III. To avoid injuries, particular attention was devoted to  
86 daily feedback obtained from the triathletes. Throughout the entire study, heart rate was recorded  
87 during training to ensure that the triathletes adhered to prescribed training. At the end of phases II and  
88 III, the triathletes performed a maximal incremental running test on a 340-m indoor running track. To  
89 ensure that performance variations during the maximal incremental runs were due to the global  
90 training regimen and not to the training session(s) performed the day before each test, the subjects  
91 were required to respect a 24 h rest period before each maximal incremental run session.

92

### 93 *Assessment of energy intake*

94

95 During the 48 h prior, each maximal oxygen uptake ( $\dot{V}O_{2max}$ ) test, the triathletes were  
96 required to follow a nutritional plan in order to ensure muscle glycogen store resynthesis. They were  
97 allowed access to a buffet-type array of breakfast and meals foods and instructed to eat until satiety  
98 was reached. Breakfast consisted of a variety of macronutrients from both solid and liquid energy  
99 sources. The selected foods included an assortment of cereals, bread, fruit, yogurt, milk, juice, ham  
100 and cheese. In the lunch and dinner meals, athletes ate a mixed salad as starter, then white meat during  
101 lunch and fish during dinner. The side plate consisted of a mixed of 50% carbohydrates (i.e., pasta,  
102 rice, noodles) and 50% of vegetables (i.e., green beans, broccoli, tomatoes). One fruit and one yogurt  
103 were added as dessert, for lunch and dinner.

104

105

### 106 *Maximal running test*

107

108 The triathletes completed a maximal incremental running test on a 340-m indoor track to  
109 determine their  $\dot{V}O_{2max}$  and the velocity at which  $\dot{V}O_{2max}$  occurred ( $v\dot{V}O_{2max}$ ). The test began at 11

110 km·h<sup>-1</sup> and the speed was increased by 1 km·h<sup>-1</sup> every 3 minutes until volitional exhaustion. A rest  
111 period of 1-min was provided between each running step. The triathletes followed a cyclist travelling  
112 at the required velocity to ensure that the subjects were respecting the imposed pace. Visual marks  
113 were set at 20 m intervals along the track. The cyclist received audio cues via an mp3 player; the cue  
114 rhythm determined the speed needed to cover 20 m. The coefficient of variation of running speed  
115 between the tests pre- and post-phase III for each running step was subsequently calculated in order to  
116 assess the reproducibility of this parameter between the two tests.

117

### 118 *Physiological parameters*

119

120 Peripheral venous blood samples were taken from an antecubital vein of participants before  
121 each running test. Samples were drawn into non-additive tubes under sterile conditions. Serum was  
122 separated from whole blood by centrifugation at 1,000 g for 10 min at room temperature. An  
123 OLYMPUS 2700 analyzer (Beckman Coulter, Brea, USA) was used for simultaneous assay with  
124 reagents from the manufacturer of Creatine Kinase (CK). Plasma adrenalin and noradrenalin were  
125 measured in high-performance liquid chromatography with electrical detection (Laboratoire Medibio,  
126 Montargis, France).

127

### 128 *Metabolic parameters*

129

130 Between each increment, blood samples were taken from the participants' ear lobes during a  
131 1-min rest period and analyzed using a Lactate Pro system (36). Oxygen uptake ( $\dot{V}O_2$ ) and expiratory  
132 flow ( $\dot{V}_E$ ) were recorded breath-by-breath with a telemetric system collecting gas exchanges (Cosmed  
133 K4b<sup>2</sup>, Rome, Italy) (11), which was calibrated before each test. Heart rate values (HR) were monitored  
134 every second using a Polar unit. Expired gases and HR values were subsequently averaged every 5 s  
135 and were analysed (i.e., mean value) on time periods corresponding to the last 30s of each running  
136 step.  $\dot{V}O_{2max}$  was determined at exercise cessation when a plateau in  $\dot{V}O_2$  despite an increase in  
137 running speed was observed. If the subjects did not demonstrate any plateau in  $\dot{V}O_2$ , the test was  
138 considered to be maximal, when the respiratory exchange ratio value exceeded 1.15 and maximal HR  
139 value was over 90% of the predicted maximal value. The lactate threshold (LT) was assessed  
140 according to the D-max method previously described by Cheng et al. (7).

141

### 142 *Biomechanical parameters*

143



144           *Kinetic measures.* An area of biomechanical data collection was installed in a particular  
145 location of the indoor running track. This area was equipped with six adjacent force platforms  
146 (Z2074AA, Kistler, Switzerland) embedded in the track and covered with a layer of tartan, so as to not  
147 influence or disturb the triathletes while running. The total platform surface was approximately 6.6 m  
148 long and 0.6 m wide and the output signals of the six platforms were acquired in series at 1000 Hz.  
149 This length enabled data recording of at least four leg support phases (two left-side and two right-side  
150 supports) regardless of the running speed. This device gathered, for each instant of the support phase,  
151 the lateral (Fx), anteroposterior (Fy) and vertical (Fz) components of the force exerted by the  
152 triathletes on the ground. The data collected were propulsion (PI<sub>mn</sub>) and braking impulses (BI<sub>mn</sub>),  
153 peak vertical impact (Rz1<sub>n</sub>), maximum peak vertical force (Rz2<sub>n</sub>), support (dS), aerial (dA) and  
154 braking durations (dB<sub>n</sub>). Impulses and forces were normalized to body weight (x 1000 for impulses).  
155 Braking duration was normalised to support duration.

156

157           *Kinematic measures.* The movement acquisition system was a Vicon optoelectronic device  
158 (Oxford, United Kingdom), which uses 12 T10 cameras (resolution: 1megapixels) to follow and  
159 record in 3D the position of set retroreflective (passive) spherical markers. The acquisition frequency  
160 was set at 200 Hz. To reduce the effects of sliding of the markers, the triathletes were dressed in tight  
161 fitting outfits and markers were fixed with double-sided tape and their contact was reinforced with  
162 elastic adhesive strips.

163           Recordings from the force-platform and the video acquisition systems were synchronized.  
164 Depending on the running speed, the triathletes ran between one and three times in this area. The data  
165 collected were step length (L<sub>xn</sub>) and width (L<sub>yn</sub>), which were normalized to leg length and analyzed  
166 using mean values for each running stage.

167

168           ***Cognitive performance.***

169

170           During the maximal incremental running test, subjects had to respond to audio stimuli  
171 occurring in the second half of each 3-minute running stage.

172

173           *Double-task.* The system was comprised of two modified nunchuks (Nintendo Wii, Tokyo,  
174 Japan), an mp3 player and recorder, earphones and linking audio cables. Nunchuks were chosen based  
175 upon their light-weight and ergonomic design. To avoid any confusion, the upper analog stick was  
176 removed, the middle finger button was locked in the pressed down position and only the forefinger  
177 button was kept functional. Custom electronics allowed forefinger button actions to be recorded along  
178 with the given audio stimuli. The whole system weighed approximately 70 g.

