Effect of Orally-administered Silver Nanoparticles (Ag-NPs) on Some Biochemical Parameters in Kidney of Rats

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Abstract

The research evaluated the oral administration of silver nanoparticles on the tissue morphology and biochemical parameters of kidneys in rats, using (0.2 and 0.4) mg/kg B.W for four days and 0.08,0.008 mg/kg weight For 30 days . The result showed significant increase in the level of serum creatinine and serum urea for animals treated with silver nanoparticles relative to the control at the dose of (0.08 and 0.008) mg/kg B.W for 30 days. As for the histological study of kidneys showed sever changes at the dose (0.08 and 0.008) mg/kg compared with the dose 0.2 and 0.4 mg/kg for 4 day which include infiltration of inflammatory cells ,haemorrhage, and degenerated and necrotic changes in renal tubules epithelium.

Keywords*: Rats, AgNPs, histopathology of Kidney.*

Introduction

Silver nanoparticles (AgNPs) accumulated in body organs and has a toxic effects especially on kidney [(1)]. (AgNPs) have tremendous potentials in medical devices due to their excellent antimicrobial properties.The nanoparticles has a small size which have appositive effect on using as these mechanical ,electrical and chemical. These characters also have a side effect as a biological and toxicological reactivity [(2)] .This will element the using of silver nanoparticles. The ingestion of silver nanoparticles can accumulated on parenchymatous organs which cause toxic damage hepatic and renal tissues [(3)].

The harmful effect of kidney function from any material that can be described as nephrotoxicity [(4)] . One indication of nephrotoxicity is a change in renal function as assessed by the glomerular filtration rate (GFR), blood urea nitrogen (BUN), serum creatinine (SCr), or urine output; however, nephrotoxicants can induce kidney damage without changing any established clinical marker of renal function. For example, studies have shown that proximal tubule necrosis in male Sprague Dawley rats exposed to gentamicin can be as high as 75% prior to any increases in BUN or SCr [(5)].

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Materials and Methods

Animal test was performed with compliance of the local ethics committee. A group of 25 BALB/c rats of about 4 weeks (weighting 48.2± 3.0 g) were purchased from Medical Faculty of Mosul University and then transferred to the laboratory. The animals were housed in a single group and maintained on commercial pellet diet, given deionized water ad libitum and kept in plastic cages. After 2 weeks' acclimation, they were randomly divided into three groups (each with two replications): the control (1) groups, with 5 rats and Ag-NPs group(2) (0.2 mg/kg,0.4 mg/kg,0.08mg/kg and 0.008 gm/kg). and carried out for 30 days alternatel The animals were kept fasting over night before. The blood and kidneys were harvested and prepared for biochemical and histopathological examinations, respectively.

The silver nanoparticles characters :

The size of silver nanoparticles used in this study is about 200-300 nm, preparation by SIGMA-ALDRICH company (< 100 nm).

The Histological Analysis and Biochemical Analysis

Biochemical tests used for evaluation the toxic effect of silver NPs on the rats used in this study the estimation of Urea and creatinine were done in the serum by using commercial kits: Katal for urea and Creatinine. All analyzes were performed in biochemical analyser from Bioplus (Celm SB-190) at 37°C.

(b) Histological Analysis.The fixed specimens at 10% neutral buffer formalin are processing ,then embedded in paraffin, sectioned at 4.5- 5 µm by using rotary microtome then staining by hematoxylin and eosin then examined by light microscope for studying the histological changes [6].

Statistical Analysis

A statistical analysis of the results of this search was performed using one –way of ANOVA table for multiple comparisons . A post-hoc method(Tukey) was used in the graph pad prism 5 progarm to evaluate a meaning level of p. 0.05

Results

The study results showed no clinical signs and no mortality was seen in all groups of animals when compared with non-treated group has occurred in each the samples, as well as, no abnormalities in the appearance and behavior of AgNPs treated rats in comparison with non- treated ones..

The mean level of blood urea in groups (0.2,0.4,0.08 and 0.008) showed a significant increase ($p \leq 0.05$) compared with the control group Fig(1).

