WHITE PAPER

SPORTS SUPPLEMENT MANUFACTURING

Recent Studies Reveal Potentially Harmful Ingredients — Demonstrating Need for GMPs, Ingredient Testing and Product Certification

In the fight for market share, sports supplement manufacturers and marketers are always searching for the next great innovation — a new ingredient or formulation that will give their product and their customers an edge. But the sports supplement business is under increasing scrutiny from regulators, consumers and the media. How do manufacturers balance the fundamental need for innovation with their responsibility to ensure the quality and safety of their products? It's not always easy. In fact, many companies are failing miserably.

Sports supplement usage is widespread in many sports – from running and cycling to baseball and bodybuilding. And it's not limited to elite athletes and professionals. Students, recreational athletes and "weekend warriors" are a significant segment of the fast-growing sports nutrition market. The supplement industry as a whole is growing at a fast pace, up from $4 billion annually in 1994 to more than $38 billion in 2016[1].

But dietary and sports supplements sometimes contain banned substances and mislabeled ingredients – just ask the many elite athletes who've been suspended from their sports due to inadvertent doping. While suspension from competition is a concern for elite athletes, it's not the greatest risk for most athletes and consumers. Researchers at NSF International, Harvard Medical School and other institutions have identified ingredients in supplements that can cause adverse health events, including liver damage, cardiac arrest and even death.

Suspension from competition is a concern for elite athletes, but it's not the greatest risk for most consumers.

Recent studies co-authored by NSF International's John Travis and researchers from Harvard Medical School, the National Center for Natural Products Research (NCNPR) at the University of Mississippi, and the National Institute for Public Health and the Environment in the Netherlands (RIVM) found that some supplements in the global market contain harmful ingredients

1 Nutritional Business Journal, 2016 NBJ Supplement Business Report
www.newhope.com/products/2016-nbj-supplement-business-report

Recent research revealed untested and potentially harmful compounds such as DMAA, DEPEA, DMBA and oxilofrine in over-the-counter supplements.

and contaminants, including drugs and untested compounds that are not always listed on the label. The research also revealed untested and potentially harmful compounds such as DMAA, DEPEA, DMBA and oxilofrine in over-the-counter supplements[2]. These ingredients are often deceptively labeled as botanical extracts such as geranium oil, dendrobium extract and pouchong tea extract – making it difficult for consumers and athletes to choose a supplement based on the ingredient listings alone.

Travis and his co-investigators are currently studying an untested and potentially harmful new ingredient used in some weight loss products.

This white paper explores recent examples of sports supplement contamination, potential causes and solutions for manufacturers and retailers who want to ensure quality and safety in the sports supplement supply chain. It also addresses:

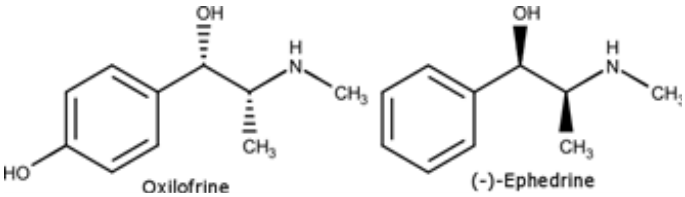
- How athletes and consumers protect themselves from potentially harmful ingredients
- What sports supplement manufacturers do to ensure their ingredients and finished products are safe
- The ingredients most commonly associated with untested and potentially harmful compounds

A PATTERN OF UNTESTED AND POTENTIALLY HARMFUL COMPOUNDS

Scientists at NSF International, Harvard Medical School, the National Center for Natural Products Research (NCNPR) at the University of Mississippi, and the National Institute for Public Health and the Environment in the Netherlands (RIVM) have studied sports supplement ingredients since 2004 and found many examples of untested and potentially harmful compounds in products available worldwide.

Oxilofrine

Unapproved Ephedrine-Like Stimulant Oxilofrine Found in 14 Dietary Supplement Products



In 2016, the research team found the unapproved pharmaceutical stimulant oxilofrine in 14 over-the-counter dietary supplement products. Their research was published in the peer-reviewed journal Drug Testing and Analysis. Oxilofrine is easily disguised or unlisted on labels, posing serious health risks to consumers.

