

Laser flash photolysis study of the photochemistry of ninhydrin and its 5-methoxy derivative in benzene solution

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Abstract

Laser flash photolysis of ninhydrin (**1**) and its 5-methoxy derivative **4**, in benzene solution, yields readily detectable transients, which were assigned to their corresponding triplet excited states. By employing this time-resolved technique, rate constants for hydrogen abstraction, electron transfer and energy transfer processes were measured. The quenching rate constants obtained are independent of the ninhydrin structure, being only dependent on the quenching process. These values range from $1.3 \times 10^6 \text{ M}^{-1} \text{ s}^{-1}$ (cyclohexa-1,4-diene) to $5.9 \times 10^9 \text{ M}^{-1} \text{ s}^{-1}$ (DABCO) for ninhydrin (**1**) and from $1.8 \times 10^6 \text{ M}^{-1} \text{ s}^{-1}$ (cyclohexa-1,4-diene) to $7.3 \times 10^9 \text{ M}^{-1} \text{ s}^{-1}$ (DABCO) for 5-methoxyninhydrin (**4**). The quenching rate constants obtained, characteristic of an $n\pi^*$ carbonyl triplet, indicate that the photochemistry of **1** and **4** is controlled by the 1,3-dicarbonyl chromophore, despite the presence of the 5-methoxy group in **4**.

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Keywords: Ninhydrin; Triplet state; Laser flash photolysis; Hydrogen abstraction; Electron transfer; Energy transfer

1. Introduction

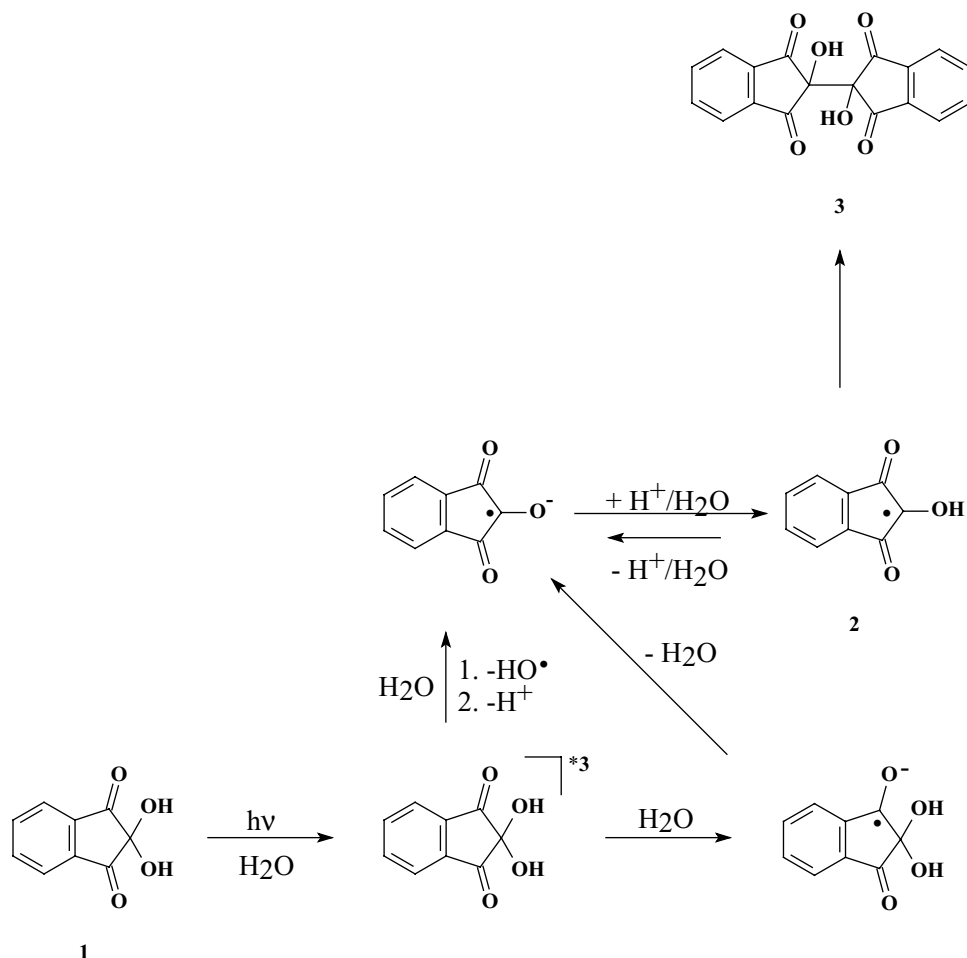
Ninhydrin (2,2-dihydroxy-1,3-indanedione) (**1**) has been employed as a very important analytical tool in several fields, such as chemistry, biochemistry, agricultural and forensic sciences, with several reviews dealing with the synthesis and applications of ninhydrin and analogs [1–4].

