Two Roads Converged in a Yellow Dye: Unusual Spectral Broadening in the Emission of Auramine-O Possibly Caused by Low-Friction Intramolecular Rotation

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Supporting Information

ABSTRACT: Steady-state and time-resolved optical techniques were employed to study the rather complex relaxation of excited states of Auramine-O (AuO) in several liquids at room temperature. We found three relaxation times in the decay of the pump–probe signals of the excited states of AuO. We focused our study on the short time decay, with a duration of within about 150–300 fs. We found that the temporal changes of the emission band of AuO could be divided into three behaviors, depending on the solvent characteristics. In dimethyl sulfoxide (DMSO), a hydrogen-bond-accepting solvent, AuO shows, at short times, a relatively broad emission band with small changes in its peak position and width as a function of time. In acetonitrile and in acetic acid, both hydrogen-bond-donating solvents, we found large changes in the band peak and width as a function of time. Dichloromethane is a solvent lacking strong solvent interactions, it is apolar and is neither a strong hydrogen-bond-donator nor a strong hydrogen-bond-acceptor. For AuO in dichloromethane we found an oscillation with a time constant of 200 fs in the emission band with small changes in its peak position and width as a function of time.

INTRODUCTION

In the late 1950s, two independent research efforts were initiated, progressing separately, eventually to converge in the study of the photoisomerization-process characteristics of Auramine-O (4,4’-(imidocarbonyl)-bis(N,N-dimethyl) aniline).

The first research effort followed a path motivated by an engineering point of view and focused on the harnessing of light for mechanical work. In the half century that followed Richard Feynman’s 1959 lecture, which carried the title “There’s plenty of room at the bottom,” molecular devices gained ever-growing attention in the new field dubbed nanotechnology. Among the Feynman-advocated molecular servomotors developed, the synthetic light-driven molecular rotor occupied a major role following the two pioneering works published on the matter by Kelly and co-workers and by Feringa and co-workers, in the 1999, volume 401, of the journal Nature.

Research following the second path was a natural extension to the field of histological methods and focused on the exploitation of molecular photophysical properties for the sensing and probing of a fluorescent dye’s microenvironment. Rapid development in the field of molecular biology demanded a broader toolkit of microscopic imaging techniques, requiring an exhaustive search for stains that possess unique function-trait qualities. In a publication dating back to 1956, Oster and Nishijima reported a study on the reduction in the fluorescence quantum yield of AuO in glycerol with rise in temperature. They concluded that it derives from the rotation rate of intramolecular chemical bonds. AuO, whether in mixture with rhodamine B or by itself, is a stain for acid-fast mycobacteria. Following up on the work by Oster and Nishijima, its photophysics and photochemistry in several solvents have been extensively studied.

Haidakker and Theodorakis made a connection between the two fields, pointing out the microenvironmental sensing, rather than manipulation, capabilities of molecular rotors. Stsiapura and co-workers introduced Thioflavin-T (ThT), a dye used for the detection of amyloid-fibril formation, into the molecular-rotor conversation, thus strengthening the connection between the two gemini fields. These works maintained that twisted intramolecular charge transfer (TICT) between the two sides of a chemical bond was the very essence of a molecular rotor. The TICT process and its effect on the molecular-fluorescence quantum yield and lifetime have been the aim of such theoretical models as the Forster–Hoffman model, the Bagchi–Fleming–Oxtoby model, and Agmon’s inhomogeneous model for p-HBDI, and the chromophore of the green fluorescent protein (GFP) is based on the Agmon–Hopfield model. More recently, Olsen and McKenzie also offered explanations for the effects of a dark
state and TICT in the relaxation processes of p-HBDI, stressing the role played by conical intersections.22–25 Glasbeek and co-workers26,27 developed previous models by Barbara, Fonseca, and co-workers28,29 to establish a model formed specifically for AuO. In a later iteration, Meech and co-workers30–33 added time-dependent friction34,35 to the model. Following employment of the Glasbeek model for AuO to model the nonradiative-decay dynamics of ThT36 and of a subsequent comparison work on the fluorescence temperature dependence of the two molecules,37 the convergence of the two roads was finally complete. We owe this realization to the insight obtained from works by, and correspondence with, Prof. Stephen R. Meech.38

The shared theme in this family of molecules, accounting for the service of whichever of the above-mentioned applications sought, is the existence of a different preferred conformation of a chemical bond in the ground and the excited states of such molecules. While decaying to its ground state, the fluorescent dye would decay by a tunable rate, which depends on the viscosity, thus allowing the control or recognition of the properties of the surrounding solvent.26,36 In the case of AuO, a controversy still remains as to whether or not there exists a barrier in the excited-state potential-energy curve.39,40

