

THE UEFA EURO 2012 ANTI-DOPING PROGRAMME – SCIENTIFIC REVIEW

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ABSTRACT: The final tournament of the UEFA European Football Championship is one of the top sporting events in the world, and a high-profile event of this kind requires a well-planned and well-executed anti-doping programme to ensure the integrity of results in the competition. UEFA EURO 2012 presented a unique logistical challenge, with the tournament spread across two countries, both covering a large geographical area. This paper discusses the planning and delivery of both the pre tournament out-of-competition (OOC) testing programme and the in-competition (IC) programme, as well as reviewing the activities of doping control officers (DCOs), the whereabouts programme and assessing the sample collection and transport process. The analytical approach applied is also discussed, along with an overview of the distribution of T/E ratios and blood parameters.

KEY WORDS: soccer, doping control, football championship, blood profiling, sports doping

Introduction

The final round of the UEFA European Football Championship is a tournament for the top national teams in European men's football and is held every four years. The 2012 tournament was contested by 16 teams, which reached the final round via a series of qualification matches held over the preceding two years. That tournament was the 14th to be staged by UEFA and the first to be staged in the neighbouring countries of Poland and Ukraine. The tournament began in Warsaw on 8 June 2012 and ended with the final in Kiev on 1 July 2012. Teams listed in Table 1 qualified for the final tournament, and a total of 367 players were registered to take part (23 from each country, with the exception of one team that registered only 22 players)

UEFA has many years of experience with the planning of in- and out-of-competition doping controls in elite football and operates an annual anti-doping programme for all of its national and club competitions. The programme uses an experienced team of doping control officers (DCOs), established sample transport procedures and a network of World Anti-Doping Agency (WADA)-accredited laboratories across Europe to ensure maximum effectiveness. As the final

round of the UEFA European Football Championship is one of the world's top sporting events and UEFA's flagship tournament at national level, it is imperative that an effective anti-doping programme is in place to deter and detect doping, seeking to ensure that all results are achieved fairly and without the use of prohibited performance-enhancing substances. Consequently, the aim for UEFA at UEFA EURO 2012 was to adapt its established anti-doping processes to ensure an effectively planned and executed anti-doping

TABLE 1. TEAMS QUALIFIED FOR THE FINAL ROUND OF THE UEFA EUROPEAN FOOTBALL CHAMPIONSHIP

Group A	Group B	Group C	Group D
Poland	Netherlands	Spain	France
Greece	Denmark	Italy	England
Russia	Germany	Republic of Ireland	Ukraine
Czech Republic	Portugal	Croatia	Sweden

programme at the tournament. This was a significant challenge, with the tournament taking place in two host countries, each with four host cities spread over a wide geographical area, and only one WADA accredited laboratory in the two countries at which to analyse samples.

The tournament's anti-doping programme involved both pre-tournament out-of-competition testing of competing squads at their preparatory training camps and a full programme of in-competition testing at all matches in the tournament. Testing was supplemented by a pre-tournament education and information programme for participating teams and players.

Applicable rules

The UEFA Anti-Doping Regulations comply with those of FIFA, as well as the standards established by the WADA. These regulations were in force at the tournament. For the purposes of WADA's Prohibited List, UEFA regulations specify that the tournament's in-competition period commences 24 hours before the first match of the tournament and ends 24 hours after the final match. This meant that all pre-tournament samples were analysed on the basis of an out-of-competition analytical menu and all tournament samples (including those collected between matches) were analysed on the basis of an in-competition analytical menu. Anti-doping rule violations (ADRVs) and associated penalties were as specified in FIFA's regulations and the World Anti-Doping Code (WADC) [1].

Doping control officers

Selection

The collection of samples for the pre-tournament and tournament programmes was conducted solely by UEFA DCOs and blood collection officers (BCOs). DCOs are only qualified to collect urine samples, while BCOs are qualified to collect both urine and blood samples. UEFA manages a team of approximately 40 DCOs across Europe, all of whom are medical doctors with many years of experience in conducting doping controls for UEFA, national anti-doping organisations (NADOs) and other international sports federations.

DCOs were selected to collect samples for the tournament and pre-tournament programmes on the basis of criteria such as doping control experience and aptitude, proximity to the test venue, nationality (to limit suggestions of bias), recent blood collection experience, and languages spoken. For the pre-tournament programme, nine BCOs and 12 DCOs were used to collect samples, with some undertaking multiple assignments on consecutive days. Doping controls were normally conducted during teams' scheduled training sessions (with samples collected in places as diverse as Dublin, Moscow and Visby), and the DCOs/BCOs then delivered the samples to the WADA accredited laboratory in Warsaw by plane. For the in-competition programme, a team comprising six DCOs and six BCOs was selected, with officers working in the same designated pairs at all of their appointed matches. The DCOs/BCOs were based permanently in Warsaw, close to the tournament laboratory, and travelled by plane

to each game on the morning of the match, before returning on the first flight the following morning.

Training and performance monitoring

All UEFA DCOs are trained to collect urine samples in accordance with the WADA International Standard for Testing (IST) [2] as part of their role in UEFA's annual anti-doping programme, so no new procedural training was required for the tournament programme. For BCOs, specific instructions on procedures and the use of blood sampling equipment were provided in advance of their first assignment in connection with the tournament. In addition, all DCOs were briefed collectively on arrival in Warsaw for the in-competition programme by means of a preparatory workshop covering documentation, equipment, player selection and logistical procedures. During the tournament, DCOs were required to attend a debriefing session after each doping control to review procedures and provide feedback in the event that improvements could be made for the next match.

