Journal of Pharmaceutical and Applied Chemistry
An International Journal

http://dx.doi.org/10.18576/jpac/030110

Spectrophotometric Study and Determination of Ibuprofen and Lornoxicam Drugs via Their Reaction with Copper (II) Reagent and Their Biological Activities

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Received: 5 Sep. 2016, Revised: 11 Nov. 2016, Accepted: 13 Nov. 2016.
Published online: 1 Jan. 2017.

Abstract: The reactions of Ibuprofen (**IBU**) and Lornoxicam (**LOR**) drugs with copper tetramine and copper sulphate reagents respectively were studied spectrophotometrically at optimum conditions of, reactant concentration, temperature, time, λ_{max} and pH. Applying of molar ratio method (MRM) on the Ibuprofen and Lornoxicam copper ion-pairs, indicated (1:1) stoichiometric ratio. In addition, validity of Beer's Law was carried out at 265 and 260 nm, for Ibuprofen and Lornoxicam products, respectively. Results of analysis were statistically validated. The linearity ranged from 2.063 to 103.14 and 3.72 to 74.36 µg ml^{-1} of recovery values 99.95 to 100.40 and 99.98 to 100.00 %, for **IBU** and **LOR** respectively. The low values of standard deviation (SD = 0.009 to 0.533 and 0.059 to 0.844) and relative standard deviation (RSD = 0.037 to 0.577 and 0.063 to 0.635 %) refer to the accuracy and precision of the proposed method for spectrophotometric micro-determination of these drugs in pure and in their pharmaceutical formulations in comparison with official methods. The robustness and procedure validation were assayed in intra and inter days. The solid ion-pairs of these drugs with copper have been separated and identified by elemental analyses and FT-IR spectroscopic tool and they were found to be biologically active toward some kinds of Tribolium confusum common insect species in flour mills and treated areas and their adults. These ion-pairs were found to be more biologically active than their drugs.

Keywords: Ibuprofen, Lornoxicam, Spectrophotometry, Pharmaceutical formulations, Solid copper ion-pairs, Biological activities.

1 Introduction

Spectrophotometry is the most common analytical technique used for analysis and determination of drugs and / or pharmaceutical formulations. Non – steroidal anti – inflammatory drugs (NSAIDs) are drugs with analgesic, antipyretic and anti – inflammatory effects. Ibuprofen (**IBU**) and Lornoxicam (**LOR**) are most famous members of this group of drugs [1, 2]. Ibuprofen is chemically 2[4-(2-methyl propyl) phenyl] propanoic acid. The molecular formula is $C_{13}H_{18}O_2$, and molecular weight is 206.28 [3, 4]. The most recent methods for determination of ibuprofen included chromatographic [5-8], electrochemical [9,10] and spectrophotometric techniques [11-16].

Lornoxicam (**LOR**) is (3*E*)-6- chloro -3-[hydroxyl (pyridine-2-ylamino) methylene]-2- methyl-2,3- dihydro -4*H*-thieno[2,3-*e*] [1,2] thiazin -4-one-1,1-dioxide. The molecular formula is $C_{13}H_{10}ClN_3O_4S_2$ and molecular weight is 371.82 [17]. In the literatures, few analytical methods for determination of Lornoxicam using UV Spectroscopy, HPLC and polarography in plasma and pharmaceutical formulation have been reported [18–22].

The fundamental role of copper and the recognition of its complexes as important bioactive compounds *in vitro* and in *vivo* aroused an ever-increasing interest on these agents as potential drugs for therapeutic intervention in various diseases. The vast array of information available for their bio-inorganic properties and mode of action in several biological systems, are creating an exciting scenario for the development of a novel generation of highly active drugs with minimized side-effects which could add significantly to the current clinical research and practice [23]. Current interest in Cu complexes is stemming from their potential use as antimicrobial, antiviral, anti-inflammatory and antitumor agents, enzyme inhibitors or chemical nucleases. Markedly, the biochemical action of Cu complexes with Non-Steroidal Anti-Inflammatory Drugs (NSAIDs) has been studied [24]. Numerous Cu(II) complexes of NSAIDs

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showing enhanced anti-inflammatory and anti-ulcerogenic activity, as well as reduced gastrointestinal toxicity compared to the un-complexed drug, have been prepared and structurally characterized [24].

