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Cocaine testing in fitness-to-drive assessments: comparison between hair analysis, urinalysis and self-reports

Test na obecność kokainy do oceny zdolności do prowadzenia pojazdów: porównanie między analizą włosów, badaniem moczu i samoceną kierowcy

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Abstract

Introduction: Cocaine is one of the most commonly used illicit substance and may have a devastating impact on road safety. The detection of cocaine use is therefore of great significance in assessing fitness-to-drive. The aim of this study is to compare anamnestic declaration of cocaine use with the results of immunoassays in urine and the results of hair analysis in the fitness-to-drive assessment procedure used in Switzerland. We hypothesised that hair analysis will detect a higher number of clandestine cocaine users than urinalysis.

Material and methods: Anamnestic declarations as well as the results of urinalysis and hair analysis were collected from fitness-to-drive reports. Urinalysis was performed with an immunoassay. Hair samples were analysed using liquid chromatography coupled with tandem mass spectrometry (LC-MS-MS).

Streszczenie

Wprowadzenie: Kokaina jest jedną z najczęściej używanych nielegalnych substancji psychoaktywnych i może wpływać destrukcyjnie na bezpieczeństwo w ruchu drogowym. Dlatego przy ocenie zdolności do prowadzenia pojazdów mechanicznych bardzo ważne jest uwzględnianie używania kokainy. Celem badania jest porównanie deklaracji kierowcy co do używania kokainy z wynikami immunologicznych badań moczu i analizy włosów przy ocenie zdolności do prowadzenia pojazdów w Szwajcarii. Stawiamy hipotezę, że analiza włosów pozwala wykryć większą liczbę użytkowników kokainy niż analiza moczu.

Materiał i metody: Z raportów dotyczących oceny zdolności do prowadzenia pojazdów uzyskano zarówno deklaracje kierowców, jak i wyniki analizy moczu i włosów na obecność narkotyków. Analizę moczu wykonywano, posługując się testem immunologicznym. Próbki włosów analizowano za pomocą

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Results: Of a total population of 311 subjects (mean age 32.5 years), 38 admitted consumption of cocaine, in 7 cases the urine immunoassay was positive and 84 subjects tested positive for cocaine in hair analysis. The detection rate in hair analysis for cocaine was 12 times greater than that for urine testing and 1.4 times greater than that for anamnestic declaration. Apart from cocaine, the investigated population consumed cannabis (21 of 81 subjects), amphetamines (30 of 81 subjects) as well as, more rarely, opiates and methadone (included substitution treatment).

Conclusions: The study results reveal that hair analysis is more effective than urinalysis and self-reports in identifying cocaine users in a fitness-to-drive assessment. Hair analysis provides long-term information about cocaine use and is therefore a useful tool for the identification and follow-up of cocaine users. Hair analysis should be included in every assessment of fitness-to-drive in subjects suspected of cocaine abuse.

Key words: Cocaine, Hair analysis, Urinalysis, Urine testing, Fitness-to-drive, Driving under influence, Self-report

chromatografii cieczowej sprzężonej ze spektrometrią masową typu tandem (LC-MS/MS).

Wyniki: Z ogólnej liczby 311 badanych (średni wiek 32,5) 38 osób potwierdziło konsumpcję kokainy. W przypadku 7 osób wynik analizy moczu był pozytywny, podczas gdy w analizie włosów pozytywny wynik uzyskano u 84 osób. Stwierdzono, że analiza włosów jest 12 razy bardziej efektywna w wykrywaniu kokainy niż analiza moczu i 1,4 razy bardziej wiarygodna niż deklaracje kierowców. Poza kokainą badani używali konopi indyjskich (21 osób na 81), amfetaminy (30 na 81), zdecydowanie rzadziej – opiatów i metadonu (włączając w to leczenie substytucyjne metadonem).

Wnioski: Wyniki badań wykazały, że w identyfikacji użytkowników kokainy przy ocenie zdolności do kierowania pojazdami analiza włosów jest bardziej efektywna niż analiza moczu i deklaracje kierowców. Analiza włosów dostarcza „długoterminowej" informacji dotyczącej używania kokainy w okresie kilku miesięcy wstecz i w związku z tym jest przydatnym narzędziem do monitorowania użytkowników kokainy. Analiza ta powinna być zawsze wykorzystywana do oceny zdolności do prowadzenia pojazdów w przypadku osób podejrzewanych o nadużywanie kokainy.