Orally administred of silver material to animals no large increase serum Creatinine levels was observed relative to the control in rats treated (0.2 and 0.4) mg/kg B.W for four days Fig(1),with contrast in rats which treated with (0.08 and 0.008)mg/kg B.W for 30 days, the level of serum urea and creatinine are significant increase relative to the control Fig(2).

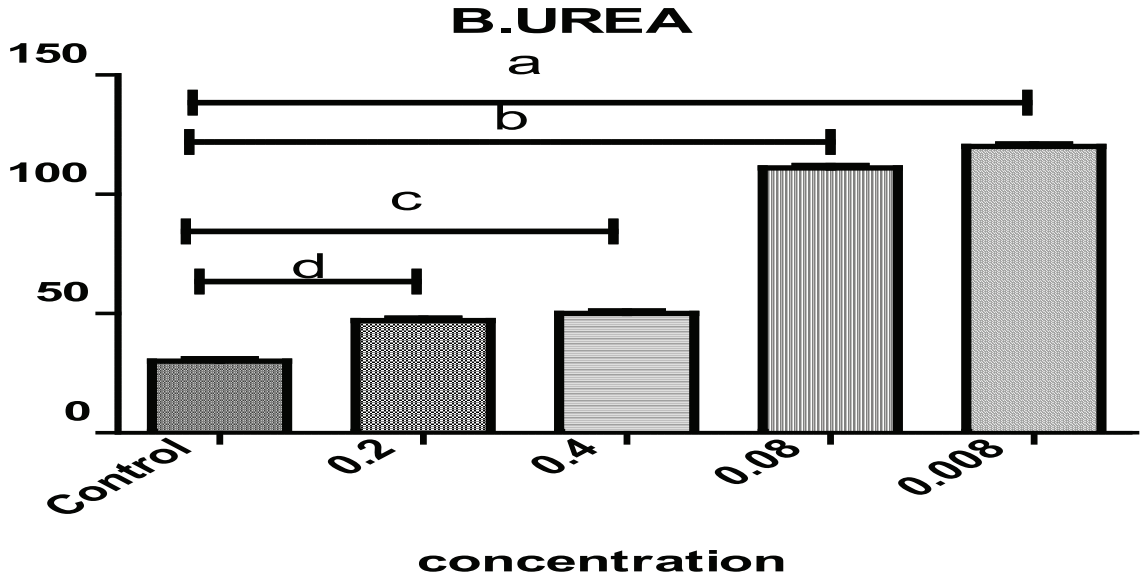


Fig (1) : Effect of different doses of AgNPs (0.2,0.4,0.08 and 0.008 mg/kg) on Blood Urea

