

カテコールアミン類の分子内水素結合における協同効果

石内俊一， 藤井正明

東工大・資源研

アドレナリンに代表されるカテコールアミン神経伝達物質（図1）は、これまでの研究の結果、気相中で特異的に少数のコンフォメーションしかとらないことを見いだしている[1,2]。この原因の一つとして、カテコールOH基とアミン鎖の双極子-双極子相互作用により、特定のコンフォメーションが強く安定化されていることが挙げられる。さらに、特異的に安定化されたコンフォメーションでは、アミン鎖における分子内水素結合が、双極子-双極子相互作用による安定化を受けない場合に比べて強くなることも見出した[1-3]。このメカニズムを明らかにするため、種々のカテコールアミン関連分子および水クラスターに対して2重共鳴レーザー分光法を適用し、その構造と水素結合強度を検討した。

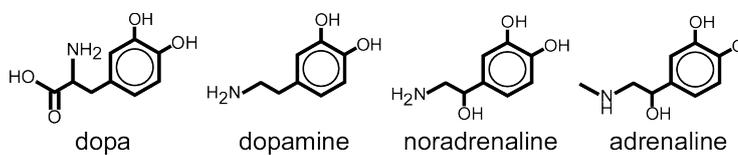


図1 カテコールアミン神経伝達物質

図2にドーパおよびその関連分子であるフェニルアラニン、チロシン及びその水クラスターの赤外スペクトルを示す。ここに取り上げた分子は、共通のアミン（アミノ酸）鎖をもち、ベンゼン環のフェノールOH置換基の数や環境が異なる。フェニルアラニン及びチロシンは気相中でそれぞれ6個[4]及び12個[3]のコンフォマーをもつが、図中の赤外スペクトルはそれぞれの最安定のコンフォメーションのもので、カルボキシ基のOH基がアミノ基の窒素原子との間で水素結合を形成している。チロシンのコンフォメーションは、解析の結果、アミノ酸鎖が6種類のコンフォメーションをとり、それぞれがフェノールOH基の2種類の配向をもつことによって合計12種類存在することが分かった[3]。一方、フェノールOH基の隣にもう1つOH基を有するドーパでは単独のコンフォメーションしか観測されない[1,2]。そのアミノ酸鎖の構造はチロシンの最安定構造のそれと一致することが赤外スペクトルの解析から明らかになった[2]。また、図に示したフェノール・水クラスターの赤外スペクトルは、チロシンの最安定コンフォマーがフェノールOH基と水の酸素原子の間に水素結合を形成した構造に対応する。従って、図2に示した分子のアミノ酸鎖のコンフォメーションは全て共通である。

ここで、分子内水素結合したカルボキシOH基の伸縮振動に注目すると、フェノールOH基とは水素結合の様な直接相互作用は存在しないにも関わらず、フェノールOH基の導入及びその水素結合形成に伴い低波数シフトしていることが分かる。即ち、これらの変化に伴いアミノ酸鎖の水素結合強度が増すことを示している。これは、フェノールOH基及びその周辺の水素結合形成により大きな部分双極子モーメントを形成し、これがアミノ酸鎖に分極を誘起した結果、アミノ酸鎖の水素結合強度が増したものと考えられる。実際に自然結合軌道(NBO)解析によって得られた部分電荷を基にアミノ酸鎖及びフェノールOH基の部分双極子モーメントを見積もると、アミノ酸鎖の部分双極子モーメントの大きさとアミノ酸鎖内の水素結合強度に正の相関があることが分かった（表1）。アミノ酸鎖の部分双極子モーメントの増大は、上に述べた様にフェノールOH

