Electron deficient alkynes reactivity against binucleophilic compounds

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Final Degree Project
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June, 6th 2018
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ACKNOWLEDGEMENT

I would first like to thank my final degree project supervisor Prof. John Wallis of Nottingham Trent University. He allowed me to develop my project in his organic research group and he guided and helped me in everything that I have need during this period. I have to say thank you to all my laboratory mates, especially to Dr Onur A. Sahin, Jonathan Bristow and Sara Shehata to support and teach me day to day in the laboratory.
Actually, I would like to thank all Nottingham Trent University staff that allowed me to do my project.

I would also like to thank to Chemistry Faculty of Universitat Rovira I Virgili and all the professors that I had along my degree studies.

Finally, I must express my very profound gratitude to my family and to my friends for give me support and continues encouragement through the process of developed this project.

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1. Abstract

Nowadays, bacteria have developed resistance against many antibiotics, for this reason, it is needed to look for new antibiotics. With the aim of finding products that could be converted in compounds with antibacterial activity, the reactivity of two electron deficient alkynes against binucleophilic species was tested and promising results were obtained. Theses two alkynes are (E)-dimethyl hex-2-en-4-ynedioate (DMHD) and dimethyl 2,4-hexadiyndioate (dialkyne).

On the one hand, in reference to DMHD, its reactivity was tested against phenylhydrazine, methylhydrazine and hydrazine. In the first two cases the results were not good according to our objective. Anyway, the reactivity of DMHD against hydrazine seemed to be promising. Two interesting products with structures that suggest to be converted in compounds to be tested as new antibiotics were found. These products are a solid lactam (E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoat) and an oily diester (methyl-2-(1'H-4',5'-dihydro-3'-methoxycarbonyl-pyrazol-5'-yl) acetate).

On the other hand, the reactivity of dialkyne against N,N'-dimethylthiourea was tested. In this case two interesting products were isolated too but in very little amounts. This low yield was caused due to the fact that dialkyne synthesis was not efficient. For this reason, the procedure to obtain the dialkyne needs to be improved. Despite this fact, it was proved that the chemistry of the dialkyne against binucleophilic species is possible. Hence, the next step could be test its reactivity against hydrazine which is thought that could generate interesting products to convert to compounds for testing new antibiotics.

2. Objectives

The general aim of this project was to explore some new chemistry of two electron deficient alkynes with nucleophilic species. These alkynes are: (E)-dimethyl hex-2-en-4-ynedioate (DMHD) and a dialkyne, dimethyl 2,4-hexadiyndioate.

The objective about studying this chemistry was to isolate products which could be converted to compounds for testing as new antibiotics.

If we focus the attention in the specific objectives of this project:

- DMHD reactivity against hydrazine.
- DMHD reactivity against phenylhydrazine.
- DMHD reactivity against methylhydrazine.
- Dialkyne reactivity against N,N'-dimethylthiourea.
- Dialkyne reactivity against hydrazine.

3. Introduction[1]

First of all, it has to be said that this project was developed at Nottingham Trent University, in the School of Science and Technology. Specifically, it has been developed in the organic group leaded by John Wallis.

This project is based in the searching of molecules that could be converted in compounds with antibacterial activity.

Bacteria are single-cell prokaryotes microorganisms that could produce infections and diseases to humans. For this reason, something is needed to fight against them and this is the antibiotics function. They are drugs that have got the aim of kill bacteria or avoid their reproduction.

It should be said that before bacteria were known, some civilizations like Chinese or Greek used antibacterial herbs or potions many centuries ago.

However, bacteria were not identified until 1670s by Anton van Leeuwenhoek and it was not until nineteenth century that they were linked with disease. Once this link was discovered, many scientists started to look for effective antibacterial agents and many different kind of antibiotics effective against different type of bacteria were discovered along nineteenth century.

Nowadays, main problem of these drugs is that bacteria have developed abilities to resist them. According to World Health Organisation, antibiotic resistance is raising high levels in all parts of the world and if this growing continues, old diseases will come back and advanced surgical procedures will not carry out due to the risks of infection.

For all these reasons, we have to look for new antibiotics and this search will never end.

If we talk about antibiotics action and their successful, we have to relate it with the fact that they can attack bacterial cell without attacking human cells because of the different properties between bacterial and human cell. We can observe this differences in the following table (Table 1).

Table 1 Differences between human and bacteria cells.

Cell properties	***Bacteria***	***Human***
Nucleus	Inexistent	Defined
Structures	Simple	Varied and complexes
Metabolism	Enzymes to catalyse some vital reactions	They don't require these enzymes
Cell membrane and cell wall	Both of them	Cell membrane

In addition, talking about the mechanism of antibacterial action, there are five main mechanisms:

- Inhibition of cell metabolism.
- Inhibition of bacterial cell wall synthesis.
- Interactions with plasma membrane.
- Disruption of protein synthesis.
- Inhibition of nucleic acid transcription and replication.

The attention is going to be focus on antibiotics that could inhibit bacterial cell wall synthesis. The two most important antibiotics that use this mechanism are penicillin and cephalosporin. The structures of these two types of antibiotics contain similarities

with the molecule structure of the compounds that it would be try to isolate in this project. For this reason, it is thought that the new molecules could have similarities in the activity with penicillin and cephalosporin.
In the following section is going to be explained the mechanism of action for penicillin. The mechanism of action for cephalosporin is not going to be explained due to the fact that it is very similar to penicillin one.

3.1 Mechanism of action for penicillin

The penicillin effectiveness and selectivity is based in the fact that bacteria cell have got a wall and animal cells do not have this wall. This wall is important to bacteria because it allows them to survive in different environmental conditions.
The wall is a peptidoglycan structure that it could be synthetized because the action of different enzymes. Penicillin is able to inhibit the action of one of these enzymes, exactly the transpeptidase enzyme which is the responsible of a cross-linking reaction. If this reaction it is not completed, the cell wall will become weak and it will not be able to protect the cell. The action mechanism for transpetidase enzyme and mechanism action for its inhibition followed by penicillin are shown in figure 1.

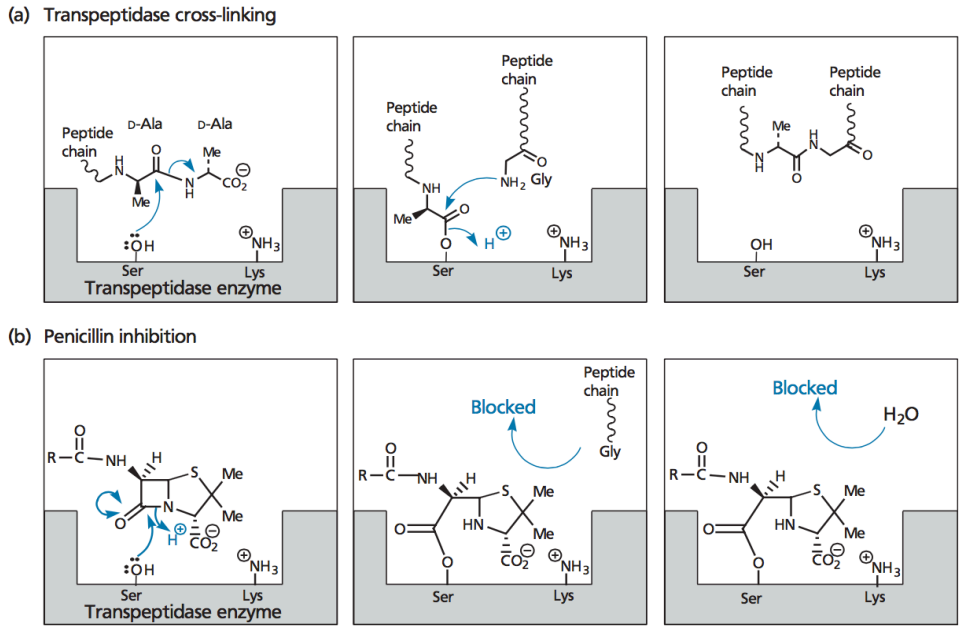


Figure 1 Transpeptidase and penicillin mechanism of action.

In figure 1a the action mechanism of transpeptidase enzyme it is shown. This enzyme allows a cross-linking reaction essential to synthetized the peptidoglycan wall. In the first step nucleophilic attack is done by transpeptidase enzyme serine at peptic carbonyl. The peptide bond between two D-Alanines (D-Ala) is broken because of this nucleophilic attack and terminal D-alanine leave the active site. After that, another peptide chain enters to the active site where a terminal glycine reacts with the alanine group to form a new peptide bond, displacing it from the transpeptidase enzyme serine and linking the two peptide chains.
In figure 1b the inhibition mechanism of the enzyme followed by penicillin is described. It is thought that the transition-state conformation adopted by D-Ala-D-ala during the cross-linking reaction is similar to penicillin structure. This similarity allows penicillin to

achieve the active site instead of D-Ala-D-Ala. Once penicillin is in the active site a nueoclephilic attack is done by the serine at penicillin carbonyl lactam. This attack causes the break of the amide bond in the same way that causes the break of D-Ala-D-ala bond. The difference between these two procedures is that penicillin has got a cyclic structure so once the amide bond is broken, the penicillin do not leave the active site and blocks the active site access to other peptide chain or water.

3.2 Relation structure reactivity penicillin and cephalosporin

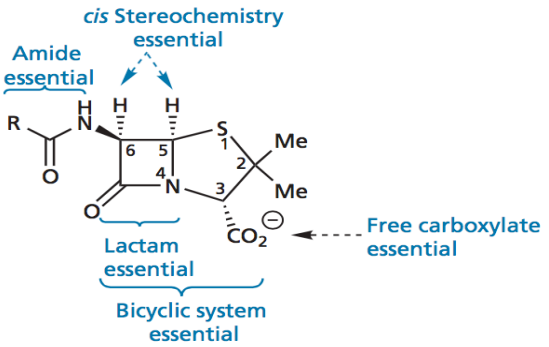


Figure 2 Penicillin structure.

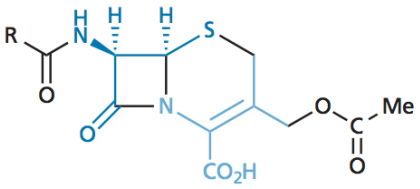


Figure 3 Cephalosporin structure.

In figures 2 and 3 the structure of penicillin and cephalosporin are shown. In the following lines the relation between their structures and their activity is going to be explained.

The first thing that it should be said is the importance of the presence of the strained β-lactam ring. The ring strain is favoured by the fact they are bicyclic systems. This structure allows the amide group to be weaker than in a non-cyclic amide. This effect is caused because on the one hand, the nitrogen hybridization of a non-cyclic amide is sp^2, so its geometry is trigonal planar. On the other hand, the nitrogen hybridization of a lactam is sp^3 because the strain of the ring forces nitrogen to adopt a pyramidal geometry. Due to these different geometries, the overlap between carbonyl π-orbitals and nitrogen lone pair is less efficient in lactams than in non-cyclic amides.

The bond weakness of lactams makes them more reactive against a nucleophilic attack which is important in the transpeptidase inhibition.

This kind of amide bond is the one that is going to be imitated in the molecules that are going to be synthetized in this project.

Other important points of these two antibiotics structures are the cis stereochemistry of the protons in the lactam ring and the acylamino side chain.

Finally, other important property is the free carboxylate group that allows penicillin to be administrated as salts. Furthermore, this ion could interact with the charged nitrogen of a lysine residue in the active site.

4. Theoretical foundation

In this section, mechanisms about the different reactions that had been done are going to be proposed. In addition, properties such as the reactivity of some compounds are going to be discussed trying to understand the reaction mechanisms.

4.1 Synthesis of (E)-dimethyl hex-2-en-4-ynedioate[2] (DMHD)

The mechanism proposed to obtain DMHD was methyl propiolate organo-catalyzed self-coupling. The organo-catalyst used was 1,4-diazabicyclo[2.2.2]octane (DABCO). The mechanism reaction could be explained by the catalytic cycle described in figure 4.

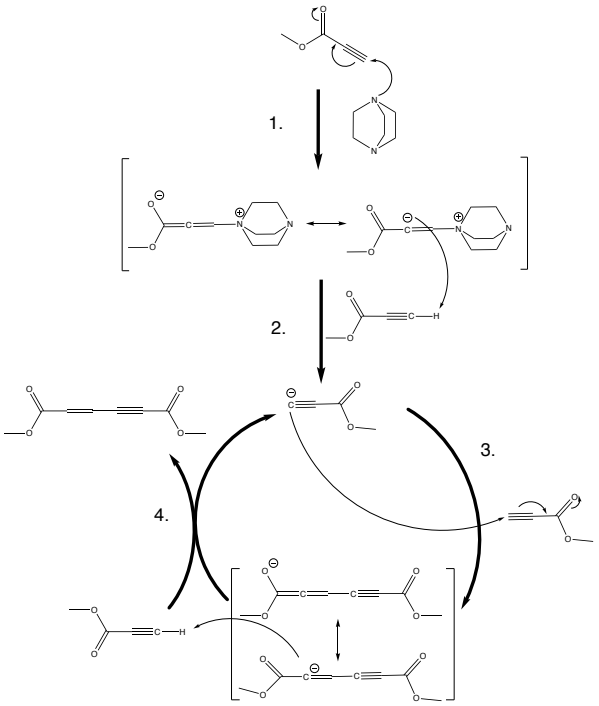


Figure 4 Catalytic cycle about DMHD synthesis.

- Steps 1 and 2 correspond to activation of methyl propiolate.

In the first step the nitrogen lone pair of DABCO acts as a nucleophile to attack the electrophilic carbon of methyl propiolate.

In the second step an acid-base reaction allows the deprotonation of methyl propiolate and its activation.

- The catalytic cycle (steps 3 and 4) starts once methyl propiolate is activated.

In the step 3 methyl propiolate deprotonated acts as nucleophile and attacks the electrophile carbon of a methyl propiolate molecule. The product of this reaction is an anionic intermediate.

This anionic intermediate reacts with other methyl propiolate molecule by an acid-base reaction (Step 4). In this step DMHD and an activated molecule of methyl propiolate are generated.

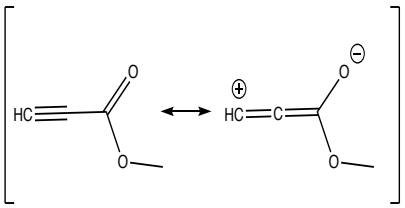


Figure 5 Methyl propiolate resonance structures.

One aspect that should be highlighted of this mechanism is the electrophile character about the alkyne terminal carbon of methyl propiolate. This property could be explained by figure 5. The conjugation of the triple bond with the ester group allows the terminal carbon to acquire the electrophile character.

4.2 Reactivity of DMHD against nucleophiles with two nucleophilic centres

DMHD is a molecule with different electrophilic centres, for this reason it was interesting to understand its reactivity against a nucleophile such as hydrazine. At the same time new interesting molecules could be isolated and converted on new compounds for testing antibiotic activity.

It could be said that DMHD contain 6 electrophilic centres (Figure 6). These six centres correspond to two carbonyl carbons and their α and β positions. The electrophilic

character of these two positions could be explained by the DMHD resonance structures (Figure 6).

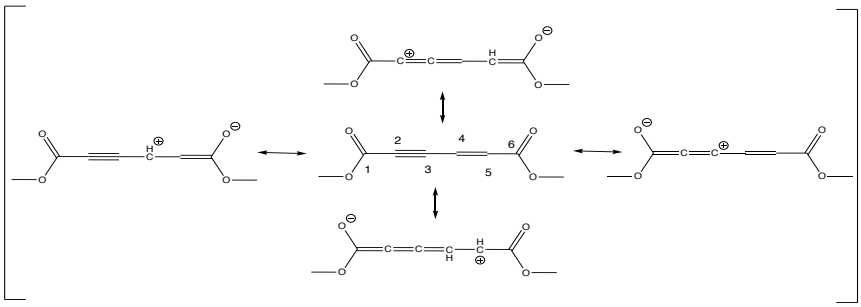


Figure 6 DMHD resonance structures.

On the one hand, the electrophilic character of β-carbonyl positions (3 and 4) is originated by the conjugation between the carbonyl and double/triple bound.
On the other hand, the electrophilic character of α-carbonyl (2 and 4) is caused by the double and triple bond conjugation.

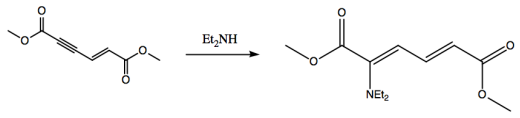


Figure 7 Reaction between DMHD and diethylamine.

Previous studies about reaction between DMHD and nucleophiles were done. First of all, the reactivity against diethylamine was tested (Figure 7). The reaction was studied in different solvents. One of them was methanol and the nucleophilic addition was produced on a *sp* carbon at α-carbonyl position.
Further investigations should be made to study the nucleophilic additions modes to DMHD. For this reason, reactions between this compound and different compounds with two nucleophilic centres were reported[3]. These studies could help to understand the reaction between DMHD and hydrazine. It was demonstrated that different thioureas attacked only at *sp* carbons of DMHD and the adjacent carbonyl carbon. In addition, the thiourea's sulphur always attacked at *sp* carbons and its nitrogen attacked at carbonyl carbon.
Two examples of these studies were the reactions of N-methylthiourea and imidazoline-2-thione with DMHD are shown in figure 8 and 9.

