Run for Science (R4S): the history of a successful project of precision and laboratory medicine in sport and exercise

Giuseppe Lippi[1], Federico Schena[2]

[1]Section of Clinical Biochemistry, University of Verona, Verona, Italy; [2]School of Sport and Exercise Sciences, University of Verona, Verona, Italy

Contributions: (I) Conception and design: All authors; (II) Administrative support: None; (III) Provision of study materials or patients: None; (IV) Collection and assembly of data: All authors; (V) Data analysis and interpretation: All Authors; (VI) Manuscript writing: G Lippi; (VII) Final approval of manuscript: All authors.

Correspondence to: Prof. Giuseppe Lippi. Section of Clinical Biochemistry, University Hospital of Verona, Piazzale LA Scuro, 37100, Verona, Italy. Email: giuseppe.lippi@univr.it.

Abstract: Since ancient times sports has drawn the attention of scientists from a large number of disciplines, including social sciences, physiology, medicine and rehabilitation. Although it is now unquestionable that regular performance of physical exercise is associated with a lower risk of developing many human diseases such as cancer, cardiovascular disorders, diabetes, rheumatic diseases, sarcopenia and frailty, some issues remain to be addressed. These typically include the amount of exercise needed to improve health and fitness without overcoming the putative health befits, along with the insufficient recognition of biological pathways that can be modulated by active lifestyle. The still incomplete knowledge of these aspects is contributing to the growth of several scientific initiatives, specifically aimed to study changes and individual adaptations underlying regular sport practice. The bases, context and early achievements of one of such initiatives, called "Run for Science", will be summarized in this article. In particular, the various editions of this event have allowed to make new discoveries and tangible advancements in human biology and personalized laboratory medicine, including the observation that cardiac troponins transitorily increase in blood after endurance exercise, that running performance is influenced by two previously unappreciated laboratory parameters (i.e., serum alpha-amylase and mean platelet volume) and that the DNA may undergo a transitory oxidative damage during exercise. More recently, the information gathered from the Run for Science helped discovering important aspects linking physical exercise with the lower risk of cancer, cardiovascular disease and asthmatic reactions. Additional analyses are underway, and specific focus will be placed on sports epigenetics, with the aim to define how recreational sport interplays with gene expression and modifies the individual response to exercise.

Keywords: Sport; physical activity; laboratory medicine; personalized medicine

Received: 28 March 2017; Accepted: 15 April 2017; Published: 27 April 2017.
doi: 10.21037/jlpm.2017.04.01
View this article at: http://dx.doi.org/10.21037/jlpm.2017.04.01

Sport, fitness and wellbeing

The history of sports is probably as old as mankind. Some cave paintings found in the Lascaux caves (France), which can be dated back to nearly 15,300 years ago, clearly represent sprinting and wrestling competitions taking place sometime in the Upper Paleolithic Era (1). Throughout the following periods of human history, and more precisely since the organization of the ancient Olympics games in Athens in the eight century BC, sport has then become an important social institution, a mass phenomenon, and even a business-generating industry. Notably, the broad diffusion of sports around the globe has also importantly boosted the development of the so called "sports medicine" or "exercise medicine", whose origins can be traced to the early Hindu and Chinese culture, throughout Hippocrates and Galen, up to recent times (1).

