Sweat Testing in Opioid Users with a Sweat Patch

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Abstract

For many years, toxicologists have detected the presence of drugs of abuse in biological materials using blood or urine. In recent years, remarkable advances in sensitive analytical techniques have enabled the analysis of drugs in unconventional samples such as sweat. In a study conducted in a detoxification center, sweat patches were applied to 20 known heroin abusers. Subjects wore the patch with minimal discomfort for five days. During the same period, two urine specimens were also collected. Target drugs analyzed either by gas chromatography–mass spectrometry (GC–MS) or liquid chromatography–mass spectrometry (LC–MS) included opiates (heroin, 6-monoacetylmorphine, morphine, codeine), cocaine (cocaine, benzoylecgonine, ecgonine methyl ester), Δ^9-tetrahydrocannabinol, benzodiazepines (nordiazepam, oxazepam), amphetamines (amphetamine, methamphetamine, methylenedioxyamphetamine [MDA], methylenedioxymethamphetamine [MDMA], methylenedioxyethylamphetamine [MDEA]), and buprenorphine. Patches were positive for opiates in 12 cases. Heroin (37–175 ng/patch) and/or 6-acetylmorphine (60–2386 ng/patch) were identified in eight cases, and codeine exposure (67–4018 ng/patch) was determined in four cases. When detected, heroin was always present in lower concentrations than 6-acetylmorphine, which was the major analyte found in sweat. Cocaine (324 ng/patch) and metabolites were found in only one case. Δ^9-Tetrahydrocannabinol (4–38 ng/patch) was identified in nine cases. Benzodiazepine concentrations were very low, ranging from 2 to 44 and from 2 to 15 ng/patch for nordiazepam and oxazepam, respectively. MDEA (121 ng/patch) and its metabolite, MDA (22 ng/patch), were detected in one case. Buprenorphine, which was administered as therapy under close medical supervision, was detected in the range 1.3–153.2 ng/patch with no apparent relationship between the daily dose and amount excreted in sweat. All the urine tests were consistent with the sweat findings, but to identify the same drugs it was necessary to test two urine specimens along with only one sweat specimen. It was concluded that sweat testing appears to offer the advantage of being a relatively noninvasive means of obtaining a cumulative estimate of drug exposure over the period of a week. This new technology may find useful applications in the treatment and monitoring of substance abusers, as the patch provides a long-term continuous monitor of drug exposure or noncompliance.

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Introduction

Given the limitations of self-reports of drug use, testing for drugs of abuse is important for most treatment programs, both for monitoring the progress of the patient and for assessing the effectiveness of particular interventions in controlled clinical trials.

For many years, analysts have detected the presence of drugs in biological materials using blood and urine. In recent years, advances in sensitive analytical techniques have enabled the analysis of drugs in unconventional biological samples such as sweat. Researchers have known since 1911 (1) that drugs are excreted by the body in sweat, but no one has developed a practical solution to the problem of capturing sweat before testing until recently. Occlusive bandages consisting of one to three layers of filter paper or pieces of cotton, gauze, or towel were proposed to collect sweat. Significant advances have been made during the past years to develop a sweat patch technology, which was recently developed by Sudormed™ (Santa Ana, CA) and marketed by Pharmchem™ Laboratories (Menlo Park, CA) under the tradename Pharm-Chek™. The sweat patch acts as a specimen container for nonvolatile and liquid components of sweat, including drugs of abuse. Sweat components are collected on a special absorbent pad, which is located in the center of the patch. Nonvolatile substances from the environment cannot penetrate the transparent film, which is a semipermeable membrane over the pad that allows oxygen, water, and carbon dioxide to pass through the patch and leaves the skin underneath healthy. Over a period of several days, sweat saturates the pad, and drugs that are present are retained.

To date, only a few applications of the sweat patch have been described (2–4). In these studies, the authors concluded that sweat testing appears to offer the advantage of being a relatively noninvasive means of obtaining a cumulative estimate of drug exposure over a period of several days. More recently, to

validate this new technology, we described a series of sweat excretion experiments with subjects who were receiving single oral doses of codeine, phenobarbital, or diazepam (5,6). Four types of experiments were performed: cumulative excretion, time course of excretion, influence of the site of patch application, and dose–concentration relationship.

