How to pack a glass column with soft gels

Preparation

a. Material quantity calculation: $V_{bed} = \frac{\pi d^2}{4} * l \qquad V_{material} = V_{bed} * f_{compression}$

b. Note the ratio of settled material and supernatant in the storage vessel.

c. Homogenise the material.

d. Fill the required amount of packing material in a graduated cylinder. Adjust to the ratio in b.

$$Amount_{material} = V_{material} + solvent$$

e. Remove the fines.

f. Wash the packing material with packing solvent.

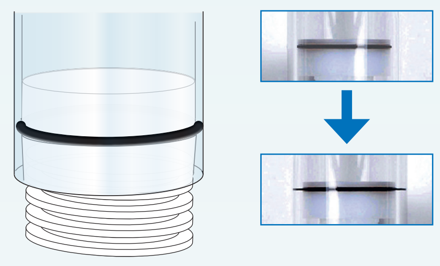
g. Adjust the required slurry concentration.

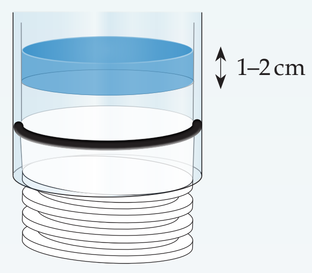
h. Prepare the system and the column by flushing both with packing solvent.

i. Insert the lower piston and tighten the O-ring (if required).

j. Assemble a packing adapter (if required).

k. Flush the pistons to remove air in the frits.





Packing

a. Homogenise the slurry.

b. Pour the slurry down the opposite site of the glass wall in order to avoid the entry of air.

c. Fill the column completely with packing solvent.

d. Insert the piston by putting it in an angle to the glass edge.

e. Close the column.

f. Start the flow stepwise until the required flow rate and packing pressure is reached.

g. Wait until the column bed has formed out and the bed length doesn't change anymore. Mark the bed position under flow. The bed is now consolidated.

h. Remove the packing adapter (if required).

i. Lower the upper piston until you reach the top of the column bed.

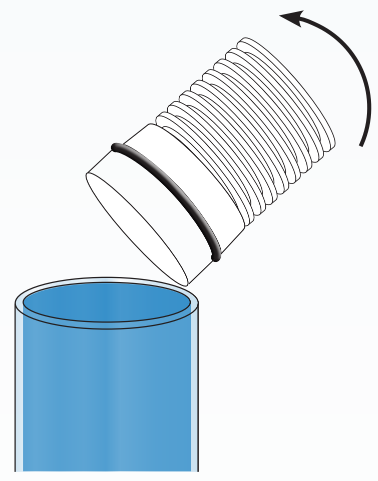
j. Start the flow again stepwise, until you reach the required flow rate and packing pressure.

k. Wait until the bed length does not change anymore. Mark the bed length under flow.

l. Lower the piston and start the flow again. Repeat this until the bed length remains constant.

m. Lower the piston until you reach the last mark (mark under flow). The bed is now compressed.

n. The column is now finally packed.



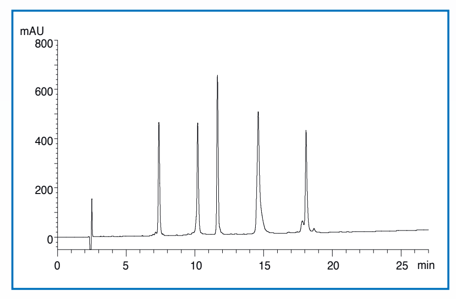
Qualification

a. Equilibrate the column with 5 column volumes.

b. Carry out the qualification corresponding to the manufacturer recommendations or your own guidelines.

c. Repeat the qualification 3 times.