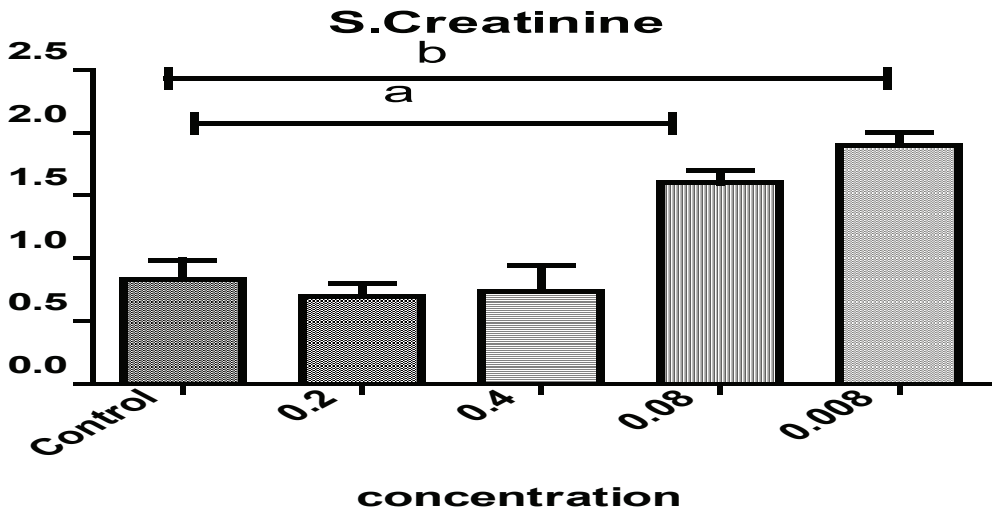


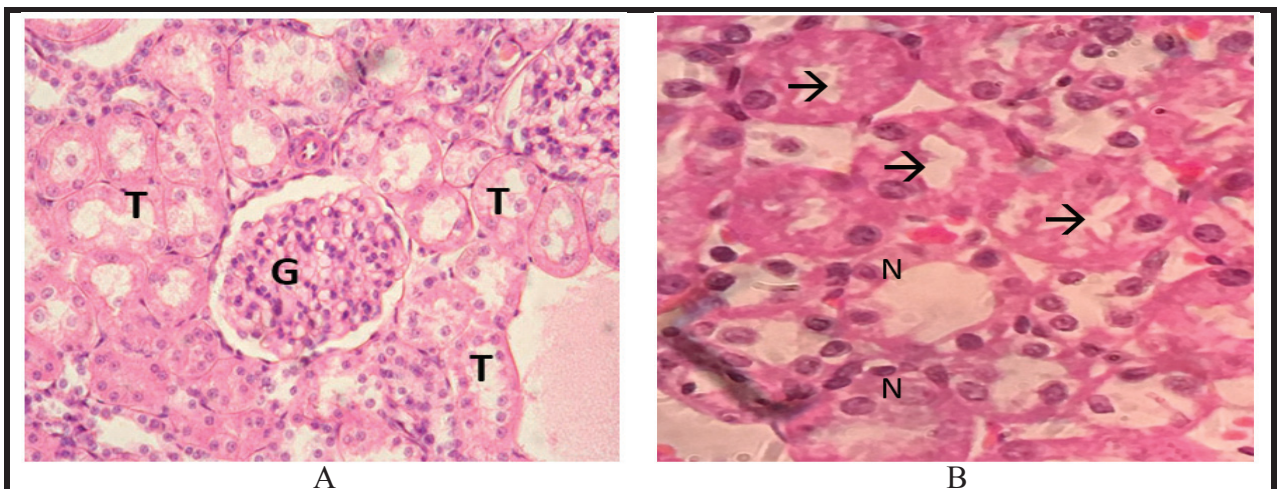
Fig (2): Effect of different doses of AgNPs (0.2,0.4,0.08 and 0.008 mg/kg) on Serum Creatinine

Histological Fidings:

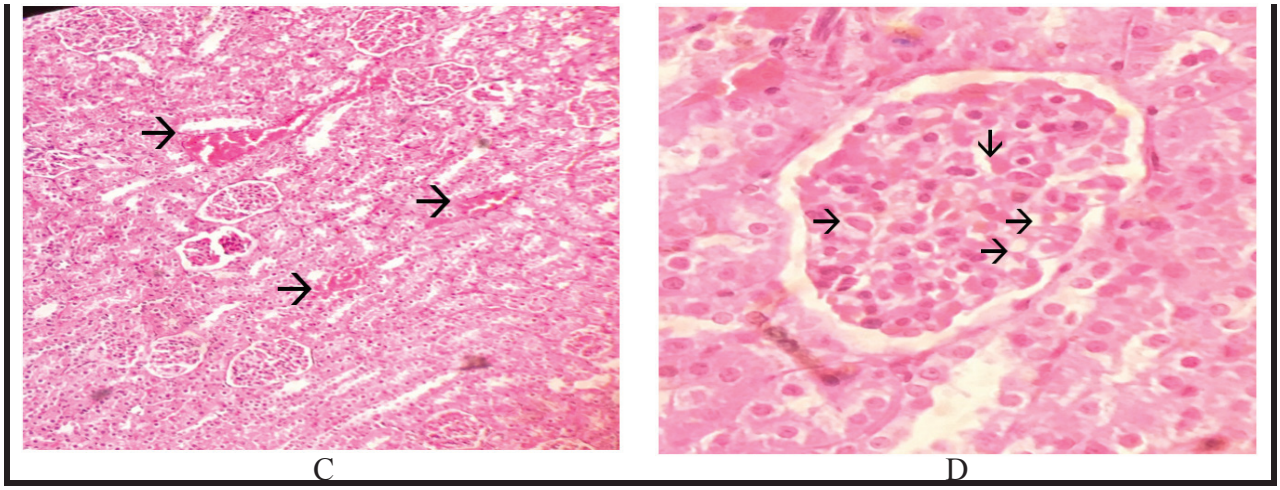
The sections of tissues were prepared for morphological changes after exposure to Ag nanoparticles and observed under a microscope. Histology examination of kidney specimens were evaluted in the three compennts ,tubular,glomerular ,and interstitial tissues. The histological examination of control group showed normal architectural of renal tubules, glomeruli and interstitial tissues with no histological changes Fig(3-A).