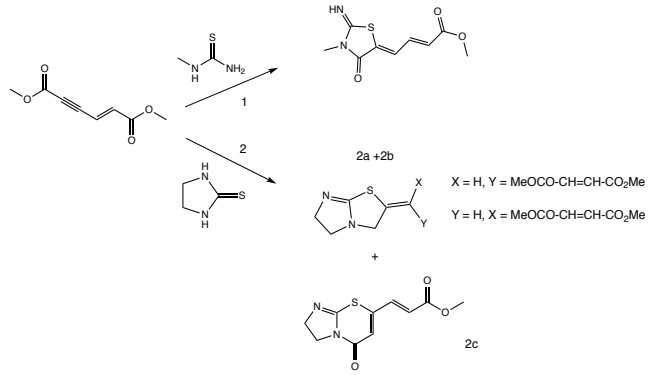


Figure 8 Reactions between DMHD and N-methilthiourea (1) and imidazoline-2-thione (2).

1) Sulphur of N-methylthiourea attacks at *sp* carbon in α-carbonyl position and nitrogen attacks at carbonyl carbon forming a five member ring.
2) Sulphur of imidazoline-2-thione attacks at *sp* carbon in α-carbonyl position and nitrogen attacks at carbonyl carbon originating a five member ring. (Products 2a and 2b).
Sulphur of imidazoline-2-thione attacks at *sp* carbon in β-carbonyl position and nitrogen attacks at carbonyl carbon generating a six member ring (Product 2c).

Other interesting reaction that was reported[3] is the reaction between DMHD and 1,2-diaminobenzene. In this case, it is showed that amines could attack at same positions as thioureas but they can attack at DMHD sp^2 carbons too (Figure 9).

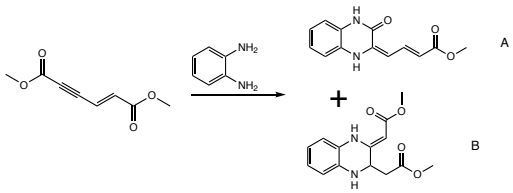


Figure 9 Reaction between DMHD and 1,2-diaminobenzene.

A) 1,2-diaminobenzene attacks at *sp* carbon in α-carbonyl position and at carbonyl position creating a six member ring (Product A).
B) 1,2-diaminobenzene attacks at *sp* carbon in β-carbonyl position and at sp^2 carbon in β-carbonyl position creating a six member ring (Product B).

4.2.1 Reaction between DMHD and hydrazine

Two products were isolated in the reaction between DMHD and hydrazine. One of them is a white solid and the other is a yellow oil. The first one fits with a lactam (methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate) and the second one is a diester (methyl-2-(1'H-4',5'-dihydro-3'-methoxycarbonyl-pyrazol-5'-yl) acetate).

4.2.1.1 Methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate synthesis

The proposed mechanism for the lactam E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate synthesis is the one in figure 10.

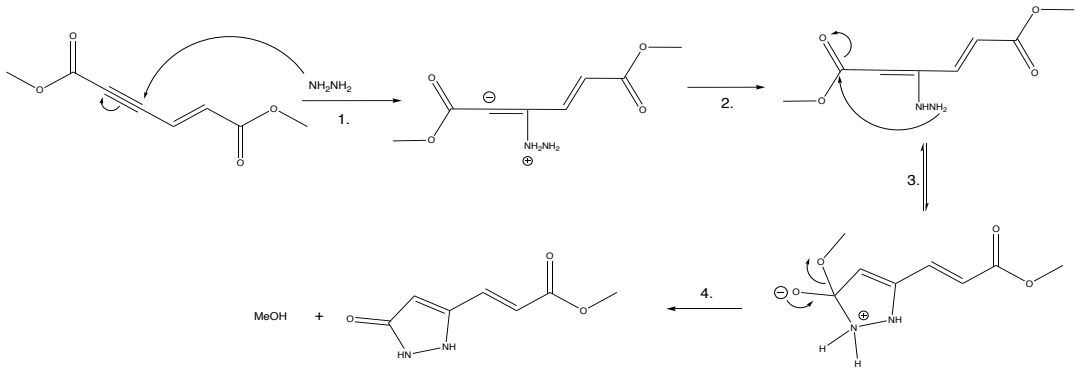


Figure 10 Mechanism E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate synthesis.

- Step 1: nucleophilic attack of hydrazine at *sp* carbon on β-carbonyl position.
- Step 2: intramolecular protonation.
- Step 3 and 4: typical intramolecular nucleophilic acyl substitution. It includes a nucleophilic attack of the hydrazine at the carbonyl group allowing the formation of a tetrahedral intermediate. After that, an alkoxy group is expulsed and this group catches the proton of the nitrogen following an acid-base reaction. According to Baldwin rules[4,5], 5-exo-trig cyclization was produced which is a favoured type of cyclization.

Paying attention to chemoselectivity of this reaction it could be explain that hydrazine attacks preferably at *sp* carbon on β-carbonyl position due to the fact that lactam is the major product of the reaction. The electrophilic character of this centre is caused by the conjugation between triple bond and ester group that allows *sp* carbon on β-carbonyl position to have partial positive charge as it was showed at

figure 6. This preference to attack *sp* carbon instead to sp^2 carbon was shown in the previous studies about thioureas[3] too.

4.2.1.2 Methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate synthesis

The proposed mechanism for the diester E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate synthesis is the one in figure 11.

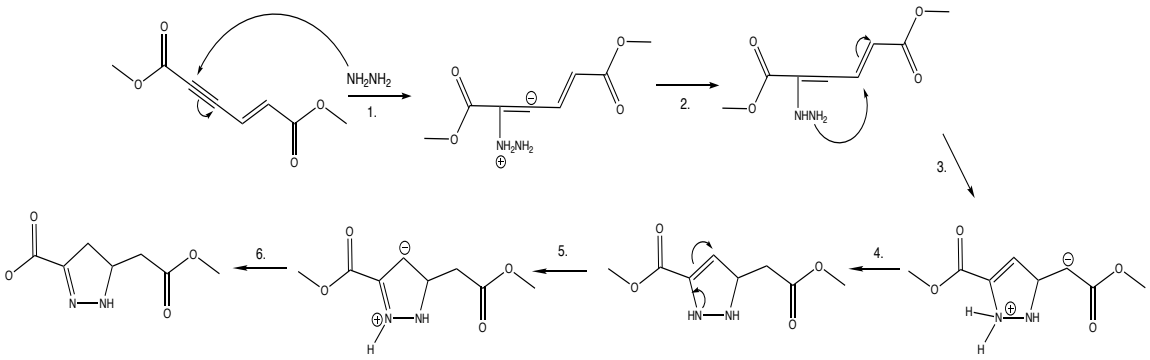


Figure 11 Mechanism of E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate synthesis.

- Steps 1 and 3 are nucleophilic attacks of the hydrazine at two different electrophile centres of DMHD. Both of these attacks are followed by a intramolecular protonation r (steps 2 and 4).
- Step 5 corresponds to an electronic rearrangement because of the possible conjugation between the nitrogen lone pair and the double bond of the cycle.
- Step 6 fits with intramolecular protonation as steps 2 and 4.

Talking about reaction chemioselectivity it is shown that despite the diester is a minor product it is possible that DMHD sp^2 carbon is attacked by a nucleophile. This kind of attack it was not shown in thioureas but it was shown in 1,2-diaminobenzene[3].

It was proposed this mechanism for the formation of diester where the first nucleophile attack it was done in the *sp* carbon of DMHD and the second attack was done at sp^2 carbon for two reasons:

- In other cases the *sp* carbon seemed to have a higher electrophilic character that sp^2 carbons.
- If the nucleophilic attacks were in the order showed in the figure 11 and the Baldwin rules[4] are followed the cyclization would be 5-exo-trig. If the attacks were in the opposite order, the cyclization would be 5-endo-dig. According to studies about the different kind of cylacitions[5] the 5-exo-trig is more favoured than 5-endo-dig.

4.2.2 Reaction between DMHD and methylhydrazine

Considering the precedents about DMHD reactivity against compounds with two nucleophilic centres, it was expected that methylhydrazine would attack at *sp* carbons of DMHD, at least the first attack was expected to be in these kind of carbons. Despite this fact, if attention is put on 1,2-diaminobenzene reaction with DMHD, it could exist a possibility that methylhydrazine attacks to DMHD sp^2 carbon.

In DMHD reaction with methylhydrazine four different products were isolated. There were achieved three different types of diesters and a lactam. The three diesters are yellow oils and the lactam is a white solid.

In all of these products methylhydrazine first attack was in a DMHD *sp* carbon as it was expected. The fact that it was not expected is that in the two main products obtained in the reaction, the first attack was done by the nitrogen that it is not attached to methyl.

The first attack to DMHD was expected to be about the nitrogen bonded to methyl because methyl is an electron-donating group, so the nucleophilic character of nitrogen bonded to methyl would be higher than the other nitrogen.

However, an explanation about this fact could be founded and it is called the α-Effect[6].

This effect explains that some nucleophiles are more nucleophilic as it would be expected because of its pKa. The explanation of this nucleophilic character increase falls in an overlap between an orbital of the nucleophilic atom and an orbital of the neighbour atom. Exactly, this overlap is produced between the orbital that contains lone pair of the neighbour atom and the orbital that contains the electrons responsible of the nucleophilic character. This overlap generates HOMO orbital energy increase and this increases the reactivity of the nucleophile.

In the case of methylhydrazine it could be said that lone pair orbital of nitrogen bonded to methyl overlap with the lone pair orbital of the other nitrogen that increased its nucleophilic character.

In the following lines is going to describe the reaction mechanism about the four products that were obtained with methylhydrazine.

4.2.2.1 Methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) acetate synthesis

The proposed mechanism for the generation of reaction major product, that it was a diester called methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) acetate, it is shown in figure 12.

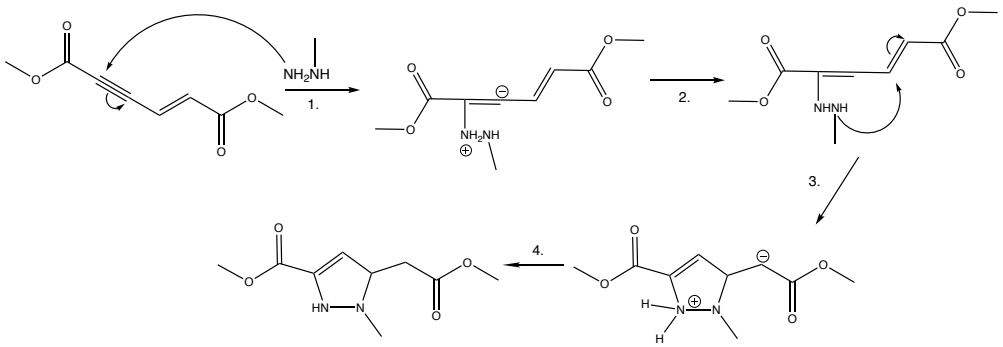


Figure 12 Reaction mechanism of methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) acetate synthesis.

Step 1 and step 3 are nucleophilics attacks that are followed by intramolecular protonations (steps 2 and 4).

About the nucleophilic attack in the step one could be highlighted that it is done at *sp* carbon as it was expected, exactly at α-carbonyl position. This attack is done by the nitrogen non attached to methyl because of α-Effect[6].

Talking about step 3, the attack is done at DMHD sp^2 carbon generating a 5-exo-trig cyclization.

4.2.2.2 Methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl) enoate synthesis

The mechanism for the synthesis of methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl) enoate is shown in figure 13. It was a totally unexpected product due to the fact that a four-membered ring is generated and this is not very stable.

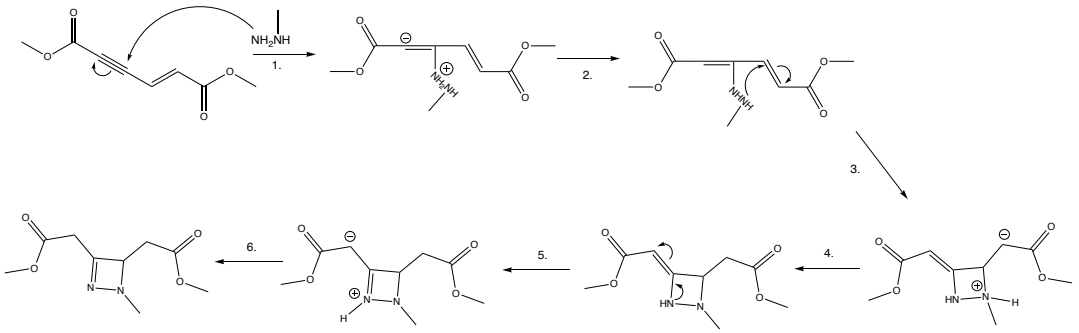


Figure 13 Reaction mechanism of methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl)enoate synthesis.

Steps 1 to 4 in the mechanism of this product are very similar to the previous mechanism. However, there are two changes that allow forming the unusual four-member ring. In the first step the nucleophilic attack is done at *sp* carbon too but this is done at β-carbonyl position.

The other different between these two mechanisms is the kind of cyclization. In this case, a 4-exo-trig cyclization is produced.

Finally, steps 5 and 6 fit with an electronic rearrangement followed by intramolecular protonation. This electronic rearrangement could be possible because an exocyclic double bond is substituted by an endocyclic double bond that is more stable.

4.2.2.3 Methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate synthesis

In the following figure (figure14) it is represented the mechanism of reaction for the synthesis of the solid product of the reaction.

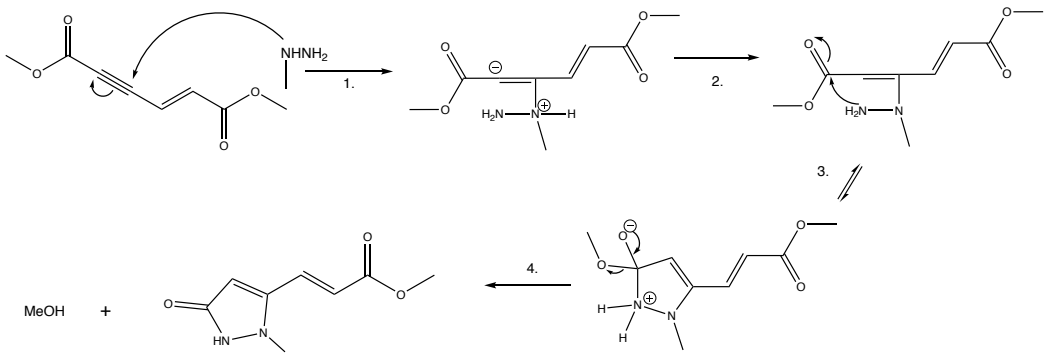


Figure 14 Reaction mechanism of methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate synthesis.

This mechanism is exactly the same that the one shown in solid lactam synthesis in the reaction of DMHD with hydrazine.

In the first step it could be observed a nucleophilic attack at *sp* carbon of β-carbonyl position followed by intramolecular protonation in step 2. It should be highlighted that in

this case the first attack is realised by the nitrogen bonded to methyl. This is the opposite order followed in the formation of the two major products of this reaction.
After that, a typical intramolecular nucleophilic acyl substitution is observed in steps 3 and 4. As it has been said, the mechanism that is followed is exactly the same as in the synthesis of the other lactam, so 5-exo-trig cyclization it is followed again.

4.2.2.4 Methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate synthesis

The proposed mechanism for achieving other diester, called methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate it could be observed in figure 15.

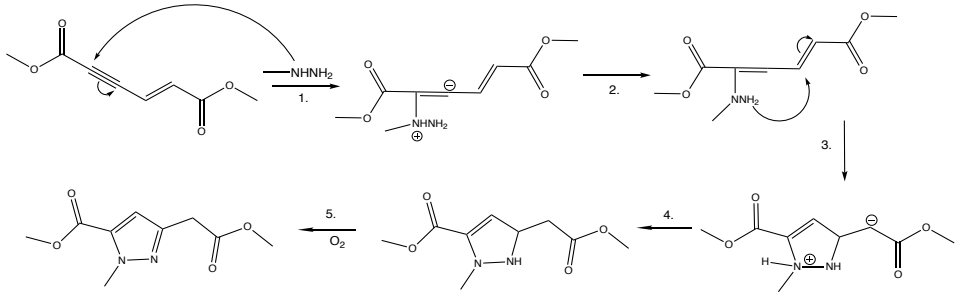


Figure 15 Reaction mechanism of methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate synthesis.

Comparing this mechanism with the mechanism of the major product (methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) acetate, it could be said that the first four steps are exactly the same and a 5-exo-trig cyclization is produced too. But there is an important differentiation. In this case the first nucleophilic attack is done by nitrogen bonded to methyl (step1).
In addition, this mechanism has got a final step (step 5) that it is not observed in the first mechanism. Step 5 fits with an oxidation that could be favoured because it allows achieving an aromatic ring.

4.3 Methyl propiolate reactivity against N,N'-dimethylthiourea

Two main products were obtained in the reaction between methyl propiolate and N,N'-dimethylthiourea. These two major products are a white solid (3-methyl-2-(methylimino)-2,3-dihydro-4*H*-1,3-thiazin-4-one) and a colourless oil (methyl (*E*)-3-(methylthio)acrylate). Their formation mechanisms are going to be explained in the following sections.
Furthermore, two more compounds were obtained in very little amounts, but it is worth explaining their formation mechanism too. These two compounds are solids: 1,3-dimethyl-2-thioxo-2,3-dihydropyrimidin-4(1*H*)-one and 1,3-dimethyl-1,2,3,4-tetrahydro-2,4-dione.

4.3.1 (Z)-3-methyl-2-(methylimino)-2,3-dihydro-4H-1,3-thiazin-4-one synthesis

The proposed mechanism for the major product formation is shown in figure 16.