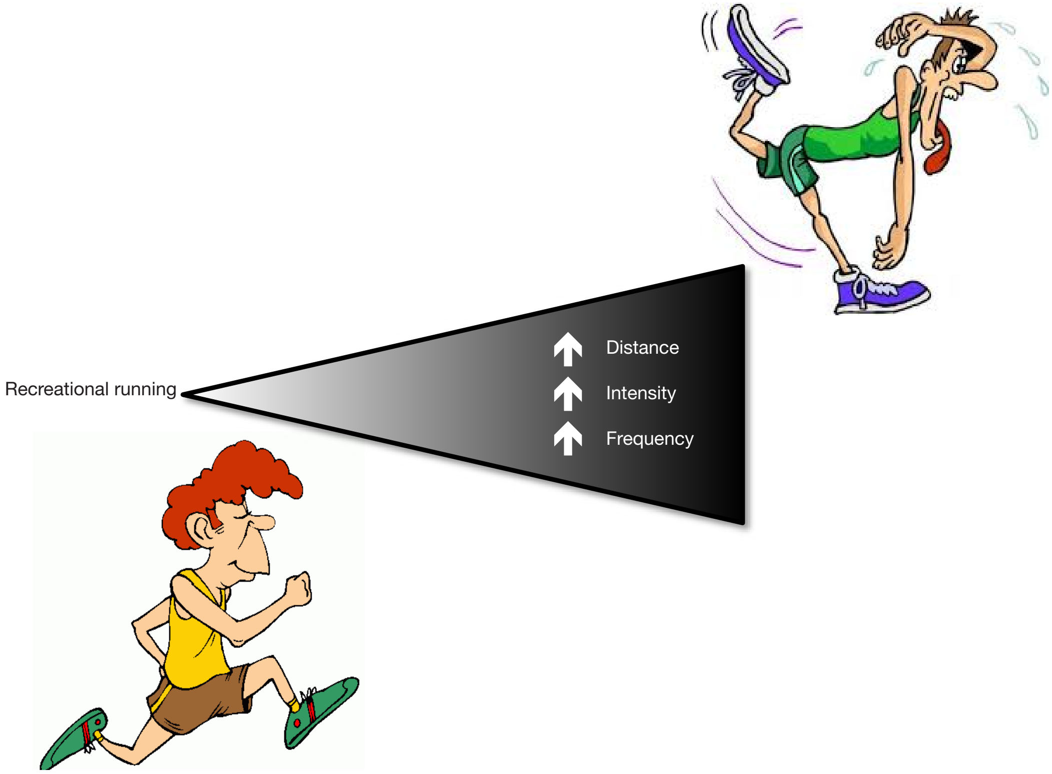

Figure 1 The challenging compromise in running distance, intensity and frequency for balancing health benefits and the risks of excessive exercise.

Since ancient times sports has drawn the attention of scientists from a large number of disciplines, including social sciences, physiology, medicine and rehabilitation. The interest of science and medicine in sports physiology is ample and multifaceted, so entailing technical research and physiological studies for optimizing or improving athletic performance, efficient management of sport injuries and recovery, along with efforts made for identifying the many biological benefits that sport may have on health and fitness (2). Since the beginning of the past century, exercise has became central to public health, and a large body of evidence has accumulated to suggest that the several biological pathways activated (or inhibited) by a physically active lifestyle may produce immense health benefits (3). It is hence now unquestionable that regular performance of physical exercise has a graded and inverse association with the risk of developing a kaleidoscope of human diseases, thus including cancer (4), cardiovascular disorders (5), diabetes (6), osteoporosis (7), rheumatic diseases (8), sarcopenia and frailty (9).

Although sports medicine has come a very long way in recent decades, and continuous research and advancements in this field are abundantly clear, some issues remain to be addressed. These typically include the minimum amount of exercise needed to improve health and fitness, the maximum allowable bout of exercise before the putative health benefits may turn into biological damage, as well as the recognition of biological pathways that can be positively modulated by exercise so that they could be directly targeted by specific health interventions and/or treatments (10) (*Figure 1*). The still incomplete knowledge of these aspects is hence contributing to the growth of several scientific initiatives and events, specifically aimed to study changes and adaptations underlying regular sport practice. The bases, context and early achievements of one of such initiatives, called "Run for Science", will be summarized in the following sections of this article.

The "context" of the Run for Science

The Run for Science, abbreviated R4S, is a research project originally conceived in the mid 2000s and officially endorsed

