

Review of Possible Sources of Exposure of Horses to Natural Products and Environmental Contaminants Resulting in Regulatory Action

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Natural products and environmental contaminants may be responsible for positive drug findings that result in regulatory actions against horse trainers. Knowledge of these potential sources of drugs may help horse trainers avoid sanctions. Author's address: Analytical Toxicology Laboratory, College of Veterinary Medicine, The Ohio State University, 601 Vernon L. Tharp St., Columbus, OH 43210. © 1997 AAEP.

1. Introduction

The low limits of detection of modern analytical methods such as immunoassays and mass spectrometry have increased the ability of racing analysts to detect drugs, their metabolites, and other analytes in test samples. The use of these methods has resulted in positive chemical findings for a number of previously unreported substances and for other substances at concentrations that were not previously reported. Some of these substances, such as the alkaloids scopolamine and atropine and the cocaine metabolite benzoylecgonine, are probably present in test samples as a result of environmental contamination. Other substances, such as the xanthine alkaloids caffeine and theobromine and the alkaloids morphine, ephedrine, phenylpropanolamine, norpseudoephedrine, and pseudoephedrine, may be present as a result of the administration of various natural products to horses. An identification of the sources of these substances is frequently difficult but may assist trainers in avoiding or reducing sanctions. The purpose of this review is to identify some

known or probable sources of drugs as environmental contaminants or as components of natural products as an aid to veterinarians who are sometimes consulted by trainers concerning such matters. The review also includes a discussion and summary of the components of some Chinese remedies used by horse trainers.

2. Materials and Methods

Information contained in this review was obtained from relevant scientific literature, quarterly and annual reports of the Association of Official Racing Chemists from 1987 to 1996, transcripts of administrative hearings of racing officials, and product information bulletins. Furthermore, interviews were conducted with several racing officials, defense attorneys, racing analysts, trainers, veterinarians, and veterinary pharmacologists.

3. Results

Several of the xanthine alkaloids (e.g., caffeine, theobromine, and theophylline) have been reported

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from official racing samples collected in the U. S. The number of reports increased after an enzyme-linked immunosorbent assay (ELISA) for caffeine became commercially available in 1990. Investigations have revealed that some of these findings are attributed to the feeding of various xanthine-containing beverages, foods, natural remedies, nutritional supplements, or contaminated feeds.

Positive findings for the related alkaloids ephedrine, pseudoephedrine, phenylpropanolamine, and norpseudoephedrine were reported during the past decade. In at least two cases, all four alkaloids were found together in the urine sample along with caffeine. These findings are attributed to the administration of herbal products containing the dried plant stem of *Ephedra sinica* Staph. or other *Ephedra* species as well as caffeine. These products are sold under various names such as Herbal Ecstasy and are used as central nervous system stimulants.

Several reports for the presence of morphine in official samples have appeared in the past decade. These findings have often been vigorously contested by horse trainers who claim no knowledge of the use of morphine or morphine-containing products in their stables. One trainer attributed the morphine in the urine sample to unintentional feeding of poppy seeds containing morphine. Bagels were identified as the possible source of the poppy seeds in the stable. The stewards exonerated the trainer when the expert witness for the racing board testified that the feeding of poppy seeds to horses resulted in morphine concentrations greater than those measured in the official sample.

Hordenine and bufotenine have been reported from official samples collected in the U. S. and Europe. Hordenine was once believed by some analysts to be used illicitly by trainers as a substitute for ephedrine. Later, after it was demonstrated that hordenine is a normal constituent of hay made from reed canary grass (*Phalaris arundinacea* L.), reports of positive findings for hordenine from the U. S. largely ceased. Two of the findings for bufotenine were vigorously disputed by the trainers, particularly because proposed sanctions for this hallucinogen were severe. The hearing officer recommended dismissal of the charges against both trainers after an expert witness for the trainers testified that the oral administration of 5-methoxy-*N,N*-dimethyltryptamine (an occasional constituent of reed canary grass hay) to horses resulted in the appearance of bufotenine in the urine without any evidence for the administered substance.

Several recent reports for scopolamine and atropine from official samples have been attributed to the ingestion of plant materials or seeds from jimsonweed (*Datura stramonium* L. and *D. meteloides* L.) and other *Datura* species. Sanctions against several trainers were not imposed after an expert witness for the racing board testified that the administration of seeds from jimsonweed (*D. meteloides*) to horses resulted in the identification of both atropine

and scopolamine in the urine in the same relative concentration ratio as was found in the official samples. Other possible sources of this alkaloid mixture were sought but none were identified.

