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Abstract

Background: study the acute and middle term (4 weeks training) effect of cycling training on fat blood hematological parameters, urine, fatigue, and general health in recreational well-trained cyclists.

Methods: 19 cyclists undergone 5 blood sampling: 1) before and after an incremental maximal ramp test 7 days before day 0 (D-0); 2) before and after 1 hour exhaustion trial test at baseline (D-0) and after 28 days of training (D-28). Age 34,5 years (±9,5); weight 74,87 kg (±6,6); height 177,3 cm (±5,2); BMI 26,3 (±4,9); VO2max 53,75 ml/kg/min (±6,01); km week 314,7 Km (±137,1).

Results: Acute effect was strong elevating WBC from 6,27±2,34 10^3 /ul to 9,01±3,63 10^3 /ul, an increase in LDL and Total CHOL, in this respect, existing literature is controversial. No changes in body weight or blood pressure was observed after 1 month of regular training albeit lipid profile significantly improved, as well as GOT.

Conclusions: effect of a short incremental bout of exercise was to temporary elevated all the blood parameters except MCH and MCHC. A month of intensive training (km week: 314,7 Km ±137,1) significantly improved blood lipids profile with no permanent effect on WBC, blood pressure or body weight, but improved post effort lactate concentration and fatigue perception. Hematuria is confirmed to be a rare occurrence in recreational cyclists. Data can be useful for training monitoring and comparisons with similar groups of athletes, where there is a lack of information in literature and for comparing exercise effects.

Key words: exercise tolerance, hematological test, lactate, urinalysis, cycling.

Introduction

Recreational cycling is a wide practiced recreational sport and with aging, the risk of cardiovascular diseases increase. One of the main aims of aerobic exercise is to lowering cardiovascular risk through improving blood lipids profile [1]. However, is not completely clear the effect of continuous exercise in lowering blood lipoproteins and even the effect on other blood and urine parameters in recreational sportsmen and surprisingly, there are very few studies on the topic with sometime conflicting results in the athletic populations [2]. In addition, few studies examined the alterations in many common or critical laboratory parameters in controlled (lab) conditions, using standardized and controlled protocols. Despite its diffusion, it exist few studies on the acute and long time effect of cycling in improving fat profiles of recreational cyclists, a very large category of practioners. At our knowledge, it exist only one study on the acute effect of cycling on blood lipids, that show after 90 min of cycling at 50% of VO^2peak, an increase in LDL and HDL cholesterol and a decrease in triglycerides (TRI), with no changes in total cholesterol [3]. The efficacy of different low volume (30-40 minutes , 3 times per week at 50% of HRR-heart rate reserve-for 12 weeks) training regimes in middle age male recreational bikers (+60 years old) [4] has been studied. Results show positive effects on body weight reduction and systolic and diastolic blood pressure (83±7 mmHg vs 80±5 mmHg and 140±8 mmHg vs 135±11 mmHg), CHOL, (216,3±4 mg/dl at baseline vs 210,9 ± 4,3 mg/dl) and LDL (120,6±4,2 mg/dl vs 116,9±4,7 mg/dl). Effect of aerobic exercise on lowering blood pressure has also been observed in non-athletic population undergone training [5] while metanalysis showed a significant effect of aerobic training in comparison to no effect of resistance training in decreasing plasma lipids of 0,10 -0,8 mg/dl in general population. Middle term decrease of CHOL, LDL, and TRI, with an increase in HDL of 0.05 mg/dl was also observed [6]. Other studies in young soccer players[7] showed a middle term a decrease in CHOL from 155,6±25,7 mg/dl to 151,5±8,7 mg/dl, in TRI from 88,4±41,7 mg/dl to 87,9 ±43,79 mg/dl, in LDL from 89,9±20,9 mg/dl to 86,2±10,11 mg/dl and in HDL-C from 51,4±8,4 mg/dl to 50,78±5,11 mg/dl. A meta-analytic study[8], evidenced high-intensity aerobic training resulted only in the improvement of high-density lipoprotein cholesterol. However, an explanation of these dissimilar results can be the non-homogeneity in the exercise load, kind of exercise, and level of participants. One acute effect of the exercise bout on the immune response was strong elevating white blood cells (WBC) from 6,27±2,34*10^3/ul to 9,01±3,63*10^3/ul. When comparing prerace specimens with those within 4 hours after the marathon in 32 runners, WBC counts increased (5.5 ± 0.2 *10^3/ul to 17.4±1,5*10^3/ul)[9] (Siegel, et al., 2001)[10]. Hemoglobin depletion in urine is also a rare finding after effort[11]. Transaminases (GOT and GPT) has been indicated as markers of liver (GPT) and muscle (GOT) damage [12] , thus is important to assess their behavior with exercise. Few information exist in literature about the changes in other