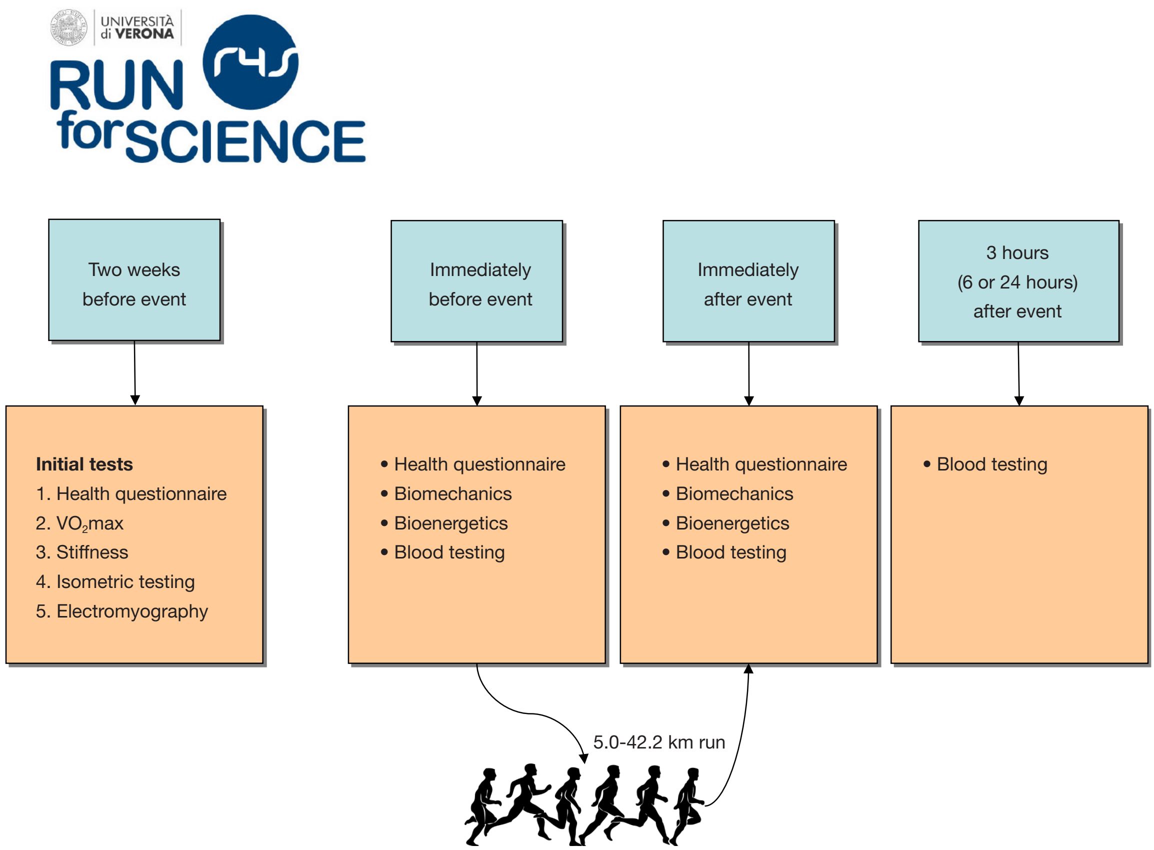

Figure 2 Typical program and organization of the "Run for Science". VO_2max, maximal oxygen uptake.

by the University of Verona with its definitive designation in 2014. The event, which has increasingly involved Italian, European and American scientific institutions, has been organized annually for the past four years, and the last edition was held in April 2, 2017. The purpose of this project is to investigate several biochemical, metabolic and physical aspects regarding endurance running, basically entailing the evaluation of muscle efficiency along with the assessment of psychological, cardiovascular and metabolic adaptation. Middle distance running has been recognized as the most suitable type of exercise for these studies, since it does not require specific talent or highly-specialized and expensive equipment, can be practiced in almost every environmental condition, and is hence the most affordable and convenient sport discipline for the general public.

The annual initiative is usually organized between the end of March and the beginning of April, to meet the most suitable conditions of temperature and humidity for running. The event typically involves more than 200 volunteers, aged between 20 to 90 years, and entails anthropometric measurements (i.e., weight, height, body composition), cognitive assessment, conditional adaptations (strength, elasticity, sensitivity), functional measures (energy expenditure and optimization), individual running performance and biochemical testing (i.e., blood sampling) before and after the trial. The day of the event the amateur athletes will be running an endurance distance (according to the different study protocols, this may vary between 5.0 and 42.2 km, but most athletes will compete a 21.1 km, half-marathon run). Before being enrolled in the study, all the volunteer subjects need to provide a written consent. The study has been originally approved by the local Ethical Committee (Department of Neurological, Neuropsychological, Morphological and Movement Sciences, University of Verona) and is performed in accordance with the Helsinki Declaration.