Steady state photolysis of ninhydrin (**1**) in water yields the pinacol hydrindantin (**3**) as the major product, besides to minor products derived from the decarbonilation of **1** [5,6]. The dynamics of this reaction in benzene and in aqueous solution was recently investigated by our group employing laser flash photolysis and electron paramagnetic resonance (EPR) [7]. In benzene, the triplet excited state was readily formed, which in the presence of suitable sources of hydrogen underwent hydrogen abstraction with the formation of the corresponding ketyl radical **2**. However, in water no triplet excited state could be observed. A long-lived transient assigned to the radical anion derived from **1** was detected, which upon protonation at low pHs (<1) led to the formation of the ketyl radical **2**. The formation of hydroxyl radicals is ambiguous and the yield of this reactive species, if formed, is very low (Scheme 1).

The triplet excited state of aromatic ketones abstracts hydrogen atoms from a variety of hydrogen donors including hydrocarbons and alcohols [8–10]. For amines, the reaction occurs through a fast electron-transfer, followed by a proton transfer [11]. This reactivity is associated to the electrophilic character of the carbonyl oxygen of the triplet state having $n\pi^*$ configuration. For those ketones showing lowest triplet state of $\pi\pi^*$ nature, the reactivity towards hydrogen donors is substantially lowered. For alkyl aryl ketones the two triplet states, $n\pi^*$ and $\pi\pi^*$, are very close in energy. Several factors, such as solvent effects or the introduction of electron-donor substituents (methyl or methoxyl groups) in the *ortho* and/or *para* position of the aromatic ring, are responsible for an inversion of these excited states. Extensive studies have been made of hydrogen abstraction and spectroscopic properties of several *para*-methoxy-substituted phenyl alkyl ketones [12–17]. However, virtually nothing is known concerning the photochemistry of cyclic 1,3-dicarbonyl compounds substituted by a methoxy group in the aromatic ring.

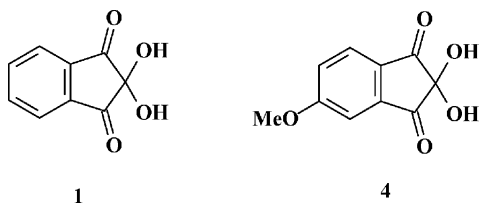
In this work we show by laser flash photolysis studies that the photochemistry of ninhydrin (**1**) and its 5-methoxy derivative, i.e. 5-methoxyninhydrin (2,2-dihydroxy-5-methoxy-1,3-indanedione) (**4**), in organic solvents such as benzene or acetonitrile, is independent of the structure of the ninhydrin and is distinct than that observed in aqueous solution. We also show that, in the former sol-

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Scheme 1.

vents, the photochemistry of **1** and **4** involves the formation of their corresponding triplet excited state, which are readily quenched in the presence of several hydrogen donors, electron donors, and olefins.



2. Experimental

2.1. Materials

The solvents employed were Aldrich Spectrograde and were used as received. Ninhydrin and 5-methoxyninhydrin were recrystallized from hot water and activated charcoal, which was removed by filtration. A yellow solution remained from which pale yellow crystals were collected af-

ter cooling, in both cases. 1,3-Cyclohexadiene, β -carotene, 1,4-cyclohexadiene, phenol, 1,1-diphenylethylene, *trans*-stilbene, *cis*-stilbene, 1,4-diazabicyclo[2,2,2]octane (DABCO), and triethylamine were purchased from Aldrich and purified when necessary.

