A computational exploration of some transnitrosation and thiolation reactions involving CH_3SNO, CH_3ONO and CH_3NHNO

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Nitric oxide (NO) is a biologically active species and its carrier molecules RXNO (X = S, O, NH) have drawn significant attention recently. In the present work, the CBS-QB3 level of theory was used to study the transnitrosation and thiolation reaction between MeXNO (X = S, O, and NH) molecules and three reactive forms of the methanethiol: the neutral molecule, MeSH, the anion, MeS^-, and the radical, $MeS^{\bullet}$. The transnitrosation and thiolation reactions between MeXNO and MeSH have the highest barriers, both with and without a molecule of water assisting. Reactions with MeS^- proceed with much lower barriers, while reactions with radical $MeS^{\bullet}$ have the lowest barriers. Comparing the reactions of MeXNO (X = S, O, NH), both transnitrosation and thiolation are more favorable for X = S than X = O or NH.

Introduction

The endogenous production of nitric oxide (NO) is associated with crucial biological events, such as endothelium-dependent relaxation, neurotransmission, and cell mediated immune response.[1,2] Since NO is a highly reactive, short-lived free radical, biological systems have evolved specific systems for NO storage, transport, and delivery. *S*-Nitrosothiols (RSNO) have been shown to be potent smooth muscle relaxants and inhibitors of platelet aggregation, and are the best candidates for the endogenous storage and transport of NO.[3–5] *S*-Nitrosylation is also an important mechanism for post-translational regulation of many classes of proteins.[6]

N-Nitroso compounds have been extensively studied because of their potent carcinogenic activities.[7] In 1956, nitrosodimethylamine was reported to induce liver cancer when fed to rats.[8] Later it was found that nitrosamines could alkylate proteins and nucleic acids.[9,10] As a result of these early findings, *N*-nitrosamines are generally considered as carcinogens.[7] *O*-Nitroso compounds, such as *tert*-butyl nitrite (TBN), amyl nitrite (AMN), and isoamyl nitrite (IAMN), have been used clinically as angina pectoris relieving agents for more than a century.[11,12] It has been shown that the vasodilator effect of these agents is exerted by releasing NO.[13] Currently, specific attention has been focused on *S*-nitroso compounds which are directly involved in many NO related biological functions. Extensive experimental studies have been carried out on the generation, stability and reactivity of these nitroso compounds.[12,14–28]

Two aspects of these nitroso compound activities are of particular interest in the present study. One is transnitrosation which contributes to NO transfer but does not involve free NO during the procedure. In aqueous solution, experiments show that sulfur-to-sulfur transnitrosation is a second order reaction with a barrier height in the range of 13–18 kcal mol^{-1}. The reaction rate depends on pH, suggesting that RS^- is the reactive species.[26,29–33] This is supported by Houk's work[34] which has shown that transnitrosation between thiol and nitrosothiol proceeds by the attack of a nucleophilic thiolate anion on an electrophilic RSNO through a novel anionic $RSN(O)SR^-$ intermediate. This intermediate has recently been characterized by ^{15}N and ^{1}H NMR.[35] Sulfur-to-nitrogen transnitrosation has also been observed experimentally.[36] Whether it plays a biological role in the generation of nitrosamines needs further research; nevertheless, it is a possible pathway for NO trafficking.

Another important reaction is thiolation in which both homolytic and heterolytic cleavage of the X–NO bond contribute to NO release. Several groups have reported the homolytic and heterolytic bond dissociation energies (BDEs) of selected nitroso compounds.[14,19,37,38] It has been shown that heterolytic BDEs are on average 29 ± 3.5 kcal mol^{-1} higher than homolytic BDEs, and the latter are in the range of 25–32 kcal mol^{-1}. Given these bond dissociation energies, thermal dissociation of these bonds is difficult under physiological conditions. However, previous studies revealed that metal ions, such as Cu^+, Fe^{2+}, and Hg^{2+} can catalyze the decomposition of *S*-nitroso compounds.[39]

Several theoretical studies have been focused on the bond dissociation energies of X–NO bonds and numerous studies of the kinetics of RXNO reactions have been reported *in vitro*.[14,15,19,20,28,36–38,40,41] However, elementary reactions of nitroso compounds are not yet well understood. In this paper, we present some further explorations of possible mechanisms for transnitrosation and thiolation reactions under a variety of conditions.

$$MeSH + MeXNO \rightleftharpoons MeSNO + MeXH \quad (1)$$

$$MeSH + MeXNO \rightleftharpoons MeSXMe + HNO \quad (2)$$

Depending on the conditions and the solvent, the reactive form of the thiol can be in the neutral molecule, MeSH, the anion, MeS^-, or the radical, $MeS^{\bullet}$. In aqueous solution, the pK_a of MeSH

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is 10.3. At pH = 7.0, it exists mainly as neutral MeSH, but there is also a significant concentration of the anion, MeS^-. In organic solvents, the pK_a is much higher; for example in MeCN, the pK_a of MeSH is 26.3 and only the neutral form of MeSH is present. The radical $MeS^\bullet$ can be generated from MeSNO photochemically or by thermal decomposition. Therefore, we have examined each of these three forms as reagents in transnitrosation and thiolation reactions.

Methodology

Molecular orbital calculations were carried out using a development version of the Gaussian series programs[42] and the CBS-QB3 method developed by Petersson and co-workers.[43,44] This method uses B3LYP[45–47] density functional theory and the 6-311G(2d,d,p) basis set to calculate optimized geometries and vibrational frequencies, and single point calculations at CCSD(T), MP4SDQ, and MP2 levels with smaller basis sets. The known asymptotic convergence of pair natural orbital expansions is used to extrapolate the MP2 calculations with a finite basis set to the estimated complete basis set limit. The CBS-QB3 level of theory has a mean absolute deviation of 0.87 kcal mol^{-1} for comparisons in the G3 validation set of atomization energies, proton affinities, ionization energies and electron affinities. It has also been shown to give reliable bond dissociation energies and heats of reactions for nitric oxide compounds.[14,34,37] Free energies of solvation were estimated using the conductor-like polarizable continuum model (CPCM)[48] computed using the B3LYP[45–47] density functional theory with the 6-31G(d,p)[49,50] basis set and gas phase B3LYP/6-31G(d,p) optimized geometries. Enthalpies in solution were obtained by adding the solvation energy difference to the CBS-QB3 gas phase enthalpies. We chose the smallest molecules MeSNO, MeONO and MeNHNO as models. Since previous work showed that the *cis* conformer is the most stable one for primary RXNO compounds,[14,19] our study is based on the *cis* conformer only.

Results and discussion

Transnitrosation

As a free radical, NO has a very short life-time. However, NO storage molecules such as RXNO significantly increase its life-time and enable NO to be transported in biological systems. An important reaction of RXNO molecules is transnitrosation, in which NO can be transferred from one molecule to another without involving free NO. Binding to an RX group alters the stability of NO and transnitrosation reactions can lead to a variety of molecules, possibly including *N*-nitrosamines which are known to be harmful. In this section we examine transnitrosation reactions involving MeXNO (X = S, O, NH) with neutral methanethiol, MeSH, the anion, MeS^-, and the radical, $MeS^\bullet$. For consistency the reactions are written and discussed in the MeXNO → MeSNO direction, but some reactions are energetically more favorable in the reverse direction.

MeSH + MeXNO Transnitrosation reactions. Fig. 1 shows the structures and energetics for sulfur-to-sulfur transnitrosation, MeSH + MeSNO → MeSNO + MeSH, in the gas phase. This would also be a suitable model for reactions in a non-polar solvent where the neutral form of methanethiol is more stable than the thiolate. Because the transition state (TS) involves a four-membered ring, the barrier is rather high (40.9 kcal mol^{-1} relative to the reactant complex). From this transition state, the reaction proceeds to an intermediate that is 38.4 kcal mol^{-1} more stable than the TS. A similar intermediate has been found by Houk in transnitrosation reactions with thiolate, MeS^- attacking MeSNO.[34] However, the barrier for transnitrosation with MeS^- is much lower since it does not require the breaking of an S–H bond. For both MeSH and MeS^-, the reaction proceeds from the intermediate over a second transition state that is the mirror image of the first, and then on to products.

Barriers that involve hydrogen transfer in a four membered ring can be dramatically lowered by adding one or two molecules of water.[51–60] Therefore, we studied the reaction catalyzed by a molecule of water. As shown in Fig. 2, the energy profile along the reaction coordinate is qualitatively similar to the uncatalyzed reaction, with two transition states separated by an intermediate. In the transition state, the water accepts a proton from MeSH and donates a different proton to the nitrogen of MeSNO. The barrier height is lowered significantly, but is still relatively high (33.4 kcal mol^{-1} above the reactant complex) and comparable to the homolytic dissociation energy of MeSNO (CBS-QB3: 32.4 kcal mol^{-1}).[14,19,37,38] In the absence of water, a second molecule of MeSH can catalyze the reaction, though the effect may not be as large. The effect of bulk solvent can be modeled implicitly with the CPCM polarizable continuum method (Fig. 2). When measured from the reactant complex, the barriers in water and

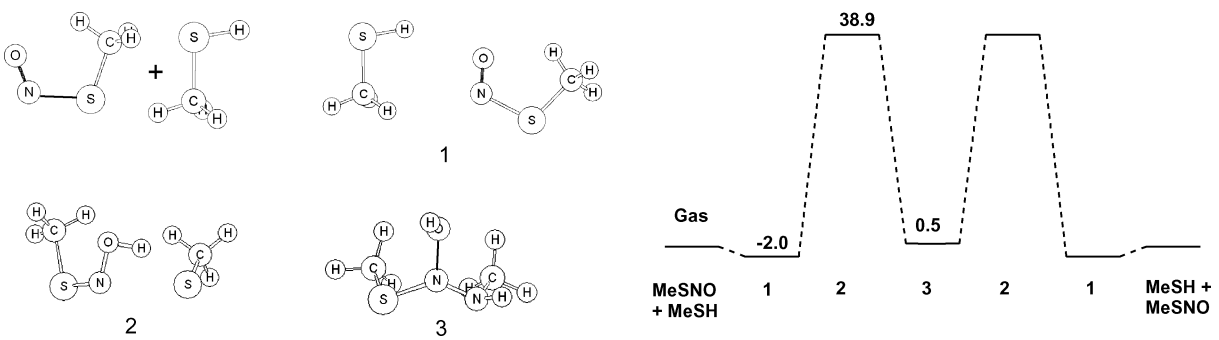

Fig. 1 Transnitrosation between MeSH and MeSNO in the gas phase at the CBS-QB3 level of theory.