The U.S. Food and Drug Administration (FDA) took action on April 4, 2016 to remove oxilofrine from the marketplace, issuing warning letters to seven companies that listed the compound on their product labels. According to the FDA, oxilofrine is an illegal dietary ingredient, which means products containing oxilofrine are considered misbranded under the law.

Oxilofrine has been studied in animals and humans and found to cause heart effects t similar to ephedrine, a compound banned by the FDA in 2004 due to serious side effects. Since then, scientists at NSF

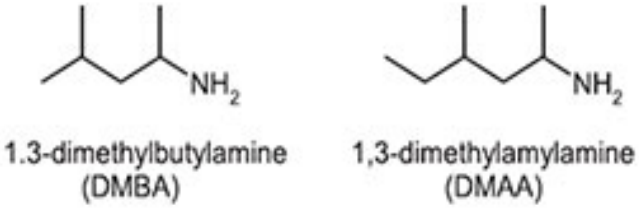
2 Cohen, P. A., Avula, B., Venhuis, B., Travis, J. C., Wang, Y.-H., and Khan, I. A. (2016) Pharmaceutical doses of the banned stimulant oxilofrine found in dietary supplements sold in the USA. Drug Test. Analysis, doi: **10.1002/dta.1976**.

International have discovered several unapproved and potentially dangerous replacement stimulants such as DEPEA, DMAA, DMBA and now oxilofrine in dietary supplements.

According to the research, 26 adverse events have been reported in the Netherlands linked to supplements containing oxilofrine. These supplements led to nausea and vomiting, tachycardia, chest pain and cardiac arrest. Additionally, oxilofrine is often disguised on labels as "methylsynephrine" or "extract of *Acacia rigidula*."

DMBA

Unapproved Synthetic Stimulant DMBA Found in Multiple Dietary Supplements



The chemical structures of DMBA and DMAA. Note that DMBA has one chiral center and that DMAA has two chiral centers.

In 2014, researchers from NSF International, Harvard Medical School and the National Institute for Public Health and the Environment in the Netherlands (RIVM) found an unapproved synthetic stimulant — 1, 3-Dimethylbutylamine or DMBA — in 12 over-the-counter dietary supplements. The findings were published in the peer-reviewed journal Drug Testing and Analysis.

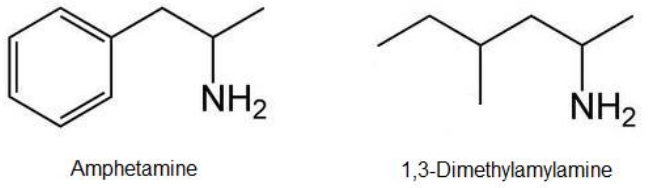
"While regulatory authorities work to remove harmful stimulants such as ephedrine and DMAA from supplements, new synthetic stimulants such as DMBA continue to crop up to take their place."

John Travis,
Senior Research Scientist, NSF International

The chemical structure of DMBA is similar to other banned stimulants like DMAA and ephedrine. DMAA was banned by regulatory agencies in the United States, United Kingdom, the Netherlands, Brazil and elsewhere because of its links to negative health events such as strokes, heart failure and sudden death. There are no known safety studies on DMBA, and its health effects are entirely unknown.

DMAA

Chemical Substance Acts Like Amphetamine and Is Not Naturally Occurring



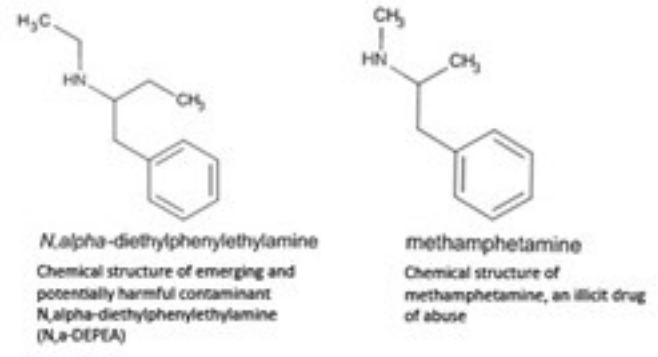
In 2013, researchers at NSF International and the U.S. Army Research Institute of Environmental Medicine (USARIEM), determined that 1,3 dimethylamylamine, also known as DMAA, is not "natural" in origin and should not be used as an ingredient in dietary supplements. The research findings support research conducted by academic research laboratories around the world.