2.2. General techniques

UV-vis spectra were recorded in a Varian Cary 3E. Laser flash photolysis (LFP) experiments were carried out on an Edinburgh Analytical Instruments LP900. Samples were contained in a 10 mm \times 10 mm cell made from Suprasil quartz tubing and were deaerated by bubbling with oxygen-free nitrogen for 20 min. The samples were irradiated with the third harmonic ($\lambda = 355$ nm, ~ 5 ns, 40 mJ/pulse) of a Nd/YAG Surelite laser. Sample concentration was chosen in order to give an absorption at the wavelength of excitation of 0.3. Stock solutions of quenchers in the same solvent employed in sample preparation were prepared so that it was only necessary to add microliter volumes to the sample cell in order to obtain appropriate concentrations of the quencher.

3. Results

Excitation ($\lambda = 355$ nm) of ninhydrin in acetonitrile led to the observation of a transient spectrum with absorption maxima at 540 and 580 nm and an end absorption below 360 nm (Fig. 1). This spectrum is similar to that previously obtained when benzene or diethyl ether was used as the solvent [7].

The decay of this transient, either in benzene or in acetonitrile, followed first order kinetics from which a lifetime of $11 \pm 2 \mu\text{s}$ was obtained. This lifetime is independent of the monitoring wavelength, which indicates that only one transient was formed in the photolysis of **1** in both solvents. The triplet nature of this transient was determined by quenching experiments employing 1,3-cyclohexadiene ($E_T = 52.4$ kcal/mol), which is an efficient quencher of triplet excited states, [18] with the quenching reaction leading to the deactivation of the excited state back to its ground state. The quenching plot followed Eq. (1) and was linear, with the quenching rate constant observed ($k_q = (1.5 \pm 0.1) \times 10^9 \text{ M}^{-1} \text{ s}^{-1}$) being slightly lower than the diffusion-control limit.

$$k_{\text{obs}} = k_0 + k_q [\text{quencher}] \quad (1)$$

Further evidence that the transient observed for **1** in benzene or in acetonitrile corresponds to a triplet excited state came from the quenching by β -carotene ($E_T = 21$ kcal/mol) [18]. In this type of experiment, the kinetics of the decay for the ninhydrin triplet (at 570 nm) as well as of the growth for the β -carotene triplet (at 530 nm) are followed. A diffusion-controlled quenching rate constant was obtained for the decay of ninhydrin monitored at 570 nm. In the present case, the overlap of the β -carotene triplet–triplet absorption spectrum with that of ninhydrin prevented measurement of the quenching rate constant by monitoring the growth of the signal at 530 nm. However, at a large concentration of the quencher, the triplet–triplet absorption spectrum for β -carotene, obtained by energy transfer from ninhydrin in benzene solution could be observed (Fig. 2). When hydrogen atom donors are used as

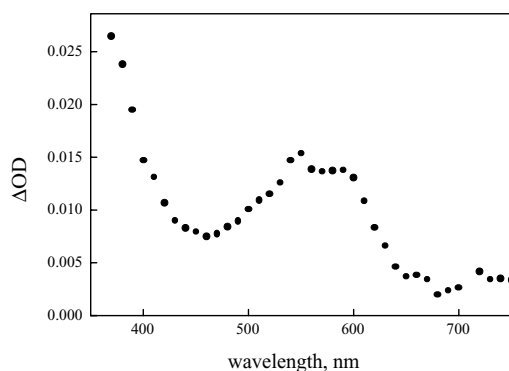


Fig. 1. Transient absorption spectrum recorded after 355 nm laser excitation of ninhydrin (**1**) in acetonitrile.

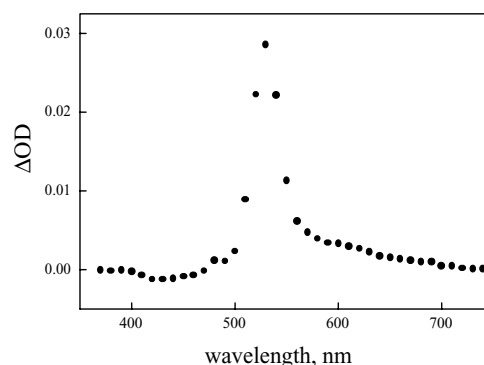
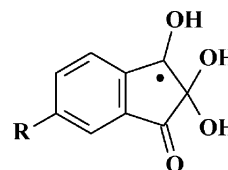


