

DRUGS OF ABUSE: ON-SITE SALIVA SAMPLING AND ANALYSIS

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ABSTRACT: A rapid way to prove the impairment of drivers is saliva analysis. We investigated a new collection method combined with an immunochromatographic teststrip. The results indicate that D,L-amphetaminesulfate can be detected at concentrations up to 500 ng/ml in whole saliva.

KEY WORDS: Saliva; Drugs of abuse; DUI.

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INTRODUCTION

Since driving under the influence of drugs is more and more recognized as a problem for traffic safety over the last few years police is searching for a suitable rapid on-site test [1, 2]. In contrast to alcohol there are only a few tests available at to prove the use of illicit drugs on the roadside routinely. For this saliva is in the focus of interest [3]. Saliva is the only body fluid which can be obtained noninvasively, rapidly and which gives information about the level of impairment at the time of testing due to its relationship to blood levels. We report on a new sampling system for the collection of saliva and the direct analysis with an immunochromatographic teststrip.

METHODS AND MATERIALS

A lateral flow immunchromatographic teststrip based on a competitive detection system was tuned for analyzing amphetamines in saliva. A 20 µm nitrocellulose membrane was used. To detect antibody binding we used gold labeled detection antibody. During the tuning we used aqueous solutions of DL-amphetaminesulfate (Sigma Inc.) to compare sensitivity. A negative test should have both lines, e.g. detection and control, with equal intensity. A positive test should have no visable intensity of the detection line.

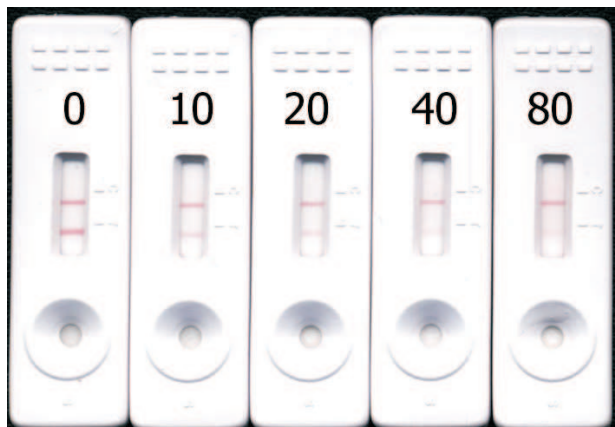


Fig. 1. Teststrip with aqueous solution (units in ng/ml).

The testsystem consists of this strip in combination with a wiping pin which is placed on the sample pad of the strip. Both is held in a housing (see Figure 2). After saliva is collected in the mouth buffer solution is added on the pin. The saliva is washed of the pin onto the teststrip. The corresponding result can be read out within minutes after adding the buffer.

Different wiping pins and procedures were evaluated for collecting saliva. We in-

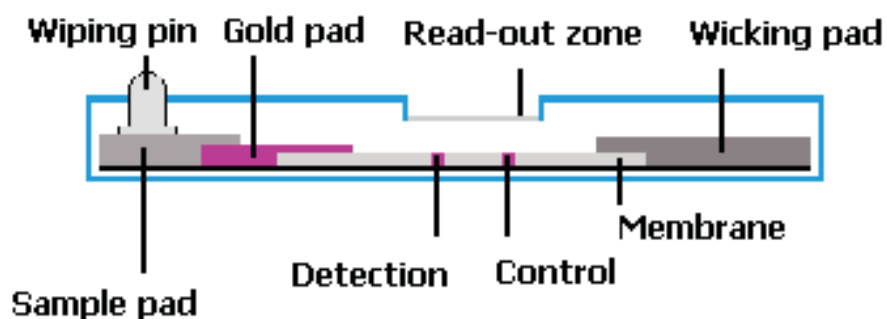


Fig. 2. Immunochromatographic teststrip.

vestigated wiping the tongue 1–5 times or for different periods of time. The collected amount of saliva was measured.

In a typical experiment we spiked whole saliva from different subjects with a concentrated D,L-amphetamine sulphate solution. We used concentrations also found in vivo (300–2000 ng/ml D,L-amphetamine sulphate) [5]. This spiked saliva was put on the pin and washed on the strip with a certain volume of buffer solution and the test was utilized after 2 min.

To make the test more sensitive we changed the preparation of the strips. The detection line was made more intense than the control line and we proposed a positive test if the detection line was less intense than the control line.

RESULTS AND DISCUSSION

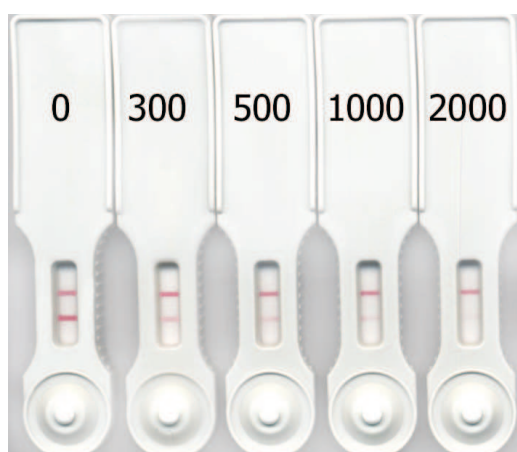


Fig. 3. Teststrip with 10 μ l of spiked saliva and buffer solution.

After tuning the test a cut-off of 80 ng/ml D,L-amphetamine sulphate was found with aqueous solutions. If the tongue is wiped for 10 sec with a wiping pin the results indicate that a reliable amount of saliva is collected, e.g. 7–10 μ l. If the tongue is wiped 1–5 times the volume of saliva is not sufficient. Executing the test with spiked saliva from different subjects we found great variations of sensitivity. With the changed proposals the results don't spread as much as before. It is possible to reliably detect concentrations of up to 500 ng/ml D,L-amphetamine sulphate.

Our results show the general suitability of testing saliva with a wiping procedure. We found changes in sensitivity interindividually and also with different samples from the same subject, which corresponds with other findings [4]. In the described procedure, the volume of saliva which is collected is not known for each test. Therefore the collection has to be standardized (for example with a sampling system which collects a defined volume of saliva).

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