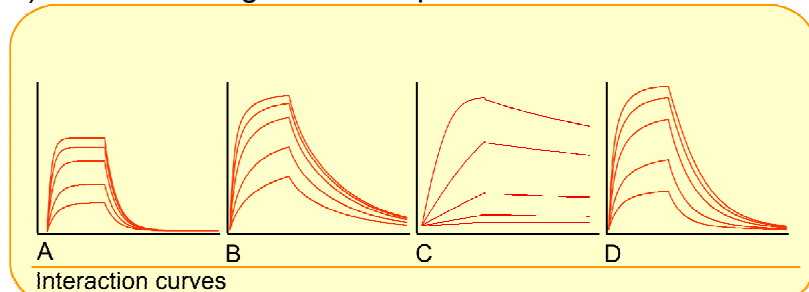


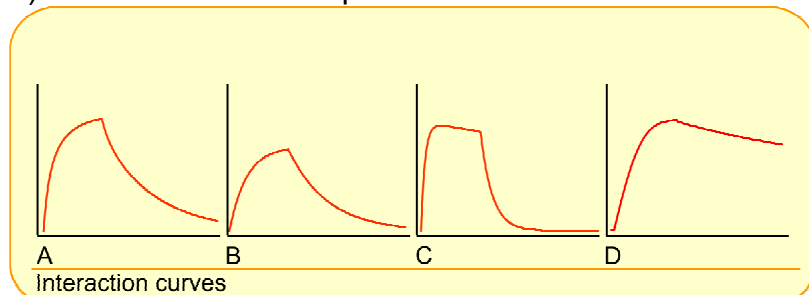
## Sensorgram quiz answers.

1) Which sensorgram has exponential interaction curves?



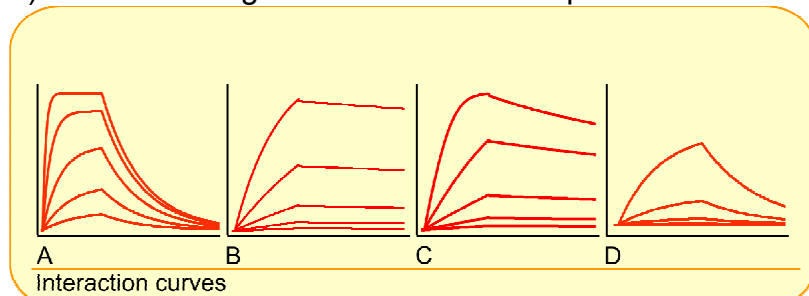
A: exponential B: heterogenic C: mass transport D: conformational change

2) Which curve is an exponential interaction curve with mass-transfer?



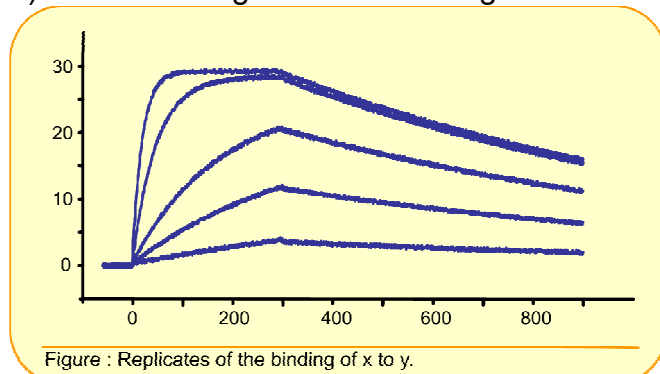
A: heterogeneous B: exponential C: mixed analyte D: mass transport

3) Which sensorgram does not have exponential interaction curves?



