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European Journal of Experimental Biology, 2014, 4(2):351-356

ISSN: 2248 –9215
CODEN (USA): EJEBAU

Acetaminophen as antibacterial agent and its effect on antibiotic susceptibilities of some pathogenic bacteria

Zainab R. Abdul-Hussein

Dept. of Biology, College of Science, Basra University, Iraq

ABSTRACT

The effect of acetaminophen extracted from paracetamol was investigated as antibacterial agent against Escherichia coli, Staphylococcus aureus, and Klebsiella pneumonia isolated from urinary tract infection ,and Pseudomonas areuginosa from wound infection, also antibiotic susceptibilities of acetaminophen exposed bacteria towards 10 antibiotics was investigated. Acetaminophen affect the growth of all studied bacteria with increasing of the concentration of the drug. Also, susceptibilities of bacteria towards antibiotic was affected when compared with non-exposed (control)bacteria.

Key words: Acetaminophen, Antibacterial, Pathogenic bacteria

INTRODUCTION

Paracetamol or acetaminophen, chemically named N-acetyl-p-aminophenol, is a widely used analgesic (pain reliever) and antipyretic (fever reducer). Paracetamol is classified as a mild analgesic. It is commonly used for the relief of headaches and other minor aches and pains and is a major ingredient in numerous cold and flu remedies.

It is not generally classified as an non-steroidal anti-inflammatory drugs NSAID because it exhibits only weak anti-inflammatory activity.

To date, the mechanism of action of paracetamol is not completely understood. The main mechanism proposed is the inhibition of cyclooxygenase(COX), and recent findings suggest that it is highly selective for COX-2[1].While it as analgesic and antipyretic properties comparable to those of aspirin or other NSAIDs, its peripheral anti-inflammatory activity is usually limited by several factors, one of which is the high level of peroxides present in inflammatory lesions. However, in some circumstances, even peripheral anti-inflammatory activity comparable to NSAIDs can be observed [2].

Because of its selectivity for COX-2 it does not significantly inhibit the production of the pro-clotting thromboxanes [1] The COX family of enzymes are responsible for the metabolism of arachidonic acid to prostaglandin H2, an unstable molecule that is, in turn, converted to numerous other pro-inflammatory compounds. Classical anti-inflammatories such as the NSAIDs block this step. Only when appropriately oxidized is the COX enzyme highly active [3,4]. Paracetamol reduces the oxidized form of the COX enzyme, preventing it from forming pro-inflammatory chemicals. This leads to a reduced amount of *prostaglandin E2* in the CNS, thus lowering the hypothalamic set-point in the thermoregulatory center[5, 6]

MATERIALS AND METHODS

Bacterial Isolates
Different pathogenic bacteria, i.e. *E. coli, Staphylococcus aureus* and *Klebsiella pneumonia* from urinary tract infection and *Pseudomonas aeruginosa* from wound infection were used in the present study.

Extraction acetaminophen
Acetaminophen was extracted by powdering (29) gm. of paracetamol in (100) ml of boiling ethanol with stirrer, centrifugation, after that supernatant was dried at room temperature in dark[7]

Preparation of stock solution
To obtain the stock solution (100000µg/ml) of Acetaminophen ,0.1gm from the extract was dissolved in 1 ml of DMSO (Dimethyl sulfoxide).Then a serial dilution (10-10000 µg/ml) from the stock solution were prepared.

Preparation of bacterial culture
One colony from each bacterial stock culture was inoculated in 4 ml nutrient broth and incubated at 37^{o}C for 24 hours.

Antibacterial assay
0.1 ml from each bacterial broth was spreaded on Muller Hinton agar plate by using sterile L-shape rod , then well made with sterile corkborer, after that 0.1 ml from each concentration of acetaminophen was added to well .All plates were incubated at 37^{o}c for 24 hours ,then diameters of inhibition zone were measured for antibacterial effect determination.

