Molecular Dynamics Insights into Electron-Catalyzed Dissociation Repair of Cyclobutane Pyrimidine Dimer

Liang Gao, Yuxiang Bu

School of Chemistry and Chemical Engineering, Shandong University, Jinan 250100, China

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Excess electrons are not only an important source of radiation damage, but also participate in the repair process of radiation damage such as cyclobutane pyrimidine dimer (CPD). Using ab initio molecular dynamics (AIMD) simulations, we reproduce the single excess electron stepwise catalytic CPD dissociation process in detail with an emphasis on the energy levels and molecular structure details associated with excess electrons. On the basis of the AIMD simulations on the CPD aqueous solution with two vertically added excess electrons, we exclude the early-proposed [2+2]-like concerted synchronous dissociation mechanism, and analyze the difference between the symmetry of the actual reaction and the symmetry of the frontier molecular orbitals which deeply impact the mechanism. Importantly, we propose a new model of the stepwise electron-catalyzed dissociation mechanism that conforms to the reality. This work not only provides dynamics insights into the excess electron catalyzed dissociation mechanism, but also reveals different roles of two excess electrons in two bond-cleavage steps (promoting versus inhibiting).

Key words: Excess electron, Cyclobutane pyrimidine dimer, Electron catalysis, Frontier orbital analysis, Ab initio molecular dynamics simulation

I. INTRODUCTION

As we all know, ultraviolet radiation is an important factor that induces cancerous or death of cells. Two adjacent thymine (T) in the DNA sequence can generate cyclobutane pyrimidine dimers (CPD) through cyclization at the C5/C5’ and C6/C6’ sites due to excessive far-ultraviolet radiation [1-3]. The existence of CPD can destroy the double helix structure of DNA and block the action of DNA or RNA polymerase, and it is considered to be the main source of DNA damage caused by ultraviolet radiation. Therefore, there is a lot of research work devoted to exploring the radiation
damage and repair mechanism of it [4–6]. Two different repair pathways of CPD damage were proposed in organisms: photolysis repair and nucleotide excision [7, 8], of which the former has received concentrated attention [9, 10]. Although studies have found that CPD photolytic enzymes only exist in archaea, bacteria, yeasts, insects, plants and marsupial mammals, no corresponding enzymes or analogues have been found in placental mammals represented by humans. However, by modifying the gene of mice to express CPD photolyase, it can greatly improve its resistance to skin cancer [11]. There are many explorations for the photolysis repair mechanism of CPD, e.g. direct observation of electron injection mechanism with ultrafast laser spectroscopy [1, 12, 13], observation of the process of electron transfer induced dimer cleavage by connecting CPD with a specific electron donor [14–16] to design new photolysis catalyst molecules [17, 18], and study of the effect of external factors such as magnetic field on the repair mechanism [19, 20]. There are also a large number of theoretical researches devoted to explaining the CPD configuration, reaction activation energy barrier, and potential energy surface involved in this mechanism [21–28].

The traditional explanation of the CPD photolysis repair mechanism generally adopts a single-electron step-by-step catalytic mechanism. In this mechanism, the corresponding enzymes or analogues first absorb blue photons and inject an excess electron into the CPD damage site through a series of photochemical reactions. The excess electron subsequently induces CPD to break the chemical bonds at C5–C5′ and C6–C6′ in turn, causing the dimer to return to the original adjacent thymine structures. In this type of repair mechanism, the reaction is considered to be close to spontaneous or with only a very small activation energy barrier [22, 27, 28]. After the CPD is induced to dissociate, the excess electron finally leaves the reaction area and continues to induce another dissociation repair or be re-captured by the photolyase. A theoretical study based on scanning of the static potential energy curve has also proposed a barrier-free concerted synchronous dissociation repair mechanism involving double excess electrons [29]. However, this mechanism has not yet been confirmed or reproduced by experiments or molecular dynamics simulations. As known, the polar solvent environment provided by aqueous solution is very critical for improving the electron affinity of bases or nucleotides including thymine [30, 31]. In order to better understand the traditional single-electron step-by-step catalytic repair mechanism of CPD, observe more kinetic details in it, and test the practical feasibility of the two-electron concerted synchronous dissociation repair mechanism, we conduct an ab initio molecular dynamics (AIMD) simulation study and our simulations support the single-electron mechanism and exclude the two-electron mechanism. In order to reveal the deeper reasons, we improve the interpretation model based on the frontier molecular orbital symmetry which was adopted in original two-electron mechanism, so that it can correctly match the electronic behavior observed in the existing dynamics simulations, and clarify different roles of single or double excess electrons played in different dissociation stages.

