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Evaluation of Abuse Drugs and Clinical Laboratory Tests Variations in Whole Blood & Urine Samples of Abusers

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Abstract: In this study, five hundred urine samples and five hundred blood samples of abusers were examined for the presence of alkaloid substances and abuse drugs in urines and bloods. These numbers of blood and urine samples of addicts in clinics of welfare organization, during detoxification treatment or maintenance treatment were screened for abuse drugs presence. The all of samples were tested through as a view of clinical laboratory methods. Age ranges of female patients were 35 ± 15 and age range of males patients were 45 ± 15. All patients filled questionnaire and satisfy forms too. First, all fresh urine and blood samples were examined to confirm presence drugs abuses, depend on their addiction and treatment, so all samples were confirmed by two tests. Then they were examined to other clinical laboratory tests. All data were analyzed by t-test and were Anova one way and two ways of Anova Turkey, and $p < 0.05$ was considered significant. The p-value of this study was $p = 0.0001$. The results of this study were showed that 4% of abusers had mild increase in hematocrite level and 2% of narcotic drugs abusers had mild lower level of blood sugars than normal range and 4% of participants had increase liver enzymes such ALT (alanine transferase), AST (aspartat transferase), ALP (alkaline phosphatease) and 1% of them had renal failure. Although blood level BUN (blood urea nitrogen) and creatinin were examined to evaluation of their renal failure .The results in Tabriz/Iran undrevision of welfare organization clinics were approximately showed that positive results of addiction are in each of urine and blood samples. Because some of abusers directly consumed full long time agonist or partial agonists' drugs such as methadone and buprenorphine for their maintenance therapy in clinics. Also doing test on blood samples has high importance in distinguishing and confirmation of drugs abuse in samples. Also in this study we conclude that among all drug analytical methods the cheapest and easiest test to screening opioids and other abuse drugs in urine and blood samples is strip test for rapid diagnosis, also tests on blood samples have high importance as a view point of accuracy to distinguishing of drugs abuse, and serum levels of some other parameters showed all abusers patients situation such as liver and renal dysfunction through clinical laboratory tests.

Key words: Abusers, urine and whole blood samples, strip test, thin-layer chromatography, abuse drugs, clinical laboratory tests.

1. Introduction

The essential role of clinical laboratory diagnostic tests is different depending on the availability of free access facilities in clinics or hospitals to abusers treatment. Nowadays, because of the increasing variety of abuse drugs, it makes incidence of epidemics addiction in the world. Also synthetic drugs make diagnostic and therapeutic problems among abusers. Several studies on biological samples for the detection of drugs have been reported, and these drugs are:

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morphine, codeine, heroin, methadone, tramadol, cannabis, amphetamine, methamphetamine, buprenorphine, benzodiazpines, tricyclic antidepressants, barbiturates. So they were tested in this study. Based on studies to achieve the ideal treatment choice, the rights distinguishing of drugs are recommended. For the managements of drug therapy several laboratory methods are sufficient and the most cost-effective to identifying but not accurate. The most appropriate samples for the detection of patients with drug abuse are the use of urine samples of patients. Several studies show that the detection of abuse drugs in urine. Edward J. Cone, et al. showed urine toxicology testing techniques and monitoring of opioids in patients. Rezaei-Basiri, M., et al. [1-3] evaluated screening of morphine and codeine in urine of opioid abusers by rapid and TLC analysis in Tabriz/Iran drug abuse distinguishing laboratory. Rezaei-Basiri, M., et al. specifically evaluated distinguishing of abuse drugs in urine and blood samples of abusers in Iran through rapid strip tests and thin layer chromatography methods in Tabriz/Iran toxicology laboratory too. Also Joseph Pergolizzi, et al. [4] have researched about the role and importance of urine drug testing for patients under opioid treatment [3, 5]. But it might be that there are false positive and negative between results of these studies as well as drug interactions have been reported. Also consumption of all abuse drugs has side effects on organ bodies such liver and kidneys and then evaluation of these organs is necessary through clinical laboratory tests. With this background, the studies on the blood and urine samples of abusers are suggested because they are inexpensive methods that have been used to ensure of proper treatment and follow up of abusers, and serum titers of some other blood parameters showed normal or dysfunction all abusers liver and their renals through clinical laboratory tests methods [2, 4].

