The prevalence of adverse analytical findings in european antidoping laboratories: monitoring and analysis in the Athens and **London Olympic Games**

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Summary

Introduction: Since 2003, the World Anti-Doping Agency (WADA) begins to provide annual public reports which informs about all the analysis performed and the adverse analytical findings (AAF) determined in the different accredited laboratories. **Objectives:** To identify the European laboratories and the most used substances for doping purposes, in addition to relate the adverse analytical findings (AAF) in European laboratories over three different periods of time (pre-Olympics, Olympics and post-Olympics).

Methods: Cohort study, following the recommendations of the STROBE declaration of the reports collected by the WADA between 2003-2015. The data belong to 16 European laboratories accredited by the WADA distributed in 11 groups of substances considered as doping substances. Inclusion criteria: detectable substances through the urine. Exclusion criteria: laboratories that between 2003-2015 were temporarily or definitively suspended by the WADA or appearance after 2004. The variables of years were transformed into pre-Olympics, Olympics and post-Olympics of the Olympic Games of Athens (2004) and London (2012), because both competitions were carried out in Europe.

Results: In the last 12 years reported, the most detected substance by European laboratories has been anabolic substances (52.42%), being the laboratory of Moscow (Russia) which presents the highest detection rate of this substance (3 out of 4 AAF). It is related the increase in the detection of cannabis in the European laboratories with post-Olympics periods (p=0,0001).

Conclusions: The laboratory with the highest proportion of AAF reports is Ghent (Belgium). Anabolic steroids are the most commonly detected substance in all the laboratories. There is a relationship between the detection of adverse analytical

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Medical statistics.

findings of cannabis in post-Olympics periods and the detection of anabolic steroids in pre-Olympics and Olympics periods.

Prevalencia de los hallazgos analíticos adversos en los laboratorios antidopaje europeos: análisis y seguimiento en los Juegos Olímpicos de Atenas y Londres

Resumen

Introducción: A partir del año 2003, la Agencia Mundial Antidopaje (AMA) comienza a emitir anualmente informes de carácter público donde se informa de todos los análisis realizados y los hallazgos analíticos adversos (HAA) encontrados en

Objetivos: Identificar los laboratorios europeos y las sustancias prohibidas mayormente reportadas, además de relacionar los HAA en los laboratorios europeos con tres periodos de tiempo diferentes (preolimpiadas, olimpiadas y postolimpiadas). Métodos: Estudio de tipo cohortes, siguiendo las recomendaciones de la declaración STROBE de los informes reportados por la AMA entre los años 2003-2015.Los datos estudiados pertenecen a 16 laboratorios europeos y 11 grupos de sustancias consideradas dopantes. Inclusión: sustancias detectables a través de la orina. Exclusión: tantos los laboratorios que entre 2003-2015 fueran suspendidos temporal o definitivamente por la AMA en Europa, como los de aparición posterior a 2004. Se transformaron las variables de años en preolímpicos, olímpicos y postolímpicos de los Juegos Olímpicos de Atenas (2004) y Londres (2012), por realizarse ambas competiciones en Europa.

Resultados: La sustancia más detectada por los laboratorios europeos en los últimos 12 años reportados han sido los anabolizantes (52,42%), siendo el laboratorio de Moscú (Rusia) el que mayor detección en dicha sustancia presenta (3 de cada 4 HAA). Se relaciona el aumento de la detección del cannabis en los laboratorios europeos con periodos postolímpicos (p=0,0001). Conclusiones: El laboratorio europeo que proporcionalmente detecta mayor número de HAA es Ghent (Bélgica). Los anabolizantes son la sustancia mayormente detectada en todos los laboratorios. Existe una relación entre la detección de HAA de cannabis en periodos postolímpicos y de anabolizantes en periodos preolímpicos y olímpicos.

Palabras clave:

Dopaje. Control antidopaje. Epidemiología. Estadística médica.