II. SIMULATION AND CALCULATION METHODS

We selected a hydrated CPD molecule (see FIG. 1 for the structural formula and corresponding site number) in a 15.05 Å × 15.05 Å × 15.05 Å periodically repeated cubic unit cell constructed with 100 water molecules as the representative system, and carried out a sufficient pre-processing balance in the electrical neutral state (5 ns classical molecular dynamics simulation plus 5 ps AIMD simulation). Our AIMD simulations are realized by the CP2K/Quickstep software package [32], adopting the BLYP functionals [33, 34] and DFT-D3 dispersion correction [35], with GTH pseudopotentials [36, 37], 300 Ry cutoff for plane wave basis set and TZV2P Gaussian group [38]. All pre-processing and simulation processes use a regular ensemble (NVT), with the Nosé-Hoover thermostat [39] to maintain the temperature of 300 K.
FIG. 2 The snapshots of the spatial distribution of the singly occupied molecular orbital (SOMO) during the single electron catalyzed dissociation process of CPD in aqueous solution. The yellow/green isosurfaces correspond to $-0.02/0.02$, respectively. The selected representative snapshot configurations. (A) The excess electron initially localized but does not break the C5–C5′ bond, (B) the C5–C5′ bond is broken, (C, D) the C5–C5′ bond is broken but the C6–C6′ bond is not, (E) the C6–C6′ bond is also broken, (F) CPD is dissociated completely.

III. RESULTS AND DISCUSSION

A. Single excess electron catalyzed dissociation

By vertically injecting a single excess electron into the pre-processed CPD aqueous solution, we dynamically observe the single-electron step-by-step catalytic dissociation process (FIG. 2), in good agreement with the experimental analysis. According to the time evolution of radius of gyration ($r_g$) of excess electron (FIG. 3(A)), we find that although it still has a tendency to diffuse state at the first time of injection by entering the bottom of the conduction band (CBM) of the water phase, the localization process to CPD also began almost immediately. This phenomenon stems from the good electron affinity of pyrimidine base molecules, especially when they are in a polar aqueous environment. By analyzing the time evolution of the Mulliken population of the spin density (FIG. 3(B)), we find that as the localization of excess electron is completed within nearly 100 fs, and almost all of the excess spin density is concentrated on CPD finally. In a more detailed analysis, we also find that the distributions of excess electrons at different sites of CPD show an evolution in space and time. In this localization process, the excess electrons are preferentially captured by the carbonyl group (C=O) at 4-site of CPD. By the end of localization, about 80% of excess electrons are concentrated at this site. But after that, the excess spin density transfers to the adjacent 5-site of CPD. By 200 fs, the 5-site has absorbed 80% of the excess spin density distribution from the whole system, and the remaining 20% is still concentrated at 4-site. In the corresponding time interval, the C5–C5′ bond cleavage of CPD is also completed simultaneously, and finally free radicals left at the 5/5′-stite, which corresponds to the accumulation of excess spin density there. The C5–C5′ bond length increases sharply from the initial 1.5 Å to about 2.75 Å (FIG. 3(C)), while the relative C6–C6′ bond length does not change significantly, indicating that the single excess electron dissociation process of CPD is a spontaneous step-by-step process. At the same time as the C5–C5′ bond breaks, there is a rapid drop in the energy level of the singly occupied molecular orbital (SOMO) of excess electron (FIG. 3(A), about 2 eV), and the adjacent highest doubly occupied orbital (HDMO) energy level rises (about 1.5 eV). The energy level gap between them almost disappears and the degenerate states appear. By observing the snapshots in FIG. 2, we can find that before and after the C5–C5′ bond breaking, the C6–C6′ bond also undergoes dissociation. Until the
FIG. 3 In the single electron catalyzed dissociation process of CPD, relevant electronic information and structural information evolve over time. (A) Time evolution of the radius of gyration \(r_g\) of excess electron and the energy level of the frontier molecular orbitals, (B) time evolution of the Mulliken spin density distribution at different sites of CPD, (C) time evolution of bond lengths and distances between the geometric centers of two pyrimidine rings of CPD, (D) time evolution of the dihedral angles of CPD where P1 and P2 denote the ring planes of two thymine moieties, respectively.