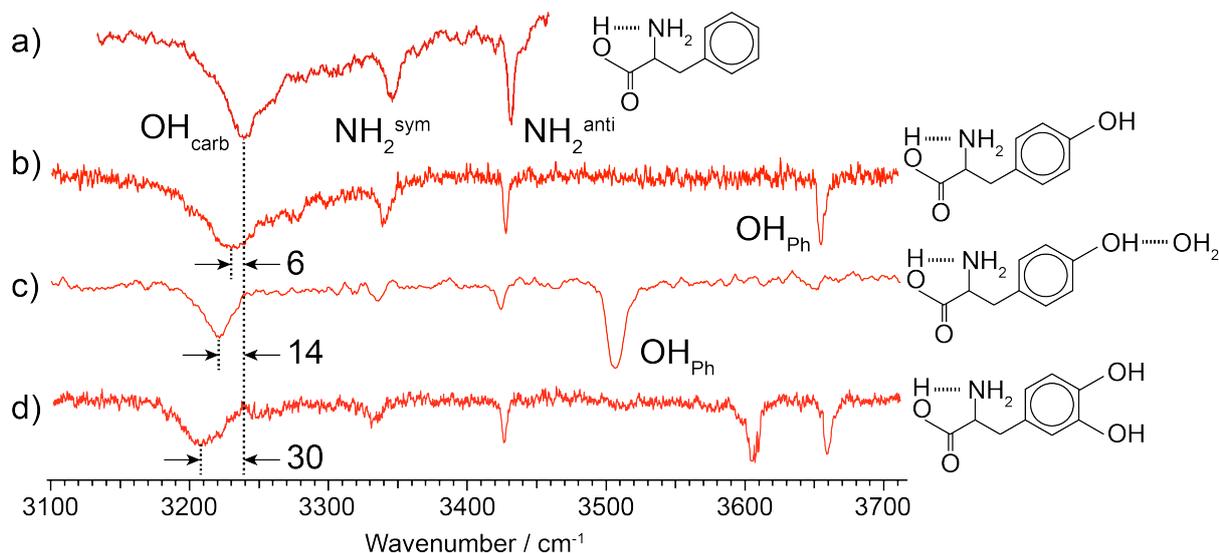


図2 a) フェニルアラニン[4]、b) チロシン[3]、c) チロシン・水クラスター及び d) ドーパ[2]の赤外スペクトル

基部位の双極子モーメントの増大によって誘起されたと考えられるが、表2を見るとフェノールOH基周辺の部分双極子モーメントはドーパよりチロシン・水クラスターの方が大きい値となっているにも関わらず、アミノ酸鎖の部分双極子モーメントはドーパの方が大きい。これは、それぞれのフェノールOH基周辺の部分双極子モーメントの方向の違いが原因であると考えられる。ドーパではフェノールOH基及びアミノ酸鎖の部分双極子モーメントがほぼ反平行になっており、そのためフェノールOH基の部分双極子モーメントがアミノ酸鎖に効率的に分極をもたらしたと考えられる。一方、チロシン・水クラスターでは水との水素結合を最適化する様にはたらき、その結果、アミノ酸鎖とフェノールOH基の部分双極子モーメントは反平行配置から大きくずれ、アミノ酸鎖の分極誘起が効率的ではなくなったと考えられる。

これらの結果は、分子内水素結合における協同効果を系統的に観測したものであると言える。このような協同効果は、分子の離れた部分の構造が別の部分の柔軟性に影響を及ぼすことを示しており、分子の柔軟性を考える上で重要な懸案事項の1つであることが示された。

表1 部分双極子モーメントの大きさ (mD)

	アミノ酸鎖	フェノール OH 基
チロシン(最安定コンフォマー)	1385	561
チロシン(最安定コンフォマー)・水クラスター	1389	1466
ドーパ	1402	1009

【参考文献】

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Gas phase spectroscopy of acetaminophen – Reassignments of conformers by electronic and IR spectra

W. Y. Sohn, S. Ishiuchi, M. Fujii

Chemical Resources Laboratory, Tokyo Institute of Technology

Acetaminophen (AAP, see Fig. 1), which is regarded as one of enzyme inhibitors, and has been considered a drug for antipyretic, analgesic and common cold, has been studied by gas phase spectroscopy. Beames and Hudson reported the first electronic spectrum of AAP in the gas phase by using laser desorption method to vaporize the sample.[1] This report showed that only one conformer exists in AAP. However, it had been doubted because it has OH group at *para*-position of aromatic ring with respect to amide group. As barrier

