

## Effect of Short Time Morphine Addiction on the Kidney Tissue in Rat

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**Abstract:** Opium is an important substance that abuses and has very pharmacological and pathological effects. Morphine is the main substance of opium. In the recent study, it has been tried to identify the short effects of morphine on the kidney pathology of Rats. The randomized double blind study was designed. Twenty rats were selected and divided in two groups and maintained under routine laboratory conditions and similar intervention. One group had been addicted orally by morphine powder and control were not addict. After three weeks, all rats were anesthetized and underwent autopsy. Their kidneys were removed en-block and sent to pathology laboratory and reviewed by one pathologist. The major considered variables were tubulo-interstitial and glomerular changes, glomerular amount and size on light microscope view. Glomerular, tubular and vascular changes, glomerular counts, glomerular size, and cortical thickness showed no significant differences between groups of study. Only Lymphocytic infiltration was higher frequency in addict group, significantly  $p = 0.007$ . It seems short course of morphine consumption is very little effect on kidney, only Lymphocytic infiltration observe in addicted renal tissue which, may be precursor of other changes in kidney. It is better long term studies are implemented in this course.

**Key words:** Morphine, addict, kidney, rat

### INTRODUCTION

In human and experimental animals, morphine is extensively metabolized via conjugative and oxidative pathways to metabolites that exhibit pharmacological activity<sup>[1]</sup>. Although only 10% of a dose of morphine is excreted unchanged in urine. The kidneys play a major role in the excretion of its metabolites<sup>[2,3]</sup>. Drug metabolizing enzymes are known to exist in the mammalian kidney and morphine is one of many drugs that have been suggested to undergo renal metabolism. Clinical observations have shown higher level of renal disease in opium-addicted patients and there were many few studies about this subject. The effect of morphine on renal histopathology has been investigated rarely. The present study was designed to investigate the effect of morphine on microscopic renal tissue changes in the rats.

### MATERIALS AND METHODS

A double blind randomized experimental trial was designed. Twenty male Spruge-Dawley rats with between 250-300 g were selected. They were divided to two groups randomly. One of them were addicted by morphine powder by rapid method of morphine dependency that examined by other researchers<sup>[4-8]</sup>. Morphine hydrochloride powder (Daru Paksh Ltd. Tehran-Iran) was used for addiction. After 3 weeks all

cases and controls rats examined with Naloxane  $2 \text{ mg kg}^{-1}$  to check dependency. Signs of dependency were controlled by a check list<sup>[4-8]</sup>. Whereas rat exhibited dependency signs, were being taken for granted addict. Then they were anesthetized by Ketamine (Merk Co. Germany) and underwent autopsy. Their kidneys were removed en-block and sent to pathology laboratory in Formalin. After fixation process, pathologist provided paraffin based block of renal tissue and sliced them. Four kind of staining were performed for each specimens:

- Hematoxilin-Eosin for routine evaluation
- Periodic acid shift for evaluation of basement membrane changes
- Masson tri chromium staining for evaluation of tubulo-interstitial changes
- Jones staining for evaluation of glomerular and mesenchymal changes<sup>[9]</sup>.

All stained slices review from the viewpoint of tubulo-interstitial and glomerular changes, glomerular amount and size by light microscope with 400 in 5 fields.

Pathologist was blinded about groups and all blocks were review two times. The study process was approved by the committee of research at Kerman medical sciences university.

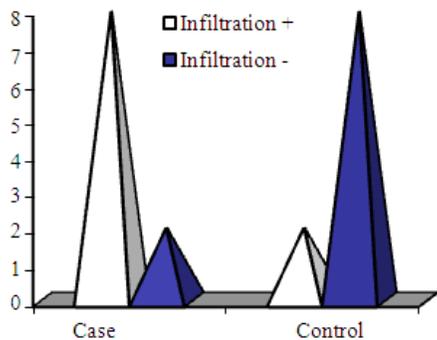


Fig 1: Frequency of lymphocyte infiltration in two groups,  $\chi^2 = 7.2$ , DF = 1,  $p = 0.007$

For data analysis the SPSS 11.5 was used. Comparison of numeric variable like amount, size, and thickness of glomerous' was carried out by two sample t-test, and results were represented as mean $\pm$ SD. Nominal data were compared between groups by Chi squared test. Difference was considered to be statistically significant at  $p < 0.05$ .