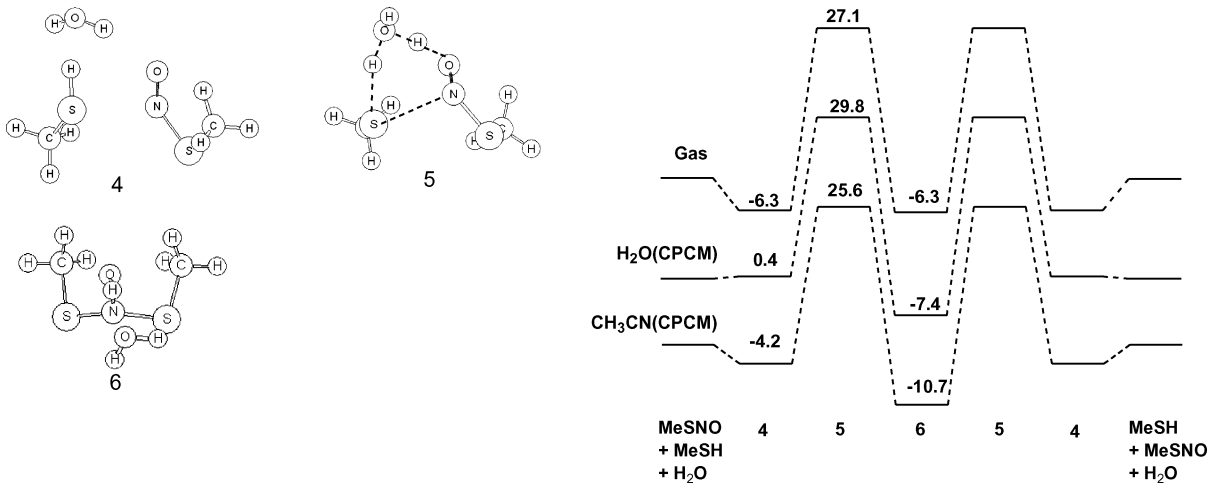

Fig. 2 Transnitrosation between MeSH and MeSNO with one molecule of water assisting in the gas phase and in solution (water and CH_3CN using the CPCM model).

acetonitrile are only 4 kcal mol^{-1} lower than in the gas phase. Thus solvation lowers the barrier by only a modest amount, provided one molecule of water is included explicitly to assist in the proton shuttle.

The reaction profiles for MeSH + MeNHNO ⇌ MeSNO + $MeNH_2$ and MeSH + MeONO ⇌ MeSNO + MeOH (Fig. 3–6) are similar to the sulfur case in that there are two high energy transition states separated by an intermediate. The reaction for MeONO is slightly exothermic in the gas phase and in solution, whereas the reaction for MeNHNO is slightly endothermic. For both systems, the gas phase barriers are higher than for transnitrosation with MeSH and comparable to the MeX–NO homolytic bond dissociation energies (BDE: 43.9 and 49.0 kcal mol^{-1}, respectively at CBS-Q level of theory).[19] Introducing an explicit molecule of water brings the barriers relative to the reactant complex into the same range as for MeSH. Similar to the sulfur case, adding the effect of solvents *via* CPCM cause only a 2–4 kcal mol^{-1} change in the barrier height relative to the reactant complexes.

MeS$^-$ + MeXNO Transnitrosation reactions. In experiment, the rate of transnitrosation between thiols is increased by an increase of thiol acidity, elevated pH, and enhanced electrophilicity of the nitrosothiol. These results suggest a mechanism involving the attack of a nucleophilic thiolate anion on an electrophilic RSNO. As mentioned above, Houk has found a low energy pathway for transnitrosation between MeS$^-$ and MeSNO *via* a novel intermediate, MeSNO(−)SMe.[34] The optimized structures

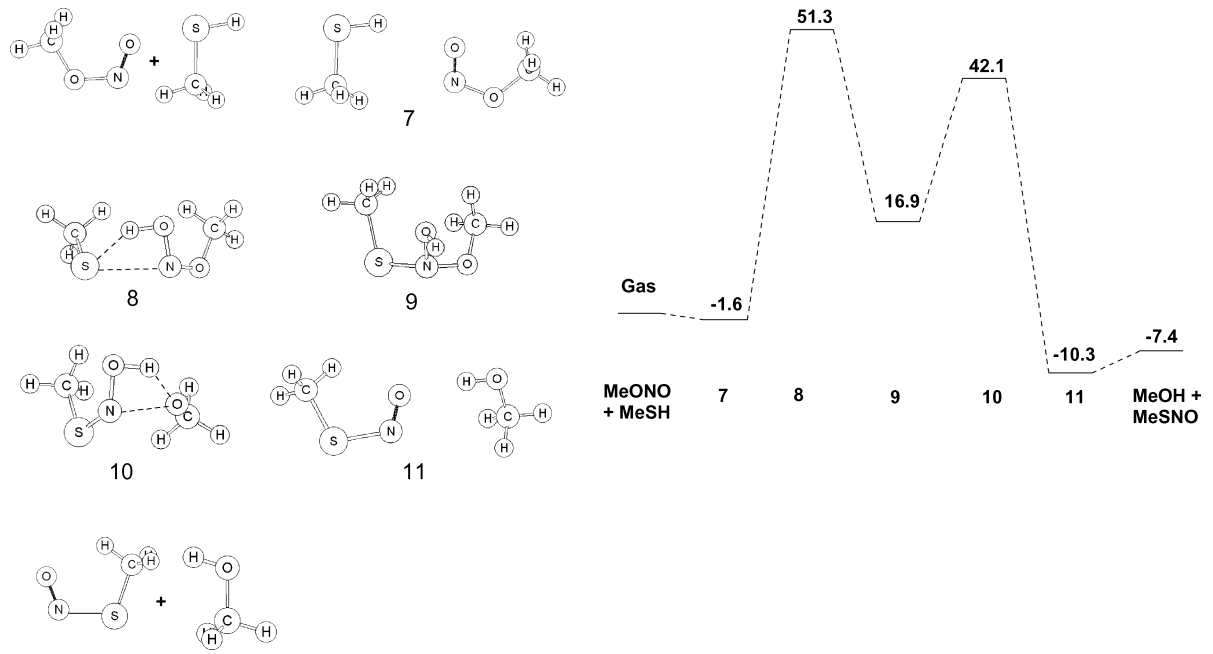

Fig. 3 Transnitrosation between MeSH and MeONO in the gas phase at the CBS-QB3 level of theory.

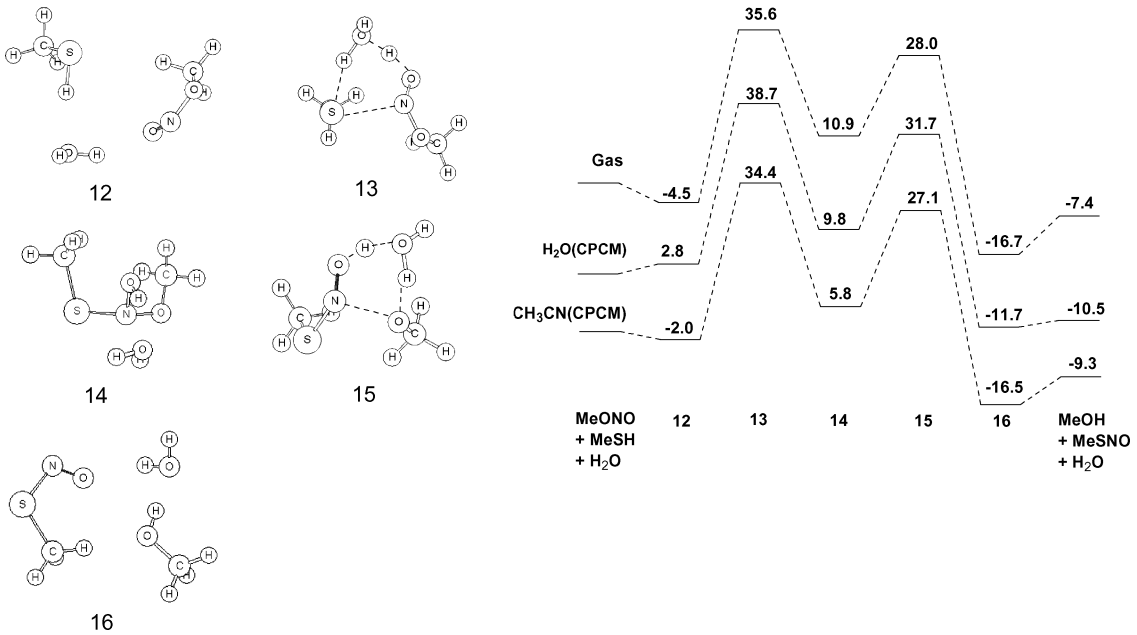

Fig. 4 Transnitrosation between MeSH and MeONO with one molecule of water assisting in the gas phase and in solution (water and CH_3CN using the CPCM model).

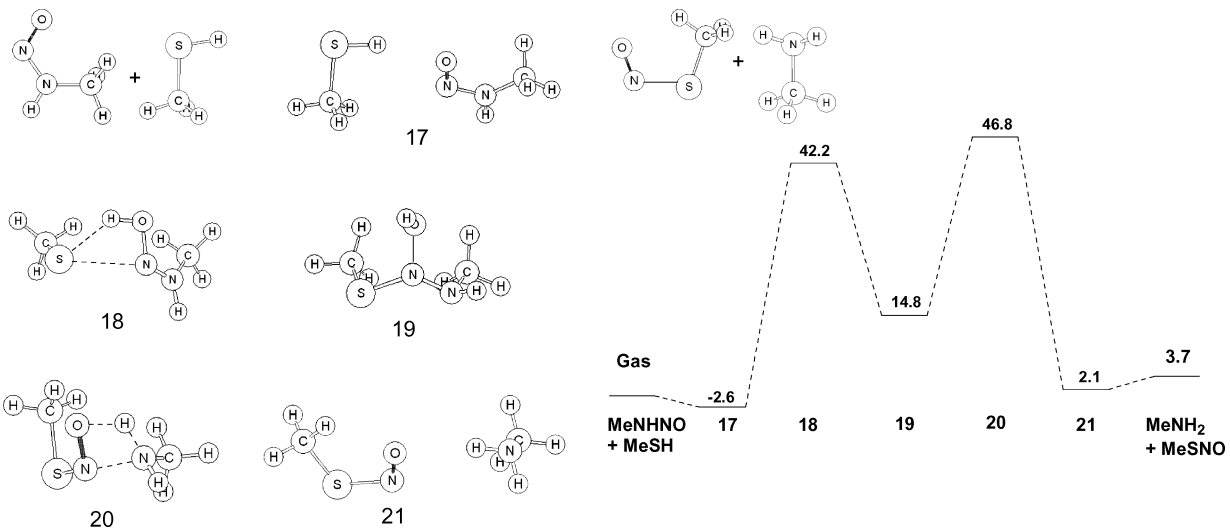

Fig. 5 Transnitrosation between MeSH and MeNHNO in the gas phase at the CBS-QB3 level of theory.

for reaction MeS^- + MeSNO in CBS-QB3 level of theory are shown in Fig. 7 for comparison with MeS^- + MeXNO (X = O, NH). For the reaction between MeS^- and MeONO (Fig. 8), the first transition state and the anionic intermediate are *ca.* 5 kcal mol^{-1} above the reactant complex (before correction for zero point energy and higher order correlation effects, the transition state is 0.98 kcal mol^{-1} higher than the intermediate). For MeS^- and MeNHNO (Fig. 9), no transition states could be located and the energy increases monotonically from reactant complex to products. The reactions of MeS^- with MeXNO (X = O, NH) are both quite endothermic (18.4 and 50.1 kcal mol^{-1}, respectively) and are unlikely to occur in the gas phase or in non-polar solvents without some means of stabilizing the MeX^- anions. These energetics are consistent with the fact that the gas phase basicity of MeO^-, and $MeNH^-$ are 24.3 and 43.8 kcal mol^{-1} greater than MeS^-.