Another topic of research that is very relevant to this paper is the time scale of photoisomerization. Tahara and co-workers used a combined spectroscopic and computational approach, utilizing transient impulsive Raman spectra together with high-level quantum-chemical calculation, to study photoisomerization from cis- to trans-stilbene.41 In addition, there are two studies that, in our view, are of particular significance for the questions raised here. The first was made in 1999 by Sawada and co-workers.42 In their study, the authors employed the ultrafast lensing effect (ULE) and found a short relaxation time component for AuO in low-viscosity solvents, which they ascribed to friction arising from hydrodynamic friction rather than from polarity/dielectric relaxation. The second was a study conducted under much more extreme conditions. In 2006, in a state-of-the-art experiment by Moskun and co-workers,33 the linear-response theory was challenged, and its refutation was demonstrated for a CN rotor.

In the current study, we used steady-state absorption and emission techniques as well as time-resolved emission and pump–probe techniques in order to study the spectroscopic properties of AuO. AuO has four possible intramolecular rotation axes. These are the two identical N,N-dimethylamino groups of each of the two dimethylamine moieties around the central bridge. In the current study we focused our attention on the ultrashort events occurring within the time window of the first 300 fs after the electronic excitation of AuO by a short (∼100 fs) laser pulse. We found that within this short time window, in certain solvents that lack hydrogen-bond-accepting capabilities, the emission band of AuO shifts linearly with time to the red by about 1400 cm⁻¹. The bandwidth broadens linearly with time also by ∼1400 cm⁻¹, and the asymmetry of the emission band increases. We found that the time-resolved emission of AuO in dichloromethane exhibits coherent oscillation with a time period of 200 fs. We attribute the short time component of the transient spectra to the rotation of the dimethylamino groups of the aminilens of AuO. This rotation arises from the transition from its ground-state distribution at about 30° to the excited state also at a larger angle (90°). In nonviscous solvents that lack hydrogen bonding, the rotational motion of the dimethylamino group is underdamped, and this results in the appearance of the oscillation in the time-resolved emission signal.

## EXPERIMENTAL SECTION

Auramine-O was purchased from Sigma-Aldrich. All measurements were carried out with fresh solutions of AuO at the desired concentration and in the chosen solvent. Acetonitrile, trifluoroethanol, and other chemicals used in this study were of analytical grade and were purchased from Aldrich and Merck.

Multichannel transient-absorption measurements were conducted with the use of a pump–probe system (Helios, Ultrafast Systems) in a cuvette with a 2 mm optical path. The data were analyzed with commercial software (Surface Xplorer, Ultrafast Systems). The samples were pumped at a frequency of 500 Hz with 80 fs pulses generated by an optical parametric amplifier (TOPAS, Light Conversion) tuned to 415 nm. To avoid degradation, the samples were continuously stirred inside the cuvette. The temporal chirp of the white-light continuum probe was measured in D₂O under identical experimental conditions, and the data were corrected for temporal chirp with the use of a built-in algorithm.

The fluorescence up-conversion technique was employed in this study to measure the time-resolved emission of curcumin at room temperature. The laser used for the fluorescence up-conversion was a cavity-dumped Ti:sapphire femtosecond laser (, Coherent), which provides short, 150 fs, pulses at about 800 nm. The cavity dumper operated with a relatively low repetition rate of 800 kHz. The up-conversion system (FOG-100, CDP, Russia) operated at 800 kHz. The samples were excited by pulses of ∼8 mW on average at the SHG frequency. The time response of the up-conversion system is evaluated by measuring the relatively strong Raman Stokes line of water shifted by 3600 cm⁻¹. It was found that the fwhm of the signal is 300 fs. Samples were placed in a rotating optical cell to avoid degradation.

The steady-state emission and absorption spectra were recorded by a Horiba Jobin Yvon FluoroMax-3 spectrophotometer and a Cary 5000 spectrometer.

## RESULTS

### Steady-State Absorption and Emission Measurements

Figure 1 shows the steady-state excitation and emission spectra of AuO in neat 1-propanol. The fluorescence quantum yield of AuO in linear alcohols (methanol to 1-pentanol) depends on the solvent viscosity. The average emission decay time τₑ is ultrashort, where τₑ = \( \frac{1}{I(t)} \) and I(t) is the normalized fluorescence intensity at time t. In ethanol, the viscosity at room temperature is 1.1 cP, and τₑ measured at 520 nm is only ∼2 ps.