A: exponential B: exponential C: mass transport D: exponential

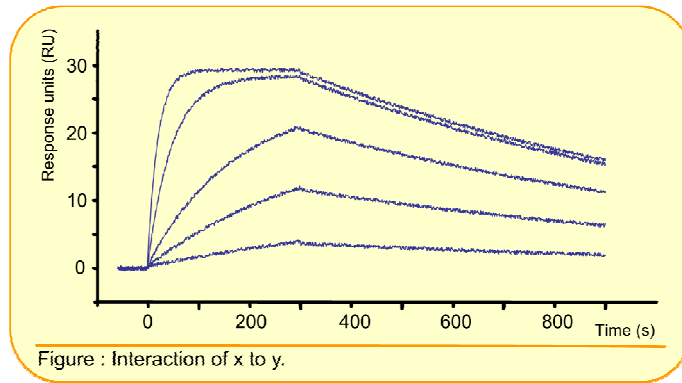
4) What is wrong with this sensorgram ?



- A: the legend is missing
- B: there are no replicates
- C: the bulk effect is too big
- D: not all curves go to steady state

Answer A.

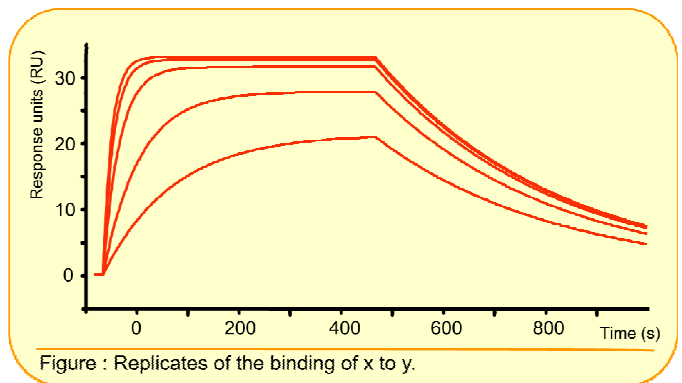
5) What is wrong with this sensorgram?



- A: the legend is missing
- B: there are no replicates
- C: the concentration range is too narrow
- D: not all curves go to steady state

Answer B

6) What is wrong with this sensorgram?

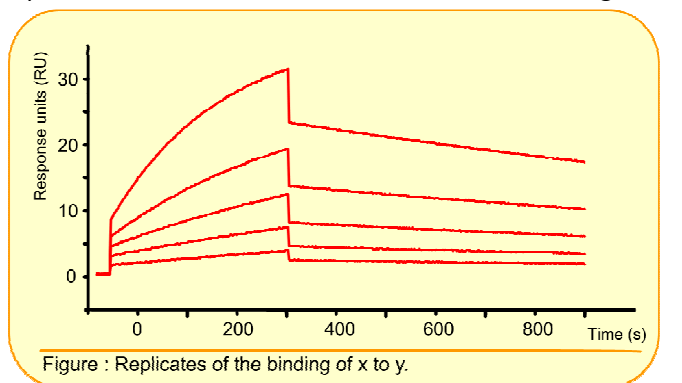


- A: the legend is missing
- B: there are no replicates
- C: the concentration range is too narrow
- D: not all curves go to steady state

Answer C:

The lower range (0.1 – 1 times  $K_D$ ) is missing.

7) What should be solved first before fitting?