179           Audio stimuli were delivered through earphones and consisted of 30 single and double, high-  
180 and low-pitched tones, randomly spaced in a 90s mp3 file. When hearing a single low-pitched or

181 double high-pitched tone, the triathlete was required to press down the left nunchuk button. Upon  
182 hearing a single high-pitched or double low-pitched tone, the triathlete was required press the right  
183 nunchuk button. All triathletes were instructed to respond as fast as possible. One week before the first  
184 maximal incremental running test, they received an mp3 test file for training, and repeated this training  
185 prior to each maximal incremental running test.

186 High- and low-pitched tones were respectively set as 5000 Hz and 150 Hz sine waves. Such  
187 frequencies allowed the triathletes to unequivocally distinguish high- from low-pitched tones. Single  
188 tones consisted of a 200 ms sine wave and double-tones consisted of two 70 ms sine waves  
189 interspaced with 80 ms, which resulted in a 220 ms stimulus. Such durations made it impossible for  
190 the triathletes to initiate any decision process before they had heard the entire stimulus.

191 It is well established that perceived loudness depends on tone (15, 37) and duration (33, 34).  
192 Single and double, high- and low-pitched tones amplitudes were adjusted in accordance to equal-  
193 loudness contours (often referred to as Fletcher-Munson curves) so that they met the international  
194 standard ISO 226 specifications (ISO 2012). During the medical assessment, subjects underwent an  
195 audiogram to ensure none of them had any hearing impairment.

196 The 30 stimuli were introduced in random order into a 90 s mp3 file and were separated with a  
197 random duration such that two consecutive stimuli were interspaced by at least 500 ms. A different file  
198 was played for each running stage so that it was not possible for the subject to learn the stimuli  
199 arrangement inside a file.

200 Data were processed in OriginPro 8.1 (OriginLab, Northampton, MA) with a custom-written  
201 script that returned, for each running stage, the percentage of false answers (excluded < 200 ms).

202

203 *Questionnaires.* The effect of the training regimen was also recorded through the assessment  
204 of the *perceived sensations* of subjects. The subjects were tested at rest and during the maximal  
205 incremental tests.

206 The Mindeval system was used to collect the data at rest (Mindeval GydléInc. Québec,  
207 CANADA). It is comprised of a web interface with a database and a stand-alone application. In the  
208 Pre- and -Post conditions, participants entered their personal key and answered questions within three  
209 areas related to pain, tiredness, and well-being, using a visual analogic scale. The software records the  
210 location of the indicator with a number ranging between 0 (no pain) and 100 (maximum pain). The  
211 collected data was stored on a secured server. Before the initiation of the study, triathletes were  
212 accustomed to the software, and the questions relative to their subjective sensations were thoroughly  
213 explained.

214 The rating of perceived exertion (RPE) was measured verbally using the Borg scale (3) during  
215 the maximal running test. This scale measures the subjective sensations accompanying the exercise.  
216 The scale and its purpose were carefully explained to each triathlete before each incremental test. The

217 triathletes were instructed to give a general RPE, a muscular RPE and a ventilatory RPE, immediately  
218 at the end of each running step and at exercise cessation.

219

### 220 *Data and statistical analyses*

221

222 The effect of the training regimen was analysed using the magnitude of variation between the  
223 beginning and the end of phase III for every parameter investigated. To reduce the effect of inter-  
224 individual differences in performance level, subsequent analyses were performed for three relative  
225 intensity levels of exercise determined for each triathlete at the end of phase III: low intensity running,  
226 lactate threshold (LT) and at exhaustion. Each parameter was compared with its respective value  
227 measured for the same running speed at the beginning of phase III. For all triathletes, the low intensity  
228 running was set at 13 km·h<sup>-1</sup> because: i) A very low coefficient of variation of running speed was  
229 indeed reported until this intensity (coefficient of variation of 3.93 and 2.24 at 12km·h<sup>-1</sup> and 13 km·h<sup>-1</sup>,  
230 respectively); ii) this running velocity was at least 2 km·h<sup>-1</sup> lower than LT for all triathletes.

231

232 Statistical analysis was performed using Statistica software for Windows (Statsoft, version  
233 7.0, Statistica, Tulsa, Oklahoma, USA). For the statistical procedure, the level of significance was set  
234 at  $p < 0.05$ .

235

236 *Assessment of the OR syndrome.* In order to determine the reproducibility of performance  
237 during the maximal running test and to identify OR athletes in the IT group, ICC (intraclass  
238 correlation coefficient) and confidence interval at 100% of performance variation were calculated for  
239 the NT group. To be diagnosed as OR, athletes of the IT group had to reveal a performance decrement  
240 higher than the lowest reproducibility value reported for the NT group (OR threshold). Using that  
241 procedure, the IT group was divided in two subgroups. When the subjects of the IT group  
242 demonstrated a performance decrement higher than OR threshold, they were considered truly  
243 overreached (OR group). When this assumption was not confirmed at the end of the overload period,  
244 they we were not considered overreached (n-OR group).

245

246

247

### 248 *Discriminant analyses*

249

250 Three stepwise discriminant analyses (DA) were conducted to determine the ability of the  
251 different variables measured during exercise to distinguish between NT, n-OR and OR groups and  
252 subsequently predict group membership. The criterion used to determine whether a variable entered  
253 the model (i.e., discriminant function) was Wilk's Lambda, which measures the deviations within each

254 group with respect to the total deviations. The sample-splitting method initially included the variable  
255 that most minimized the value of Wilk's Lambda, provided the value of F was greater than a certain  
256 critical value. The next step was pairwise combination of the variables with one of them being the  
257 variable included in the first step. Successive steps were performed in the same manner, always with  
258 the condition that the F-value corresponding to the Wilk's Lambda of the variable to select has to be  
259 greater than the aforementioned "entry" threshold. If this condition was not satisfied, the process was  
260 halted, and no further variables were selected in the process. Before including a new variable, an  
261 attempt was made to make some of those already selected if the increase in the value of Wilk's  
262 Lambda was minimal, and the corresponding F-value was below a critical value. Wilk's Lambda,  
263 canonical correlation index, and percentage of subjects were computed as indicators of OR predictive  
264 capacity.

265

266 The first DA (DA1) was performed on all the tested subjects (NT, n-OR and OR groups: 24  
267 subjects tested at 3 running intensities) using all the variables tested in the study (n = 21). It was used  
268 to determine if some variables would allow to identify three groups of triathletes according to their  
269 training regimen and their performance decrement during the protocol. The second DA (DA2)  
270 excluded the n-OR group (NT and OR groups: 19 subjects at 3 running intensities, see below for the  
271 justification of the 19 subjects) using all the variables measured (n = 21). This analysis was performed  
272 to identify the most valuable variables in classifying triathletes of NT and OR groups as overreached  
273 or not. The discriminating variables with their respective Wilk's lambdas and p-value, canonical  
274 correlation ( $r_c$ ) and classification percentage were noted. Considering that markers of OR should be  
275 applicable in training practice (32), a third additional DA (DA3) was performed to investigate the  
276 minimal number of variables allowing a reasonable discrimination between the OR and NT groups.

