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ON THE MECHANISM OF INDIRUBIN FORMATION BY REACTION OF ISATIN WITH 2-AMINO-ACETOPHENONE

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ABSTRACT

A failed intent recently published in order to explain indirubin formation from isatin and 2-amino-acetophenone has been commented and discarded. In this paper two routes to indirubin using the same reactants are provided after the initial Knoevenagel reaction. In the first proposal the proximity of the aromatic amino group to the methine in the reaction product suggests C-N formation by amination of carbanion via hydride elimination, as references inform. This proximity was confirmed in a 3D chemical structure. This oxido-cyclization yields indirubin.

The other route starts with the lactam resonance structure. Reaction of the enolate with water gives an intermediate to lactim; this is conjugated and permits reaction with the aromatic amino group and can be cyclisation.

Carbanion formation at the methine and reaction with aerial oxygen yields a peroxide which reacts with water. The electrodotic enol in the five-member ring forms a carbon-carbon double bond and expulsion of a hydroperoxide, yielding indirubin.

Keywords: Aerial Oxidation, Amination of Carbanion; 3D Chemical Structures; Hydride Elimination; Knoevenagel Reaction; Lactim

1 INTRODUCTION

In the pioneering studies of Baeyer for the synthesis of indigo, he obtained always, in more or less proportions, two products: indigo (indigo blue) and indirubin (indigo red) which was termed firstly purpurin. Indirubin was always obtained in minor quantity, (secondary product), [1]. Since then, various synthetic routes to indirubin have been described because this compound and some derivatives have many biological applications, for instance treatment of leukemia, [2].

Recently, indirubin has been prepared from isatin and 2-amino-acetophenone [3], but the authors failed to explain how indirubin is formed since an erroneous reaction mechanism was advanced, without arriving to the target molecule.

In this communication we provide two routes to indirubin using the same reactants, and the electron flow is given in each step.

This paper is a follow up of our studies on reaction mechanism, [4-8].

2 ANTECEDENTS

Chemistry of the reactants. Isatin was obtained for the first time by Laurent in France, [9, 10] by oxidation of indigo with nitric acid. The reaction mechanism has been given, and the laboratory method has been translated to English from the French, [1].

Baeyer synthesised isatin by nitrosation of oxindole (2-indolinone) via its enol (2-hydroxyindole. Nitroso oxindole was reduced to amino group and oxidised to carbonyl by cupric chloride, yielding isatin, [11, 12].

For the obtention of 2-amino-acetophenone see [13].

3 DISCUSSION

In an article about the preparation of indirubin from isatin and 2-amino-acetophenone [3], there is a failed intent to explain indirubin formation by this route. The authors propose that the product obtained after a Knoevenagel condensation cyclises by reaction of the amino group with the α,β-unsaturated amido group present in the isatin moiety. This is an error because the amide resonance [14, 15] hasn't been taken into account, and the proposed electron flow doesn't occur because there is no carbonyl available. For instance, the amide resonance explains the passage from the lactam form to the lactim (imidol), see [16, 17]. By the way, the correct polarization in the reaction intermediate is in the α,β-unsaturated ketone, as indicated in Figure 1.

So, the next step is obsolete and besides, it contains a positive quaternary nitrogen atom, a protonation that is inadmissible in alkaline medium. Also, a proposed electron flow is fictitious since it cannot end at a sp3 carbon atom. Finally, the authors do not arrive to the target molecule, indirubin, because they propose false intermediates and an oxidation step is missing.

Now let´s see the correct sequence for indirubin formation from isatin and 2-amino-acetophenone. In Figure 2 are the reactants and the reactive species. Reaction occurs at the keto group in C-3, not at the carbonyl at C-2 of the lactam due to the previously mentioned amide resonance. The first product in this Knoevenagel reaction [18, 19] is the alcohol. We obtained the 3D image of this intermediate and is very interesting to observe that the benzene ring is out of plane respect to the isatin moiety, Figure 4.

The next step is dehydration of the alcohol, forming a double bond at C-3 in the isatin fragment. The 3D image of a rotamer shows again that the benzene ring is out of plane, and also the proximity of the amino group to the methine group, Figure 4.. Formation of a carbanion at this carbon permit reaction with the amino group via hydride elimination. This C-N bond formation is an oxidative reaction [20, 21], and in our case also a cyclization. This way indirubin has been formed.