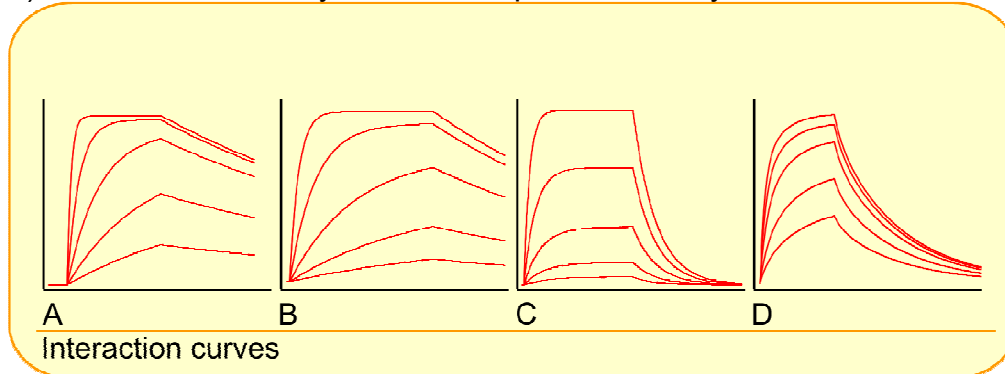


- A: Association time should be longer
- B: These are not exponentials
- C: Make dissociation time longer
- D: Match flow and analyte buffer

Answer D

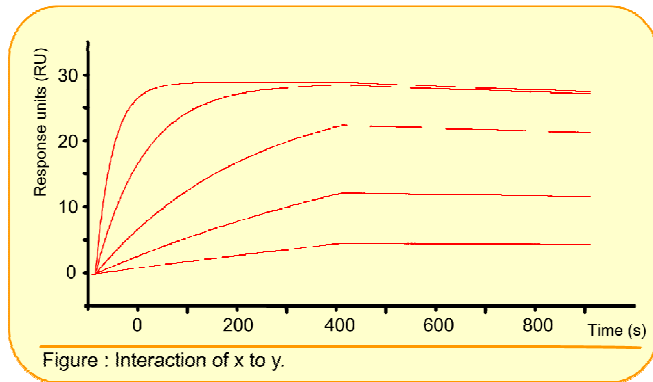
There is a difference between flow buffer and analyte buffer causing a jump.

8) Which dataset can you use for equilibrium analysis?



Answer D: only in C all the curves come to steady state.

9) What should be optimized before fitting?



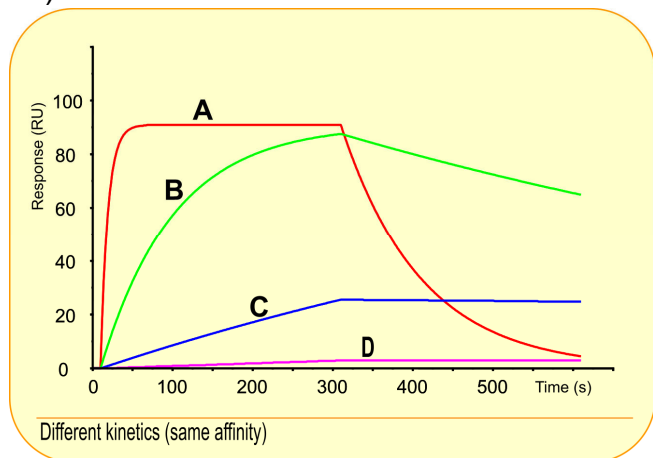
- A: association time longer
- B: longer dissociation time
- C: use higher analyte concentrations
- D: use lower analyte concentrations

Answer B.

For a reliable dissociation measurement at least 5% of the response should be dissociated.

10) Which curve has the fastest dissociation?

11) Which curve has the fastest association?



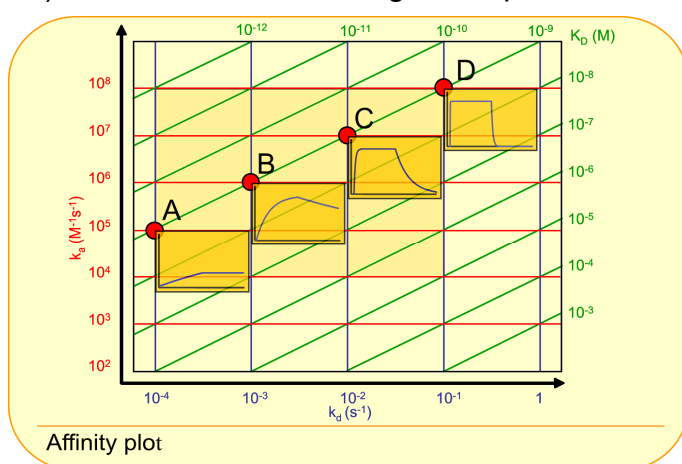
- A: curve A
- B: curve B
- C: you should know the analyte concentration
- D: you should know the  $R_{max}$

10) because dissociation is concentration independent you can deduce it is A.