277

278 *Parameters evolution*

279

280 Since this protocol involved a relatively small number of subjects (n < 32) and the data  
281 obtained did not always meet the assumptions of normality, as assessed visually by normal probability  
282 plot and by the Shapiro-Wilk test, non-parametric statistical analyses ensued. A Friedman rank test  
283 was undertaken to evaluate the statistical differences in time for each group and a Mann-Whitney test  
284 was completed to assess significant differences between NT and OR groups. The results are expressed  
285 as the mean value with standard deviation ( $\pm$  SD).

286

287

288 **RESULTS**

289

290 All the subjects successfully completed the prescribed training program in both NT and IT  
291 groups.

292

### 293 *Assessment of the OR syndrome*

294

295 An intra-class correlation test (ICC) was used to classify the subjects from the IT group as  
296 overreached (OR group) or non-overreached (n-OR group). First, the reproducibility of the  
297 performance of the NT group was measured using the ICC test (see method). ICC value was very high  
298 (ICC = 0.98), with a performance repeatability ranging between 0.6 to 1.8% (mean: 0.9%). On the  
299 basis of this analysis, a decrement of performance of greater than 1.8% was used as the criteria to  
300 discriminate the OR subjects in the IT group. Subsequent analysis showed that only 11 of the 16  
301 triathletes that complete the overload training were considered as truly OR group). The five other  
302 subjects of the IT group were not diagnosed OR.

303

### 304 *Performance*

305

306 In the OR group, the running performance decreased on average by  $4.4 \pm 1.1\%$  between the  
307 beginning and the end of the intensified training period ( $18.3 \pm 0.2 \text{ km}\cdot\text{h}^{-1}$  and  $17.6 \pm 0.3 \text{ km}\cdot\text{h}^{-1}$ ,  $p <$   
308  $0.001$ , pre- and post-overload period, respectively). When expressed in total running distance covered  
309 during the incremental test, this decline represented  $13.3 \pm 3.2\%$ .

310

### 311 *Physiological parameters*

312

313 Both the NT and OR groups were first submitted to the same initial 4 week training protocol  
314 (phases I and II in Figure 1). As shown in Table 2, the physiological variables values measured at the  
315 end of phase II were not significantly different between the two experimental groups. The OR group  
316 then completed a training program with 40% increase in load (phase III).

317

318 *Metabolic parameters.* At the end of the overload period (phase III), a decrease of HR and  $[\text{La}^-]$   
319  $_{\text{b}}$  values was observed for the OR group for the two submaximal intensities and at exhaustion (Table  
320 2a). In contrast, no significant variation was observed for these two parameters for the three running  
321 intensities in the NT group. These variations in HR and  $[\text{La}^-]_{\text{b}}$  values were significantly different for  
322 OR and NT groups for all the running intensities (compare the numerical values in columns 3 and 6 of  
323 Table 2a). No significant differences in  $\dot{V}\text{O}_2$  and  $\dot{V}_{\text{E}}$  values were observed between the two groups  
324 before and after phase III.

325

326 *Blood parameters.* No significant statistical difference in [CK] was observed in the OR group  
327 during phase III ( $234 \pm 142$  and  $257 \pm 157$  UI.L<sup>-1</sup>, pre- and post- phase III,  $p = 0.07$ ). No significant  
328 variation was observed either in the NT group for this parameter during the same period ( $180 \pm 83$  and  
329  $161 \pm 49$  UI.L<sup>-1</sup>, pre- and post- phase III, respectively,  $p = 0.48$ ). Similarly, there were no significant  
330 differences in plasma catecholamine concentrations in both groups before and after phase III ( $p >$   
331  $0.37$ ). Similarly, there were no significant interaction (time x training regimen) for plasma [CK] ( $p =$   
332  $0.17$ ), adrenalin ( $p = 0.88$ ) and noradrenalin ( $p = 0.90$ ) at rest.

333

### 334 *Cognitive performance*

335

336 There was no difference between groups at rest ( $-5.5 \pm 11.2\%$ ,  $-4.3 \pm 3.4\%$ , for NT and OR  
337 groups, respectively,  $p = 0.39$ ), low intensity ( $-1.2 \pm 4.5\%$ ,  $-2.0 \pm 5.5\%$ , for NT and OR groups,  
338 respectively,  $p = 0.69$ ) and lactate threshold ( $-1.9 \pm 8.7\%$ ,  $1.3 \pm 9.2\%$ , for NT and OR groups,  
339 respectively,  $p = 0.52$ ). In contrast, the OR group demonstrated a significant decrease in performance  
340 at exhaustion than the NT group ( $8.7 \pm 11.3\%$  and  $-12.1 \pm 17.9\%$ , for NT and OR groups, respectively,  
341  $p = 0.04$ ).

342

### 343 *Biomechanical parameters*

344

345 Except dS (support duration) at LT (lactate threshold) ( $-11 \pm 12$  ms and  $2 \pm 6$  ms, for OR and  
346 NT groups, respectively,  $p = 0.01$ ), no significant interaction effect was reported for all the 9  
347 parameters investigated at three running speeds ( $p > 0.05$ ) (Table 2b).

348

### 349 *Perceived sensations*

350

#### 351 *At rest*

352

353 The OR triathletes reported increased sensations of pain ( $16 \pm 24$  and  $53 \pm 26$ ,  $p < 0.01$ , before  
354 and after the overload period, respectively) and tiredness ( $20 \pm 18$  and  $85 \pm 11$ ,  $p < 0.001$ , before and  
355 after the overload period, respectively). In contrast, there was no significant difference for these two  
356 parameters during the same period for the NT group ( $28 \pm 32$  and  $18 \pm 13$ , for pain,  $38 \pm 16$  and  $38 \pm$   
357  $24$ , for tiredness, before and after phase III, respectively,  $p > 0.05$ ). There was a significant difference  
358 in the change in pain ( $p = 0.03$ ) and tiredness ( $p < 0.001$ ) between the OR and NT groups. Well being  
359 sensation demonstrated no significant change in both groups before and after phase III ( $76 \pm 17$  and  $61$   
360  $\pm 31$ ,  $p = 0.23$ , for OR group,  $73 \pm 22$  and  $73 \pm 20$ ,  $p = 0.72$ , before and after the overload period,  
361 respectively).