Therefore; the aim of the present study is to study the nature of reaction of copper (II) with analgesic, antipyretic and anti – inflammatory to develop a new simple, rapid, reliable and precise UV spectrophotometric method for analysis of these drugs and their formulations. Also, spectroscopic study aimed to shed light on the nature of the formed complexes both in solution and in solid state and their biological activities.

2 Experimental

2.1 Materials

All reagents of analytical grade and double distilled water were used without further purification throughout the research. Authentic samples of Ibuprofen and Lornoxicam drugs supplied by Arab Drug Co. (Egypt) were used. Tablets of Ibuprofen (Brufen Tablets) and Lornoxicam (Xefo Tablets) specified as 200 mg /tablet and 8 mg / tablet respectively were produced by Sedico (Egypt). Ammonia solution (4 % v/v), phosphoric acid, acetic acid, boric acid were supplied by BDH respectively. Also, anhydrous sodium carbonate and sodium hydroxide (AR) were supplied by BDH.

2.2 Solutions

$5x10^{-2}$ M solution of anhydrous sodium carbonate was prepared by dissolving accurate weight in distilled water by using 250 mL volumetric flask. 10^{-3} M stock solution of copper sulphate reagent was prepared by dissolving accurate weight in double distilled water by using 100 mL volumetric flask. Copper tetramine reagent was prepared and tested. Universal buffer solutions were prepared as recommended by Briton and Robinson [25].

2.2.1 Standard stock solutions

10^{-3}M (206 µg. ml^{-1} , 372 µg. ml^{-1}) of standard stock solutions of pure Ibuprofen and Lornoxicam drugs were prepared separately in two 100 mL volumetric flasks by dissolving accurate weights 0.0206 g and 0.0372 g respectively in sufficient amount of 0.05 M Na_2CO_3 solution and the volume of each solution was completed to the mark with double distilled water.

2.2.2 Sample solutions

For pharmaceutical preparation, ten tablets specified as (400 mg /tablet) and (8 mg /tablet), for **IBU** and **LOR**, respectively, were weighed and powdered separately. Equivalent amount of powder to one tablet of each separate drug was weighed and dissolved in sufficient amount of 0.05 M Na_2CO_3 solution. The solution of each drug was transferred into separate 50 mL volumetric flasks and the volume completed to the mark with double distilled water.

2.3 Procedures

Spectrophotometric study of Ibuprofen (**IBU**) and Lornoxicam (**LOR**) drugs with copper tetramine and copper sulphate reagents respectively, is involved for selection of proper conditions, such as λ_{max}, pH, temperature, molar ratio method (MRM) and abeyance to Beer's Law. The selected proper parameter is that gives the highest molar absorptivity (ε, L mol^{-1} cm^{-1}).

2.3.1 Preparation of working standard solution and construction of calibration curves

Working solutions with different concentrations were prepared by pipetting 0.1 – 5 ml of pure standard Ibuprofen drug solution into separate 10 ml volumetric flasks, 5 ml of 10^{-3} M copper tetramine reagent was added and each volume completed to the mark with double distilled water. Also, solution with different concentrations were prepared by pipetting 0.1 – 1.9 ml of pure standard Lornoxicam drug solution into separate 10 ml volumetric flasks, 2 ml of 10^{-3} M copper sulphate reagent was added and each volume completed to the mark with distilled water. Spectrophotometric measurements were recorded at 265 and 260 nm respectively. The calibration curves were constructed and molar ratio method (MRM) [26] was applied.

Solutions of 10, 30, 70 and 14.87, 29.74, 66.92 µg. ml^{-1} of Ibuprofen and Lornoxicam drug solutions respectively were used for microdetermination of pharmaceutical formulations with the same manner of standard drugs. Spectrophotometric measurements were studied within (intra) and in between (inter) days in order to investigate the validation and robustness of applied methods.