Several sports supplements have tried to market DMAA as a natural constituent of geranium or its extract, but there is no credible scientific evidence to support that claim. Under the Dietary Supplement Health and Education Act (DSHEA), any naturally occurring ingredient can be sold over the counter as a dietary supplement, provided it was marketed and sold prior to

the signing of the act. Products containing DMAA have been linked to health problems and at least five deaths. As a result, the U.S. FDA issued several warning letters to manufacturers who have formulated their products with DMAA and have urged people to avoid these products. Several professional and Olympic athletes have lost their eligibility to compete due to DMAA. Stores and online retailers still sell products that contain DMAA, despite the FDA ban.

N,a-DEPEA

Emerging and Potentially Harmful Adulterant Found in a Dietary Supplement



In 2013, scientists at NSF International found evidence of an emerging and potentially harmful adulterant called N,alpha-diethylphenylethylamine (N,a-DEPEA) in a popular dietary supplement product. The substance has a chemical structure similar to methamphetamine and was found in a consumer dietary supplement product called Craze. Additionally, the substance (N,a-DEPEA) was not disclosed on the label. The substance was found as part of a collaborative testing project conducted by scientists at NSF International, Harvard Medical School (HMS) and the National Institute for Public Health and the Environment (RIVM) in the Netherlands.

In separate testing, NSF International scientists also detected N,a-DEPEA in a different supplement called Detonate by Gaspari Nutrition. Products containing this ingredient have been linked to several failed drug tests. DEPEA's addictive and pharmacological properties are unknown.

Half of all Americans take dietary or sports supplements, including multivitamins, minerals and herbs, to help with overall health, weight loss and strength training efforts.

SOURCE: U.S. CENTERS FOR DISEASE CONTROL AND PREVENTION (CDC)

OTHER NOTEWORTHY RESEARCH

In 2004, a study funded by the International Olympic Committee and published in the International Journal of Sports Medicine, found that 15 percent of 634 supplements tested from 13 countries contained steroids prohibited in sport, none of which were declared on the label.[3]

The **Journal of the American Medical Association** found that dietary supplements account for half of all Class 1 drug recalls in the United States. The research, supported by the Canadian Institutes of Health Research, showed that over a nine-year period (2004–2012), 51 percent of the U.S. Food & Drug Administration's (FDA) recalls involved dietary supplements containing unapproved ingredients or drugs. During this period, 465 products were recalled, of which 237 were dietary supplements.[4]

3 H. Geyer1 , M. K. Parr1 , U. Mareck1 , U. Reinhart1 , Y. Schrader1 ,W. Schänzer1. Analysis of Non-Hormonal Nutritional Supplements for Anabolic-Androgenic Steroids - Results of an International Study. Int J Sports Med 2004; 25(2): 124-129. DOI: 10.1055/s-2004-819955

4 Harel Z, Harel S, Wald R, Mamdani M, Bell CM. The Frequency and Characteristics of Dietary Supplement Recalls in the United States. JAMA Intern Med. 2013;173(10):929-930. doi:10.1001/jamainternmed.2013.379.

TOP 10 SUPPLEMENT INGREDIENTS TO AVOID

As an avid runner, NSF International's John Travis knows first-hand how supplements can support your performance and help you feel better. But as a chemist specializing in dietary supplements, he also knows that some supplements on the market contain ingredients that can have adverse effects on your long-term health. While most of the ingredients themselves are legal, many have health consequences when mixed with other ingredients and medications. Even exercise levels can impact how these ingredients affect your health. For example, when you're working out with an elevated heart rate, caffeine has an entirely different effect on your body compared to when you are at rest.

Supplements can be beneficial as long as you're aware of which ingredients to avoid. Here are the top 10 ingredients to avoid when purchasing or manufacturing dietary and sports supplements:

1. **Yohimbine:** This can appear on the label as erex, testomar, yocon, yohimar or yohimbe. Because of its chemical make-up, it is an alpha-blocker (which dilates blood vessels) and can cause serious harm to people already prescribed alpha-blockers or taking other drugs that dilate blood vessels. This includes Viagra, certain antidepressants and drugs containing nitrates, such as those that treat heart conditions.

