THE SYNTHESIS AND EVALUATION OF COMBINE DRUG ACTION OF CODEINE AS SUPPLEMENTARY DRUG WITH DAKIN-WEST B-ACETAMIDOKETONES

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Abstract:
β-acetamido ketones were synthesized in high yields under mild condition via four component Dakin-West reaction which carried out by condensation of aromatic aldehydes, enolizable ketones, acetonitrile and acetyl chloride in presence of mixed metal oxide CuO-SnO_2 as versatile catalyst. The biological study of compounds codeine was used as supplementary drug in combination of the resultant Dakin-West compounds. Interesting results were obtained in accordance with the principle of 'Synergism'. It was observed that, synthesized β-acetamidoketones and supplementary drug, Codeine worked together to produced results which were not independently obtainable.

Keywords: Combine drug action, Dakin-West reaction, Synergism, Supplementary drug, Codeine.

Introduction

The combination of two or more drugs implies to either synergism or antagonism [1-5].Synergism and antagonism corresponds to higher and lower antimicrobial activity than would be expected from the individual activity due to simple addition effect.[6] The physical interaction in these regards does not commonly but occasionally causes contact between the different drug under consideration[7]. More roughly saying, the activity of drug moiety X is affected by drug Y due to molecular interaction of drug with biological system and various microbial, physiological and biochemical effect of such interaction[8]. There for one can say that through the said study the observation about the individual drug and its effect on cellular system would be documented.

In order to fallows mechanistic study on antimicrobial drug such interaction in vitro and in culture cell medium are commonly used. In order to controlling infectious dieses the combine drug therapy experiment can be carried out with antimicrobial agent.[9,10] In order to avoid failure of single microbial agent more molecules can be used in combination. The positive result may also be obtained by using such kind of combine drug therapy as its lead to reduction in doses of drugs, maximise the antimicrobial effect and quenches the side effect on the other hand if the combination of two drug minimise the effect of one another corresponds to base of the concept antagonism, and is not generally adapted. [11,12] Hence it becomes necessary to know the pattern of interaction in between two or more drug before the choice of treatment.

Synergy, in general, may be defined as two or more things functioning together to produce a result not independently obtainable [13]. The term *synergy* comes from the Greek word *syn-ergos*, meaning "working together". In

the context of organizational behaviour, following the view that a cohesive group is more than the sum of its parts, synergy is the ability of a group to outperform even its best individual member. These conclusions are derived from the studies conducted by Jay Hall on a number of laboratory-based group ranking and prediction tasks [14]. Synergy is literally everywhere around us, and within us; it is unavoidable. Here are just examples: About 2,000 separate enzymes are required to catalyze a metabolic web. But if remove one of the more critical of these enzymes, say the hexokinase that facilitates glycolysis, the process would not go forward. Water has a unique set of emergent, combinatorial properties that are radically different from those of its two constituent gases. But simple mixing of the two gases together without a catalyst like platinum, synergism will not be reflected. Synergy has been advanced by Robert Corning as a hypothesis on how complex systems operate. Environmental systems may react in a non-linear way to perturbations, such as climate change, so that the outcome may be greater than the sum of the individual component alterations. Synergistic responses are a complicating factor in environmental modelling [15]. Pest synergy would occur in a biological host organism population where, for example, the introduction of 'Parasite A' may cause 10% fatalities, and 'parasite B' may also cause 10% loss. When both parasites are present, the losses would normally be expected to total less than 20%, yet in some cases, losses are Significantly greater. In such cases it is said that the parasites in combination have a synergistic effect.[16]

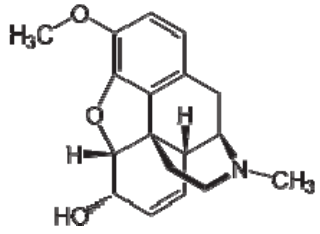


Fig. Codeine

Drug synergy can occur in biological activity and because of pharmacokinetics. Shared metabolic enzymes can cause drugs to remain in the bloodstream much longer in higher concentrations than if individually taken.[17]

At present period, many researchers are moving towards the preparation, characterization and biological screening of β-acetamidoketone compounds.[18-23]

From the literature survey, it was revealed that, no work is carried out regarding the study of synergism by using various biologically active compounds synthesized by multicomponent reactions; in particular the combine drug action study of β-acetamidoketone and Codeine is thruster area.[24] By taking this fact into consideration, very recently we have done the synthetic and biological evaluation of industrially important chemical moieties through green protocol. In continuation of these efforts, our research group had decided to evaluate the synergistic combine drug action of β-acetamidoketone compounds and Codeine (as supplementary drug) by using the pathogenic Gram positive and Gram negative micro organisms namely; *Staphylococcus aureus*, *Escherichia coli, Shigella dysentariae, Klebsiella pneumoniae, Proteus mirabilis, Salmonella typhi, Bacillus subtilis, Bacillus megatherium, Proteus vulgaris* and *Pseudomonas aeruginosa.*