11) because association is concentration dependent you can not say anything about the association by looking at a curve.  
The answer is C.

12) Which curve has the fastest association?

13) Which curve has the highest equilibrium constant?



- A: A
- B: They are all the same
- C: D
- D: C is higher than B

12) just follow the lines  $C = 10^7$  and  $D$  is  $10^8 \text{ M}^{-1} \text{ s}^{-1}$

13) they all lie on the same green line of  $K_D = 10^{-9} \text{ M}$

14) Which fitting result should you report?

$k_a$	$k_d$	$K_D$
( $M^{-1}s^{-1}$ )	( $s^{-1}$ )	(M)
$1.1 \pm 0.1 E5$	$4.7 \pm 0.2 E-3$	$4.27 E-8$

A

$k_a$	$k_d$	$K_D$
(1/Ms)	(1/s)	(M)
$1.1 \pm 0.1 10^5$	$4.7 \pm 0.2 10^{-3}$	$4.27 10^{-8}$

B

$k_a$	$k_d$	$K_D$
( $M^{-1}s^{-1}$ )	( $s^{-1}$ )	(M)
$1.1 \pm 0.1 10^5$	$4.7 \pm 0.2 10^{-3}$	$4.27 \pm 0.46 10^{-8}$

C

$k_a$	$k_d$	$K_D$
( $M^{-1}s^{-1}$ )	( $s^{-1}$ )	(M)
$1.1 \pm 0.1 10^5$	$4.7 \pm 0.2 10^{-3}$	$4.3 \pm 0.5 10^{-8}$

D

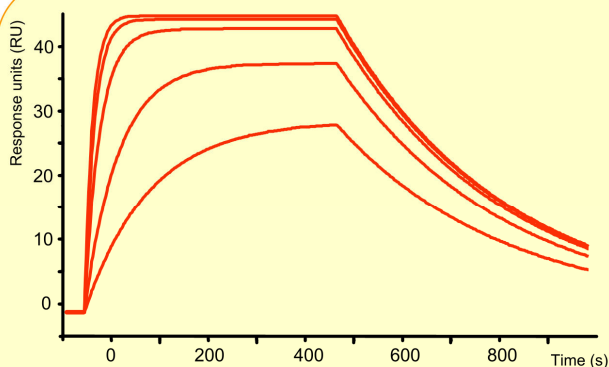
Fitting results

Answer D

Absolute correct would be

$k_a$ :  $\text{mol L}^{-1} \text{s}^{-1}$

15) What is bad in this sensorgram presentation ?



- A: Injection time too long
- B: Concentration range not balanced
- C: Response too high
- D: Not all curves reach steady state

Answer B

16) What can you do when you have this sensorgram?