The aim of this report is to compare the usefulness of urine and sweat for the monitoring and managing of patients on buprenorphine substitution maintenance.

Materials and Methods

Chemicals

Methanol and acetonitrile were HPLC grade (Merck, Darmstadt, Germany). Other chemicals were analytical grade and were provided by Merck. All drugs and deuterated internal standards were purchased from Radian (Austin, TX), with the exception of SKF 525A (proadifen) which was obtained from Smith, Kline, and French Laboratories (Herts, U.K.).

Samples

Twenty heroin addicts participated in the study. All subjects met routine clinical criteria indicating opioid addiction, such as ten years of intravenous heroin abuse. In addition, some subjects were mothers with newborns, and some subjects were developing diseases such as AIDS or hepatitis C. Subjects were not paid for participation. According to the medical staff, a positive result would not exclude the subject from continuing buprenorphine therapy.

All subjects were verbally informed about the procedure and gave a verbal informed consent agreement. Sweat patches were applied to the outer portion of the upper arm or back. The selected skin site for patch placement was gently cleaned with a 70% isopropanol swab before application. In all cases, the patch was applied on a Monday morning and removed 5 days later, on Friday, by pulling an edge of the adhesive backing, taking care not to touch the absorbent pad. After removal of the patch, the pad was stored in plastic tubes at –20°C. At the same time, on day 0 and day 5, a urine specimen was collected and was frozen until analysis.

Sweat patches were provided by Pharmchem Laboratories. No financial support was obtained for this study.

Analysis of sweat patches

The target drugs were extracted from the absorbent pad in 5 mL methanol in presence of 100 ng of the following deuterated internal standards: morphine-d_3, codeine-d_3, 6-monoacetylmorphine-d_3, cocaine-d_3, benzoylecgonine-d_3, ecgonine methyl ester-d_3, Δ^9-tetrahydrocannabinol-d_3, buprenorphine-d_4, nordiazepam-d_5, oxazepam-d_5, amphetamine-d_5, methamphetamine-d_5, methylenedioxyamphetamine-d_5, methylenedioxyethylamphetamine-d_5, methylenedioxymethamphetamine-d_5, and SKF 525A. The tubes were shaken for 30 min on an orbital shaker at 200 rpm. Then the methanol solution was divided into 3 portions: 1 mL for buprenorphine testing, 1 mL for amphetamines testing, and the remainder for the other compounds. The methanol was evaporated to dryness, in the presence of 100 µL of isopropanol–HCl (99:1, v/v) in the case of amphetamines.

Buprenorphine was identified and quantitated by liquid chromatography (LC) coupled to ionspray mass spectrometry. A 2-µL portion of the reconstituted residue was injected on a Waters (Milford, MA) to Nova Pak C_{18} (150 × 2.0-mm i.d.) column. The mobile phase was acetonitrile–ammonium acetate buffer (2mM, pH 3.0) (80:20, v/v) with a flow rate of 200 µL/min and a postcolumn split of 1:4. Detection was performed on a Perkin-Elmer (Foster City, CA) Sciex API-100 mass analyzer.

Amphetamine and related compounds were identified after derivatization with 100 µL heptafluorobutyric anhydride (Sigma, St Louis, MO) and 50 µL ethyl acetate for 30 min at 70°C, and then analyzed by gas chromatography–mass spectrometry (GC–MS).

The other drugs were derivatized by silylation with 40 µL *N,O,*-bis(trimethylsilyl)trifluoroacetamide (BSTFA) plus 1% trimethylchlorosilane (Interchim, Montluçon, France) for 20 min at 60°C, and then analyzed by GC–MS.

