**Calculating the spectral overlap integral**

***1. Time domain representation***

A core concept in Forster Resonance Energy Transfer (FRET) is the ***spectral overlap integral*** which measures the degree of energetic similarity between a donor and an acceptor chromophore. It appears in the definition of the Forster rate,

where and are the fluorescence spectrum of the donor and absorption spectrum of the acceptor as functions of either energy or frequency. While this is a good formulation for illustrating the concept of ***resonance*** in FRET it is not easy to evaluate since no general expressions exist for and . A more convenient representation is the ***time domain*** which is related to the frequency domain via the Fourier transform,

For reasons that will become apparent we will take advantage of the fact that and are real functions, meaning they lack imaginary parts and are therefore equal to their own complex conjugate,

Using the identity that,

we get,

This is essentially a reflection of ***Parsival’s theorem*** (if you wanted to know). and are fluorescence and absorption ***response functions*** respectively which, unlike and , can be expressed in a simple analytical form. They are derived from Kubo’s line-shape formulation of linear response theory and are,

is the ***frequency of the 0-0 transition*** of the donor/acceptor,

Where is the ***0-0 transition energy***. is the so-called ***reorganization energy*** of the donor, which characterizes the vibrational relaxation that occurs on the excited state following excitation and on the ground state following de-excitation. Due to the symmetry of the displaced oscillator model of molecular excitations the fluorescence peak of a chromophore is red-shifted by an amount relative to it absorption peak. This is the so-called ***Stokes shift***. Lastly, is known as a ***line broadening function***. A simple way to think about this is that for a given chromophore,

1. determines the peak position of the absorption spectrum.
2. determines the peak of the fluorescence.
3. determines the width and vibronic structure of the spectra.

It is very important to notice the symbol in Eqns. (6) and (8) which implies a ***complex conjugate*** operation. Both the response functions and the line broadening functions are ***complex*** (having ***real*** and ***imaginary*** parts). If we define and complex number as,

then,

As such,

The line broadening function, , for chromophore is defined by the ***fluctuation-dissipation theorem*** as,

where is the so-called ***spectral density*** of the system-bath interaction. The meaning and functional form of are discussed below but here we merely assume that it is a known, real function of frequency. Similarly, the ***reorganization energy*** is defined,

However, we have generally already defined the reorganization energy when constructing the spectral density. However, there is a subtlety that we will discuss below.

has an important property that will help us recast Eqn. (6) in a more convenient form,

This means that,

We can split up the integral in Eqn. (6) to give,

If we separate the real and imaginary parts of Eqn. (20),

We now see that,

So,

This last step may look like pointless mathematical messing around but it is far easier to numerically evaluate an improper integral with only one infinite limit than one with two. One of the things we notice is that while and are complex the overlap integral is real. However, this is only true if it is evaluated carefully. Brute force methods will generally leave some residual imaginary component. Therefore we make the stipulation that we are only interested in the real part of the integral,

***2. Evaluating the line-broadening function***

To evaluate integral in Eqn. (24) we need to define and and treat their real and imaginary parts separately,

where,

and,

It is important to remember that while is a complex function both and are real functions. Since the ***integrands*** of Eqn. (26) and (27) are quite complicated it can be useful to break them up further,

where the ***integrand functions*** are defined,

It is ***very important*** to note that the that appears in and is simply a variable that we integrate over. It is not the ***transition frequency of the chromophore***. Therefore, the computational procedure is this,

1. Compute the integrand functions , , , and .

In **lineshapes.py**  is named **gReInt**while is called **gImInt**.

2. Use a numerical function integration routine (such as Gaussian quadrature) to obtain , , and .

In **lineshapes.py** is named **gRe** and is named **gIm**.

3. Assemble and .

In **lineshapes.py** this is done with **gt**.

***3. Evaluating the integral***

In addition to the line broadening functions we require the transition frequencies, and , and the reorganization energy of the donor, . These parameters are obtained by fitting the absorption and fluorescence spectra of the relevant chromophores but there are a few things we need to consider when dealing with .

The reorganization energy is a measure of ***vibronic coupling***. In other words it quantifies how fluctuation of the nuclear coordinates effect the electronic transition energy/frequency. As such it is totally defined by the spectral density function as illustrated by Eqn. (16).

For chlorophyll people generally use the phenomenological spectral density of Marcus and Renger,

where and are natural nuclear vibrational frequencies and , and are coupling constants. This spectral density is rather finely parameterized and in addition to absorption and fluorescence can reproduce a large range of non-linear spectra. It doesn’t contain as an explicit parameter so one would have to evaluate Eqn. (16), albeit only once. Since we are really only interested in the absorption and fluorescence spectra and the low resolution time-resolved fluorescence, we don’t really need a detailed spectral density. The Qy peak is essentially just a featureless Lorentzian so we can use something much simpler. We will use the ***over-damped Brownian oscillator*** (ODO) model,

where is the reorganization energy and is the correlation time of bath fluctuations. As a simple proof,

The carotenoid S1 state has a non-trivial vibronic structure and so we must use a more complicated spectral density composed of one ODO and two under-damped vibrational modes,

With this we have a slight problem. While this spectral density gives us a correct overall line shape it is basically a fudge. The true spectral density cannot be decomposed in this manner. This results in two problems:

1. It introduces anomalous shift in the absorption spectrum so that the 0-0 line does not occur at .
2. Eqn. (16) will give an excessively large Stokes shift.

Problem (1) is solved by adding a correction to the line shape function,

Problem (2) is solved simply by setting the Stokes shift as .

Now that we have all of the parameters is possible to numerically evaluate the integral in Eqn. (24). The best way to do it is not to brute force a solution but to break it up into two parts,

Note that both of the terms in Eqn. (39) are explicitly real.

1. **Spectral\_density.py** contain several spectral density functions. They each return 3 variables. CAR\_2MODE[0]= is the spectral density value, CAR\_2MODE[1]= is used for the Stokes Shift, CAR\_2MODE[2]= is used for the anomalous shift. ODO[0]=, ODO[1]= and ODO[2]=0.0. ***CHL\_RENGER is not finished*** CHL\_RENGER[0]= while CHL\_RENGER[1]= CHL\_RENGER[2]=0.0. If one wishes to use CHL\_RENGER then one will either have to calculate elsewhere or modify it to calculate and return CHL\_RENGER[1]=.

2. **gt** in **lineshapes.py** returns three variables. gt[0] is the actual value and gt[1] is the error on the numerical integration (used for de-bugging only). If you are using an ODO or carotenoid spectral density then gt[2] is the reorganization energy (which is obtained from the spectral density function). If you are using the Renger spectral density then gt[2]=0. ***This needs correcting!***

3. Define the response functions and .

**NOTE:** while **At** in **Spectrum\_mol.py** is correct, **Ft** is incorrect and should be entirely rebuilt from nothing.

4. Numerically evaluate the integral.