Słowa kluczowe: kokaina, analiza włosów, analiza moczu, badanie moczu, zdolność do prowadzenia pojazdu, prowadzenie pojazdu pod wpływem, samoocena kierowcy

Introduction

Illegal drug use is a global problem: The World Health Organization (WHO) estimates that in 2012 between 162 and 324 million people (3.5-7.0% of the world's population aged 15-64) have used drugs at least once in the previous twelve months. Cannabis is the most commonly used substance in Europe, with an annual prevalence of 4.3% of those aged 15-64, followed by cocaine (0.7%), amphetamine-type stimulants and opioids (mostly heroin) [1].

Even in a small European country such as Switzerland with approximately 8.2 million inhabitants, the number of cocaine users in 2010 was estimated about 25,445 to 44,275 persons, who consumed around 3,770 to 7,540 kg cocaine (sales volume 369-520 million Swiss francs or US-$, respectively) [2]. Cocaine, chemically an alkaloid from the tropane group, is obtained mainly in South America from the leaves of the coca plant (bot. *Erythroxylum coca* Lam.) [3, 4]. It has a stimulating, activity-increasing and euphoric effect. Cocaine users can experience depression when the effects wear off as well as psychotic episodes. Cocaine affects the cardiopulmonary system by increasing respiratory and heart rate as well as blood pressure, and has a vasoconstrictive effect [5]. It is also known to have a local anaesthetic effect [6, 7].

The consumption of psychoactive substances is one of the major factors affecting driving performance leading to accidents [8-10]. In a study, the odds of being involved in a collision was 2.11 for cocaine users compared to non-users [11].

The "classical" method for detecting a drug use is immunoassay-based tests in urine or blood [12, 13]. For several years, hair analysis is used with increasing frequency for detecting the use of illicit drugs (and other substances) in different settings such as forensic populations, occupational screening and fitness-to-drive assessments [14-26]. Hair analysis and urinalysis have their specific advantages and limitations. Urinalysis is a simply performed and rather low-price method. The most important limitations are false-positive results due to immunological cross-reactions and the possible adulteration of the urine specimen, e.g. by dilution or substitution [27, 28]. In contrast, immunological cross-reactions do not exist in hair analysis performed by evidentiary methods such as liquid chromatography or gas chromatography coupled with mass spectrometry. However, the results may be difficult to interpret in the case of a possible external contamination [29, 30]. Furthermore, hair analysis can be manipulated by some hair cosmetics, e.g. bleaching. In most cases, such a treatment can easily be verified when taking a hair sample. There are products that promise to reduce drug concentration in hair, but these products seem to have only a slight effect: in a study, a decrease in cocaine concentration of 5% after the use of a "cleaning shampoo" was observed [31].

In Switzerland, a driver's licence authority is obliged by law to commission a fitness-to-drive assessment:

a) if drugs with a high addictive potential are found with a driver (drug consumption not needed) or
b) in case of driving under influence (DUI) of a drug [32].

These assessments were performed by specialised physicians certified by the Swiss Society of Legal Medicine ("specialist in traffic medicine") [33]. The key task of such an assessment is the objective clarification of the drug consumption pattern of the investigated subject, preferably over a period of several months. If the use of an illicit drug can be proven, an abstinence period of 6-12 months is demanded from the driver to re-grant the licence.

Before the introduction of hair analysis, only urinalysis were performed in fitness-to-drive assessments in Switzerland. Nowadays, the Swiss Society of Legal Medicine recommends for fitness-to-drive examinations urinalysis for assessing cannabis consumption and hair analysis for the use of other drugs as opiates, cocaine and amphetamines [34]. It is assumed that hair analysis will discover more clandestine cocaine users than urinalysis because of the longer time window of detection, but there are no published studies investigating if the use of hair analysis instead of urinalysis has objective advantages in fitness-to-drive assessments as carried out in Switzerland.

The aim of this study was to compare self-reports and the results of urinalysis and hair analysis for the detection of cocaine consumption in the fitness-to-drive assessment procedure used in Switzerland. We hypothesised that hair analysis will detect a higher number of clandestine cocaine users than urinalysis.