**Pump–Probe Transient Spectroscopy.** Figure 2 shows the transient pump–probe spectra of AuO in neat acetonitrile.

There are four panels in the figure showing the spectra at different times. Figure 2a shows the transient pump–probe spectra at short times, up to 0.3 ps. The sample is excited at 500 Hz by a short laser pulse of 80 fs at 410 nm. The sample is probed by a supercontinuum in the spectral range of 440–800 nm. At these short times, the main part of the signal is negative, which we assign to the ground-state absorption bleach at λ < 470 nm. At longer wavelengths, the negative signal is assigned to the excited-state emission. The emission intensity increases up to about 0.23 ps, which is longer than the short finite response time—120 fs—of the pump–probe optical setup.
interesting point to notice is that the peak position and bandwidth of the emission spectrum are time-dependent. The spectrum shifts to the red by about 1400 cm$^{-1}$ within 0.2 ps, and the bandwidth increases from about 1600 cm$^{-1}$ to about 3000 cm$^{-1}$. A relatively small absorption in the near IR appears at the same rate as that of the emission. It is also noticed that the bandwidth and the spectral-peak position of this absorption depend on time. At short times this peak absorption is at about 580 nm with a rather small bandwidth, and at longer times the peak position is at $\sim$680 nm with a large bandwidth.

Figure 2b shows the transient pump–probe spectra in the intermediate time window from 0.3 to 0.9 ps. In this time window, the main fluorescence band decays with an average decay time of about $\sim$400 fs. The emission band peak shifts further to the red by about 1300 cm$^{-1}$ from 520 nm to about 555 nm.

Figure 2c shows the third time window of the pump–probe time-resolved spectra of AuO in acetonitrile from $t = 0.9$ to 1.8 ps. The fluorescence intensity at 1.8 ps drops to zero. The emission band peak is at about 555 nm and is asymmetric with a weak near-IR tail. A rather narrow absorption band at 490 nm grows in as time progresses and reaches a maximum intensity at 1.8 ps with nearly constant band-peak position and bandwidth.

Figure 2d shows the long-time window of the AuO transients from 1.8 to 50 ps. Within this time scale the intensity of the transient absorption band with a peak at 490 nm decreases and reaches zero intensity at about 50 ps. The band-peak position shifts to the blue as time progresses.

The pump–probe signals of AuO in acetonitrile and acetic acid show unusual time-dependent spectral changes during the first $\sim$300 fs. The signal intensity increases as a function of time beyond what is expected because of the finite laser time width of 80 fs fwhm and the pump–probe system response of $\sim$130 fs. The emission band reaches its maximum intensity at 300 and 333 fs for acetonitrile and acetic acid, respectively. Zero time for the measurements is determined by the maximum intensity of the solvent Raman line.

Figure 3a,b shows the time-resolved pump–probe spectra of AuO in acetonitrile at several times up to 300 fs. Figure 3a shows the signal of AuO in acetonitrile as obtained by the Helios pump–probe system in the spectral range $\sim$420–800 nm. The spectra consist of a strong emission band with a maximum, the position of which depends on time with a band peak at 520 nm at $t \approx$ 300 fs, and a weak absorption band with a peak at $\sim$600 nm of an intensity of $\sim$1/20 of that of the
In our analysis of the emission band we ignore the overlap of the weak absorption.

Figure 3b shows the inverted and normalized transient pump−probe spectra shown in Figure 3a over a spectral range of 15 000−22 500 cm$^{-1}$. We have omitted the first transient spectra at $t = 0$ and $t = 0.033$ ps in which the Raman signal is large and show the fit to a log-normal spectral function of the spectra (as described in the next section). As seen in the figure, the fits are rather good. As is clearly seen in Figure 3b, the shape and position of the emission spectra up to 300 fs depend strongly on time. As time progresses, the value of the full-width half-maximum increases; the band maximum shifts to the red; and the band asymmetry increases.

**Spectral Fitting.** We fit the shape and height of the emission band seen in the pump−probe transient spectra for several measured times over the time range of 0.1−0.3 ps, by a log-normal spectral-line-shape function. The details of the fitting function and parameters are given in the Supporting Information (SI).