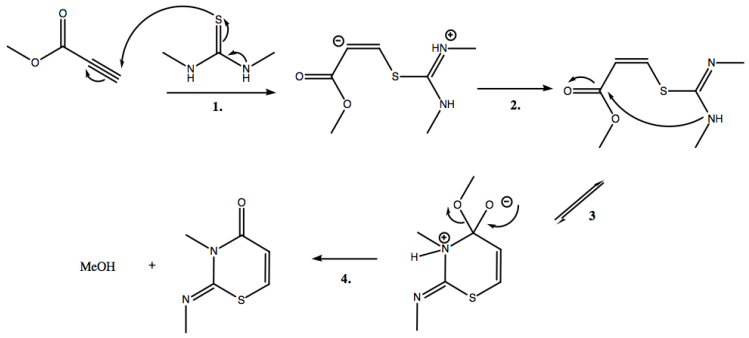


Figure 16 Reaction mechanism for (Z)-3-methyl-2-(methylimino)-2,3-dihydro-4H-1,3-thiazin-4-one synthesis.

- Step 1: This step fits with a nucleophilic cis-addition of the N,N'-dimethyltiourea to dimethyl propiolate. This is possible because of sulphur nucleophilic character and the electrophilic character of methyl propiolate β-position (explained in figure 6).
- Step 2: Acid-base reactions between methanol and the result product of the nucleophilic cis-addition allow carbonyl α-position protonation and nitrogen deprotonation. The stereochemistry of the resulting product is Z.
- Steps 3 and 4: these steps correspond to an intramolecular nucleophilic acyl substitution. According to Baldwin[4] rules a 6-exo-trig cyclization could be observed and this type of cyclization is favoured[5].

4.3.2 Methyl (E)-3-(methylthio)acrylate synthesis

The proposed mechanism for the colourless oil formation is shown in figure 17.

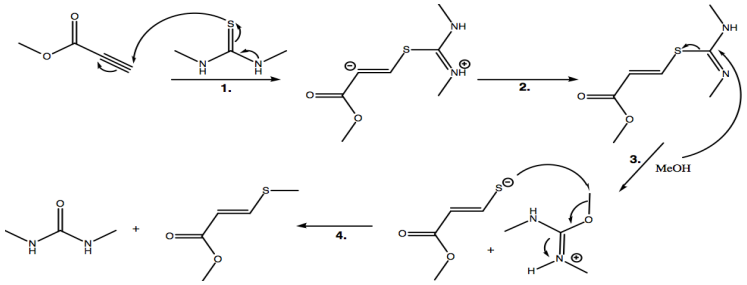


Figure 17 Reaction mechanism for (E)-3-(methylthio)acrylate synthesis.

- Steps one and two are similar to the previous mechanism, but there is an important different. In this case it could be observed a nucleophilic trans-addition and the stereochemistry of the second step resulting product is E. This stereochemistry does not allow the intramolecular cyclization as in the previous mechanism.
- Step 3: methanol nucleophilic attack at electrophilic centre of the product resulting in step 2. This nucleophilic attack causes the molecule splitting in an anion and a cation.
- Step 4: anion nucleophilc attack to the cation to achieve the interest product (E)-3-(methylthio)acrylate and N,N'-dimethylurea.

4.3.3 1,3-dimethyl-2-thioxo-2,3-dihydropyrimidin-4(1H)-one and 1,3-dimethylpyrimidine-2,4(1H,3H)-dione synthesis

The reaction mechanisms of these two minor products are going to be explained in this section. The mechanism is very similar for both of them, 1,3-dimethyl-2-thioxo-2,3-dihydropyrimidin-4(1*H*)-one (figure 18) and 1,3-dimethyl-1,2,3,4-tetrahydro-2,4-dione (figure 19).

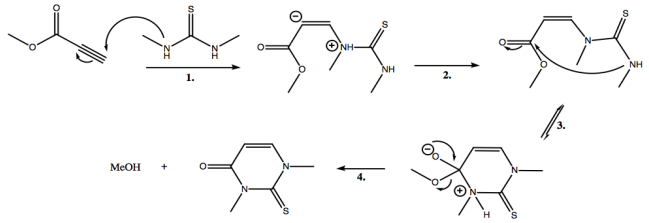


Figure 18 Reaction mechanism for 1,3-dimethyl-2-thioxo-2,3-dihydropyrimidin-4(1H)-one.

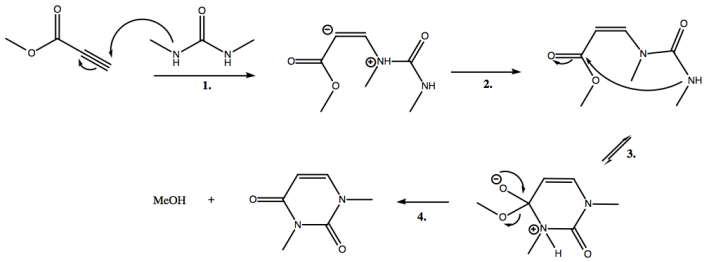


Figure 19 Reaction mechanism for 1,3-dimethyl-1,2,3,4-tetrahydro-2,4-dione.

The mechanism followed in both cases is the same that it was explained in 16 for the major product. For this reason, both mechanism show a nucleophilic cis-addition (step 1), followed by acid-base reaction with methanol (step 2) and an intramolecular nucleophilic acyl substitution with 6-exo-trig cyclization (steps 3 and 4).

The differences between these mechanism and the one for (Z)-3-methyl-2-(methylimino)-2,3-dihydro-4H-1,3-thiazin-4-one are:

- Figure 18→ The first nucleophilic attack is not done by sulphur, in the case, it is done by nitrogen lone pair.
- Figure 19→ In this case N,N'-dimethylthiourea is not involved in the reaction. The compound that reacts with methyl propiolate is N,N'-dimethylurea. This urea it could be originated in formation of (E)-3-(methylthio)acrylate (figure 17).

4.4 Dimethyl 2,4-hexadiyndioate synthesis and its reaction with N,N'-dimethylthiourea

The formation mechanism about dimethyl 2,4-hezadiyndioate and the products that were isolated in its reaction with N,N'-dimethylthiourea are going to be discussed in this section.

4.4.1 Dimethyl 2,4-hexadiyndioate synthesis

It is known that dimethyl 2,4-hexadiyndioate synthesis consists in oxidative Glaser-Hay homocoupling of methyl propiolate. Despite this kind of alkyne couplings are very useful and they have been studied extensively, their mechanism is still under debate.

A simplified explanation (figure 20) about a quiet recent computational study[7] is going to be described in this section.

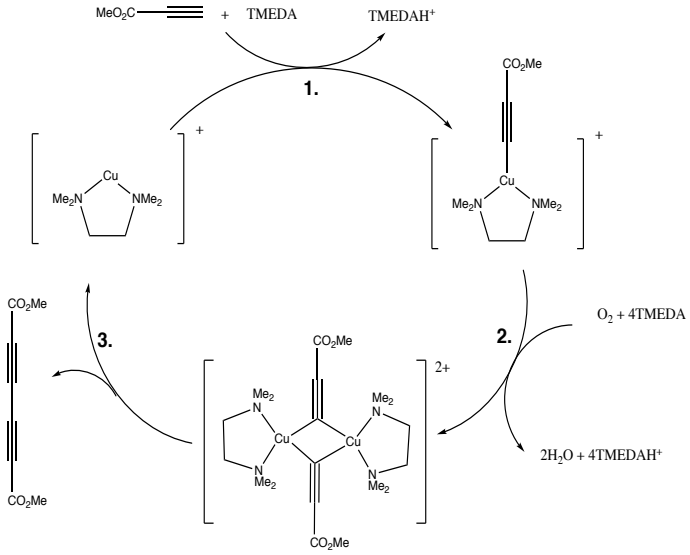


Figure 20 Catalitic cycle about dimethyl 2,4-hexadiyndioate synthesis.

- The first step consists in methyl propiolate deprotonation because an acid-basic reaction with tetramethylethylendiamine (TMEDA) and its coordination to Hay-Catalyst (cooper (I)-TMEDA complex).
- The second step represents the oxidation of copper I to copper II by oxygen that is reduced to water.
- Step three shows a reductive elimination where dimethyl 2,4-hexadiyndioate is formed and Hay-Catalyst is recovered.

4.4.2 Dimethyl 2,4-hexadiyndioate reactivity against N,N'-dimethylthiourea

First of all it is important to speak about dimethyl 2,4-hexadiyndioate properties as electrophile. It is thought that this dialkyne contains 6 different electrophilic centres (figure 21) as DMHD. The explanation about the presence of these 6 electrophilic centres in the dialkyne is the same that the one gave to DMHD. Anyway, there is an important different between these two compounds: the electrophilic character it could be different for each centre in DMHD but in the dialkyne there are only three types of electrophilic centres (1=6, 2=5 and 3=4) due to its symmetrical structure.

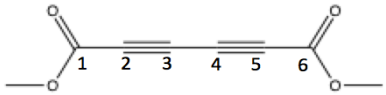


Figure 21 Electrophilic centres about dimethyl 2,4-hexadiyndioate

According to previous studies about thioureas with DMHD[3] described in section 4.2 it was expected that two molecules of N,N'-dimethylthiourea could react with the dialkyne. It was thought that sulphur could attack to positions 2 and 4 and nitrogen could attack to positions 1 and 6 generating a cyclization.

In the reaction between dialkyne and N,N'-dimethylthiourea three products were obtained. One of them was the bicyclic product expected (bis-(3-methyl-2methylimino-4-oxo-1,3-thiazolidinylidene)ethane). The other product (methyl-(2-(methoxycarbonyl-methylidene)-3methyl-2-methyimino-thiazolidin-acetate) was the result to the addition of the thiourea at the two β-carbonyl positions of the dialkyne. Finally, the last product (dimethyl E,Z-3,3'-thiobis(acrylate)) was obtained because of the reaction between the thiourea and some methyl propiolate that remained without react in the synthesis of dialkyne.

The proposed formation mechanisms of these three products are going to be exposed in the following sections.

Bis-(3-methyl-2methylimino-4-oxo-1,3-thiazolidinylidene)ethane formation mechanism

The proposed mechanism for the synthesis of this solid bicycle is shown in figure 22.

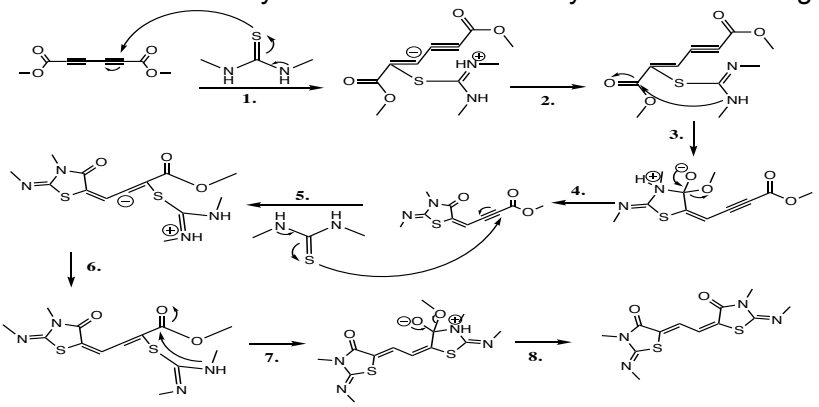


Figure 22 Bis-(3-methyl-2methylimino-4-oxo-1,3-thiazolidinylidene)ethane formation mechanism.

- Step 1: N,N'-dimethylthiourea sulphur nucleophilic addition at dialkyne α-carbonyl position as it was expected.
- Step 2: Acid-base reactions between methanol and the result product of the nucleophilic addition allow carbonyl β-position protonation and nitrogen deprotonation.
- Steps 3 and 4: these steps correspond to an intramolecular nucleophilic acyl substitution. According to Baldwin[4] rules a 5-exo-trig cyclization could be observed and this type of cyclization is favoured[5].

Steps 4 to 8 are exactly the same as 1 to 4, but in this case a thiourea second molecule reacts with the part of the dialkyne that remained without react.

Methyl-(4(-2-(methoxycarbonyl-methylidene)-3-methyl-2-methyimino-thiazolidin) formation mechanism

The suggested mechanism for methyl-(4(-2-(methoxycarbonyl-methylidene)-3-methyl-2-methyimino-thiazolidin) acetate synthesis is exposed in figure 23.

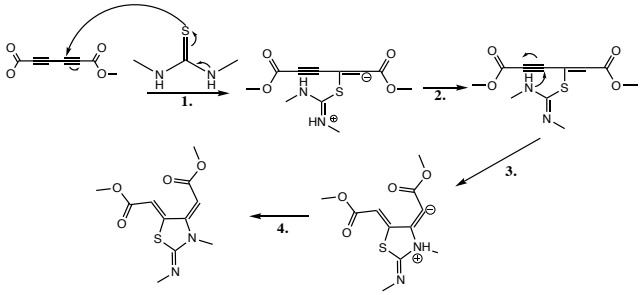


Figure 23 Formation mechanism methyl-(4(-2-(methoxycarbonyl-methylidene)-3-methyl-2-methyimino-thiazolidin).

- Step 1 corresponds to N,N'-dimethylthiourea sulphur nucleophilic addition at dialkyne β -carbonyl position.
- Step 3: nitrogen intramolecular nucleophilic addition at free dialkyne β-carbonyl position. In this step it could be observed a 5-exo-dig cyclization which is a favoured kind of cyclization.

Step 2 and 4: Acid-base reactions between methanol and the result products of the nucleophilic additions allow carbonyl α-positions protonation and nitrogen deprotonation.

Dimethyl E,Z-3,3'-thiobis(acrylate) formation mechanism

The mechanism for the formation of the product resulting of the reaction between methyl propiolate and N,N'-dimethylthiourea is showed in figure 24.

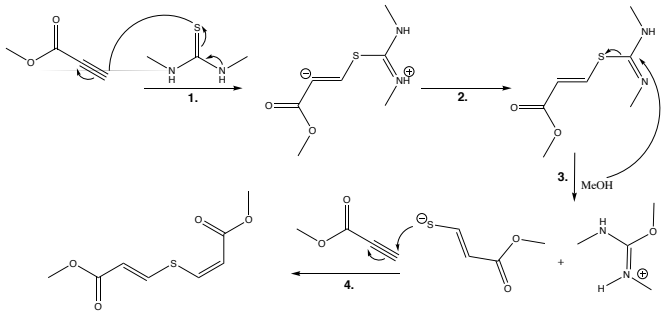


Figure 24 Dimethyl E,Z-3,3'-thiobis(acrylate)formation mechanism

- Step 1 is a nucleophilic cis-addition of N,N'-dimethylthiourea sulphur to methyl propiolate.
- Step 2 fits with acid-basic reaction between the addition product and methanol.
- Step 3 shows that the intramolecular cyclation is not possible as in other cases due to the alkene stereochemistry. For this reason, methanol is able to do a nucleophilic attack and split the molecule in an anion and a cation.
- In the step 4 it could be observed a nucleophilic trans-addition of the anion to methyl propiolate . After that, an acid-basic between the product of the addition and the cation of the previous step allowed the formation of the product.

It should be highlighted that the stereochemistry of the alkene generated in step 2 is E and the stereochemistry of the alkene formatted in the last step is Z.

5. Experimental section

The experimental procedures that had been followed are explained in detail in this section.

5.1 Reaction between DMHD and hydrazine

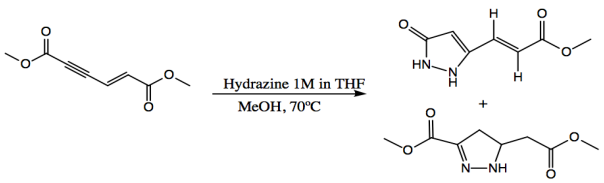


Figure 25 Reaction scheme

Hydrazine solution 1M in THF (0.3g, 0.34mmol hydrazine, 0.06eq) and methanol (10mL) were added to a 100mL flask with a guard tube filled with $CaCl_2$. The solution was stirred for 5 minutes and DMHD (1g, 5,95mmol, 1eq.) was added to the stirred solution. The mixture was stirred and heated to 70ºC overnight. After that the mixture was dark orange but the DMHD remained without react. More hydrazine solution was added (0.15g, 0.17mmol hydrazine) and the mixture was heated to 70ºC for 3 hours. The mixture was cooled in ice for 10 minutes and 50mL of diethyl ether were added. A little bit of white precipitate appeared but it was impossible to filtrate it by vacuum filtration due to the small size of the particles. Finally it was filtrated with filter paper.

The reaction was not going in the correct way due to the fact the amount of hydrazine was not enough. It was a problem of calculation, because hydrazine solution 1M in THF was used instead of use monohydratated hydrazine.

The diethyl ether was evaporated from the liquid phase and the correct amount of hydrazine solution 1M in THF was added (6mL, 6mmol hydrazine, 1eq. hydrazine). 3mL of methanol were added and the mixture was stirred for one hour (the flask had got a guard tube filled with $CaCl_2$). The following step was heating to 70ºC for 3 hours. Then, the mixture was cooled in ice for 40 minutes. Some precipitate appeared, 30mL of diethyl ether were added and the mixture was cooled again. The solid was separated by vacuum filtration and washed with diethyl ether.

Finally, column chromatography was done to separate the different compounds of the liquid phase. The mobile phase was ethyl acetate/cyclohexane (1:1), the polarity was increased until ethyl acetate/cyclohexane (3:1) and the column was washed with methanol solution 10% in ethyl acetate.