2. **Phenethylamines:** This class of chemical compounds is sometimes listed as PEA or buried in a complicated ingredient name such as B-phenylethylamine or N-methylphenylethylamine. It is particularly dangerous for people taking monoamine oxidase inhibitor (MAOI) antidepressants because these drugs allow PEA to reside in the body longer, making its effects—such as dizziness and elevated heart rate—much more potent.

3. **Geranium:** Several sport supplement products have tried to pass off a harmful off-patent drug known as DMAA as a natural constituent of geranium or its extract. It is not related to geranium, and it has been associated with numerous adverse health events and even several deaths. Several professional and Olympic athletes have lost their eligibility to compete due to DMAA. Even though the U.S. FDA has banned this ingredient it may still show up in products in stores and online, so make sure to avoid it.

4. **Any ingredient with "andro" in the name:** Androstenedione or andro is a steroid hormone that is a precursor of testosterone. Many other steroids will also have "andro" within an ingredient name, such as 1-androsten-3b-ol-17-one and 4-chloro-17a-methyl-andro-4-ene-3,17b-diol. These are considered schedule 3 controlled substances, and nearly all major sports organizations, including the World Anti-Doping Agency, the U.S. Military and the Olympic Committee, have banned these steroids. Side effects include behavioral changes, heart disease, depression, high cholesterol and mood swings.

THE STATE OF THE SUPPLEMENT INDUSTRY

> The supplement industry has grown from $4 billion in 1994 to $38 billion in 2016. [5]

> In 1994 there were approximately 4,000 dietary supplements in the marketplace; in 2009 there were more than 55,000.

> Adverse event reporting has increased every year since 2006.

> There are more than 15,000 dietary supplement manufacturing facilities, with more than half located outside the United States.

> Of all these registered dietary supplement manufacturing facilities, less than 3 percent are audited annually in an average year.[6]

- When they are audited, two-thirds are found to have significant deficiencies and most have multiple or severe violations.

5 Nutritional Business Journal, 2016 NBJ Supplement Business Report **www.newhope.com/products/2016-nbj-supplement-business-report**

6 Kapoor, A., and Sharfstein, J. M. (2016) Breaking the gridlock: Regulation of dietary supplements in the United States. Drug Test. Analysis, 8: 424–430. doi: **10.1002/dta.1892**.

BUYER BEWARE: ADULTERATED FINISHED PRODUCTS

You can't always trust what's on the label. Just ask the elite athletes who have been suspended for inadvertent doping. Here's an example the labeled ingredients in a sports nutrition product:

Supplement Facts
Serving Size: 25 g (1 scoop)
Servings per container: 20

Amount per serving		% DV
Calories	40	
Total Fat	0	0%
Total Carbohydrate	5.5 g	1.8%
Protein (whey protein isolate, whey protein concentrate)	4 g	8%
Magnesium (aspartate)	15 mg	3.8%
Potassium (aspartate)	37 mg	1.1%
Creatine Monohydrate	5,000 mg	*
D-Ribose	5,000 mg	*
L-Glutamine	4,000 mg	*
L-Arginine Alpha Ketoglutarate	2,500 mg	*
Taurine	1,000 mg	*

*Daily Value (DV) not established.

Other Ingredients: Citric Acid, Lecithin, Natural & Artificial Flavors, Stevia Extract (leaf).

And here's what independent testing revealed was **actually** in the product:

- Terbutaline 110 ng/g (Beta-2 agonist)
- Morphine 13 ng/g (Pain killer)
- Amphetamine 92 ng/g
- N-Methylphenethylamine 1500 ng/g
- beta-Methylphenethylamine 220 ng/g
- Octopamine 310 ng/g (athletic banned stimulant)

While this product was marketed as a sports supplement, it contains at least six banned substances on World Anti-Doping Agency (WADA) and Major League Baseball (MLB) anti-doping lists.