For MeS^- + MeSNO and MeONO, the largest solvent effect is for the energy of the reactant and product complexes to the isolated species. By contrast solvent has only a modest effect on the barriers relative to these complexes. Because of the high barrier for X = O, sulfur-to-sulfur transnitrosation occurs much more ready than oxygen-to-sulfur transnitrosation by this mechanism. The large solvent effect computed for MeS^- + MeNHNO in aqueous solution is probably an artifact.

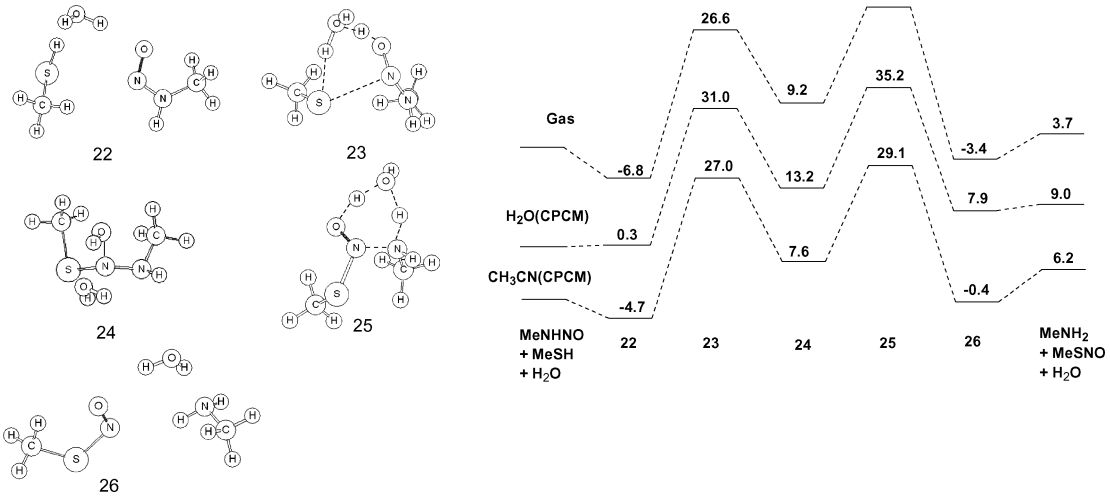

Fig. 6 Transnitrosation between MeSH and MeNHNO with one molecule of water assisting in the gas phase and in solution (water and CH_3CN using the CPCM model).

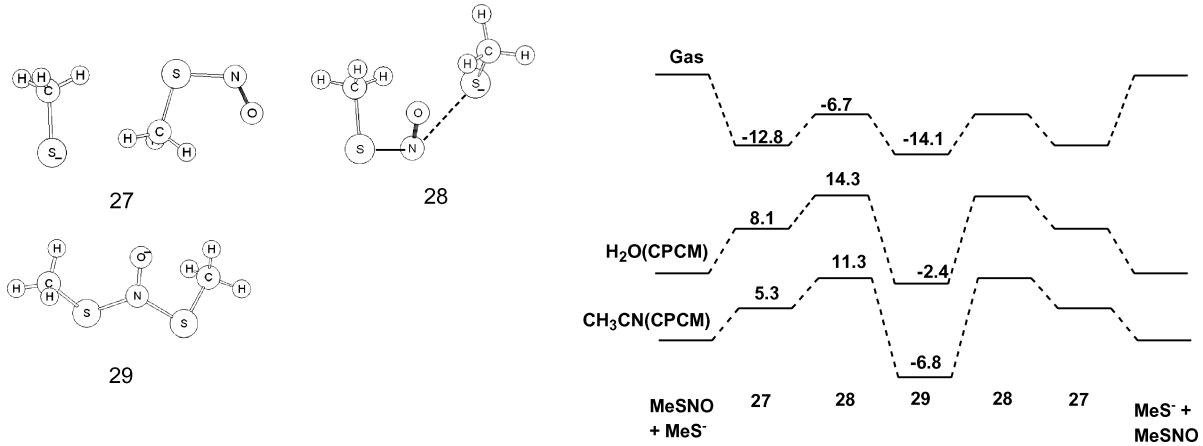

Fig. 7 Transnitrosation between MeS^- and MeSNO in the gas phase and in solution (water and CH_3CN using the CPCM model).

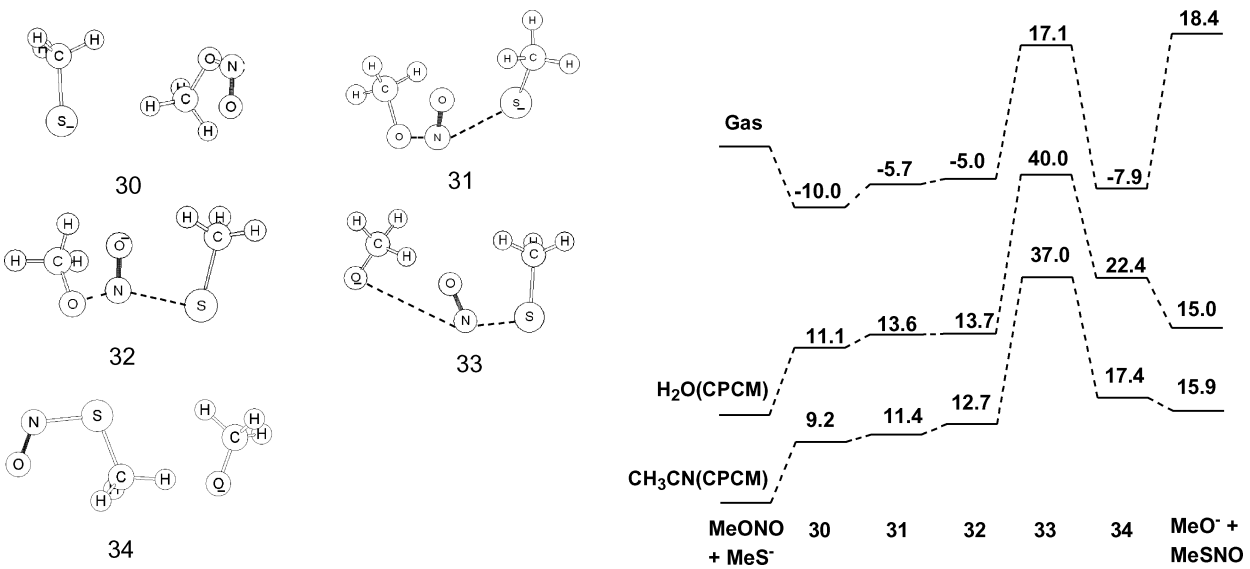

Fig. 8 Transnitrosation between MeS^- and MeONO in the gas phase and in solution (water and CH_3CN using the CPCM model).

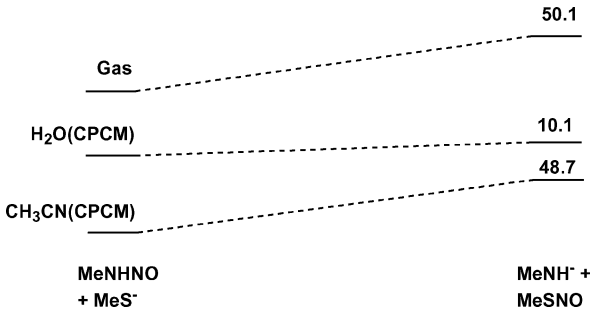

Fig. 9 Transnitrosation between MeS^- and MeNHNO in the gas phase and in solution (water and CH_3CN using the CPCM model).

MeS• + MeXNO Transnitrosation reactions. In solution, thiol radicals can be produced either by thermal dissociation or by photodissociation. MeS• + MeSNO form a weakly bound complex in the gas phase (Fig. 10). However, a relaxed scan shows that there is no barrier for the direct reaction between MeS• + MeSNO to form the intermediate which is 10.8 kcal mol^{-1} lower than reactant complex. Thus NO can be very easily transferred between MeS• and MeSNO. The reactions between MeS• and MeXNO (X = O, NH, shown in Fig. 11 and 12) have higher barriers (15.5 and 24.6 kcal mol^{-1}, respectively) and are both endothermic (10.8 and 16.5 kcal mol^{-1}, respectively). However, compared with the corresponding reactions between MeS^- and MeXNO (X = O, NH), these two reactions are less endothermic, especially for X = NH. In contrast to the reactions with MeS^- anion, solvation causes only modest changes in the energetics of reactions between MeS• radical and MeXNO. The barriers for MeS• in the gas phase and in solution are lower than the homolytic bond dissociation energy for the corresponding MeX–NO bond (32.4, 43.9, and 49.0 kcal mol^{-1} for X = S, O, and NH respectively), and are consistent with the trend in the BDEs.

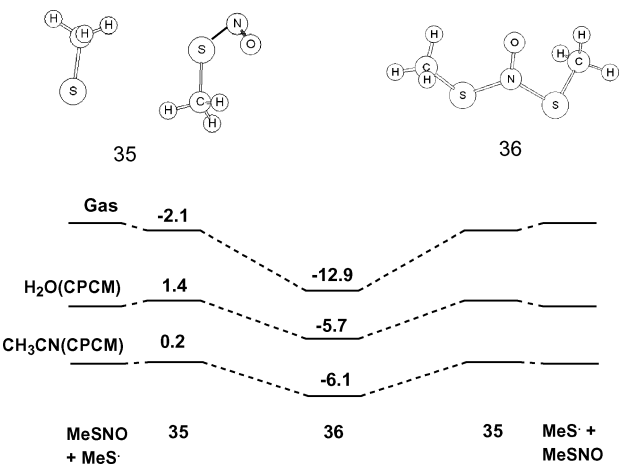

Fig. 10 Transnitrosation between MeS• and MeSNO in the gas phase and in solution (water and CH_3CN using the CPCM model).

Thiolation

In vivo, thiolation occurs in competition with transnitrosation and may be associated with the release of NO which contributes the bioactivity of *S*-nitrosothiols. It has been established that *S*-nitrosothiols are sensitive to both photolytic[61,62] and transition metal ion-dependent decomposition[39] but are stable in the presence of transition metal ion chelators in the dark. A recent theoretical study demonstrates that Cu^I complexation promoted degradation of *S*-nitrosothiols is the result of S–NO bond weakening by the formation of a Cu^I–RSNO complexes.[63] In the present work, we limit our study to non-metal-catalyzed mechanisms. Similar to transnitrosation, thiolation can be also considered *via* neutral MeSH, anion MeS^-, or radical MeS•.