common blood and urine markers immediately after effort [3]. Our aims were to assess the acute (after a maximal short trial) and middle term (4 weeks, after 1 hour exhaustive endurance trial), modifications of hematological blood and urine parameters in recreational well trained recreational cyclist. We also want to assess the middle term effect of endurance training on basic health parameters of recreational cyclists, as rest heart rate, blood pressure and fatigue perception.

Materials and methods

19 amateur male well trained cyclists undergone 5 blood sampling: 1) before and after an incremental maximal ramp test till exhaustion 7 days before day 0 (D-0); 2) before and after 1 hour exhaustion trial test at baseline (D-0) and after 28 days of training (D-28). Mean age of the subjects was 34,5 years (±9,5), body weight 74,87 kg (±6,6), height 177,3 cm (±5,2), BMI 26,3 (±4,9), VO^2max 53,75 ml/kg/min (±6,01), km per week 314,7 km (±137,1). The subjects can be classified as "recreational road cyclists" [13] and ethical committee clearance was granted by Ethical committee of University of Bologna. The subjects filled a questionnaire aimed at assessing alcohol and caffeine usage, protein, carbo and fats and water consumption. Reported diet did not change over the observation period. Clinical anamnesis was performed in order to exclude any past or ongoing pathology, which could interfere with the trials and the absence of drugs intake. VO_2 max (ml/kg/min) was assessed one week prior the D-0, with an incremental ramp test on the cycloergometer (Lode Corival, The Nederlands) with ramp of 20 watts per minute until exhaustion. The VO_2max was measured during 3 min at max intensity in the exhaustion trial. After the incremental test and the exhaustion trial, blood lactate (mM/L) was measured at 3, 6 and 9 minutes, and the highest value (usually observed at 6 minute) was retained.

Fatigue protocol at the cycloergometer consisted of a light warm up followed by: 10 min at 70% of VO^2max; 9 minutes at 50% VO^2max + 1 min at 90%VO^2max x 4, ending with 3 min at VO^2max for a total of 60 min exhaustion test. Every 10 minutes the subjects were asked to rate the effort using the Borg RPE scale[14]. Heart rate was measured during the 1-hour bout and in the 10 subsequent minutes. Lactate was measured at 3, 6, and 9 min post effort, after the VO2max test (MAX) and after the fatigue protocols. In order to assess the intensity of the effort, the subjects were asked to rank the effort in the last 10 minutes of the exhaustion on a Borg 20 visual analog scale. The subject practiced the same protocol on road at least three times per week for the subsequent 28 days. Urine sampling were taken twice at rest before trials at D-0 and D-28. The following biochemical parameters were measured on fasting blood sample, collected, frozen and processed with standard laboratory methods. Parameters have been corrected for plasma volume changes. The parameters

that was collected refer to White blood cell (WBC, 10^3/ul), a general marker of inflammation, Red Blood Cell concentration (RBC, 10^6/ul), Hemoglobin (HGB, g/dl) and Hematocrite (HCT, %). Also, we considerate the Mean Corpuscular Volume (MCV, fl), Mean Content of Hemoglobin (MCH, pg), Mean Concentration of Hemoglobin Content (MCHC, g/dl), Red Blood Cell Distribution wide (RDW, %), Glicemy (mg/dl). Moreover, we measured a few parameters of organ function, as GPT (U/L), a marker used for liver and muscle damage; Transaminases YGT and GOT, marker of muscle damage[12]. Finally HDL-Cholesterol (mg/dl), total Cholesterol (CHOL, mg/dl), Triglycerides (TRI, mg/dl) and LDL-Cholesterol (LDL, mg/dl).