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Introduction

Each year increasingly sophisticated detection methods appear in the fight against doping, which, along with improvements in education and research, promote the anti-doping campaign and the harmful effects of doping on human health, integrity and the fundamental values of participating in sport^{1,2}. For this reason, the WADA produces a list of doping substances, which is modified and re-edited each year due to constant research into substances, methods and technological advances that may alter the health of athletes³.

The World Anti-Doping Agency (WADA) was created in 1999, and emerged as a private foundation subject to Swiss law. Its main objective is to promote and coordinate the fight against doping in sport on an international level. Since 1999 anti-doping controls have been regulated, but it was not until 2005 that a regulation protocol was created, validating the compulsory nature of the World Anti-Doping Code⁴.

From 2003, the WADA began to distribute annual public reports, giving information about all the analyses carried out in all WADA-accredited laboratories. These reports can be obtained via the ADAMS IT programme (Anti-Doping Administration & Management System), which was created with the purpose of coordinating anti-doping control activities and managing the location of athletes, both in and out of competitions^{5,6}.

Due to the major social and economic involvement of sport in our society, success has become increasingly important for athletes, clubs and trainers, who aim to renew contracts and continue to participate among the sporting elite, and who often even turn a blind eye to established regulations^{7,8}. The physical and mental demands of top-level sport mean that some athletes turn to the consumption of illegal substances in the quest to improve their physical performance. Considered the most important international competition for sport in general, the Olympic Games (OG) have become a unique scenario for investigating potential illegal substance abuse activities in professional sport.

Until now the relationship of AAF in high level competitions such as the OG has not been researched, as research targeting this kind of event is fundamentally based on the economic impact and repercussions in the different countries where these competitions are held9-11, on sociosanitary aspects such as the propagation of the Zika virus in the Rio de Janeiro OG in 2016¹¹, or on health risk factors in mass sporting events¹². To develop new tools to detect illegal substances, it is important to know which are the most commonly used substances in doping, investigating new methods of fighting and detecting substances and illegal methods. To do this it is essential to know the prevalence of illegal substance abuse in sports as well as its geographic distribution. Identifying the most commonly used group of substances by athletes will enable us to discover the most frequent attitudes towards doping held by those that aim to deceive, and will give us a useful prevention tool. On the other hand, discovering the distribution of the different substances by years could be interesting, as we already know that in the years leading up to the Olympics, even the same year, classifying championships are held for participation in the OG. For optimum athlete performance, a good training planning is essential, based on objectives and the championships in which he/she wishes to participate¹³. This way, and knowing that anti-doping controls are more numerous in this kind of competition, we could find out if there is a relationship between the different years and the AAF in Europe.

Therefore, the study objectives are to describe the most detected WADA-banned substances; to identify accredited European laboratories that detect the largest number of AAF; and to find out the proportion of AAF in terms of European anti-doping controls between 2003 and 2015; as well as relating and defining the detection of AAF with pre-Olympic, Olympic and post-Olympic years in European anti-doping laboratories.

Material and method

Study design

An observational, analytical, longitudinal and retrospective cohort study was carried out - following the recommendations of the STROBE¹⁴ statement - of the reports given by the WADA between 2003-2015¹⁵⁻²⁷.

Data extraction

The study data belongs to 16 WADA-accredited European laboratories on 11 groups of substances considered to be doping.

Process

In the WADA reports all the variables of interest were detected and codified, and were transferred to a 2010 Microsoft© Excel database for the initial data registration.

Substances that could be detected exclusively through urine were included in the study.

Laboratories that were either temporarily or permanently suspended by WADA in Europe between 2003-2015 were both excluded from the study, as well as those that appeared after 2004, for not providing enough information and with the aim of homogenising the sample.

Statistical analysis

All the analyses were performed with the IBM SPSS Statistics 21 programme.

For the study of the descriptive statistics, 16 accredited European laboratories were used, with the selected time period between 2003 and 2015.