Figure 4a shows the position of the emission band peak of AuO in acetonitrile as a function of time in the time region 0−300 fs after an excitation laser pulse (fwhm ∼80 fs) is applied. The peak after 66 fs is at about 20 600 cm$^{-1}$, whereas after 266 fs it is at 19 200 cm$^{-1}$. The total shift within 200 fs is ∼1400 cm$^{-1}$, a rather large red shift over an ultrafast period of time. The shift is linear in time, and this may hint at inertial motion.
of either the solvent or an intramolecular rotation of a moiety of AuO.

The solvation correlation function is given by

\[ S_i(t) = \frac{\nu_i(t) - \nu_i(\infty)}{\nu_i(0) - \nu_i(\infty)} \tag{2} \]

where \( \nu_i(t) \) is the emission-peak position in wavenumber units at time \( t \), and \( \nu_i(0) \) and \( \nu_i(\infty) \) are the emission-peak positions at \( t = 0 \) and at a long time \( (t \approx 300 \text{ fs}) \), respectively. Figure 4b shows the solvation correlation function which is calculated from the emission-band peak position shown in Figure 4a.

The \( S_i(t) \) of coumarin 153 in many solvents has been measured by Maroncelli and co-workers. It was found that \( S_i(t) \) for many solvents spans three decades of time and could be approximated by a four-exponential function \( S_i(t) = \sum_{i=1}^{4} a_i \exp(-t/\tau_i) \). In AuO at the short time of 300 fs, \( S_i(t) \) decays linearly rather than exponentially with time.

AuO, alone and in a mixture with coumarin 102, was also chosen by Mark Berg to test his hypothesis on the contributions of dynamic homogeneity and dynamic heterogeneity as sources for line broadening for nonexponential contributions of dynamic homogeneity and dynamic heterogeneity. The Glasbeek model assigns a diffusion constant to the rotational motion of the two aniline groups around the central carbon bridge. The diffusion motion of the excited-state population modifies the width of the initial population distributions and controls the population movement along the adiabatic-potential surface as a function of the rotational angle. Therefore, it is expected that both the emission spectral width and the emission red shift obey a SQRT time dependence.

The rate of the emission-band shift in acetic acid is slower by about 7% than that in acetonitrile, while the solvent viscosity of acetic acid (\( \eta_{\text{acetic}} = 1.15 \text{ cP} \)) is about three times greater in acetic acid than in acetonitrile (\( \eta_{\text{acetonitrile}} = 0.35 \text{ cP} \)). The comparison suggests that the process that shifts the emission band to the red at the ultrafast time of up to 300 fs is not linearly related to viscosity.

**Global Analysis.** Figure S1 in the SI shows the global analysis of the transient pump–probe spectra of AuO in acetic acid and in acetonitrile shown in Figures 2 and 3. The global fit to three exponents provides three time constants for the excited-state dynamics of AuO in these solvents. For acetic acid, these are 322 ± 150 fs, 2.2 ± 0.8 ps, and 65 ± 6.5 ps. Global analysis is not a good analysis tool when spectral band-shift is the main time-dependent process that takes place. The global-analysis results of the AuO pump–probe transient spectra support the conclusion that three major processes occur in the
excited state. We claim that there is distinct short-time behavior that takes place within 300 fs; this is also supported by the global-fit spectra. Our analysis shows that the position, bandwidth, and asymmetry of the emission peak are time-dependent within this short time period. This could not be seen in the global-fit spectra. The intermediate time constant of about 0.4 ps for acetonitrile and 2.2 ps for acetic acid is related to the fluorescence decay by the adiabatic coupling mechanism linked to the aniline rotation. The intermediate decay time depends on the solvent viscosity. This topic was extensively studied by Glasbeek and co-workers,26 by Meech and co-workers,30 and by others. The third and long decay-time component of about 65 ps in acetic acid is associated with the relaxation of the dark excited-state absorption $S_{1}^{D} \rightarrow S_{0}^{D}$. The long decay time is attributed to the nonradiative decay of the dark state and the repopulation of the ground state.

Figure 8a and 8b show a comparison of the time-dependent red band shift with the fwhm of the AuO emission in acetonitrile and acetic acid, respectively.

As seen in both panels, the rate of increase of the fwhm and the rate of the red band shift of AuO in both solvents are similar. In the case of a linear solvation response, a simple relationship should exist between the spectral broadening and shift that result from solute–solute interactions.50–52 Specifically, if $\sigma$ represents the inhomogeneous broadening of either the absorption or emission spectrum, it should be related to the shift of the spectrum due to nuclear repolarization by

$$\Delta \nu = \frac{\sigma^2}{kT}$$

The validity of this relationship is independent of any particular solvation model. We indeed find that the result of the linear-response theory holds for the large spectral change of AuO emission within the first 300 fs after excitation.