The following biochemical parameters were measured on urine: specific weight, pH, Albumin, Sugar, Urobiline, Bilirubin, hemoglobin, Nitrates, Acetone, Leucocitary Esterase, deposits, leucocytes. In order to avoid hemoconcentration, subjects have been kept constantly hydratated checking they can drink water ab libitum during the 1-hour bout. Blood pressure was measured at rest 5 and 10 minutes after the cessation of the exhaustion trials at D-0 and D-28. Blood pressure at rest was in normal range for all the subjects according to their age. Statistical analysis was performed using a T test for paired samples with IBM-SPSS v.20 software, with significance level set at 0,5%. All the subjects participating in the study gave their informed consent. Clearance was given by the University of Bologna ethical committee.

Results

Body weight remained unchanged over the 28 days in total group (74,87±6,48 kg D-0, 74,64±6,19 kg D-28). VO^2max measured during the last bout of 3 minutes at VO^2max during the fatigue protocol, improved significantly between D-0 and D-28 (49,8±9,93 ml/kg/min vs. 59,8± 8,7 ml/kg/min; p = 0,22). In the incremental test performed one week before D-0, VO2max was 53,75 ±6,01 ml/kg/min. Heart rate measured at minute 10 of the recovery, significantly decrease from D-0 to D-28 (105 vs 99, p = 0.04). Blood pressure (mm/Hg) at 10 minutes post exercise didn't change significantly between D-0 and D-28 (max and min: 114±16 mm/Hg and 82±13 mm/Hg vs 120±14 mm/Hg and 82±11 mm/Hg).

Urine. Specific weight at D-0 was 1013,6 ±5,9 mg and 1016±9,8 mg at D-28, without any statistically significant differences and pH remained unchanged too (5,85±0,76 and 5,7±0,76) as well as albumin, sugar, bilirubin, hemoglobin, nitrates, acetone, leucocytary esterase, microscopic deposits, leucocytes, which remained absent, while urobiline remained stable at 0.2 mg/dl after 1 month training. Only one subject showed a presence in urine of HGB at 1 month confirm that hematuria is a relatively rare post exercise finding as previously observed[11]. The results of Borg's

test indicate a close value to maximal intensity effort: D 0 19±1,5 and D 28 19±1 score points, without any significant difference. The higher value for lactate was retained and is reported in Table I. Maximum lactate concentration significantly decreased between D-0 and D-28.

[Table I]

Results for hematological parameter prior and after the incremental test are reported in Table II.

[Table II]

All parameters significantly increased except MCH and MCHC.

Results for hematological parameters at D-0 and D-28 in the pre-trial conditions are shown in Table III.

[Table III]

In the graph 1, are shown the effects of the 28 days of training.

[Graph 1]

Discussion

All parameters show an increase pre/post trial except MCH and MCHC. This result can indicate a lack of water shift into the erythrocytes during the trial, as demonstrated before[15]: we can hypothesize the "ab libitum" water drinking during the trials, is not enough to guarantee water shift in the erythrocytes, which have a limited capacity to store water, not connected with water ingestion. We find an acute increase in LDL and CHOL, contrary to other studies in marathoners who found a decrease in LDL, and no change in cholesterol[16], but according to another study which showed similar transient results in cyclists[17]. While on the long term the decrease of HDL, LDL and CHOL are in accord with others studies in young soccer players[7]. They show a decrease in CHOL from 155,6±25,7 mg/dl to 151,5±8,7 mg/dl, in LDL from 89,9±20,9 mg/dl to 86,2±10,11 mg/dl and in HDL from 51,4±8,4 mg/dl to 50,78±5,11 mg/dl. One acute effect of the exercise bout on the immune response was strong elevating WBC from 6,27±2,34*10^3/ul to 9,01±3,63*10^3/ul, according to other studies [9] which compared prerace specimens with those within 4 hours after the marathon in 32 runners who measured an increased WBC (5.5 ± 0.2 to 17.4±1,5). We observed a significant increase of HGB and HCT only the acute measurements, while other studies in marathoners didn't observed hematocrit and hemoglobin changes[10]. Contrary to findings after a marathon run[18] , in our study RBC and MCV increased, this seems a characteristic of cycling effort. Depending on the study, hematocrit decreased[18],[19] not changed[20] or increased [10,21] after a marathon. Besides water loss, the increase in hematocrit can be attributed to the breaking of red blood cells in

