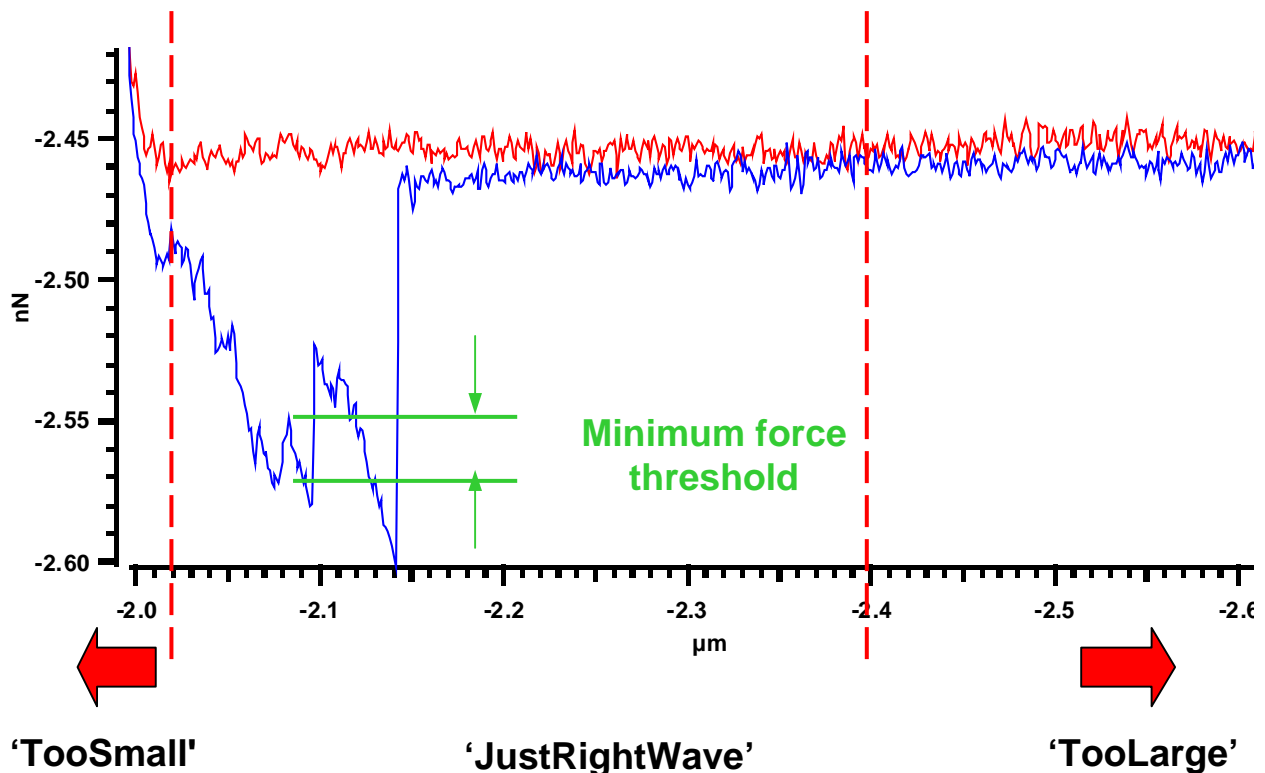


Instructions for pull_off_histogram_v9 (RTV9)

Description-

This version of Russell Taylor's (UNC-CH Computer Science) force-distance curve (FC) analysis procedure not only determines all the dissociation values above a user-defined threshold (in pN) for multiple-event force curves, it also finds these values within a user-defined distance from the zero point. The zero point is determined by averaging the last 10% of the points in the retract curve data, which is used to represent the free air deflection. Version 9 also generates histograms of the number of curves with no events above the defined threshold (0 jumps), 1 event (1 jump), 2, and so on. This new feature will allow probability to be generated more easily.

This distance procedure allows the omission of non-specific events like tip-substrate interactions (typically the first event), or if you know a specific distance that you expect a dissociation to occur. The program generates three separate histograms with regard to the distance window: one labeled 'histogram', which is the desired one that determines dissociation values above the user-defined force threshold AND within the user defined parameters (*called 'JustRightWave'*); a histogram called 'histogram-small', which represents events found above the user defined force threshold, but occur before the user defined distance window; and one labeled 'histogram-large', which represent the values above the force threshold and beyond the desired distance window. The figure below gives a sense of these two user defined criteria: this procedure would recognize this wave as having three 'jumps' in it.



RTV9 Procedure:

Go to File→open procedure→pull_off_histogram_v9 (where ever the file is on your machine); the RTV9 procedure file opens. You can change the smoothing number in the procedure, or change the default parameters (called variables, towards bottom of procedure window) that come up in parameter dialogue (below), just so you don't have to type them in every time if you have many similar data sets. Make sure to click 'compile' at the bottom of the procedure window to activate those changes.

In command line type: pull_off_histogram(); hit return. A dialogue comes up that asks you where the data set is; once you tell it where to find it, the following parameter dialogue comes up-

Parameter	Value
Velocity of retract in nm/Second:	1800
Minimum nm from zero force to pull-off:	20
Maximum nm from zero force to pull-off:	500
Bin size in pN:	5
PicNewtons per nm:	60
Minimum jump in pN:	30

Velocity of retract in nm/second: this value is necessary so program can calculate correct distances (it does it in a time dimension- this is a multiplier to make things simpler for the user)

Minimum nm from zero force to pull off: This value determines the shortest time distance from the zero point that the procedure determines events (jumps) from.

Maximum nm from zero force to pull off: this value determines the upper distance limit from the zero point that the procedure determines events (jumps) from.

Bin Size in pN: this determines bin size in histograms

PicoNewtons per nm: spring constant of cantilever

Minimum jump in pN: this is the minimum threshold force the procedure will count as an event.

Hit 'Continue' button and it churns the data. When complete, three histograms come up (just right, small and large). Do with them what you will. Two more histogram are generated that tell you how many curves had no events, how many had one, two, etc. One histogram tells you this information regarding only the 'just right wave'; the others tell you regarding all the waves in the data set.

Other Igor tasks-

Say you want to know the exact values (in table form) from this sorting procedure for each of the individual histograms generated: go to windows → new table; from the 'Columns to Edit' menu, the following selections will give you:

Countall: shows how many jumps (above minimum force threshold) per curve were detected

Countgood: shows how many jumps (above minimum force threshold) *AND* within the distance window were detected per curve

histwave_countgood: shows how many curves in the data set (above minimum threshold *AND* within distance criterion) had 0 jumps, 1 jump; 2, on so on

histwave_countall: shows how many curves in the entire data set (regardless of minimum threshold *AND* within distance criterion) had 0 jumps, 1 jump; 2, on so on

Histwave_small: shows number of curves detected below the defined distance window; x- axis is Force (in pN) & dependent on the chosen number of bins in histogram

Histwave_large: shows number of curves detected above the defined distance window; x- axis is Force (in pN) & dependent on the chosen number of bins in histogram

JustRightWave: gives the individual force values above minimum threshold *AND* within distance criterion, in table form. Scroll down to end to get number- remember 1st curve has 0000 suffix, so add 1 to what ever point the last one is labeled as.

Outwave: three columns-outwave[][0] gives forces (in pN) at that events' respective distance from zeropoint (outwave[][1]) per curve number/suffix (outwave[][2]).

TooSmallWave: gives force value(s) (in pN) of the jump/event detected *below* the defined distance window; scrolling to end tells how many total in this category

TooLargeWave: gives force value(s) (in pN) of the jump/event detected *above* the defined distance window; scrolling to end tells how many total in this category