During the tournament, DCOs' performance was monitored by a team of observers consisting of members of UEFA's anti-doping unit, the UEFA Anti-Doping Panel and the UEFA Medical Committee. In addition, the Chairman of the UEFA Anti-Doping Panel was present for the full duration of the tournament to review the DCOs' activities.

Equipment and logistics

Transport of samples

The large distances between the host cities (Warsaw, Wrocław, Gdansk and Poznan in Poland, and Kyiv, Donetsk, Kharkiv and Lviv in Ukraine) made ground transport of samples unfeasible, particularly considering the time restrictions on the safe transport of blood samples and the tight time frames between matches for the reporting of sample results. Consequently, all samples were transported by plane (i) on the day the sample was collected for pre-tournament out-of-competition samples or (ii) on the morning after the match (on the first available flight) for in-competition samples. This required the careful planning of flights for DCOs, as well as the prior agreement of domestic and international airlines and customs authorities to ensure the safe and unimpeded transport of samples as cabin baggage in the custody of DCOs. During the tournament, this process was facilitated by the provision of advance notification of DCOs' travel plans to the local organising committee (LOC) airport co-ordinators based in each host city airport, as well as by written letters of agreement from airlines. The pre-notification process was acceptable in terms of disclosure risk, as all tournament matches were to involve testing, so the prior notification of dates did not reveal any aspect of the test distribution plan. Other testing conducted during the tournament was not disclosed to airport or customs staff until samples had been collected.

No problems were experienced with the cross-border transport of samples by DCOs, either by road or by plane, despite some delays to scheduled flights' departure times during the competition period.

Sample collection kit

Collecting and transporting samples, particularly blood samples, always presents a unique challenge, with the need to ensure that the integrity of samples is maintained during the journey from the collection site to the laboratory. This is particularly difficult where travel over longer distances is required. The kit used has to be reliable, trusted by the players and teams, and – perhaps most importantly – easy for DCOs and BCOs to use, store and transport.

All sample collection kits and associated equipment used in connection with the tournament were supplied by the company Berlinger from Switzerland, which also supplies kits for UEFA's annual anti-doping programme. Berlinger is one of the leading suppliers of equipment for anti-doping programmes in international sport.

Urine and blood samples were collected using standard Berlinger Bereg kits. Blood samples were taken with a butterfly needle. Four separate tubes were needed for whole blood and serum in order to store all of the samples (an "A" and "B" sample for each). As blood and serum samples need to be analysed shortly after collection, and must be kept at a very stable temperature (between 4 and 12°C), cooling equipment was required to maintain this temperature for at least 48 hours. The Nanocool box was used for this purpose for all tournament and pre-tournament blood and serum samples, with a Q Tag device to monitor the temperature during transit. Nanocool boxes were prepared by BCOs for out-of-competition testing and by the Warsaw laboratory for in-competition testing. All blood samples were securely refrigerated in the doping control station until the collection of samples had been completed, at which point the Nanocool box was activated by the BCO.

Management of Therapeutic Use Exemptions (TUEs)

Management of TUEs followed UEFA's usual procedure, which complies with WADA's International Standard for TUEs [3]. All TUE applications for players had to be submitted to UEFA 21 days before the start of the tournament (with the exception of emergency cases, which could be reviewed at shorter notice if they occurred just prior to the start of the tournament).

Between 1 May 2012 and 2 July 2012, UEFA granted one TUE for glucocorticosteroids. This was the only TUE applied for in connection with the tournament.

Out-of-competition testing programme

The test distribution plan for the pre-tournament programme was complicated by a combination of (i) the unique legal arrangements in European football, whereby UEFA has jurisdiction over players only when they are competing in its competitions, and (ii) the short period of time that teams had to prepare for the tournament after the completion of their domestic club seasons. In effect, a three week window was available in which to conduct out-of-competition doping controls. This was the time between the point at which selected players met for pre-tournament training for the first time and

the point at which teams left their training camps for the host countries. The start date for training camps is usually determined by the date that the domestic and European club season ends and will also depend on the countries in which the majority of the team's players play their club football (which is often not their national league). The first participating team to meet for a pre tournament training camp did so on 9 May 2012, while the last team met up for the first time on 24 May 2012.

Team whereabouts

To enable UEFA to plan its programme, teams were required to supply their overall pre tournament schedule approximately one month in advance of the tournament (by 27 April 2012). As of 1 May 2012 (or the week of the first training camp if it was later), all 16 participating teams had to submit detailed whereabouts information to UEFA via weekly reports/schedules for the following week, which had to be submitted every Friday before 12.00 CET, in accordance with the standard procedure applied by UEFA in the UEFA Champions League. Whereabouts submissions consisted of training camps/sessions, friendly matches, the address of the training ground or match venue, and the address of the hotels/training camps where the team was staying.

Whereabouts violations were handled in accordance with the UEFA Anti-Doping Regulations, with teams subject to team non-compliances for failing to submit accurate information (or provide updates) about the location of the team or individual players, and with players subject to individual non-compliances for failing to update UEFA in advance regarding their absence from collective team activities, where that resulted in them missing doping controls.