The qualitative variables are expressed as counts and frequencies. The graphic summaries are expressed using bar charts and pie charts.

To study the difference between pre-Olympic, Olympic and post-Olympic periods, the year variables were transformed into pre-Olympic, Olympic and post-Olympic years of the OG of Athens (2004) and London (2012), as both were competitions held in Europe. The years corresponding to the Peking OG (2008) were therefore excluded. Data relating to 2006 and 2010 was also excluded so that the recoded variable in the post-Olympic year corresponded to a calendar year, just like the pre-Olympic and Olympic years. The AAF data was relativized

depending on the total analysis of each year to obtain a homogenous variable and so as to carry out the non-parametric tests.

After carrying out the normality tests, it was established that the data grouping was not homogenous, which is why non-parametric tests were carried out between two independent samples (Wilcoxon), with the CI being 95% (p = 0.05). The size of the effect was also calculated for the difference between the pre-Olympic, Olympic and post-Olympic time periods.

Results

Anabolic substances are those most frequently detected by European laboratories, followed by cannabis, glucosteroids and beta-agonists (Figure 1).

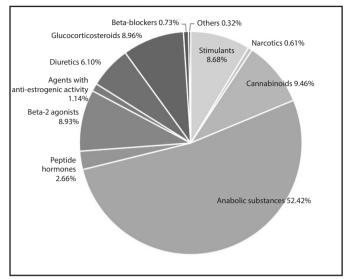
The European laboratories that proportionally detected the greatest number of AAF are Ghent (Belgium) (5.09%), Paris (France) (4.91%) and Madrid (Spain) (3.50%) (Table 1).

The proportion of AAF with regard to the anti-doping controls carried out between 2003 and 2015 in each laboratory barely exceeded 5% of all the samples analysed (Figure 2).

Detection of doping substances in the pre-Olympic, Olympic and post-Olympic years

The WADA reports were classified into time periods, cataloguing the years depending on their proximity to the OG: 2003 and 2011 were

Figure 1. Percentual representation of AAF in European laboratories between 2003 and 2015.



pre-Olympic years; 2004 and 2012 were Olympic years; 2005 and 2013 were post-Olympic years.

The data analysis displays the relationship between the detection of illegal substances in the different periods of time, with the presence of cannabis being particularly relevant in the post-Olympic period (Z=4.397; p=0.0001; SE=0.63) and anabolic substances in the Olympic period studied (Z=3.269; p=0.001; SE=0.47) (Table 2).

Table 1. AAF by substances reported in European Laboratories between 2003-2015.

Laboratory	Substances											
	Stim	Narc	Cann	Anab	Horm	Beta2	Antio	Mask	Gluco	Beta B	Others	Total AAF
Seibersdorf, Austria	102	1	128	922	34	91	24	135	58	10	0	1505
Ghent, Belgium	498	37	544	1932	65	312	58	195	240	23	0	3904
Helsinki, Finland	21	1	32	533	9	101	10	51	23	7	0	788
Paris, France	398	65	1030	1947	211	563	34	305	1181	49	10	5793
Cologne, Germany	372	26	194	2490	87	171	52	242	202	35	6	3877
Kreischa, Germany	88	10	86	748	17	180	22	96	88	16	4	1355
London, United Kingdon	m 251	9	102	437	39	51	20	53	17	7	1	987
Athens, Greece	160	1	105	680	21	42	12	96	75	3	0	1195
Rome, Italy	273	20	295	1514	164	352	15	206	263	26	36	3164
Oslo, Norway	105	6	77	830	17	145	10	60	100	5	31	1386
Lisbon, Portugal	114	3	216	649	30	62	8	98	133	38	6	1357
Moscow, Russia	259	8	114	2379	34	30	52	299	71	14	0	3260
Barcelona, Spain	68	4	83	603	47	189	31	115	73	6	3	1222
Madrid, Spain	219	19	258	1413	78	464	12	166	332	14	11	2972
Stockholm, Sweden	82	1	42	1024	8	379	23	37	287	2	0	1885
Lausanne, Switz	134	9	118	876	102	101	29	55	100	8	7	1539
Total	3144	220	3424	18977	963	3233	412	2209	3243	263	115	36189