5. **Bitter orange:** This botanical ingredient can have many other names such as biarade, seville or sour orange. When ephedrine (or ephedra) was banned, many weight-loss products used bitter orange instead because it has similar effects, such as constricting blood vessels and increasing blood pressure and heart rate. It is often combined with other similar compounds to create a stimulant "cocktail" that can contribute to cardiac events.

6. **Germander:** This is a member of the mint family, but germander has been linked to liver damage. In some instances, manufacturers have mistakenly used germander in place of skullcap, a known herbal remedy also in the mint family. The USDA conducted a study of 13 skullcap-containing supplements purchased online and found that only five actually contained skullcap and four contained the potentially toxic germander, so do your research before buying a supplement containing skullcap.

7. **Guarana:** Guarana is often combined with similar botanical or herbal ingredients containing caffeine, such as yerba mate, kola nut and/or green tea. This causes a caffeine stacking effect because the seeds of the guarana plant contain twice the concentration of caffeine as coffee beans. Yet supplements containing guarana do not always properly list the caffeine content on their labels. Consuming too much caffeine can cause caffeine toxicity which includes symptoms such as rapid heartbeat, anxiety, gastrointestinal disturbance and dizziness, among others.

8. **Yerba mate extract:** This herbal ingredient can contribute to elevated caffeine levels, but it also exposes users to cancer-causing substances depending on how it is processed. When harvested, yerba mate is traditionally dried or smoked over a wood fire, imparting its signature smoky flavor. When organic material such as the yerba mate leaf is combusted, cancer-causing nitrosamines can develop. This effect is further concentrated when yerba mate is processed into an extract or supplement ingredient.

9. **Kratom:** Derived from the kratom tree that grows in Southeast Asia, kratom contains mitragynine alkaloids that exhibit some of the same pharmacological effects as opium. This ingredient is newer to the U.S. dietary supplement scene. Products containing this ingredient have been recalled as consumption may lead to nausea, dizziness, constipation and, in worst cases, hallucinations and delusions. FDA currently has an import ban on this ingredient and products that contain it.

10. **Bael tree fruit:** This fruit contains a harmful chemical called aegeline, which was found in the weight-loss supplement OxyElite Pro. This product was linked to 50 cases of liver damage including two transplants and one death. It can appear on product labels as N-[2-hydroxy-2(4-methoxyphenyl) ethyl]-3-phenyl-2-propenamide.

QUALITY AND SAFETY START WITH GOOD MANUFACTURING PRACTICES (GMPS)

In the United States, Good Manufacturing Practices (GMPs) are a system of procedures and documentation, written or analytical, to ensure that the product has the identity, strength, composition, quality and purity that it is represented to possess.

GMPs include:

- Standard operating procedures (SOPs)
- Formulation records
- Storage procedures
- Equipment testing
- Cleaning protocols

For products sold in the U.S., manufacturers must register with the U.S. FDA. This means they are on the FDA's radar for inspections.

Common Problems in Manufacturing Facilities

As the global leader in dietary supplement GMP facility registration, NSF International conducts hundreds of GMP audits in dietary supplement manufacturing facilities each year. According to NSF International, the safety of sports supplements is often compromised by three common failures in manufacturing facilities:

- Failure to verify the identity of a dietary ingredient prior to use
- Failure to follow written procedures for QA operations
- Failure to verify the finished product

Reputable companies can seize this opportunity to showcase their products' safety, verify label claims and differentiate themselves in the marketplace.

INCREASED SCRUTINY BY REGULATORS AND LAW ENFORCEMENT

During the last few years, regulators and law enforcement agencies have started taking a closer look at the supplement industry. In Europe, over 2.6 tonnes of counterfeit and substandard food supplements were seized in 2014 as part of the Interpol/Europol Operation OPSON IV. Also in 2014, the Italian Competition Authority fined a manufacturer 250,000 for using unauthorized health claims in promoting a food supplements.

In the United States, the U.S. Department of Justice has pursued civil and criminal cases against more than 100 dietary supplement companies for supplements containing unlisted ingredients or making unsupported claims since 2014. The U.S. FDA has seized products and shut down several manufacturers for violations of U.S. regulations (21 CFR Part 111) and courts are handing down hefty fines and contempt sentences.