2.4 Preparation of solid ion-pairs

The solid ion-pairs of Ibuprofen and Lornoxicam drugs with copper (II) were prepared by addition of a solution of appropriate weight of metal salt of 0.125 g (0.5 mmol) copper sulphate penta hydrate in 50 ml water to a 50 ml solution of 0.103 g (0.499 mmol) **IBU** and 0.186 g (0.5 mmol) **LOR** , respectively. Appropriate weight of each drug dissolved in 2 ml of 0.05 M Na_2CO_3 and bidistelled water. The resulted solid ion-pairs appeared as colored precipitates. The precipitates leaved for 10 mins until completely settled. The obtained solid complexes were separated, filtered and washed with suitable solvent using a Hearch funnel of suitable pores. Moreover, the obtained solid ion-pairs were dried in vacuum desiccator. The yield of each solid ion-pair was calculated. The physical properties of these ion-pairs were studied (color, mp, solubility, etc.). Elemental analyses (C, H, and N) and FT-IR were made at the Microanalytical Center of Cairo

University. The performed analyses aiming to elucidate the structures of prepared ion-pairs.

2.4.1 Elemental Analyses and Spectroscopic studies

a- Determination of the metal content of the prepared ion-pairs

Accurately weighed portion (0.0501, 0.0247g respectively) of the prepared ion-pairs was placed in two separate Kjeldahl flasks. A mixture of concentrated nitric and hydrochloric acids (aquaregia, 1:3) added to powdered complex with gradual heating. After evaporation of each mixture near dryness and complete digestion, the remained solutions had faint blue color. Each solution was then diluted to a 10 mL with bidistelled water and the copper content was determined by titration of 1 mL of each solution against 0.01M standard EDTA solution, using Murexide indicator by recommended procedure [27]. The molecular weights of the given ion-pairs were calculated from its copper content titrimetrically determined, using the equation (1):

$$W = M \times V \times M.Wt. / 1000 \quad (1)$$

Where W = weight of solid digested ion-pair, M = the obtained molarity of Cu (II) , V = 10 ml of ion-pair solution, M.Wt. = molecular weight of digested ion-pair.

b- The other elements like C, H, N, X, and S were analyzed and determined in Micro analytical center of Cairo University.

2.4.2. Biological Activity of drugs and their complexes

a- Insects and commodity

Adult of *Tribolium confusum* were laboratory reared on wheat flour at 27.5 ±1.5ºC and 70% ± 5% (R.H.) according to the method of Frederic *et al*.[28] with some modifications.

***b- Bioassay and statistical analysis*:**

T. confusum adult was topically treated with 10µ of each compound according to the protocol described by Delobel *et al*.[29] as follows: Thirty insects divided on three replicates (10 adult/replicate) were topically and mortality was then monitored after 24 hr. Thirty adults of control experiment were used in three replicates without treatment. The adult mortality was estimated according to Abbot [30]. Estimation of LD50 values was made using Finney analysis [31].

2.5. Instruments

Spectrophotometer, spectronic model 601-Milton Roy (USA), UV – Vis with matched quartz cell of 1 cm optical length was used for spectrophotometric measurements in the wavelength range of 200 – 800 nm. Automatic micropipette (10 – 100 µL) model Accupipette USA, was used to measure the very small volumes, Glass micro pipettes were used to measure the large volumes, Sensitive analytical balance [0.0001g, SCALTEC (Germany)], pH/ mV-meter Model 701 A/digital ion analyzer and Magnetic stirrer theromostated hot plate (VELP-Europe). Infrared spectra were recorded on a Perkin-Elmer FT-IR type 1650 spectrophotometer in the region 4000 - 400 cm^{-1} as KBr discs.

