

Application note #19

## **AUTOMATIC CONCENTRATION DEPENDANCE STUDIES**

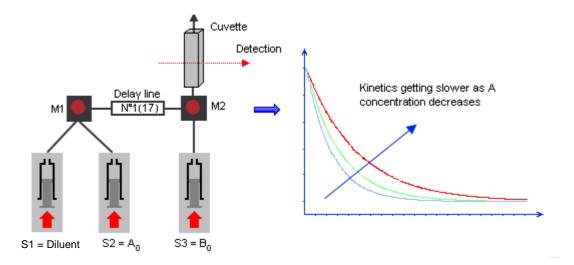
Keywords: automatic concentration variation, dilution,

Concentration dependence studies follow reaction kinetics while varying the concentration of one of the reagents. These studies are tedious and time consuming when performed manually.

Bio-Logic stopped flow systems are designed so that a series of automatic concentration steps for a complete study, can be done quickly. In a couple of minutes with an outstanding precision, a series of concentration can be done 20 times more rapidly compared to competitive instruments! Indeed other stopped flows are unable to propose such kind of program due to poor instrument modularity <sup>(1)</sup>.

Bio-kine is the only software proposing automation of mixing ratio.

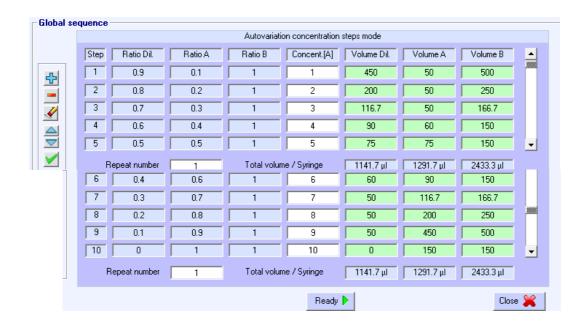
This note illustrates the use of the concentration dependence function of the SFM by following the concentration dependency of A in the reaction A + B, while keeping concentration B constant. It is done using 3 syringes and 2 mixers according to the following diagram.



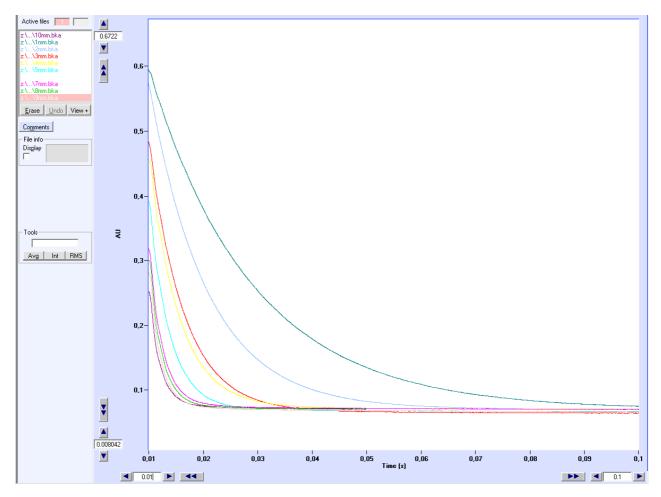
Solutions are loaded in the stopped flow, with mixer M1 changing the concentration of A, and thus diluting solution loaded in syringe 2. The second mixer M2 starts the A+B reaction. A short delay line is installed between M1 and M2 to minimize use of reagent A. The user indicates initial concentrations of A and B, and then the mixing ratios to apply between (A + diluent) and B at each step in mixer 2. The user builds the sequence by varying the ratio "diluent/A" in mixer 1 or by changing the final concentration of A. Once all parameters are set, Biokine calculates the shot volumes per step. The sequence can be customized by the user if desired.

In this example, 140  $\mu$ M DCIP (Dichlorophenolindophenol) is mixed with sodium ascorbate, with the concentration varied automatically from 10mM to 1mM. The program is given below

(1): to perform concentration dependence studies on single drive mechanism stopped-flow, user needs to prepare all concentration manually and is thus exposed to experimental error. System also needs to be refill at each concentration. With a series of 10 concentrations, loss of time will be 20 times compare to Bio-Logic's SFM.



When the sequence is ready, ten shots are performed from lower concentration to higher concentration, and the results are displayed in real time.



Please contact us for more information