MeSH + MeXNO Thiolation reactions. The uncatalyzed thiolation reaction between MeSH and MeSNO has a prohibitively high barrier in the gas phase. The optimized geometries and energetics are shown in Fig. 13 and 14. Without a molecule of water, the barrier is *ca.* 12 kcal mol^{-1} higher than transnitrosation in gas phase. Inclusion of an explicit water lowers the difference to 9 kcal mol^{-1} and adding bulk solvent reduces the difference to 3–5 kcal mol^{-1}.

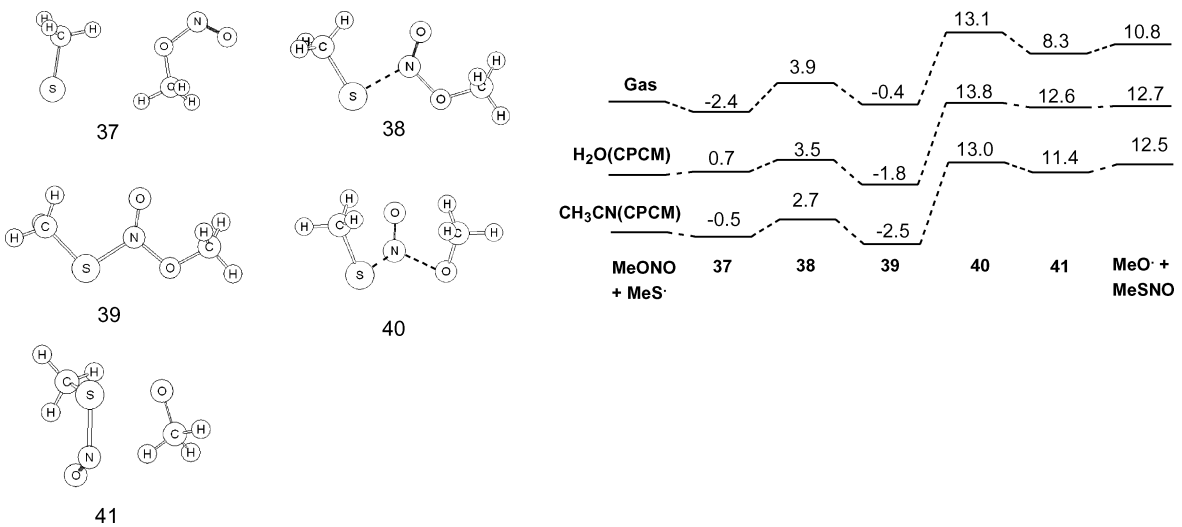

Fig. 11 Transnitrosation between MeS• and MeONO in the gas phase and in solution (water and CH_3CN using the CPCM model).

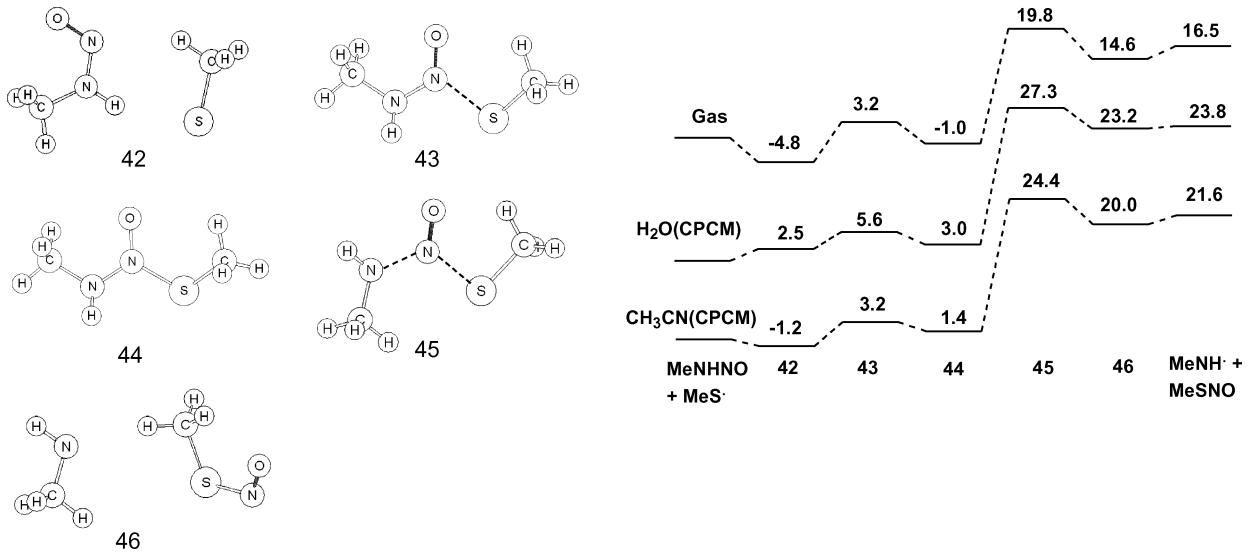

Fig. 12 Transnitrosation between MeS• and MeNHNO in the gas phase and in solution (water and CH_3CN using the CPCM model).

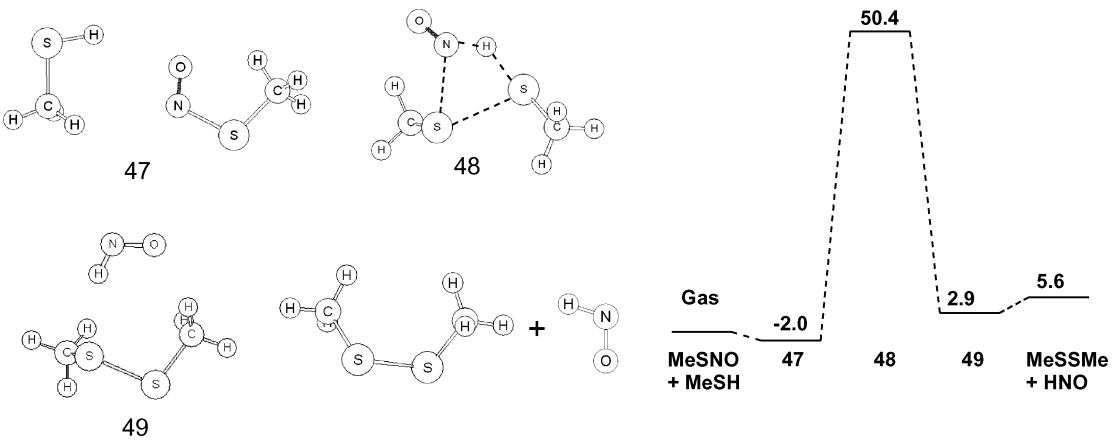

Fig. 13 Thiolation reaction between MeSH and MeSNO in the gas phase at the CBS-QB3 level of theory.

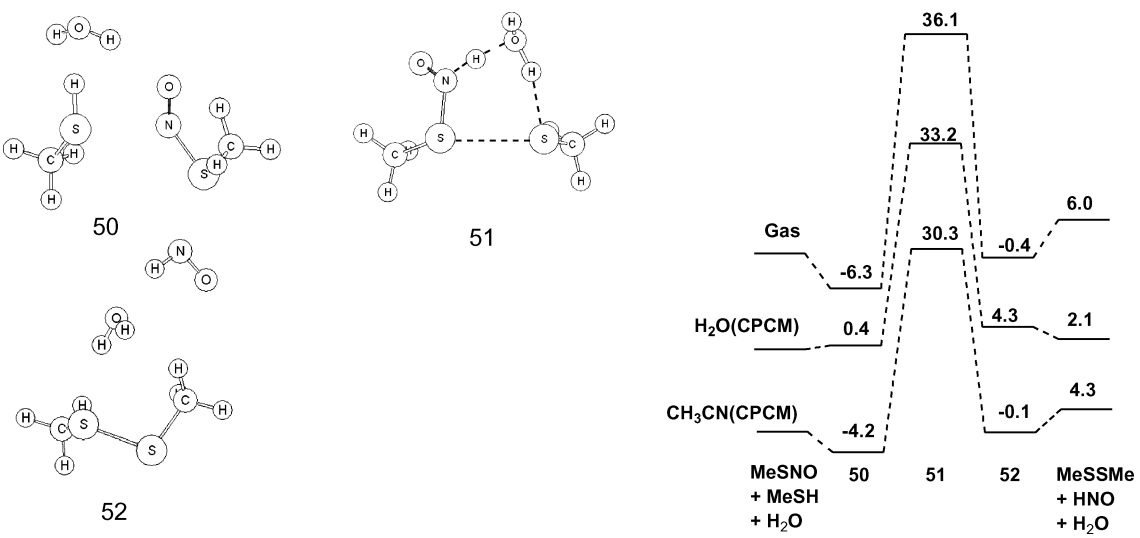

Fig. 14 Thiolation reaction between MeSH and MeSNO with one molecule of water assisting in the gas phase and in solution (water and CH_3CN using the CPCM model).

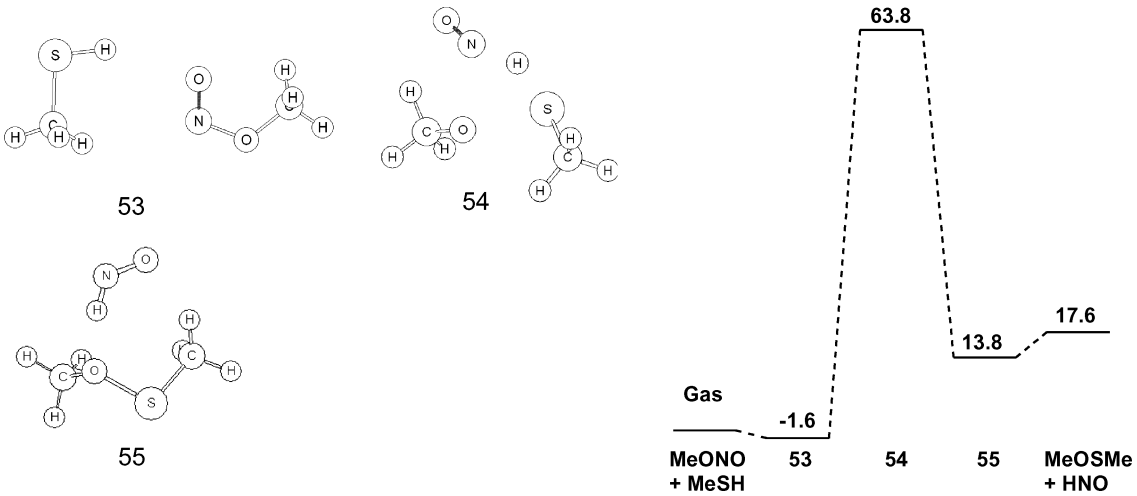

Fig. 15 Thiolation reaction between MeSH and MeONO in the gas phase at the CBS-QB3 level of theory.