3 Results and Discussion

The drugs under this study are given in Fig 1a for IBU and Fig b for LOR;

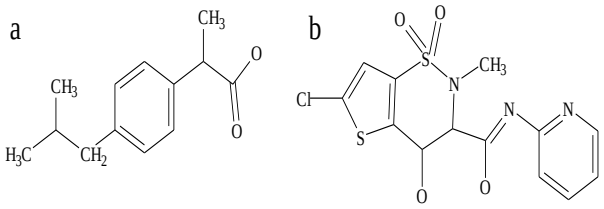

Fig. 1: a- Structure formula of Ibuprofen (**IBU**), b- Structure formula of Lornoxicam (**LOR**).

3.1 Selection of suitable wavelength

The spectra of Ibuprofen (**IBU**), copper (II) tetramine (R_1) and that of **IBU** – Cu (II) product show that **IBU** λ_{max} = 220 nm, R_1 has λ_{max} = 615 nm and **IBU** - R_1 has a λ_{max} = 265 nm. Therefore, the selected wavelength to do further study for the reaction of **IBU** and R_1 is that of the product (265 nm) occurs faraway from that of both drug and reagent.

The spectra of Lornoxicam (**LOR**), copper (II) sulphate (R_2) and that of **LOR**-R_2 product show that LOR λ_{max} = 375 nm, R_2 has λ_{max} = 810 nm and **LOR**- R_2 has a λ_{max} = 260 nm. Therefore, the selected wavelength to do further study for the reaction of **LOR** and R_2 is that of the product (260 nm) occurs faraway from that of both drug and reagent.

3.2. Effect of pH on spectra of drugs and reaction products

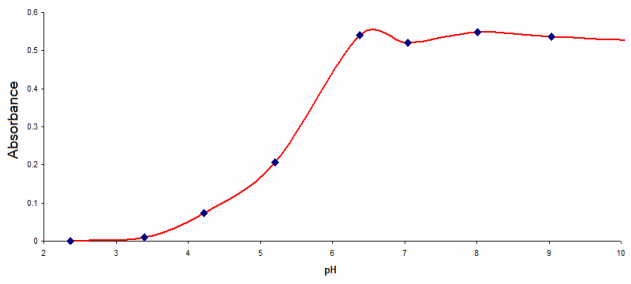

Fig. 2: Effect of pH on the spectrum of 2.5 x 10^{-4} M IBU-R_1 complex.

Table1. Analytical parameters used for spectrophotometric determination of pure **IBU** and **LOR** drugs by the proposed method.

Parameters	drugs	
	IBU	**LOR**
Reagent	Copper tetramine	Copper sulphate
Temperature (°C)	45	30
λ_{max} (nm)	265	260
pH	6.5	5.0
Beer's law ($\mu g.ml^{-1}$)	2.06 – 103.14	3.72 – 74.36
LDL ($\mu g.ml^{-1}$)	2.04	3.35
HDL ($\mu g.ml^{-1}$)	103.16	74.70
R^2	0.995	0.999
Regression equation (* Y)	Y = 0.0965 + 0.4	Y = 0.823x + 0.163
Molar absorptivity ($mol^{-1}\ cm^{-1}$)	1.18×10^4	9.54×10^3
SD	0.009 – 0.533	0.059 – 0.844
RSD %	0.037 – 0.577	0.063 – 0.636
Sandell sensitivity ($\mu g.cm^{-2}$)	0.017	0.035
Recovery %	99.95 – 100.40	99.98 – 100.00

Table 2 Intra – day spectrophotometric micro-determination of pure drug by proposed method

drug	[wt] taken ($\mu g.ml^{-1}$)	[wt] found ($\mu g.ml^{-1}$) ± SD	Recovery (%)	SD[a]	RSD(%)[a]
IBU	10.31	10.31± 0.0017	99.98-100.02	0.0017	0.0165
	41.26	41.26± 0.0239	99.94-100.06	0.0239	0.0579
	72.198	72.198 ± 0.0261	99.96-100.04	0.0261	0.0362
LOR	3.718	3.718± 0.0094	99.75-100.25	0.0094	0.2528
	37.18	37.18±0.0203	99.95-100.05	0.0203	0.0546
	70.65	70.65± 0.0345	99.95-100.05	0.0345	0.0488