lengths of the two bonds both increase to about 4 Å, the single electron catalyzed dissociation of CPD declares its completion. After the dissociation, the excess spin density is redistributed between different sites of two thymine molecules: the 6-stite is scored about 60%, while the 4-site carbonyl group retains the remaining 40%. The bond lengths of C5–C6 and C5′–C6′ are slightly shortened, which corresponds to their return of the double bond characteristics after thymine separation. The biggest change is the energy level of the frontier MOs. During the dissociation process of the C6–C6′ bond, the degeneracy between HDMO and SOMO is broken and their gap quickly increases to \(\sim 3\) eV. At this time, SOMO is mainly distributed over and between the two thymine bases (with the help of weak \(\pi-\pi\) stacking), with a higher energy level \((-4\) eV). Therefore, it is not difficult to understand that the excess electron at this time has the opportunity to return to the conduction band minimum of solvent phase, which corresponds to its role as a catalyst in the single electron catalyzed dissociation mechanism.

In order to clarify the internal driving force of CPD single electron catalyzed dissociation mechanism, we also further analyze the changes in its molecular structure. We fit the six-membered pyrimidine ring planes (P1/P2) of two thymine structural units (T1/T2) by the least square method, and obtain the dihedral angle and geometric center distance between them. The distance between the geometric centers (T1–T2) can be combined with the C5–C5′ and C6–C6′ bond lengths to reflect the changes in the relative positions of the structural units during the dissociation process. The dihedral angle between P1 and P2 and the dihedral angle C5–C6–C6′–C5′ reflect the relative orientation of the structural units to a certain extent. Together, they reflect the huge structural tension in the original CPD molecule, which is the internal driving force for dissociation. We find that before dissociation, the two pyrimidine ring planes of CPD present a dihedral angle of about 50°, which makes C5–C5′ and C6–C6′ close enough to form a four-membered ring structure, but it noticeably runs counter to the tendency of parallel stacking between \(\pi\) bonds of two pyrimidine units. The dihedral angle of C5–C6–C6′–C5′ is also not in a flat
state, and has an angle of about 20°. This aspect is the natural chair conformation tendency of a non-aromatic four-membered ring, and it is also to accommodate the steric hindrance of adjacent groups at the four sites. We notice that in the neutral conformation, the two methyl groups at C5–C5′ have a clear tendency to repel each other, which inevitably makes much greater structural tension here than C6–C6′. Moreover, the 5/5′-site is also closer to the preferential capturing groups of excess electrons (i.e. the 4/4′-site carbonyl groups), and thus it has a higher rank to accept excess electron. Therefore, we propose that the dissociation of the C5–C5′ bond is a fixed-point release of the structural tension accumulated in the original dimer molecule with the help of excess electron. In the process of the C5–C5′ bond breaking, the C5–C6–C6′–C5′ dihedral angle also rapidly increases to 45°, showing the huge steric hindrance between adjacent methyl groups at 5-sites. At the same time, the dihedral angle of P1–P2 is reduced to 35°, which naturally conforms to the orientation of π-π stacking in parallel between the imidazole rings. However, since there is still a bond between C6 and C6′, the geometric center distance of P1–P2 is almost unchanged (only slightly increased by 0.5 Å). Although such a short C6–C6′ distance increases the repulsion between different groups, it may also strengthen the π-π stacking to a certain extent. Thus, we can find that in the 300 fs transition period between the bond cleavage of C5–C5′ and C6–C6′, except for the dihedral angle of P1–P2 is still evolved in the direction of decreasing, other parameters almost do not change. After the C6–C6′ bond cleavage, the distance between the geometric centers of P1–P2 increases simultaneously, and the dihedral angle of P1–P2 is reduced to 10°, corresponding to the final release of structural tension. The two thymine bases after dissociation have almost parallel orientations, but the increase in their distance after the C6–C6′ bond disappears and also weakens the π-π accumulation between them, resulting in a reduction in their ability to hold the excess electron together, so that the excess electron can easily leave after completing the catalyst mission.