Antibiotic sensitivity of acetaminophen exposed bacteria
0.1 ml of 24 hours acetaminophen exposed bacterial broth culture was spreaded on Muller Hinton agar. Susceptibility of isolates to different antibiotics were tested following Kirby Bauer disc diffusion method against selected antibiotics(Bioanalse), namely: chloramphenicol(10 mcg), tetracyclin(10 mcg), vancomycin(10 mcg), amikacin(10 mcg), erythromycin(15 mcg), Amoxiclav(amoxilin/clavulamic acid)(20/10 mcg), cefataxime(10 mcg), canamycin(30 mcg), ciprofloxacin (10 mcg), amikacin(10 mcg) and imipenem(10 mcg).

RESULTS AND DISCUSSION

Antibacterial screening of acetaminophen
The finding of the present study revealed that all concentrations of acetaminophen extracted from paracetamol tablets reduced the growth of each of the islolated bacteria from urinary tract infection i.e. *E. coli* and *Staphylococcus aureus, Klebsiella pneumonia* or *Pseudomonas aeruginosa* isolated from wound infection.The growth of all bacterial species were inhibited with increasing of drug concentration.

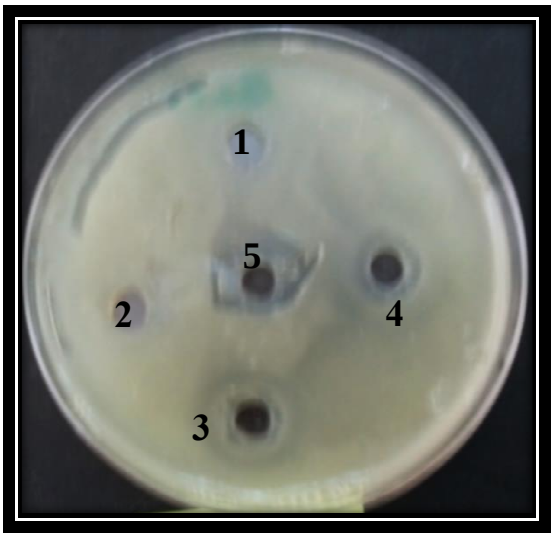
Table (1) antibacterial effect of acetaminophen

Acetaminophen	concentration *				
Bacteria	**1**	**2**	**3**	**4**	**5**
Pseudomonas aeruginos	**12**	**12**	**14**	**18**	**20**
E coli	**10**	**12**	**12**	**16**	**18**
Staphylococcus aureus	**0**	**0**	**10**	**12**	**22**
Klebsiella pneumonia	**0**	**10**	**10**	**12**	**16**

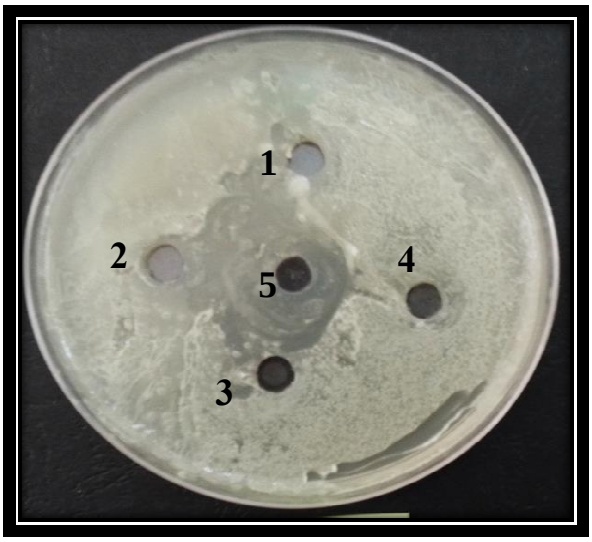
**=(1=1*10,2=1*10^{2},3=1*10^{3},4=1*10^{4}.5=1*10^{5})*

Paracetamol is part of the class of drugs known as "aniline analgesics"; it is the only such drug still in use today[8] It is not considered a non-steroidal anti-inflammatory drugs NSAIDs because it does not exhibit significant anti-inflammatory activity (it is a weak COX inhibitor)[9].