Table 3 Inter – day spectrophotometric micro-determination of pure drugs by proposed method

drug	[wt] taken ($\mu g.ml^{-1}$)	[wt] found ($\mu g.ml^{-1}$) ± SD	Recovery (%)	SD[a]	RSD(%)[a]
IBU	10.31	10.31 ±0.0099	99.90-100.09	0.0099	0.0959
	41.26	41.26 ± 0.0195	99.95-100.05	0.0195	0.0473
	72.20	72.20 ± 0.0482	99.93-100.07	0.0482	0.0668
LOR	3.718	3.718 ± 0.0239	99.36-100.64	0.0239	0.6428
	37.18	37.18 ± 0.0559	99.85-100.15	0.0559	0.1503
	70.65	70.65 ± 0.0857	99.88-100.12	0.0857	0.1213

[a] Mean values for six replicates at 6 days.

Table 4 Spectrophotometric micro determination of **IBU** drug in different pharmaceutical formulations by proposed and official methods

Sample	Proposed		Official[4, 24]		% Recovery		[a]SD	
	[Drug][1]	µg mL^{-1}	[Drug]	µg mL^{-1}				
	Taken	Found	Taken	Found	Proposed	Official (4 ,31)	Proposed	Official [4,31]
IBU in Brufen Tablet (200 or 400 mg/Tablet)	10	10	*50	*49.59	100.00	*99.18	0.0162	*0.88
	30	29.91	**400	**393.48	99.70	**98.37	0.0359	**1.47
	70	69.99			99.99		0.075	

*British Pharmacopoeia (2002).
** Indian pharmacopoeia (1996). [a]SD values for five replicate

From the pH data in Figure (2), it is clear that the suitable pH is 5.0-6.5 for studying **IBU**-Cu (II) and **LOR**-Cu (II) ion-pairs respectively.

3.3 Effect of Temperature on the drug reaction products

From the data obtained, it is found that, the suitable temp for **IUB**-R_1 and **LOR** –R_2 ion-pairs are 45 and 47°C respectively.

3.4 Stoichiometry of reagents and drugs

The stoichiometric ratio of each drug with described

reagent was determined by the molar ratio method [26]; the results referred to the formation of 1:1 [R]: [drug] ion-pairs/or metal complexes. These proposed structures (Fig 3) need more evidences after separation and structure identification of solid reaction products.

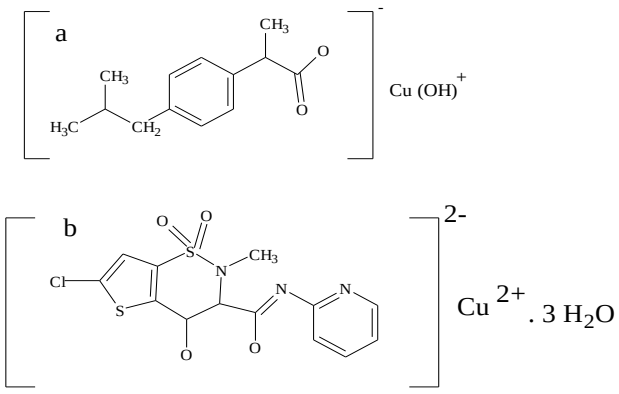

Fig. 3. A- Proposed structure of **IBU-Cu (II)** ion pair**, B-** Proposed structure of **LOR-Cu (II)** ion pair

Therefore the selected proper conditions used for micro-determination of **IBU** and **LOR** drugs in pure and in pharmaceutical formulations using copper (II) reagents are λ_{max} = 265 and 260 nm, Time = 15 and 5 min, Temp. = 45 and 47ºC, pH = 6.0 and 5.0 and stoichiometric ratio of reactants = 1:1.