### B. Double excess electron catalyzed dissociation

Under the unrestricted open-shell approximation, after vertical injection of double excess electrons in a singlet state, we do not obtain a complete barrier-free synchronous dissociation process as expected in Ref.[29], instead, the dissociation stops at the C5–C5′ breaking. Although we try to extend the simulation time to more than 5 ps or change the initial configuration, we fail to observe the bond cleavage of C6–C6′, which has to make us doubt the feasibility of this double electron mechanism in reality. After analyzing in detail the behavior of double excess electrons when they participate in the C5–C5′ bond breaking, and comparing with the previous single excess electron stepwise catalytic mechanism, we find that the roles of two excess electrons at this time are not as expected in the literature [29]. By taking a snapshot of the spatial distribution of molecular orbitals occupied by the double excess electrons of different spins (α or β) (FIG. 4), we clearly find that the spin state of the entire system undergoes a series of transformation from the closed shell (CS) diffuse state, to the broken-symmetry singlet state (BS) with a single excess electron localized at CPD, and finally to the end of returning to the CS localized state with only the C5–C5′ bond cleavage. The gyration radius ($r_g$) of double excess electrons and the squared spin value ($\langle S^2 \rangle$) of the whole system more quantitatively reflect this process (FIG. 4(A)): the α excess electron finishes its localization to the CPD earlier before 30 fs, but the β excess electron is still retained in the conduction band minimum, making the system appear in the BS state with $\langle S^2 \rangle$=1. This is very similar to the phenomenon we get in the two-electron reduction process of carbon dioxide molecules and can also be attributed to the slower molecular and hydration structure adjustment than charge transfer. Thus, the system does not immediately provide enough possibilities for localization of the second electron, resulting in the short-time charge separation state. In this system, the existence of the BS state continues until around 130 fs. With the cleavage of the C5–C5′ bond, the HOMO energy level occupied by the localized excess electron decreases, and the diffuse excess electron still in the conduction band minimum flows into this orbital synchronously, making whole system return to the CS state. In the single electron catalytic mechanism, the C5–C5′ bond breaking also creates free radical characteristics, which is also conducive to localization of the second excess electrons. However, we cannot confirm whether the existence of double excess electrons provides an additional boost to the C5–C5′ bond breaking, because the changes in the electronic or molecular structure in FIG. 5 are very similar to the corresponding stage in FIG. 3.
FIG. 4 The snapshots of spatial distribution of the highest occupied molecular orbital (HOMO) during the two-electron catalyzed dissociation process of CPD. The yellow/green isosurfaces correspond to $-+0.02$, respectively, and the corresponding different spins are also marked in the upper right corner. The selected representative snapshot configurations include: (A) the closed shell singlet diffuse state after vertical injection of two excess electrons, (B) one of excess electrons localizes and the other continues to diffuse, resulting in a broken-symmetry open-shell singlet state, (C) the diffuse electron also localizes synchronously with the C5–C5′ bond cleavage, (D, E) after the C5–C5′ bond breaking.

FIG. 5 In the double electron catalyzed dissociation process of CPD, relevant electronic and structural information evolve over time. (A) Time evolution of the radius of gyration ($r_g$) of excess electrons and the $\langle S^2 \rangle$ value of the whole system, (B) time evolution of the energy level of the frontier molecular orbitals, (C) time evolution of bond lengths and the distances between the geometric centers of the two pyrimidine rings of CPD, (D) time evolution of dihedral angles of CPD where P1 and P2 denote two thymine molecular planes, respectively.