While the evalution of the kidney specimens of animal treated with silver nanoparticles using (0.2 and 0.4 mg/Kg B.W.)for four days showed acute necrosis of epithelial cell lining renal tubules associated with swelling of epithelial cells lining renal tubules lead to stenosis the lumen which appeared a star shape (Fig 3-B).generalized congestion of blood vessels (Fig 3-C). While the glomerular altration characterized by vacuolar degeneration and necrosis of mesengial cells(Fig 3-D).

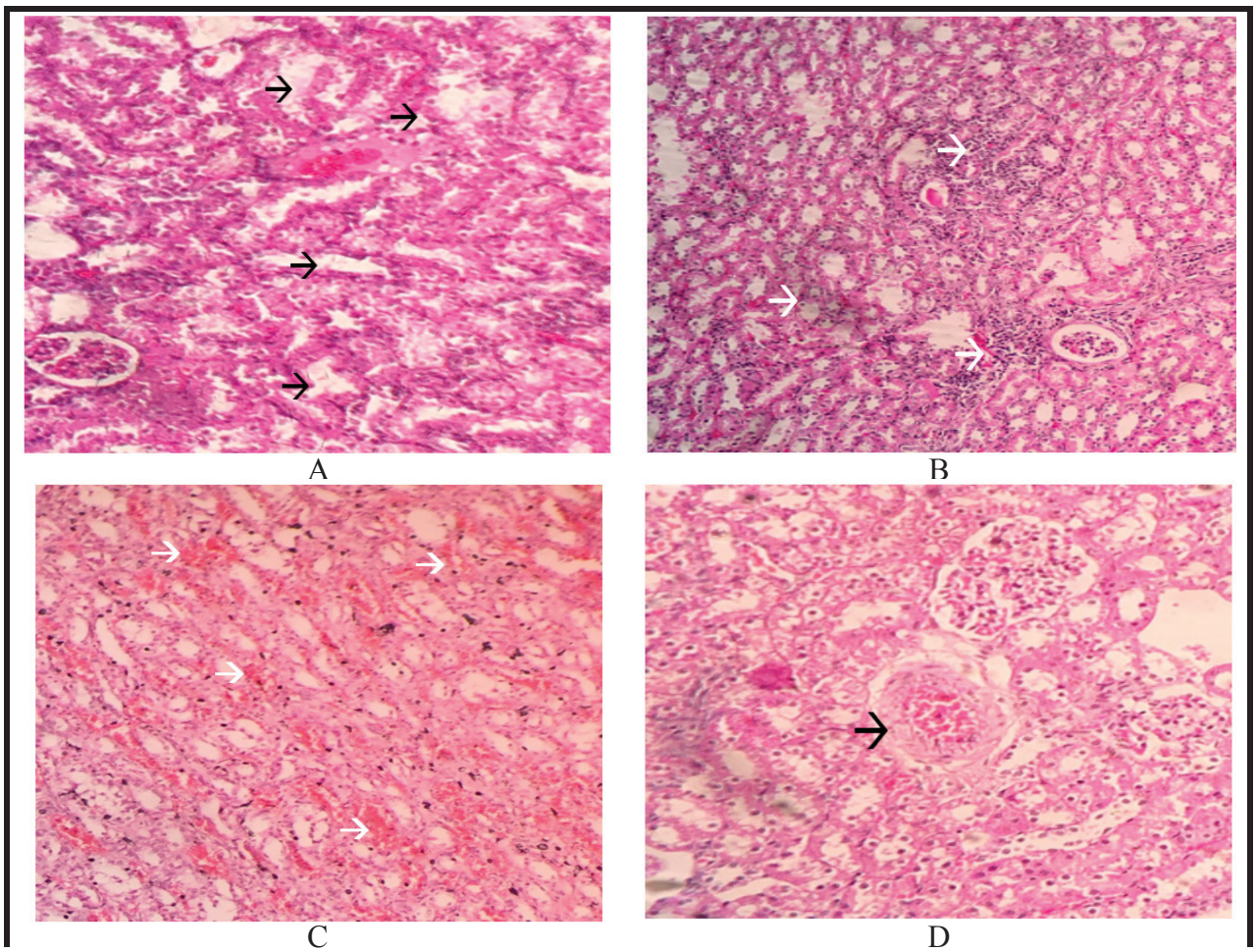
While the histological examination of kidney specimense of animals treated with(0.08 and 0.008 mg/ Kg B.W.)of silver nanoparticles for 30 days showed histological changes more sever than those treated for four days these changes including thickening in the basement membrane of renal tubules with necrosis of epithelial cell lining renal tubules(Fig 4-A). Focal infiltration of inflammatory cells(macrophages,lymphocytes) (Fig 4-B), sever heamorrhage in the interstitial tissues (Fig 4-C) thickening in the wall of blood vessels (Fig 4-D),. Also glomeruli suffered from increased the cellularity of glomerular tuft (Fig 4-E).Other section showed sloughing and desquamation of epithelial cells lining renal tubules in the lumen of tubules there is cellular debris (Fig 4-F). Also there was distortion of the basement membrane of renal tubules and glomeruli.