In order to prove the validity and applicability of the proposed methods and reproducibility of the results, five replicate experiments, at five concentrations of standard Ibuprofen (**IBU**), Lornoxicam (**LOR**) and their pharmaceutical formulations, were carried out under proper selected conditions (λ_{max}, pH, Temp, and time). The obtained analytical parameters for the micro – determination including molar absorptivity, Sandell sensitivity (S), standard and relative standard deviations, LDL and HDL and recovery % for each drug are given in Table 1**.**

The obtained results of analytical parameters for spectrophotometric micro – determination of pure **IBU** and **LOR** drugs show that the recovery values were in the ranges of 99.95 – 100.40 %, 99.98 – 100.10 % for **IBU** and **LOR** drugs, respectively. The Sandell sensitivity (S) was found to be 0.017 and 0.035 µg.cm$^{-2,}$ respectively. The LDL and HDL were found to be 2.04 – 103.16 and 3.35 – 74.70 µg.ml^{-1} for **IUB** and **LOR** drugs, respectively. The SD values were in the ranges of 0.009 – 0.533, 0.059 – 0.844, respectively. In addition to, the RSD values were in the ranges of 0.037 – 0.577 %, 0.063 – 0.636 %, respectively. These values refer to the accuracy and precession of the suggested procedures in micro-determination of both drugs in pure states.

3.5 Validity of Beer's Law

Under the optimum conditions Beer's Law is valid in the concentration ranges of 2.06 – 103.14 and 3.72 – 74.36 µg. ml^{-1} for IBU and LOR respectively. Above these limits, negative deviations were observed. The correlation coefficient values were found to be 0.995 and 0.999, for **IBU** and **LOR** drug products respectively. Therefore, these standard calibration curves can be used for micro-determination of these drugs within and in between days in order to assay validity and robustness of the suggested procedures.

3.6 Intra and inter- day measurements

Table 2 shows the obtained values of intra – day determination for different concentrations of the pure drugs by proposed method for five replicates of each drug concentration.

It is found that the recovery values were in the ranges of 99.94 – 100.06 and 99.75 – 100.25 % for **IBU** and **LOR** drugs, respectively. The SD values were in the ranges of 0.0017 – 0.0261 and 0.0099 – 0.0345, respectively. Also, the RSD values were in the ranges of 0.0165 – 0.0579 and 0.0488 – 0.252%, respectively. These values refer to the high accuracy, reliability and precision of the applied procedures.

Table 3 shows the obtained values of standard and relative standard deviations of inter – day measurements of pure drugs by proposed method for five replicates of each drug concentration.

It is found that the recovery values were in the ranges of 99.93 – 100.09 and 99.36 – 100.64 % for **IBU** and **LOR** drugs, respectively. The SD values were in the ranges of 0.0099 – 0.0482 and 0.0239 – 0.0857, respectively. Also, the RSD values were in the ranges of 0.0473 – 0.0959 and 0.1213 – 0.6428 %, respectively. These data refer to the accuracy, validity and robustness of the suggested procedures.

Tables (4 and 5) show the results obtained for the spectrophotometric determination of **IBU** and **LOR** drugs in pharmaceutical formulations by the proposed and official methods [4, 32].

The recovery values of **IBU** obtained by proposed method were ranged between 99.83 – 100.17 %, but for **IBU** obtained by reported official method were 98.37 and 99.18 %. Also, SD of **IBU** obtained by proposed method was ranged between 0.0171 – 0.0358, but SD for **IBU** obtained by reported official method was 0.88 and 1.47 [4, 32].

On the other hand, the obtained results by proposed method for **LOR** determination, as in Table 5, it is found that the recovery values were between 99.82 – 100.18 % and SD values were between 0.0275 – 0.0683. But the recovery value obtained by reported official method [33, 34] was 100.71 %. This study reveals that the proposed method for determination of **IBU** and **LOR** drugs are simple, selective and sensitive with reasonable precision and accuracy more or less on the same level like official ones. Moreover, the proposed method can be used as alternative method for the

Table 5 Spectrophotometric microdetermination of **LOR** drug in different pharmaceutical formulations by proposed and official methods

Sample	Proposed		Official (32,33)		% Recovery		[a]SD	
	[Drug] µg mL^{-1}		[Drug] µg mL^{-1}					
	Taken	Found	Taken	Found	Proposed	Official	Proposed	Official[32,33]
LOR in Xefo	7.4364	7.4421	4.00	4.02	100.08	100.50	0.0238	0.008
Tablets	29.7456	29.7512			99.69		0.1193	
(4 or 8 mg/Tablet)	66.927	66.9826	8.00	7.95	100.08	99.38	0.0575	0.011

[a]SD values for five replicates.