A 1.5-µL portion of the derivatized extract was injected through the column (HP-5 MS capillary column, 30 m × 0.25-mm i.d.) of a Hewlett-Packard (les Ulis, France) 5890 GC coupled with an HP 5989B engine. Injector temperature was 260°C, and splitless injection was employed with a split-valve off-time of 0.75 min. The flow of helium through the column was 1 mL/min. The temperature column was programmed to rise from an initial temperature of 60°C held 1 min, to 290°C at 30°C/min and held at 290°C for the final 6 min. Benzodiazepines were detected using negative chemical ionization, whereas other drugs were detected by electron impact.

Analytes were identified and quantitated based upon comparison of the retention times and the relative abundance of three confirming ions with the deuterated internal standards. SKF 525A was used as internal standard for heroin.

Analytical parameters were determined in the previous studies (5,6). Standard curves were constructed by addition of drug analytes and deuterated standards to drug-free absorbent pad. The assays were linear in the range tested. The limits of detection for the target compounds were in the range from 0.01 to 2.0 ng/patch with a minimum signal-to-noise ratio of 3. The extraction recoveries were higher than 83% for all drugs. Within-run and between-run coefficients of variation were less than 16% for all drugs (Table I).

Urine samples were screened using fluorescent polarization immunoassay (FPIA) conducted on an Abbott ADx, and confirmation of the positives was performed with standard GC–MS procedures (5,7–9). To be considered positive, concentrations in sweat had to be higher than the lowest concentration tested in the linearity validation. In urine, specimens were considered positive using the positive cutoff proposed by the manufacturer.

Results and Discussion

Subjects wore the patch with minimal discomfort for five days. However, a few individuals developed a slight skin

irritation after exposure to the sun. It was possible for each subject to continue his or her normal hygiene practices. Nobody accidentally abraded the patch.

Although less common than urine collection, no one refused to wear the patch. It was generally necessary to explain the nature of the study before applying the patch, which was considered a curiosity by the subjects rather than a device for control.

Twenty subjects were recruited for this study. Sweat patches and urine specimens were tested for opiates (heroin, 6-monoacetylmorphine, morphine, codeine), cocaine (cocaine, benzoylecgonine, ecgonine methyl ester), cannabis (Δ^9-tetrahydrocannabinol), benzodiazepines (nordiazepam, oxazepam), amphetamines (amphetamine, methamphetamine, methylenedioxyamphetamine, methylenedioxymethamphetamine, methylenedioxyethylamphetamine), and buprenorphine. The analytical development of screening procedures allowed the simultaneous determination in sweat of several drugs of abuse, such as opiates, cocaine, cannabis, and benzodiazepines. During the study, no pharmaceutical other than buprenorphine was administered to the subjects. Results of the sweat patch analysis are presented in Table II. Patches were positive for opiates in 12 cases. Heroin and/or 6-monoacetylmorphine were identified in eight cases (67%), and codeine exposure was determined in four cases (33%). Heroin, when detected, was always present in lower concentrations than 6-monoacetylmorphine. In some cases, Cone et al. (2) reported higher heroin concentrations. The unique finding of heroin in sweat is of particular interest to document drug exposure of the subject. Its detection leads to a therapeutically useful change in the relationship between patients and clinical staff, particularly when the patient is informed of the result.

As is the case for hair testing (10), 6-monoacetylmorphine appears to be the major analyte in sweat after heroin intake. Therefore, care is necessary to prevent the conversion of 6-monoacetylmorphine to morphine. As heroin street samples generally contain codeine, this is also detected in sweat in cases of heroin abuse. Morphine is a metabolite of codeine and is detected in urine and blood when codeine is used. This is also the case when sweat is tested, and the amount of morphine is about 0–10% the amount of codeine, which appears consistent with a previous report (5).

Cocaine and its metabolites were detected in only one case, indicating that cocaine use by heroin addicts is infrequent in France. This is consistent with epidemiological studies (11). Cocaine was the major analyte excreted in sweat. Smaller amounts of ecgonine methyl ester and benzoylecgonine were present. Contrary to hair testing (7), ecgonine methyl ester was present in greater amounts than benzoylecgonine, which was also observed in a previous study (2).

Δ^9-Tetrahydrocannabinol, the active ingredient of cannabis, was detected in nine cases with concentrations ranging from 4 to 38 ng/patch and a mean value of 16 ng/patch.