The typical program and organization of the Run for Science are shown in *Figure 2*, and are also available in a dedicated website (11). As regards the four previous editions officially endorsed by the University of Verona as

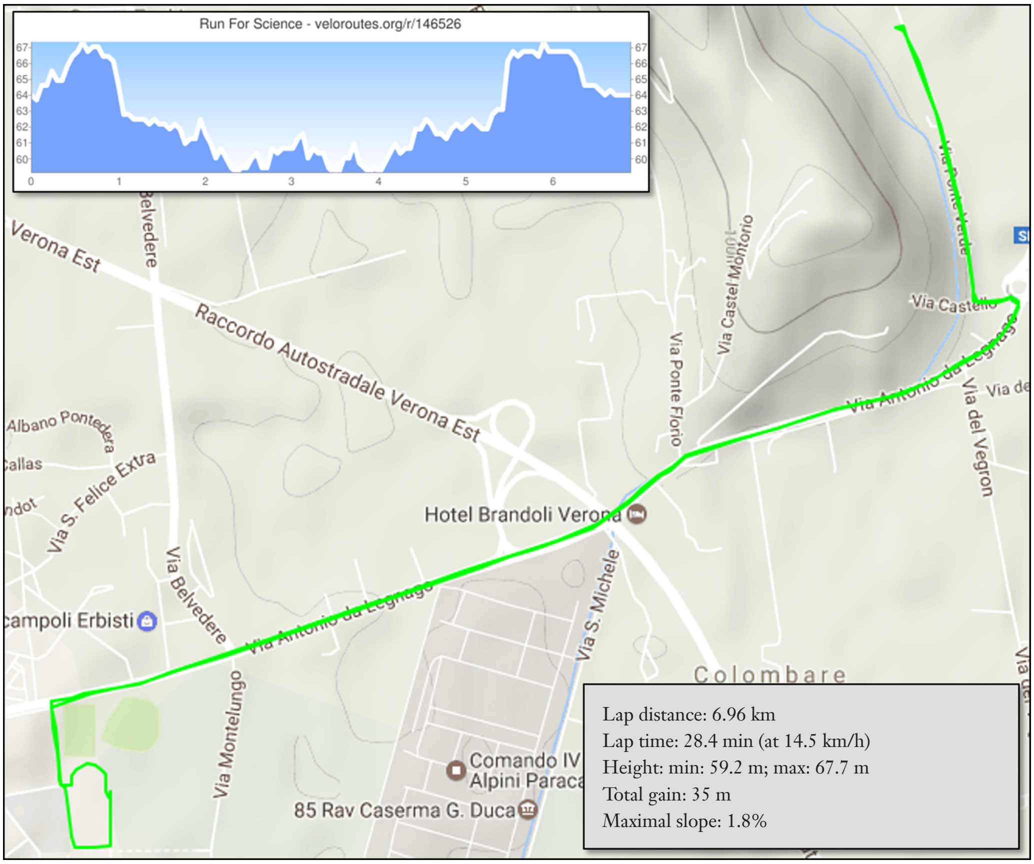

Figure 3 Description of the route of the "Run for Science".

"Run for Science" (i.e., between 2014 and 2017), data were prevalently collected from athletes performing 21.1 km (half-marathon) or a 42.2 km (marathon) runs, equipped with a heart rate (HR) monitor, and running at exercise intensity comprised between 75% and 85% of their maximal oxygen uptake (VO_2max). The study population consists of recreational athletes engaged in non-professional teams of amateur runners (training regimen usually ranging between 180 and 260 min per week; VO_2max comprised between 40–60 mL/kg/min; body mass index ranging between 20 and 30 kg/m^2). Maximal aerobic capacity is measured in each subject approximately two weeks before the event, by means of a running test on treadmill and using breath by breath ergospirometric system. After familiarization, all runners are requested to undergo a progressive incremental test, starting from the habitual running pace and then increasing the speed by 0.5 km/h every one minute, until reaching volitional exhaustion. The study project requires the subjects to rest for at least 36 hours before the event, avoiding intake of medications known to alter endurance performance or biochemical and metabolic profile, and avoiding ingestion of food for 3 hours after the run. The trial usually starts at around 9.00 AM and the route always develops on a virtually identical circuit. Briefly, the ~7 km circuit is located 5 km from the center of the town of Verona (Italy), and develops with a maximum of 35 m vertical gain (1.8% maximal slope) (*Figure 3*). Participants are free to drink ad libitum during the run, but are not allowed to ingest food until a second blood sample has been taken after crossing the finishing line. The median time of completing the 21.1 km trial is around 115 min (range usually comprised between 100–140 min). Blood samples are always taken at baseline (i.e., 10 to 15 min before the start of the run), immediately after crossing the finishing line (i.e., post-run), as well as 3 hours after the end of the run. On occasion, and according to the specific study design, additional samples may also be collected (i.e., 6 and 24 hours after the end of the run). The blood samples are immediately transferred to the laboratory of clinical chemistry and hematology of the University of Verona, within specific transport boxes, under controlled conditions of time and temperature. Upon arrival in the laboratory, the samples needing immediate testing are rapidly analyzed (i.e., for obtaining the complete blood count), whereas those requiring preparation for obtaining serum and different types of plasma (i.e., EDTA, lithium-heparin or citrate plasma) are centrifuged, divided in several aliquots and stored at −70 °C. This organization has allowed the construction of a large biorepository of biological samples over time, from which the specimens can be easily retrieved when additional studies are planned. Preanalytical variability is always seen as a critical issue in clinical trials (12), so that the highly standardized and strictly monitored procedures used for collecting blood and for sample management should be considered peculiar strengths of the Run for Science initiative.