2. Materials and Methods

2.1 Study Population

The study included two groups of addict, women with aged 35 ± 15 and men with age range of 45 ± 15 have been recorded. First of all consent forms and questionnaires and also all satisfy forms filled for patients. Information such as age, gender, type of materials, medicines for the treatment and history of the forms has been recorded. All questionnaires and also all satisfy forms after filling confirmed in ethical committee of welfare organization [6]. And 100 mL of urine samples and 10 mL of whole blood samples were collected to their all clinical laboratory examines. Table 1 showed that demographic characteristics of men abusers and women abusers involved in the study. All abusers data were collected on 2016 and 2017.

2.2 Preparation of Samples

All blood and urine samples of all patients were tested using rapid strips test. And 1 mL of all study samples such as blood samples before and after detoxification or MMT duration were centrifuged in 3,000 g for 5 minutes and prepared. Then the serums of blood samples were removed and were exposed to rapid strip tests and the positive and negative and suspicious cases were recorded and reported. In this study for the presence of abuse drugs in blood and urine samples detection limit were > 300 mg/mL to drugs sensitivity and considered positive with using rapid strip tests [7].

Table 1 Demographic characteristics of men abusers and women abusers involved in the study.

Parameters	Men abusers (n = 250)	Women abusers (n = 250)	Statistical analysis
Age (years) (mean ± SD)	45 ± 15	35 ± 15	$p = 0.17$
Years of methadone consumption(mean ± SD)	3.4 ± 0.5	3.2 ± 0.7	$p = 0.24$
Years of buprexin consumption (mean ± SD)	1.43 ± 0.54	1.5 ± 0.51	$P = 0.33$

T-test, $p < 0.05$; significance considered.

2.3 Samples Analysis

All urine samples with using rapid strip have been tested before and after detoxification or MMT (Methadone Maintance Therapy) duration. Also all blood samples before and after detoxifications have been tested with strips. Finally all of urine samples to confirm the presence of abuse drugs have been tested with using thin layer chromatography TLC (thin layer chromatography). Whole blood samples of all abusers were examined to other clinical laboratory tests too.

2.4 List of Abuse Drugs

THC (tetrahydrocanabinol), tramadol, methadone, buprenorphine, morphine, codeine, heroin, MDMA (methylendioxymethamphetamine), amphetamines, methamphetamine, and cannabis are the most basic nature of them [8].

2.5 Statistical Analysis

Before and after detoxification in each group, Anova one and two way statistical analysis and after detoxification t-test among groups of men and women were used for statistical analysis.

3. Results

The results in Tabriz/Iran undrevision clinics of welfare organization were approximately showed that all of study populations had positive results in each of urine and blood samples. The results showed that the patients who encountered in the process of detoxification and MMT terms, their blood samples due detoxification and treatment have been positive results. In the same study the blood test results of all groups after the detoxification were approach negative with rapid strip tests. Results of this study showed that all patients who consumed drugs, such as buprenorphine and methadone to maintenance treatment have positive results in blood test. Also, all of abusers who received maintenance treatment were positive results in urine test that have been reported. The analysis of this study showed that some positive results and false negative test results can be seen in the urine samples of patients. Then the results of this study showed that all patients had positive in results of blood tests because they were used abuse drugs. The 2% results of urine samples were reported false negative by the rapid methods; these samples have been approved positive through TLC procedures. It also has been recorded in 10% of cases through TLC with drug interaction, because patients have used other drugs or medications. According to abusers questionnaire forms attendance information some of the results of this study are showed in Tables 2-5. Table 1 shows it is not significantly demographic characteristics between men abusers and women abusers involved in the study. So Table 2 shows rapid results in duration of treatment in blood samples of women abusers. Table 3 shows rapid strip tests results of blood samples during of before and after detoxification between men and women abusers. Table 4 expresses that the urine test results have shown drug interaction in MMT process of some cases of men and women abusers. Also urine rapid test results during the period of detoxification in men and women groups of study population were shown in Table 5 [9, 10]. The results of this study showed that 4% of abusers had mild increase in hematocrite level and 2% of narcotic drugs abusers had mild lower level of blood sugars than normal range, and 4% of participants had increase liver enzymes such ALT (alanine transferase), AST (aspartat transferase), ALP (alkaline phosphatease) and 1% of them had renal failure. Although blood level BUN (Blood Urea Nitrogen) and creatinin were examined to evaluation of their renal failure [11, 12, 22]. Fig. 1 shows some percentage of abusers parameters variations such as liver and renal function and blood tests through their treatment following.

Table 2 Rapid strip test results in duration of treatment with methadone or buprenorphine in blood samples.