Material and methods

Study population

All medical records from individuals with a history or suspicion of drug use examined between 2010 and 2012 in the Department of Traffic Sciences of the Institute of Forensic Medicine, University of Bern, were retrospectively analysed. Further inclusion criteria were complete results of hair analysis and urine rapid tests, no cosmetic hair treatment as well full anamnestic information. The reports were drawn up in the context of a fitness-to-drive assessment at the request of a driving licence authority. The competent Ethics Committee of the Canton of Berne explained that, for retrospective data analyses, as in this case, no permission must be obtained from the subject (Decision KEK-No. Z048/13). The University privacy policy has been observed.

Anamnestic information

As part of the assessment, physicians collected anamnestic declarations concerning drug use. The physicians were all trained by one of the authors (MP) for minimalising interviewer bias and the anamnestic assessment was based on the recommendations of the Swiss Society of Legal Medicine referring to this matter [34]. The complete history of drug consumption was evaluated though the focus was on the time period overviewed by hair analysis. The assessments took one to two hours to complete (including physical examination and collection of hair and urine). Furthermore, information about the use of alcohol,

medication, personal medical history, and possible cosmetic hair treatments were collected.

Urinalysis

Each person gave a urine sample during the course of the assessment. The urine was collected in a disposable cup with no additives and analysed immediately after delivery by trained personnel. To prevent urine manipulation, an inspection and a temperature measurement was carried out; urine samples with a temperature of < 32°C and with sensory abnormalities (colour, smell, foam formation, flocculation, crystalline sediment and the like), nitrite > 500 mg/L and pH < 3 or > 11 were considered to be manipulated and not used.

The urinalysis was performed with a Drug Screen Multi-12A®-Test Panel according to the manufacturer's guidelines (Nal von Minden GmbH, 47441 Moers, Germany). The performing person carried out the reading of the tests visually. In this test, there is a panel of twelve single sandwich immune assays for opiates, methadone, cocaine, amphetamine, methamphetamine, 3,4-methylenedioxy-methamphetamine (MDMA, ecstasy), cannabinoids, benzodiazepines, barbiturates, buprenorphine, tramadol and tricyclic antidepressants. The immunoassay for cocaine has a cut-off value of 300 ng/ml (calibrator benzoylecgonine).

Analysis of hair samples

At least two hair samples were taken from each subject. They were taken from the vertex or the occiput of the head. If the head hair was too short or the test subject refused consent to provide a sample of head hair, body hair from the legs, arms or chest was shaved. If consent was provided to take a sample of scalp hair, a lock as thick as a pencil was tied with a string and then cut as close to the skin as possible using a scissor. The length of the scalp stubble was measured. Head or body hair was protected from light in wrapped aluminium foil and immediately sent to the analysing laboratory.

Hair samples were washed with three solvents (water, acetone, diethylether, 4 mL each) prior to cutting into approx. 1-3 mm pieces with scissors. 30 mg of the homogenised material was extracted by adding 1 mL methanol in a 2 mL micro-vial and treatment in an ultrasonication bath for two hours. Extracts were evaporated to dryness under a nitrogen-stream, and re-dissolved in 100 µL of HPLC-eluents A/B 95/5 (v/v). The LC-MS/MS system consisted of a binary gradient HPLC 1100 (Agilent) and a Qtrap 3200 hybrid triple-quadrupole ion trap tandem-mass spectrometer (Sciex). Solvent A was 0.1% formic acid/water and solvent B was 0.1% formic acid in acetonitrile. Gradient elution was performed from 5% solvent B linear to 95% B in 7 minutes with a flow rate of 0.4 mL/min. Separation was performed with a Luna PFP reversed phase column 150 x 2.0 mm, 5 µm (Phenomenex). Two fragment-ions of each compound (cocaine, benzoylecgonine, ecgonine methylester and cocaethylene) were monitored, and one transition of each of their deuterated standards – which had been added to the methanol prior to extraction. Limits of quantitation cocaine and metabolites were 100 pg/mg, however, for cocaine a threshold was set to 500 pg/mg according to SOHT guidelines to avoid detection of hair contamination [35]. Besides cocaine and its metabolites, the following substances were included in the same procedure: amphetamine, methamphetamine, MDMA, MDEA, MDE, morphine, 6-monoacetyl-morphine, codeine, methadone, EDDP, tramadol – with their deuterated standards. Furthermore, fragment-ions of heroine, LSD, modafinil, zolpidem and methylphenidate were monitored as well, without any deuterated analogues included in the analysis.