Table 6 Analytical and physical data copper (II)-Drugs ion-pairs

Complex	Color (Yield, %)	m.p (°C)	Found(calcd)					
			C	H	N	S	Cl	M, M.Wt?
IBU-Cu (II) ($CuC_{13}H_{17}O_2.H_2O$), mole mass = 286.84	Blue (92.39)	230 - 235	54.77 (54.39)	6.4 (6.97)	—	—	—	21.76 (22.16)
LOR-Cu(II) ($Cu\mathbf{C}_{13}H_{10}ClN_3O_4S_2.3H_2O$), mole mass = 489.37	Yellowish-Green (88.08)	220 - 225	31.67 (31.88)	2.68 (3.27)	7.22 (8.58)	7.05 (13.08)	8.69 (7.25)	12.87 (12.99)

M = Cu (II), L_1 = IBU anion (ligand= $C_{13}H_{17}O_2$, mole mass = 206.29 g/mol),
L_2 = LOR anion (ligand = $C_{13}H_{10}ClN_3O_4S_2$, mole mass = 371.8192 g/mol).

Table 7. Effect of **IBU** and **LOR** drugs and of copper-drug ion-pairs on the Tribolium confusum insects

Drugs compounds		
Conc. %	IBU	LOR
10	8	21
30	20	40
50	41	73
LD_{50}	55	35
Control	00	00
. Copper-drug complexes		
Conc. %	Cu (II)-IBU	Cu (II)-LOR
10	21	3
30	50	20
50	86	41
Control	00	00

routine determination of **IBU** and **LOR** drugs in pure and in their pharmaceutical formulations.

3.7 Structure investigation of solid ion-pairs

The separated solid copper ion-pairs were analyzed by elemental analyses and found to have general formulae of **IBU**-Cu (II) ($CuC_{13}H_{18}O_2.HO$) and that of **LOR**-Cu(II) ($CuC_{13}H_{10}ClN_3O_4S_2.3H_2O$) . The data obtained are given in Table (6).

The structural formulae of these ion-pairs had been studied by comparison of FT-IR of the drugs and of these compounds. The FT-IR of **IBU** refers to the bands of $\nu OH_{stretch}$ (at 3092-2870cm^{-1}), νOH_{bend} (at 2729-2543 cm^{-1}), $\nu C\text{-}O_{bend}$ (at 588-421 cm^{-1}) of νCOO (at 937-747 cm^{-1}) and of νbenzene ring (at 1508-1008 cm^{-1}). These bands are shifted to higher values of wave numbers in the corresponding **IBU**-Cu complex [$\nu OH_{stretch}$ (at 3422 cm^{-1}), νOH_{bend} (at 3955-2870 cm^{-1}), $\nu C\text{-}O_{bend}$ (at 853-549 cm^{-1}) of νCOO (at 1513-1294 cm^{-1})] except that of benzene ring is shifted to lower values of wave numbers [νbenzene ring (at 11118-1070 cm^{-1})]. These data means that carboxylic strongly shared in formation of **IBU**-Cu compounds which leads to lowering of the electron density over the benzene ring.

There is another peak appeared at 62-588 cm^{-1} which may be attributed to the νCu-O bond. These data confirm the proposed structure of **IBU**-Cu in Fig (3a). The **LOR** drug FT-IR shows νOH (at 3200-3600 cm^{-1}), νNH_{amide} (at 3350-3500 cm^{-1}), νC=O (amide) (at 1620-1680 cm^{-1}), νC-O (amide) (at 1025-1200 cm^{-1}) and νCONH (at 1680-1700

cm^{-1}). In case of FT-IR of **LOR**-Cu; these bands are shifted the higher values of wave numbers [νOH (at 3450- 3650 cm^{-1}), νNH_{amide} (at 3371-3400 cm^{-1}), νC-O (amide) (at 1155-1191 cm^{-1}) and some of them are shifted to lower wave numbers such as νC=O(amide) (at 1572-1628 cm^{-1}),and νCONH (at 1628-1670 cm^{-1}). This confirms the sharing of both amide group in enol form and OH of heterocyclic ring in formation **LOR**-Cu as given by the proposed structure in Fig (3b).