Figure 9a shows the transient pump–probe spectra of AuO in 1-propanol at several times up to $\sim$220 fs after excitation.

The band intensity at $t \approx 0$ is already near the maximum intensity, whereas in both acetonitrile and acetic acid the band intensity shows an increase between $t = 0$ and about 300 fs. The emission-band intensity of AuO in 1-propanol decreases with time. The emission-band peak position of acetic acid at $t = 0$ is at 21 200 cm$^{-1}$, whereas in acetonitrile it is much smaller, about 1300 cm$^{-1}$.

The bandwidth increase with time is rather small compared to the fwhm at $t = 0$. At 220 fs the fwhm reaches a maximum value of 2300 cm$^{-1}$, while at $t = 0$ it is $\sim$1900 cm$^{-1}$. For acetonitrile, the fwhm increase in this time window is $\sim$1700 cm$^{-1}$, greater by about a factor of 4. The emission-band peak position of acetic acid at $t = 0$ is at 21 200 cm$^{-1}$, whereas in acetonitrile it is at 20 600 cm$^{-1}$.

Figure 9b shows both the peak position and the fwhm of the emission band of AuO in 1-propanol in the first 220 fs after the laser-pulse excitation. The band position is almost fixed at about 20 300 cm$^{-1}$; the fwhm increases slightly, by about 500 cm$^{-1}$; and the asymmetry also shows a slight increase with time. These results differ from those shown in Figures 3 and 6 for the emission band of AuO in both acetonitrile and acetic acid.
Figure 10 shows the transient pump–probe spectra of AuO in polyvinylpyrrolidone (PVP) film.

In PVP film, we assume that all the intramolecular rotational motions are hindered, and therefore large spectral changes with time are not expected. The normalized emission spectra shown in Figure 10b show, as expected, only a minor band shift within the time window 0–4 ps. The full-width half-maximum of the signal increases with time, but at a much slower rate than in acetonitrile. The fwhm is large at all times. The band-peak position at \( t = 0 \) is already at a rather low energy \( \tilde{\nu}_p \approx 19400 \) cm\(^{-1} \) compared to 20700 cm\(^{-1} \) in acetonitrile. We assume that AuO in PVP film is in the ground-state conformation, and segment rotation of both the dimethylamine groups, as well as that of the aniline groups around the center bridge, is prevented. An important finding of the transient pump–probe spectra of AuO in PVP is that no transient absorption at 490 nm at long times, that we assigned to the dark state \( S_{1D} \rightarrow S_{nD} \), is observed. We therefore conclude that rotation of the aniline is prevented, and so AuO in PVP does not cross to the dark state. A nonradiative process still exists, even in the fluorescing state, but the lifetime of the fluorescing state is rather long, about 90 ps.

Figures S2a and S2b in the SI show the transient pump–probe spectra of AuO in DMSO. DMSO is a polar (\( \pi^* = 1.0 \)) and also basic solvent with large hydrogen-bond-accepting (HBA) capabilities (\( \beta = 0.76 \)) and lacks hydrogen-bond-donating (HBD) capabilities (\( \alpha = 0 \)). As seen in the transient spectra also in DMSO, the spectrum intensity rises sharply over the instrument-response time. In contrast to the spectra in both acetonitrile and acetic acid for which HBA is rather small\(^{49} \) (\( \beta = 0.40 \) and 0.45, respectively), their HBD \( \alpha = 0.19 \) and 1.12. The bandwidth and peak band position in the transient spectra of AuO in DMSO depend only slightly on time, whereas in acetic acid and acetonitrile, both depend linearly on time and increase by about 1400 cm\(^{-1} \) within the first 300 fs after excitation.