height of OH rotation is sufficiently high, the molecule can be easily frozen at each local minimum. It means that it should have at least two conformers along OH orientation in gas phase. As expected, it was demonstrated that AAP has two conformers in gas phase by S. J. Lee et al.[2] They distinguished two different conformers by UV-UV hole burning (HB) spectroscopy. The most intense band was assigned to *cis*-conformer, and a weak transition which is located 33 cm^{-1} low-frequency from the origin of the *cis*-conformer was re-assigned to *trans*-conformer from the previous assignment of a hot band of the methyl rotational level. Though it seems that the previous work solved the questions on the variety of conformers in AAP, the reported spectra enhance questions. The most serious problem is that the HB spectrum of the *cis*-isomer gives the transition at the origin of the *trans*-isomer. If their assignment is correct, no signal should appear at the transition of another species. Even though the overlap of transitions between two conformers is possible, it is very difficult for the transition at the origin of the *trans*-isomer to be shown in the HB spectrum of the *cis*-isomer because contribution of *trans*-isomer at the origin of *cis*-isomer should be small.

Thus, we motivated to measure the HB spectra of AAP independently with the advanced laser desorption molecular beam spectrometer. The thorough spectral cooling enables us to clearly observe vibronic structures. In present study, the HB spectroscopy shows co-existence of four independent species in the resonance enhanced multi photon ionization (REMPI) spectrum of AAP (Fig. 2).[3] The simplest interpretation for four species is that there is four isomers arose from OH orientation (*cis*(OH)- and *trans*(OH)-) and *cis* and *trans* isomers for amide bond (*cis*(amide)- and *trans*(amide)-; see Fig. 1). However, as depicted in Fig. 1, it is not reasonable because the *cis*(amide)-isomers locate more than 3 kcal/mol higher in energy than the *trans*(amide)-isomers. For this energy difference, comparable population in a supersonic jet cannot be expected.

To solve this puzzling problem, we considered internal rotational levels of methyl group (Fig. 3), and

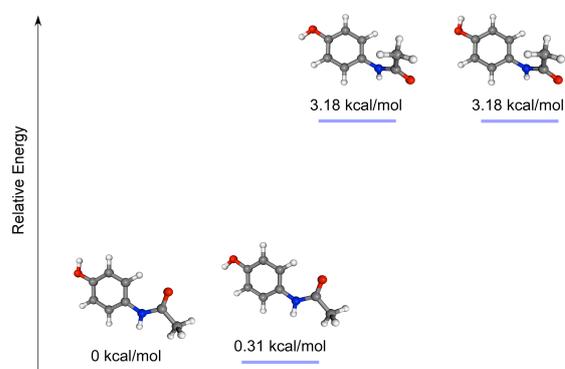


Fig. 1 Ground state optimized structures and relative energies of each conformer of AAP.