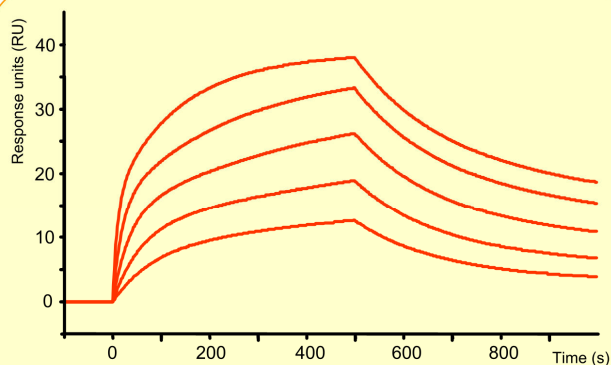
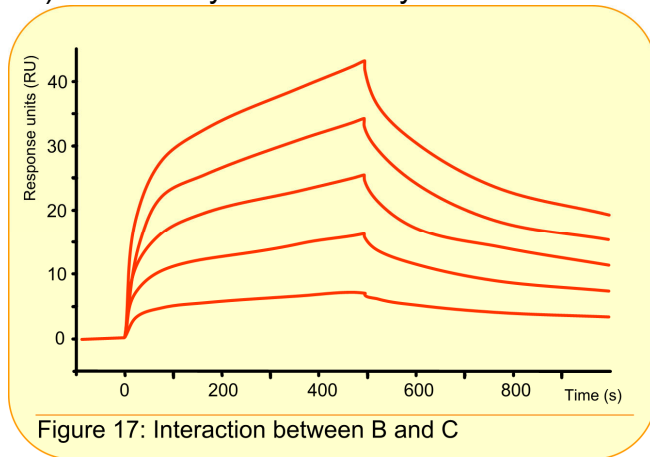


Figure 16: Interaction between A and G

- A: check the analyte for purity
- B: reverse the ligand and analyte
- C: A + B
- D: make the flow rate faster

Answer C

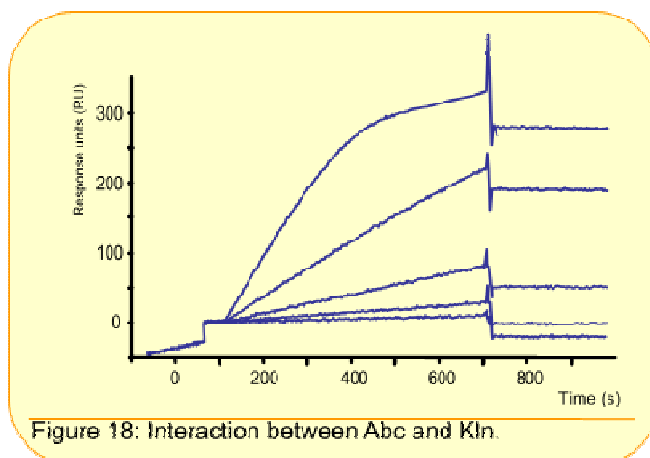
17) What can you do when you have this sensorgram?



- A: check the analyte for purity
- B: reverse the analyte and ligand
- C: try an other immobilisation technique
- D: A + B + C

Answer D

18) What can you do to optimize this interaction?



- 1: lower ligand concentration
- 2: match buffers better
- 3: equilibrate better
- 4: use higher flow rate

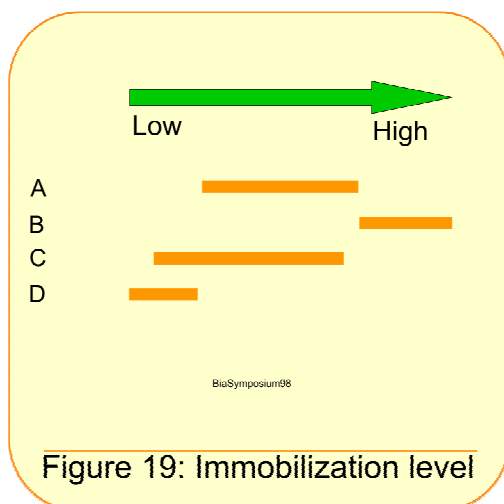
- A: 1 + 2
- B: 3 + 4
- C: 1 + 2 + 3
- D: 2 + 3 + 4

Answer C.

Not D: Although higher flow rates diminish mass transport it is better to lower the ligand concentration by making a new surface.

19) How much ligand should you immobilize for analyte concentration measurements?

