Contamination of dietary supplements and positive drugs tests in sport

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The use of dietary supplement is widespread at all levels of sport, as indeed it is in the general population. Reviews of the published literature (involving a meta-analysis of 51 published surveys involving 10,274 male and female athletes), however, suggest that the use of supplements is more prevalent in athletes (46%) than in the general population (35-40%), while use is more prevalent still among elite athletes, with 59% reporting supplement use (Sobal and Marquart, 1994).

Athletes who take supplements often have no clear understanding of the effects of the supplements they are using, but it seems clear that supplements should be used only after a careful cost-benefit analysis has been conducted. On one side of the balance are the rewards, the most obvious of which is an improved performance in sport, and on the other side lie the costs and the risks. Vitamin and mineral supplements are generally perceived as being harmless, and the one-a-day multivitamin tablet is seen as an insurance policy "just in case". Many herbal products are also used, even though there is little or no evidence to support their claimed benefits. The fact that most of these supplements enjoy only brief periods of popularity before disappearing from the marketplace suggests that any benefits perceived by athletes are not strong

enough to warrant continued use or recommendation to friends and colleagues.

More exotic supplements, many of which have names that suggest an anabolic action and with promotional material to match, have become a prominent feature on the shelves of sports nutrition stores in the last decade or two. Some of these products make extravagant claims about building bigger, stronger and faster muscles, repairing the damage caused by hard training, resisting infections and illnesses, and preventing chronic fatigue. "Fat-burning" and weight loss products are also extremely popular with athletes, many of whom are seeking to achieve unrealistically low levels of body fat. The products boast impressive results, often supported by unsubstantiated endorsements, usually come with a price tag to match too, but for the athlete who is training to the limit of endurance, no price seems too high.

Unlike drugs, the regulations governing the purity of dietary supplements and of the claims that can be made as to their purity and efficacy are somewhat lax. The passing of the Dietary Supplements Health and Education Act (1994) by the US Congress resulted in a considerable liberalisation of the regulations regarding the manufacture and sale of nutritional supplements. The current regulations in the US and in many other countries permit the unrestricted sale of substances that are closely related to testosterone and other anabolic androgenic agents. The American Food and Drugs Administration (FDA), prompted in part by a number of fatalities associated with supplement use, is now proposing to introduce legislation that will regulate the manufacture and distribution of dietary supplements. At present, however, dietary supplements are not evaluated by regulatory agencies and inaccurate labelling of ingredients is known to be a problem. Internet selling has also effectively removed most of the national controls that are in place to protect the consumer.

For the elite athlete liable to testing for the use of prohibited pharmacological agents, the lack of regulation governing dietary supplements poses unique problems, and there is now compelling evidence that dietary supplements may be responsible for at least some of the positive doping results recorded by athletes. This has long been recognised to be true in the case of some mild stimulants,

including especially CAFFEINE and ephedrine, that are present in over the counter herbal tonics. Many published studies have shown contamination of supplements with prohibited compounds, and others have shown that some products do not contain any measurable amount of the substances identified on the label while others may contain up to 150% of the stated dose (Parasrampuria et al, 1998, Gurley et al, 2000). Where relatively expensive ingredients are involved, it seems that some products contain little or no active ingredient (Green et al, 2001).

Nandrolone in sport

Nandrolone is the popular name for the anabolic androgenic steroid more properly known as 19-nortestosterone. This drug is closely related, in both structure and function, to the male sex hormone testosterone, which plays a role in building muscle and in aggressive behaviour. Both of these effects might be of benefit to competitive athletes and there is a long history of abuse of anabolic androgenic steroids in sport: these agents have been used whenever athletes need strength, power, muscle bulk, aggression or rapid recovery during periods of intensive training or competition. Nandrolone is effective, but is easily detected. After injection of nandrolone esters, a positive urine test is likely to result for some months, but oral nandrolone is quickly cleared from the system.

Nandrolone itself is not measured in urine samples collected for athletes by the doping authorities, but rather one of its metabolites (19-norandrosterone) is measured. 19-norandrosterone (and the other metabolite, 19-noraetiocholanolone) may be detected in the urine if nandrolone has been taken either orally or by injection, but these compounds can also arise from metabolism of the prohormones of nandrolone, 19-norandrostenedione and 19-norandrostenediol, if these have been taken. 19-norandrostenedione (and its related compound androstenedione, which is commonly referred to as "Andro") which is a precursor of testosterone, are sold to athletes in many countries as

nutritional supplements rather than as drugs. The evidence that athletes benefit from the use of these compounds is not strong, but they have been endorsed by successful performers in sports where they are not banned, including especially baseball. Athletes are often more convinced by positive endorsements from successful performers (androstenedionne is not prohibited in baseball and has been heavily promoted by star performers in that sport) than they are by negative results from carefully controlled scientific studies.