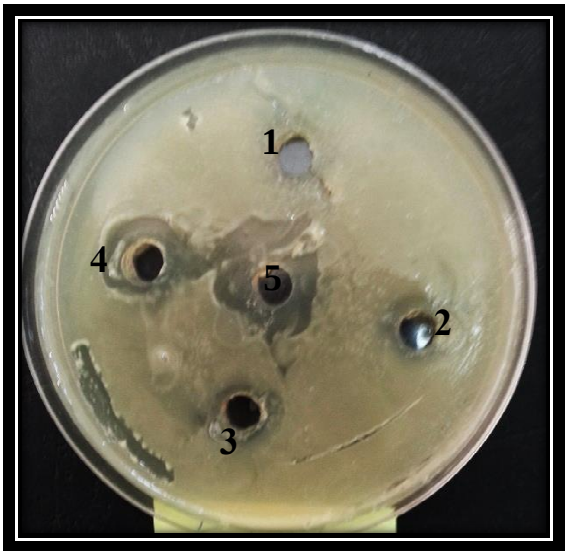
In his study of in vitro activity of ibuprofen and acetaminophen on bacteria Al-Janabi found out that acetaminophen affect the growth of each of *Staphylococci aureus, Bacillus subtilis, E. coli, Enterobacteraerogenes, Enterobacter cloacae, Salmonella typhi and Paracoccusyeei*[10]. Complexes of acetaminophen with Co, Ni or Fe element (not acetaminophen alone) revealed variable inhibitory effects on *E. coli*, while *Serratia* and *Bacillus subtilis* didn't affect by any of these complexes[11].



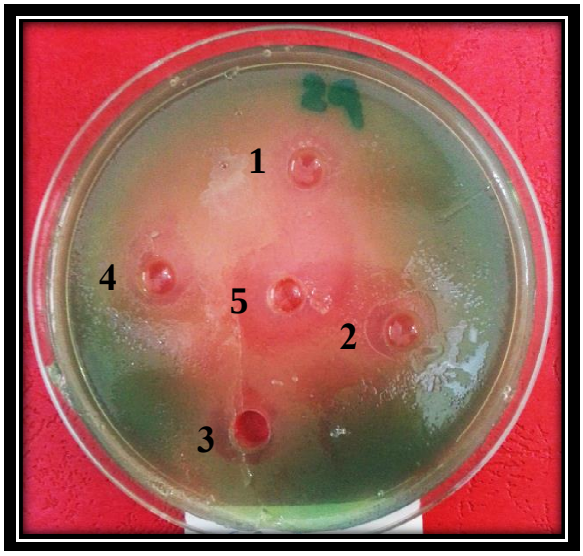
Klebsiella



Staphylococcus



E. coli



Pseudomonas

Figure (1) Antibacterial effect of different concentration of acetaminophen against *Klebsiella pneumonia, Staphylococcus aureus, E. coli, and Pseudomonas aeruginosa*(1=1*10,2=1*10^2,3=1*10^3,4=1*10^4.5=1*10^5)

Antibiotic susceptibilities

Although acetaminophen showed little effect against studied bacteria (inhibition zones ranged from (10-20 µg/ml), but it increased the susceptibilities of these bacteria towards antibiotics. Antibiotics susceptibility test showed that bacteria isolated from urinary tract infection were the most affected organism ,as it shown in table(2) figure (2&3),acetaminophen increased the susceptibilities of *E. coli* and *Staphylococcus aureus* when compared with control group (non-exposed), so results revealed that non exposed *E. coli* was resistant to vancomycin, erythromycin, and Amoxicalv (amoxilin /clavulamic acid), while the exposed bacteria were sensitive to these antibiotic, also, acetaminophen increase the susceptibilities of *E. coli* to chloramphenicol, impenemin, cefatxime and ciprofloxacin. Antibiotics susceptibility of *Klebsiella pneumonia* was highly affected when exposed to acetaminophen when compared with non-exposed bacteria which was resistant to both of erythromycin and amoxiclav, while the diameter of inhibition zone of exposed these antibiotics against exposed bacteria was 28mm for erythromycin and 22mm for amoxiclav. Also susceptibilities of this bacteria to other antibiotics was affected when exposed to acetaminophen. Antibiotics susceptibility of *Staphylococcus aureus* was highly increased when exposed to acetaminophen.