Beside being officially endorsed by the University of Verona, many other partners have been increasingly involved, so that the current team comprises scientists from the University of Torino (Italy), University of Rome (Italy), University of Brescia (Italy), University of Milan (Italy), University Hospital of Parma (Italy), the Research Center Mountain Sports and Health (CeRiSM) (Italy), Rome Foro Italico (Italy), New York University School of Medicine (US), German Sport University in Cologne (Germany) and University of Valencia (Spain).

Former achievements

Since the 2007, when the first official event has been held, up to the 2017 (i.e., the date of the last edition), the Run for Science has engaged over 1,000 recreational athletes, thus allowing to achieve extraordinary successes in physiology and laboratory sport medicine, which were especially useful for better identifying and interpreting the short- and medium-term variations of many biochemical and hematological tests in response to exercise, and permitting to unravel previously overlooked pathways in health and disease. Most of the important scientific breakthroughs that have been made in the various editions of the R4S are summarized in *Table 1* (13-28). Some of these represent new discoveries or tangible advancements over former scientific knowledge. This especially refers to the observation that the concentration of cardiac troponins measured with highly-sensitive immunoassays transitorily increases after endurance exercise, that the running performance is influenced by two previously unappreciated laboratory parameters (i.e., serum alpha-amylase and mean platelet volume), and that the DNA may undergo a transitory oxidative damage during endurance exercise. More recently, the results of the R4S helped discovering important aspects linking physical exercise with a lower risk of cancer (i.e., acute decrease of cancerogenic bile acids in the circulation), cardiovascular disease (i.e., short- and long-term reduction of low-density lipoproteins) and asthmatic reactions (i.e., acute decrease of circulating eosinophils in blood). Additional analyses are underway, as allowed by the large availability of biological specimens obtained in the various editions of the event.

Future perspectives and conclusions

Although the various editions of the Run for Science have allowed to achieve some important results for understanding the personal response to exercise and for identifying a number of individual determinants of performance (*Table 1*), we have already planned future studies in the field of personalized laboratory medicine. In particular, specific focus is being placed on sports epigenetics, with the aim to define how recreational sport interplays with gene expression and modulates the individual response to exercise (29). Running economy and performance both depend on many physiological factors (e.g., anthropometric variables, functional characteristics, volume and intensity of training) but also exhibit high inter-individual variations, which are not fully explainable by sport physiology. Moreover, the health benefits of physical exercise may differ from one subject to another, so that a more accurate understanding of individual changes will be necessary to define personalized strategies aimed to lowering the risk of many human disorders (30). Therefore, a deep search of gene-to-environment interplay, so including physical exercise, will probably offer the most attracting opportunity for a tailored approach in prevention and care of many human diseases (31), but may also offer new opportunities for the global struggle against doping, which is not limited to top-class athletes but deeply plagues also recreational sports (32).

In the long and windy road towards personalized (precision) laboratory medicine, we really hope that the many important findings obtained during the various editions of the Run for Science may allow the replication of this project at other latitudes and in different countries, to help establishing a new paradigm for assessing individual biochemical responses to endurance exercise.