II.EXPERIMENTAL

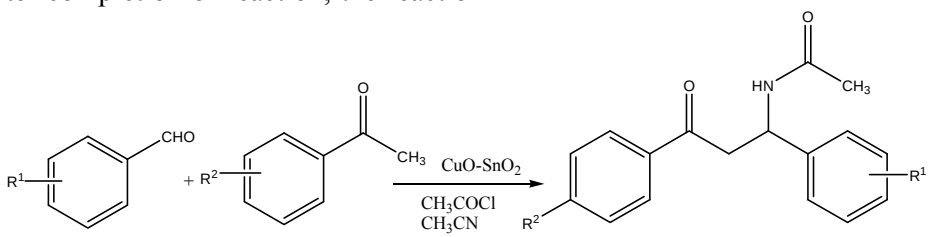
a) General:

All commercially available chemicals and reagents were purchased from Aldrich and used without further purification. The melting points of all the synthesized compounds were recorded in precision digital melting point apparatus, Model MP-D and are uncorrected. The IR spectra of the synthesized compounds were recorded on Nicolet Instruments Corporation, USA make MAGNA 550 spectrometer. The PMR spectra were recorded on Varian, USA make Mercury plus-300 MHz NMR spectrometer. The GC-MS analysis of synthesized compounds was performed on Hewlett Packard make GCD-1800A EI source analyzer at Sophisticated Analytical Instrument Facility (SAIF), IIT Bombay, Powai, Mumbai, India.

b) Typical Experimental Procedure For The Preparation of *β*-acetamido ketones(I_a-I_h)

In typical synthesis of β-acetamido ketones, mixture of aromatic aldehyde (10 mmol), enolizable ketone (10 mmol), acetyl chloride (10 mmol) and acetonitrile (10 mmol) was well

stirred (Scheme :1)in presence of $CuO\text{-}SnO_2$ for the appropriate time (as mentioned in Table-2). The progress of reaction was monitored by TLC. After completion of reaction, the reaction mixture was extracted with ethyl acetate. Purification of product was carried out on silica gel before evaporation of solvent.



R^1=H,Cl,Me,OH,NO_2,OCH_3,$OCOCH_3$,
R^2=H,Me,Cl,4-NO_2,3-OMe,4-Cl,4-Br

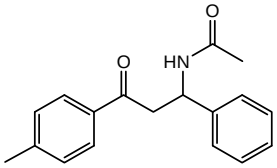
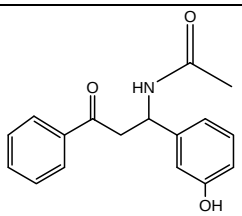
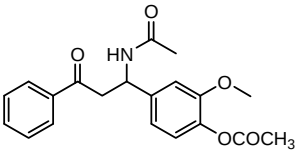
Scheme 1: Synthesis of β-acetamido ketones

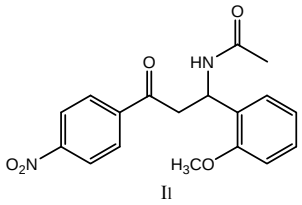
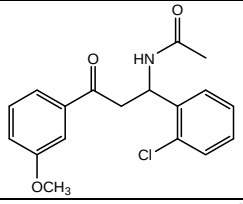
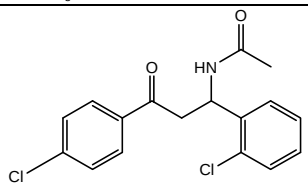
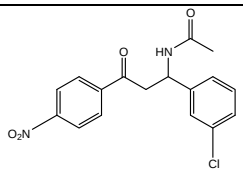
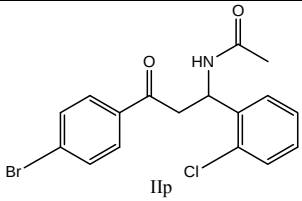
Reaction conditions: aromatic aldehydes =10 mmol, enolizable ketone =10 mmol, acetyl chloride =10 mmol and acetonitrile =10 mmol, $CuO\text{-}SnO_2$ catalyst, All compounds are well characterized by spectroscopic techniques.

Following β- acetamido ketones were synthesized:

Reaction conditions: aromatic aldehydes =10 mmol, enolizable ketone =10 mmol, acetyl chloride =10 mmol and acetonitrile =10 mmol, $CuO\text{-}SnO_2$ catalyst, All compounds are well characterized by spectroscopic techniques.

Table No.1 : List of the biologically evaluated β- acetamido ketone compounds synthesized by four component reaction.