Fig. 2. Triplet–triplet absorption spectrum for β -carotene obtained from energy transfer from ninhydrin triplet in benzene.

quenchers for triplet ketones with $n\pi^*$ character, hydrogen abstraction occurs leading to the formation of the ketone ketyl radical [19,20]. Thus, a quenching rate constant of $(1.3 \pm 0.1) \times 10^6 \text{ M}^{-1} \text{ s}^{-1}$ for the triplet excited state of **1** by 1,4-cyclohexadiene was obtained, fully in accord with a previous value of $(1.8 \pm 0.1) \times 10^6 \text{ M}^{-1} \text{ s}^{-1}$ reported in the literature [7]. It is worth noting, for this quencher is possible to observe the ketyl radical derived from **1**, i.e. **5a** ($R = \text{H}$), which shows an end absorption below 360 nm and is structurally distinct of **2**, already described in the literature [7] (see Scheme 1).



5 a: $R = \text{H}$
b: $R = \text{OMe}$

For phenol, the rate constant is still in accord with the formation of the $n\pi^*$ triplet state of **1** in benzene or acetonitrile (Table 1). In this case a new transient was observed, with

Table 1
Second order rate constants for the quenching of the triplets of ninhydrin (**1**) and 5-methoxyninhydrin (**4**) in benzene solution

Quencher	k_q ($\text{M}^{-1} \text{ s}^{-1}$) ^a	
	1	4
β -Carotene	5.1×10^9	5.3×10^9
1,3-Cyclohexadiene	1.5×10^9	1.8×10^9
1,4-Cyclohexadiene	1.3×10^6	1.8×10^6
Phenol	1.4×10^8	1.9×10^8
1,1-Diphenylethylene	3.2×10^6	2.7×10^6
<i>trans</i> -Stilbene	1.2×10^7	1.5×10^7
<i>cis</i> -Stilbene	2.3×10^8	1.7×10^8
DABCO	5.9×10^9	7.3×10^9
Triethylamine	3.8×10^9	3.9×10^9

^a Estimated to be accurate to $\pm 10\%$.

maximum at 400 nm, which was assigned to the phenoxyl radical.

Table 1 also shows second order quenching rate constants for several other quenchers, such as olefins (1,1-diphenylethylene, *cis*- and *trans*-stilbene) and electron donors (triethylamine and DABCO). In all cases, linear plots following Eq. (1) were obtained and representative quenching plots for **1** are shown in Fig. 3A. For the olefins, the quenching rate constants observed may result from the involvement of contributions from physical and chemical quenching. On the other hand, the electron donors quench the triplet excited state of **1** in a diffusion-controlled mode. No evidence for the formation of a new transient was obtained with any of these quenchers.

Laser excitation (355 nm) of 5-methoxyninhydrin (**4**) in benzene solution leads to the formation of a transient which show absorption maxima at 450, 560 and 600 nm (Fig. 4), with the maxima in the long wavelength region of the spectrum being slightly shift to the red when compared to those of the parent compound, i.e. **1**. This transient decays by first order kinetics with a lifetime of $3.0 \pm 0.3 \mu\text{s}$. Its triplet character was confirmed by quenching by β -carotene at a diffusion-controlled rate in this solvent. Like ninhydrin, this