5.2 Reaction between DMHD and phenylhydrazine

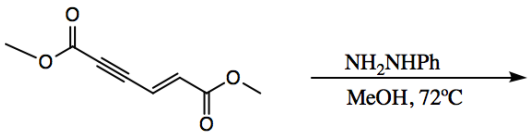


Figure 26 Reaction scheme

Phenylhydrazine (0.53, 4,76mmol, 1eq.) and methanol (10mL) were added to a 100mL flask with a guard tube filled with $CaCl_2$. The solution was stirred for five minutes and DMHD (0.8g, 4,76mmol, 1eq.) was added. The orange mixture was stirred for one hour but the reagent didn't react (checked by TLC). For this reason, the reaction mixture was heated to 72ºC overnight (19 hours).

Then, the reaction mixture was cooled in ice for one hour. 35mL of diethyl ether were added to the mixture, some precipitate appeared and the mixture was cooled in ice again. The following step was filtrating the solid by vacuum filtration and cleaning it with diethyl ether.

Finally, the liquid phase was purified by column chromatography. The mobile phase used was cyclohexane/ethyl acetate (6:1). The polarity was increased until ethyl acetate/cyclohexane (4:1).

5.3 Synthesis of (E)-dimethyl hex-2-en-4-ynedioate[8]

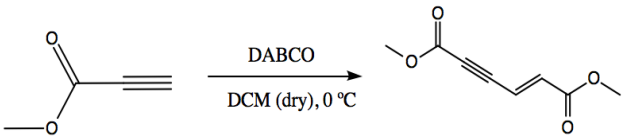


Figure 27 Reaction scheme.

Dry DCM (40mL) and methyl propiolate (2mL, 20.48mmol, 1 eq.) were added to a 250mL two-neck flask. The flask was put in a bath of water, NaCl and ice, due to the fact the solution had to be at 0ºC.

The colourless solution was stirred for 5 minutes. After that, DABCO (24.68mg, 0,22mmol, 0.01eq.) was added to the stirred solution and the mixture was stirred for 5 minutes. The mixture changed its colour with the time. Firstly it was colourless, later it was pink and finally it became brown.

After the mixture was stirred for 5 minutes the presence of methyl propiolate in the reaction mixture was checked with a TLC (mobile phase: cyclohexane/diethyl ether 7:1).

When presence of methyl propiolate could not be appreciated in reaction mixture, solvent was evaporated in vacuum line.

After that, the brown solid obtained was purified by column chromatography with cyclohexane/diethyl ether 7:1 as mobile phase.

5.4 Reaction between DMHD and hydrazine

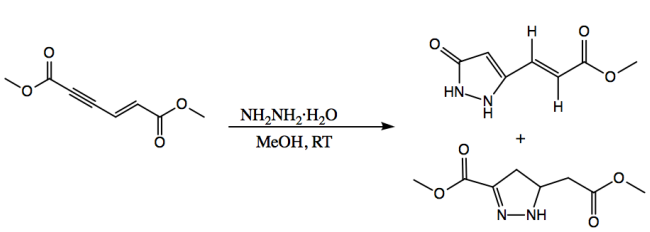


Figure 28 Reaction scheme.

Monohydrated hydrazine (0.3g, 5,99mmol, 1eq) and methanol (10mL) were added to a 100mL flask with a guard tube filled with $CaCl_2$. The solution was stirred for 5 minutes and then DMHD (1g, 5,95mmol, 1eq.) was added to the stirred solution. The reaction mixture was yellow.

After the reaction mixture was stirred for 4 hours some precipitate appeared and it was stirred overnight.

Reaction mixture was filtered by vacuum filtration and the white solid was cleaned with methanol.

Trying to isolate some solid that could remain in the liquid phase methanol was evaporated (except 2mL) and 30mL of diethyl ether were added. The mixture was stirred for 15 minutes, then it was cooled in ice and brown gelatine appeared.

After that, solvent was evaporated and the gelatine was dissolved in methanol. Then it was filtered by vacuum filtration and more white solid was isolated.

Finally, a chromatography column was done to isolate more white solid and other components that were in the liquid phase. The mobile phase used was ethyl acetate/cyclohexane (5:1), its polarity was increased until ethyl acetate/cyclohexane (7:1) and the column was washed by 10% methanol in ethyl acetate.

5.5 Reaction between DMHD and methylhydrazine

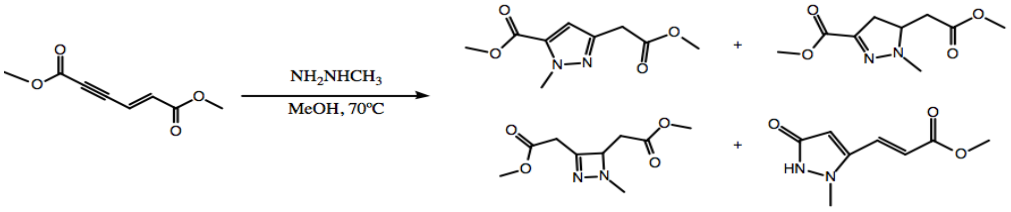


Figure 29 Reaction scheme.

Methyl hydrazine (0.28g, 6.08 mmol, 1eq) and methanol (10mL) were added to a 100mL flask with a guard tube filled with $CaCl_2$. The solution was stirred for 5 minutes and later DMHD (1g, 5,95mmol, 1eq.) was added to the stirred solution. The reaction mixture was dark yellow and it was stirred overnight.

After that, the DMHD did not react (checked by TLC) and the mixture was heated to 70ºC for four hours. The DMHD remained without react, so the mixture was cooled and two drops of methylhydrazine were added with a Pasteur Pipette. The mixture was stirred for two hours but some reagent remained without react. For this reason, five drops of methylhydrazine were added and the mixture was heated to 70ºC for four hours.

The next step was evaporated almost all methanol (2mL were left) and 30mL of diethyl ether were added. An oil appeared but it was impossible to achieve its solidification.

Finally a chromatography column was done using cyclohexane/ethyl acetate (2:1) as mobile phase. The polarity was increased until ethyl acetate/cyclohexane (4:1) and the column was washed by 10% methanol in ethyl acetate.

5.6 Reaction between methyl propiolate and N,N'-dimethylthiourea

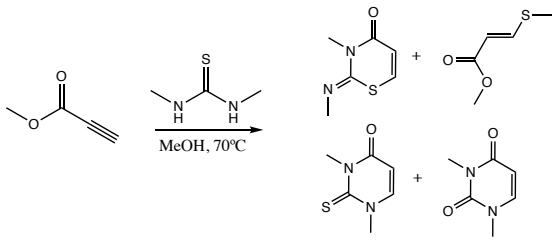


Figure 30 Reaction scheme.

A solution of N,N'-dimethylthiourea (1.24g, 11.84mmol, 1eq) in methanol (10mL) were introduced to a 100mL flask. The flask was introduced in an ice bath and methyl propiolate (1g, 11.89mmol, 1eq) was added to the solution. The mixture was stirred overnight. The reaction was monitoring by TLC and after being stirred overnight N,N'-dimethylthiourea remained without react. For this reason, the reaction mixture was heated until 70ºC for 6 hours.

After that, the different components of the reaction mixture were separated by column chromatography. The mobile phase used was cyclohexane/ethyl acetate 5:1, its polarity was increased until ethyl acetate/cyclohexane 5:1 and it was washed with a 10% methanol solution in ethyl acetate.

5.7 Dimethyl 2,4-hexadiyndioate synthesis[9] and its reaction with N,N'-dimethylthiourea

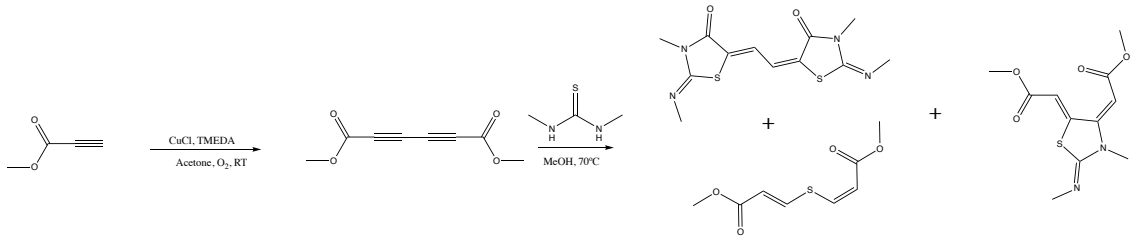


Figure 31 Reaction scheme.

Hay catalyst was prepared by stirring at room temperature CuCl (0.118g, 1.189mmol, 0,1eq) and tetramethylethylenediamine (TMEDA) (0.06mL, 0.4mmol, 0.03eq) in acetone (2.5mL) for 40 minutes.

A solution of methyl propiolate (1g, 11.89mmol, 1eq) in acetone (7mL) was introduced into a 50mL three-necked flask provided with a cold finger and the Hay catalyst was added. Air was bubbling to reaction mixture for 2 hours. After that, the solvent was evaporated and the resulting residue was dissolved in diethyl ether 10mL and it was filtered. The liquid phase was washed twice with HCl (5%, 2 x 12.5mL). The organic layer was dried over anhydrous $MgSO_4$.

A solution of N,N'-dimethylthiourea (1.24g, 11.84mmol, 1eq) in methanol (10mL) were introduced to a 100mL flask. The flask was introduced in an ice bath and the organic layer was added.

The mixture was stirred for 2 hours and a yellow precipitate appear. It was filtered twice by vacuum filtration and cleaned with diethyl ether.

However, the TLC showed that there was N,N'-dimethylthiourea without react in the liquid phase and it was stirred overnight.

After that, N,N'-dimethylthiourea remained without react in the reaction mixture. For this reason the diethyl ether was evaporated, methanol (10mL) was added and the liquid phase was heated until 70ºC for 6 hours. The TLC showed different spots and it was decided to run a chromatography column. The mobile phase used was cyclohexane/ethyl acetate 8:1, the polarity was increased until ethyl acetate/cyclohexane 5:1 and it was washed with a 10% methanol solution in ethyl acetate.

5.8 Reagents properties[10]

Table 2 More significant reagents properties.

Substance	*Purity (%)*	*Toxicity*	*Handling*
Methyl propiolate	99		Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.
DABCO			Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.
Hydrazine solution	1.0 M in THF		Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.
Phenylhydrazine	97		Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.
Hydrazine monohydratated	98		Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.
Methylhydrazine	98		Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.
TMEDA	99		Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.
CuCl	≥99%		Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.
N,*N*′-Dimethylthiourea	99		Use laboratory coat, safety goggles, nitrile gloves and fume cupboard.

6. Results and discussion

In this section the results of different reactions are going to be described and discussed. In addition, the characterization of the different products that were obtained is going to be explained too.

6.1 Synthesis of (E)-dimethyl hex-2-en-4-ynedioate (DMHD)

DMHD's synthesis was done twice following the literature[8]. The first attempt was done in little scale with the aim of checking that the procedure described was good. Once this fact was checked, the reaction was repeated in a larger scale. The results of these two attempts are shown in table 2.

Table 3 Results about two attempts of DMHD's synthesis.

Attempt	*Theoretical product amount (g)*	*Product obtained (g)*	*Yield (%)*
1	1.89	1.73	92
2	5.00	4.98	99

In both cases white solid was obtained. It should be said that the literature procedure followed[8] was very efficient. DMHD was obtained with large yields, the procedure was quick and simple (five minutes of reaction and purification by column chromatography) and the product isolated was very pure. It has to be highlighted that DMHD isolated with this procedure was purer than the DMHD that it was had in the beginning. TLC justified this higher purity. Two spots were shown in the TLC for the old DMHD and only one spot was observed for the DMHD synthetized by the new procedure.
Furthermore, the new procedure was quicker and more efficient than the one that it was followed few years ago to achieve DMHD.

6.1.1 DMHD characterization

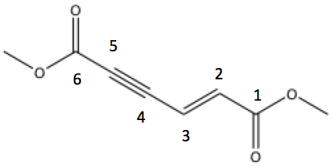


Figure 32 DMHD structure.

DMHD was characterised by ^{13}C and ^{1}H NMR spectra. Its characterization and its comparison with the literature are going to be explained in this section. It has to be said that the results for only one of the attempts are going to be shown, because the characterization for both attempts has been nearly the same.

On the one hand, ^{13}C spectra were registered in deutered chloroform ($CDCl_3$) as in the literature, but the radiation source used was 100MHz and in the literature was used 75MHz. Chemical shift (δ) values and their assignments for DMHD are described in table 3.

Table 4 Chemical shift (δ) ^{13}C values for DMHD in the literature and experimental ones.

Assignment	C1	C6	C3	C2	C5	C4	OCH_3	OCH_3
Literature δ (ppm)	165.10	153.50	135.10	121.70	86.70	81.80	53.00	52.30
Experimental δ (ppm)	165.10	153.53	135.04	121.71	86.70	81.75	53.10	53.20

Looking to table 3 it could be said that ^{13}C chemical shifts obtained for the synthetized DMHD were nearly the same as the ones in the literature.

On the other hand, ^{1}H spectra were registered in the same way that in the literature, deutereted chloroform ($CDCl_3$) at 300MHz. Chemical shift (δ) values and their assignment for DMHD are shown in table 3. Furthermore, integration, multiplicity and coupling constant are described for each signal.

Table 5 Chemical shift (δ) ^{1}H values for DMHD in the literature and experimental ones.

	δ (ppm)	*Integration*	*Multiplicity*	*Coupling constant (J) in Hz*	*Assignment*
Literature	6.79	1 H	d	16.00	C3H
Experimental	6.80			16.01	
Literature	6.47	1 H	d	16.00	C2H
Experimental	6.48			16.01	
Literature	3.82	3 H	s		OCH_3
Experimental	3.83				
Literature	3.79	3 H	s		OCH_3
Experimental	3.80				

Observing table 4 it could be said that the results for ^{1}H spectra for the synthetized DMHD match very well with literature values.
Taking in count the results obtained for ^{13}C and ^{1}H spectrum for the synthetized DMHD and their comparison with the literature it could be said that DMHD was correctly synthetized.

6.2 Reaction between DMHD and hydrazine

Reaction between DMHD and hydrazine was repeated three times. The results in the first attempt were not the ones expected. For this reason, the reaction was repeated changing some conditions. Finally, the reaction was repeated a third time with the conditions of the second time but in a larger scale with the aim of confirm some results. There were isolated a product and a by-product. The first one, it was the desired product and it was a white solid. This solid is a lactam called methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate and their results are shown in table 6.

Table 6 Results for the lactam called methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate.

Attempt	*Theoretical product amount (g)*	*Product obtained (g)*	*Yield (%)*
1	1.0000	0.0138	1
2	1.0000	0.3414	34
3	2.0000	0.7802	39

The by-product is an oily diester called methyl-2-(1'H-4',5'-dihydro-3'-methoxycarbonyl-pyrazol-5'-yl) acetate and their results are exposed in table 7.

Table 7 Results for the diester called methyl-2-(1'H-4',5'-dihydro-3'-methoxycarbonyl-pyrazol-5'-yl) acetate

Attempt	*Theoretical product amount (g)*	*Product obtained (g)*	*Yield (%)*
1	1.1905	0.1500	13
2	1.1905	0.0000	0
3	2.3811	0.9791	12

It could be observed that the results are very different between the first attempt and the second and third one. A very little amount of the main product was isolated in the first attempt so reaction conditions were changed.
Two main changes were done between the first attempts and the other two. First of all, the reagents used were different. In the first attempt a DMHD synthetized few years ago and hydrazine solution 1M in THF were used. For the other two attempts new DMHD was synthetized with a method that allowed obtaining a purer compound and monohydrated hydrazine was used instead of hydrazine solution 1M in THF.
The other different in the procedure followed between the first and the others attempts was that in the first one the reaction mixture was heated until 72ºC and in the other attempts the reaction was done at room temperature.
Other aspect that could seem weird is that in the second attempt the by-product was not isolate but it has got an explanation. The main product is insoluble in the reaction mixture so it was isolated by filtration cause of its precipitation (it was not possible in the first attempt, the precipitates obtained in the first attempt were plenty of impurities or they were mixtures of different compounds). After the filtration, it was seen by TLC that some solid remain in the reaction mixture mixed with other by-products. For this reason a column chromatography was done to separate the components of the mixture. It has to be said that the only interesting components of the mixture were the solid and the oil by-product. There were isolated other components but they were only very little amounts and they were not pure or they were mixtures.
In the first and third attempts the solid and the oil were separated in the column chromatography. This separation was not possible in the second attempt and a mixture of both of them was obtained. Despite this fact, they could be separated adding to them diethyl ether and stirring them for overnight. After that, the solid was isolated by vacuum filtration and the solvent of the liquid phase was evaporated to isolate the oil. The problem was that when the oil NMR 1H spectrum was checked it seemed that the oil has decomposed or it was mixed with other compounds.
To continue with results discussion the characterization of the solid product and the oil by-product are going to be explained in two next sections.

6.2.1 Methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate characterization

The white solid lactam was characterized by X-Ray crystallography and by ^{1}H and ^{13}C NMR spectra.
All X-ray characterizations were done by single crystal X-ray diffraction data were measured on an Agilent Xcalibur diffractometer equipped with a Sapphire detector and an 700 series Cryostream low temperature system.

NMR characterization

^{1}H and ^{13}C results for the white solid are shown next to figure 33.

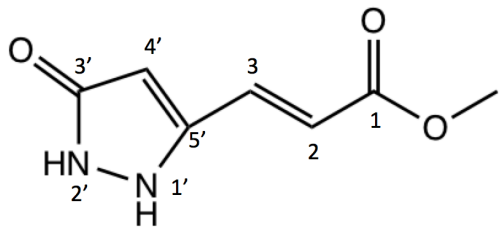


Figure 33 Methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate structure.