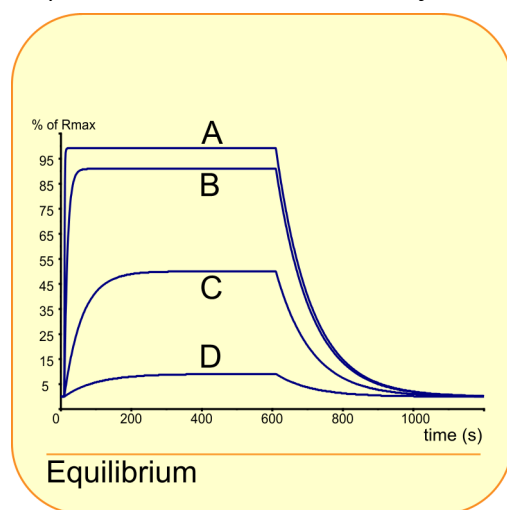
20) How much ligand should you immobilize for kinetic analysis?



- A: low to high
- B: high
- C: low to medium
- D: low

19) Answer B.  
20) Answer D.

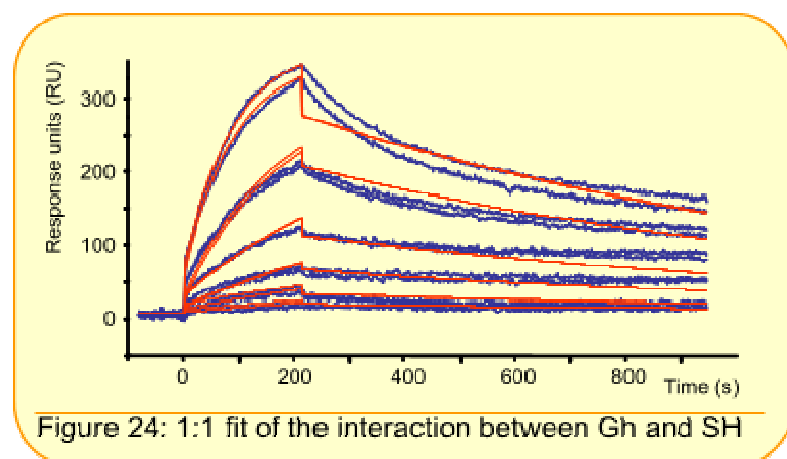
- 21) Which curve is at equilibrium (steady state)?  
 22) Which curve is saturating the ligand?  
 23) Which curve has an analyte concentration comparable to the  $K_D$ ?



A: curve A  
 B: curve B  
 C: curve C  
 D: all four curves

21) Answer D  
 22) Answer A  
 23) Answer C

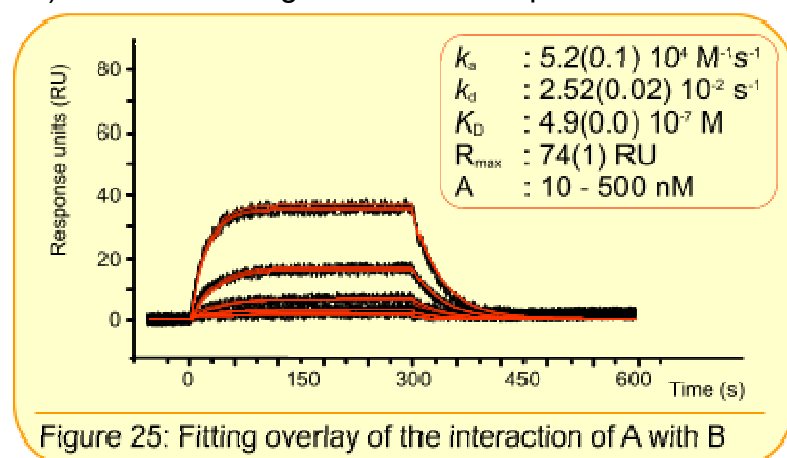
- 24) When you have this fitting as a result. What can you do?



A: lower ligand concentration  
 B: check the ligand for purity  
 C: use higher flow rate  
 D: A + B

Answer D.

- 25) Are the values given in the inset plausible with this sensorgram?