159 the foot's plantar circulatory bed during running[22] and even if this is not the case of cycling, a
160 certain degree of compression on the plantar surface of the feet is present. In addition, the
161 controversial observations in previous studies showing an RBC (hemoconcentration) increase,
162 hypothesized it can be due to differences in fluid intake and environmental conditions (e.g. hot). In
163 our study, where subjects were allowed (and controlled) to drink ab libitum, we observed a
164 significant acute increase in all hemoglobin parameters.

165 We observed on a long term a positive effect on the lipid profile and glycemic indexes, as
166 confirmed in previous studies[2]. The acute increase of WBC observed pre-post trial at D-0, was not
167 present after one month of training, being transitory. Probably a longer period is necessary to
168 observe a lasting effect on WBC. It was observed[23] an increase in WBC, which is an important
169 indicator of immune function, after 1 year of training in recreational soccer players. Mean
170 hemoglobin content also show an increase on the long term. GOT, MCV and MCH showed a
171 significant difference at 1 month. Being GOT a marker of muscle damage[12], a reduction in this
172 parameter, indicate a better state of training.

173 **Conclusions**

174 Recreational cycling population is an at risk group for cardiovascular disease, mainly due to
175 overweight. Accumulation of arterial cholesterol and triglycerides is a co-factor in heart diseases.
176 For this reason and because is a low impact activity, cycling is highly recommended to middle aged
177 men. Aging is also associated with increased visceral fat and body weight. There are few data in the
178 literature about normal hematological values for this population of recreational sportsmen, and
179 contradictory results on the changes of basic hematological parameters with training, despite the
180 large diffusion of this recreational activity. In our study, we observed an acute post exercise
181 increase of GLI, TRI, HDL-C and LDL-C and CHOL, WBC, and hematocrit, differently from other
182 studies who show an increase in LDL, HDL and a decrease in TRI and no acute changes in CHOL
183 in runners [3]. Endurance trials of running and cycling seems thus to have a different acute effect on
184 acute blood fat turnover. On the long term, we observed a decrease in plasma CHOL, HDL and
185 LDL, and a slight increment in the MCV and MHC, without any significant change in the others
186 blood parameters. We did not observe, except in one case, hematuria, confirming it is a rare event
187 as previously observed[11] . Metanalysis studies[8] show that high-intensity aerobic training results in
188 improvement only in high-density lipoprotein cholesterol in normal population. However, when
189 considering athletic populations, the improvement affects all the lipid profile, and we confirm this
190 finding. This result can be ascribed to the strong biochemical reactions triggered by training. We
191 provided the evidence that recreational cycling has a beneficial long-term effect on the lipid profile
192 in a different way respect to running, even if we did not observed any positive effect on further

blood pressure reduction or fatigue perception, probably because our subjects have reached their optimal values. Cycling seems to differ from running in the effect on hematological parameters. The data provided can also be useful for the comparison with other groups of trained recreational cyclists. A limitation of the present study is a lack of control on the diet of the subjects during the study period, which could have affected some hematological parameters. Further studies on recreational cyclists, should clarify the effective exercise protocols able to improve the lipid profile.

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Notes

The authors have no conflicts of interest.