THE SOLUTION: INDEPENDENT TESTING AND CERTIFICATION

As these high-profile cases illustrate, the actions of a few bad manufacturers can irrevocably harm brand reputation and reflect negatively on the entire supplement industry. Fortunately, reputable companies can seize this opportunity to showcase their products' safety, verify label claims and differentiate themselves in the marketplace. Independent third-party certification of dietary and sports supplements and ingredients helps ensure safer, quality products.

Three Levels of Compliance and Certification

So how do reputable manufacturers demonstrate the quality of their products and verify the accuracy of label claims? Independent, third-party organizations like NSF International currently certify to three levels of compliance and certification standards.

- Regulatory compliance with Good Manufacturing Practices.
- Certification to NSF/ANSI 173: *Dietary Supplements*, the official American National Standard for dietary supplement products.
- Independent, third-party certification with testing on a lot-by-lot basis for over 270 athletic banned substances. The NSF Certified for Sport® program is an example of this most rigorous level of certification.

Let's take a closer look at what is required to achieve the highest level of independent third-party certification, which involves all three steps above.

Regulatory Compliance and GMPs

In the United States, supplement companies are required to ensure their products are not contaminated, mislabeled or harmful, and to comply with regulatory standards such as 21 CFR Part 111, Good Manufacturing Practice (GMP). Testing and certification ensures ingredients meet these requirements for quality and safety, provides documented evidence of ingredients and test results, and verifies label claims.

GMP guidelines require processes and documentation to assure a product has the identity, strength, composition, quality and purity that appear on its label. GMP guidelines apply to dietary supplement, ingredient and raw material manufacturers and to distribution, warehousing and packaging companies. GMP regulations also require identification of all raw ingredients.

Verifying GMPs involves assessing the physical plant and grounds, personnel, equipment, production and process control systems, holding and distribution

processes, recordkeeping, and procedures for handling recalls, product returns and product complaints.

Product Certification to the American National Standard for Dietary Supplements

Next, supplement manufacturers can work toward dietary supplement certification. In addition to passing twice annual GMP audits, certification requires products to be tested to verify compliance with NSF/ANSI 173: *Dietary Supplements*, the official American National Standard for dietary supplement products. This includes:

- Label claims and content verification, as well as contaminant testing. Certification verifies the contents of the package are the same as the label and that there are no harmful levels of specific contaminants.
- Verification of product formulation and label claims through a toxicology review.
- Ongoing monitoring to verify compliance through periodic auditing and testing.

Contaminant testing includes metals that pose health risks (like lead, mercury, arsenic, cadmium and chromium VI), microbial contaminants, aflatoxins, pesticides and herbicides. Testing can also detect fillers and allergens, even at very low abundance (down to a few molecules of DNA). This helps prevent possible allergic reactions and ensures manufacturers are getting the ingredients they are paying for. The standard also provides criteria for determining that GMPs were followed in the production of dietary supplements.

Certification to NSF/ANSI 173: *Dietary Supplements* provides a means to source safer products, raw materials and ingredients. Regardless of where ingredients are sourced, proper testing and qualification of suppliers is paramount to maintaining control over the supply chain and ensuring the quality and safety of finished products.

NSF Certified for Sport® supplements are tested on a lot-by-lot basis for over 270 athletic banned substances from the World Anti-Doping Agency (WADA), NSF Annex B, NFL and MLB prohibited substance lists.

Sports Supplement and Ingredient Testing and Certification

Finally, supplement manufacturers can take an extra step and work toward independent certification for safer use by athletes. For example, the NSF Certified for Sport® program builds on the NSF/ANSI 173: *Dietary Supplements* standard by screening for athletic banned substances. This rigorous certification program was developed with regulatory, sports industry and consumer groups to help athletes and consumers choose supplements that do not contain banned substances.

Because each product is unique, certification involves customized test methods relevant to the particular type of supplement. For example, ingredients known to contain aristolochic acid are assayed for it, botanicals are screened for pesticides and glycerin products are tested for diethylene glycol.