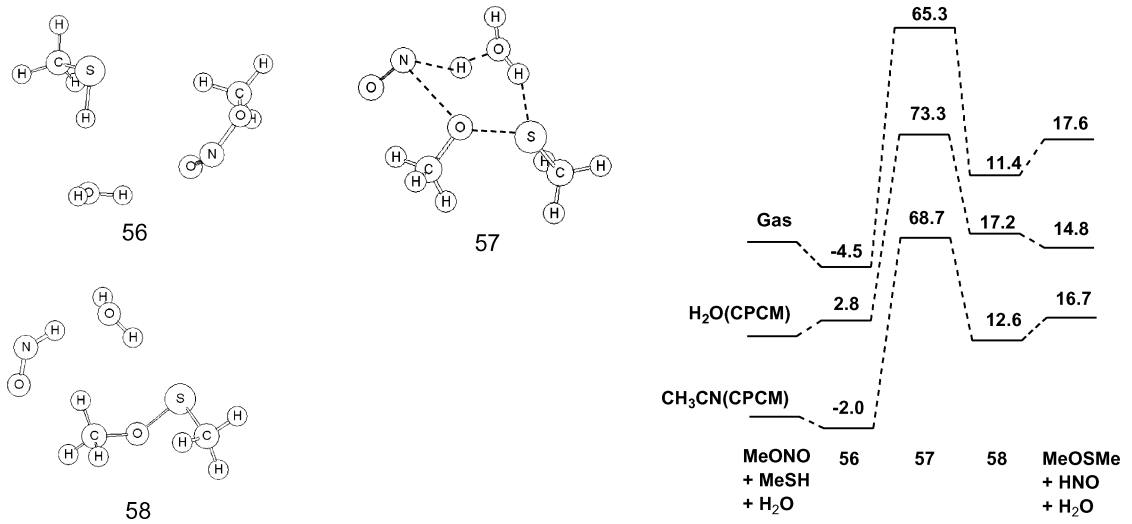

Fig. 16 Thiolation reaction between MeSH and MeONO with one molecule of water assisting in the gas phase and in solution (water and CH_3CN using the CPCM model).

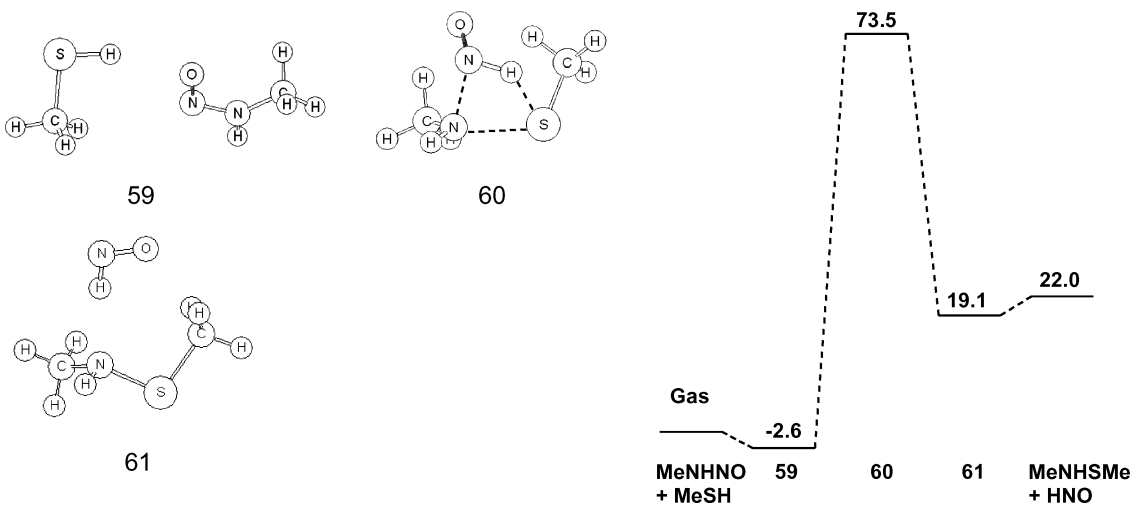

Fig. 17 Thiolation reaction between MeSH and MeNHNO in the gas phase at the CBS-QB3 level of theory.

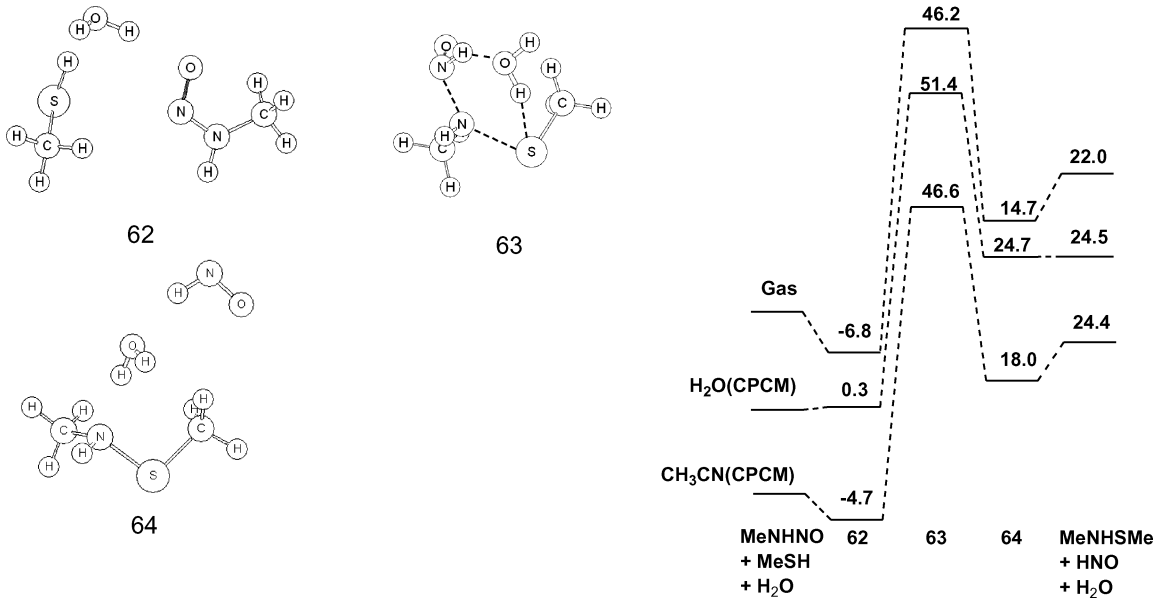

Fig. 18 Thiolation reaction between MeSH and MeNHNO with one molecule of water assisting in the gas phase and in solution (water and CH_3CN using the CPCM model).

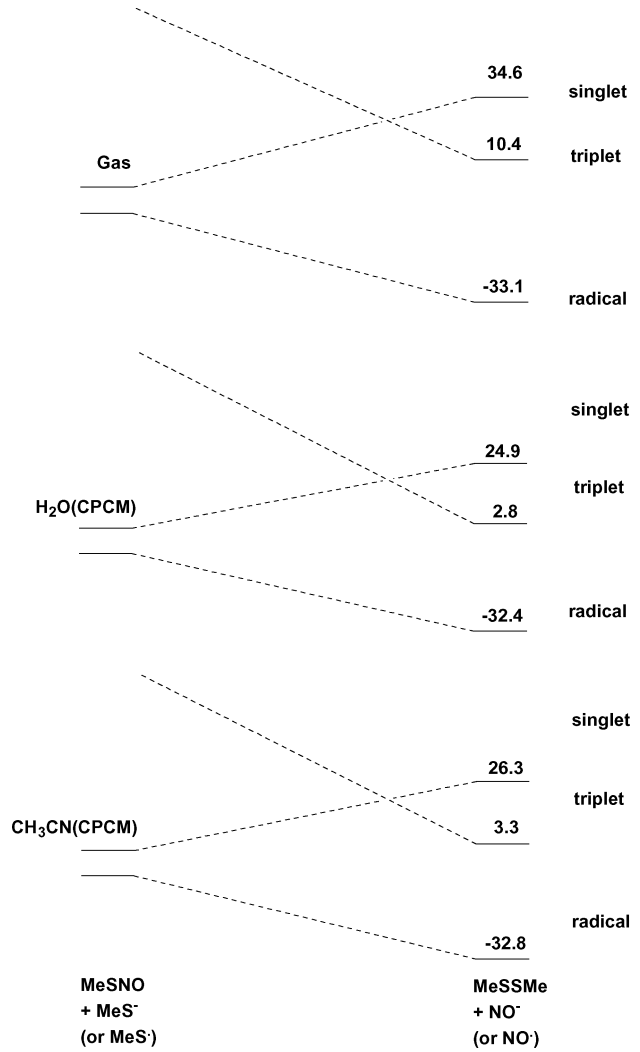

Fig. 19 Thiolation reaction between MeS^-, $MeS^\bullet$ and MeSNO in the gas phase and in solution (water and CH_3CN using the CPCM model).

The uncatalyzed thiolation reactions of MeSH with MeONO and MeNHNO have even high barriers than with MeSNO. Addition of an explicit water lowers the barriers for MeNHNO but not for MeONO. Bulk solvent does not change the barrier heights or reaction energies significantly (Fig. 15–18).

MeS^- + MeXNO Thiolation reactions. Thiolation *via* MeS^- is complicated by the fact that the singlet NO^- is higher in energy than the ground state triplet NO^- (~17 kcal mol^{-1} experimentally,[64,65] 24.2 kcal mol^{-1} at CBS-QB3). The crossing from singlet to triplet surface must occur somewhere along the reaction path. For the present study, it is sufficient to calculate the stationary points along reaction path on singlet and triplet surface separately. The optimized structures and energies at CBS-QB3 level of theory are shown in Fig. 19, 20, 21. On a singlet surface, all three of these reactions are significantly endothermic (energies of the isolated products relative to isolated reactants are 34.6, 46.6, and 51.0 kcal mol^{-1} for MeXNO, X = S, O, and NH, respectively), and consequently the barriers are also very high. For X = O and NH (Fig. 20 and 21), the transition states are 40.3 and 72.9 kcal mol^{-1} above the corresponding reactant complex, respectively. For X = S, a relaxed scan along the reaction path shows that transition state does not exist, which means that MeSSMe + NO^-(singlet) react without a barrier producing MeS^- + MeSNO. On a triplet surface, a relaxed scan indicates that MeS^- + MeSNO also does not have transition state. However, the triplet MeSSMe + NO^- product is 24.2 kcal mol^{-1} more stable than the singlet case. For X = O and NH, both in the gas phase and in solvent, the singlet and triplet surfaces cross just before or just after the transition states (Fig. 16 and 17). For X = O (Fig. 20) the transition state is 1.0 kcal mol^{-1} higher than the product complex at the B3LYP/6-311G(2d,d,p) level of theory, but additional electron correlation effects stabilize the transition structure more than the product complex. In the gas phase the reactions are *ca.* 20 kcal mol^{-1} more endothermic than for thiolation *via* thiol. In solution, the barriers for thiolation *via* thiolate are lower than *via* neutral