Hydride ions are very bad leaving groups but in this case the reaction can be considered as a hydride transfer which is much better; that is, transfer to an unreacted keto group (in isatin or in the acetophenone) forming an alkoxide. The latter reacts with aerial oxygen and a five-member cyclic mechanism restores the keto group with elimination of a hydroperoxide, instead of the hydride ion.

Another route to indirubin is via the zwitter ion in amide resonance. Reaction of the enolate with water gives a transient intermediate to lactim, whose conjugation permits cyclisation as indicated in Figure 3. Then, carbanion formation at the methine group and reaction with oxygen yields a peroxide which is neutralized by water. The 2D structure indicates steric hindrance; however, in the 3D model there is no problem because the heterocycles are almost perpendicular (Figure 4).

Molecular oxygen is a high energy molecule and this kind of molecules has relatively weak bonds. This makes oxygen an electron acceptor, [22, 23].

Finally, the electrodotic property [24, 25] of the enol in the five-member ring forms a carbon-carbon double bond and elimination of a hydroperoxide ion (oxidation step) and indirubin results.

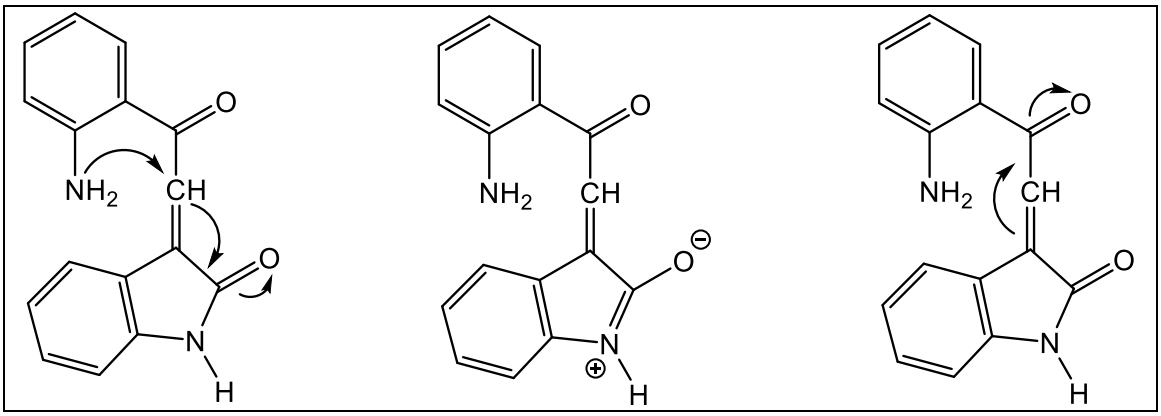


Figure 1: Erroneous electron flow, and two correct polarizations

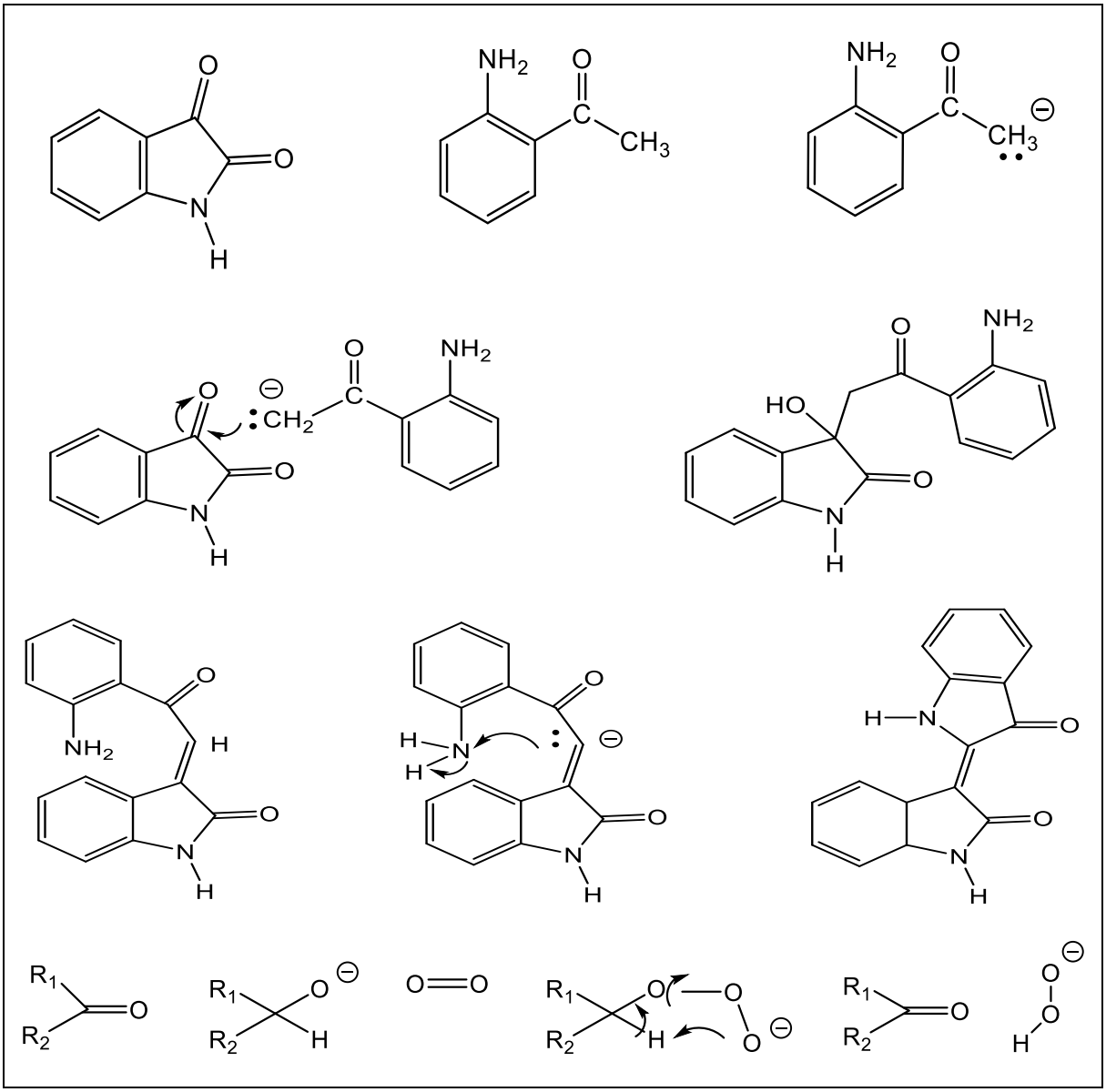


Figure 2: First route to indirubin from isatin and 2-amino-acetophenone

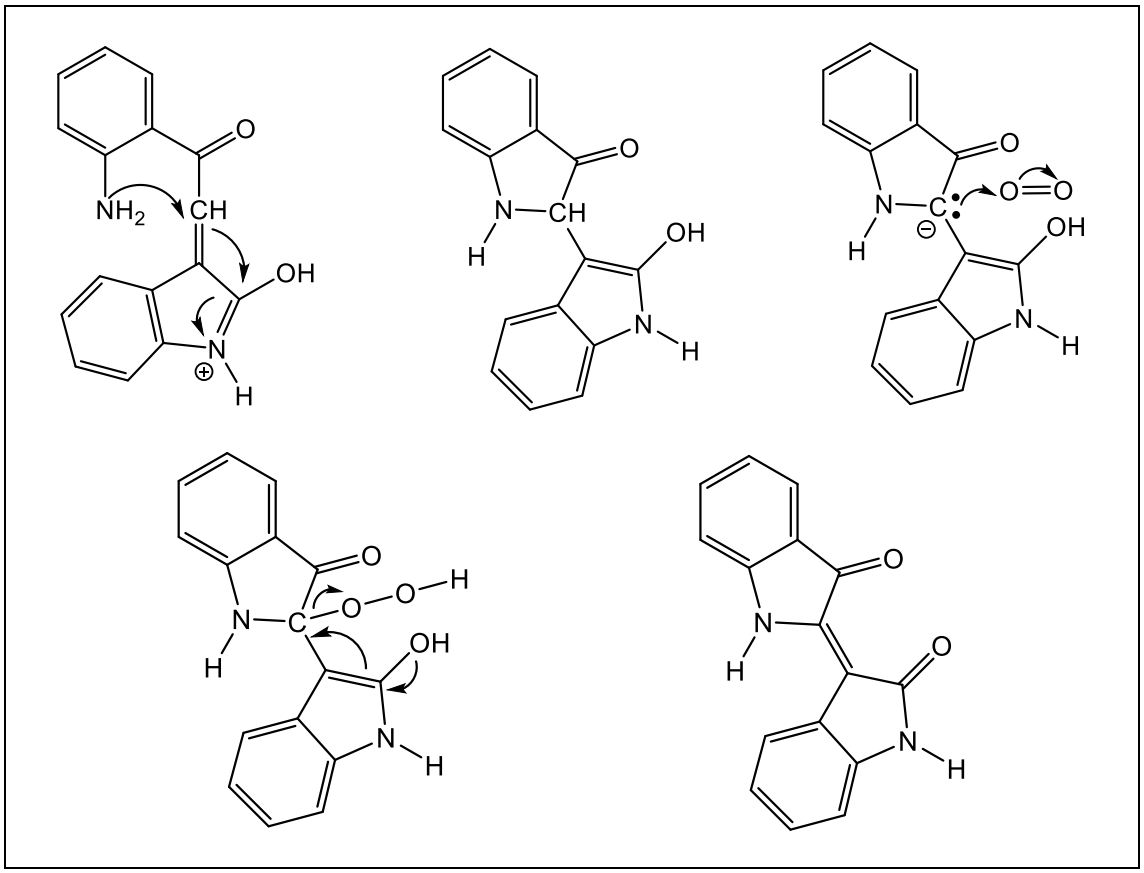


Figure 3: Second route to indirubin formation