Sports supplements are tested on a lot-by-lot basis for over 270 athletic banned substances from the World Anti-Doping Agency (WADA), NSF Annex B, NFL and MLB prohibited substance lists. Testing covers various pharmacological activity classes, including anabolic steroids (like testosterone and stanozolol), stimulants (like amphetamine and DMAA), diuretics (like chlorothiazide and bumetanide), beta agonists (like albuterol and salmeterol), beta blockers (like atenolol and metoprolol), narcotics (such as morphine), cannabinoids (like THC), hormones and masking agents. This list is monitored and updated as new substances are discovered or banned. This is why the NSF Certified for Sport® program is used by the NFL, NHL, MLB, PGA, LPGA, Canadian Centre for Ethics in Sport (CCES) and the New York City Police Department.

INCREASED DEMAND FOR CERTIFIED SPORTS SUPPLEMENTS

Rob Ninkovich learned a costly lesson in September 2016. The New England Patriots defensive end was suspended for four games after testing positive for a banned substance — one he wasn't even aware he'd taken. Ninkovich told U.S. cable network ESPN, "Any supplement I've ever used was bought at a store. I was unaware something I bought had a substance in it that would give me a positive test because it wasn't listed [as an ingredient on the label]."

His story is all too common. Liverpool football player Mamadou Sakho. Welsh runners Rhys Williams and Gareth Warburton. Canadian Boxing champ Lucian Bute. Elite athletes all over the world are learning that dietary supplements sometimes contain banned substances and mislabeled ingredients. Under strict anti-doping rules, testing positive for banned substances can lead to lengthy suspensions – even for athletes like these who violated the rules unintentionally.

"One thing I have learned is that if a supplement is not NSF certified, there are no regulations that ensure that what is on the label is 100 percent accurate. That is a hard lesson for me to learn at this stage in my career, but I take responsibility for it. It's a mistake I made and it hurts that I won't be there for my teammates," Ninkovich said.

LABORATORY TEST METHODS

To comply with both U.S. and European regulations, nutritional product manufacturers are required to test incoming ingredients and products to confirm their identity, purity and potency. This sort of testing is particularly important when sourcing ingredients from new suppliers or from countries without robust regulatory structures. But without an advanced degree in chemistry, it can be difficult to know which tests are appropriate in each situation.

How do chemical tests differ from microbiological tests? And what can we learn about dietary supplement ingredients from DNA sequencing? There are three main types of testing that are important to the dietary supplement industry:

- Microbiological testing
- Chemical testing
- DNA testing or sequencing

Microbiological tests are typically used for contaminant testing and label claims verification. Contaminant testing in dietary supplement ingredients usually includes screening for yeast and mold, *Salmonella*, *E. coli* and *Staphylococcus*. Microbiological tests can also be used to quantify total aerobic microbial count and to verify label claims related to probiotic strains and activity.

Chemical testing includes many different test techniques and methods that can be used to produce a variety of qualitative and quantitative measures. This includes:

- Contaminant testing for heavy metals such as arsenic and cadmium
- Label claims verification for minerals like calcium, sodium and potassium
- Label claims verification for qualitative identification and quantitative measurement of botanicals and dietary supplement ingredients
- Screening for athletic banned substances

Because each product is unique, laboratory procedures and test methods must be customized to meet specific analytical needs. Some of the techniques used in these methods include:

- **High Performance Thin Layer Chromatography (HPTLC) –** This technique is particularly useful for testing botanical ingredients. It can identify the ingredient and differentiate between various plant parts, such as the root, leaves or stem. HPTLC is used to authenticate samples by comparing a "fingerprint" of chemical bands in a sample to

the "fingerprint" of a known reference material. This technique can be used to identify processed samples such as finished products, oils and extracts. However, HPTLC is limited by the complexity of the sample material. For example, it may have limited value with a material that contains multiple botanicals.