Table 1 Summary of the most important findings emerged from the various editions of the "Run for Science" (13-28)

Parameter	Variation after the run			
	Immediately after	3 hours after	24 hours after	Additional findings
DNA double-strand breaks	↑	N/A	N/A	
Alkaline phosphatase (ALP)	↑	–	–	
Alanine aminotransferase (ALT)	↑	–	–	
Aspartate aminotransferase (AST)	↑↑	↑	↑	
Alpha-amylase	↑	–	–	Predicts running performance
B-type natriuretic peptide (BNP)	↑↑↑	↑↑	↑	
Bile acids	↓	N/A	N/A	
Bilirubin	↑↑	↑↑↑	↑	
Cardiac troponins	↑↑	↑↑↑	↑	
Creatine kinase (CK)	↑↑	↑↑↑	↑↑↑	
Gamma-glutamyl transferase (GGT)	↑	↑	–	
Glomerular filtration rate (GFR)	↓↓↓	↓	–	
Iron	↑↑	N/A	N/A	
Lactate dehydrogenase (LDH)	↑↑	↑↑↑	↑	
Low density lipoproteins (LDL)	↓	N/A	N/A	
Lipoprotein(a)	–	N/A	N/A	
Myoglobin	↑↑	↑↑↑	↑	
Osteocalcin	↑	–	–	
Parathyroid hormone (PTH)	↑↑/	–	–	
Total cholesterol	↓	N/A	N/A	
Triglycerides	↑	N/A	N/A	
Urea	↑	N/A	N/A	
Uric acid	↑↑	N/A	N/A	
White blood cells (WBC)	↑↑	↑↑↑	–	
Neutrophils	↑↑	↑↑↑	–	
Lymphocytes	↓	↓	–	
Monocytes	↑	↑↑	–	
Eosinophils	↓↓	↓↓↓	–	
Basophils	↑	↑↑	–	
Red blood cells	↓↓	↓↓	↓	
Hemoglobin	↓	↓	↓	
Reticulocytes	–	↓	↓	
RDW	↑	N/A	N/A	
Platelets	↑↑	–	–	
Mean platelet volume (MPV)	↑↑	–	–	Predicts running performance

↑, increased compared to baseline; ↑↑, highly increased compared to baseline; ↑↑↑, extremely increased compared to baseline; ↓, decreased compared to baseline; ↓↓, highly decreased compared to baseline; ↓↓↓, extremely decreased compared to baseline; –, unvaried compared to baseline; NA, not assessed; RDW, red blood cell distribution width.

Acknowledgments

Funding: None.

Footnote

Conflicts of Interest: Both authors have completed the ICMJE uniform disclosure form (available at http://dx.doi.org/10.21037/jlpm.2017.04.01). Giuseppe Lippi serves as the unpaid Editor-in-Chief of *Journal of Laboratory and Precision Medicine* from November 2016 to October 2021. The authors have no other conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