As demonstrated previously (6), benzodiazepine concentrations are low in sweat. Excretion in sweat appears to be maximal with basic drugs that have high partition coefficients and pK_a

Table I. Analytical Parameters After Addition of Drug Analytes to Drug-Free Absorbent Pads

Drugs	Linearity (ng/patch)	Limit of detection (ng/patch)	Recovery (%)
Heroin	10–300 (r*= 0.995)	1.0	93.8
6-Acetylmorphine	10–5000 (r = 0.995)	0.5	89.9
Morphine	10–1000 (r = 0.996)	0.5	91.6
Codeine	10–500 (r = 0.997)	0.5	94.2
Cocaine	10–1000 (r = 0.998)	0.5	95.3
Benzoylecgonine	10–300 (r = 0.994)	1	88.2
Ecgonine methyl ester	10–300 (r = 0.993)	2.0	86.8
Tetrahydrocannabinol	2–50 (r = 0.997)	1.0	83.0
Amphetamine	10–500 (r = 0.999)	0.5	91.6
Methamphetamine	10–500 (r = 0.998)	0.5	89.3
MDA†	10–500 (r = 0.996)	0.5	89.9
MDMA	10–500 (r = 0.997)	0.5	90.6
MDEA	10–500 (r = 0.998)	0.5	90.7
Nordiazepam	0.5–50 (r = 0.997)	0.01	93.1
Oxazepam	0.5–50 (r = 0.994)	0.01	92.6
Buprenorphine	1–200 (r = 0.996)	0.2	85.4

*Correlation coefficient.
†Abbreviations: MDA, methylenedioxyamphetamine; MDMA, methylenedioxymethamphetamine; MDEA, methylenedioxyethylamphetamine.

Table II. Results of the Sweat Patch Analyses*

Subject	HER†	6-MAM	MOR	COD	THC	NOR	OXA	Miscellaneous
1	–	117	42	36	38	10	3	
2	157	1835	113	189	–	–	–	
3	–	–	40	4018	18	44	3	
4	–	–	–	–	–	–	–	
5	–	–	–	–	–	15	2	
6	–	–	–	–	–	–	–	
7	–	–	–	–	–	–	–	324 (COC), 58 (BZE), 89 (EME)
8	–	–	–	–	–	–	15	
9	87	958	81	56	–	–	–	
10	–	–	–	67	–	2	–	
11	–	–	–	–	27	–	–	
12	–	–	–	–	4	–	–	
13	–	328	145	10	14	–	–	
14	–	60	29	40	9	–	–	
15	175	2386	271	139	14	4	–	
16	37	108	165	85	–	–	–	
17	–	–	110	1812	–	–	–	
18	–	–	–	–	–	–	–	
19	–	–	21	206	6	–	–	121 (MDEA), 22 (MDA)
20	–	431	181	89	11	31	4	

*All the concentrations are in ng/patch.
†Abbreviations: HER, heroin; 6-MAM, 6-monoacetylmorphine; MOR, morphine; COD, codeine; COC, cocaine; BZE, benzoylecgonine; EME, ecgonine methyl ester; NOR, nordiazepam; OXA, oxazepam; THC, Δ^9-tetrahydrocannabinol; MDA, methylenedioxyamphetamine; MDEA, methylenedioxyethylamphetamine.

values close to the value of sweat pH, near 5.0 (12). The sweat patch is operating as an ion trap for the group of drugs that are weak bases and have pK_as around 8.0 (3). The presence of low nordiazepam concentrations is consistent with a pK_a of 3.5 for the drug. Oxazepam, which is more polar, will probably not diffuse across skin membranes as well as the parent drug does.

Finally, the identification of methylenedioxyethylamphetamine (121 ng/patch) and its metabolite, methylenedioxyamphetamine (22 ng/mg), represents the first report of excretion in sweat. Again, the parent drug, which was more apolar, was present in a greater amount than the metabolite.