Urinary tract infection is one of the commonest bacterial infections. The Enterobacteriaceae are the most frequent pathogens detected, causing 84.3% of UTI [12,13].

Rising antibiotic resistance among uropathogens, and especially the emergence of multi-drug resistant clonal groups, has provided urgency to the development of novel preventative and therapeutic strategies[14].

Paracetamol and/or non-steroidal anti-inflammatory drugs (NSAIDS) are of use for symptomatic relief. However, recent research suggests that some NSAIDs may also have the capacity to treat UTI caused by *E. coli*[15].

The effect of acetaminophen on the susceptibility of *Pseudomonas aeruginosa* against antibiotic was less significant than urinary tract bacteria (table 2, figure 2).

Acetaminophen exposed and non-exposed *Pseudomonas aeruginosa* was resistant to six tested antibiotic except (amikacin, gentamycin, impenemin, and ciprofloxacin).

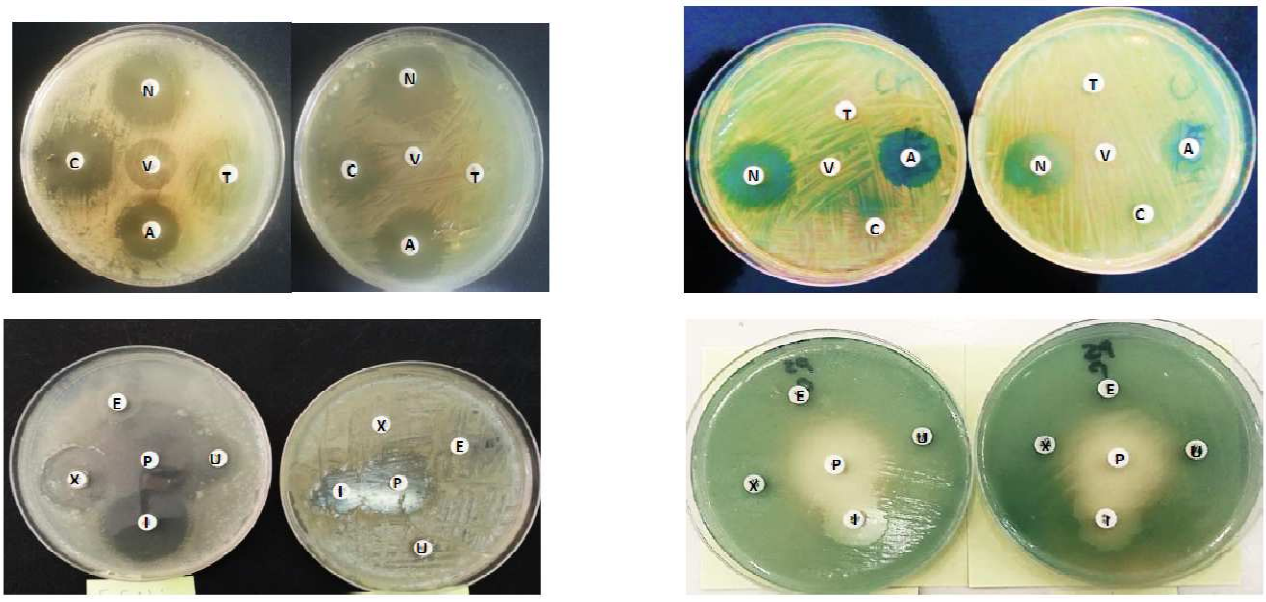
Many studies showed that *Pseudomonas aeruginosa* isolated from wound infection was multi dr*ug* resistant .*Pseudomonas aeruginosa*is naturally resistant to-lactams, including broad-spectrum cephalosporins, quinolones, chloramphenicol and tetracyclines, mainly because of the very low permeability of their cell wall. Moreover, *P. aeruginosa* is characterized by the production of inducible cephalosporinase, active efflux and poor affinity for the target (DNA gyrase)[16].