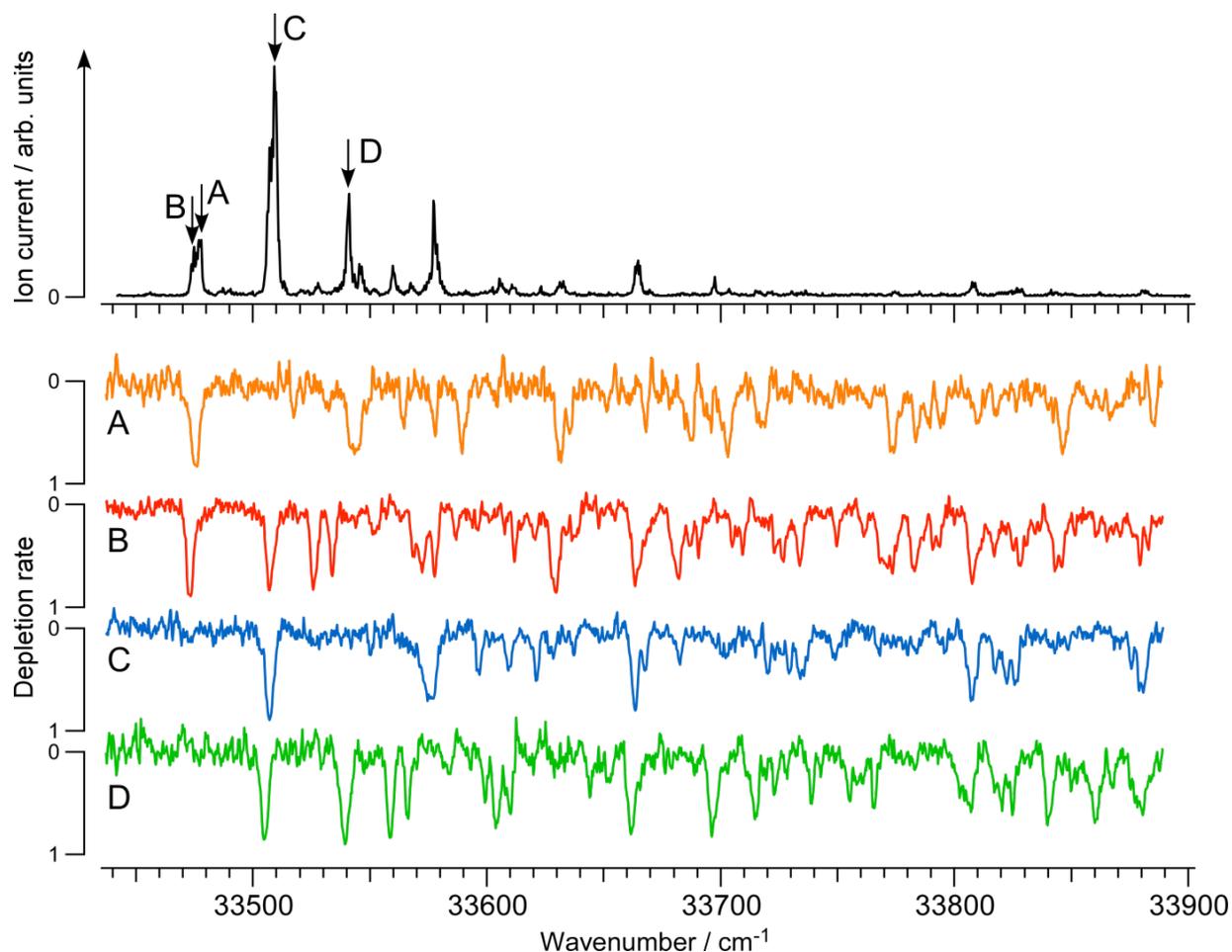


Fig. 2 REMPI spectrum (upper trace) and HB spectra (bottom trace) observed by probing A to D bands in REMPI spectrum.

could explain the 4 different HB spectra and their vibronic levels successfully.[3] It was concluded that A and B are derived from *trans*(OH) with *trans*(amide) isomer, but B arises from vibrationally excited level of methyl internal rotation ($1e$). Similarly, C and D come from *cis*(OH) with *trans*(amide), but D arises from $1e$ level. Thus, it is concluded that AAP has two rotational isomers along the OH orientation in the molecular beam.

References

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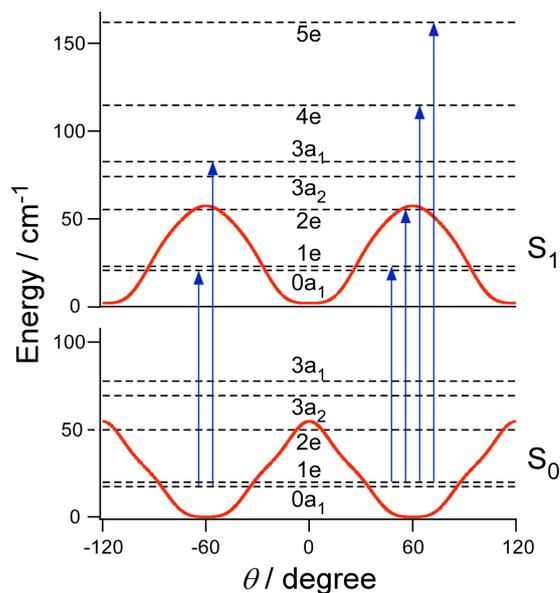


Fig. 3 The energy levels of methyl internal rotation with the potential curves.