Stim: Stimulants; Narc: Narcotics; Cann: Cannabinoids; Anab: Anabolic substances; Horm: Peptide hormones; Beta-2 agonists: Antio: Agents with anti-estrogenic activity; Mask: Masking agents/Diuretics; Gluco: Glucocorticosteroids; Beta-Beta-blockers; Other: others; Total AAF: total number of adverse analytical findings.

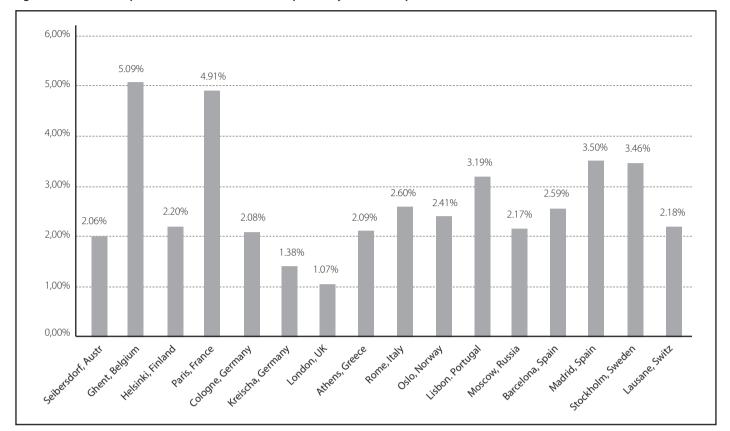


Figure 2. Percentual representation of the total AAF reported by the 16 European laboratories between 2003 and 2015.

However, the Wilcoxon test for related samples did not reveal differences in any of the periods analysed in terms of beta-agonist substances (pre-OG: Z = -0.507; p = 0.612. OG-post: Z = 1.7; p = 0.089. Post-pre: Z = 1.368; p = 0.171), anti-estrogenic substances (Z = 1; p = 0.317. OG-post: Z = 1; p = 0.317. Post-pre: Z = 0.378; p = 0.705), as well as diuretics and masking agents (Z = 1.375; D = 0.169. OG-post: D = 0.096. Post-pre: D = 0.204; D = 0.096.

Discussion

The latest scandals called "State doping" in the McLaren report in Russia²⁸ are based on statements from Grigory Rodchenkov, the exdirector of the anti-doping laboratory for the Winter Olympics in Sochi 2014, in which Russia allegedly, concerned about its poor results in the Winter Olympics in Vancouver 2010, decided to initiate a process to conceal urine samples with traces of illegal substances, and allegedly provide doping substances and methods to athletes with the best chances of winning competitions. The scandal, which directly affected the participation of Russian athletes in the Olympic and Paralympic Games in Rio 2016, opens the debate about procedure and rigor in anti-doping controls. According to the information obtained in this study, the Moscow laboratory is the European laboratory with the second highest number of anti-doping controls over the past 12 years, but its

AAF detection is very low (2.17% of almost 150,000 anti-doping controls). Despite this, it is the laboratory to detect the most anabolic substances in its AAF. It is worth highlighting that the data reported in this study belongs exclusively to the samples analysed by the different laboratories. WADA-accredited laboratories perform anti-doping controls on the athletes in their countries, and also analyse national and international competition samples, meaning that the samples from each laboratory are not necessarily samples from exclusively national athletes in the laboratory where the anti-doping control was carried out. This is one of the reasons why the aim is to compare the presence of the substance in the different Olympic time periods, thus ruling out laboratories as such. Initially it could be thought that in the years running up to the Olympics and during the Olympic year itself, there could be an increase of illegal substance detection, as this is the period when the most classifications for the OG take place. However, after analysing the results, it has been observed that significant differences can be seen between the three periods depending on the substances studied.