Sample collection and analysis

Following a recommendation by the UEFA Anti-Doping Expert Panel, the UEFA Executive Committee decided that both blood and urine samples should be collected at each doping control, both prior to and during the tournament. This was a continuation of the policy adopted at the 2008 tournament, which had been the first time that both blood and urine analysis had been implemented at a major international sports event. In total, 284 players were tested in connection with the 2012 tournament on the basis of an analytical menu that was devised by experts from leading WADA accredited laboratories in Europe and approved by the UEFA Anti-Doping Expert Panel. The menu included partial screening for out-of-competition samples and, in addition, testing for artificial haemoglobin, blood transfusion (whole blood), blood parameters (whole blood), hGH (serum), EPO (urine), CERA (serum), [4,5,6,7] SARMS (urine – as part of the partial OOC screen) and plasticisers (urine), as well as IRMS analysis (which can identify endogenous and exogenous intake of anabolic steroids, providing a complete individual steroid profile).

All out-of-competition doping controls conducted pre-tournament involved ten players from the team undergoing blood, urine and serum tests with no notice at the team's pre-tournament training

camp. All participating teams were visited at least once prior to the start of the tournament, and a total of 160 tests were administered (Table 2). Teams were responsible for organising a suitable doping control station (DCS), either at the training venue or at the hotel where the team was staying.

All out-of-competition samples were analysed at the laboratory of the Department of Anti-Doping Research at the Institute of Sport in Warsaw to ensure consistency of analysis and results across samples. All procedures were supervised by experienced experts from other WADA accredited laboratories during the pre- and in-competition phase. All samples were received by the laboratory within 24 hours of collection and were analysed within 48 hours of receipt

TABLE 2. OUT-OF-COMPETITION ANTI-DOPING LABORATORY'S ANALYTICAL MENU AND DISTRIBUTION

Partial screen for OOC samples (urine)	Ten players per team
EPO (urine)	Five players per team
IRMS (urine)	Two players per team, and only if required by WADA technical documents
SARMS (urine)	Ten players per team
CERA (serum)	Ten players per team
HGH (serum)	Ten players per team
Blood parameters (whole blood)	Ten players per team
Blood transfusion	Two players per team
Plasticisers (useful when combined with blood parameters)	Ten players per team

(with the exception of IRMS analysis, which was conducted within 96 hours) (Table 3). All samples collected in the out-of-competition programme came back negative.

In-competition testing programme

Doping controls were conducted after every match at the tournament, with two players from each team drawn to undergo testing, plus two reserves. Reserves were available to be tested in the event that a selected player sustained an injury that rendered him incapable of completing the test (which did not occur at any tournament match). Any of the 23 squad players could be drawn for testing, regardless of whether they actually participated in the match.

32 doping control chaperones (four per stadium) were appointed to assist with the in-competition programme from the pool of tournament volunteers. Where possible, chaperones came from the national anti-doping organisations of the host countries. In Poland, five chaperones were experienced anti-doping officers from Komisja do Zwalczenia Doping w Sporcie (Polish Commission Against Doping in Sport), and all other chaperones were selected from the volunteer pool.

All chaperones were trained by UEFA's anti-doping unit in advance of the tournament via a specially designed distance learning programme. In addition, a support service was made available within the anti-doping unit to ensure that any questions regarding the role could be answered by experienced staff. Finally, all chaperones were given a full pre-match briefing by a DCO before their first match at the tournament.

The in-competition programme commenced with the first tournament match on 8 June 2012 between Poland and Greece, and ended on 1 July 2012 with the final between Spain and Italy.

TABLE 3. OVERVIEW OF THE DELIVERY CONDITIONS OF THE OUT-OF-COMPETITION SAMPLES (20 X BLOOD AND 10 X URINE SAMPLES PER MISSION) TO THE WARSAW LABORATORY INCLUDING TEMPERATURE INFORMATION (Q-TAG SYSTEM)

Collection date	End of collection*	Delivery date	Travel time**	Q-Tag nformation	Max. temperature (°C)
18 May 2012	14.25	19 May 2012	22 h 20 min	OK/OK	
21 May 2012	14.20	22 May 2012	18 h 20 min	Not activated/ Not activated	
21 May 2012	16.01	22 May 2012	17 h 44 min	OK/OK	
23 May 2012	19.30	24 May 2012	15 h 45 min	OK/Alarm	— / 18.4
23 May 2012	14.40	24 May 2012	21 h 45 min	OK/OK	
23 May 2012	20.20	24 May 2012	16 h 35 min	OK/Alarm	— / 24.6
24 May 2012	14.40	25 May 2012	18 h 45 min	OK/Broken	
25 May 2012	14.40	25 May 2012	8 h 35 min	Alarm/OK	> 16 / —
25 May 2012	16.30	26 May 2012	21 h 40 min	OK/Alarm	— / 19.4
26 May 2012	13.00	26 May 2012	10 h 40 min	OK	
27 May 2012	14.46	27 May 2012	8 h 26 min	OK/OK	
28 May 2012	14.10	28 May 2012	7 h 20 min	OK	
28 May 2012	14.15	29 May 2012	22 h 15 min	OK/OK	
29 May 2012	20.40	30 May 2012	15 h 15 min	Alarm/OK	22.5 / —
31 May 2012	21.10	1 June 2012	18 h 30 min	Alarm	23.2
2 June 2012	20.45	3 June 2012	19 h 40 min	OK/OK	

Note: * download time of the last sample of urine/blood prior to transport to laboratory, ** time from the end of collection to delivery to the laboratory

Doping control stations (DCSs) at tournament stadiums were of extremely high quality, as most of the stadiums were newly built, with specific consideration given in the planning stage to the design and location of the facility. All DCSs used at the tournament were inspected by members of the UEFA Medical Committee and anti-doping unit staff at site visits conducted in 2011 and 2012.