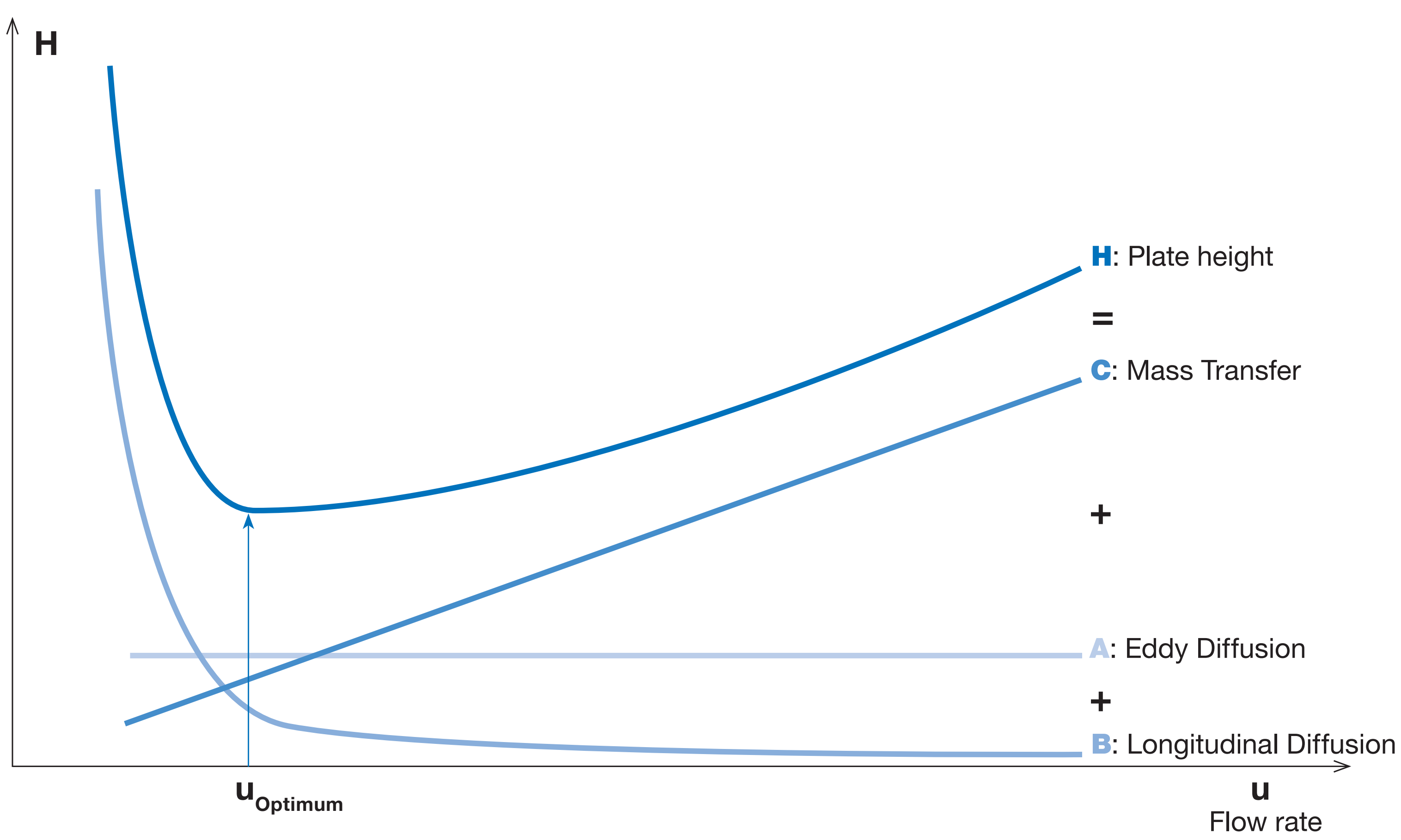
d. Evaluate the qualification. If required, unpack the column and re-pack it.



What impact does the packing quality have on the chromatographic results?

The basis for every chromatographic separation is the correct choice of stationary phase. Only this way can an optimum resolution be achieved. Of course, the packing material needs to be packed into the column as efficiently as possible for robust and reproducible results.

The importance of an ideal packed column bed results from the van Deemter equation. With a homogenously packed column, the peak broadening is reduced. Therefore, the Eddy Diffusion is a direct measure for the packing quality of a column.



$$H = A + \frac{B}{u} + C \cdot u$$

A B C

Where

H = Plate height

A = Eddy Diffusion

B = Longitudinal Diffusion

C = Mass Transfer

u = Flow rate

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