^{1}H NMR (300MHz, DMSO):
δ = 12.21 (1H, S, NH), δ = 9.81 (1H, S, NH),
δ = 7.34 (1 H, d, J=16.00Hz, C3),
δ = 6.41 (1 H, d, J=16Hz, C2),
δ = 5.93 (1 H, s, C4'), δ =3.69 (3 H, s, OCH_3).
^{13}C NMR (100MHz, DMSO): δ = 166.54(C1),
δ = 162.07 (C3'), δ = 138.18 (C5'),
δ = 131.58 (C3), δ = 117.30 (C2),
δ =91.07 (C4'), δ = 51.53 (OCH_3).

Talking about the signals of ^{13}C spectrum, the carbonyl signals could be highlighted. As it was expected, the two carbonyls appear at different chemical shift due to the fact that lactam carbonyl is more shielded than the ester carbonyl.
Focusing in the pyrazol ring, it could be said that the signals for C4' and C5' are the ones expected taking in count the literature about pyrazol[11].
Furthermore, the signals for α-carbonyl and β-carbonil (position C2 and C3) are the typical for this kind of carbons.
Finally, the typical signal for methyl ester carbon appeared at 51.53ppm.

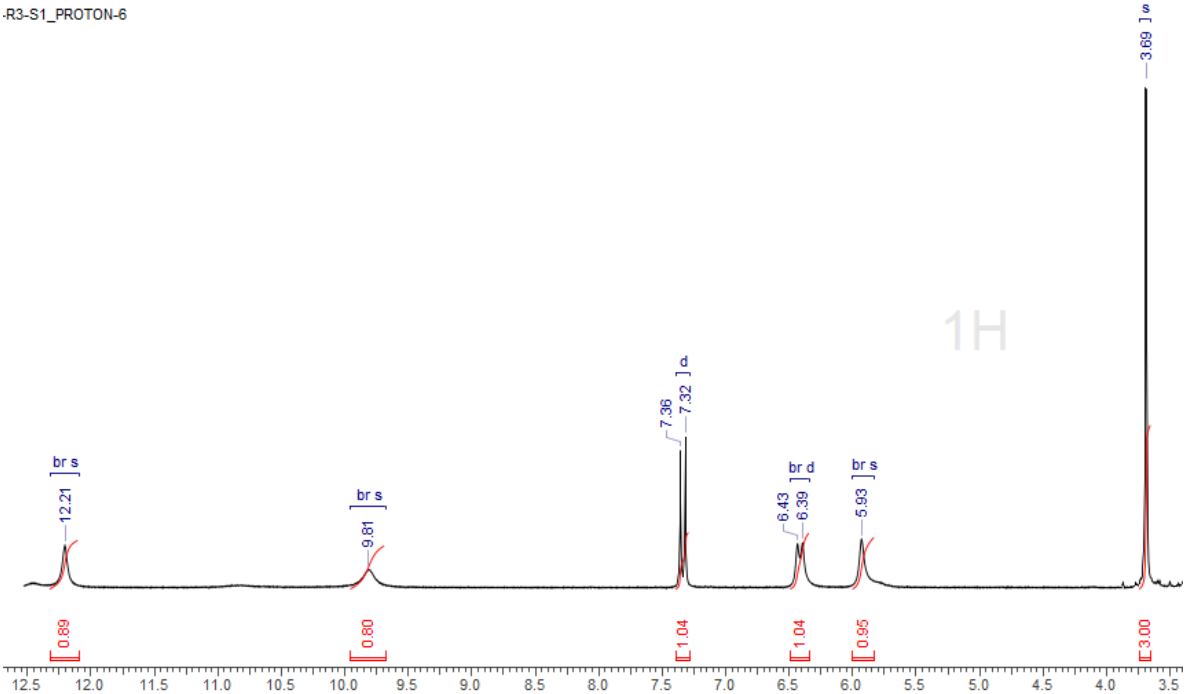


Figure 34 Methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate structure.

In figure 34 it could be seen the lactam ^{1}H NMR spectrum. In this spectrum there are two broad signals 12.21 and 9.81 ppm that fit with the protons bonded to nitrogen.
Other fact to highlight about this spectrum is that there are two doublets. The coupling constants for these doublets are 16Hz. This is the typical coupling constant for trans alkenes. This is the reason why these two doublets are related for the protons in positions C2 and C3 that are couple to each other.

Furthermore a singlet that integrates one proton is observed at 5.87ppm and this signal fits with proton at position C4'.
Finally, at 3.69ppm could be observed a singlet that integrates 3 protons which correspond to protons about ester's methyl.

X-Ray crystallography characterization

Crystal from the solid was grown in methanol. It was analysed by X-Ray crystallography. The structure obtained for the crystal was the one that is shown in figure 35 and its data is described below figure 35.
The crystal structure showed an enol instead of showing the lactam structure that it was expected. For this reason the bond-lenghts measured in the crystal were compare with the literature[12]. This comparison it is described in tables 8,9 and 10.

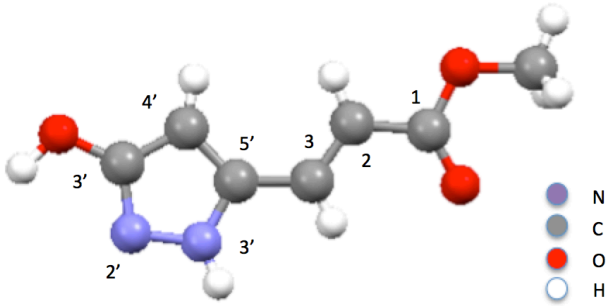


Figure 35 Methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate crystal structure.

Crystal data for **methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate** : $C_7H_8N_2O_3$, M_r = 168.15, triclinic, a = 5.8909(5), b = 7.8681(8), c = 9.0673(8) Å, V = 389.22(7)Å^3, Z = 2, $P\bar{1}$, D_c = 1.426 g/cm^3, μ = 0.114 mm^{-1}, T = 150 K, 1794 reflections (R_{int}=0.0226), 1413 with $F^2 > 2\sigma$, R(F, $F^2 > 2\sigma$) =0.1149 , R_w (F^2, all data) = 0.1259.

Table 8 C-H and O-H bond lengths

Bond	C-H (OCH_3)	C2-H	C3-H	C4'-H	O-H
Experimental (Å)	0.980	0.963	0.898	0.945	0.963
Literature (Å)	1.066	1.077	1.077	1.077	0.967

Table 9 C-X bond lengths (X=O,N)

Bond	O-CH_3	C=O	C1-O	C5'-N	C3'=N	C3'-OH
Experimental (Å)	1.445	1.214	1.334	1.345	1.341	1.343
Literature (Å)	1.453	1.199	1.332	1.357	1.329	1.333

Table 10 C-C and N-N bond lengths

Bond	C1-C2	C2-C3	C3-C5'	C5'-C4'	C4'-C3'	N-N
Experimental (Å)	1.471	1.330	1.452	1.378	1.392	1.356
Literature (Å)	1.488	1.340	1.455	1.369	1.410	1.366

On the one hand, observing the previous tables could be highlighted that the crystal bond lengths correspond to an enol form due to the fact the bond lengths that were obtained fit very well with the ones in the literature.
On the other hand, the ^{1}H and ^{13}C spectra fit very well with the lactam form.
For this reason it is thought than and equilibrium between two tautomers is possible and these two tautomers could be in equilibrium because of a tautomerization reaction (Figure 36).

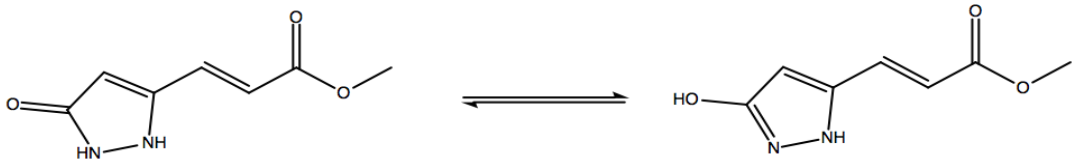


Figure 36 keto-enol tautomerism methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate structure.

The only possible explanation to the fact that in NMR is observed the keto form and in X-Ray crystallography is shown an enol form is: the keto form is the most important when the compound is in solution and when the compound is crystallised the major form is the enol one.

6.2.2 Methyl-2-(1'H-4',5'-dihydro-3'-methoxycarbonyl-pyrazol-5'-yl) acetate characterization

The characterization of the yellow oily diester was done by ^{1}H and ^{13}C NMR spectra. In addition this characterization was supported by a two-dimensional NMR spectroscopy technique. This technique was HMQC (Heteronuclear Multiple-Quantum Correlation) and it could show the connections between hydrogens and carbons. This is possible because this two-dimensional technique shows chemical shift correlation between protons directly bonded to a carbon.
NMR results obtained are described under figure 37.

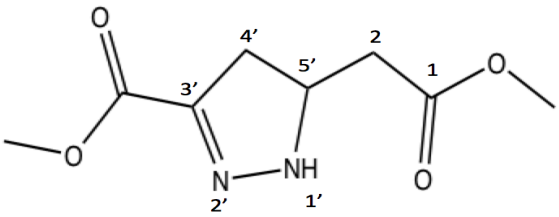


Figure 37 Diester structure.

^{1}H NMR (300MHz, $CDCl_3$): δ = 6.66 (1H, S, NH), δ = 4.28 (1 H, m, C5'), δ = 3.82 (3H, s, CH_3OCOC3'), δ = 3.71 (3 H, s, $OC1OCH_3$), δ =3.08 (1 H, dd, J=16Hz, J=12Hz C4'H), δ =2.60 (3 H, m, $C2H_2$/C4'H).
^{13}C NMR (100MHz, $CDCl_3$): δ = 171.94(C1), δ = 163.12 ($H_3COCOC3'$), δ = 142.0 (C3'), δ = 58.05 (C5'), δ = 52.30 ($H_3COCOC3'$), δ =52.14($OC1OCH_3$), δ = 38.82 (C2),
δ = 36.19 (C4').
It could be seen that ^{13}C signal for the two carbonyl carbon are different. The explanation for this difference is that carbonyl (C1) is not conjugated with a double bond, but the other carbonyl is conjugated. For that reason, the C1 chemical shift is higher than chemical shift of the other carbonyl.
The rest ^{13}C signals are the typical ones for the type of carbon than they represent. These carbons could be classifieds in four groups:

- Carbons about ester's methyls.
- Two secondary carbons with sp^3 hybdridation (C4' and C2).
- One tertiary carbon with sp^3 hybdridation (C5').
- One disubstituted sp^2 carbon (C3').

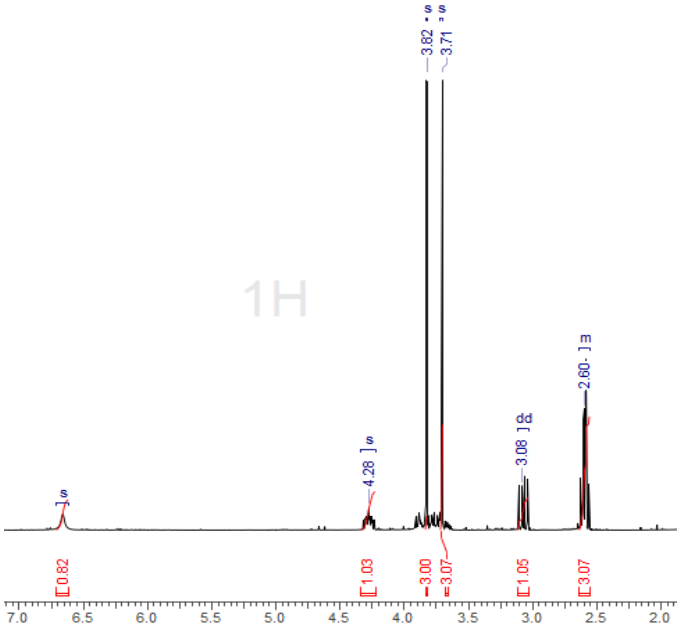


Figure 38 Diester ^{1}H NMR spectrum.

In figure 38 is shown the diester ^{1}H spectrum. In this figure it is worth highlighting the proton bonded to nitrogen broad peak at 6.66. As we expected the integration is nearly one. Furthermore, the two singlets at 3.82 and 3.71 that integrate three each one corresponding to ester methyls.

The other signals are going to be explained after the expansion that is shown in figure 39.

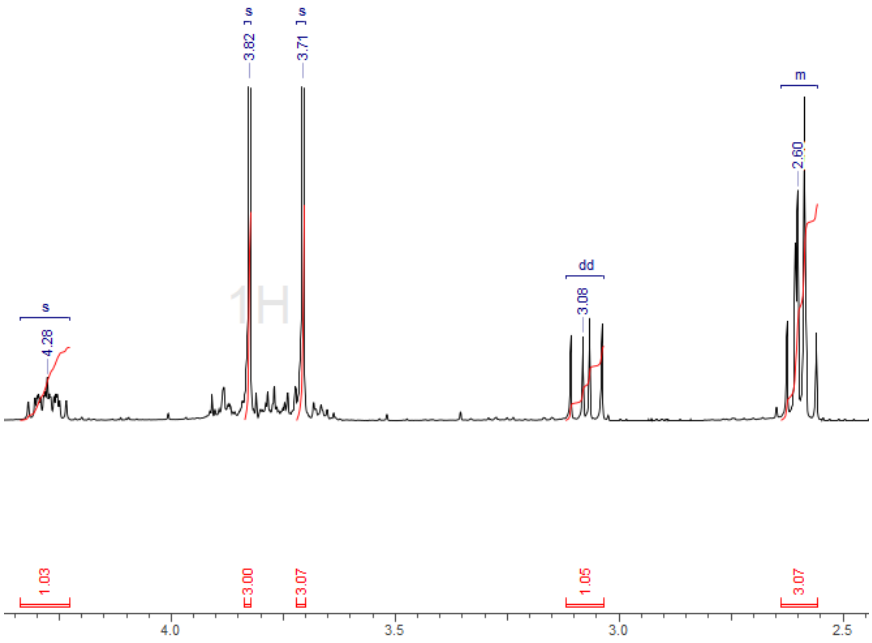


Figure 39 Expansion diester ^{1}H NMR spectrum.

It could be observed that 4.28ppm signal is a multiplete that integrates one, so it fits very well with proton bonded to C5' due to the fact that it could be coupled with the two protons in C2 and the other two in C4'.

About the signal at 3.08ppm it could be said that is a doublet doublet and its integration is 1. For this reason, it fits with a proton bonded to C4' that could be coupled with the

other proton attached to same carbon and proton bonded to C5'. In addition, if attention is paid on the coupling constants (J=16Hz and J=12Hz), it could be said that the higher one correspond to the coupling with the proton bonded to same carbon and the lowest one could represent the coupling with the proton in the neighbour carbon.
Finally, the signal at 2.60ppm it is thought that contain three protons because its integration is 3. These three protons could fit with the two in C2 and the one in C4'. The two in C2 are couple to each other and to proton on C5'. Furthermore, the proton in C4' could be coupled with proton in C5'. For this reason the signal showed it is a multiplet.
Finally, it is important highlighting that the previous deductions were supported by HMQC. ^{1}H signal at 2.60ppm was correlated with two different carbons, so it helped on the deduction that this signal represent the protons bonded to two different carbons. Furthermore, 3.08 and 4.48ppm were correlated with a carbon each one.

6.2.3 Applications

Two interesting molecules were obtained in this reaction.
On the one hand, speaking about the major product it could be said that this solid lactam it could not have properties as a new antibiotic. Anyway, it could be modified to achieve this goal (Figure 40). Furthermore, a simple method to synthesize this lactam has been found.

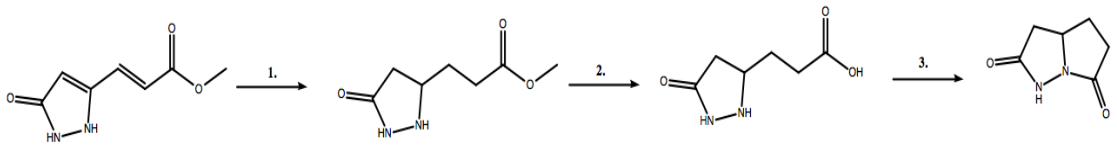


Figure 40 Possible modifications to the solid lactam with the aim of achieving a molecule for testing its antibiotic activity.

In the following lines the steps to achieve the bicyclic compound and the reason because it could be an interesting structure are going to be explained.

- 1st step (reduction)→It is necessary to reduce the double bounds of the molecule, for this reason is needed to found a mild reducer that allows the selective double bond reduction without cause the carbonyl reduction.
- 2nd step (oxidation) → The ester must be oxidised with the aim of increase the electrophilic character of the ester carbonyl.
- 3rd step (cyclization)→The final step is a cyclization that it could be done using an intramolecular nucleophilic acyl substitution.

It is thought that this bicyclic structure could work as an antibiotic due to the fact that its structure it is similar to penicillin. For this reason, it could be an inhibitor of bacterial cell wall synthesis.
The aim property of this bicyclic compound is that it contains two weak amide bonds. This is one of the important properties that allow penicillin activity.
In this case, the weakness of these bounds it is caused because the overlap between carbonyl π -orbitals and nitrogen lone pair is small. There are two reasons that explains this poor overlap:

- Nitrogen hybridization of these two lactams is sp^3, so its geometry is trigonal planar due to the strain caused by the rings.
- α-effect[6] appears due to the fact that there are two nitrogens bonded. This effect originates a lone pair polarization of the nitrogen and this fact

makes the overlap between carbonyl π-orbitals and nitrogen lone pair difficult.

On the other hand, speaking about the oily diester product the first thing that it should be said is that its yield was little. Anyway, it is a molecule that could be transformed trying to achieve a molecule that could be test as a new antibiotic (Figure 41).