Anabolic substances

This kind of substance has been linked to sports with strength specialities - such as weightlifting and throwing - and to its high detection in laboratories in central and eastern European areas²⁹, as the main effect of

Table 2. Comparison between competitive periods and adverse analytical findings.

	Olympics ^b - Pre-Olympics ^c			I	Post-Olympics ^b Olympics ^c	-	Post-Olympics ^b - Pre-Olympics ^c			
	SE	Z	р	SE	Z	р	SE	Z	р	
Stimulants	0.35	-2.41 ^b	0.016	0.34	-2.355b	0.019	0.46	-3.189 ^b	0.001	
Narcotics	0.20	-1.414 ^b	0.157	0.29	-2°	0.046	0.14	-1°	0.317	
Cannabinoids	0.13	-0.931 ^b	0.352	0.42	-2.876 ^c	0.004	0.63	-4.397°	0.0001	
Anabolic substances	0.04	-0.28 ^b	0.779	0.47	-3.269°	0.001	0.30	-2.045°	0.041	
Hormones	0.17	-1.207 ^b	0.227	0.39	-2.674 ^b	0.007	0.41	-2.838 ^b	0.005	
Beta2-agonists	0.07	-0.507 ^b	0.612	0.25	-1.7 ^b	0.089	0.20	-1.368 ^b	0.171	
Anti-estrogenic	0.14	-1 ^b	0.317	0.14	-1°	0.317	0.05	-0.378 ^c	0.705	
Masking	0.20	-1.375 ^b	0.169	0.24	-1.663°	0.096	0.03	-0.204 ^c	0.839	
Glucosteroids	0.36	-2.474 ^b	0.013	0.06	-0.411°	0.681	0.37	-2.554 ^b	0.011	
Beta-blockers	0.20	-1.414 ^b	0.157	0.29	-2°	0.046	0.12	-0.816 ^c	0.414	
Others	0.29	-2.041 ^b	0.041	0.00	0	1°	0.29	-1.983 ^b	0.047	

SE: size of effect based on r (r < 0.029 = small; r = 0.03 = medium; r > 0.031 = large). Z: difference between the averages. p: value of p. based on the positive ranges. cbased on the negative ranges.

anabolic substances is to increase muscle mass and strength. Available literature usually relates the use of anabolic substances in combination with other illegal substances such as diuretics or beta-agonists, as they boost the anabolic effect or aim to mask its possible detection in antidoping controls³⁰. The information taken from this study reveals that there is greater detection of anabolic substances in the Olympic and pre-Olympic periods, but the same relation has not been found in the same periods for beta-agonists and diuretics.

Stimulants

The most used stimulants are ephedrine and cocaine. Their presence increases significantly in the post-Olympic periods studied (2005 and 2013), though this fact may be due to the use of stimulants as a social habit rather than as a substance to enhance sporting performance³¹. According to the 2015 official reports from the Spanish Observatory of Drugs and Addictions, in 2013 some 2.2% of the Spanish population claimed to have consumed cocaine at least once in the previous 12 months³². Cocaine remains detectable in urine for 3 to 5 days, but its metabolites can be detected for a long time afterwards. According to a study performed in the Rome anti-doping laboratory, the AAF discovered in athletes were metabolites with minimum amounts of this substance, relating more to social habits as opposed to doping in sport³⁴.

Narcotics

Narcotics are used in sport to speed up recovery time and/or to mask symptoms of an injury during competition³⁴. In this study, it was revealed that in the years running up to the Olympics, European laboratories detected a greater presence of narcotics than at any other time. This phenomenon could be due to sporting activity for classification for the different OG sports.