Many different androgenic anabolic steroids, including nandrolone and testosterone itself, have been used by athletes over the years, and well-established measures are in place to detect abuse of these compounds (Verroken and Mottram, 2003). According to the figures released by UK Sport, 22 of the 110 positive findings (out of a total of 5406 tests conducted) in 2000-2001 were for anabolic agents, making this the second largest category of substances after stimulants (UK Sport, 2001). Many different anabolic agents are used in sport, and there is often a clear attempt by the athlete to gain an unfair advantage, but the apparent increase in the prevalence of nandrolone positive cases in recent years has generated much controversy, and the picture has not been entirely resolved (Kohler and Lambert, 2002). A closer examination of the statistics published by the IOC, however, does not entirely support this impression. Although there has been a steady increase in the number of nandrolone cases detected worldwide, these represent an almost constant fraction of the total number of tests carried out by IOC-accredited laboratories. If data from the first two years of nandrolone testing are ignored, almost exactly 0.25% of all tests yield a positive nandrolone case each year.

The Cologne results

A paper published in November 2000 provided the first reliable evidence of steroid contamination of dietary supplements (Geyer et al, 2000). This study reported the results of analysis carried out on legitimate dietary supplements, none of which declared on the label that they contained steroids, none of which would reasonably be expected to contain prohibited compounds, and none of which gave any warning to athletes that problems might result from their use. Analysis of these supplements established the presence of nandrolone,

testosterone and other steroids. When the supplements were fed to healthy volunteers in the recommended doses, they gave positive nandrolone urine tests, with urinary concentrations of up to 623 ng/ml (the threshold for a positive test is 2 ng/ml for men and 5 ng/ml for women). The supplements tested were identified as Chrysin, Tribulus Terrestris and Guarana, none of which should give a positive test, but the names of the companies producing these supplements were not revealed.

The Cologne laboratory followed this up with a much larger survey (dopinginfo.de, 2002a). Between October 2000 and November 2001 a total of 634 non-hormonal nutritional supplements were obtained in 13 countries from 215 different suppliers. The supplements were bought at retail outlets in the various countries (578 samples = 91.2 %), on the internet (52 samples = 8.2%) and by telephone (2 samples = 0.3 %). Two samples (0.3 %) were sent for analysis by the IOC. 289 supplements were from companies that were known to sell pro-hormones, and 345 supplements came from companies which did not sell prohormones. Testosterone and nandrolone, together with their metabolic precursors, were identified by gas chromatography/mass spectrometry after isolation from the supplement matrix. This required a different extraction process from that normally used for urine analysis and other components in the products gave problems with the analytical process in a significant number of cases.

Eleven different anabolic androgenic steroids were identified in these supplements. Of the 634 samples analysed, 94 (14.8 %) contained hormones or prohormones that were not declared on the label. Reliable data could not be obtained on a further 66 samples (10.4 %) because of the matrix effects referred to above. Of all positive supplements, 23 samples (24.5 %) contained compounds related to nandrolone and testosterone, 64 samples (68.1 %) contained prohormones of testosterone, and 7 samples (7.5 %) contained prohormones of nandrolone. A total of 49 supplements contained one steroid, but 45 contained more than one steroid and 8 products contained five or more different steroid compounds

Most of the positive supplements were bought in the Netherlands (25.8 %), in Austria (22.7 %) in the UK (18.8 %) and the USA (18.8 %). According to the labels on the products, all of the supplements that

were found to contain steroids were produced by companies located in one of only five countries (the USA, the Netherlands, the UK, Italy and Germany), although products purchased in other countries were also found to be contaminated. 21.1 % of the nutritional supplements from prohormone selling companies contained anabolic androgenic steroids, whereas proportionally fewer (9.6 %) of the supplements from companies not selling prohormones were positive. The positive supplements had a highly variable content of anabolic androgenic steroid (from 0.01 µg/g to 190 µg/g) and this varied between tablets or capsules within the same container. Excretion studies on healthy volunteers given nandrolone prohormones in doses of as little as 1 µg resulted in urinary concentrations of the nandrolone metabolite norandrosterone above the cut-off limit of the IOC for several hours.