## RESULTS

Among histopathological variables which were studied; glomerular, tubular and vascular changes showed no significant differences between groups of study.

In addict and control groups mean glomerular counts were 14.6 $\pm$ 1.2 and 15.5 $\pm$ 0.7 in order so the difference was not statistically significant ( $p = 0.59$ ).

Also mean cortical thickness was 208.8 $\pm$ 27 micron in addict group and 178 $\pm$ 15.5 micron in control group, Statistically no significant difference could be found between groups. The next variable which was analyzed was glomerular size in diameter. Mean glomerular diameter in addict and control groups were 25.4 $\pm$ 1.8 and 24.2 $\pm$ 0.8 which were not different statistically ( $p = 0.85$ ).

Lymphocytic infiltration in renal cortex was found in eight addict rats versus two rats in control group. The difference was statistically significant ( $p < 0.007$ , DF = 1,  $\chi^2 = 7.2$ ) (Fig. 1).

## DISCUSSION

There are several substances, which are released from opium in the body. Morphine, which is the main alkaloid in this group, is thought to play the main role in what opium does in beings. Morphine has several effects on physiology, histology and immune system of the body. Some of them, which are known, are listed below.

Weakening immune system, reduction of total body oxygen consumption and hypoxia, stimulating antidiuretic hormone release from kidneys, peripheral and visceral vasodilatation<sup>[10]</sup>.

Kidney is one of the main sites of morphine metabolism<sup>[10]</sup> so renal problems could be expected in morphine addiction. Johnson showed that application of morphine in rats could result in significant degenerative changes in kidneys. Increased pedocytic microinjections were one of the important changes in this study<sup>[11]</sup>.

Toupalik and coworkers showed that acute tubular nephritis is found in chronic heroin addicts and explained that this is the result of repeated hypoxia in kidneys<sup>[12]</sup>.

Dettmeyer and coworkers found inflammatory reactions like lymphomonocytic glomerulonephritis. In a significant proportion of kidneys of addicts<sup>[13]</sup>.

Chronic tubulointerstitial disease with tubular atrophic changes and monocytic infiltration is another known problem, which is well described in chronic application of analgesics.

According to the high prevalence of opium addiction in our community and the clinical experiences that show a higher incidence of kidney problems in opium addicts comparing to non-addicts and also regarding the studies mentioned above we decided to design this study.

The differences of mean cortical thickness, glomerular count and glomerular diameter were not significant between two groups of study. But reversed variations like increased cortical thickness and glomerular diameter con-committed with decreased glomerular count in addicts can show primary stages of a renal pathology.

Kidneys respond to stress by hypertrophy but not hyperplasia. According to these reversed variations and just slightly high p-values 0.06, and 0.059. One may conclude that sample size or addiction duration could be the reason. As mentioned above duration of addiction could be important. In this study, we found primary stages of renal pathology. Maybe continuing the study could reveal and show the differences between two groups.

Something important found in this study is significantly higher proportion of rats with lymphocytic infiltration in kidneys among addict rats comparing to the non-addict ones. This could show presentation of kidney problems in addict rats. The most important factor responsible for this is hypoxia in kidneys. As mentioned before, morphine could decrease systemic and therefore renal blood flow by peripheral and visceral vasodilatation<sup>[10]</sup>. Decreased total oxygen consumption is another factor, which could increase hypoxia. Kidney is sensitive to hypoxia and responds it by infiltration of inflammatory cells.

## CONCLUSION

Anyhow, we recommend more studies in cellular and molecular levels and different addiction durations

to be done in order to show what are the changes caused by morphine.

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