In-competition samples were analysed for the full WADA analytical menu plus additional screening for EPO, HGH, blood transfusion and SARMS, with IRMS analysis conducted as required [8,9] (Table 4). All samples were received by the laboratory within 24 hours of collection and were analysed within 24 hours of receipt (with the exception of EPO and IRMS analysis, which was conducted within 48 hours). All results were reported before the team's next match. All samples were analysed by the WADA-accredited laboratory in Warsaw under the supervision of experienced experts from other WADA accredited laboratories.

TABLE 4. IN-COMPETITION ANTI-DOPING LABORATORY'S ANALYTICAL MENU AND DISTRIBUTION (31 MATCHES)

Full screen for IC samples	Four samples per match
EPO (urine)	Four samples per match as of quarter-finals
IRMS (urine)	If requested by WADA technical documents
SARMS (urine)	Four samples per match
HGH (serum)	Four samples per match
Blood transfusion (whole blood)	Four samples per match as of semi-finals

All 124 players' blood and urine samples collected in the in-competition programme were returned as negative. On three occasions, blood could not be successfully collected from a player (with two of these incidents involving the same player). Following the conclusion of the tournament, all samples were placed in storage (at -20°C) and will be retained for re-analysis by UEFA as required.

Transport to the laboratory

Overview of the delivery conditions of IC samples (standard four blood and four urine samples per match) to the Warsaw laboratory is presented in Table 5.

Results and discussion

Sample analysis

In accordance with WADA's quality control requirements, tournament samples were analysed to the highest possible standards. Analysts working in the laboratory in Warsaw were trained prior to the tournament in the Lausanne and Seibersdorf laboratories, and pre-tournament training was conducted in Warsaw by experts from

the Lausanne laboratory. Training was also offered by manufacturers providing new analytical equipment.

Pre-analytics

Table 6 summarises the pH and specific gravity of the urine samples (A samples) collected during in-competition and out-of-competition testing.

Urine testing

Out-of-competition urine samples were tested for the following prohibited substances included on WADA's 2012 Prohibited List [10]:

- S1: Anabolic agents
 - 1. Anabolic androgenic steroids (AASs)
 - 2. Other anabolic agents
- S2: Peptide hormones, growth factors and related substances
 - 1. Erythropoiesis-stimulating agents
 - 2. Chorionic gonadotropin (CG) and luteinising hormone (LH)
 - 5. Growth hormone
- S3: Beta-2 agonists
- S4: Hormone and metabolic modulators
 - 1. Aromatase inhibitors
 - 2. Selective oestrogen receptor modulators (SERMs)
 - 3. Other anti-oestrogenic substances
 - 5. Metabolic modulators: peroxisome proliferator activated receptor δ (PPAR δ) agonists (e.g. GW 1516)
- S5: Diuretics and other masking agents
 - M1. Enhancement of oxygen transfer
 - 1. Blood doping
 - 2. Artificially enhancing the uptake, transport or delivery of oxygen, including efaproxiral (RSR13)

In-competition urine samples were also screened for:

- S6: Stimulants
- S7: Narcotics
- S8: Cannabinoids
- S9: Glucocorticosteroids

In the pre-tournament testing programme, samples from two of the ten players tested in each team were analysed for synthetic testosterone or testosterone precursors using gas chromatography combustion isotope ratio mass spectrometry (GC/C/IRMS), with other samples analysed in this way if required by WADA technical documents. At the in-competition stage, analysis using GC/C/IRMS was performed only when required by WADA technical documents [8,9].

The distribution of testosterone to epitestosterone (T/E) ratios in out-of-competition and in competition samples is shown in Figure 1 and Figure 2.

All samples with a T/E ratio in excess of 4.0 were analysed using IRMS in accordance with the requirements laid down in the WADA

TABLE 5. OVERVIEW OF THE DELIVERY CONDITIONS OF IC SAMPLES (STANDARD FOUR BLOOD AND FOUR URINE SAMPLES PER MATCH) TO THE WARSAW LABORATORY, INCLUDING TEMPERATURE INFORMATION (Q-TAG SYSTEM)