Table (2) Antibiotic susceptibilities of acetaminophen exposed bacteria

Antibiotics / Bacteria	Non exposed										Exposed									
	C*	V	A	N	T	E	U	I	X	P	C	V	A	N	T	E	U	I	X	P
Pseudomonas	0**	0	16	21	0	0	0	20	0	32	0	0	20	23	0	0	0	24	0	40
E coli	17	0	31	27	0	0	0	18	6	28	32	19	23	29	25	10	30	32	20	50
Staphylococcus	23	19	26	27	22	20	0	20	0	22	32	20	30	30	25	25	0	28	0	26
Klebsiella	25	22	30	27	20	0	0	18	10	30	36	22	32	42	36	28	22	30	16	48

**C= chloramphenicol(10 mcg),T=tetracyclin(10 mcg), V=vancomycin(10 mcg), A= amikacin(10 mcg), E=erythromycin(15 mcg), U=Amoxicalv(amoxilin/clavulamic acid)(20/10 mcg), X= cefataxime(10 mcg), N=Gentamycin(30 mcg), P= ciprofloxacin (10 mcg) and I= imipenem(10 mcg)*

*** Diameter of inhibition zone (mm)*

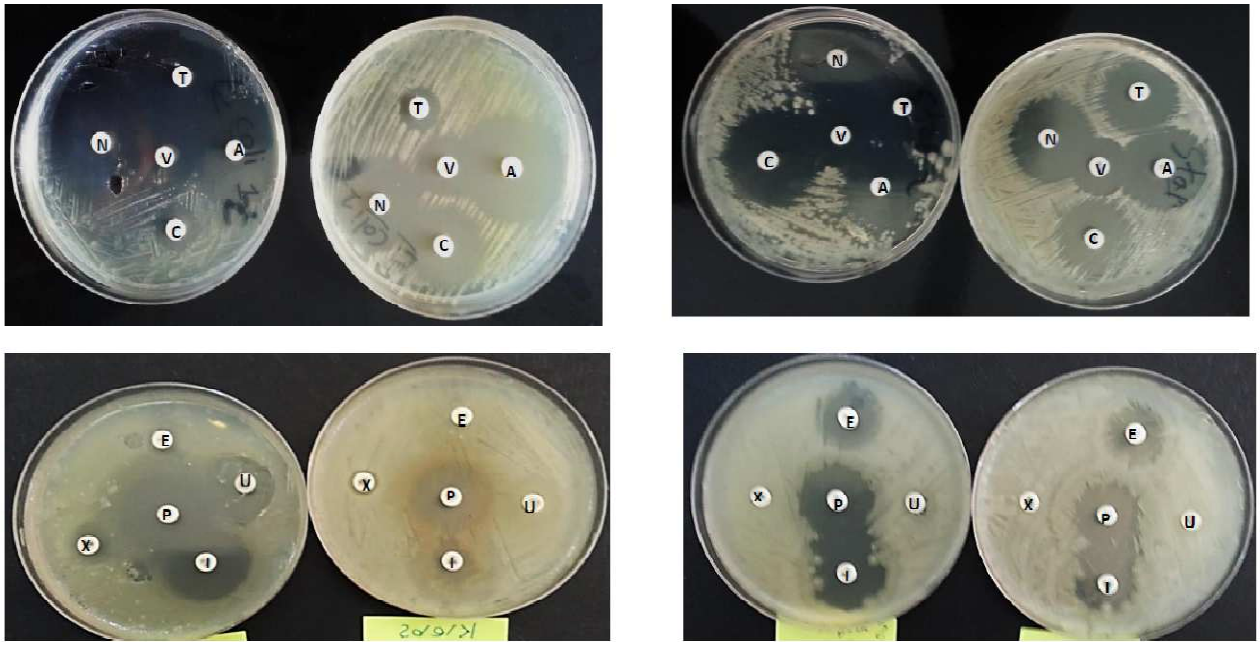


E. coli ***Pseudomonas aeruginosa***

Figure (2) Diameters of inhibition zone measurements (mm) of acetaminophen exposed *E. coli* and *Pseudomonas aeruginosa*and their sensitivity to different antibiotics