**Auramine-O in Dichloromethane.** Figure 11 in panels a and b show the time-resolved pump–probe signal of AuO in dichloromethane, measured at different wavelengths. Dichloromethane is a polar and very weak protic solvent with low viscosity (\( \eta = 0.44 \) cP) at room temperature. The Kamlet–Taft solvent parameters are\(^{49} \) \( \pi^* \approx 0.82, \beta = 0.10, \) and \( \alpha = 0.13. \) A striking feature in the signal is the oscillation seen at 200 fs after the signal peak. We believe this phenomenon stems from an inertial intramolecular rotation, which we assign to the N,N-dimethyl group; from a nearly “parallel” orientation with respect to the phenyl ring of the aniline in the ground state to a perpendicular position in the excited state. We assume that the transition dipole moment between the excited state and the ground state of AuO depends on this rotation. At short wavelengths the signal intensity decreases since the emission band shifts to the red with time. When the angle of the dimethylamino group is at minimum potential, the angular velocity is large, and in dichloromethane the solvent friction is small; therefore, the rotation angle continues to increase beyond the minimum-potential point. The potential energy increases at angles larger than that for the minimum potential, and thus the restoring force of large angles brings the system back to the equilibrium position at minimum potential. The under-damped oscillation seen in the time-resolved emission at many wavelengths in the spectral region 450–610 nm shown in
Figure 11 is a consequence of such under-damped rotation. This oscillation is not observed in other solvents, including acetonitrile, with even lower viscosity.

Figure 12a,b shows the transient pump–probe spectra of AuO in dichloromethane.

The intensity of the spectra reaches a maximum at the rather short time of about 0.17 ps, whereas in acetonitrile and acetic acid, the fluorescence intensity reaches the maximum value at twice as long a time as than in dichloromethane, about 0.3 and 0.33 ps, respectively. We explain this large time difference by the low friction in the rotational motion of the dimethylamino group in dichloromethane. The shape of the AuO emission band in dichloromethane is much more symmetric than in acetic acid and acetonitrile at long times as well. These results could also be explained by the rather small friction of the solvent that enables rotation of the dimethylamino group under-damped oscillation.

**DISCUSSION**

**Main Findings:**

1. Three time constants are observed for the relaxation of electronically excited AuO.

2. Within the short time window of $\tau \sim 300$ fs, an emission band at $\sim 500$ nm is formed.

3. In non-HBA or weak HBA solvents, the intensity of the emission band increases; the band shifts to the red; the bandwidth increases; and the band asymmetry increases with time.

4. In HBA solvents like DMSO and 1-propanol at this short time window, the asymmetry, width, and position of the emission band change only slightly with time.

5. We find oscillation with a time period of $\sim 200$ fs in the time-resolved emission of the emission band of AuO in dichloromethane.

6. At the intermediate time window from $300$ fs to a few picoseconds, the intensity of the emission band decreases, and the band maximum position shifts to the red. In alcohols at long times the band maximum is at $\sim 560$ nm. The decay of the emission-band intensity depends on the solvent viscosity.

7. An absorption band with a peak at $\sim 490$ nm is formed at the same rate as the decay of the emission band. The average decay times of the emission band in methanol ($\eta = 0.6$ cP), ethanol ($\eta = 1.2$ cP), 1-propanol ($\eta = 2.3$ cP), and 1-butanol ($\eta = 2.5$ cP) are 0.81, 1.86, 3.7, and 5.0 ps, respectively.

8. The transient absorption decay time is much longer than the fluorescence decay. In most of the solvents studied, the decay times are in the range of 10 to about 30 ps.

9. The decay time of the transient absorption at 490 nm of AuO in dichloromethane is exceptionally long—$\tau \sim 200$ ps—compared to $\sim 20$ ps in acetonitrile.

**Short-Time Transient Spectroscopy of Auramine-O.** In this study we focused our attention on the short time window of the first 300 fs after excitation. Most of the previous time-resolved studies of AuO described the relation between the solvent viscosity and the rather short fluorescence lifetime. It was found that the AuO fluorescence lifetime in many solvents could be linearly related to the solvent viscosity. This observation and the long-time transient absorption led Glasbeek and co-workers to suggest a model that relates the fluorescence decay and the time-dependent band shift to the solvent viscosity. A short description of this model is given in the SI.

The model fit to intermediate and long times was satisfactory and explained both the viscosity dependence of the fluorescence decay and the red band shift with time. In contrast to the success of the model in fitting the time-resolved spectra at intermediate and long times, at short times, the short wavelengths show a much faster decay time as well as rapid red band shift. This could not be fitted by a model in which a single time-independent diffusion coefficient is assumed. Meech suggested that at short times the diffusion coefficient of the two-aniline-ring rotation should be time dependent since the friction should be treated as dielectric friction.

In the current study, we provide an alternative explanation and propose that the rapid decay of the AuO emission arises from the intramolecular rotation of about 60° of the N,N-dimethyl groups around the C−N bond. The rotation time we find is about 300 fs when the solvent friction controls the overdamped motion. In the non-hydrogen-bonding solvent dichloromethane, the hydrogen-bond interaction of AuO with the solvent is zero, and the rotation time is $\sim 200$ fs. Oscillation is seen in the time-resolved emission (see Figure 11) as a consequence of the under-damped rotational motion of the dimethyl groups of the N,N-dimethylaniline moiety.