Buprenorphine was administered under close medical supervision. Subjects were asked to take their medication in the presence of the nurses. The drug was analyzed using LC–ionspray mass spectrometry because of thermal degradation that occurs with GC–MS. Results of the sweat patch analyses, along with the daily dose, are presented in Table III. Four subjects, who were at the end of their therapy, were no longer taking buprenorphine. When administered, buprenorphine was excreted in sweat in the range 1.3–153.2 ng/patch. Norbuprenorphine was never detected, except in the single case of a high buprenorphine concentration (153.2 ng/patch). In this specimen, the norbuprenorphine concentration was 3.1 ng/patch. The correlation between daily dose and buprenorphine in sweat appears to be very poor, with a correlation coefficient (r) of 0.427. Therefore, sweat testing seems to be a qualitative test rather than a quantitative means to estimate the amount of drug ingested. There were substantial intersubject differences in the amount of buprenorphine detected in sweat for the same dose. This may be explained by the differences in sweat excretion rates between the individuals.

Table III. Concentrations of Buprenorphine in the Sweat Patches After Controlled Administration

Subject	Daily buprenorphine dose (mg)	Buprenorphine in sweat (ng/patch)
1	–	ND*
2	–	ND
3	–	ND
4	–	ND
5	0.4	7.5
6	0.8	8.5
7	0.8	5.6
8	1.6	3.5
9	2.0	4.1
10	2.4	5.8
11	2.4	1.3
12	2.4	14.9
13	2.4	33.2
14	3.0	31.6
15	3.2	153.2
16	3.2	26.8
17	3.2	10.4
18	3.2	58.1
19	3.2	21.3
20	6.0	47.9

* ND = not detected.

Urine specimens were collected at the beginning and at the end of patch wear. Urinalysis was performed by GC–MS to identify the target compounds. When a patch tested positive for heroin exposure (presence of heroin or 6-monoacetylmorphine), 6-monoacetylmorphine was always found in one or both corresponding urine samples. Similar findings were observed in the case of codeine exposure where only morphine and codeine were simultaneously identified in urine and sweat. Therefore, sweat can be accurately used to differentiate heroin and codeine abuse based on the presence of heroin or 6-monoacetylmorphine. Testing sweat for heroin or 6-monoacetylmorphine leads the toxicologist to use GC–MS and not immunoassay, although the latter procedure can find applications in drug screening (13,14). For the other drugs, urine findings and sweat findings were also in agreement. No subject was urine positive and sweat negative. Also, no subject was sweat positive and urine (based on the sum of the two specimens) negative. However, if only one urine test were performed, it would have been possible to observe sweat positive and urine negative subjects in six cases. These cases involved opiate exposure in three subjects who had used heroin, two subjects who had used codeine, and the only subject positive for cocaine. Sweat tested positive for opiates and cocaine in 13 cases. The first specimens of urine, collected on day 0, tested positive for opiates in 10 cases. The second specimens of urine, collected on day 5, tested positive in nine cases for opiates and in one case for cocaine. Sweat testing was able to show drug exposure after urine collection (the first urine specimen) and after the drug had been eliminated from the body (the second urine specimen) and total metabolization had occurred. Therefore, the sweat patch appears to be more effective than urine in detecting the use of opiates and cocaine. The same observations were presented by Fogerson et al. (13). Sweat testing appears to be as effective as two urine tests performed each week to detect a single drug exposure. These findings can be observed with drugs having urinary detection times of 2–3 days, depending on immunoassay cutoffs. Compounds with a longer elimination half-life, such as benzodiazepines or cannabinoids, were positive in all urine samples. In such cases, one would observe a positive sweat and a negative first urine specimen only when the drug is taken for the first time during the time of patch application.

Although still early in development, testing individuals for illicit drugs with sweat patches increases the window of drug detection to one week. As proposed by Cone et al. (2), this period can be increased to several weeks. The test appears to be very sensitive as the administration of low doses of cocaine (approximately 1–5 mg) produces detectable amounts in sweat (2).