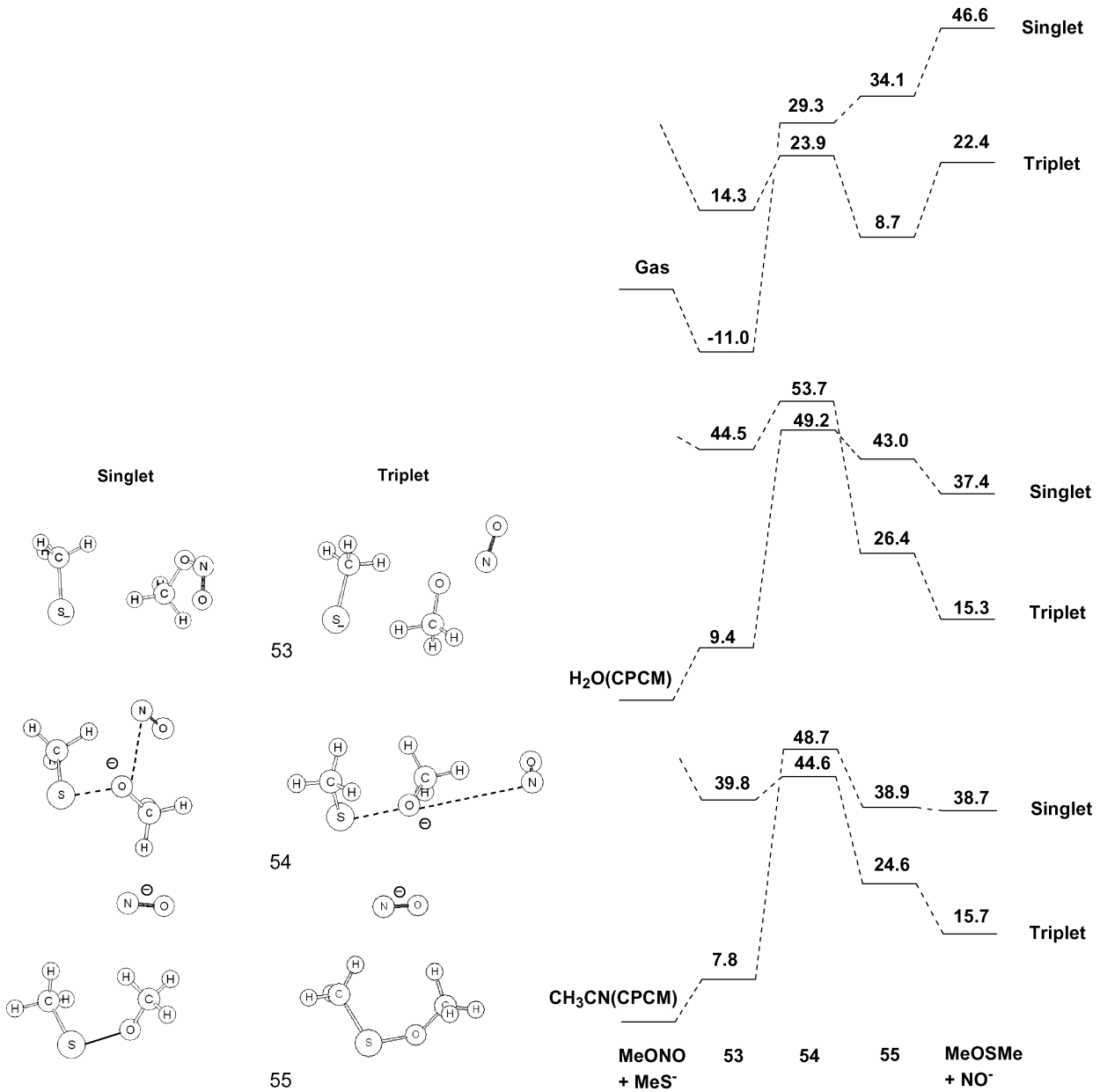

Fig. 20 Thiolation reaction between MeS^- and MeONO in the gas phase and in solution (water and CH_3CN using the CPCM model).

MeSH. The calculations for X = S suggest that solvation by water or acetonitrile may lower the energetics by *ca.* 10 kcal mol^{-1}, but the barriers are still very high. Computational and experimental work find the pK_a of NO^- is around 11.4 indicating that NO^- tends to be protonated in water around pH = 7 (However, HNO can dimerize readily to give hyponitrous acid which dehydrates to give N_2O).[66–68] Potentially, protonation could occur during the course of the reaction, leading to more reasonable energetics. In addition, the fast consumption of NO^- by reaction with oxygen or by other mechanisms may make this reaction thermodynamically more favorable.[28,69]

MeS• + MeXNO Thiolation reactions. Similar to transnitrosation, thiolation reactions with MeS• radical are facile (Fig. 19, 22 and 23). In particular, the reaction of MeS• with MeSNO to form a disulfide bond is barrierless. This is in accord with the autocatalytic effect of MeS• on the decomposition reaction of MeSNO at high MeSNO concentration and the increase of the decomposition rate at the increase concentration of MeSH.[17] Reactions are exothermic and the barriers are modest between MeS• and MeXNO (X = NH, O). Optimized structures and energetics are shown in Fig. 22 and 23. Solvation does not change the barrier height significantly and the reactions remain exothermic.

Summary

Molecular orbital calculations have been used to study transnitrosation and thiolation reactions. Depending on the reaction conditions, neutral MeSH, anion MeS^-, and radical MeS• are all possible reactive species. Barriers for neutral MeSH are very high, but can be lowered by the participation of an explicit water

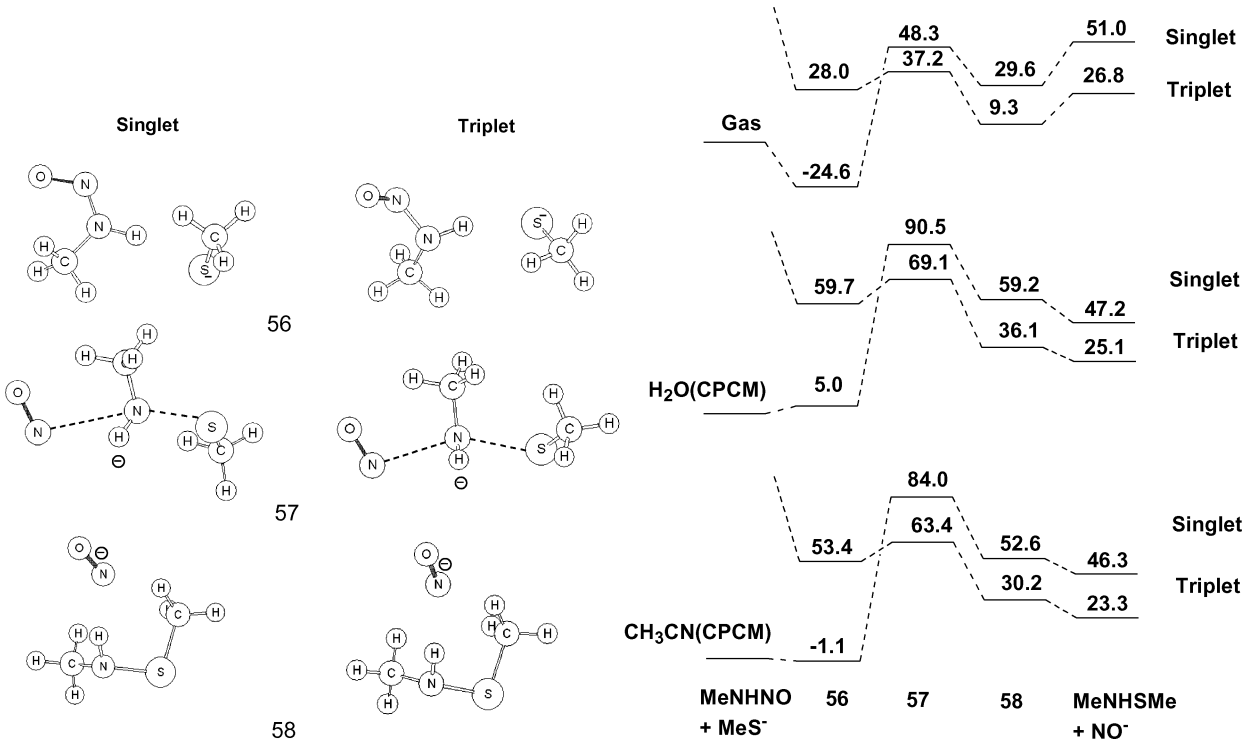

Fig. 21 Thiolation reaction between MeS^- and MeNHNO in the gas phase and in solution (water and CH_3CN using the CPCM model).

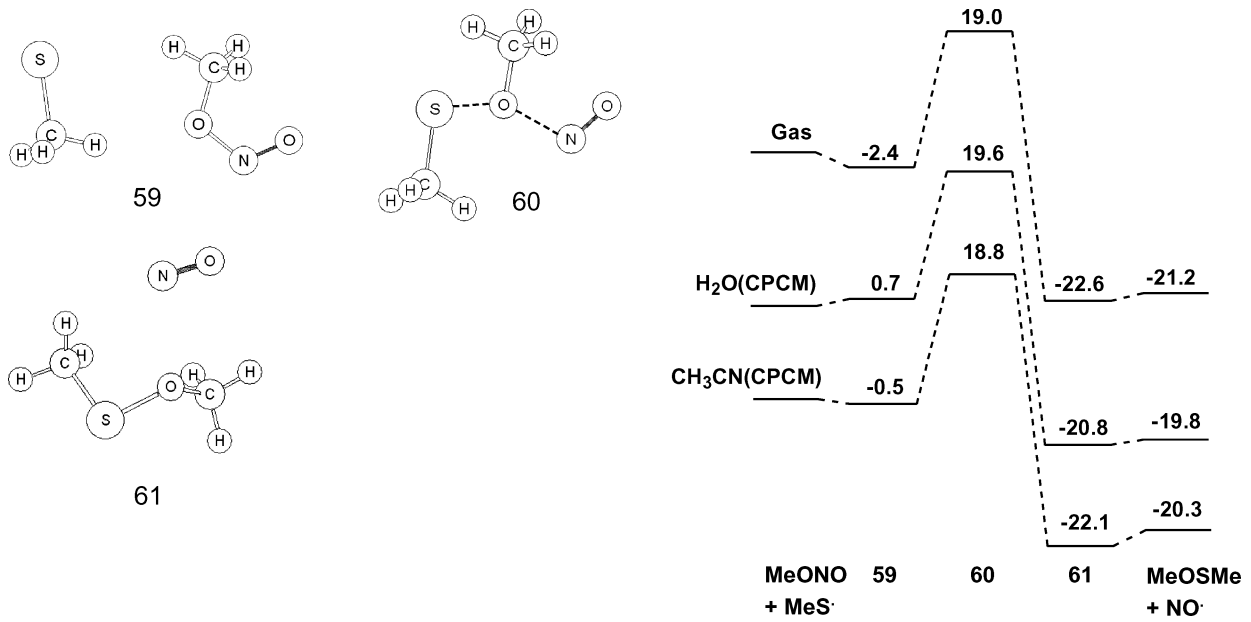

Fig. 22 Thiolation reaction between $MeS^•$ and MeONO in the gas phase and in solution (water and CH_3CN using the CPCM model).

molecule. Formation of the anion MeS^- brings down the barrier heights even more. In each case, $MeS^•$ radicals reacting with RXNO (X = S, O, NH) have the lowest barrier for both transnitrosation and thiolation reactions. The transnitrosation and thiolation reaction of RXNO are more favorable for X = S than for X = O or NH.