In this study, the hair sample was counted as positive for cocaine when cocaine, benzoylecgonine, ecgonine methyl ester and/or cocaethylene were detected in the hair. In segmented hair samples, only the result of segment close to the head was included.

Statistical analysis

The data was analysed using the SPSS Statistics 20 program for Windows (SPSS Inc., Chicago, USA). The results shown in Tables I-III and Figure 1 were obtained by using descriptive data analysis. The comparison of the nominal-scaled variable «sex» was performed using the χ^2 test. The interval-scaled variables (not normally distributed, tested using the Kolmogorov-Smirnov-test) were compared by using the Mann-Whitney-test. The level for statistical significance was set to $p < 0.05$.

■ Results

The study was performed in a population of 311 subjects, 284 men and 27 women aged 18-62 years

Table I. Characteristics of the sample

Factors	All subjects (*n* = 311)	Subjects negative for cocaine in hair analysis (*n* = 227)	Subjects positive for cocaine in hair analysis (*n* = 84)	Significance*
Gender, *n* (%)				
Male	284 (91.3)	205 (90.3)	79 (94)	NS
Female	27 (8.7)	22 (9.7)	5 (6)	
Age in years, mean (SD)	32.5 (8.8)	32.5 (8.8)	32.61 (8.9)	NS
DUI of illicit drugs offences, mean (SD)	0.52 (0.7)	0.52 (0.7)	0.52 (0.7)	NS
DUI of alcohol offences, mean (SD)	0.46 (0.8)	0.45 (0.9)	0.49 (0.8)	NS
DUI of illicit drugs and alcohol offences, mean (SD)	0.22 (0.4)	0.24 (0.4)	0.15 (0.4)	NS
Other road traffic act offences, mean (SD)	1.27 (2.0)	1.22 (1.8)	1.39 (2.4)	NS
Type of hair, *n* (%)				Not performed
Scalp	226 (73)	178 (78.4)	48 (57.1)	
Legs	66 (21)	38 (16.7)	28 (33.3)	
Chest	14 (4)	8 (3.5)	6 (7.1)	
Arms	5 (2)	3 (1.3)	2 (2.4)	
Time frame of detection in hair analysis in months, mean (SD)	4.3 (1.5)	4.46 (1.5)	4.0 (1.5)	$p = 0.002$

**Negative vs. positive for cocaine in hair analysis*
DUI – driving under influence, NS – non-significant, SD – standard deviation

Table II. Comparison of the prevalence for drug consumption: hair analysis versus urinalysis and self-reports

	Hair analysis	
	Positive for cocaine (*n*)	Negative for cocaine (*n*)
Self-reports		
Positive for cocaine (*n*)	30	8
Negative for cocaine (*n*)	54	219
Urine test		
Positive for cocaine (*n*)	7	0
Negative for cocaine (*n*)	77	227

Table III. Comparison of the prevalence for drug consumption: urinalysis versus self-reports

	Urine test	
	Positive for cocaine (*n*)	Negative for cocaine (*n*)
Self-reports		
Positive for cocaine (*n*)	7	31
Negative for cocaine (*n*)	0	273

(mean age 32.5 years). Characteristics of the study population can be found in Table I.

Head hair specimens were obtained from 226 subjects and non-head hair specimens from 85 subjects (66 subjects provided leg hair, 14 chest hair and 5 arm hair). The mean time frame of detection by hair analysis in all subjects was 4.3 months (range 1-12 months). The group that tested positive for cocaine use had a statistically significant shorter time frame of detection (Table I).

38 subjects admitted the use of cocaine. Of these, 7 showed also a positive result in urinalysis and 30 tested positive in hair analysis (Table II and III).

Of the 273 subjects who denied cocaine consumption, no one tested positive in urinalysis, but in 54 cases a positive result was obtained in hair analysis (Table II and III).