362

363 *During exercise*

364

365 There was a significant difference in  $\Delta\text{GenRPE}$  (general perceived exertion change) was  
366 observed at exhaustion ( $+1.8 \pm 1.4$  and  $+0.1 \pm 1.3$ ,  $p = 0.02$ ) between the OR and NT groups, however  
367 there were no-statistical differences at low ( $+2.1 \pm 3.1$  and  $-0.4 \pm 1.0$ ,  $p = 0.05$ ) and LT intensities  
368 ( $+2.2 \pm 2.4$  and  $+0.1 \pm 1.8$ ,  $p = 0.08$ ). The  $\Delta\text{MuscRPE}$  (muscular perceived exertion change) was  
369 significantly different between NT and OR groups at Low ( $+4.1 \pm 3.2$  and  $+0.0 \pm 1.0$ ,  $p < 0.01$ ) and  
370 LT intensities ( $+3.3 \pm 2.2$  and  $+0.8 \pm 1.1$ ,  $p = 0.02$ ), but not at exhaustion ( $+3.3 \pm 2.0$  and  $+1.7 \pm 1.4$ ,  $p$   
371  $= 0.10$ ). Finally, the training load did not influence  $\Delta\text{VentRPE}$  (ventilatory perceived exertion change)  
372 for the three running intensities ( $p > 0.20$ ).

373

374 *Discriminant analyses*

375

376 The DA1 was performed on all the tested subjects using all the variables tested in the study. It  
377 was used in order to determine if some variables would allow identification of three groups of  
378 triathletes according to their training regimen and performance decrement during the protocol. DA1  
379 indicated the presence of two significant discriminant functions ( $p < 0.01$ ). As a linear combination of  
380 discriminating variables, the analysis resulted in canonical coefficients for the first function being  
381 derived so that the group means on the function were as different as possible. The coefficient for the  
382 second function was also derived to maximize the differences between the group means as long as the  
383 values on the second function were not correlated with those on the first function. The discriminant  
384 functions were used to compute the position of the triathlete's data in the discriminant space (Figure  
385 2). The horizontal direction corresponded to function 1, with the lateral separation among the three  
386 groups indicating how much they were distinguished on this function. The vertical axis corresponded  
387 to function 2, with the vertical separation indicating the manner in which the groups were  
388 distinguished in a way unrelated to the way they were separated on function 1 (40). Using this analysis  
389 87.5% of the NT, n-OR and OR subjects were classified in the correct group (Table 3). With three  
390 groups, 33.3% of correct predictions are possible with pure random assignment (24). In summary,  
391 DA1 showed that we could discriminate the three groups of athletes using the variables measured.

392

393 The second DA (DA2) excluded the n-OR group using all the variables measured. It was  
394 performed to identify the most valuable variables in classifying triathletes of NT and OR groups as  
395 overreached or not. It indicated the presence of one significant discriminant function ( $p < 0.001$ ). The  
396 discriminant function was interpreted by examining the standardized coefficients (see Table 4a) in  
397 order to ascertain which variables contributed most to determining scores on the function. The larger  
398 the magnitude of the coefficient, the greater the contribution of that variable to the discriminant  
399 function.  $\Delta\text{HR}$  (heart rate variation) made the greatest contribution to scores on that function followed

400 by  $\Delta dS$  (stance phase duration change),  $\Delta dA$  (aerial phase duration change),  $\Delta[La^-]_b$  (blood lactate  
401 concentration change) and  $\Delta Lxn$  (step-length change) with a lesser contribution from the three other  
402 factors selected in the model ( $\Delta PImn$ , propulsive impulse change;  $\Delta Lyn$ , step largeness change;  
403  $\Delta muscRPE$ , muscular perceived exertion change). The classification procedure correctly placed 98.2%  
404 of the triathletes of NT and OR groups into their respective groups (see Table 4b). The probability by  
405 chance with two groups would have been 50.0%. The extent to which all parameters were valuable  
406 and necessary in DA2 was determined via a stepwise procedure. A forward stepwise procedure was  
407 utilized whereby the individual variable that provided the greatest univariate discrimination was  
408 selected first and was then paired with each of the remaining variables one at a time, to determine the  
409 combination which produced the greatest discrimination. This analysis included the 8 selected  
410 variables of DA2 in the following order of decreasing discriminating power:  $\Delta HR$ ,  $\Delta[La^-]_b$ ,  $\Delta PImn$ ,  
411  $\Delta dS$ ,  $\Delta dA$  and  $\Delta Lxn$ . All these variables made a significant ( $p < 0.05$ ) contribution to discrimination  
412 between NT and OR groups, while no statistical significant contribution were observed for both  $\Delta Lxn$   
413 and  $\Delta MuscRPE$  (Table 4c). In summary, DA2 ranked 8 of the 21 variables measured as valuable to  
414 discriminate between OR and NT groups.

415

416 Considering that only a limited number of markers of OR could practically be applied in the  
417 training environment, a third additional DA (DA3) was performed. It investigated the minimal number  
418 of variables allowing a reasonable discrimination between the OR and NT groups. When the variables  
419 was restricted to  $\Delta HR$  and  $\Delta[La^-]_b$  (i.e., the two most valuable variables in DA2), the classification  
420 score still reached 89.5% (Table 5). The classification function coefficients determined by DA3 could  
421 be used in an equation to determine the likelihood of an individual triathlete to be classified as OR  
422 using variables measured during exercise:

423

$$424 \text{ OR index} = 0.17 \times \Delta HR + 0.89 \times \Delta[La^-]_b + 1.36$$

425

426 Where  $\Delta HR$  and  $\Delta[La^-]_b$  represent heart rate and blood lactate concentration changes, respectively. As  
427 illustrated in Figure 2, using that formalism, a negative value strongly suggests a state of OR.

428

429

## 430 **DISCUSSION**

431

432 The main findings of this study were that: (i) Combining physiological, biomechanical and  
433 cognitive variables were useful to assess overreaching (OR) in endurance trained athletes after an  
434 overload period; (ii) multidimensional analysis showed that heart rate and blood lactate concentration  
435 changes were the most important factors in discriminating between control and OR athletes; (iii) while  
436 motor control did not appear to be altered during an incremental running test with OR, cognitive



437 performance was impaired at exhaustion in OR subjects compared to the controls; (iv) the  
438 physiological perturbations associated with OR were coherent with perturbations of the autonomic  
439 nervous system activity; (v) these results led to the proposal that an index based on two variables  
440 could assist in the diagnosis of OR in endurance athletes.

441

442 At the end of the overload training period, a 4.4% decline in maximal running speed was  
443 observed in the OR group. Given that the daily variation of this test was <1.8% in the NT group, the  
444 decline in performance could be attributed to the effects of the intensified training protocol. This  
445 reduction in performance was in line with the 5.4% decrement reported by Halson et al. (17) in OR  
446 cyclists with a similar incremental protocol. When expressed in total running distance during the  
447 incremental test, this decrease in performance represented 13.3% in the OR group. A similar decrease  
448 was observed by Lehmann et al. (26), who showed an 8% decline in total running distance during an  
449 incremental exercise test in middle- and long-distance runners. Additionally, in our study, the OR  
450 triathletes reported a large increase in perceived fatigue at rest, while no significant variations were  
451 assessed in the NT group. Reduced physical performance and increased fatigue are two of the common  
452 criteria for diagnosing OR (18), which confirmed that these athletes were not adapting to the  
453 prescribed overload training. It allowed us to conduct further comparison with the NT athletes (i.e.,  
454 normal training group) to determine discriminate markers of OR/OT.