- **High Performance Liquid Chromatography (HPLC) –** This technique is used to identify and measure supplement ingredients such as vitamins, caffeine and amino acids. HPLC can also measure specific marker compounds found in botanical ingredients and finished products. It can sometimes identify adulteration based on the chemical profiles of markers. Depending on the method, several compounds can be analyzed at once using this technique. Identifications are based on comparison of the detection characteristics, such as retention time, to that of an authentic reference standard.
- **Gas Chromatography (GC) –** GC is also used to identify and measure specific marker compounds found in botanical ingredients, oils, extracts and finished products. This technique can potentially identify cases of adulteration. This is accomplished by comparing the chemical profile of specific markers in a material to their expected ratios. For example, saw palmetto has a distinct fatty acid profile. If the fatty acid profile of a material differs from the reference materials or profile, the material may be adulterated.
- **High-Resolution, Accurate-Mass Mass Spectrometry (HRAM-MS) –** This is an important and highly powerful tool. This instrumentation has the capability to accurately measure the mass of a substance to the third decimal place and beyond. For example, it could measure the mass of caffeine to 194.080 Da. When coupled to a GC or HPLC, this technique provides a potent detection technique capable of discriminating between components of complex materials.
- **Nuclear Magnetic Resonance (NMR) Spectroscopy –** This remarkable tool can be used to determine the relative position of atoms within a molecule, which helps reveal the chemical structure of a substance. It is a powerful tool for identifying single components. It can also be used for complex mixtures, such as the analysis of aloe inner leaf gel material.

Of course, this isn't an exhaustive list of scientifically valid techniques and methods used in the analysis of dietary supplements, but it's a good place to start.

DNA testing is becoming increasingly common in the dietary supplement industry. While DNA barcoding has become synonymous with "DNA testing," there are numerous other ways to perform DNA testing.

DNA testing is a valuable authentication tool when used in combination with chemical and microbiological testing. Chemical testing generally requires a comparison of a material against a known reference material, which can be problematic if you are unsure of the contents of the test sample. DNA testing identifies which species are present in a sample. At that point, it is possible to follow up with additional chemical testing to determine the chemical composition of the material.

AN OPPORTUNITY TO DEMONSTRATE QUALITY AND SAFETY

The actions of a few bad manufacturers can irrevocably harm brand reputation and reflect negatively on the entire supplement industry. Fortunately, reputable companies can seize this opportunity to showcase their products' safety, verify label claims and differentiate themselves in the marketplace.

In 2017, more and more athletes are looking for certified products. The dietary supplements industry plays a major role in protecting supplement safety and consumers are increasingly savvy, demanding transparency and trust. Being able to prove that what is on the label is what is in the product is beneficial for producers, suppliers, retailers and consumers.

Testing and certification increase product quality and reduce the risk of adverse events caused by unidentified ingredients. Testing and certification also reduce the risk of litigation and regulatory action. When products are independently tested and certified, consumers can be confident that those products will not result in accidental doping or adverse health effects.

Learn more about NSF International's GMP facility registration and product certification for dietary supplements at **www.nsf.org**.

Learn more about NSF Certified for Sport® at **www.nsfsport.com**.

OUR MISSION

NSF International is dedicated to being the leading global provider of public health and safety-based risk management solutions while serving the interests of all stakeholders, namely the public, the business community and government agencies.

ABOUT THE AUTHOR

John Travis

SENIOR RESEARCH SCIENTIST
NSF INTERNATIONAL

John Travis has more than 20 years of experience as an analytical chemist specializing in the analysis of dietary supplements. As Senior Research Scientist at global public health organization NSF International, Travis analyzes hundreds of dietary supplement products each year for various contaminants, emerging drugs and harmful compounds. Utilizing techniques ranging from gas chromatography to high-performance liquid chromatography to mass spectrometry, John has developed and validated analytical methods for numerous marker compounds and trace contaminants. He is an active member of the Association of Analytical Communities (AOAC), participating on expert review panels for carotene and lutein, and is a member of the AOAC stakeholder panel for dietary supplements.

Travis is also a subject matter expert on athletic banned substances and was instrumental in the development of the screening methods used for the NSF International's Certified for Sport® program, which now screens products for more than 270 banned substances on the World Anti-Doping Agency, National Football League, Major League Baseball and National Collegiate Athletic Association lists. Travis is currently involved with the analysis of pharmaceutical agents and illicit drugs, stimulants and other prohibited substances as both adulterants and contaminants in dietary supplements and functional foods, co-authoring scientific papers on ingredients of concern including stimulants drugs DMAA, DEPEA and DMBA found in dietary supplements.