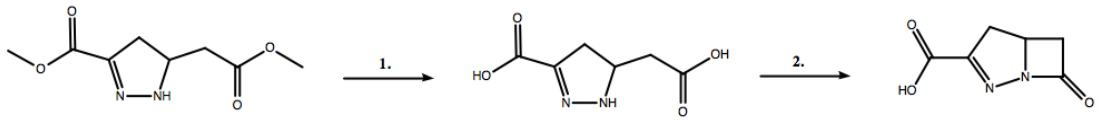


Figure 41 Possible modifications to the oily diester with the aim of achieving a molecule for testing its antibiotic activity.

Two modifications must be done in the diester to achieve a structure very similar to penicillin.

- 1st step→ esters oxidations are needed to increase the electrophilic character of carbonyls.
- 2nd step→ cyclization that it could be done using an intramolecular nucleophilic acyl substitution.

It could be observed that the structure of this possible new compound is very similar to penicillin, for this reason, it is thought that it could works as an inhibitor of bacterial cell wall synthesis.

As in the lactam case, the most important property of this new compound is its weak amide bone. The reasons for this weakness are the same that the ones explained before: the sp^3 hybridization of amide nitrogen and the α-effect[6].

Furthermore, this new compound contains other similarity with penicillin. This similarity is that it contains a free acid carboxylic group that it could be deprotonated.

6.3 Reaction between DMHD and phenylhydrazine

It could not be isolated any interesting products in the reaction between DMHD and phenylhydrazine. It was obtained a little amount (0.04g) of a precipitate from the reaction mixture. It was analysed by NMR and its ^1H spectra showed that it was not a pure substance.

The liquid phase of the reaction mixture seemed to contain different components because some spots were observed in the TLC. A chromatography column was run trying to separate the different compounds. After that, it seemed that two little amounts (0.13 and 0.18g) of two components were isolated. Once these fractions were analysed by NMR, it was shown that they were mixtures of more than one compound.

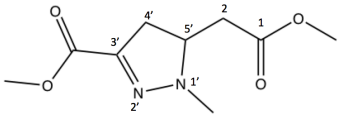
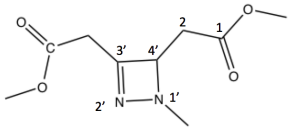
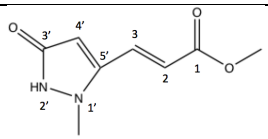
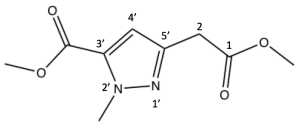
It has to be highlighted that this reaction was done with the DMHD synthetized few years ago and the reaction mixture was heated until 72ºC. These conditions are very similar to the ones following in the first attempt of reaction between DMHD and hydrazine that it did not work. For this reason, it could be worth to try the reaction with phenylhydrazine with the new DMHD and new reaction conditions.

6.4 Reaction between DMHD and methylhydrazine

Four different products were obtained in the reaction between DMDH and hydrazine. These four products are three yellow oily diesters and one solid, the names of these three diesters are: methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) acetate, methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl) enoate and methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate. The solid is white, it

is a lactam and it is called methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate.
The results about this reaction are described in table 7.

Table 11 Reaction between DMDH and hydrazine results

Product name	***Structure***	***Theoretical product amount (g)***	***Product obtained (g)***	***Yield (%)***
Methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) acetate		1.2739	0.2596	20
Methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl) enoate		1.2739	0.1600	13
Methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate		1.0834	0.0602	6
Methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate		1.2620	0.0608	5

Considering the previous studies about reactivity of DMHD against two nucleophilic centres, it was expected that the major product would be the solid lactam.
Contrary to this, the solid it is not the major product. An explanation to this fact could be that on the one hand, the solid obtained in this reaction is soluble in the reaction mixture. On the other hand the solid product was insoluble in the reaction mixture and its precipitation was easy in the previous reaction with hydrazine.
The other three products are soluble in the reaction mixture too. For this reason, the reaction mixture was a liquid phase that contained the four products and they were separated by chromatography column.
Other aspect that should be highlighted about reaction results is that for the formation of the three oily diesters, one of the methylhydrazine nucleophilic attacks were done at sp^2 carbon of the double bond, exactly at β-carbonyl position.
In the following section is going to be explained the characterization for each product. Different NMR techniques were used for the four products and X-Ray crystallography was used for the solid one too.

6.4.1 Methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) acetate characterization

This yellow oily diester was characterized by different NMR techniques. First of all it was analysed by ^{1}H and ^{13}C NMR spectra. The information obtained with these two one-dimensional NMR spectroscopy analyses was not enough. For this reason, two-dimensional NMR spectroscopy analyses were required. It was used HMQC with the aim of guess the connections between carbons and protons like in the characterization of the oil obtained in the reaction with hydrazine. Furthermore, other two-dimensional NMR technique was used in this case. This technique is called COSY (correlation

spectroscopy) and it could allow us deducing which protons are coupling with each other.
^{1}H and ^{13}C signals and useful information about them is shown under figure 42. After that, ^{1}H NMR spectrum is shown on figure 43.

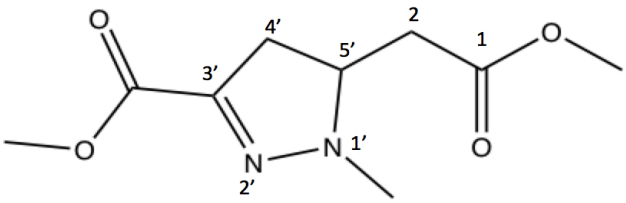


Figure 42 Structure Methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl).

^{1}H NMR (300MHz, $CDCl_3$): δ = 3.81 (3 H, s, OCH_3), δ = 3.81 (1 H, m, C5'), δ = 3.70 (3 H, s, OCH_3), δ = 3.25 (1 H, dd, J = 12Hz, J = 16Hz, $C2H_2$), δ = 3.00 (3 H, s, NCH_3), δ = 2.80 (1 H, dd, J = 4Hz, J = 16Hz, $C2H_2$), δ = 2.68 (1 H, dd, J = 16Hz, J = 16Hz, $C4'H_2$), δ = 2.55 (1 H, dd, J = 8Hz, J = 16Hz, $C4'H_2$).
^{13}C NMR (100MHz, $CDCl_3$): δ = 171.17(C1), δ = 162.95(C3'CO2Me), δ = 139.80(C3'), δ = 64.65(C5'), δ = 52.24(OCH_3), δ = 52.13(OCH_3), δ = 39.43(NCH_3), δ = 38.07($C2H_2$), δ = 37.28($C4'H_2$).

The explanation for ^{13}C signals is nearly the same that the one that was done to diester obtained in the reaction with DMHD and hydrazine.
The only different thing is that in this case there is one more signal that corresponds to the methyl bonded to nitrogen. As it was expected, this signal appears in similar chemical shift than the signals for esters' methyl, the only fact that could be explained is that the chemical shift for methyl attached to nitrogen carbon is lower than the chemical shift for the other two methyls. This effect is logical because the carbon bonded to nitrogen is more shielded than the carbons bonded to esters' oxygen.

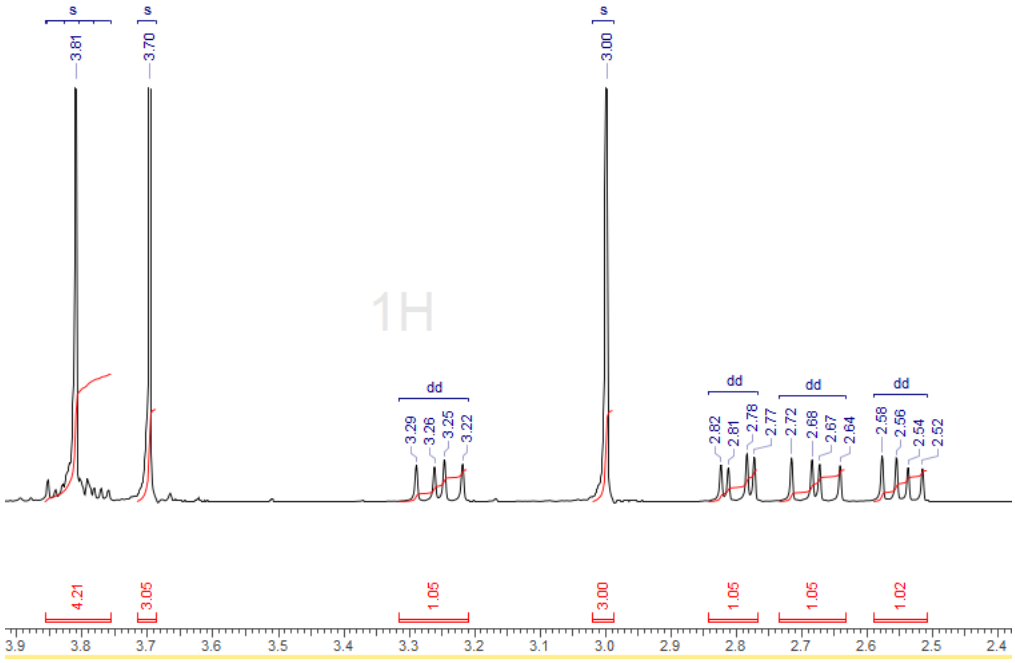


Figure 43 Methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) ^{1}H NMR spectrum.

First of all the clearest signals are going to be explained. Two singlets that integrates three protons each one could be seen. One of them appeared at 3.70ppm and it fits with the methyl of one of the esters. The other singlet appeared at a lower chemical shift (3.00ppm) and it could be related with the methyl attached to nitrogen.

It could seem that at 3.81ppm it was other singlet, but it was deduced that this signal was formed by a singlet and a multiplet for different reasons:

- It could be seen a multiplet in the base line.
- The signal integrated 4 protons.
- Proton connection with two different carbons is shown in HMQC.
- Coupling between this signal and other doublets doublets is shown in COSY.

For all this reasons, the conclusion was that at 3.81ppm there is a singlet that integrates 3 and this fits with the methyl of the ester protons. In addition, in this signal there is a multiplet that integrates one proton and it is related with the proton in position 5'. The multiplicity of this signal could be explained by the fact that this proton is coupled with the protons at positions 2 and 4'.

Talking about the doublets doublets it is shown that there are four of them and the integration for each one is one proton. For this reason it could be thought that this signals represents the four protons in positions 2 an 4'. The explanation about the multiplicity of these signals is simple. Each proton at position 2 and 4' is coupled to the proton attached to the same carbon and to the proton at position 5'. Considering the coupling constants of these signals it could be observed that each signal has got a coupling constant of 16Hz and other smaller coupling constant. The 16Hz coupling constant represents the coupling with the proton attached to the same carbon and the other one represents the coupling with the proton at position 5'.

Finally, the exactly assignment for this doublets doublets was possible because of HMQC that showed signals 3.20 and 2.80 attached to C2 and signals 2.68 and 2.55 attached to C4'.

6.4.2 Methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl) enoate characterization

This oily diester product was totally unexpected because it has got a four-member ring and this type of ring is difficult to be formed. Furthermore, it was surprising that after the previous diester, it was the most abundant product. However, it could be found an explanation considering the methylhydrazine position attacks at DMHD. The two attacks were done at β-carbonyl positions, one at *sp* carbon and the other one at sp^2 carbon. Both positions are electrophilic centres because of the conjugation of the double/triple bond with each ester, so they could be attacked by nucleophiles such as methylhydrazine.

This product was characterized by different NMR techniques too. There were used two one-dimensional techniques (^{1}H and ^{13}C) and used a two-dimensional technique too (HMQC).

The assignment of each signal it is shown after figure 44.

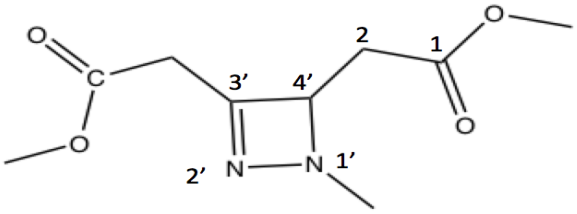


Figure 44 Methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl) enoate structure.

^{1}H NMR (300MHz, $CDCl_3$): δ = 3.76 (3 H, s, OCH_3), δ = 3.69 (3 H, s, OCH_3), 3.63 (1 H, t, J=12Hz, C4'H), δ = 3.38 (1 H, d, J=16Hz, C3'CH_2), δ = 3.30 (1 H, d, J=16Hz, C3'CH_2), δ = 3.06 (2 H, d, J=12Hz, C2H_2), δ = 2.86 (3 H, s, NCH_3).

^{13}C NMR (100MHz, $CDCl_3$): δ = 171.37 (C1), δ = 169.66 (C3'CH_2<u>C</u>O_2Me), δ = 147.07 (C3'), δ = 69.85 (C4'), δ = 52.61 (OCH_3), δ = 52.35 (OCH_3), δ = 43.23 (NCH_3), δ = 39.88(C3'CH_2), δ = 35.82(C2H_2).

The explanation for the ^{13}C signals it is very similar that the ones gave before to the other diesters.

Despites this facts, there are two things that could be interesting to highlight. Because of the formation of a four-member ring, in this case there are two secondary sp^3 carbons (C2 and C3'-<u>C</u>) with similar chemical shift. In addition, it could be observed that in this case the two carbonyl carbons have got very similar chemical shifts. This is not a surprising fact because they have similar chemical environment.

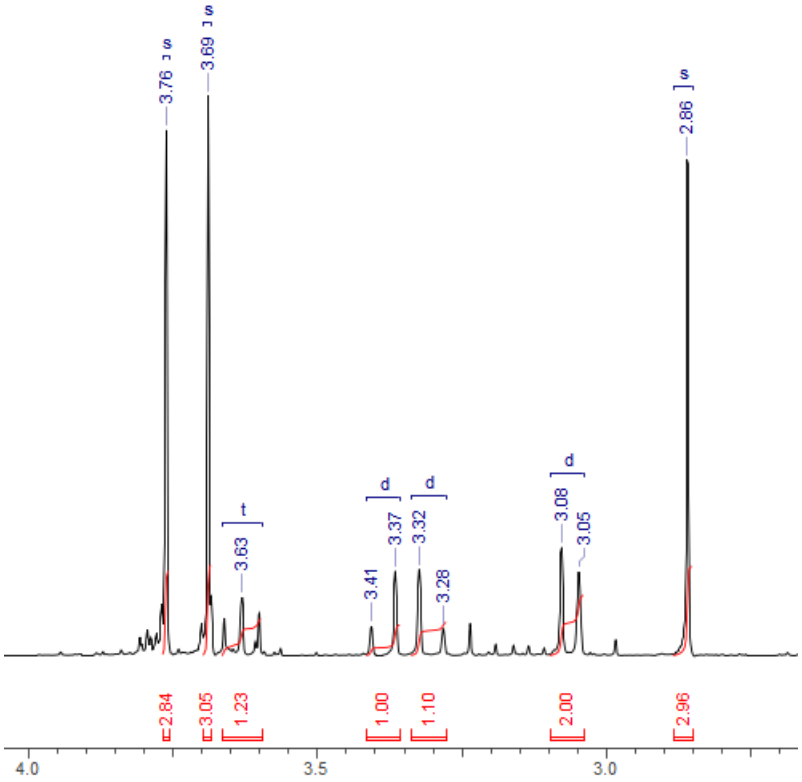


Figure 45 Methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl) enoate structure ^{1}H spectra

The explanation about the ^{1}H NMR spectrum for this diester and his structure deduction is going to be explained in the following lines.

Three singlets that integrate three protons each one appeared at 3.76, 3.69 and 2.86ppm. The two signals with the highest chemical shift are related with the methyls of the two esters. The other singlet at 2.68ppm fits with protons in the methyl bonded to nitrogen.

A triplet could be seen at 3.63 an its integrations is a little bit bigger than one, but it could be caused by some little impurities in the base line. This signal correspond to proton at C4' that is coupled with the two protons in C2. This fact is justified because the couple constants for this triplet and for the doublet at 3.06 pm are 12Hz. The doublet at 3.06ppm integrate two protons and it is related with the two protons at C2.

Finally there are two more doublets at 3.30 and 3.38ppm that integrate one proton each one. These signals fits with the two protons bonded at carbon situated between the ester and C3'. These protons are coupled to each other and its explained by the fact the coupling constant of each doublet is the same (16Hz). An aspect that should be highlighted is that these two doublets seem to be a quadruplet, but they have this shape because of roof effect. This effect appears when the difference on frequency between two signals it is similar to its coupling constant value. This fact cause that outer lines (far from the chemical shift of the other proton) decrease its intensity and inner lines (lines closer form the chemical shift of the other proton) increase its intensity.

6.4.2Methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate characterization

The solid lactam was characterized by ^{1}H and ^{13}C NMR spectra. It was possible to grow crystals about this solid so it was characterized by X-Ray crystallography too.

NMR characterization

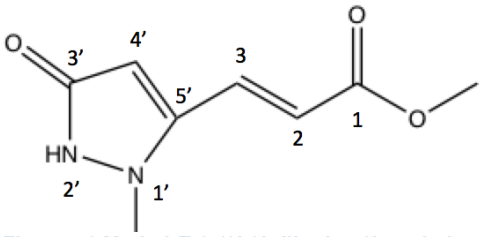


Figure 46 Methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate structure.

^{1}H NMR (300MHz, $CDCl_3$): δ = 11.42 (1 H, s, NH), δ = 7.43 (1 H, d, J=15.78Hz C3), δ = 6.30 (1H, d, J=15.78Hz, C2), δ = 5.87 (1 H, s, C4'), δ = 3.78 (3 H, s, OCH_3), δ = 3.74 (3 H, s, NCH_3).
^{13}C NMR (100MHz, $CDCl_3$): δ = 166.68(C1), δ = 161.35(C3'), δ = 139.40(C5'), δ = 129.33 (C3), δ = 121.17 (C2), δ = 89.43 (C4'), δ = 52.00 (OCH_3), δ = 35.62 (NCH_3).