Collection date	End of collection*	Delivery date	Travel time**	Q-Tag nformation	Max. temperature (°C)
8 June 2012	21.25	8 June 2012	25 min	OK	
8 June 2012	23.30	9 June 2012	7 h 40 min	OK	
9 June 2012	22.00	10 June 2012	17 h 15 min	OK	
10 June 2012	21.25	11 June 2012	9 h 55 min	OK	
10 June 2012	00.35	10 June 2012	15 h 45 min	OK	
11 June 2012	23.18	12 June 2012	19 h 7 min	OK	
11 June 2012	01.10	11 June 2012	6 h 30 min	OK	
12 June 2012	21.00	13 June 2012	10 h 55 min	OK	
12 June 2012	01.40	12 June 2012	9 h 20 min	Alarm	> 12
13 June 2012	21.45	14 June 2012	15 h 10 min	OK	
13 June 2012	00.15	13 June 2012	35 min	OK	
14 June 2012	21.48	15 June 2012	8 h 52 min	OK	
14 June 2012	00.45	14 June 2012	15 h 10 min	OK	
15 June 2012	23.25	16 June 2012	16 h 45 min	OK	
15 June 2012	01.10	15 June 2012	10 h 30 min	OK	
16 June 2012	01.20	16 June 2012	8 h 25 min	OK	
17 June 2012	00.15	17 June 2012	35 min	OK	
17 June 2012	00.25	17 June 2012	7 h 5 min	OK	
18 June 2012	00.40	18 June 2012	15 h 20 min	OK	
18 June 2012	00.30	18 June 2012	15 h 40 min	OK	
19 June 2012	02.00	19 June 2012	5 h 8 min	OK	
19 June 2012	00.28	19 June 2012	6 h 52 min	OK	
20 June 2012	00.10	20 June 2012	10 h 30 min	OK	
20 June 2012	01.30	20 June 2012	15 h 50 min	OK	
22 June 2012	00.16	22 June 2012	34 min	OK	
22 June 2012	23.45	23 June 2012	7 h 15 min	OK	
24 June 2012	01.55	24 June 2012	14 h 40 min	OK	
25 June 2012	02.34	25 June 2012	7 h 1 min	OK	
28 June 2012	02.15	28 June 2012	16 h 25 min	OK	
29 June 2012	00.11	29 June 2012	24 min	OK	
2 July 2012	01.54	2 July 2012	7 h 36 min	OK	

Note: * download time of the last sample of urine/blood prior to transport to laboratory, ** time from the end of collection to delivery to the laboratory

International Standard for Laboratories [11,12,13]. No indication of the use of synthetic testosterone or testosterone precursors was identified. The Warsaw laboratory also sent all steroid profile parameters to UEFA in order to compare those results with the results of previous analysis.

TABLE 6. OUT-OF-COMPETITION ANTI-DOPING LABORATORY'S ANALYTICAL MENU AND DISTRIBUTION

	pH		Specific gravity	
	OOC samples	IC samples	OOC samples	IC samples
Total samples	160	132	160	132
Mean	6.08	5.87	1.018	1.014
Median	6.00	5.69	1.017	1.011
Min.	5.08	5.07	1.003	1.002
Max.	7.72	7.75	1.035	1.036
SD*	0.55	0.53	0.008	0.009
CV**	9%	9%	0.8%	0.9%

Note: * standard deviation, ** coefficient of variation

The presence of recombinant erythropoietin was tested for using ultrafiltration, isoelectric focusing (IEF), double blotting and chemiluminescence detection [14,15,16,17]. For suspicious profiles, an SDS PAGE test was conducted straight after screening [18]. All results were negative. The percentage of samples with suspicious profiles was approximately 19% for out-of-competition samples and approximately 6% for in-competition samples.

Each of the OOC samples was analysed for the presence of plasticiser metabolites [19]. Although the presence of elevated levels of these indicators is not unambiguous proof of the use of blood transfusion, this represents important additional evidence alongside the blood passport programme [20]. No suspicious results were detected.

Blood parameter distribution

Blood parameters were only analysed in the out-of-competition programme. A full blood count was obtained for all blood samples, and the stimulation index (OFF score) and abnormal blood profile score (ABPS) were calculated [21,22,23]. Blood parameters were