The names of the prohibited supplements and of the companies that supplied them have not been published, but the products included amino acids and protein powders, Creatine, Carnitine, Ribose, Guarana, Zinc, Pyruvate, Hydroxy MethylButyrate, Tribulus Terrestris, Vitamins and minerals, and Herbal extracts. Many of these products are in common use among athletes. Although there have been many calls from athletes, coaches, team doctors and others for the names of contaminated products to be made public, this is probably not helpful, for several reasons. There is no certainty that other products, not named on such a list, are "safe", and some analyses have shown that contamination levels may vary greatly within a single product batch and even within a single bottle. This brings potential problems of litigation, and the laboratory conducting the tests would be liable to legal action from the manufacturers or suppliers. There have also been suggestions that products shown to be contaminated with steroids may experience a growth in sales among the major part of the athletic population that is not liable to testing for drugs.

Supporting evidence

The IOC-accredited laboratory in Vienna repeated the Cologne study with a smaller number (54) of supplements purchased in Austria. Twelve of these (22%) contained prohibited steroids, the same as the proportion of products purchased in Austria in the Cologne tests (23%). Unlike the German results, however, the identities of the

companies and the individual products have been published on the Internet (dopinginfo.de, 2002b).

The assumption has generally been that the presence of these steroid compounds is the result of accidental contamination during the manufacturing or packaging process rather than deliberate adulteration of the products in an attempt to increase their efficacy (Geyer et al, 2001). The amounts of steroids detected have been extremely variable, even within a single batch, but have generally been extremely small. The presence of small amounts of testosterone and related prohormones is not generally a problem as the testing procedures allow sufficient latitude to cope with small variations introduced by the ingestion of these compounds. Nandrolone testing, however, is much more specific and ingestion of as little as 1 μg of nandrolone of 19-androstenedione may result in a transient positive test. In addition to the results of Geyer et al (2000) which showed positive tests for nandrolone after ingestion of contaminated supplements, Delbeke et al (2002) have also shown that ingestion of contaminated supplements in amounts substantially less than the recommended dose could result in urinary 19-norandrosterone concentrations above the doping threshold for periods of up to 120 h. This relatively prolonged period makes the retrospective identification of the cause of a positive test more difficult to establish.

In 2002, the same laboratory found methanedienone in a supplement bought in England (Gmeiner, 2002). This drug was present in high amounts, enough to have an anabolic effect, but also enough to produce serious side effects, including liver toxicity and carcinogenicity. This finding arose because of a positive finding for methanedienone in a routine test on an athlete who denied having used the drug. Subsequent investigation identified this steroid in supplements used by this athlete. Samples of this product purchased by the laboratory were also found to contain substantial amounts of methanedienone. These results were later confirmed by the IOC laboratory in Cologne (Geyer et al, 2002). Unlike most of the cases involving steroids related to nandrolone and testosterone, this is not a trivial level of contamination and raises the possibility of deliberate adulteration of the product with the intention of producing a measurable effect on muscle strength and muscle mass. The prospect

of adverse health effects at these high doses also raises real concerns.
In 2003 the Cologne laboratory reported the results of the analysis for caffeine and ephedrine of 110 different supplements that did not include these compounds on the ingredients list declared on the product label. Of the 110 products analysed, 14 were found to contain caffeine and 2 to contain ephedrine (Parr et al, 2003). It is not clear how or why these compounds came to be present in these products, but it may well be that this was an attempt by the manufacturers to increase sales by ensuring a noticeable subjective response after ingestion of these products. Caffeine has been removed from the lists of doping substances issued by the IOC and WADA, but was prohibited at the time of this test.
There are numerous other reports in the literature of contamination of dietary supplements with toxic and prohibited compounds that are not identified on the label (Catlin et al, 2000; Kamber et al, 2000). Advising athletes to avoid the use of dietary supplements is not the solution, as they are reluctant to discontinue the use of supplements that offer a clear advantage. Dispensing with testing for nandrolone and other prohibited substances is not an option either, and nor is the abandonment of the strict liability rule.

A solution to the problem?

Various attempts have been made to address this problem, but there is no easy solution. Only a very small fraction of purchasers are affected by the presence of vanishingly small levels of prohibited drugs, so it may be unrealistic to apply pharmaceutical standards to the manufacture of food supplements. Equally, the athlete whose career and livelihood may be at stake deserves protection. Various schemes have been proposed for the testing of supplements with a view to producing approved lists, but such schemes face formidable difficulties. Contamination may vary from batch to batch and it has even been shown that some tablets or capsules within a single bottle may contain steroids while others do not. Uncertainties in the analytical process may affect at least 10% of supplements analysed. Administration of material to volunteers and testing of urine also raises real ethical and practical difficulties.
The only answer is for each athlete must carry out a cost benefit

analysis before using any supplement, but it is not at present possible to quantify some of the elements of this analysis. Supplements should be used only when there is good evidence of efficacy and safety, and athletes must consider the consequences of even a small risk of a positive test.