The explanation for ^{13}C signals is exactly the same that the one did to the solid obtain in the reaction between DMHD and hydrazine (methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate).
The only different with this explanation is that in this case there are two signals for methyl carbons. The one with a higher chemical shift was assigned to OCH_3 because it is less shielded than NCH_3 due to the fact oxygen is more electronegative than nitrogen.

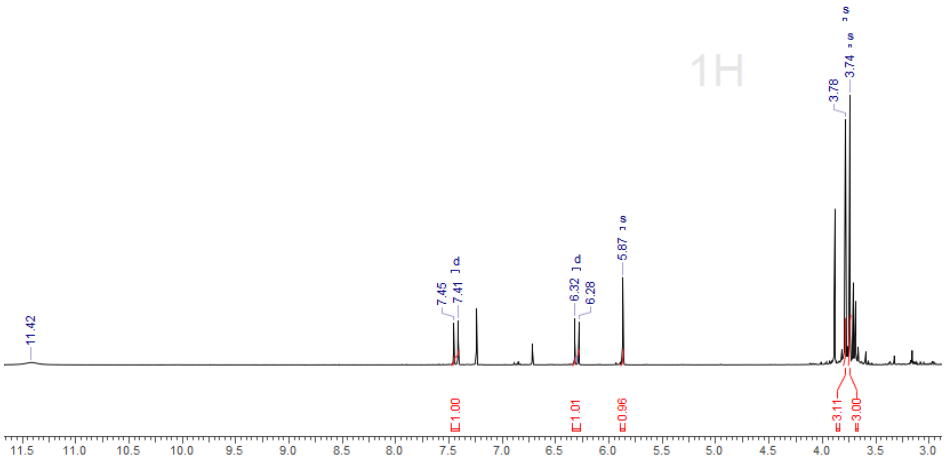


Figure 47 Methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate ^{1}H spectrum.

In figure 47 it could be seen lactam ^{1}H NMR spectrum. In this spectrum there is a broad signal at 11.42ppm that fits with the proton bonded to nitrogen.
They could be observed two singlets that integrates 3 protons each one and they correspond to methyls. 3.78ppm signal fits with OCH_3 and 3.74ppm signal with NCH_3.
Other fact to highlight about this spectrum is that there are two doublets. The coupling constants for these doublets are 15.78Hz, this is the typical coupling constant for trans alkenes. It is why these two doublets are related eith the protons in positions C2 and C3 that are couple to each other.
Furthermore a singlet that integrates one proton is observed at 5.87ppm and this signal fits with proton at position C4'.
Finally it should to be highlighted that in the methyl region some more signals could be observed. Their intensities are lower than the peaks related with CH_3 and NCH_3, so these extra peaks appeared because of the presence of some impurities.

X-Ray crystallography characterization

Crystal from the lactam was grown in methanol and they were analysed by X-Ray crystallography. The structure obtained for the crystal was the one that is shown in figure 48 and its data is described below figure 48.

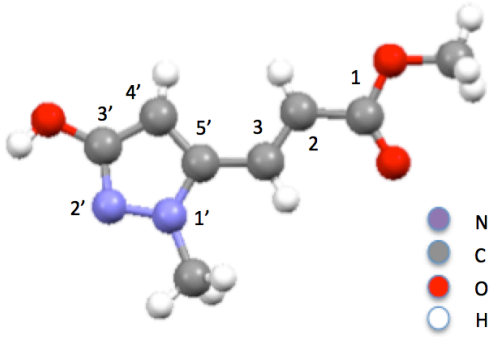


Figure 48 Methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate crystal structure

In the same way that in the solid obtain in reaction between DMHD and hydrazine, the crystal structure did not show a lactam, it showed an enol. For this reason, the bond-lenghts measured in the crystal were compare with the literature[12]. This comparison it is described in tables 12,13 and 14.

Crystal data for **Methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate**: $C_8H_{10}N_2O_3$, M_r = 182.18, triclinic, a = 4.6014(5), b = 9.4905(11), c = 10.7215(12) Å, V = 427.56(10) Å^3, Z = 2, $P\bar{1}$, D_c = 1.306 g/cm^3, μ = 0.104 mm^{-1}, T = 150 K, 1963 unique reflections (R_{int}=0.0342), 1541 with $F^2 > 2\sigma$, R(F, $F^2 > 2\sigma$) =0.0779 , R_w (F^2, all data) = 0.1543.

Table 12 C-H and O-H bond lengths

Bond	C-H (OCH_3)	C-H (NCH_3)	C2-H	C3-H	C4'-H	O-H
Experimental (Å)	0.980	0.980	0.950	0.950	0.950	0.843
Literature (Å)	1.066	1.066	1.077	1.077	1.077	0.967

Table 13 C-X bond lengths (X=O,N)

Bond	O-CH_3	C=O	C1-O	C5'-N	C3'=N	C3'-OH	N-CH_3
Experimental (Å)	1.443	1.204	1.335	1.351	1.331	1.347	1.453
Literature (Å)	1.453	1.199	1.332	1.357	1.329	1.333	1.469

Table 14 C-C and N-N bond lengths

Bond	C1-C2	C2-C3	C3-C5'	C5'-C4'	C4'-C3'	N-N
Experimental (Å)	1.467	1.326	1.443	1.379	1.393	1.353
Literature (Å)	1.488	1.340	1.455	1.369	1.410	1.366

In the same way as was explained to the solid obtain in the reaction between DMHD and hydrazine:

- Results obtained for the crystal show that the structure fits with an enol form.
- ^1H and ^{13}C spectra indicates that the solid is a lactam

In this case it was thought that and equilibrium between two tautomers is possible too (Figure 49).

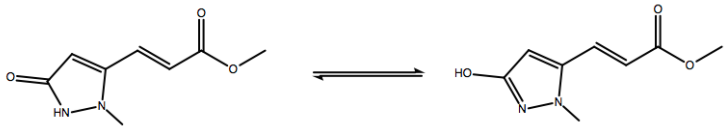


Figure 49 keto-enol tautomerism methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate.

The conclusion about this structure is the same one gave for the solid of the other reaction: the keto form is the most important when the compound is in solution and when the compound is crystallised the major form is the enol one.

6.4.4 Methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate characterization

The less abundant product isolated was a yellow oily diester too. This product was characterized by three different NMR techniques. First of all ^{1}H and ^{13}C spectra were registered and after that HMQC was done to support the characterization.

The signals assignments for ^{1}H and ^{13}C are shown next to figure 50.

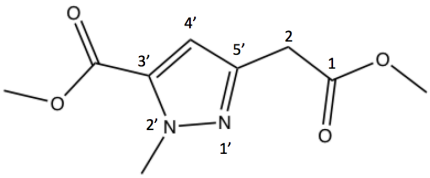


Figure 50 methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate structure.

1H NMR (300MHz, CDCl3): δ =6.78 (1H, s, C4'), δ =4.12 (3 H, s, NCH_3), δ =3.85 (3 H, s, OCH_3), δ =3.70 (1 H, s, OCH_3) δ =3.66 (2 H, s, C2).

^{13}C NMR (100MHz, $CDCl_3$): δ = 171.00(C1), δ = 160.28 (C3'-CO), δ = 143.77 (C5'), δ = 133.16 (C3'), δ = 110.04 (C4'), δ = 52.33 (OCH_3), δ = 52.02 (OCH_3), δ = 39.49(NCH_3), δ = 33.92 (C2).

In this case, the ^{13C} spectra explanation is the same that the explanation for the major product. The only difference is that in this case in position 5' there is a sp^2 disubstituted carbon instead of sp^3 carbon. Anyway, the signal for this carbon is the one expected for this carbon type.

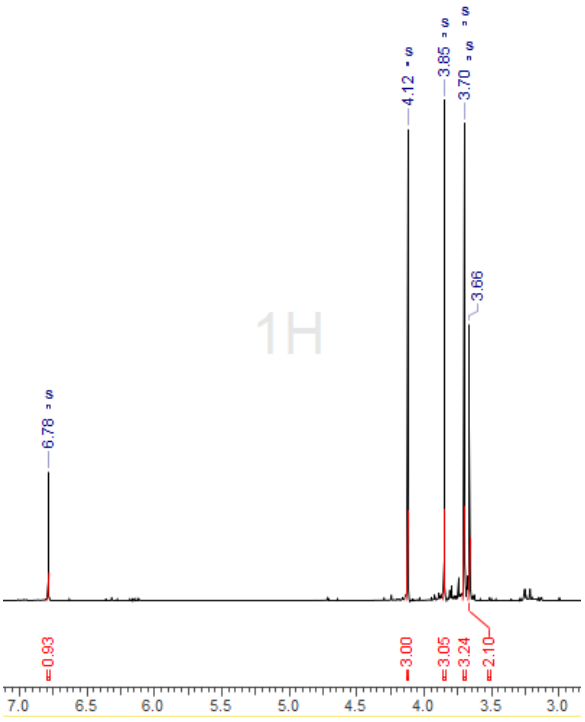


Figure 51 Methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate ^{1}H spectrum.

There are not possible proton couplings in this diester, so all ^{1}H signals are singlets.

There are three singlets that integrate three protons each one and they correspond to methyl's protons. About their chemical shift it could be highlighted that in this case the chemical shift for the protons on methyl bonded to nitrogen is higher than the esters' methyl protons. The assignment for the signal of these two kinds of protons it was possible because HMQC was used.

The other two singlets correspond to $C2H_2$ and to C4'H. They were easy to assign due to its different integration.

6.4.5 Applications

After results discussion about reaction between DMHD and methylhydrazine, it should be highlighted that only one of the products could be converted to a compound with antibacterial activity. The problem is that this compound is the minor product: methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate. It is thought that this diester could be reduced, oxidized and cyclized as is shown in figure 53. Following these steps a compound similar to penicillin could be achieved.

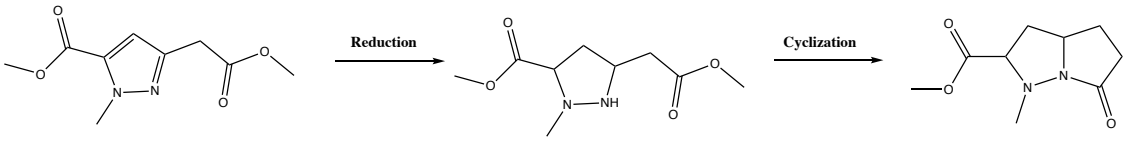


Figure 52 Possible steps to convert methyl-2-(3'-methoxycarbonyl-2'-metyl-pyrazol-5'-yl) acetate to a compound that could be useful as antibiotic.

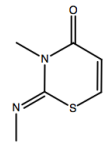
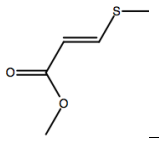
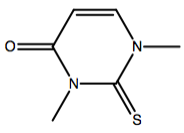
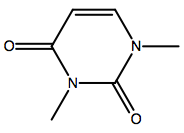
Focusing in the others products, the structure of the main product that is the oily diester called methyl-2-(4',5'-dihydro-3'-methoxycarbonyl-1'-methyl-pyrazol-5'-yl) acetate and the solid one (methyl-E-3-(2',3'-dihydro-1'-methyl-3'-oxopyrazol-5'-yl)-prop-2-enoate), is similar to the interesting solid and oily products obtain in the reaction between DMHD and hydrazine. The problem with the two products obtained in the reaction with methylhydrazine is the position of the methyl attached to nitrogen. This position makes impossible its cyclization and hence, the structure similar to penicillin could not be achieved.

Finally, the product with the four-membered ring (methyl-2-(1',4'-dihydro-3'-(methoxycarbonyl)methyl-1',2'-diazet-4'-yl) enoate) could not be cyclized because the cyclization would mean achieve a structure with two four-membered rings.

6.5 Methyl propiolate reactivity against N,N'-dimehtylthiourea

The aim of this reaction was to know the reactivity of methyl propiolate against N,N'-dimethylthiourea. The reaction results are shown in table 15. Knowing this reactivity could be helpful to understand other interesting reactions, for example the next reaction that would be explaining. This reaction is between dimethyl hexa-2,4-diynedioate and N,N'-dimethylthiourea.

Table 15 Results about reaction between methyl propiolate and N,N'-dimethylthiourea

Product name	***Structure***	***Theoretical product amount (g)***	***Product obtained (g)***	***Yield (%)***
3-methyl-2-(methylimino)-2,3-dihydro-4*H*-1,3-thiazin-4-one		1.8580	0.4084	22
methyl (*E*)-3-(methylthio)acrylate		1.5723	0.2542	16
1,3-dimethyl-2-thioxo-2,3-dihydropyrimidin-4(1*H*)-one		1.8580	0.0538	3
1,3-dimethyl-1,2,3,4-tetrahydro-2,4-dione		1.6669	0.0453	3

Taking account the results, it could be said that the yields for these reaction products are low. Despite this fact, conclusions about the reactivity of methyl propiolate against N,N'-dimethylthiourea could be extracted. It is obvious that there are two major products and two minor products. In addition, these results are very coherent with the expected reactivity.

On the one hand, the two major products are the result of nucleophilic addition of N,N'-dimethylthiourea sulphur at methyl propiolate electrophilic centre. The difference between these products is that the first is the result of trans-addition that allows a posterior cyclization. The second product is the result of cis-addition and the stereochemistry of the resulting compound of this addition does not allow the cyclization. It should be highlighted that in these nuclepophilic additions are involved the most nucleophilic centre in N,N'-dimethilthiourea (S) and the most electrophilic centre in methyl propiolate (β-carbonyl position).

On the other hand, the two minor products are result of nitrogen nucleophilic additions. The nucleophilic character of this centre is lower than the one for N,N'-dimethylthiourea sulphur. For this reason, it is logical that these two are the minor products.

Furthermore, it must be said the formation of last product in the table it is possible because of the formation of N,N'-dimethylurea. This urea it could be originated in the formation of (E)-3-(methylthio)acrylate so it is reasonable that it has got such a low yield.

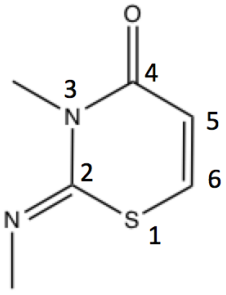
For all these reasons, it is thought that the most probable attack of N,N'-dimethylthiourea to methyl propiolate is done by sulphur at β-carbonyl position, but this attack could be trans or cis.

The characterization of these products is going to be explained in the following section. These characterizations were done by ^{1}H and ^{13}C NMR and mass spectrum. Actually, the structures of the products were predicted by NMR analysis and they were confirmed checking their molecular weight by mass spectrum.

The mass spectra were obtained by gas chromatography (Agilent Technologies 7890B) followed by mass detector (Agilent Technologies 5977B MSD).

6.5.1 (Z)-3-methyl-2-(methylimino)-2,3-dihydro-4H-1,3-thiazin-4-one characterization

The signals for ^{1}H and ^{13}C NMR spectra and the Mass-Spectrum signals for the major product are shown in figure 53.



^{1}H NMR (300MHz, $CDCl_3$): δ = 7.33 (1 H, d, J=8Hz, C6H), δ = 5.39 (1 H, d, J=8Hz C6H), δ = 3.74 (3 H, s, NCH_3), δ = 3.30 (3 H, s, NCH_3).

^{13}C NMR (100MHz, $CDCl_3$): δ = 177.67(C4), δ = 160.40(C2), δ = 143.40(C6), δ = 105.02 (C5), δ = 45.16 ($C2=NCH_3$), δ = 34.84 ($N3CH_3$).

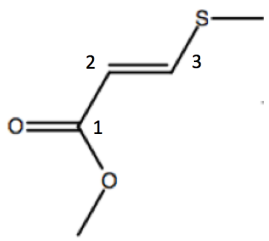
GC-MS (EI): m/z (%)= 156(100), 55 (27).

Figure 53 (Z)-3-methyl-2-(methylimino)-2,3-dihydro-4H-1,3-thiazin-4-one structure. ^{1}H and ^{13}C NMR spectra signals. GC-MS peaks.

Paying attention to NMR results different facts could be highlighted. First of all the two doublets in ^{1}H spectrum and its coupling constants suggests the presence of

disubstituted alkene with Z stereochemistry. This fact was confirmed with the two typical peaks in ^{13}C for α,β–insaturated carbonyl to α and β positions.
Other significant peaks in ^{13}C spectrum were the one for lactam carbonyl carbon (177.67 ppm) and the one for imine carbon (160.40ppm).
Finally, the signals for the protons of two metyls bonded to nitrogen were shown in the ^{1}H spectrum and its carbon signals were observed in ^{13}C spectrum.

6.5.2 Methyl (E)-3-(methylthio)acrylate characterization



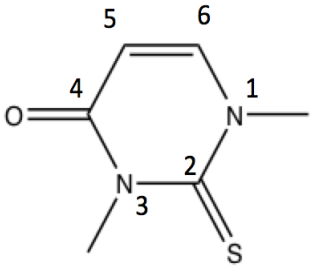
^{1}H NMR (300MHz, $CDCl_3$): δ = 7.72 (1 H, d, J=16Hz, C3H), δ = 5.62 (1 H, d, J=16Hz, C2H), δ = 3.69 (3 H, s, OCH_3), δ = 2.30 (3 H, s, SCH_3).
^{13}C NMR (100MHz, $CDCl_3$): δ = 165.60(C1), δ = 147.19(C3), δ = 112.64(C2), δ = 51.40 (OCH_3), δ = 14.25 (SCH_3).
GC-MS (EI): m/z (%)= 132(73), 117 (59), 101(100).