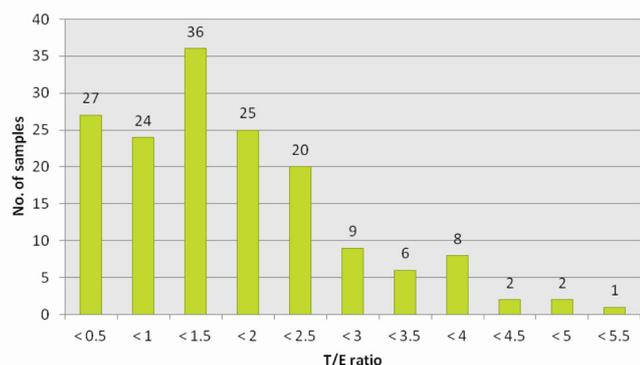


FIG. 1. DISTRIBUTION OF TESTOSTERONE TO EPITESTOSTERONE (T/E) RATIOS IN OUT-OF-COMPETITION SAMPLES AT UEFA EURO 2012

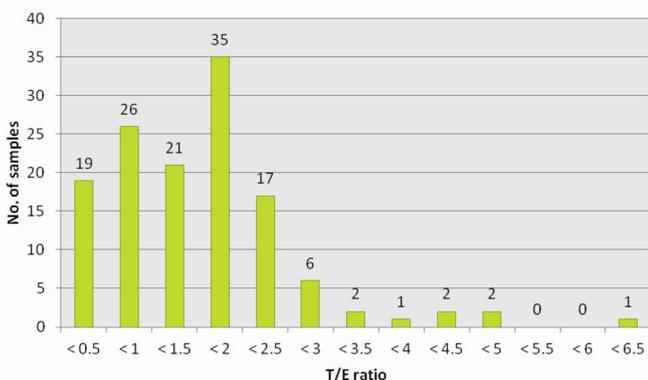


FIG. 2. DISTRIBUTION OF TESTOSTERONE TO EPITESTOSTERONE (T/E) RATIOS IN IN-COMPETITION SAMPLES AT UEFA EURO 2012

not analysed in-competition, as in competition blood test results were shown at UEFA EURO 2008 [24] to be affected by dehydration. This had led to elevated levels of some parameters, meaning that the analysis was of little value (Figure 3, Figure 4, Figure 5). The conditions required for athlete blood passport (ABP) tests were not fulfilled either, as players were not required to rest for two hours post-match.

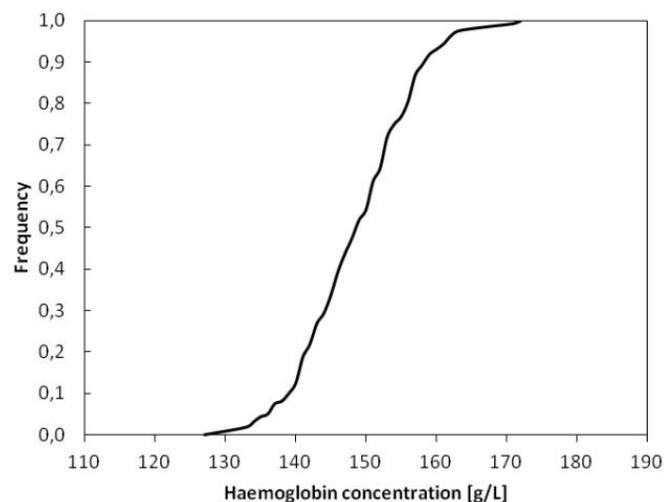


FIG. 3. CUMULATIVE FREQUENCY DISTRIBUTION OF HAEMOGLOBIN CONCENTRATION (G/L) IN OUT OF COMPETITION SAMPLES AT UEFA EURO 2012

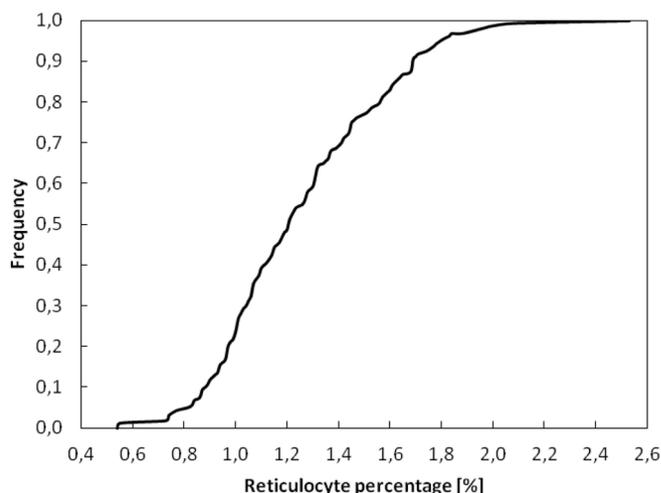


FIG. 4. CUMULATIVE FREQUENCY DISTRIBUTION OF RETICULOCYTE PERCENTAGES IN OUT-OF-COMPETITION SAMPLES AT UEFA EURO 2012

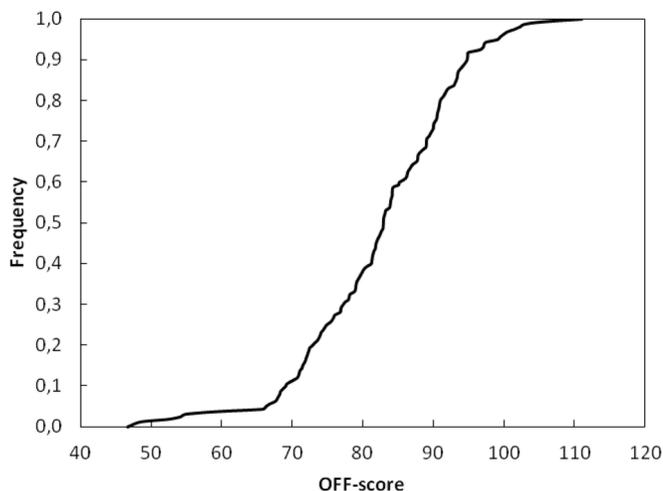


FIG. 5. CUMULATIVE FREQUENCY DISTRIBUTION OF OFF SCORES IN OUT-OF-COMPETITION SAMPLES AT UEFA EURO 2012

Blood parameters are determined in a harmonised manner thanks to an external quality control programme for blood samples in which all WADA-accredited laboratories participate [25]. All laboratories use the same protocol and quality controls to ensure that results can be interpreted consistently and compared across laboratories. All blood parameters from samples collected in connection with UEFA EURO 2012 were sent to UEFA by the Warsaw laboratory to allow comparison with previous results.

Growth hormone testing

Serum was tested for the presence of human growth hormone in both the out-of-competition and in-competition programmes (Table 7). All HGH tests were prepared using a special chemiluminescence immunoassay kit for HGH [26]. All results were expressed by means of two measured concentrations: the concentration resulting from the “REC” assay (corresponding roughly to the recombinant HGH) and the concentration resulting from the “PIT” assay (corresponding to all HGH forms circulating in the blood).