	Women	Men	Consumption history	Methadone or buprenorphine
No	Age	Age	Month or year	> 300 ng/mL
1	24	40	2 yr	+
2	28	31	2 yr	+
3	35	58	3 yr	+
4	30	40	3 yr	+
5	50	55	4 yr	+
6	47	59	3 yr	+
7	25	50	2 yr	+
8	33	56	4 yr	+
9	39	59	4 yr	+
10	35	39	2 yr	+
11	45	41	3 yr	+
12	46	39	2 yr	+
13	50	50	5 yr	+
14	50	40	3 yr	+
15	23	30	1 yr	+
16	22	30	1 yr	+
17	33	39	3 yr	+
18	49	60	4 yr	+
19	48	57	5 yr	+
20	22	30	10 months	+

>300 ng/mL: positive results (+), N = 20, rapid strip test, blood samples.

Table 3 Rapid strip tests results of blood samples during before and after detoxification.

	Women	Men	Before detoxification	After detoxification
No	Age	Age	> 300 ng/mL	< 300 ng/mL
1	50	60	+	-
2	48	46	+	-
3	46	45	+	-
4	41	41	+	-
5	35	39	+	-
6	23	30	+	-
7	22	37	+	-
8	20	38	+	-
9	21	32	+	-
10	24	30	+	-
11	31	31	+	-
12	32	36	+	-
13	47	39	+	-
14	49	40	+	-
15	48	42	+	-
16	46	40	+	-
17	46	45	+	-
18	49	49	+	-
19	50	50	+	-
20	38	37	+	-
21	35	39	+	-
22	39	36	+	-
23	31	32	+	-
24	39	35	+	-
25	29	35	+	-
26	22	39	+	-
27	26	38	+	-
28	26	30	+	-
29	21	30	+	-
30	22	59	+	-

> 300 ng/mL: positive results (+), < 300 ng/mL: negative results (-) N = 60, rapid strip test, opiod (heroin, morphine, codein, cannabis, metamphetamine) detection in abusers men and women blood samples. Anova two way, $p = 0.0001$.

Table 4 Urine test results have shown drug interaction in MMT process of some cases.

	Women	Men	Consumption history	Methadone buprenorphine
No	Age	Age	Month or year	>300 ng/mL
1	49	55	2 yr	±
2	48	44	3 yr	±
3	46	41	2 yr	±
4	44	40	1 yr	±
5	41	40	4 yr	±
6	25	30	1 yr	±
7	49	60	5 yr	±
8	50	59	5 yr	±
9	21	32	10 months	±
10	22	31	2 yr	±
11	24	35	1 yr	±
12	50	58	4 yr	±

Age > 35, N = 12, drug interactions have after detoxification through TLC (thin layer chromatography) method, ± (drug interaction).

Table 5 Urine rapid strip test results during the period of before and after detoxification in population study of men and women.

	Women	Men	Before detoxification	After detoxification
No	Age		> 300 ng/mL	< 300 ng/mL
1	50	60	+	-
2	50	59	+	-
3	44	58	+	-
4	49	57	+	-
5	43	55	+	-
6	47	50	+	-
7	41	49	+	-
8	50	44	+	-
9	43	41	+	-
10	41	44	+	-
11	40	40	+	-
12	40	39	+	-
13	42	58	+	-
14	42	42	+	-
15	28	39	+	-
16	27	30	+	-
17	26	31	+	-
18	25	33	+	-
19	34	39	+	-
20	23	30	+	-
21	32	35	+	-
22	21	30	+	-
23	20	45	+	-
24	28	45	+	-
25	29	43	+	-
26	23	44	+	-
27	25	56	+	-
28	28	31	+	-
29	38	31	+	-
30	35	32	+	-
31	33	60	+	-
32	31	55	+	-
33	48	46	+	-
34	50	39	+	-
35	49	30	+	-

> 300 ng/mL: positive results (+), < 300 ng/mL: negative results (-) N = 70, rapid strip test, abuser drugs (AM, MA, MDMA, canabis, tramadol, opiods…) detection in men and women abusers urine samples. Anova two way, $p = 0.0001$.