Recent experimental work shows that the *S*-transnitrosation reaction is facile and several orders of magnitude faster than *S*-thiolation reaction at pH 7 and room temperature.[21,41] Under these conditions, the neutral thiol is the majority species, but there is a significant amount of thiolate. Since the mechanisms for both reactions involving neutral thiol have very high barriers, the reactions involving thiolate species will contribute the most. The mechanisms involving radical $MeS^•$ have the lowest barriers; however, the concentration of radical $MeS^•$ in the solution is very low, making the contribution from the radical $MeS^•$ mechanism very small. At high temperatures or in the presence of transition metals or in light, the radical mechanism may contribute more.

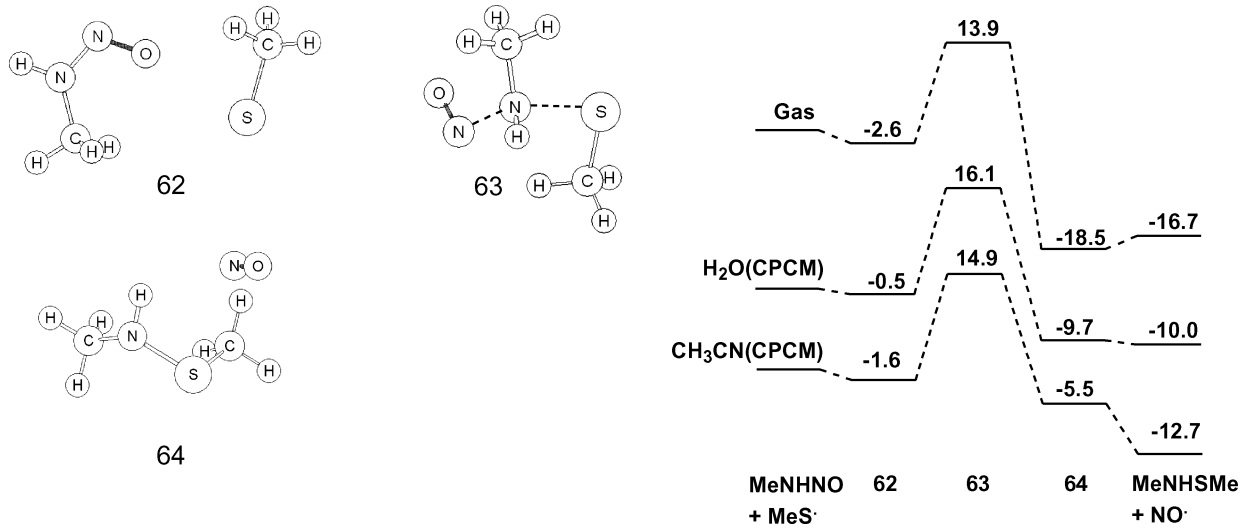

Fig. 23 Thiolation reaction between MeS• and MeNHNO in the gas phase and in solution (water and CH_3CN using the CPCM model).

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References

1 S. Moncada, R. M. J. Palmer and E. A. Higgs, Nitric-Oxide-Physiology, Pathophysiology, and Pharmacology, *Pharmacol. Rev.*, 1991, **43**, 109–142.

2 P. G. Wang, M. Xian, X. P. Tang, X. J. Wu, Z. Wen, T. W. Cai and A. J. Janczuk, Nitric oxide donors: Chemical activities and biological applications, *Chem. Rev.*, 2002, **102**, 1091–1134.

3 G. Gabor, N. Allon and H. H. Weetall, Are thiols the carrier of nitric oxide in biological systems? A kinetic model, *Microchem. J.*, 1997, **56**, 177–187.

4 A. Hirayama, A. A. Noronha-Dutra, M. P. Gordge, G. H. Neild and J. S. Hothersall, *S*-Nitrosothiols are stored by platelets and released during platelet-neutrophil interactions, *Nitric Oxide*, 1999, **3**, 95–104.

5 P. R. Myers, R. L. Minor, R. Guerra, J. N. Bates and D. G. Harrison, Vasorelaxant Properties of the Endothelium-Derived Relaxing Factor More Closely Resemble *S*-Nitrosocysteine Than Nitric-Oxide, *Nature*, 1990, **345**, 161–163.

6 K. L. Rossman, C. J. Der and J. Sondek, GEF means go: Turning on Rho GTPases with guanine nucleotide-exchange factors, *Nat. Rev. Mol. Cell Biol.*, 2005, **6**, 167–180.

7 W. Lijinsky, *Chemistry and Biology of N-nitroso Compounds*, Cambridge University Press, Cambridge, 1992.

8 P. N. Magee and J. M. Barnes, The production of malignant primary hepatic tumours in the rat by feeding dimethylnitrosamine, *Br. J. Cancer*, 1956, **10**, 114.

9 P. N. Magee and E. Farber, Toxic liver injury and carcinogenesis. Methylation of rat-liver nucleic acids by dimethylnitrosamine *in vivo*, *Biochem. J.*, 1962, **83**, 114.

10 P. N. Magee and T. Hultin, Toxic liver injury and carcinogenesis. Methylation of proteins of rat-liver slices by dimethylnitrosamine *in vitro*, *Biochem. J.*, 1962, **83**, 106.

11 L. T. Brunton, Use of nitrite in angina pectoris, *Lancet*, 1867, 97–98.

12 L. J. Ignarro, Nitric oxide: A unique endogenous signaling molecule in vascular biology (Nobel lecture), *Angew. Chem., Int. Ed.*, 1999, **38**, 1882–1892.

13 B. Cederqvist, M. G. Persson and L. E. Gustafsson, Direct demonstration of no formation *in vivo* from organic nitrites and nitrates, and correlation to effects on blood pressure and to in vitro effects, *Biochem. Pharmacol.*, 1994, **47**, 1047–1053.

14 C. Baciu and J. W. Gauld, An assessment of theoretical methods for the calculation of accurate structures and S–N bond dissociation energies of *S*-nitrosothiols (RSNOs), *J. Phys. Chem. A*, 2003, **107**, 9946–9952.

15 N. Bainbrigge, A. R. Butler and C. H. Gorbitz, The thermal stability of *S*-nitrosothiols: Experimental studies and *ab initio* calculations on model compounds, *J. Chem. Soc., Perkin Trans. 2*, 1997, 351–353.

16 A. R. Butler and P. Rhodes, Chemistry, analysis, and biological roles of *S*-nitrosothiols, *Anal. Biochem.*, 1997, **249**, 1–9.

17 M. G. de Oliveira, S. M. Shishido, A. B. Seabra and N. H. Morgon, Thermal stability of primary *S*-nitrosothiols: Roles of autocatalysis and structural effects on the rate of nitric oxide release, *J. Phys. Chem. A*, 2002, **106**, 8963–8970.

18 A. P. Dicks, P. H. Beloso and D. L. H. Williams, Decomposition of *S*-nitrosothiols: The effects of added thiols, *J. Chem. Soc., Perkin Trans. 2*, 1997, 1429–1434.

19 Y. Fu, Y. Mou, B. L. Lin, L. Liu and Q. X. Guo, Structures of the X–Y–NO molecules and homolytic dissociation energies of the Y–NO bonds (Y = C, N, O, S), *J. Phys. Chem. A*, 2002, **106**, 12386–12392.

20 L. Grossi and P. C. Montevecchi, A kinetic study of *S*-nitrosothiol decomposition, *Chem.–Eur. J.*, 2002, **8**, 380–387.

21 N. Hogg, The kinetics of *S*-transnitrosation—A reversible second-order reaction, *Anal. Biochem.*, 1999, **272**, 257–262.

22 D. R. Noble and D. L. H. Williams, Nitrosation products from *S*-nitrosothiols *via* preliminary nitric oxide formation, *J. Chem. Soc., Perkin Trans. 2*, 2002, 1834–1838.

23 R. J. Singh, N. Hogg, J. Joseph and B. Kalyanaraman, Mechanism of nitric oxide release from *S*-nitrosothiols, *J. Biol. Chem.*, 1996, **271**, 18596–18603.

24 S. P. Singh, J. S. Wishnok, M. Keshive, W. M. Deen and S. R. Tannenbaum, The chemistry of the *S*-nitrosoglutathione glutathione system, *Proc. Natl. Acad. Sci. U. S. A.*, 1996, **93**, 14428–14433.

25 J. M. Tullett, D. D. Rees, D. E. G. Shuker and A. Gescher, Lack of correlation between the observed stability and pharmacological properties of *S*-nitroso derivatives of glutathione and cysteine-related peptides, *Biochem. Pharmacol.*, 2001, **62**, 1239–1247.

26 K. Wang, Z. Wen, W. Zhang, M. Xian, J. P. Cheng and P. G. Wang, Equilibrium and kinetics studies of transnitrosation between *S*-nitrosothiols and thiols, *Bioorg. Med. Chem. Lett.*, 2001, **11**, 433–436.

27 D. L. H. Williams, The chemistry of *S*-nitrosothiols, *Acc. Chem. Res.*, 1999, **32**, 869–876.

28 P. S. Y. Wong, J. Hyun, J. M. Fukuto, F. N. Shirota, E. G. DeMaster, D. W. Shoeman and H. T. Nagasawa, Reaction between *S*-nitrosothiols and thiols: Generation of nitroxyl (HNO) and subsequent chemistry, *Biochemistry*, 1998, **37**, 5362–5371.