TABLE 7. HGH RESULTS FOR BLOOD SAMPLES COLLECTED DURING TESTING

	OOC blood samples		IC blood samples	
	REC	PIT	REC	PIT
Total samples	160		120	
No. of samples above LOQ*	129	160	113	120
Percentage below LOQ	19%	0%	6%	0%
	OOC samples above LOQ		IC samples above LOQ	
No. of ratios	129		113	
Mean of ratios	0.469		0.428	
Median of ratios	0.432		0.423	
SD**	0.182		0.133	
Mean CV*** of ratios	39%		31%	
Min.	0.178		0.855	
Max.	0.182		1.133	
WADA cut-off	1.81		1.81	

The ratio of the two concentrations was calculated in accordance with WADA guidelines [27]. Only negative samples were observed.

Non-steroidal anti-inflammatory drugs (NSAIDs)

NSAIDs were detected in a large number of samples, both in-competition and out-of-competition. A relationship was found between the duration of the tournament and the number of samples containing NSAIDs. The most frequent compound was diclofenac, which was used more towards the end of the tournament.

Nicotine

Very high concentrations of nicotine were observed in several samples. This high concentration is very rarely observed in routine anti-doping samples.

Declaration of medication

As is standard practice for doping controls, players were required to declare any medication that had recently been taken or administered prior to the control. Of the 124 players tested at the tournament, 65 (52%) declared that they had taken some sort of medi-

cine during the last three months, while 59 (48%) declared that they had not taken any medicine, or only vitamins or minerals.

Of the players who declared the recent use of medication, 4% declared that they had been given a local cortisone injection during the last three months and 54% (i.e. 35 of those 65) declared that they had taken (or were taking) NSAIDs. Thus, 28% of the players tested at the tournament had taken NSAIDs during the previous three months.

These findings suggest that treatment of musculoskeletal problems with local corticosteroids is uncommon in football, whereas treatment with NSAIDs is more frequent. However, the figure for NSAIDs is lower than at UEFA EURO 2008, where 44% of players tested reported having taken NSAIDs.

10% of the players tested declared that they had taken sleeping pills on account of insomnia during the last three months.

Conclusion

The anti-doping programme conducted at UEFA EURO 2012 continued UEFA's commitment to ensuring that the highest possible standards of doping deterrence and detection are applied at its major competitions. Following on from the process begun in 2008, all samples were analysed for the most advanced possible range of prohibited substances, using blood and urine samples processed at a laboratory accredited by WADA under the direction of some of the leading anti-doping laboratories in Europe. None of the samples collected returned positive results, although interesting levels of nicotine and NSAIDs (which are not prohibited) were detected during the tournament. The testing and analytical programme employed is an excellent example of high quality co-ordinated doping controls at a major team sport tournament.

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REFERENCES

- World Anti-Doping Agency, World Anti-Doping Code, 2012. Available at http://www.wada-ama.org/Documents/World_Anti-Doping_Program/WADP-The-Code/WADA_Anti-Doping_CODE_2009_EN.pdf accessed 19 October 2012.
- World Anti-Doping Agency, International Standard for Testing, January 2012. Available at http://www.wada-ama.org/Documents/World_Anti-Doping_Program/WADP-IS-Testing/2012/WADA_IST_2012_EN.pdf, accessed 23 October 2012.
- World Anti-Doping Agency, International Standard for Therapeutic Use Exemptions, January 2011. Available at http://www.wada-ama.org/Documents/World_Anti-Doping_Program/WADP-IS-TUE/2011/WADA_ISTUE_2011_revJanuary-2012_EN.pdf, accessed 12 November 2012.
- Lönnerberg M., Dehnes Y., Drevin M., Garle M., Lamon S., Leuenberger N., Quach T., Carlsson J. Rapid affinity purification of erythropoietin from biological samples using disposable monoliths. *J. Chromatogr. A* 2010;1217,7031-7037.
- Reichel C., Abzieher F., Geisendorfer T. SARCOSYL-PAGE: a new method for the detection of MIRCERA- and EPO-doping in blood. *Drug Test. Anal.* 2009;1,494-504.
- Reichel C. SARCOSYL-PAGE: a new electrophoretic method for the separation and immunological detection of PEGylated proteins. *Methods Mol. Biol.* 2012;869,65-79.
- Lamon S., Giraud S., Egli L., Smolander J.,