Solvation dynamics show that there is a fast solvation component with a characteristic time, shorter than 100 fs, in many solvents. In the current experiment on AuO the short characteristic time component is longer than the solvation dynamics component with $\tau \sim 250 \pm 50$ fs. This longer decay time, by a factor of 2, is in favor of the interpretation of
intramolecular rotation of the rather small dimethylamino group of the aminilen of AuO. The rotation time of the aniline group of a ThT molecule, a molecule similar to AuO, was calculated by Mukamel and co-workers. They found a rotation time of about 400 fs in an unperturbed molecule.

Estimation of the Inertial Twisting Time of the Dimethyl Groups. An estimation of the moment of inertia, \( I \), of the dimethyl groups around the C–N bond is given by

\[
I = mr^2
\]

where \( m \) is the mass of the two dimethyl groups, \( m = 30 \text{ au} \), and \( r \approx 4.98 \times 10^{-26} \text{ kg} \). The distance perpendicular to the rotation axis of the center of mass of the CH3 is about 1.21 Å, and so \( I = 4.98 \times 10^{-26} \times (1.21 \times 10^{-10})^2 \approx 7.3 \times 10^{-46} \text{ kg m}^2 \).

The rotation rate of the dimethyl groups depends on the angular dependence of the potential in the excited electronic state. Sawada and co-workers calculated the angular-energy dependence of the excited electronic state in vacuum of both the ground and excited states. They found that the minimum energy is at about 30°. The rotation angular-energy dependence of their calculation is probably much too large. We therefore calculate the rotational frequency of the dimethylamino groups by assuming that the potential difference between the initial angle of 60° and the final 90° position is only 100 mV. This relatively small driving potential will be transformed into angular kinetic energy

\[
\frac{1}{2}J\omega^2 = \frac{1}{2}I\omega^2 = \frac{1}{2}m(r\omega)^2 = \frac{1}{2}I\omega^2 = \omega^2 \approx \frac{\omega}{k_B T} \approx 1.6 \times 10^{13} \text{ Hz}
\]

and so, the inertial free-rotation time \( \tau_{\text{ff}} \approx 150 \text{ fs} \).

The rapid fluorescence-band red shift and the increase in the bandwidth of AuO in many solvents takes about 300 fs longer than the system response and is slower by about a factor of 2 than the inertial free rotation of the dimethylamino groups under 100 mV driving force. Friction with the solvent surrounding the dimethylamino groups is obviously expected to lower the rotation frequency and to bring about an increase in rotation time. Experiments presented here show that the fluorescence band undergoes large changes in about 300 fs in solvents with small or non-hydrogen-bond-accepting abilities. The most striking observation is that the fluorescence time-dependence signal of AuO in dichloromethane, as shown in Figure 11, exhibits an unmistakable 200 fs oscillation. This solvent is a weak protic and strong polar solvent with very small hydrogen-bond donating or hydrogen-bond-accepting capabilities. We explain this oscillation as a signature of the underdamped rotation of the dimethylamino low-friction condition. The oscillation arises as a consequence of an overshoot of the minimum energy, but it eventually reaches the minimum potential. In the hydrogen-bond solvents, the rotation of the dimethylamino group is strongly influenced by the solvent friction and the rotation overdamped, and no such oscillations are observed.

Mukamel and co-workers used ab initio calculations to study the time-resolved photoisomerization of the ThT molecular rotor. The measured nonradiative rates of ThT are somewhat lower than that of AuO, but the overall solvent-viscosity dependence of the nonradiative rate is similar. The chemical structure of ThT is shown in Scheme S1 in the SI. ThT contains an aniline group and a methylbenzothiazole ring. The rotation of the two-ring subsystem is responsible for the high nonradiative rate. They found, as previous studies have shown, that a high barrier exists at \( \Phi_2 = \pm 60° \) for the rotation of the dimethylamino group. Another finding was that a variation of the twist angle of the dimethylamino groups (\( \Phi_2 \)) in Figure 1 of ref 53 occurs during the simulation time of the S1 state, and the dimethylamino groups oscillate in the potential-energy well with \( |\Phi_2| < 50° \). The evolution of both twist angles, \( \Phi_1 \) and \( \Phi_2 \), is related to their vibrational modes. The \( \Phi_2 \) twist-angle mode is ~96 cm\(^{-1}\) and \( \Phi_2 \), and that of the dimethylamino rotation with respect to the benzene ring is ~85 cm\(^{-1}\). In our current work we see, in the time-resolved emission signals, a ~200 fs oscillation which could be related to the ~85 cm\(^{-1}\) oscillation that Mukamel and co-workers found in their simulation. They observed a rotation time of about 400 fs in an unperturbed molecule.