Gas phase spectroscopy and anharmonic vibrational analysis of the 3-residue peptide Z-Pro-Leu-Gly-NH₂ by laser desorption supersonic jet technique

S. Ishiuchi, M. Fujii

Chemical Resources Laboratory, Tokyo Institute of Technology

The interplay between gas-phase infrared (IR) spectroscopy and accurate quantum chemical calculations has brought innovative progress to the study of the secondary structure of peptides. One of the main emphases of these studies is obviously to reveal the intrinsic structure of polypeptides, which assists us to understand the solvent effect on their structures

concerning folding and denaturalization. One of the main interests for studying peptides is the elucidation of local preferences and the competition among them. Different secondary structures, such as γ -folds, β -strands, β -turns and 3_{10} -helices, are characterized by different intramolecular hydrogen-bonding (H-bonding) networks of the types, C_5 , C_7 , C_{10} and C_{13} , where C_n indicates an H-bond network consisting of an n -membered ring, $N-H\cdots\pi$, and exhibit distinct spectroscopic signatures. The N-H stretching transition (Amide A band), which is highly sensitive to this interaction, shows a red shift upon the formation of an H-bond, and thus provides precise information about the H-bond network. In general, the extent of the red shift is greater with stronger interactions. Hence, the amide A band is expected to exhibit an increasing amount of red shifts for the following interaction: $N-H\cdots\pi < C_5 < C_{10} < C_7$. [1]

In this work, we applied the gas phase spectroscopy to Z-PLG-NH₂ (Z= benzyloxycarbonyl, P=Pro, L=Leu, G=Gly) (Fig. 1), which is a partial peptide of a neuro-peptide “oxytocin”. By analyzing the electronic spectrum of Z-PLG-NH₂, it was found that the peptide possesses almost single conformation in gas phase. So we measured IR spectrum of the main conformer at NH stretch region (Fig. 2). Five distinctly sharp and strong transitions are observed at 3319, 3341, 3366, 3447, and 3516 cm⁻¹. Only the N-H stretching frequencies of Z-PLG-NH₂ are expected to be observed in this spectral range, and all of the bands should be assigned to N-H stretching vibrations. However, Z-PLG-NH₂ has only four N-H oscillators, namely, Leu-NH, Gly-NH, and two of the terminal -NH₂ group, and thus the assignment is non-trivial. The observed five transitions in the N-H stretching frequency region are expected to be due to splitting by anharmonic coupling. The IR spectrum and its tentative assignments were reported in our previous paper.[2] The previous analysis mainly relied on analogous vibrational bands of related molecules, and no discussion was possible about the splitting of bands.

To establish a structural assignment, we performed anharmonic vibrational structure calculations by the second-order vibrational quasi-degenerate perturbation theory (VQDPT2) based on a vibrational self-consistent field (VSCF) zeroth-order description. The calculations were performed by using the SINDO program package developed by Dr. Yagi (who is now in Sugita group at Riken) [3]. Fig. 3 shows a simulated IR spectrum of conformer A obtained by VQDPT2. The calculated vibrational eigenstates are expressed as a linear combination of the VSCF configuration function. Thus, for the sake of

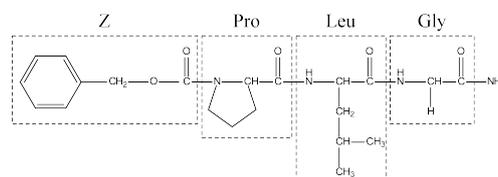


Fig. 1 Z-Pro-Leu-Gly-NH₂.