- Jarsch M., Stubenrauch K.G., Hellwig A., Saugy M., Robinson N. A high-throughput test to detect C.E.R.A. doping in blood. *J. Pharm. Biomed. Anal.* 2009;50,954-958.
8. WADA Technical Document TD2004EAAS – Reporting and Evaluation Guidance for Testosterone, Epitestosterone, T/E Ratio and other Endogenous Steroids, Version. 1.0, August 2004. Available at, http://www.wada-ama.org/Documents/World_Anti-Doping_Program/WADP-IS-Laboratories/Technical_Documents/WADA_TD2004EAAS_Reporting_Evaluation_Testosterone_Epitestosterone_TE_Ratio_EN.pdf accessed 12 November 2012.
 9. World Anti-Doping Program: Guidelines for the reporting and management of elevated T/E ratios and endogenous steroids, Version 2.0, October 2011. Available at http://www.wada-ama.org/Documents/Resources/Guidelines/WADA_Guidelines_Reporting_Elevated_TE_Ratios_2.0_EN.pdf accessed 22 November 2012.
 10. World Anti-Doping Agency, The 2012 Prohibited List, January 2012. Available at http://www.wada-ama.org/Documents/World_Anti-Doping_Program/WADP-Prohibited-list/2012/WADA_Prohibited_List_2012_EN.pdf accessed 24 November 2012.
 11. World Anti-Doping Agency, International Standard for Laboratories, Version 7.0, January 2012. Available at http://www.wada-ama.org/Documents/World_Anti-Doping_Program/WADP-IS-Laboratories/ISL/WADA_Int_Standard_Laboratories_2012_EN.pdf accessed 27 November 2012.
 12. Piper T., Mareck U., Geyer H., Flenker U., Thevis M., Platen P., Schänzer W. Determination of 13C/12C ratios of endogenous urinary steroids: method validation, reference population and application to doping control purposes. *Rapid Commun. Mass Spectrom.* 2008;22,2161-2175.
 13. Saudan C., Emery C., Marclay F., Strahm E., Mangin P., Saugy M. Validation and performance comparison of two carbon isotope ratio methods to control the misuse of androgens in humans. *J. Chromatogr. B Analyt. Technol. Biomed. Life Sci.* 2009;877,2321-2329.
 14. Lasne F., Martin L., Crepin N., de Ceaurriz J. Detection of isoelectric profiles of erythropoietin in urine: differentiation of natural and administered recombinant hormones. *Anal. Biochem.* 2002;311,119-126.
 15. Lasne F., Martin L., Martin J.A., de Ceaurriz J. Isoelectric profiles of human erythropoietin are different in serum and urine. *Int. J. Biol. Macromol.* 2007;41,354-357.
 16. Reihlen P., Völker-Schänzer E., Majer B., Schänzer W. Easy-to-use IEF compatible immunoaffinity purification of Erythropoietin from urine retentates. *Drug Test. Anal.* 2012;4,813-817.
 17. WADA Technical Document TD2009EPO – Harmonization of the Method for the Identification of Recombinant Erythropoietins (i.e. Epoetins) and Analogues (e.g. Darbepoetin and Methoxypolyethylene Glycol-Epoetin Beta), Version 2.0, September 2009. Available at http://www.wada-ama.org/Documents/World_Anti-Doping_Program/WADP-IS-Laboratories/Technical_Documents/WADA_TD2009EPO_Harmonization_of_Method_Identification_Recombinant_Erythropoietins_Analogues_EN.pdf accessed 4 December 2012.
 18. Reichel C., Kulovics R., Jordan V., Watzinger M., Geisendorfer T. SDS-PAGE of recombinant and endogenous erythropoietins: benefits and limitations of the method for application in doping control. *Drug Test. Anal.* 2009;1,43-50.
 19. Monfort N., Ventura R., Latorre A., Belalcazar V., López M., Segura J. Urinary di-(2-ethylhexyl)phthalate metabolites in athletes as screening measure for illicit blood doping: a comparison study with patients receiving blood transfusion. *Transfusion* 2010;50,145-149.
 20. Giraud S., Robinson N., Mangin P., Saugy M. Scientific and forensic standards for homologous blood transfusion anti-doping analyses. *Forensic Sci. Int.* 2008;179,23-33.
 21. Sottas P.E., Vernece A. Current implementation and future of the Athlete Biological Passport. *Bioanalysis* 2012;4,1645-1652.
 22. Sottas P.E., Robinson N., Rabin O., Saugy M. The athlete biological passport. *Clin. Chem.* 2011;57,969-976.
 23. WADA Technical Document TD2010BAR – Blood Analytical Requirements for the Athlete Biological Passport, Version 1.0, January 2010. Available at http://www.wada-ama.org/Documents/World_Anti-Doping_Program/WADP-IS-Laboratories/Technical_Documents/WADA_TD2009EPO_Harmonization_of_Method_Identification_Recombinant_Erythropoietins_Analogues_EN.pdf accessed 4 December 2012.
 24. Vouillamoz M., Thom C., Grisdale R., Saugy M., Giraud S., Robinson N., Gmeiner G. and Geisendorfer T. Anti-doping testing at the 2008 European football championship. *Drug Test. Anal.* 2009;1,485-493.
 25. Quality Control Center Switzerland – CSCQ, Programme name: WADA - EQAS Haematological module. Available at <http://www.cscq.ch> accessed 9 November 2013.
 26. Bidlingmaier M., Suhr J., Ernst A., Wu Z., Keller A., Strasburger C.J., Bergmann A. High-sensitivity chemiluminescence immunoassays for detection of growth hormone doping in sports. *Clin. Chem.* 2009;55,445-453.
 27. World Anti-Doping Agency, Guidelines hGH isoform differential immunoassays for anti doping analyses, Version 1.0, June 2012. Available at http://www.wada-ama.org/Documents/Science_Medicine/Anti-Doping-Labs-Guidelines/WADA_SC-Guidelines_hGH%20Differential%20Immunoassays_EN_June10.pdf, accessed 4 December 2012.