Cont... **Figure (3)**



Fig(3): (3-A) Light Micrograph Section Kidney of Control group of Rat showing architectural of renal tubules normal (T) and glomeruli (G),10X ; (3-B) sections for tubular analysis in kidney from 0.2 mg/kg AgNS treated rats. acute necrosis of epithelial cell(N) lining renal tubules associated with swelling lead to stenosis the lumen(à) 40X ; (3-C) micrographs sections for tubular analysis of kidney from (0.2 and 0.4) mg/kg AgNS treated rats. Generalized congestion of blood vessels . 4X ; (3-D) sections of kidney from(0.2 and 0.4) mg/kg AgNS treated rats. vacuolar degeneration and necrosis of mesengial cells (à) 40X , (Haematoxylin and Eosin stain).



Cont... **Figure (4):**

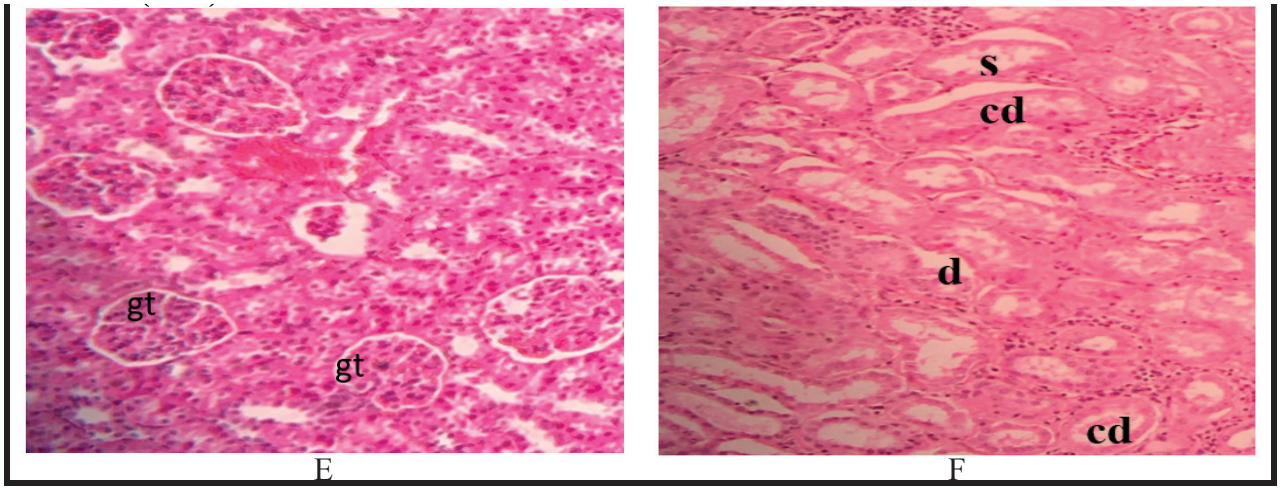


Figure (4): Light micrographs sections (4-A) kidney from(0.08 mg/kg)AgNS treated rats. thickening in the basement membrane of renal tubules with necrosis of epithelial cell lining renal tubules(à),10X ; (4-B) sections for tubular analysis of kidney from(0.08 and 0.008) mg/kg AgNS treated rats. Focal infiltration of inflammatory cells(à) (macrophages , Lymphocytes), 4X ; (4-C) sections for tubular analysis of kidney from(0.08 and 0.008) mg/kg AgNS treated rats. sever heamorrhage in the interstitial tissues(à) ,10X ; (4-D) sections for tubular analysis of kidney from(0.08 and 0.008) mg/kg AgNS treated rats. Foamy thickening in the wall of blood vessels(à), 10X ; (4-E) sections for tubular analysis of cortical region in kidney from(0.08 and 0.008) mg/kg AgNS treated rats. Foamy glomeruli suffered from increased the cellularity of glomerular tuft (gt), 400X ; (4-F) section of rat kidney following oral exposure to silver nanoparticles at different dosages (0.08 and 0.008) mg/kg AgNS treated rats. Show sloughing (s) and desquation of epithelial cells lining renal tubules in the lumen of tubules there is cellular debris (cd) and distortion (d)of the basement membrane of renal tubules, 4X (Haematoxylin& eosin stain).

Table (1) showed the scoring of histological changes of kidney in the treated group and control group.

Table 1: Effect of AgNPs (0.2,0.4,0.08 and0.008 mg/kg) for 30 days on tubular qualified criteria; The following grading scheme was used to score the histological alterations: (−) absent; (+) mild; (++) moderate; (+++) severe

histological alteration	Control	0.2	0.4	0.08	0.008
Basement membrane irregularity	-	+	+	+	+
swelling cell	-	+	-	+	++
Necrosis	-	++	+	+	-
Congestion	-	+	++	+	+++
Vacuolar degeneration	-	+	+	+	+
Infiltration of inflammatory	-	-	-	+	+