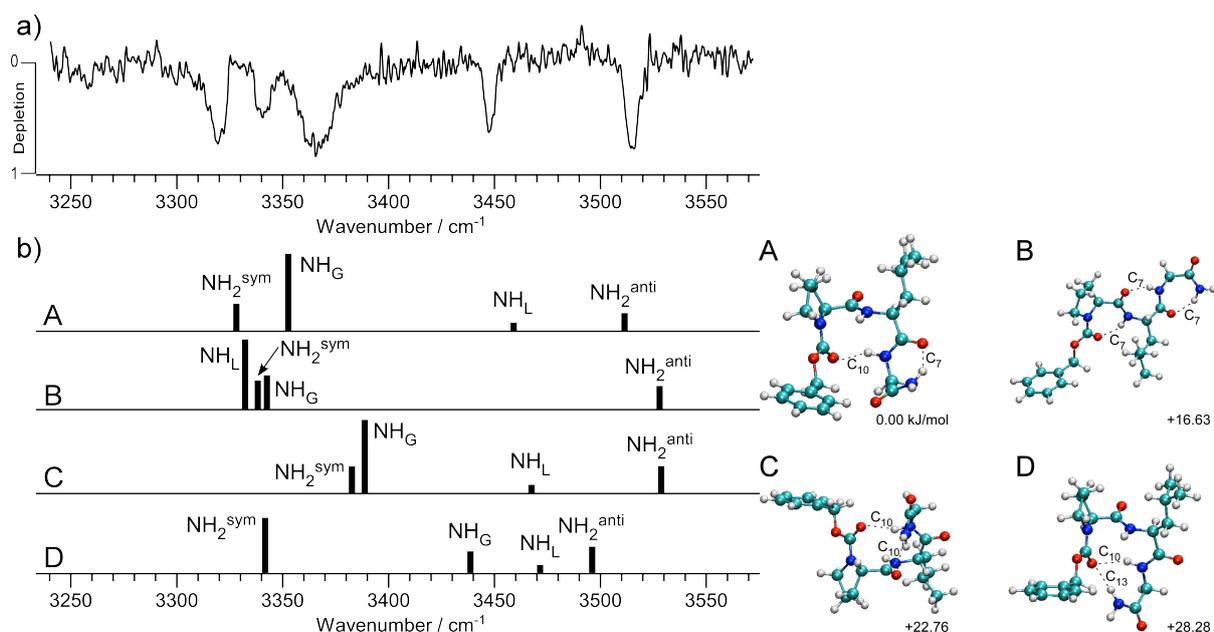


Fig. 2 IR spectrum of Z-PLG-NH2 (a) and simulated ones of 4 stable conformers A~D.

understanding, each transition is color-coded according to the weight of the symmetric and anti-symmetric stretching modes of the NH_2 , N-H stretching modes of Gly and Leu, and other modes. Anharmonic vibrational analysis reveals that the symmetric stretching mode of NH_2 contributes to the two experimentally observed transitions at 3319 and 3341 cm^{-1} , thus confirming the assignment of the splitting. The resonance partner of the anharmonic splitting is mainly a combination tone of NH_2 bending and C=O stretching of Gly. In addition, it is found that the origin of the broad band at 3366 cm^{-1} is the anharmonic coupling between the N-H stretching mode of Gly and the low-frequency modes in which the peptide main chain moves collectively. The strong coupling between them is plausible from a physical point of view, because the N-H bond of Gly forms a C_{10} H-bond network with the O=C bond of the Z-group and this hydrogen bond should be the mostly affected by the collective motion of the peptide main chain.

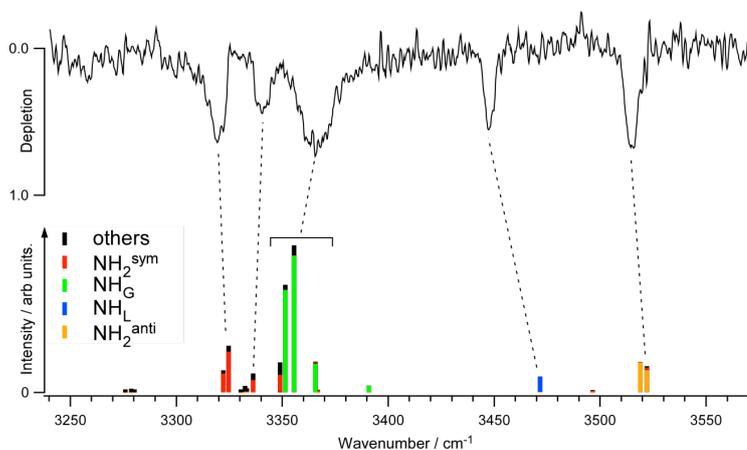


Fig. 3 Theoretical anharmonic IR spectrum of conformer A obtained by VQDPT2 method compared with the recorded IR dip spectrum.

References

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