### SUMMARY AND CONCLUSIONS

We used steady-state and time-resolved optical techniques such as fluorescence up-conversion and pump–probe spectroscopy to study the excited-state dynamics of Auramine-O (AuO), a molecular rotor, in several solvents at room temperature. We found that electronically excited AuO relaxes to the ground state in three time steps.

The shortest decay time component is associated with a strong emission with a band maximum at about 500 nm. The decay time is about 150–300 fs and is attributed to a twist motion of the dimethylamino group of the two anilines of AuO (see Scheme 1). We found three behavior characteristics of the emission bands at short decay times. In polar solvents that have HBA properties, after the excitation process by an 80 fs, 410 nm laser pulse, the fluorescence bandwidth, peak position, and asymmetry change only slightly with time up to about 300 fs. The solvents we used that belong to this category are DMSO and 1-propanol.

The solvents in the second group we used are polar but either lack or exhibit weak HBA. Acetic acid is a mild acid with \( pK_a \approx 4.7 \) and has a high HBD property but lacks HBA property, and acetonicitrile has very small HBA properties and rather low HBD. Both belong to this second group. In these two solvents, we observed profound changes in the AuO emission-band characteristics as a function of time within the short time window. The band-peak position shifts to the red; the bandwidth increases; and the band asymmetry increases as well, as a function of time. In acetonitrile, both the band shift and the bandwidth increase by about 1400 cm\(^{-1}\) within 250 fs in a linear dependence on time.

The third type of solvent that affects the time dependence of the AuO emission band is one that is polar but lacks both types of hydrogen bonds, namely, HBA and HBD, and also has low

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**Scheme 1. Molecular Structure of Auramine-O**

![Scheme 1. Molecular Structure of Auramine-O](image)
viscosity. Dichloromethane (DCM) belongs to this group. We found that the AuO emission band in DCM shows a rather small band shift and bandwidth increase within the short time window of ~200 fs. The fluorescence time dependence shows distinct oscillation with a time constant of about 200 fs (see Figure 11). We attribute the oscillation to an under-damped twist motion of the dimethylamino group of the two AuO anilines.

The second time constant that is associated with the excited-state dynamics of AuO is attributed to the rapid decay of the fluorescence band and further red band shift from about 510 nm to about 550 nm in hydrogen-bonding solvents like alcohols. In the case of linear alcohols like methanol to 1-pentanol, this intermediate time dynamics depends on the viscosity of the solvent. This fluorescence decay was successfully explained by a model first introduced by Glasbeek and co-workers.26 The model is based on adiabatic coupling between two excited states: the initially strongly fluorescing excited state and a charge-transfer dark state. The coupling strength depends on the angle of rotation between the two aniline-ring planes. When the angle is 90°, the coupling strength reaches a maximum, and the oscillator strength of the mixed excited states is ~0.01 of that of the fluorescing state. Thus, the fluorescence intensity falls as a function of time, and this gradual decrease in the intensity is a function of the rotation of the aniline from an initial, nearly parallel structure, to its final perpendicular structure.

The third time constant is associated with a rather long decay time of a dark state that we attribute to the S1 → S0 transition. The decay time is about 10−20 ps in hydrogen-bonding liquids with medium viscosity. It is much longer (about 200 ps) in dichloromethane, which lacks hydrogen-bonding properties.

It is in the shortest decay time component of AuO in solvents such as DCM, where under-damping of the rotation is evident, that we find the most compelling argument for convergence of the two fields, that of fluorescent molecular rotors and that of histological dyes like AuO. Such oscillatory behavior strikes us as a defining characteristic for the promise of fluorescent molecular rotors, which constitute a great portion of the servomotor candidates developed since Richard Feynman’s talk in 1959. We therefore allow ourselves the liberty to conclude with a paraphrase on Robert Frost’s renowned poem “The Road Not Taken”: two roads converged in a yellow dye. Once converged, they can now be trodden to serve both aims at once.

**ASSOCIATED CONTENT**

**Supporting Information**

A Molecular structure of Thioflavin T (ThT) B. Time-resolved pump—probe spectra of AuO. This material is available free of charge via the Internet at http://pubs.acs.org.

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