All subjects with a positive urinalysis tested positive in hair analysis (Table II). In 8 cases hair

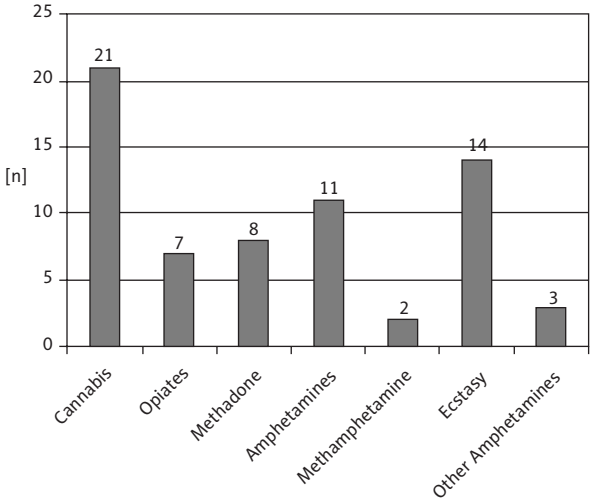

Figure 1. Number of positive tested hair samples for all analysed drugs (for cannabis based on self-reports)

analysis was negative for cocaine despite of anamnestic declaration of use.

There were no statistically significant differences in gender, age and investigated traffic offences (driving under influence of drugs and/or alcohol, speeding, other traffic offences) between the groups with a negative and a positive result for cocaine in hair analysis.

Other substances were found in the hair analysis that was positive for cocaine; the most common of these were amphetamine-type stimulants drugs including amphetamine, methamphetamine and MDMA. Cannabis was not analysed in the hair but 21 subjects with positive hair analysis for cocaine admitted also the consumption of cannabis (Figure 1).

■ Discussion

The use of cocaine, one of the most commonly used illicit substances, can have a devastating impact on road safety [8-11]. The detection of cocaine use is therefore of great significance in traffic medicine.

Several studies described the advantage of hair analysis for fitness-to-drive purpose [14-18]. One study presented only case reports [16], the others compared hair analysis and urinalysis [14, 15, 18] or highlighted the experience with hair analysis [17]. This study is the first comparing self-reports with the results of hair analysis and urinalysis in a Swiss fitness-to-drive assessment setting.

The present study shows that the detection rate of cocaine using hair analysis is approximately 12 times higher than urine immunoassays. However, this result can be attributed not only to purely analytical reasons. In our study, the subjects know approximately 1-2 month in advance the examination date and may thus (temporarily) end their cocaine consumption. This factor is certainly also reflected in the low number of cocaine-positive urine tests compared with the results of the hair analysis and the anamnestic data: 38 subjects admitted a cocaine use, but only 7 of them had a positive result for cocaine in the urine test. Immunoassays test is positive only within a window of a few days following cocaine consumption [36]. Furthermore, for assessing fitness-to-drive in relation to a drug abuse, the consumer behaviour over a longer period of time is of importance and not only a "snap-shot" of a few days. And for this purpose, hair analysis is an appropriate method.

In this study, the superiority of hair analysis compared with urinalysis was almost at the same degree as that described in the study of Martinez *et al.* for a male population living on the Arizona-Mexico border [37]. In comparison to this study, Dufaux *et al.* [14] and Mieczkowski *et al.* [38] described smaller positivity rates in hair for cocaine than in urine (3.8-fold and 2.3-fold respectively). That can be explained with the different circumstances of testing. In an abstinence programme, participants are interested in living drug-free for re-granting their driving licence. Quite the contrary, in a fitness-to-drive assessment, subjects try to hide their drug consumption to avoid driving licence withdrawal. Referring to the study of Mieczkowski *et al.*, it can be supposed that there is a high proportion of regular drug users in an arrestee population and so, the correlation between results of hair analysis and urinalysis is higher than in our study population.

A further advantage of hair analysis is the uncomplicated collection of the sample material. A urine collection under surveillance can be an unpleasant and embarrassing situation for the test subject. Moreover, there are almost no known manipulation possibilities for hair analysis. A hair cosmetic treatment (bleaching or colouring) which can influence the detection of cocaine can be recognised most of the time while harvesting the sample [39, 40]. For urine rapid tests, however, a variety of possibilities for manipulation have been described in the literature [41, 42].

One advantage of the urine rapid test is the readily available result if using a "bedside" test panel as in this study. The subject can be confronted directly upon a positive finding, a motivation to give correct or additional information on the consumer behaviour. But immunoassays such as the panel used by us are always fraught with the risk of false-positive results and never to be regarded as conclusive due to the mechanism of action (antigen-antibody reaction). Immunoassays for cocaine show only a few cross reactions as described in the literature [43, 44]. Using hair analysis the drug that is targeted, as well as its metabolites, is detected directly by confirmatory analysis (chromatographic methods coupled to mass spectrometry). In this study, the target was cocaine, benzoylecgonine and ecgoninemethylester. The risk of cross-reaction is thus not present.