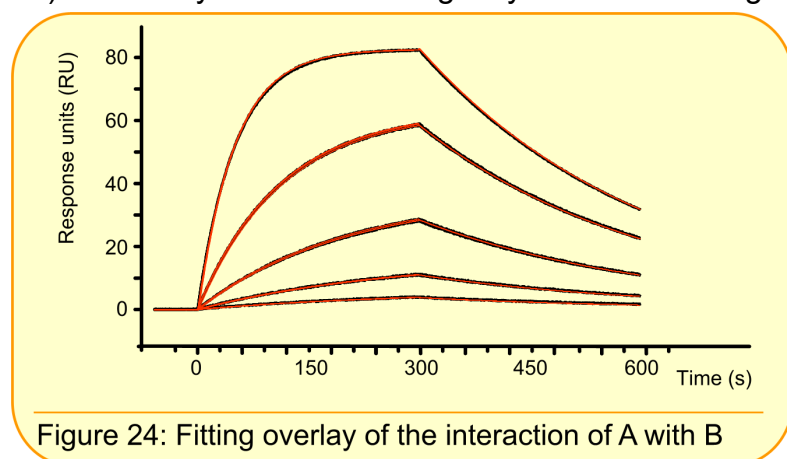


A: yes  
 B: no, dissociation looks faster  
 C: no, dissociation looks slower  
 D: no,  $R_{max}$  is to high

Answer A.

D: you can not tell this because the curves show no saturation probably due to too low analyte concentration.

26) What do you want to change if you see this fitting?



- A: nothing, this looks fine
- B: make association time longer
- C: make dissociation time longer
- D: use higher analyte concentration

Answer A.

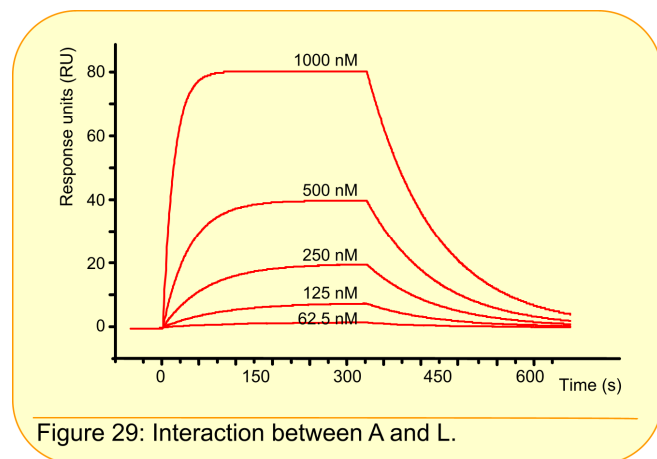
27) To calculate (fit) meaningful results you need curves      Answer C

- A: which go to  $R_{max}$
- B: which go to steady state
- C: which have curvature
- D: with a low response

28) What can you tell about the  $R_{max}$ ?      Answer B

- A: It is dependent on the  $k_a$  and  $k_d$  of an interaction
- B: It is dependent on the surface capacity and molecular weight of ligand and analyte
- C: It is dependent on de analyte concentration
- D: It is dependent on the equilibrium constant  $K_D$

29) What can you say about this sensorgram?



- A: the analyte concentration range is not wide enough.
- B: the response is not following exponential kinetics
- C: there is mass transport limitation
- D: this look a fine sensorgram

Answer D

B: you can't say that the response is following kinetics because the  $K_D$  is unknown and these analyte concentrations can below  $1 \times K_D$ . If you

know the kinetic constants it is possible to deduce how many times the  $K_D$  the analyte concentration is and you can infer of the response is following kinetics.

30) The minimal requirements in a publication are:      Answer D.

- A: sensorgram + fit overlay + kinetic values
- B: sensorgram (replicates) + fit overlay + kinetic values
- C: table with kinetic values and representative sensorgram
- D: full method used in the experiments + B