By comparing the spatial distribution of frontier molecular orbitals, we can find that in the single or double-electron mechanisms of the C5–C5′ bond cleavage, the spatial distribution (symmetry) characters are almost the same. The difference is that the increase in the occupancy of the highest occupied molecular orbital
FIG. 6 After ionization of double electrons, structural information in the C6–C6′ dissociation evolves over time. (A) Time evolution of bond lengths and distances between the geometric centers of the two pyrimidine rings of CPD, (B) time evolution of dihedral angles of CPD.

FIG. 7 The concerted synchronous double electron induced dissociation mechanism proposed in the Ref.[29].

seems to be an obstacle to the breaking of the C6–C6′ bond. Thus, we choose the structure at this time as the initial stage, and perform an AIMD simulation on the neutral system that is created by vertically ionizing the two excess electrons. As shown in FIG. 6, almost immediately after vertical ionization, the C6–C6′ bond begins to spontaneously dissociate. The process is basically the same as the second half of the single-electron catalytic dissociation, and thus we do not repeatedly describe this process here. However, we can find that for the C6–C6′ bond breaking, the presence of excess electrons in the CPD molecular orbital seems to be a disadvantage. When there is no excess electron, the dissociation happens immediately, and when there is only one excess electron, the dissociation needs to wait for a certain time. When double excess electrons exist, the dissociation is completely inhibited until the re-ionization occurs, which clearly requires additional energy.

In short, our AIMD simulations do not support the existence of a double excess electron dissociation pathway for CPD, at least until the secondary ionization excitation conditions are available, it is not an alternative pathway, although it also has spontaneous C5–C5′ bond breaking. At the same time, it should be noted that even if the ionization excitation is used to promote the dissociation, the mechanism seems to be equivalent to the single electron step-by-step mechanism, instead of similar synchronization [2+2] split mechanism of the ring predicted in Ref.[29]. In order to clarify the chemical nature, we try to analyze the mechanism based on the symmetry of the frontier molecular orbitals.

C. The frontier molecular orbital analysis

First of all, it is necessary to evaluate the barrier-free concerted synchronous double electron induced dissociation mechanism expected in Ref.[29] as shown in FIG. 7. We find that this mechanism is the reverse process (ring split) of the classic [2+2] ring-forming reaction which is a classic photoreaction in textbooks and is still widely used in various organic and photocatalytic reactions [40, 41]. Its most prominent feature is synchronization. In the explanation of the expected mech-
anism, the parts outside the core area (only four p-type orbitals of C5C5′/C6C6′) of the reaction are all ignored, making our research ideas clear enough. Before the excess electrons are added, all the occupied molecular orbitals of the system are just conducive to the bonding of C5−C5′ and C6−C6′, and the πL−πR orbital conducive to dissociation is not occupied, and thus it does not harm the stability of the structure. When double excess electrons are introduced, they naturally occupy the πL−πR orbital which weakens the bonding effect of C5−C5′ and C6−C6′. In the subsequent structural adjustment of the [CPD]2− dianion, the two bond lengths spontaneously elongate according to this trend, thereby raising the energy level of the πL+πR* orbital until it is higher than the highest occupied πL−πR orbital energy level. At this time, the ionization of double excess electrons can directly cause the bond formation at C5−C5′ and C6−C6′ to be actually cancelled, and thus the two thymine structural units are separated. Based on the potential energy curve scanning under a Cs symmetry restriction, Davis et al. [29] did not find an energy barrier in this process, and thus they proposed a [2+2]-like barrier-free concerted synchronous double electron induced dissociation mechanism.