Different letters show the statistically significant differences between treatment and control (Tukey test, $P \leq 0.05$).

Discussion

AgNPs have attracted the attention of researchers due to their application in the treatment of diseases in addition to its antimicrobial effects [7] and [8] AgNPs used in a variety of application [9]. The wide range application of nanoparticles far many biomedical and industry [10] also used in coating of surgical instrument [11] and prothesis[12]. Yet, the nanomateriales have higher surface to volume ratio that makes them more reactive and toxic due to smaller size of silver NPsthis will cause penetration through the tissue then cause cytotoxic effect [13] .

This study showed that nanoparticle administration cause increased the level of serum urea and creatinine this result to estimate effect g NPs on renal function. Serum urea and creatinine are useful indices for evaluating the status of renal functions[14]. A rise in the level of serum urea may imply impaired renal excretion [15] may be due to inactivation resulting from the affinity of the Ag NPs for thiol (-SH) groups, hereby causing functional state of proteins to change [16] .

This study showed that silver nanoparticles have a toxic effect on renal tissue which occur due to the deposition of AgNPs in the kidney which cause histological alteration So the recent study showed a histological alteration in the renal tubules , glomeruli and interstitial tissue , these alteration includes degeneration and necrosis in the tubules and glomeruli with in infiltration of inflammatory cells and haemorrhage in the interstitial tissue these changes occur as aresult of (ROS) production and release of cytokines which have a toxic effect and leading to histological alteration by causing DNA damage [17] [18] [19]

Such histological changes caused by AgNPs are evidence that supports the ability of nanoparticles to cause biochemical alteration that result in cell damage [20] .

Conclusion

The conclusion of this study that the biochemical and histological alteration of kidney induced by AgNPs due to the toxic effect of nanoparticles the severity of alteration depend up on the dose and duration of the silver nanoparticles Authors contribution

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Ethical Clearance: The Research Ethical Committee at scientific research by ethical approval of both MOH and MOHSER in Iraq

Conflict of Interest: The writers claim a manuscript has no conflicts of interest.

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