The pyridyl group of **LOR** drug shows ν values at 1581, 1030, 991and 604 cm^{-1} due to its different modes of vibrations. These modes of vibrations are shifted to lower values of wave number (at 963-786 cm^{-1}). This means that the electron cloud around pyridyl group withdrawn away due to sharing of amide group of **LOR** in complex formation. The existence of water of hydration and/or water of coordination in the spectra of the complexes render it difficult to get conclusion from the OH group of the drugs, which will be overlapped by those of the water molecules. From these data it is clear that, carboxylic group of **IBU** ligand and OH groups in an enol form of **LOR** shared in complex formation and other functional groups are affected by this sharing. Copper ion is also complete its coordination sphere with water molecules and/or OH forming drugs basic salts [35]. The OH anion is coming from the basic carbonate solutions of used drugs in preparation of these compounds.

3.8 Biological activities of drugs in comparison with their copper compounds

The biological activities of Ibuprofen, Lornoxicam drugs and their copper ion-pairs were determined according to the protocol described by Delobel et al. [28]. *Tribolium confusum* is the most common and destructive insect species in flour mills and treated areas. Adult of *Tribolium confusum* were laboratory reared on wheat flour at 27 ± 1.5 ^{o}C and 70% ± 5% (R.H) according to the method of Frederic et al. [29] with some modifications.

T. *confusum* adult was topically treated with 10 µg of each compound (**IBU**, **LOR**, **IBU**-Cu and **LOR**-Cu) according to Delobel et al. [28] protocol as follows: Thirty insects divided on three replicates without treatment. The adult mortality was estimated according to Abbot [31]. Estimation of LD_{50} values was made using probit analysis by Finney [32]. The data obtained are shown in Table (7).

From these data it is found that, **LOR** drug was the most biologically effective on *T. confusum* which caused 21, 40, and 73% mortalities after adult treatments with the concentration of 10, 30 and 50 of **LOR** comparison to no effect on the control. On the other hand, IBU drug showed least mortality (8, 20, and 41%) on *T. confusum* with the same concentrations. The order of toxicity (LD_{50}) values was found 55 and 35 % of **IBU** and **LOR** drugs, respectively. On the other hand, the solid prepared copper-drug complexes, showed that LOR-Cu complex was most biologically active than **IBU**-Cu ion-pair. It is also concluded that the presence of cupric ions in moiety of these ion-pairs enhanced biological activities of these drugs [23, 24]. This may be attributed to copper essential biological activity. The enhanced biological activity of both **LOR** drug and its copper ion-pair may be attributed to the extra bioactivity of sulpher atoms in entity of this drug and its product.

5 Conclusions

The proposed method could be applied efficiently for spectrophotometric microdetermination of the drugs under investigation in pure and / or dosage forms. Also, the proposed method requires less time for analysis, provide better RSD, LDL, HDL and high recovery percentages. Moreover, the proposed method is simple, low cost and could be easily used in a quality control laboratory for their analysis. The solid ion-pairs of these drugs with copper have been separated and identified by elemental analyse and FT-IR spectroscopic tool and they were found to be biologically active toward some kinds of Tribolium confusum common insect species in flour mills and treated areas and their adults. These ion-pairs were found to be more biologically active than their drugs. Their extra biological activities may be due to the presence of the biologically active copper (II) in moiety of these drugs ion-pairs.

Acknowledgement

The authors would like to present their grateful thanks to the Chemistry Department and Faculty of Science for the technical and chemical supports for this research to achieve its objectives.

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