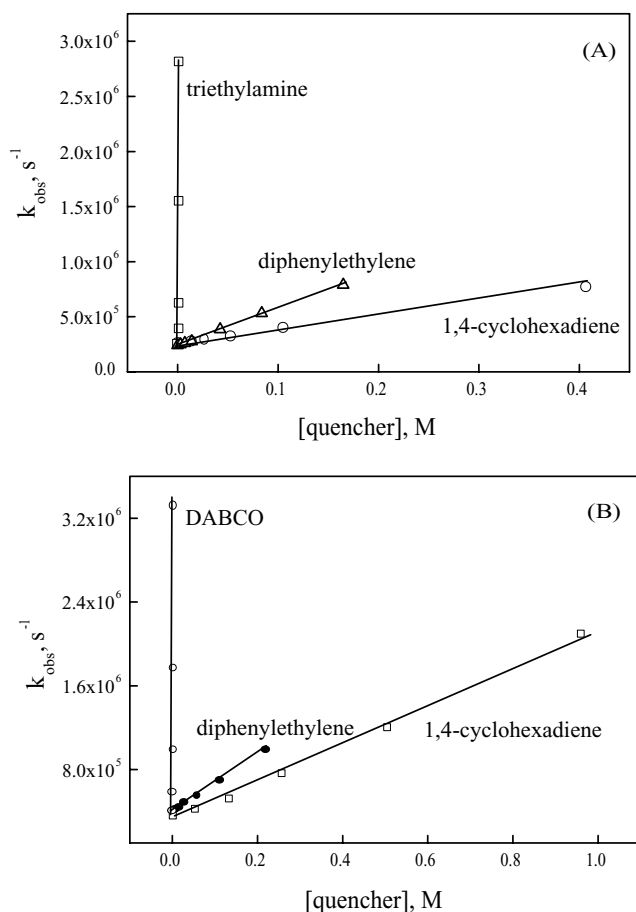


Fig. 3. Representative quenching plots of ninhydrins triplet state by several quenchers in benzene ($\lambda_{\text{exc}} = 355 \text{ nm}$): (A) ninhydrin (**1**) ($\lambda_{\text{mon}} = 570 \text{ nm}$); (B) 5-methoxyninhydrin (**4**) ($\lambda_{\text{mon}} = 560 \text{ nm}$).

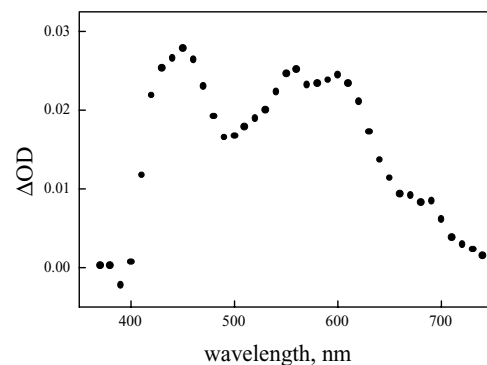


Fig. 4. Transient absorption spectrum recorded after 355 nm laser excitation of 5-methoxyninhydrin (**4**) in benzene.

quenching process results in the concomitant formation of the β -carotene triplet at 530 nm.

Using the same quenchers as those reported for ninhydrin (see above), similar quenching rate constants were obtained for 5-methoxyninhydrin (**4**) (Table 1). New transients were also observed for **4** when 1,4-cyclohexadiene and phenol were used as quenchers. These transients were assigned to the corresponding ketyl radical derived from **4** (for 1,4-cyclohexadiene), i.e. **5b** ($\text{R} = \text{OMe}$) (Fig. 5), and to the phenoxyl radical (in the phenol case). Representative quenching plots for 5-methoxyninhydrin (**4**) and employing equation 1 are shown in Fig. 3B. As for the ninhydrin case, no evidence for the formation of a new transient was obtained when olefins or electron-donors were employed as quenchers.

4. Discussion

The spectroscopic data obtained in this work for the transient derived from **1** are in excellent agreement with those reported previously in organic solvents [7]. In deaerated benzene or acetonitrile solutions this transient decays by a

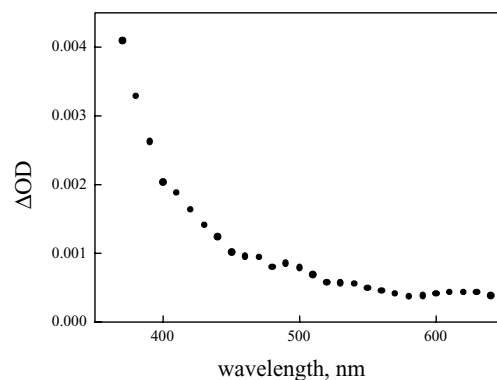


Fig. 5. Transient absorption spectrum recorded after 355 nm laser excitation of 5-methoxyninhydrin (**4**) in the presence of 0.35 M 1,4-cyclohexadiene in benzene.

first-order kinetics ($\tau = 11 \pm 2 \mu\text{s}$) with its assignment being based on the results of 1,3-cyclohexadiene and β -carotene quenching experiments. The transient spectroscopy of the 1,3-diketone **4** differs from that observed for **1**. Thus, shorter lifetimes and a red-shift of close to 20 nm were observed for its triplet, either in benzene or in acetonitrile, when compared to the triplet of **1**.