(C= chloramphenicol(10 mcg), T=tetracyclin(10 mcg), V=vancomycin(10 mcg), A= amikacin(10 mcg), E=erythromycin(15 mcg), U=Augmentin(amoxilin/clavulamic acid)(20/10 mcg), X= cefataxime(10 mcg), N=Gentamycin(30 mcg), P= ciprofloxacin (10 mcg) and I= imipenem(10 mcg)

(left Petridish= exposed, right=control)



Klebsiella pneumonia ***Staphylococcus aureus***

Figure (3) Diameters of inhibition zone measurements (mm) of acetaminophen exposed *Klebsiella pneumonia* and *Staphylococcus aureus* and their sensitivity to different antibiotics

(C= chloramphenicol(10 mcg), T=tetracyclin(10 mcg), V=vancomycin(10 mcg), A= amikacin(10 mcg), E=erythromycin(15 mcg), U=Augmentin(amoxilin/clavulamic acid)(20/10 mcg), X= cefataxime(10 mcg), N=Gentamycin(30 mcg), P= ciprofloxacin (10 mcg) and I= imipenem(10 mcg)

(left = exposed, right=control)

CONCLUSION

The present study concluded that Acetaminophen possessed antibacterial effect with increasing of the concentration. Also, susceptibilities of acetaminophen exposed bacteria towards antibiotic was affected when compared with non-exposed (control)bacteria.

REFERENCES

[1] Hinz, B.; Cheremina, O.; Brune, K. **2008,** *The FASEB journal* 22 (2): 383–390.
[2] Andersson DA, Gentry C, Alenmyr L, Killander D, Lewis SE, Andersson A, Bucher B, Galzi J-L, Sterner O, Bevan S, Högestätt ED, Zygmunt PM **2011,** *Nat Commun* 2: 551.
[3] Ohki S, Ogino N, Yamamoto S, Hayaishi O **1979,** *J. Biol. Chem.*254 (3): 829–36.
[4] Harvison PJ, Egan RW, Gale PH, Nelson SD **1986,** *Adv. Exp. Med. Biol.* 197: 739–47.
[5] Aronoff DM, Oates JA, Boutaud O **2006,** *Clin. Pharmacol. Ther.* 79 (1): 9–19.
[6] Roberts, L.J. II. & Marrow, J.D, *"Analgesic-antipyretic and Anti-inflammatory Agents and Drugs Employed in the Treatment of Gout" in, "Goodman & Gilman's The Pharmacological Basis of Therapeutics 10th Edition" by Hardman, J.G. &Limbird, L.E.* McGraw Hill, **2001** pp.687
[7] Williamson, K.L *Macroscale and Microscale Organic Chemistry Experiments*, D.C. Heath and Company, Lexington, Mass,**1989**pp. 215
[8] Bertolini A, Ferrari A, Ottani A, Guerzoni S, Tacchi R, Leone S **2006,** *CNS Drug Reviews***12** (3–4): 250–75.
[9] Viswanathan, A. N.; Feskanich, D.; Schernhammer, E. S.; Hankinson, S. E. **2008,** *Cancer Research*68(7): 2507.
[10] AL-Janabi ,A.S.(2010 *J Glob Infect Dis*. 2(2): 105–108.
[11]Lawal A, Obaleye JA. **2007**. *Biokem*;19:9-15.
[12] Wada, K. Kariyama, R. Mitsuhata, R. Uehara, S.Watanabe, T. Monden, K. and Kumon, H. ***2009,*** *Acta. Med. Okayama*. 63: 263-272
[13] Gales, AC. Jones, RN. Gordon, KA. Sader, HS. Wilke, WW. and Beach, ML**. 2000** *J. Antimicrob. Chemother.* 45: 156-163.
[14]Gupta, K., T. M. Hooton, K. G. Naber, B. Wullt, R. Colgan, L. G. Miller, G. J. Moran, L. E. Nicolle, R. Raz, A. J. Schaeffer & D. E. Soper, **2011,** *Clin Infect Dis* 52: 103-120.
[15] Mazumdar K, Dutta NK, Dastidar SG, **2006,** *In Vivo*. 20(5):613-9.

[16]Li, X.Z., D.M. Livermore and H. Nikaido, **1994,** *Antimicrob Agents Chemother*, 38(8): 1732-1741.