By analyzing the mechanism proposed in Ref.[29] carefully, we find some deviations from the actual reaction process. First, there is no Cs symmetric dimer conformation actually, which can be derived from the 20° twisted C5−C6−C6′−C5′ dihedral angle under the neutral condition. As we all know, a slight distortion of a highly symmetric structure can decrease the degeneracy of the molecular orbitals. When the distortion is sufficiently large, it is possible to change the order of energy level and affect the actual mechanism. Secondly, the expected [2+2]-like mechanism approach even requires a higher D4h symmetry restriction than Cs. In the other word, this expected mechanism introduces a hidden plane of symmetry perpendicular to the C5−C6 and C5′−C6′ bonds, forcing the changes of C5−C5′ and C6−C6′ to be always synchronously equivalent. However, in the actual environment, the 5-sites and 6-sites are clearly not equivalent, which mainly comes from two facts. One of them is the huge steric hindrance between the two methyl groups at 5-sites, causing the C5−C5′ to accumulate more structural tension than C6−C6′. On the other hand, the most accessible site for excess electrons in thymine is 4-site which is close to C5−C5′, resulting in the flow of excess electrons according to the 4→5→6 transfer mode. Thus, in our actual AIMD simulation, we can always find that the C5−C5′ bond dissociation prior to that to the C6−C6′ bond. Finally, the proposed mechanism in Ref.[29] is also difficult to explain why the traditional single electron induced dissociation mechanism is of step-by-step, although they emphasized that double electrons may be more beneficial to this mechanism. Furthermore, the non-explicit solvation model in that proposed mechanism also has an obvious defect, that is, the excess electrons forming a diffuse state by entering the conduction band minimum may be ignored. There is a large amount of real hydration environment in our AIMD simulation, which provides the potential for excess electrons to diffuse or even pre-solvated, so that when double excess electrons cannot be localized to the target at the same time, the system can experience a transitional charge separated state (i.e. open-shell singlet BS state). On one hand, it ensures that the reaction zone does not accumulate excessive negative charge instantly, and it also reduces the energy level of the localized electrons in disguise. In the non-explicit solvation model, two excess electrons can only be attached to the CPD at the same time. Because the energy level of the doubly-occupied molecular orbital is too high, it appears as a dipole bound state, which is not true in the bulk environment.

Although this literature mechanism model has many failures in describing actual problems, the idea of simplifying the complex reaction process to a small number of atoms and molecular orbitals in the core reaction area and then using basic theories such as symmetry to analyze its essence has been the standard paradigm of scientific research since ancient time. We try to continue to use this analytical idea, and carry out a more realistic analysis of the single electron step-by-step catalytic dissociation mechanism, and try to find the reason for double excess electrons to inhibit the C6−C6′ dissociation, as shown in FIG. 8. First, we support that before the dissociation of CPD, the frontier molecular orbital of the reaction zone should be regarded as having D4h symmetry. Based on this, we can understand that although the C5−C5′ independent bond-breaking behavior occurs first, the SOMO wave function occupied by excess electrons during the bond-breaking process (FIG. 2(B)) still has obvious πL−πR characteristics, that is, there is an obvious nodal plane in the direction almost perpendicular to C5−C5′ and C6−C6′. The broken bond of C5−C5′ also comes from the weakening...
of the bonding effect brought about by excess electrons entering this orbital, as well as the steric hindrance and electron flow outside the model zone.

After the C5−C5′ bond is broken, the symmetry of the system degrades to $C_{2v}$, which is in line with the actual dynamics simulation but different from the mechanism model in the literature [29]. At this time, the combination of the $\pi_L + \pi_R^*$ orbital that is not considered under the $D_{4h}$ symmetry becomes extremely important. Under the $D_{4h}$ symmetry, the $\pi_L + \pi_R^*$ and $\pi_L - \pi_R^*$ orbitals are degenerate, and because the bond formation/breaking effects on C5−C5′ and C6−C6′ cancel out each other, they inevitably are not occupied. After the symmetry deteriorates to $C_{2v}$, they naturally split due to the Jahn-Teller effect, and the energy level of the $\pi_L + \pi_R^*$ orbital is rapidly reduced due to the breaking of the C5−C5′ bond, and soon falls below the $\pi_L - \pi_R$ orbital which is not suitable for the symmetry analysis at this time. Certainly, we also need to emphasize that the huge C5−C6−C6′−C5′ torsion angle in the system clearly shows that it does not have any high symmetry, but it further strengthens the Jahn-Teller effect. In the actual reaction, the distance and orientation between C5 and C5′ at this time no longer allow them to form a bond effectively, so we can surprisingly find the bonding effect between the $\pi_L + \pi_R^*$ orbital and $\pi_L^* + \pi_R^*$ orbital is converging (only strengthening C6−C6′), and this corresponds to the emergence of a degenerate state of the energy level of the Frontier occupied molecular orbital (FIG. 3(A) and FIG. 5(B)). Moreover, we can infer that one of the major features of the $\pi_L + \pi_R^*$ orbital is the single-sided double bond structure, as shown in FIG. 2(C) and FIG. 4(D), which also supports the mechanism proposed here to a certain extent. We can also find that the essence of this split ring mechanism: excess electron can degrade the symmetry of the bonding mode of the whole system by occupying molecular orbitals with lower symmetry, thereby inducing a readjustment of the configuration and energy level alignment.