This new technology is particularly suitable in rehabilitation cases when it is important for medical personnel to get information on the behavior of the patients. Patches can be worn continuously and constitute a record of drug intake during that period. However, sweat patches are not suitable for rapid screening purposes when urine, which is immediately available, appears to be the specimen of choice. For example, driving under the influence of a drug cannot be characterized using sweat as it is necessary to wear the patch for at least 2 h to test positive (5,6).

The introduction of new testing procedures, such as the sweat patch, has changed the behavior of some patients. They often spontaneously talk about their illicit drug use and can be challenged to reduce their consumption, which can be detected by the results of the next test. Sweat testing appears to offer the advantage of being a relatively noninvasive means of obtaining a cumulative estimate of drug exposure over a long period, whereas urine is rather an incremental measure with high invasiveness.

Conclusion

In conclusion, sweat analysis may be a useful adjunct to conventional drug testing. Specimens of sweat can be more easily obtained and with less embarrassment than urine specimens. As analytical procedures generally involve GC–MS to document heroin exposure, the routine analysis of sweat is not accessible to most laboratories, but from a clinical point of view, the generated data are extremely helpful. This new technology may find useful applications in the treatment and monitoring of substance abusers.

References

1. H. Tachan. Uber den Ubergang von Arzneimitteln in den Schweiss. *Arch. Exp. Phatol. Pharmakol.* **66:** 224–46 (1911).
2. E.J. Cone, M.J. Hillsgrove, A.J. Jenkins, R.M. Keenan, and W.D. Darwin. Sweat testing for heroin, cocaine, and metabolites. *J. Anal. Toxicol.* **18:** 298–305 (1994).
3. M. Burns and R.C. Baselt. Monitoring drug use with a sweat patch: An experiment with cocaine. *J. Anal. Toxicol.* **19:** 41–48 (1995).
4. V. Spiehler, J. Fay, R. Fogerson, D. Schoendorfer, and S. Niedbala. Enzyme immunoassay validation for qualitative detection of cocaine in sweat. *Clin. Chem.* **42:** 34–38 (1996).
5. P. Kintz, A. Tracqui, C. Jamey, and P. Mangin. Detection of codeine and phenobarbital in sweat collected with a sweat patch. *J. Anal. Toxicol.* **20:** 197–201 (1996).
6. P. Kintz, A. Tracqui, and P. Mangin. Sweat testing for benzodiazepines. *J. Forensic Sci.* **41:** 851–54 (1996).
7. P. Kintz, V. Cirimele, C. Sengler, and P. Mangin. Testing human hair and urine for anhydroecgonine methyl ester, a pyrolysis product of cocaine. *J. Anal. Toxicol.* **19:** 479–82 (1995).
8. P. Kintz, D. Machart, C. Jamey, and P. Mangin. Comparison between GC–MS and the EMIT II, Abbott ADx, and Roche OnLine immunoassays for the determination of THCCOOH. *J. Anal. Toxicol.* **19:** 304–306 (1995).
9. A. Tracqui, P. Kintz, and P. Mangin. HPLC/MS determination of buprenorphine and norbuprenorphine in biological fluids and hair samples. *J. Forensic Sci.* In press.
10. P. Kintz and P. Mangin. What constitutes a positive result in hair analysis: Proposal for the establishment of cut-off values. *Forensic Sci. Int.* **70:** 3–11 (1995).
11. F. Facy and F. Laurent. La toxicomanie en France. Quelques données épidémiologiques. *Revue Prat.* **277:** 67–73 (1994).
12. H.L. Johnson and H.I. Maibach. Drug excretion in human eccrine sweat. *J. Invest. Dermatol.* **56:** 182–88 (1971).
13. R. Fogerson, J. Suttliff, and C. Walker. Testing for cocaine and opiates with the PharmChek™ sweat patch. In *Society of Forensic Toxicologists, Inc. Abstracts,* Baltimore, MD, October 9–14, 1995, p 27.
14. V. Spiehler, J. Fay, R. Fogerson, D. Schoendorfer, and R.S. Niedbala. Enzyme immunoassay validation for qualitative detection of cocaine in sweat. *Clin. Chem.* **42:** 34–38 (1996).

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