Cannabinoids

The effect produced by cannabis and its derivatives on the body are usually used to reduce anxiety and as a relaxant before sporting competitions. On the other hand, there is certain controversy regarding its use as a doping substance, as physiological effects such as an increased heart rate and blood pressure, and reduced psychomotor activity are counter-productive effects in sporting performance^{35,36}. In the results taken from this study, we can observe the strong link in pre-Olympic years compared to post-Olympic years, with pre-Olympic years producing the highest detection rate in European laboratories. In Spain, cannabis is the most frequently consumed illegal drug, especially among young people aged between 15-34 years³⁷, the age at which the majority of athletes develop their sporting careers.

Peptide hormones

This kind of illegal substances comprises a very heterogeneous group of substances, with the most common being erythropoietin. In recent years there have been various international scandals that have linked this kind of drug to sports like cycling, such as the infamous "Operación Puerto" in 2006, which broke up a doping network in Spain³⁸. In this study a greater prevalence can be observed in the detection of hormones in the post-Olympic years than in the other periods. With a lack of data of the AAF in each sport, it is not possible to link the abuse of certain illegal drugs to specific sports³³.

Glucosteroids and other substances

Glucocorticoids have an effect on the central nervous system, reducing pain and increasing the state of euphoria, thus leading to an improvement in sporting performance. The results of this study reveal that glucocorticoids were mainly detected in Olympic and post-Olympic years.

During the OG of Athens 2004, 79 AAF were detected in glucosteroids, which were studied by the IOC as they were therapeutic use exemptions³⁹. As the official reports given by the WADA only provide information about the AAF, it is not known whether these AAF were eventually considered TUE or not.

Beta-blockers

Beta-blockers are substances used to reduce the body's heart rate. It is a WADA-banned substance for just some sports, especially those that require precision or aim, such as archery, driving and motor sports. An increase of this kind of substance has been observed in the Olympic years compared to pre-Olympic and post-Olympic periods.

Study limitations

It is worth highlighting that the data reported in this study comes exclusively from the samples analysed by the different laboratories. Study limitations are the following:

- WADA-accredited laboratories perform anti-doping controls on athletes from their countries and also analyse samples from national and international competitions, which is why the samples from each laboratory are not necessarily exclusively samples from national athletes in the laboratory where the anti-doping control takes place.
- We can identify that existing data comes from official reports, but as these are illegal substances and methods, there is a black market behind all of these activities. There are studies that aim to explore this market, such as the research carried out in German anti-doping laboratories, which aim to link the AAF with the illegal drugs market, studying doping substances found in customs controls^{40,41}.
- There is heterogeneity between the first reports produced (2003-2005) and the latest ones (2012-2015), as the latter reports provide a larger amount of data.
- The sensitivity of the instruments used may vary between laboratories, as,given this is such specific machinery, the sensitivity in detecting different substances may be different.
- Furthermore, the data corresponding to the AAF reported by the laboratories has been studied, but many of the adverse analytical findings may correspond to TUE, which is why in principle they may appear as banned substances, whilst in reality they correspond to authorised pharmaceutical treatments for those athletes. This would translate as fewer athletes having truly used doping⁴².

On the other hand, despite the large number of anti-doping controls carried out in the 2003 to 2015 period, the proportion of AAF per laboratory barely exceeds 5% of the total samples analysed. However, despite the AAF figures being low, sporting institutions must defend clean sport to eradicate doping altogether. Even so, Europe is the continent with the largest number of anti-doping sanctions to date⁴³. In this respect, preventing doping should constitute a cornerstone in the sporting education of athletes, linked to ethical and social values that sport and competing represent^{44,45}. If the most frequently used substances for doping are known, as well as the sports for which these illegal substances or methods are used the most, much more specific prevention campaigns can be carried out. With this aim, and for future

research studies, it would be interesting to discover the geographical distribution and evolutionary trend of doping substances and their possible relationship with different sports, as well as studying the proportion of TUE connected to the AAF reported from each laboratory.

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