The detection of cocaine metabolites in numerous official samples has been reported since 1990. These findings coincide with the introduction of very sensitive cocaine ELISA tests, which first became available commercially around 1990. Concentrations of benzoylecgonine and ecgonine methyl ester in official urine samples have generally ranged from approximately 10 to 50 ng/ml. These concentrations are lower than the threshold concentration (150 ng/ml) for benzoylecgonine in urine samples collected from military personnel and transportation employees in the U. S. These concentrations are also considerably less than the urine concentrations reported after the administration of cocaine doses necessary to produce an effect on exercising horses. Some stewards have chosen not to penalize certain trainers with impeccable reputations and outstanding stable management whose horses have produced urine samples containing low concentrations of cocaine metabolites. These rulings have been relatively uncommon, however.

Pyrilamine and procaine are reported from numerous urine samples annually. Many of these findings are unexplained but vigorously contested. Some of these findings have been attributed to the contamination of certain vitamin products with pyrilamine and certain horse feeds with procaine from procaine penicillin contained in pig feeds that were prepared by the feed mill immediately before preparation of the horse feed. An unambiguous demonstration of these sources of contamination has persuaded some racing authorities to dismiss charges against trainers.

Chinese herbal remedies have been confiscated from horse trainers at several race tracks. These products include Yunnan Paiyao and Endurox, containing ciwujia. Although no positive reports have been attributed to the administration of these substances to horses, they and similar products are a potential source of drug violations because their manufacture is unregulated.

4. Discussion

The xanthine alkaloids caffeine, theophylline, and theobromine stimulate the heart and central nervous system, produce diuresis, and dilate the bronchioles. They are obtained from a variety of sources including coffee beans (*Coffea arabica* L. or *C. liberica* Hiern), cola nuts (*Cola nitida* Ventenat), tea (*Camellia sinensis* L.), maté leaves (*Ilex paraguensis* St. Hil.), guarana paste (*Paullinia cupana* Kunth), and cacao beans (*Theobroma cacao* L.). The alkaloids are found in various beverages and foods such as soft drinks, coffee, tea, and dark and milk chocolate. Theobromine may also be found in feed as a result of contamination with cacao husks, and caffeine is found in various health food store products such as Choline Cocktail from Twinlab. Some products con-

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tain caffeine as a result of the presence of guarana extract or guarana paste, although they may bear labels stating that they contain no added caffeine or that they contain guaranine (a synonym for caffeine). Caffeine is also found in several over-the-counter products such as Vivarin and NoDoze for appetite suppression and increased alertness.

The feeding of chocolate and chocolate candies to horses can result in positive findings for both theobromine and caffeine. Theobromine and caffeine were detected in urine samples collected from research horses administered twenty M&M chocolate peanuts daily for 8 days. Concentrations of theobromine exceeded the international threshold of 2 µg/ml for several hours after the last administration, and the concentrations of theobromine in urine samples were greater than those of caffeine at all times. Examination of the theobromine-to-caffeine concentration ratios in urine may allow veterinary pharmacologists to differentiate chocolate ingestion from caffeine administration. These interpretations obviously will require analysts to perform quantitative analyses for these alkaloids in test samples.

A positive finding for caffeine in a horse has usually been assumed to result from caffeine ingestion or caffeine administration. However, analysts from Novamann (Quebec) Inc. have recently reported that the administration of theophylline to horses results in positive findings for caffeine at peak concentrations of approximately 100 ng/ml and 200 ng/ml in blood and urine, respectively. These results confirm that caffeine is a metabolite of theophylline in the horse as it is in several other species.

Positive findings for several alkaloids such as ephedrine, pseudoephedrine, phenylpropanolamine, and norpseudoephedrine have resulted from the administration of various unregulated health food store products. Ephedrine and related alkaloids are contained in several products (e.g., Ma Huang, Ephedra, Herbal Ecstasy), whereas phenylpropanolamine appears to have contaminated others such as some Spinach Octacosanol Tablets as a result of poor manufacturing practices.

Positive findings for morphine have been attributed to the ingestion of poppy seeds from *Papaver somniferum* L. These seeds are used in the preparation of several bakery products such as poppy seed cakes and bagels. The administration of 1–5 g of commercial poppy seeds containing approximately 70 µg of morphine/g of seeds to horses resulted in detectable concentrations of morphine in urine for at least 24 h after the dose. Other sources of morphine such as feeds containing bakery byproducts contaminated with poppy seeds appear possible and may account for morphine findings when no other explanation is found. Another possible but unproved explanation for the presence of morphine in urine is the fact that morphine is an endogenous substance in mammals, although it is usually present in very low concentrations.