Various *para*-methoxy-substituted phenyl alkyl ketones have been extensively studied for spectroscopic properties. These studies show that the lifetime of the lowest triplet state of character $\pi\pi^*$ for the *para*-methoxy ketones is much longer than the corresponding non-substituted ketone, which shows triplet excited state of $n\pi^*$ character. This is a consequence of the presence of the electron-donating substituent that may switch the electronic configuration of the T_1 and T_2 states of these ketones, since they are very close in energy (~ 3 kcal/mol) [9,10,12,21,22]. Thus, the shorter lifetime for **4** when compared to **1** is a first indication that the introduction of a methoxyl group in the benzene moiety of ninhydrin must not alter the $n\pi^*$ character of its triplet excited state.

Quenching studies give support to the spectroscopic results. The reaction with 1,4-cyclohexadiene, an excellent hydrogen-donor, [23] leads to the same quenching rate constant for both. ninhydrin and 5-methoxyninhydrin. The rate constant obtained for these two 1,3-diketones is slower than those reported for $n\pi^*$ monoketones. This difference in reactivity is probably due to differences in the ΔH of the reaction, reflecting the lower triplet energy of **1** and **4**, which was estimated to be < 52 kcal/mol from quenching studies with 1,3-cyclohexadiene.

Phenols can act as electron and/or hydrogen donors, with the electron transfer being more probable in polar solvents, whereas hydrogen transfer occurs in non-polar solvents [24]. The rate constants observed for the quenching of ninhydrin and 5-methoxyninhydrin by phenol are two orders of magnitude faster than those observed for 1,4-cyclohexadiene, from which one expects a pure hydrogen atom transfer mechanism. The formation of the phenoxyl radical in the quenching process of **1** and **4** by phenol, together with the fast quenching rate constant observed, seems to indicate that the primary quenching process involves an electron transfer, followed by a fast proton transfer, that ultimately generates the phenoxyl radical.

The very fast quenching rate constant observed for ninhydrin or 5-methoxyninhydrin when in the presence of the excellent electron-donors DABCO and triethylamine indicate that a full electron transfer mechanism is operating for both quenchers.

Since for a given quencher the quenching rate constants obtained for **1** and **4** are much the same in all examples cited above, the difference in energy between the excited states T_1 ($n\pi^*$) and T_2 ($\pi\pi^*$) for ninhydrin (**1**) must be high enough to be overcome by simply adding a 5-methoxy group on its aromatic ring. Thus, it is reasonable to assume that any stabilization of the $\pi\pi^*$ state of 5-methoxyninhydrin (**4**) does not bring the energy of this level close to its $n\pi^*$ state.

In other words, one can also associate the reactivity of **4** to its lowest energy triplet of $n\pi^*$ character.

To further confirm this statement, a plot of the quenching rate constants obtained from the reaction of the triplet states of ninhydrin (**1**) and 5-methoxyninhydrin (**4**) with the different quenchers employed in this work (Table 1) shows that there is a linear correlation between these two sets of data with a slope of 1.015 ($r = 0.998$). This result leads us to conclude that for a specific quencher (hydrogen donor, electron donor or olefin), the quenching mechanism is the same for **1** or **4**, which is in agreement with a triplet excited state of $n\pi^*$ character for both ninhydrins. Finally, from the quenching rate constants obtained, characteristic of an $n\pi^*$ carbonyl triplet, one can conclude that the photochemistry of ninhydrin and 5-methoxyninhydrin is controlled by the 1,3-dicarbonyl chromophore, in spite of the presence of the 5-methoxy group in the diketone **4**.

5. Conclusion

In conclusion, it was shown by nanosecond laser flash photolysis studies that the quenching rate constants of the triplet state of ninhydrin (**1**) and 5-methoxyninhydrin (**4**) by several quenchers are independent of the ninhydrin structure. The excellent correlation observed between the quenching rate constants for the pair ninhydrin versus 5-methoxyninhydrin clearly show that the presence of the 5-methoxy substituent does not affect the reactivity of the parent compound, i.e. ninhydrin. This can be explained by the substantially high difference in energy between the $n\pi^*$ and $\pi\pi^*$ triplet states for **1**, with the change in structure being not able to perform an inversion of these two states.

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