

Calculating an unbiased fluorescence decay curve recorded by a TCSPC system with detector and electronics dead-times

The function `TCSPC_DTcorrected` calculates decay curves measured with a single-photon counting system with ps/ns temporal resolution under pulsed excitation. The function returns both an unbiased, dead-time corrected TCSPC curve as well as the uncorrected “standard” TCSPC curve. The function call is:

```
[tcspc_corrected,tcspc_standard,epsilonP] =  
TCSPC_DTcorrected(tPhoton, syncPeriod, DT_electronics, DT_detector, flag)
```

All input parameters except for `flag` are in time-units of TCSPC bins.

<code>tPhoton</code>	A vector containing the absolute photon arrival times.
<code>syncPeriod</code>	A scalar containing the time between two consecutive sync pulses (P in the paper).
<code>DT_electronics</code>	A scalar containing the dead-time value of the electronics (E in the paper).
<code>DT_detector</code>	A scalar containing the dead-time of the detector (D in the paper).
<code>flag</code>	If set to 1, the folder “subroutines” has to be added to the MATLAB path manually (see below).

The outputs `tcspc_corrected` and `tcspc_standard` of the function are the unbiased and biased decay curves, respectively. The former is normalized such that `sum(tcspc_corrected)` equals the average number of photons per excitation period (ϵP in the paper), the latter is not normalized, therefore `sum(tcspc_standard)` equals `numel(tPhoton)`. The output `epsilonP` is the average number of photons per excitation period ϵP . It can be used to obtain a corrected intensity value for the pixel by multiplying with the number of excitation cycles for this pixel via:

```
intensity=ceil( (max(tPhoton)-min(tPhoton)) / syncPeriod) * epsilonP
```

Note for improving performance:

In order not to clutter your file system, we put all subroutines into a folder called “subroutines”, which is added to the MATLAB path in the first few lines of `TCSPC_DTcorrected`. If you call this function several times, for example in a script where you evaluate many pixels, there is no need to add the subfolder to the path every time. In this case, we recommend that you set `flag` to 1 and add the folder manually / in the script that calls `TCSPC_DTcorrected` using the following code:

```
fullPathToThisFile = mfilename('fullpath');  
[path,~,~] = fileparts(fullPathToThisFile);  
addpath(genpath([path,filesep,'subroutines']));
```

Example:

A typical example is shown below. The photon arrival times were simulated for a TCSPC bin width of 64ps, $\varepsilon P = 1.5$, an excited state lifetime of 3ns, an excitation period of 50ns, an electronics deadtime of 80ns, a detector deadtime of 30ns and a total number of 5000 photons. The photon arrival times are given as the vector `tPhoton` in the file `tcspc_example.mat`.

```
syncPeriod = 782;  
DT_electronics = 1250;  
DT_detector = 516;  
load('tcspc_example.mat');  
[tcspc_corrected,tcspc_standard] = TCSPC_DTcorrected(tPhoton,...  
    syncPeriod,DT_electronics,DT_detector);  
t=0.064*(1:782);  
plot(t,tcspc_standard/sum(tcspc_standard),'-b',...  
    t,tcspc_corrected/sum(tcspc_corrected),'-r');  
xlabel('time [ns]')  
ylabel('normalized intensity [-]')  
legend('standard TCSPC curve','unbiased TCSPC curve')
```

A simple exponential fit yields decay times of 1.64ns and 2.93ns for the biased and unbiased TCSPC-curves, respectively. As expected, the unbiased curve gives a much better result.