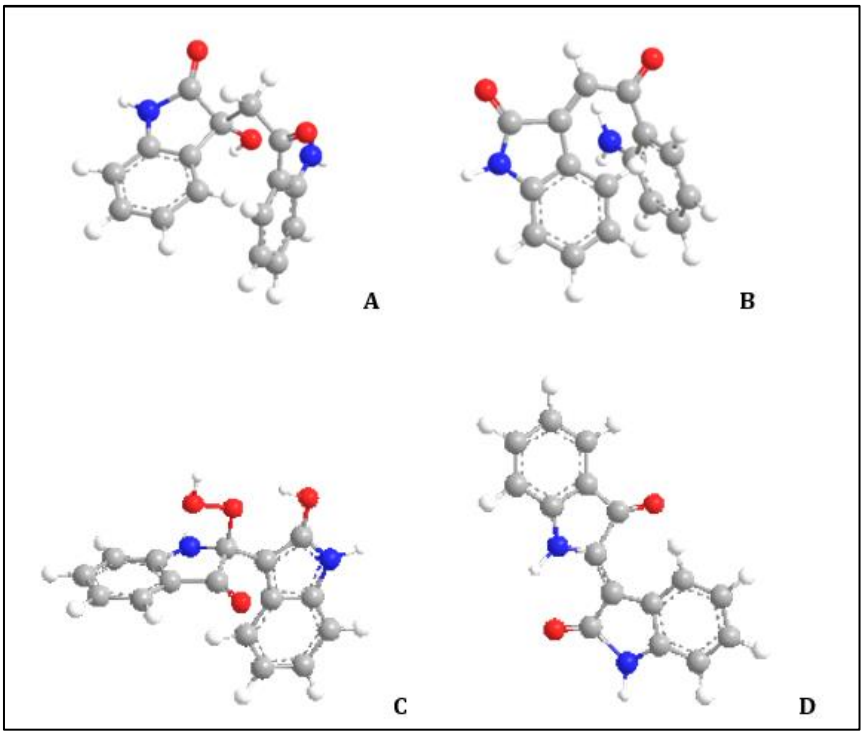


Figure 4: A: intermediate and B: final product of Knoevenagel reaction; C: hydroperoxide intermediate in second route D: indirubin

4 CONCLUSION

A failed intent to explain the formation of indirubin from isatin and 2-amino-acetophenone has been commented and discarded. Two routes to indirubin using these reactants have been provided. The use of 3D chemical structures has been very useful from different points of view, synthetic, geometric and steric, as mentioned in the text.

STATEMENTS ON COMPLIANCE WITH ETHICAL STANDARDS

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Disclosure of conflict of interest

There is no conflict of interest to declare.

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