Figure 54 Methyl (E)-3-(methylthio)acrylate structure. ^{1}H and ^{13}C NMR spectra signals. GC-MS signals.

In this case, two doublets in ^{1}H spectrum suggest the presence of disubstituted alkene too. Now the coupling constant showed that the stereochemistry was E. This fact was confirmed as in the previous case with the ^{13}C spectrum, were typical peaks for α,β–insaturated carbonyl to α and β positions were observed.
Other interesting signal detected in ^{13}C spectrum was the one for α,β–insaturated ester carbonyl (165.60ppm).
Finally, two methyls group could be identified because of their signals in ^{1}H and ^{13}C spectra. One of them showed the typical signals for methyl ester and the other one, which is bonded to sulphur presented low chemical shift in both spectra as it was expected.

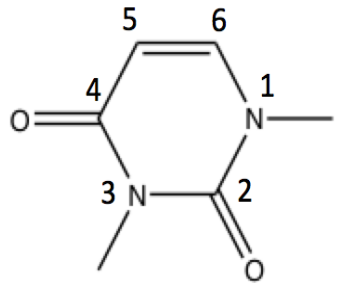
6.5.31,3-dimethyl-2-thioxo-2,3-dihydropyrimidin-4(1H)-one and 1,3-dimethylpyrimidine-2,4(1H,3H)-dione characterization

The characterization of the two minor products is going to be done at the same time due to the fact their characterizations were very similar.



^{1}H NMR (300MHz, $CDCl_3$): δ = 7.16 (1 H, d, J=8Hz, C6H), δ = 6.36 (1 H, d, J=8Hz C5H), δ = 3.39 (3 H, s, NCH_3), δ = 3.13 (3 H, s, NCH_3).
^{13}C NMR (100MHz, $CDCl_3$): δ = 165.03(C2), δ = 162.30(C4), δ = 131.95(C6), δ = 118.01 (C5), δ = 37.20 (NCH_3). δ = 29.99 (NCH_3)
GC-MS (EI): m/z (%)= 156(52), 83 (98), 71(100), 69(79), 58(52).

Figure 55 1,3-dimethyl-2-thioxo-2,3-dihydropyrimidin-4(1H)-one structure. ^{1}H and ^{13}C NMR spectra signals. GC-MS signals.



^{1}H NMR (300MHz, CDCl3): δ = 7.06 (1 H, d, J=8Hz, C6H), δ = 5.86 (1 H, d, J=8Hz C5H), δ = 3.34 (3 H, s, NCH3), δ = 3.29 (3 H, s, NCH3).
^{13}C NMR (100MHz, $CDCl_3$): δ = 172.39(C4), δ = 163.38(C2), δ = 142.66(C6), δ = 101.41(C5), δ = 37.05(N3C), δ = 27.77 (N1C).
GC-MS (EI): m/z (%)= 140(100), 55 (64).

Figure 56 1,3-dimethyl-1,2,3,4-tetrahydro-2,4-dione structure. ^{1}H NMR and ^{13}C spectra signals. GC-MS data.

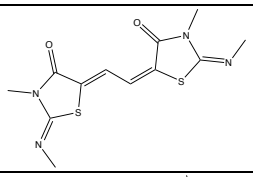
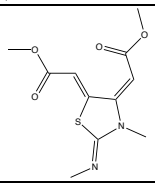
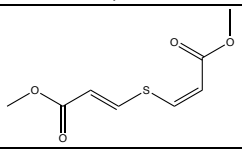
Paying attention to the results about the NMR spectra for these two products, two doublets are observed in both cases. The coupling constant of these doublets suggests as in the first case, the presence of disubstituted alkenes with Z stereochemistry. In addition, this fact is confirmed by ^{13}C spectrum. This is confirmed as in the first case because the two typical peaks for α,β–insaturated carbonyl to α and β positions.
In addition, about ^{1}H spectrum could be highlighted for these two minor products the presence of two singlets that fits with methyl protons.
It could be observed that the chemical shifts for the two products are similar but they could be differentiated because their mass spectra.
For 3-dimethyl-1,2,3,4-tetrahydro-2,4-dione the molecular weight obtained was 140 and the molecular weight for 1,3-dimethyl-2-thioxo-2,3-dihydropyrimidin-4(1H)-one was 156.

6.6 Dimethyl 2,4-hexadiyndioate synthesis[9] and its reaction with N,N'-dimethylthiourea

The results and its discussion about dimethyl 2,4-hexadiyndioate synthesis and its reaction with N,N'-dimethylthiourea are going to be described in this section.
The first thing that should be highlighted is that the yield about the dialkyne (dimethyl 2,4-hexadiyndioate) it couldn't be known. Literature procedure was followed[9] but the last step was not done because it consists in evaporated the solvent where the dialkyne was dissolved. The problem with this step is that dialkyne is very unstable to light and it discomposes very easily when it is isolated. For this reason dialkyne was obtained dissolved in ether and this organic phase was used to the reaction with N,N'-dimethylthiourea. Following this procedure it was not possible to check the success of

the dialkyne synthesis. For this reason, it was decided to do the previous reaction between methyl propiolate and N,N'-dimethylthiourea. Doing this reaction it was check if the reaction between the thiourea and this kind of alkynes was possible. Once it was known that it was possible, the reaction between dialkyne and thiourea could be possible. Arrived to this point, the reaction between the organic phase that should contain the dialkyne and the thiourea it was feasible. In the case of no obtained the expected products of this reaction, this fact would suggest that the dialkyne was not correctly synthesized.
The results for the reaction between the dialkyne and the thiourea are described in table 16.

Table 16 Results about reaction between dimethyl 2,4-hexadiyndioate and N,N'-dimethylthiourea.

Product name	***Structure***	***Theoretical product amount (g)***	***Product obtained (g)***	***Yield (%)***
Bis-(3-methyl-2methylimino-4-oxo-1,3-thiazolidinylidene)ethane		1.8474	0.0327	2
2 methyl-(4-(2-(methoxycarbonyl-methylidene))-3-methyl-2-methylimino-thiazolidin)-acetate		3.2175	0.0208	1
dimethyl E,Z-3,3'-thiobis(acrylate)		2.4071	0.013	1

Paying attention to results in table 16 it could be observed that the yields were very low. Anyway, these lows yields have got an explanation and positive conclusions could be obtained about them.
On the one hand, speaking about the reasons of the lows yields it could be said that the dialkyne was not correctly synthesized. There were two points in the procedure followed to synthesize the dialkyne that could be wrong.
The first one is that air was bubbled in the reaction mixture instead of bubbling oxygen as it was indicated in the literature, so the amount of oxygen that it was bubbled with the air could be not enough.
The other possible mistake could be the system that it was employed to do the reaction. It was observed that the level of the solvent (acetone) of the reaction mixture was decreasing during the reaction. It indicated that the acetone was being evaporated and the methyl propiolate could be evaporated too.
Furthermore, there were two more facts that indicated that the dialkyne synthesis was not very successful:

- The last product is result about reaction between methyl propiolate and the thiourea.
- A big amount of the thiourea was found in the reaction mixture after the reaction.

On the other hand, it could be said that this reaction was very useful despite the low yields. The expected product was obtained (bicycle) and other product result of the reaction between dialkyne and thiourea was isolated. For this reason, it could be highlighted that the chemistry between dialkyne and thioruea is possible but the way of obtain the dialkyne needs to be improved. Once this is achieved, it would be possible to prove the reactivity between the dialkyne and hydrazine to obtain new products which could be converted to compounds for testing new antibiotics.

The results about this reaction and the previous experience about the reactivity between DMHD with thioureas and hydrazine suggest: promising products could be obtained in the reaction between hydrazine and the dialkyne. For example one promising product could be the once show in figure 57.

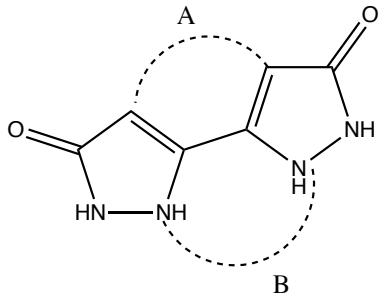


Figure 57 Possible product in the reaction between dialkyne and hydrazine.

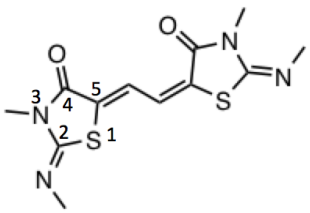
The discontinued lines in figure 57 show possible points of cyclization. Paying attention to side A it could be try a Diels-Alder reaction with a dienophile.

In reference to side B, it could be tried the reaction between the two nitrogens and a molecule with two electrophilic centres achieving a cyclization. These possible cyclized products could be tested as antibiotics.

The characterization of the three products obtained in the reaction between dialkyne and thiourea are going to be exposed in the following sections.

6.6.1 Bis-(3-methyl-2methylimino-4-oxo-1,3-thiazolidinylidene)ethane characterization

The characterization of this byclic product was done by ^{1}H and ^{13}C NMR spectra.



^{1}H NMR (300MHz, $CDCl_3$): δ = 7.20 (1 H, s, C5=CH), δ = 3.27 (3 H, s, NCH_3), δ = 3.23 (3 H, s, NCH_3).

^{13}C NMR (100MHz, $CDCl_3$): δ = 165.32(C4), δ = 148.56(C2), δ = 131.79 (C5=C), δ = 122.39 (C5=C), δ = 39.53 ($N3CH_3$), δ = 29.55 ($C2=NCH_3$).

Figure 58 Bis-(3-methyl-2methylimino-4-oxo-1,3-thiazolidinylidene)ethane structure. ^{1}H and ^{13}C NMR signals.

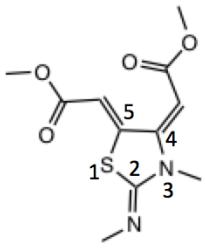
The first thing that could be highlighted about ^{1}H and ^{13}C of this bycyle structure is that total number signals of proton and carbon are the half of the total number of protons and carbons in the molecules. This effect is explained by the fact that there are isochrones protons and carbons. This isochrony is caused due to the symmetry of the molecule. This bicycled molecule could be split in two sides related by a symmetry plan.

In one hand, ^{1}H spectrum is very clear. The expected peaks about the proton in the alkene and the methyl protons are showed.

On the other hand, speaking about ^{13}C spectrum it could be said that the typical peaks for the α,β–insaturated carbonyl are showed. Furthermore, the assignment of the imine carbon (C2) was confirmed with the literature[13]. Finally, the different chemical shift of the two methyl carbons could be explained because the carbon attached to C3 could be affected by carbonyl anisotropic effect. For this reason, it is thought that the chemical shift of this carbon is higher than the one for C2.

6.6.2 Methyl-(4-(2-(methoxycarbonyl-methylidene))-3-methyl-2-methylimino-thiazolidin)-acetate characterization

This product was characterized by ^{1}H and ^{13}C NMR spectra and with a two-dimensional NMR technique, HMQC.



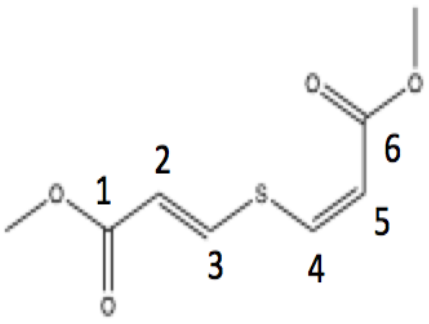
^{1}H NMR (300MHz, $CDCl_3$): δ = 9.04 (1 H, s, C5=CH), δ = 5.27 (1 H, s, C4=CH), δ = 3.90 (3 H, s, OCH_3), δ = 3.71 (3 H, s, OCH_3), δ = 3.35 (3 H, s, NCH_3), δ = 3.17 (3 H, s, NCH_3).
^{13}C NMR (100MHz, $CDCl_3$): δ=167.38(C5=CH-C=O), δ = 163.24(C4=CH-C=O), δ = 148.66 (C2), δ = 126.39 (C5), δ = 122.19 (C5=CH), δ = 97.32 (C4=CH), δ = 53.12 (OCH_3), δ = 51.27 (OCH_3), δ = 36.89 (NCH_3), δ = 34.63 (NCH_3).

Figure 59 methyl-(4-(2-(methoxycarbonyl-methylidene))-3-methyl-2-methylimino-thiazolidin)-acetate structure. ^{1}H and ^{13}C NMR signals.

Focusing in ^{1}H NMR spectrum should be highlighted the different chemical shift between the two-alkene protons. This difference was very important to the characterization because it suggests that the stereochemistry of the two alkenes is different. Due to the different stereochemistry, the high chemical shift (9.04ppm) of one of the protons is explained because it is affected by carbonyl anisotropic effect.
Furthermore, the expected signals for esters' methyls and methyls bonded to nitrogen appeared.
Paying attention to ^{13}C NMR spectrum, the expected signals for different types of methyls and the two carbonils could be identified. In addition, other clear assignment was C2 due to this type of carbon chemical shift was checked in the literature[13].
Furthermore, it has to be highlighted that only three of the four alkene carbons could be assigned. Taking in account that the rest of signals fit very well with the proposed structure, it is thought that the signal about the missing carbon it is not enough intense to be detected.
Finally, it should be mentioned that the assignment of the two alkene carbons bonded to protons has been assigned because of the HMQC were their connections with protons are showed.

6.6.3 Dimethyl E,Z-3,3'-thiobis(acrylate) characterization

The product resulting between the reaction of thiourea and methyl propiolate was characterized by ^{1}H and ^{13}C NMR spectra.



^{1}H NMR (300MHz, $CDCl_3$): δ = 7.65 (1 H, d, J=16Hz, C3H), δ = 7.14 (1 H, d, J = 8Hz, C4H), δ = 6.03 (1 H, d, J=16Hz, C2H), δ = 5.98 (1 H, d, J=8Hz, C5H), δ = 3.71 (3 H, s, OCH_3), δ = 3.69 (3 H, s, OCH_3).
^{13}C NMR (100MHz, $CDCl_3$): δ=166.47 (C=O), δ = 165.26(C=O), δ = 146.04(C3), δ = 143.83 (C4), δ = 117.99 (C2), δ = 115.70 (C5), δ = 51.83 (OCH_3), δ = 51.74 (OCH_3).

Figure 60 Dimethyl E,Z-3,3'-thiobis(acrylate) structure. ^{1}H and ^{13}C NMR signals.

Focusing about the signals about carbonyls it should be said that they present the typical ester chemical shift. The exactly assignment of each one it could not be done

because the two carbonyls have got very similar chemical shift. The same explanation could be done to methyl signals.

The assignment of the alkene protons and carbons was possible because different reasons:

- The signals were the typical for α,β–insaturated carbonyl to α and β positions.
- The couplings constants of the protons show the stereochemistry of the two alkenes. The ones with the highest couplings constants fit with E-alkene and the ones with lowest couplings constants correspond to Z-alkene.
- Furthermore, the chemical shift and the coupling constant to this kind of E-alkene was checked in the literature[14].

6 Conclusion

The reactivity of two electron deficient alkynes against binucleophilic species was tested and promising results were obtained. Theses two alkynes are (E)-dimethyl hex-2-en-4-ynedioate (DMHD) and dimethyl 2,4-hexadiyndioate (dialkyne).

On the one hand, the reactivity of DMHD against three different species with two nucleophilic centres was tested. These nucleophilic species were: hydrazine, phenylhydrazine and methylhydrazine. The results for each one were very different.
In reference to reaction with phenylhydrazine the results were not good due to the fact it was not possible isolate interesting products. The conditions of reaction between this specie and DMHD should be improved.
Speaking about the results of the reaction with methylhydrazine, it could be said that four different products were isolated. The problem was that only one of them could be interesting to convert to a compound that could work as antibiotic and it was the minor product. The other three products could not be converted to compounds for testing as antibiotics because of their structure it was not the one desired.
Despite the results for these two species were not the ones expected, the results for the reaction between hydrazine and DMHD were positive. Two different products were isolated and their structures suggest that they could be converted to compounds with antibacterial activity. The major product is a solid lactam called methyl E-3-(1'H-2',3'-dihydro-3'-oxopyrazol-5'-yl)prop-2-enoate that is very easy to be isolated. The other product was minoritary. This is an oily diester called methyl-2-(1'H-4',5'-dihydro-3'-methoxycarbonyl-pyrazol-5'-yl) acetate. It was more difficult to isolate and with a lower yield but its structure could be interesting too.

On the other hand, the reactivity of dialkyne against N,N'-dimethylthiourea was tested. It has to be highlighted that two interesting products were obtained in this reaction, but only little amount of them were generated. This low yield is caused by the difficulties to synthesize the dialkyne. For this reason this synthesis must be improve. The two interesting products synthetized are a solid with bicyclic structure called bis-(3-methyl-2methylimino-4-oxo-1,3-thiazolidinylidene)ethane and a colourless oil called 2-methyl-(4-(2-(methoxycarbonyl-methylidene))-3-methyl-2-methylimino-thiazolidin)-acetate.
Anyway, the little amount obtained of the two products suggests that the chemistry of this dialkyne with binucleophil species is possible.
The next step would be improve the dialkyne synthesis and test its reactivity against hydrazine. Taking account the results obtained with DMHD, it is thought that with the dialkyne could be possible isolate interesting products to convert to compounds with antibacterial properties.

In conclusion, the first steps in the study about the reactivity of these electron deficient alkynes against binucleophilic species has been done and the results suggest that promising compounds could be obtained with further research.

7 References

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