Compound	Name of the compound	Structure	Yield (%)	Time (h)	MP (^{0}C)
I_a	β-Acetamido-β-(phenyl)-4-methylpropiophenone		74	10	117
I_b	β-Acetamido-β-(3-Hydroxyphenyl) – propiophenone		76	12	119
I_c	β -Acetamido- β-(3-methoxy, 4-acetoxy-phenyl) Propiophenone		60	8	92

I_d	β -Acetamido- β-(2-methoxy)-4-nitro propiophenone	 II	82	10	146
I_e	β -Acetamido- β-(2-chloro)-3-methoxy propiophenone		55	10	104
I_f	β -Acetamido- β-(2-chloro)-4-chloro propiophenone		79	15	169
I_g	β -Acetamido- β-(3-chloro)-4-nitro propiophenone		70	8	172
I_h	β-Acetamido-β-(2-chloro)-4-bromo propiophenone	 IIp	72	12	192

***Reaction conditions:** aromatic aldehyde=10mmol, enolizable ketone=10mmol, acetyl chloride =10mmol and acetonitrile=10mmol, $CuO\text{-}SnO_2$ catalyst, All compounds are well characterized by spectroscopic techniques such as IR, NMR, GC-MS.

Evaluation of biological activities of β- acetamido ketones: β- acetamido ketones were tested against the bacterial species using cup plate method and their minimum inhibitory concentrations (MIC) were as determined by using broth macro dilution method. The organisms used for both these method include *Staphylococcus aureus, Escherichia coli, Shigella dysentariae, Klebsiella pneumoniae, Proteus mirabilis, Salmonella typhi, Bacillus subtilis, Bacillus megatherium,Proteus vulgaris* and *Pseudomonas aeruginosa*.

RESULTS AND DISCUSSION

The antimicrobial activity of β- acetamido ketones was assessed against the test organisms *Staphylococcus aureus, Escherichia coli, Proteus vulgaris, Bacillus Subtilis, Pseudomonas aeruginosa, Bacillus megatherium, Salmonella typhi, Shigella dysentariae Klebsiella Pneumoniae* and *Proteus mirabilis.* All bacterial species used in present investigation are known human pathogens.

The minimum inhibitary concentration (MIC) values were determined by serial dilution method. The comparative study of MIC values of the compounds is given in **Table-2.**

The results of sensitivity of various pathogenic bacteria towards the synthesized compounds in presence of Codeine as supplementary drug are tabulated. The results of sensitivity of pathogenic microbes towards the synthesized β- acetamido ketones (Ia-Ih) are shown.

For convenience, the compounds were graded as ---

01) Highly active : With MIC values > 3 to 12.5 μg/ml

02) Moderately active : With MIC values > 25 to μg/ml

03) Poorly active : With MIC values > 100 – 200 μg/ml

Majority of the compounds shows moderate activity with MIC values in the range > 3 to 200 μg/ml towards Gram positive and Gram negative micro-organisms.

The compounds Ia and Ib possess highest activity with MIC values 3 to 6.2 μg/ml. towards all the bacteria tested.

Table 2: Comparative study of MIC values of β- acetamido ketones against micro-organisms.

No.	Compounds→ Microbes↓	Ia	Ib	Ic	Id	Ie	If	Ig	Ih
1	*Staphylococcus aureus*	6.2	3.0	100	100	25	50	12.5	100
2	*Escherichia coli*	6.2	>3.0	50	25	12.5	50	12.5	50
3	*Proteus vulgaris*	>3.0	>3.0	100	25	12.5	6.2	25	100
4	*Bacillus Subtilis*	>3.0	6.2	25	25	25	100	100	25
5	*Pseudomonas aeruginosa*	6.2	6.2	25	50	6.2	12.5	25	25
6	*Bacillus megatherium*	6.2	6.2	100	50	50	100	12.5	100
7	*Salmonella typhi*	3.0	3.0	50	25	50	100	3.0	50
8	*Shigella dysentariae*	>3.0	3.0	25	12.5	25	50	50	25
9	*Klebsiella Pneumoniae*	3.0	>3.0	100	25	12.5	25	25	100
10	*Proteus mirabilis*	6.2	>3.0	25	50	25	50	12.5	25

Compounds Ie and If were less active towards all the pathogens excepting *Pseudomonas aeruginosa* and *Proteus vulgaris* respectively. Compounds Ic, Id and Ih were less active with respect to antimicrobial activity towards used pathogens. The compound Id is exceptionally sensitive towards *Pseudomonas aeruginosa*. The compounds Ig highly active in case of *Salmonella typhi.* Thus the microbes *Staphylococcus aureus, Escherichia coli, Salmonella typhi, Klebsiella pneumoniae* and *Proteus mirabilis* were comparatively resistive towards all the synthesized compounds excepting compounds Ia and If.

CONCLUSION

The supplementary drug, Codeine was found synergistic with combination of compounds Ia and Ib. Where as in case of comounds Ie, If and Ig , the results were exceptionally appreciable towards the microorganisms *Pseudomonas aeruginosa, Proteus vulgaris, Shigella dysentariae, Bacillus subtilis, Bacillus megatherium, Proteus vulgaris and Bacillus subtilis, Shigella dysentariae.*

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