29 D. J. Barnett, J. McAninly and D. L. H. Williams, Transnitrosation between Nitrosothiols and Thiols, *J. Chem. Soc., Perkin Trans. 2*, 1994, 1131–1133.
30 D. J. Barnett, A. Rios and D. L. H. Williams, NO-Group Transfer (Transnitrosation) between *S*-Nitrosothiols and Thiols. Part 2, *J. Chem. Soc., Perkin Trans. 2*, 1995, 1279–1282.
31 D. J. Meyer, H. Kramer and B. Ketterer, Human glutathione transferase catalysis of the formation of *S*-nitrosoglutathione from organic nitrites plus glutathione, *FEBS Lett.*, 1994, **351**, 427–428.
32 A. P. Munro and D. L. H. Williams, Reactivity of sulfur nucleophiles towards *S*-nitrosothiols, *J. Chem. Soc., Perkin Trans. 2*, 2000, 1794–1797.
33 R. Rossi, L. Lusini, F. Giannerini, D. Giustarini, G. Lungarella and P. Di Simplicio, A method to study kinetics of transnitrosation with nitrosoglutathione: Reactions with hemoglobin and other thiols, *Anal. Biochem.*, 1997, **254**, 215–220.
34 K. N. Houk, B. N. Hietbrink, M. D. Bartberger, P. R. McCarren, B. Y. Choi, R. D. Voyksner, J. S. Stamler and E. J. Toone, Nitroxyl disulfides, novel intermediates in transnitrosation reactions, *J. Am. Chem. Soc.*, 2003, **125**, 6972–6976.
35 L. L. Perissinotti, A. G. Turjanski, D. A. Estrin and F. Doctorovich, Transnitrosation of nitrosothiols: Characterization of an elusive intermediate, *J. Am. Chem. Soc.*, 2005, **127**, 486–487.
36 A. H. Al-Mustafa, H. Sies and W. Stahl, Sulfur-to-nitrogen transnitrosation: transfer of nitric oxide from *S*-nitroso compounds to diethanolamine and the role of intermediate sulfur-to-sulfur transnitrosation, *Toxicology*, 2001, **163**, 127–136.
37 M. D. Bartberger, J. D. Mannion, S. C. Powell, J. S. Stamler, K. N. Houk and E. J. Toone, S–N dissociation energies of *S*-nitrosothiols: On the origins of nitrosothiol decomposition rates, *J. Am. Chem. Soc.*, 2001, **123**, 8868–8869.
38 J. M. Lu, J. M. Wittbrodt, K. Wang, Z. Wen, H. B. Schlegel, P. G. Wang and J. P. Cheng, NO affinities of *S*-nitrosothiols: A direct experimental and computational investigation of RS–NO bond dissociation energies, *J. Am. Chem. Soc.*, 2001, **123**, 2903–2904.
39 J. McAninly, D. L. H. Williams, S. C. Askew, A. R. Butler and C. Russell, Metal-IonCatalysis in Nitrosothiol (RSNO) Decomposition, *J. Chem. Soc., Chem. Commun.*, 1993, 1758–1759.
40 A. P. Dicks, E. Li, A. P. Munro, H. R. Swift and D. L. H. Williams, The reaction of *S*-nitrosothiols with thiols at high thiol concentration, *Can. J. Chem.*, 1998, **76**, 789–794.
41 E. A. Konorev, B. Kalyanaraman and N. Hogg, Modification of creatine kinase by *S*-nitrosothiols: *S*-nitrosation *vs. S*-thiolation, *Free Radical Biol. Med.*, 2000, **28**, 1671–1678.
42 M. J. Frisch, G. W. Trucks, H. B. Schlegel, G. E. Scuseria, M. A. Robb, J. R. Cheeseman, J. A. Montgomery, T. Vreven, K. N. Kudin, J. C. Burant, J. M. Millam, S. Iyengar, J. Tomasi, V. Barone, B. Mennucci, M. Cossi, G. Scalmani, N. Rega, G. A. Petersson, H. Nakatsuji, M. Hada, M. Ehara, K. Toyota, R. Fukuda, J. Hasegawa, M. Ishida, T. Nakajima, Y. Honda, O. Kitao, H. Nakai, M. Klene, X. Li, J. E. Knox, H. P. Hratchian, J. B. Cross, C. Adamo, J. Jaramillo, R. Gomperts, R. E. Stratmann, O. Yazyev, A. J. Austin, R. Cammi, C. Pomelli, J. Ochterski, P. Y. Ayala, K. Morokuma, G. A. Voth, P. Salvador, J. J. Dannenberg, V. G. Zakrewski, S. Dapprich, A. D. Daniels, M. C. Strain, Ö Farkas, D. K. Malick, A. D. Rabuck, K. Raghavachari, J. B. Foresman, J. V. Ortiz, Q. Cui, A. G. Baboul, S. Clifford, J. Cioslowski, B. B. Stefanov, G. Liu, A. Liashenko, P. Piskorz, I. Komaromi, R. L. Martin, D. J. Fox, T. Keith, M. A. Al-Laham, C. Y. Peng, A. Nanayakkara, M. Challacombe, P. M. W. Gill, B. Johnson, W. Chen, M. W. Wong, J. L. Andres, C. Gonzalez, E. S. Replogle and J. A. Pople, *Gaussian 03*, Development Version, 2003.
43 J. A. Montgomery, M. J. Frisch, J. W. Ochterski and G. A. Petersson, A complete basis set model chemistry. VI. Use of density functional geometries and frequencies, *J. Chem. Phys.*, 1999, **110**, 2822–2827.
44 J. A. Montgomery, M. J. Frisch, J. W. Ochterski and G. A. Petersson, A complete basis set model chemistry. VII. Use of the minimum population localization method, *J. Chem. Phys.*, 2000, **112**, 6532–6542.
45 A. D. Becke, Density-functional exchange-energy approximation with correct asymptotic behavior, *Phys. Rev. A: At., Mol., Opt. Phys.*, 1988, **38**, 3098–3100.
46 A. D. Becke, Density-Functional Thermochemistry. 3. The Role of Exact Exchange, *J. Chem. Phys.*, 1993, **98**, 5648–5652.
47 C. T. Lee, W. T. Yang and R. G. Parr, Development of the Colle–Salvetti Correlation-Energy Formula into a Functional of the Electron-Density, *Phys. Rev. B: Condens. Matter*, 1988, **37**, 785–789.
48 V. Barone and M. Cossi, Quantum calculation of molecular energies and energy gradients in solution by a conductor solvent model, *J. Phys. Chem. A*, 1998, **102**, 1995–2001.
49 R. Ditchfield, W. J. Hehre and J. A. Pople, Self-consistent molecular-orbital methods. IX. Extended Gaussian-type basis for molecular-orbital studies of organic molecules, *J. Chem. Phys.*, 1971, **54**, 724–728.
50 P. C. Hariharan and J. A. Pople, Influence of polarization functions on MO hydrogenation energies, *Theor. Chim. Acta*, 1973, **28**, 213–222.
51 S. Antonczak, M. F. Ruizlopez and J. L. Rivail, *Ab-Initio* Analysis of Water-Assisted Reaction-Mechanisms in Amide Hydrolysis, *J. Am. Chem. Soc.*, 1994, **116**, 3912–3921.
52 C. Clavero, M. Duran, A. Lledos, O. N. Ventura and J. Bertran, Theoretical-Study of the Addition of Hydrogen Halides to Olefins-a Comparison between $(HCl)_2$ and $(HF)_2$ Additions to Ethylene, *J. Comput. Chem.*, 1987, **8**, 481–488.
53 A. Lledos and J. Bertran, Lactam–Lactim Tautomeric Interconversion Mechanism of 2-Pyridone in Aqueous-Solution, *Tetrahedron Lett.*, 1981, **22**, 775–778.
54 A. Lledos, J. Bertran and O. N. Ventura, Water-Chain Intervention in the Ketonization of Vinyl Alcohol-an *Ab initio* Study, *Int. J. Quantum Chem.*, 1986, **30**, 467–477.
55 M. T. Nguyen and A. F. Hegarty, *Ab initio* Study of the Hydration of Ketenimine (CH_2=C=NH) by Water and Water Dimer, *J. Am. Chem. Soc.*, 1983, **105**, 3811–3815.
56 M. T. Nguyen and A. F. Hegarty, Molecular-Orbital Study on the Hydrolysis of Ketene by Water Dimer-Beta-Carbon *Vs* Oxygen Protonation, *J. Am. Chem. Soc.*, 1984, **106**, 1552–1557.
57 D. A. Palmer and R. Vaneldik, The Chemistry of Metal Carbonato and Carbon-Dioxide Complexes, *Chem. Rev.*, 1983, **83**, 651–731.
58 Y. Pocker and D. W. Bjorkquist, Stopped-flow studies of carbon dioxide hydration and bicarbonate dehydration in water and water-d_2. Acid–base and metal ion catalysis, *J. Am. Chem. Soc.*, 1977, **99**, 6537–6543.
59 O. N. Ventura, A. Lledos, R. Bonaccorsi, J. Bertran and J. Tomasi, Theoretical-Study of Reaction-Mechanisms for the Ketonization of Vinyl Alcohol in Gas-Phase and Aqueous-Solution, *Theor. Chim. Acta*, 1987, **72**, 175–195.
60 I. H. Williams, Theoretical Modeling of Specific Solvation Effects Upon Carbonyl Addition, *J. Am. Chem. Soc.*, 1987, **109**, 6299–6307.
61 R. J. Singh, N. Hogg, J. Joseph and B. Kalyanaraman, Photosensitized Decomposition of *S*-Nitrosothiols and 2-Methyl-2-Nitrosopropane Possible Use for Site-Directed Nitric-Oxide Production, *FEBS Lett.*, 1995, **360**, 47–51.
62 D. J. Sexton, A. Muruganandam, D. J. McKenney and B. Mutus, Visible-Light Photochemical Release of Nitric-Oxide from *S*-Nitrosoglutathione—Potential Photochemotherapeutic Applications, *Photochem. Photobiol.*, 1994, **59**, 463–467.
63 C. Toubin, D. Y. H. Yeung, A. M. English and G. H. Peslherbe, Theoretical evidence that Cu–I complexation promotes degradation of *S*-nitrosothiols, *J. Am. Chem. Soc.*, 2002, **124**, 14816–14817.
64 C. Szmytkowski and K. Maciag, Total Cross-Section For Electron-Impact On Nitrogen Monoxide, *J. Phys. B*, 1991, **24**, 4273–4279.
65 J. Tennyson and C. J. Noble, Low-Energy Electron-Scattering By The No Molecule, *J. Phys. B*, 1986, **19**, 4025–4033.
66 M. D. Bartberger, W. Liu, E. Ford, K. M. Miranda, C. Switzer, J. M. Fukuto, P. J. Farmer, D. A. Wink and K. N. Houk, The reduction potential of nitric oxide (NO) and its importance to NO biochemistry, *Proc. Natl. Acad. Sci. U. S. A.*, 2002, **99**, 10958–10963.
67 J. M. Fukuto, M. D. Bartberger, A. S. Dutton, N. Paolocci, D. A. Wink and K. N. Houk, The physiological chemistry and biological activity of nitroxyl (HNO): The neglected, misunderstood, and enigmatic nitrogen oxide, *Chem. Res. Toxicol.*, 2005, **18**, 790–801.
68 V. Shafirovich and S. V. Lymar, Nitroxyl and its anion in aqueous solutions: Spin states, protic equilibria, and reactivities toward oxygen and nitric oxide, *Proc. Natl. Acad. Sci. U. S. A.*, 2002, **99**, 7340–7345.
69 N. Hogg, R. J. Singh and B. Kalyanaraman, The role of glutathione in the transport and catabolism of nitric oxide, *FEBS Lett.*, 1996, **382**, 223–228.