Acknowledgements

We want to express our most sincere sympathies to our colleague Andrea Sapone who passed away, and his family. Professor Sapone A., who has collaborated with us during this study, was a highly valuable and respected member of our team

	MAX	G-0	G-28
Total group	14,63±3	9,65±2,74	7,35±3*

Tab. 1. Max Blood lactate (mM/L) measured during the ramp test, after exhaustion test at baseline (G-0) , (G-1) and after 28 days (G-28) of training, in all subjects * p = 0,14;

Index	PRE	POST	p value
GLI	80,95±15,59	98,30±14,79	0,001*
YGT	19,79±7,33	21,21±6,77	0,000*
GOT	26,20±8,10	34,45±8,95	0,000*
GPT	23,40±11,13	40,80±15,16	0,000*
HDL	62,14±15,45	67,84±18,45	0,000*

CHOL	182,55±22,47	190,70±33,55	0,093
TG	81,20±49,67	84,70±32,95	0,269
LDL	88,64±16,89	94,86±21,23	0,010*
RBC	4,85±0,39	5,11±0,35	0,000*
HGB	14,77±0,92	15,36±0,85	0,000*
HCT	45,11±3,08	47,31±2,61	0,000*
MCV	92,71±4,34	92,79±4,35	0,356
MCH	30,39±1,53	30,13±1,34	0,016*
RDW	13,26±0,80	13,3±30,82	0,104
WBC	6,27±2,34	9,01±3,63	0,000*

Tab 2. Ematochemistry PRE-POST 1 hour intensive cycling on the cycloergometer in the total sample.(G 0).

GLI: Glicemia, mg/dl; YGT, GOT and GPT: Transamynases, U/L; HDL: HDL-Cholesterol, mg/dl; CHOL: total Cholesterol, mg/dl; TRI: Trygliceride mg/dl; LDL: LDL-Cholesterol, mg/dl.RBC : Red Blood Cell , 10^6/ul ; HGB: Hemoglobin, g/dl; HCT: Hematocrit , %; MCV Mean Corpuscular Volume, fl; MCH: Mean Content of Hemoglobin, pg; MCHC: Mean Concentration of Hemoglobin Content, g/dl; RDW: Red Blood Cell Distribution Wide, %; WBC : White blood cell, 10^3/ul. * significantly different

	G-0	G-28	Sig. T
GLIC	80,95±19,59	73,95±19,84	0,141
YGT	19,79±7.33	19,35±7.09	0,818
GOT	26,20±8,10	24,30±6,42	0,122
GPT	23,40±11,13	22,30±5,69	0,55
HDL	61,14±15,45	58,52±15,12	0,095*
CHOL	182,55±22,47	164,00±40,78	0,065*
TRI	81,2±49,67	87,25±64,96	0,817

LDL	88,64±16,89	83,32±17,33	0,083*
RBC	4,84±0,37	4,86±0,42	0,827
HGB	14,71±0,91	14,79±1,15	0,888
HCT	44,96±3,06	45,13±3,56	0,952
MCV	92,52±4,30	93,04±4,44	0,09*
MHC	30,3±1,51	30,49±1,51	0,462
MCHC	32,75±0,57	32,77±0,64	0,87
RDW	13,29±0,79	13,26±0,86	0,969
WBC	6,27±2,34	6,01±1,46	0,604

Tab. 3. Ematochemical parameters at G0 and G28, in the pre-TGal condition. * significantly different

GLI: Glicemia, mg/dl; YGT, GOT and GPT: Transamynases, U/L; HDL: HDL-Cholesterol, mg/dl; CHOL: total Cholesterol, mg/dl; TRI: Tryglicerides mg/dl; LDL: LDL-Cholesterol, mg/dl.RBC : Red Blood Cell , 10^6/ul ; HGB: Hemoglobin, g/dl; HCT: Hematocrite , %; MCV Mean Corpuscular Volume, fl; MCH: Mean Content of Hemoglobin, pg; MCHC: Mean Concentration of Hemoglobin Content, g/dl; RDW: Red Blood Cell Distribution Wide, %; WBC : White blood cell, 10^3/ul. * significantly different