In the subsequent C6−C6′ bond breaking process, the model zone returns to $D_{4h}$ symmetry, so the $\pi_L + \pi_R^*$ orbital becomes untimely again. As the distance between C5−C5′ and C6−C6′ increases, the energy level of the $\pi_L - \pi_R$ orbital becomes lower, while that of the $\pi_L + \pi_R^*$ orbital increases. After this series of changes, the degeneracy of the frontier molecular orbitals occupying the track is forced to increase (FIG. 3(A)). At this time, we believe that the occupation of the $\pi_L^* + \pi_R^*$ orbital by excess electrons is not conducive to the dissociation of C6−C6′. Meanwhile, the scanning study on the bulk potential energy surface of this process also reveals a tiny energy barrier (1.5−2.5 kcal/mol) for the C6−C6′ bond breaking [22, 27, 28]. Certainly, excess electrons can be excited or ionized before the C6−C6′ dissociation, but this requires additional energy. According to the Koopmans theorem, the vertical detachment en-
ergy of a single excess electron is about 2.5 eV (FIG. 3(A)). However, the obstacle caused by the single electron occupying the $\pi_{L}^{*} + \pi_{R}^{*}$ orbital does not prevent the dissociation of C6−C6′ in practice. Therefore, the leaving of excess electrons can wait until the dissociation is completed and the SOMO energy level is significantly increased, thus achieving the characteristics of the actual catalytic reaction. When we re-examine the double electron catalyzed process, we can find that localization of the second electron is synchronized with the C5−C5′ bond breaking, which may increase the occupancy of the $\pi_{L}^{*} - \pi_{R}$ orbital and increase the bond breaking rate. But this bond breaking is a spontaneous process based on the inherent structural tension of CPD, and excess electrons only act as catalysts. For the subsequent C6−C6′ bond breaking which is the rate-determining step in whole reaction, the doubly-occupied $\pi_{L}^{*} + \pi_{R}^{*}$ and $\pi_{L}^{*} + \pi_{R}$ are noticeably more unfavorable for its occurrence. Thus, we can finally assert that under actual conditions, the concerted synchronous double electron catalyzed dissociation process does not occur in practice, at least when the whole system is in the ground state, and the second excess electron actually is an inhibitor (or negative catalyst) rather than a booster (positive catalyst).

IV. CONCLUSION

In summary, we explore the process of dissociation and repair of CPD catalyzed by excess electrons through AIMD simulations. In our research, we reproduce the single excess electron step-by-step catalytic dissociation mechanism, and analyze the electron capture pathways, structural tension changes, and frontier molecular orbital changes, so as to understand the dynamics mechanism and details more intuitively and deeply. We also try to realize the concerted synchronous double electron catalyzed dissociation pathway expected in the literature on this basis, but failed. After reasonable modeling and analysis of the changes in the symmetry of the frontier molecular orbitals in the reaction zone, we obtain a more reasonable description of the dissociation process, and produce a deeper understanding of the single excess electron step-by-step catalyzed CPD dissociation mechanism in aqueous solution. At the same time, we exclude the concerted synchronous double electron induced dissociation from the perspective of basic theory and simulation, explain the distortion of the model that came to the wrong assump-

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