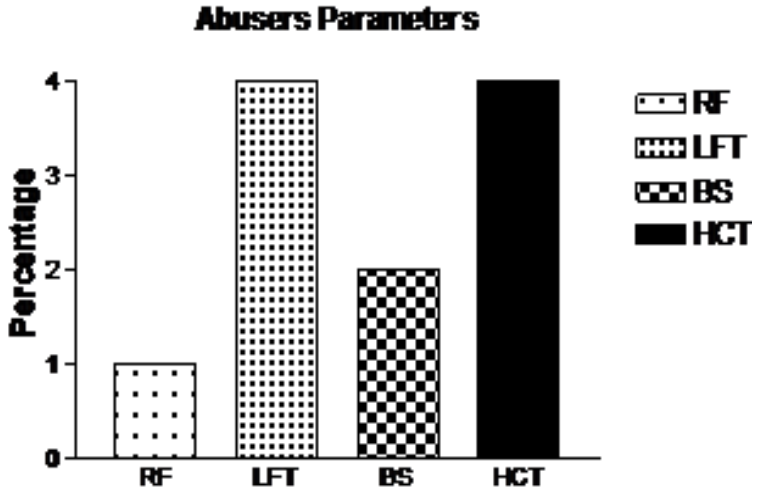

Fig. 1 Percentage of abusers parameters variations.
RF = Renal failure;
LFT = Liver function test;
Liver enzymes: ALT, AST, ALP;
BS = Blood sugars;
Decrease blood sugars;
HCT = Hematocrite increase, mild erythropoiesis.

4. Discussion

Depending on the clinical situation, selections of drug treatment and diagnostic methods are varied in different countries of the Middle East. Clinics of Iranian drug addiction care centers used buprenorphine, naloxone, naltrexone and methadone for treatment of opioid dependence and multiple protocols for laboratory diagnosis [2, 4]. According to clinical picture of abusers and facilities in different countries, diagnostic and treatment in these regions are different. The results of this study show patients who chronically and persistently consume abuse drugs or other drugs, so these substances and medications are distributed in the body and then the existence of these drugs in their body fluids is often possible [13-14]. So the materials can be found in most biological fluids. Tables 2, 3 and 5 are clearly shown positive and negative results in treatment course of MMT and detoxification between men and women abuser patients. These results suggest that the rapid strip test methods for accurate diagnosis are considered appropriate and beneficial process [15, 16]. The results of Table 4 show that most patients are over 20 years, and continuously for the treatment or detoxification has consumed drugs that the chromatography urine test results of them have shown drug interactions, then the use of TLC is very suitable to confirm of drug interactions. Obviously, such patients in addition of drug therapy for the treatment of addiction, also have used other drugs to treatment of drugs abuse side effects. The results of Tables 2 and 5 show the abusers who deny risk of their addiction and in these cases it might be in the urine samples positive and false negative results can be seen, then using blood samples are suitable to diagnose and confirm the presence of addictive drugs. Nowadays, the alternative and feasible methods to distinguish of over dose drugs are requirement in drug poisoning in hospitals and forensic medicines, in order to detection and diagnosis of other medicinal or drugs poisoning with rapid strip test kits with sensitivity above 300 ng/mL through urine samples are necessary, these kits have sensitivity to distinguishing of drugs over dose in blood samples too [16, 17]. Some of these studies demonstrate useful results for long-term treatment of methadone to opiate abusers. In the all of clinical diagnostic laboratory

centers such as addiction rehab centers and addiction clinics and psychiatric hospitals, clinical laboratories, pharmaceutical laboratory of toxicology and forensic departments routinely use from liquid phase and solid phase extraction techniques to distinguish opioids in biological samples by the TLC, and serum titers of some other blood parameters showed normal or dysfunction all abusers liver and their renals through clinical laboratory tests methods. Also according to this study the side effects of abuse drugs consumptions followed up through clinical laboratory tests methods [19]. Attendance to Fig. 1, it might be the increasing of blood hematocrite levels of some abusers that were related to oxygen decreasing during of their smoking conditions. The erythropoiesis makes increasing blood hematocrite levels. The feedback of oxygen decreasing levels causes bone marrow motivation to erythropoiesis and oxygen supplies [20, 21].

5. Conclusions

Depending on the strategy for the treatment of patients, buprenorphine and methadone maintenance therapy is used. Always blood and urine tests will be positive for these substances duration of MMT. We conclude that between all drug analytical methods the cheapest and easiest tests of opioids and drugs in urine and blood samples are strip test for rapid diagnosis and TLC is appropriate confirmation method to drug abuse distinguishing, also doing test on blood samples is high importance to distinguish of drugs abuse. The basic function of rapid strip test kits is immunochromatography. They are made and available cheaper than other methods in Iran [16]. According to background of this study, we will be proposed that to detection and diagnosis of other medicinal or drugs poisoning using blood samples and rapid strip test kits with sensitivity above 300 ng/mL are recommended. Also tests on blood samples have high importance as a view point of accuracy to distinguish of drugs abuse, and serum levels of some other parameters showed dysfunction in some of abusers' liver and their renals through clinical laboratory tests [22].

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