455

### 456 **Early detection of overreaching**

457

458 The aim of this study was to identify specific marker(s) of OR in triathletes that could be used  
459 prospectively to prevent endurance athletes from developing OT. The present results showed that a  
460 combination of 8 physiological, cognitive and biomechanical parameters changes measured during an  
461 incremental maximal running test successfully discriminated between OR and NT triathletes at 98.2%  
462 (chance probability: 50%). Indeed, with the exception of only 1/57 cases (19 triathletes, 3 running  
463 intensities), the training state of individual athletes was adequately classified. Interestingly, the  
464 stepwise discriminant analysis indicated that the  $\Delta\text{HR}$  and  $\Delta[\text{La}^-]_b$  were the two most valuable factors  
465 to discriminate between OR and NT groups. When the discriminant analysis was restricted to these  
466 two parameters, 89.5% of the triathletes were still well classified. These findings have strong practical  
467 applications as both these measures fulfil the criteria defining a usable marker for detecting OR (and  
468 OT) (32): (i) objective; (ii) not easily manipulated; (iii) applicable in training practice; (iv) not too  
469 demanding for athletes; (v) affordable for the majority of athletes and (vi) based on a theoretical  
470 framework.

471

472 We expected that alterations of the running motor patterns (i.e. stride kinematic and  
473 mechanical parameters) in triathletes could have been a valid indicator of OR. Surprisingly, we were

474 only able to detect minor modifications in the motor pattern, which used in isolation, did not  
475 distinguish OR athletes from the NT group. These observations suggest that motor control was largely  
476 preserved during the incremental exercise (at submaximal levels), regardless of training status. These  
477 findings may also partly explain why athletes can become OR/OT despite close and regular  
478 observation from coaches. Indeed, without clearly visible changes in motor patterns (i.e. noticeable  
479 changes gait), it becomes difficult to discriminate OR from other potential causes of performance  
480 decrement, which emphasizes the necessity for regular monitoring in endurance athletes, especially  
481 during periods of heavy training (43). On the basis of the present findings, we suggest to monitor HR  
482 and blood lactate concentration. Indeed, the combination of these two measures in the OR index  
483 algorithm ( $OR\ index = 0.17 \Delta HR + 0.88 \Delta [La^-]_b + 1.36$ ), could be used as an objective early warning  
484 for maladaptive training in endurance athletes.

485

## 486 **Underlying mechanisms of overreaching**

487

### 488 *The autonomic hypothesis*

489 Whilst the underlying cause(s) of OR (and OT) in endurance athlete remains to be determined  
490 (18, 45), there is an agreement that the concomitant decrease of HR and  $[La^-]_b$  reported in several  
491 studies could reveal a down-regulation of the sympathetic nervous system and/or changes in  
492 parasympathetic/sympathetic tone during OR (19, 26, 43). Two mutually non-exclusive mechanisms  
493 (i.e. centrally and peripherally mediated factors) have been suggested to underpin these physiological  
494 changes. In favor of a centrally mediated factors, Lehmann et al. (26) reported decreased nocturnal  
495 urinary norepinephrine and epinephrine excretion after an increase in training volume leading to OR.  
496 There was also a concomitant decline in submaximal and maximal heart rates along with the changes  
497 in catecholamines. In contrast, others reported decreases in heart rate and/or (20) lactate concentration  
498 in absence of catecholamine modulations (17, 43). Prolonged exposure to catecholamines resulting  
499 from intensified training and/or psychological stress may also downregulate  $\beta$ -adrenergic receptors  
500 sensitivity, and/or decrease their number (27, 47). This has been observed after exhaustive dynamic  
501 exercise (6), chronic exposure to hypoxia (14) and during a prolonged long-term period of heavy  
502 endurance training (23) or after infusion of adrenergic agonists (42).

503

### 504 *A role for cognitive factors?*

505 In the present study, the cognitive performance was preserved in all athletes at rest and  
506 submaximal intensities. Notably however, cognitive performance was reduced at exhaustion in OR  
507 athletes. These findings show that whilst cognitive measures were only marginally useful to predict  
508 OR, they were affected by OR. These observations are consistent with the threshold theory that  
509 involves two hypothetical notions (38). The first suggests that the brain has a reserve capacity and  
510 second that the brain has a threshold of impairment. According to this model, the larger the brain

511 reserve capacity and the higher the threshold of impairment, the better the tolerance of cognitive  
512 processes to different stimuli. In the context of that theory, we propose that the psychological load  
513 associated with running during the incremental test (i.e., rate of perceived exertion, RPE) only affected  
514 cognitive performance when high running speed were reached (i.e., beyond the lactate threshold). The  
515 decreased cognitive performance observed at exhaustion was in agreement with Chmura and Nazar  
516 (8), who demonstrated that it is only above lactate threshold that reaction time increased markedly  
517 during a running incremental test.

518         The coincidence of increased physical exhaustion and the large deterioration in the double task  
519 performance indicated that in OR and NT groups: (i) Running at severe intensities (i.e., above lactate  
520 threshold), are accompanied by a large cognitive load; and (ii) that these two tasks rely upon the  
521 similar cognitive resources. Moreover, since the cognitive performance showed greater decrease in the  
522 OR triathletes (despite lower running speed at exhaustion) than the control group and this occurred  
523 with an increase of both general and muscular perceived exertion, it seems that central factors may be  
524 involved in OR. This is further supported by the finding that the increased perception of exertion was  
525 not associated with higher muscle damage in the OR triathletes. Taken collectively, these results  
526 demonstrate that the attention demand of running is increased at high intensity in OR subjects, which  
527 may suggest a contribution of central fatigue in OR. These results agree with previous studies that  
528 have highlighted similarities between OR/OT athletes with chronic fatigue syndrome and major  
529 depression symptoms (2, 32). Indeed, decreased psychomotor speed has consistently been shown to be  
530 present in both depression and OR/OT athletes (32). Furthermore, a reduced performance on  
531 psychomotor speed tasks was observed in OT athletes at rest (1212, 13, 21, 31). The present  
532 investigation extends these results by showing cognitive impairment during strenuous exercise in OR  
533 athletes.

534

### 535 **Summary**

536         In order to determine discriminant markers of maladaptive training endurance athletes,  
537 comparisons were made between various physiological, cognitive and biomechanical measures in OR  
538 and non-OR triathletes during 3 weeks of increased training load. A combination of physiological,  
539 cognitive and biomechanical parameters changes measured during an incremental maximal running  
540 test successfully discriminated between OR and control at 98.2%. Heart rate and blood lactate  
541 concentration variations were the two most discriminating factors (89.5% of discrimination success,  
542 when combined).