Hordeanine and bufotenine have been detected in

urine samples collected from horses after ingestion of reed canary grass (*P. arundinacea*). The identification of bufotenine in two official test samples was attributed to the ingestion of reed canary grass hay containing 5-methoxy-*N,N*-dimethyltryptamine after it was demonstrated that horses readily metabolize 5-methoxy-*N,N*-dimethyltryptamine to yield bufotenine (Fig. 1). Demonstration of this metabolic pathway and testimony that the urine also contained several other alkaloids commonly found in reed canary grass persuaded a hearing officer to recommend no sanctions against the trainers whose horses produced urine samples containing bufotenine.

Several recent reports for scopolamine and atropine from official samples have been attributed to the ingestion of plant materials or seeds from jimsonweed (*D. stramonium* L. and *D. meteloides* L.) and other *Datura* species. The contamination of animal feeds with jimsonweed seeds is relatively common and has been responsible for poisoning of livestock. A recent report describes the detection of both atropine and scopolamine in urine samples collected from horses fed seeds and other plant materials from *D. meteloides*.¹ These investigators showed that doses as small as 6.5 g of plant material resulted in peak urine concentrations of scopolamine near 100 ng/ml 1 h after dosing with a corresponding atropine concentration approximately 10% of that of scopolamine.

Cocaine is a widely distributed environmental contaminant because of its abuse by humans. Paper currency in the U. S. is frequently contaminated with cocaine. In one study, currency from banks in 12 cities was found to contain cocaine ranging from a few nanograms to 270 µg per unit of currency.² The presence of relatively low concentrations of the cocaine metabolites ecgonine methyl ester and benzoylecgonine (Fig. 2) may result from unintentional exposure of a horse to cocaine-contaminated feeds or tack. Because cocaine is readily absorbed after dermal exposure in humans,³ dermal exposure of horses may result in the presence of cocaine metabolites in the urine. The effects of cocaine on treadmill exercise by horses were evaluated by McKeever et al.⁴ These studies demonstrated that cocaine doses greater than 100 mg administered intravenously 5 min before exercise are necessary to produce an effect on exercise. Urine concentrations of benzoylecgonine after cocaine doses such as these would be expected to be several micrograms per milliliter.^{5,6}



Fig. 1. Metabolic pathway for oxidation of 5-methoxy-*N,N*-dimethyltryptamine to bufotenine in horses.

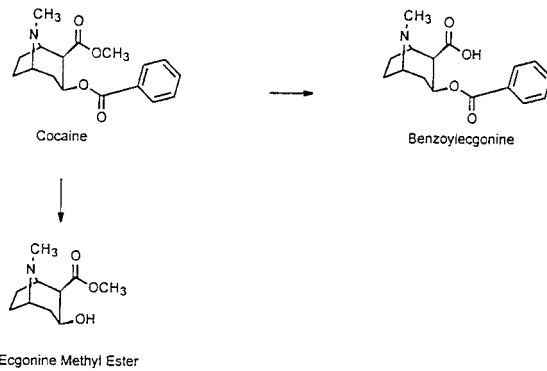


Fig. 2. Metabolic pathway for cocaine in horses.

Pyrilamine is an antihistamine drug that is contained in several nonprescription products for horses. Numerous unexplained positive findings for a major metabolite of pyrilamine have been reported. One possible explanation for some of these findings is pyrilamine contamination of Ultra-Plus and Ultra-Vite multivitamins and mineral supplements for horses.

Procaine has been identified from horse urine as a result of the ingestion of feed contaminated with procaine penicillin caused by an inadequate cleaning of equipment in a feed mill following preparation of a pig feed containing Aureo SP, an antibiotic mixture. Regulatory authorities accepted this explanation when it was demonstrated that the urine samples also contained sulfamethazine, a sulfonamide also contained in the antibiotic mixture, and the trainer obtained records from the feed company documenting their preparation of pig feed containing Aureo SP immediately before preparation of the trainer's horse feed.

Yunnan Paiyao and Endurox, containing ciwujia, are two products that are available in health food stores and nutrition centers that are used by some horse trainers to treat their horses. Use of these products may be motivated by a belief that natural products are permitted by rules of racing or that they will not be detected by laboratory tests. These products are purported to contain Chinese herbal remedies. However, the veracity of these claims has not been reported.

The findings summarized in this review provide considerable evidence that various products sold to horse trainers as well as environmental contaminants are responsible for some drug violations. The recognition of these sources by veterinarians may help trainers avoid drug violations or may aid in the defense of individuals accused of drug violations under the absolute insurer rule.

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