References

1. Snook GA. The history of sports medicine. Part I. Am J Sports Med 1984;12:252-4.
2. Berryman JW. Exercise is medicine: a historical perspective. Curr Sports Med Rep 2010;9:195-201.
3. Lippi G, Banfi G, Botrè F, et al. Laboratory medicine and sports: between Scylla and Charybdis. Clin Chem Lab Med 2012;50:1309-16.
4. Sanchis-Gomar F, Lucia A, Yvert T, et al. Physical inactivity and low fitness deserve more attention to alter cancer risk and prognosis. Cancer Prev Res (Phila) 2015;8:105-10.
5. Carnethon MR. Physical Activity and Cardiovascular Disease: How Much is Enough? Am J Lifestyle Med 2009;3:44S-49S.
6. Gill JM, Cooper AR. Physical activity and prevention of type 2 diabetes mellitus. Sports Med 2008;38:807-24.
7. Borer KT. Physical activity in the prevention and amelioration of osteoporosis in women: interaction of mechanical, hormonal and dietary factors. Sports Med 2005;35:779-830.
8. Holla J, Fluit M, van Schaardenburg D, et al. Recreational exercise in rheumatic diseases. Int J Sports Med 2009;30:814-20.
9. Iolascon G, Di Pietro G, Gimigliano F, et al. Physical exercise and sarcopenia in older people: position paper of the Italian Society of Orthopaedics and Medicine (OrtoMed). Clin Cases Miner Bone Metab 2014;11:215-21.
10. Lippi G, Schena F, Guidi GC. Health benefits of physical activity. CMAJ 2006;175:776.
11. Run for Science. Available online: http://www.r4s.it/info/. Last accessed, 27 March, 2017.
12. Lippi G, Simundic AM, Rodriguez-Manas L, et al. Standardizing in vitro diagnostics tasks in clinical trials: a call for action. Ann Transl Med 2016;4:181.
13. Lippi G, Schena F, Salvagno GL, et al. A half-marathon run does not acutely influence lipoprotein(a). Nutr Metab Cardiovasc Dis 2008;18:e37-8.
14. Lippi G, Schena F, Montagnana M, et al. Influence of acute physical exercise on emerging muscular biomarkers. Clin Chem Lab Med 2008;46:1313-8.
15. Lippi G, Schena F, Montagnana M, et al. Acute variation of osteocalcin and parathyroid hormone in athletes after running a half-marathon. Clin Chem 2008;54:1093-5.
16. Lippi G, Schena F, Salvagno GL, et al. Acute variation of estimated glomerular filtration rate following a half-marathon run. Int J Sports Med 2008;29:948-51.
17. Lippi G, Schena F, Salvagno GL, et al. Acute variation of biochemical markers of muscle damage following a 21-km, half-marathon run. Scand J Clin Lab Invest 2008;68:667-72.
18. Lippi G, Schena F, Salvagno GL, et al. Influence of a half-marathon run on NT-proBNP and troponin T. Clin Lab 2008;54:251-4.
19. Lippi G, Banfi G, Montagnana M, et al. Acute variation of leucocytes counts following a half-marathon run. Int J Lab Hematol 2010;32:117-21.
20. Lippi G, Schena F, Montagnana M, et al. Significant variation of traditional markers of liver injury after a half-marathon run. Eur J Intern Med 2011;22:e36-8.
21. Lippi G, Schena F, Dipalo M, et al. Troponin I measured with a high sensitivity immunoassay is significantly increased after a half marathon run. Scand J Clin Lab Invest 2012;72:467-70.
22. Lippi G, Salvagno GL, Danese E, et al. Variation of red blood cell distribution width and mean platelet volume after moderate endurance exercise. Adv Hematol

2014;2014:192173.
23. Lippi G, Salvagno GL, Danese E, et al. The baseline serum value of α-amylase is a significant predictor of distance running performance. Clin Chem Lab Med 2015;53:469-76.
24. Salvagno GL, Lippi G, Tarperi C, et al. Iron concentration increases after moderate endurance exercise: implications for screening of blood transfusion in sports. Drug Test Anal 2015;7:346-7.
25. Lippi G, Salvagno GL, Danese E, et al. Mean platelet volume (MPV) predicts middle distance running performance. PLoS One 2014;9:e112892.
26. Lippi G, Buonocore R, Tarperi C, et al. DNA injury is acutely enhanced in response to increasing bulks of aerobic physical exercise. Clin Chim Acta 2016;460:146-51.
27. Boccia G, Dardanello D, Tarperi C, et al. Decrease of muscle fiber conduction velocity correlates with strength loss after an endurance run. Physiol Meas 2017;38:233-40.
28. Danese E, Salvagno GL, Tarperi C, et al. Middle-distance running acutely influences the concentration and composition of serum bile acids. Potential implications for cancer risk? Oncotarget 2017. doi: 10.18632/oncotarget.17188
29. Danese E, Benati M, Sanchis-Gomar F, et al. Physiological determinants of urine and plasma myomiRNAs in recreational, middle-age athletes. J Lab Prec Med 2017. doi: 10.21037/jlpm.2017.04.04
30. Perez-Quilis C, Lippi G, Mena S, et al. PAFIYAMA syndrome: prevention is better than cure. J Lab Precis Med 2016;1:8.
31. Lippi G, Bassi A, Bovo C. The future of laboratory medicine in the era of precision medicine. J Lab Precis Med 2016;1:7.
32. Lippi G, Mattiuzzi C. Doping and anti-doping testing in sports: are we only pointing at the bright side of the moon? J Lab Precis Med 2017;1:8.

doi: 10.21037/jlpm.2017.04.01
Cite this article as: Lippi G, Schena F. Run for Science (R4S): the history of a successful project of precision and laboratory medicine in sport and exercise. J Lab Precis Med 2017;2:11.