543         The results showed that the triathletes running motor patterns were not altered until exhaustion  
544 in OR subjects. These observations could explain why athletes can become OR/OT whilst under the  
545 close supervision of a coach/scientist. Without visual marker, an external observer would have  
546 difficulty to discriminate OR from other potential causes of performance decrement. These findings  
547 also highlight that monitoring physiological responses could help preventing OR and OT. On the basis

548 of the current observations, we propose an OR index, which combines heart rate and blood lactate  
549 concentration changes after a training period could be helpful to routinely detect OR in athletes  
550 submitted to strenuous training regimen. Indeed, this algorithm may be used to monitor and  
551 prospectively guide future manipulations in training load so that the risks of OR/OT are reduced.

552         Whilst the physiological mechanisms that underlie OR/OT remain to be fully elucidated, the  
553 concomitant decrease of heart rate and blood lactate concentration changes pointed to perturbations of  
554 the autonomic nervous system as one mechanism underlying the genesis of OR. Additionally, since  
555 the double task showed that running at severe intensities was accompanied by an increased cognitive  
556 load, which is further increased with OR, it also appears that an athlete's cognitive resources are  
557 depleted during intense exercise with OR/OT. These results should be now confirmed on a larger  
558 population of athletes, involved in different sports and levels of performance.

559

- 1 **Figure 1.** Schematic representation of the experimental protocol.
- 2 **Figure 2.** Discriminant analysis scatter plots using different number of groups and variables. NT:
- 3 normal training group; n-OR: intensified training group without overreaching symptoms; OR:
- 4 intensified training group with overreaching symptoms.
- 5

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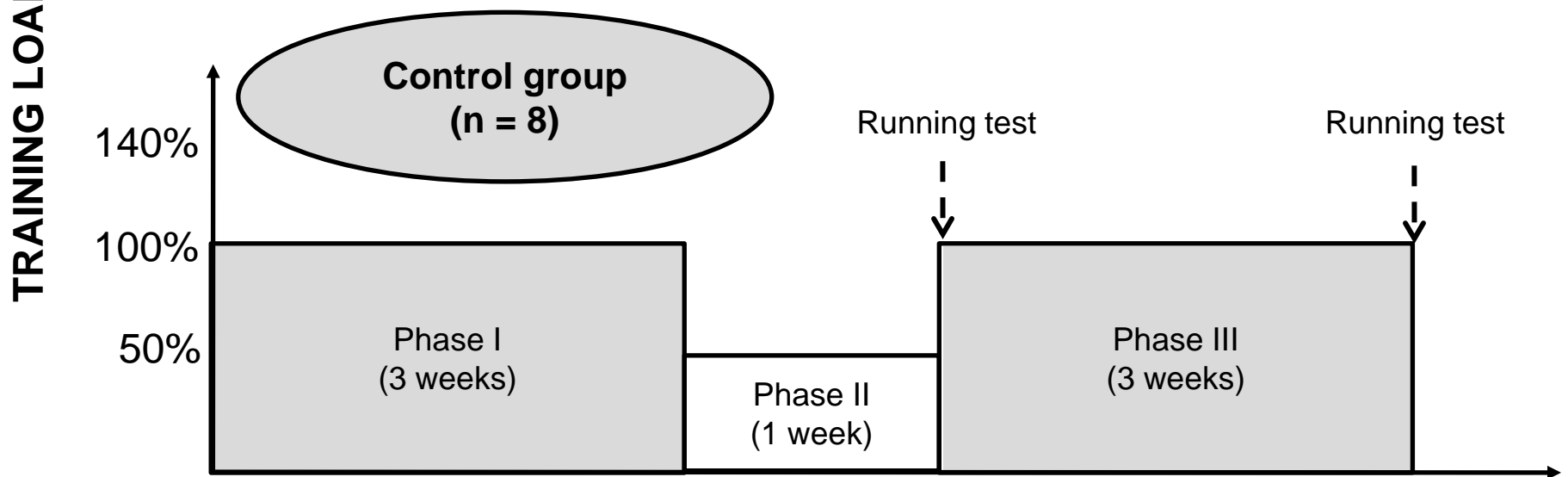
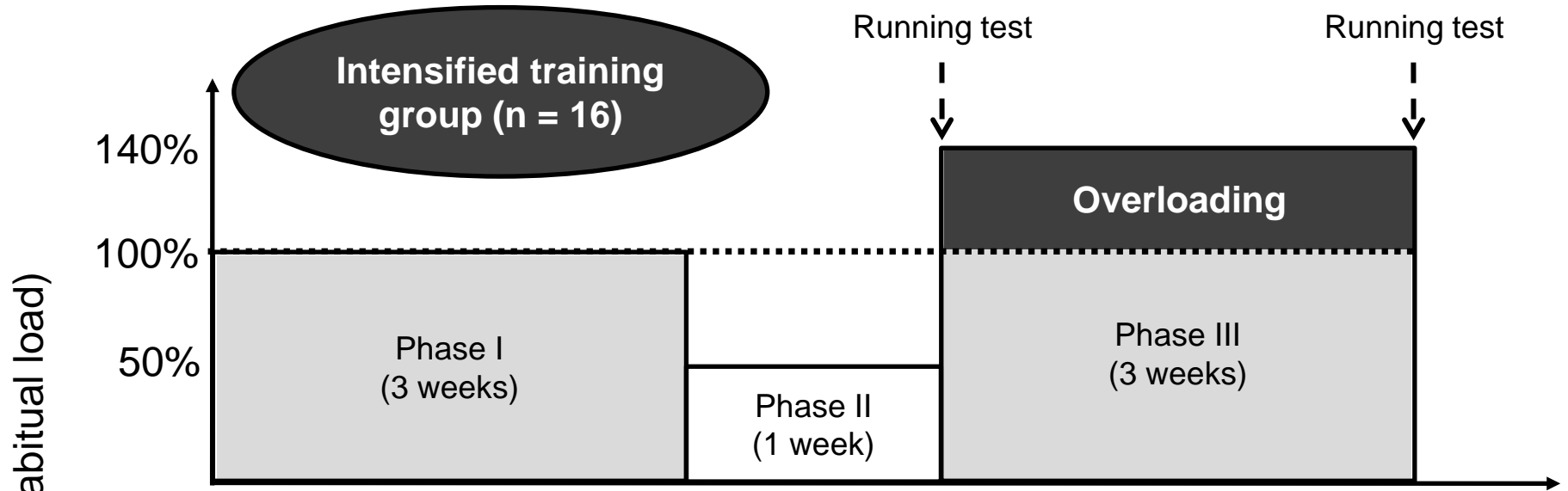
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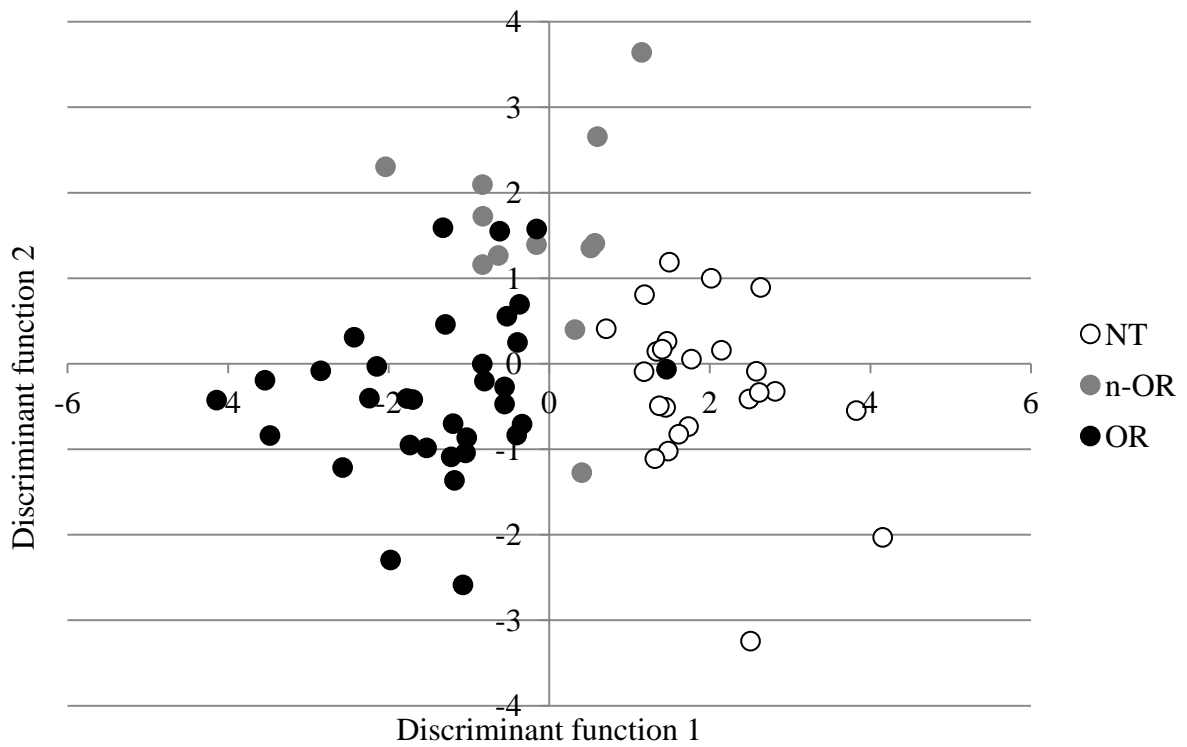
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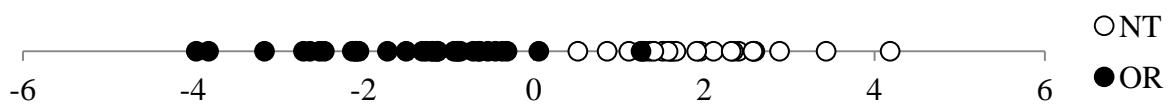
**A. Discriminant analysis 1 (3 groups, 16 variables)**