The relatively small discrepancy between cocaine evidence in hair and the anamnestic data (1.4 times) in this study can be explained by the fact that cocaine consumption had already been proven in connection with previous driving under influence of drugs offences in the most of the subjects, facts that are well known to the experts. Thus, the subjects had little reason to conceal consumption (at least in the past). Musshoff *et al.* have also noticed this observation in a population taking part in an official methadone maintenance programme [22].

Cocaine was demonstrated in 54 of 273 subjects, who denied consumption of this substance. This fact shows that the hair analysis for cocaine is also useful for monitoring cocaine abstinence. This result is also confirmed in other studies, in which a cocaine use was dramatically underreported [21, 45-50].

In 8 cases, cocaine was not detected in the hair analysis despite the subject's admission of consumption. This can be explained by consumption outside the examined period of time or by consumption in very low levels, therefore cocaine and its metabolites did not exceed the limit of detection in hair analysis.

Cocaine is often not the only substance that was consumed. Most frequently, cocaine users consumed additionally cannabis (21 of 84 subjects) and amphetamines (30 of 84 subjects), rare opiates and methadone (included substitution treatment). For cannabis, the anamnestic data was used, since THC and metabolites were not analysed by hair analysis in this study. These results reflect well the most commonly consumed drugs in Switzerland (cannabis, cocaine, amphetamines) [51]. A driving capacity assessment should not search for an isolated drug. Due to the high frequency of consumption of several substances, the screening should be carried out additionally for, as an example, opiates, cocaine, amphetamines and cannabis. The hair analysis is ideal for this.

The subject population showed a clear predominance of men. This is consistent with the data in the literature describing a higher frequency of cocaine use among men [51], and is also evidence that the present study population is comparable to the general population.

It is interesting that almost every second subject committed a driving under influence of alcohol offence and that every fourth subject was known by the authorities for driving under combined influence of drugs and alcohol. Furthermore, every subject has committed on average 1.27 other non-substance related traffic offences (crimes that lead to a warning or a driver's licence revocation, no violations which are punishable only by a fine). Educational interventions and campaigns should focus on the issue of driving under influence of drugs as well as on other traffic related hazardous behaviours (alcohol consumption, speeding etc.).

Our results show that the hypothesis of the study was correct and that hair analysis is the method of choice if drug consumption behaviour has to be controlled over a longer period as in fitness-to-drive assessments or drug rehabilitation programmes. Urine testing for drugs should be used if a short-term use is in question as in cases of suspected drug intoxication or in cases of supposed driving under influence. If drug testing is planned, the following questions should always be answered: Which time frame of detection is required? Urine testing by immune assays for cocaine can only overview a period of 2 to 3 days whereas hair analysis covers a period of several months, depending on the length of the collected hair.

The study has some limitations. It was conducted in a single centre and the results are only applicable for the studied population. Also the number of study subjects is rather low. The positive results of immunoassays in urine were not verified with confirmatory methods; but the validation of immunoassays was not aim of this study.

Conclusions

Hair analysis is more effective than urinalysis and self-reports in identifying cocaine users in a fitness-

to-drive assessment setting. In this study, the detection rate of cocaine use with hair analysis is approximately 12-fold that of urinalysis. Hair analysis should be included in every fitness-to-drive assessment in subjects suspected of cocaine use. Due to the frequent use of multiple substances, there should always be a screening of the usual drugs as part of a fitness-to-drive assessment. The indication for hair analysis or urine testing should always be dependent on the question of the required detection time frame.

Conflict of interest/Konflikt interesów

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None declared./Nie występuje.

Ethics/Etyka

The work described in this article has been carried out in accordance with the Code of Ethics of the World Medical Association (Declaration of Helsinki) on medical research involving human subjects, EU Directive (210/63/EU) on protection of animals used for scientific purposes, Uniform Requirements for manuscripts submitted to biomedical journals and the ethical principles defined in the Farmington Consensus of 1997.

Treści przedstawione w pracy są zgodne z zasadami Deklaracji Helsińskiej odnoszącymi się do badań z udziałem ludzi, dyrektywami EU dotyczącymi ochrony zwierząt używanych do celów naukowych, ujednoliconymi wymaganiami dla czasopism biomedycznych oraz z zasadami etycznymi określonymi w Porozumieniu z Farmington w 1997 roku.

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