Success rate for classification : 87.5%



**b. Discriminant analysis 2 (2 groups, 16 variables)**

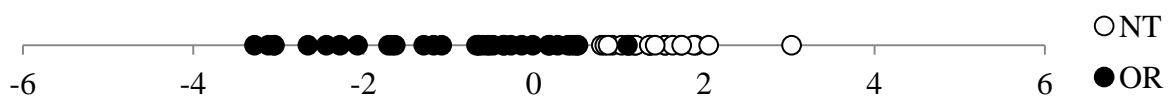
Success rate for classification : 98.2%



**c. Discriminant analysis 3 (2 groups, 2 variables)**

Success rate for classification : 89.5%

$$\text{OR index} = 0.17 \Delta\text{HR} + 0.88 \Delta[\text{La}^-]_b + 1.36$$



<b>Subject characteristics</b>	<b>Normal Training group (n = 8)</b>	<b>Intensified Training group (n = 15)</b>
<b>Age (years)</b>	32.4 ± 2.8	31.0 ± 1.4
<b>Height (cm)</b>	176.8 ± 2.1	178.7 ± 1.2
<b>Weight (kg)</b>	69.7 ± 2.6	70.6 ± 1.3
<b><math>\dot{V}O_{2max}</math> (ml.min<sup>-1</sup>.kg<sup>-1</sup>)</b>	64.9 ± 2.8	62.3 ± 1.5
<b>MAS (km.h<sup>-1</sup>)</b>	18.2 ± 0.4	18.3 ± 0.2

**Table 1.** Selected characteristics of the two experimental groups.  $\dot{V}O_{2max}$ : maximal oxygen uptake; MAS: maximal aerobic speed. Values are expressed as means ± SEM of the means. No significant difference between both groups for all the parameters.

a.

Intensity	Physiological variables	Normal Training Group (NT, n = 8)			Overreached Group (OR, n = 11)		
		Pre-Training	Post-Training	Variation	Pre-Training	Post-Training	Variation
Low	$\dot{V}O_2$ ( $\text{mlO}_2 \cdot \text{min}^{-1} \cdot \text{kg}^{-1}$ )	48.8 ± 5.0	47.9 ± 5.2	-0.9 ± 2.4	49.1 ± 1.8	49.7 ± 3.3	0.6 ± 2.1
	$\dot{V}_E$ ( $\text{L} \cdot \text{min}^{-1}$ )	92 ± 14	90 ± 13	-2 ± 5	91 ± 11	94 ± 12	3 ± 5
	HR ( $\text{beats} \cdot \text{min}^{-1}$ )	155 ± 11	154 ± 11	-1 ± 2	152 ± 13	143 ± 13**	<b>-8 ± 6<sup>#</sup></b>
	$[\text{La}]_b$ ( $\text{mmol} \cdot \text{L}^{-1}$ )	1.7 ± 0.5	1.5 ± 0.4	-0.2 ± 0.3	2.7 ± 1.0	1.9 ± 0.8**	<b>-0.8 ± 0.8<sup>#</sup></b>
LT	$\dot{V}O_2$ ( $\text{mlO}_2 \cdot \text{min}^{-1} \cdot \text{kg}^{-1}$ )	58.5 ± 3.4	58.5 ± 1.5	0.0 ± 0.6	57.4 ± 3.9	57.9 ± 6.0	0.4 ± 3.0
	$\dot{V}_E$ ( $\text{L} \cdot \text{min}^{-1}$ )	130 ± 19	131 ± 14	1 ± 8	126 ± 17	130 ± 19	4 ± 5
	HR ( $\text{beats} \cdot \text{min}^{-1}$ )	176 ± 8	175 ± 8	-1 ± 3	172 ± 9	163 ± 9**	<b>-9 ± 5<sup>##</sup></b>
	$[\text{La}]_b$ ( $\text{mmol} \cdot \text{L}^{-1}$ )	3.4 ± 0.8	3.1 ± 0.8	-0.2 ± 0.6	3.8 ± 1.1	2.5 ± 0.7**	<b>-1.3 ± 0.8<sup>#</sup></b>
At exhaustion	$\dot{V}O_2$ ( $\text{mlO}_2 \cdot \text{min}^{-1} \cdot \text{kg}^{-1}$ )	61.5 ± 3.3	61.3 ± 1.6	-0.2 ± 1.2	61.0 ± 5.2	60.9 ± 6.4	-0.1 ± 3.2
	$\dot{V}_E$ ( $\text{L} \cdot \text{min}^{-1}$ )	154 ± 17	159 ± 15	5 ± 11	162 ± 22	161 ± 23	-1 ± 11
	HR ( $\text{beats} \cdot \text{min}^{-1}$ )	182 ± 13	182 ± 12	0 ± 1	181 ± 8	173 ± 8***	<b>-8 ± 3<sup>###</sup></b>
	$[\text{La}]_b$ ( $\text{mmol} \cdot \text{L}^{-1}$ )	8.9 ± 1.1	9.0 ± 0.7	0.3 ± 0.6	8.1 ± 2.0	6.9 ± 1.7**	<b>-1.2 ± 0.2<sup>##</sup></b>

b.

Intensity	Biomechanical parameters	Normal Training Group (n = 8)			Overreached Group (n = 11)		
		Pre-Training	Post-Training	Variation	Pre-Training	Post-Training	Variation
Low	Stride length (x leg length)	1.39 ± 0.06	1.40 ± 0.08	0.01 ± 0.05	1.36 ± 0.05	1.36 ± 0.06	0.00 ± 0.03
	Support duration (ms)	243 ± 11	241 ± 18	-2 ± 10	255 ± 17	253 ± 13	-1 ± 8
	Aerial duration (ms)	112 ± 20	116 ± 21	-1 ± 10	104 ± 26	104 ± 22	-1 ± 10
	Maximum peak vertical force (x weight)	2.63 ± 0.24	2.65 ± 0.25	0.02 ± 0.07	2.52 ± 0.16	2.50 ± 0.15	-0.02 ± 0.07
LT	Stride length (x leg length)	1.65 ± 0.10	1.63 ± 0.11	-0.02 ± 0.04	1.56 ± 0.10	1.54 ± 0.11	-0.02 ± 0.05
	Support duration (ms)	211 ± 12	211 ± 17	-1 ± 8	229 ± 15	231 ± 13	2 ± 7
	Aerial duration (ms)	130 ± 23	132 ± 24	2 ± 6	120 ± 25	108 ± 23	<b>-11 ± 12<sup>##</sup></b>
	Maximum peak vertical force (x weight)	2.83 ± 0.33	2.83 ± 0.34	0.00 ± 0.07	2.67 ± 0.21	2.60 ± 0.17	-0.07 ± 0.11
At exhaustion	Stride length (x leg length)	1.79 ± 0.11	1.76 ± 0.12	-0.03 ± 0.05	1.68 ± 0.14	1.68 ± 0.14	-0.01 ± 0.06
	Support duration (ms)	199 ± 10	198 ± 16	-1 ± 10	214 ± 15	208 ± 13	-7 ± 9
	Aerial duration (ms)	130 ± 20	131 ± 19	2 ± 3	120 ± 22	117 ± 20	-3 ± 10
	Maximum peak vertical force (x weight)	2.83 ± 0.29	2.84 ± 0.28	0.01 ± 0.08	2.70 ± 0.21	2.66 ± 0.18	-0.04 ± 0.12

**Table 2.** Mean values ( $\pm$  SD) and deltas of variation of selected physiological (a) and biomechanical parameters (b) at baseline and after the training period for the normal training group and the overreached group. The data are presented for three running intensities determined at the end of the training program: Low (13km.h<sup>-1</sup>), Lactate Threshold (LT) and at exhaustion. Each parameter is presented for the same absolute running speed before and after the training period.  $\dot{V}O_2$ : oxygen uptake;  $\dot{V}E$ : expiratory flow; HR: heart rate; [La-]b: blood lactate concentration. Significantly different from pre-training at \* $p < 0.05$ ; \*\*  $p < 0.01$ ; \*\*\*  $p < 0.001$ . Significantly different from the normal training group at <sup>#</sup>  $p < 0.05$ ; <sup>##</sup>  $p < 0.01$ ; <sup>###</sup>  $p < 0.001$ .

Group	Number of cases	Predicted group			Correct
		NT	n-OR	OR	
NT	24	24	0	0	100%
n-OR	15	2	10	3	66.7%
OR	33	1	3	29	87.8%
Total	72	27	13	32	87.5%

**Table 3.** Classification matrix of discriminant analysis 1 using 3 groups and 21 variables (DA1). Each case represented one subject for one exercise intensity. NT: subjects of the normal training group; n-OR: subjects of the overload group demonstrating no clinical symptoms of overreaching; OR: subjects of the overreached group.

a.

Variable	Standardized coefficient
$\Delta$ HR	-0.74
$\Delta$ dS	-0.61
$\Delta$ dA	-0.58
$\Delta$ [La <sup>-</sup> ] <sub>b</sub>	-0.47
$\Delta$ Lxn	-0.44
$\Delta$ PI <sub>mn</sub>	-0.38
$\Delta$ Lyn	-0.26
$\Delta$ MuscRPE	0.23

b.

Group	Number of cases	Predicted group		Correct
		NT	OR	
NT	24	24	0	100%
OR	33	1	32	97.0%
Total	57	25	32	98.2%

c.

Step	Variable	Wilk's lambda	Significance level
1	$\Delta$ HR	0.39	0.0000
2	$\Delta$ [La <sup>-</sup> ] <sub>b</sub>	0.31	0.005
3	$\Delta$ PI <sub>mn</sub>	0.29	0.03
4	$\Delta$ dS	0.31	0.004
5	$\Delta$ dA	0.30	0.009
6	$\Delta$ Lxn	0.30	0.02
7	$\Delta$ Lyn	0.28	0.14
8	$\Delta$ MuscRPE	0.27	0.20

Table 4. Detailed results for the stepwise discriminant analysis using 2 groups and 21 variables (DA2): standardized canonical discriminant function coefficients (a); classification matrix (b) and summary table (c). NT: normal training group; OR: overreached group; HR: heart rate; [La<sup>-</sup>]<sub>b</sub>: blood lactate concentration; PI<sub>mn</sub>: normalised maximum peak vertical force; dS: support duration; dA: aerial duration; Lxn: normalised stride length; Lyn : normalised stride largeness; MuscRPE: muscular rate of perceived exertion.

Group	Number of cases	Predicted group		Correct
		NT	OR	
NT	24	23	1	95.8%
OR	33	5	28	84.8%
Total	57	28	29	89.5%

**Table 5.** Classification matrix of discriminant analysis using two groups and two variables ( $\Delta HR$ ,  $\Delta [La^-]_b$ , DA3). Each case represented one subject for one exercise intensity. NT: normal training group; OR: overreached group.