Pneumatic conveying systems and physical stability of monoclonal antibodies: Example of rituximab

V. Vieillard[1], K. Rilcy[1], C. Magneux[2] , A. Bellanger[2], A. Astier[1]*, M. Paul[1],
[1] *: Department of Pharmacy, Henri Mondor Hospital Group and * UMR 7054, School of Medicine, Paris 12, University. Créteil France*
[2] *: Department of Pharmacy, Pitié Salpétrière Hospital Group, Paris, France*

Introduction

Proteins such as monoclonal antibodies (mAb) are sensitive products which could undergo complex degradation pathways during various manipulation steps like transport. Aggregation is the main instability phenomenon induced by mechanical stress. Transports and especially transports with conveying systems are susceptible to induce aggregation and consequently a loss of efficiency and/or toxic effects such as immunogenicity. These pneumatic conveying systems are in place in some hospitals but are not currently used for transport of proteins since no stability data under this specific stress are available.

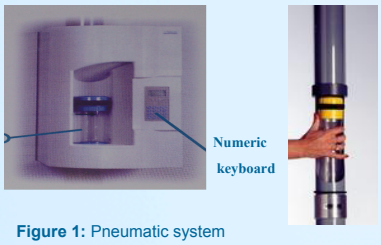


Figure 1: Pneumatic system

Manufacturer's drug information is not useful giving only sentences such as “avoid shaking”.

The objective of this study was to verify if the pneumatic conveying systems could be used to send bags containing the mAb like rituximab to clinical services.

Materials and methods

- Rituximab (RTX) was diluted in NaCl 0,9% to obtain a concentration of 1 mg/ml.
- One batch of bags (Freeflex® polyolefin bags 50 ml, Fresenius kabi) were prepared at the Department of Pharmacy, Pitié Salpétrière Hospital Group.
- Several conditions were tested:
  - ➢ presence or absence of air
  - ➢ travel time
  - ➢ number of routes (1 to 8)
- All experiments were conducted the same day.
- Various protein characterization methods were used to determine changes in physical properties of Rituximab such as mechanically induced aggregation :
  - ➢ size exclusion chromatography (SEC)
  - ➢ dynamic light scattering (DLS) describing submicronic populations and corresponding mean diameters (md)
  - ➢ turbidity (at 350 nm)
  - ➢ infrared spectroscopy

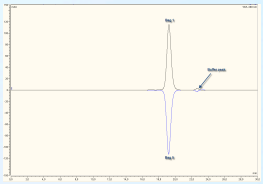
Results and Discussion

Without air

Up to 8 routes and without air into the bags, no modification was observed in comparison with the control (no route):

Figure 2: only one peak in SEC with a retention time of 19.18 ± 0.03 min and an AUC of 71.51 ± 1.83 mAU*min.

A second peak at 22.7 min was observed for all conditions corresponding to the buffer.



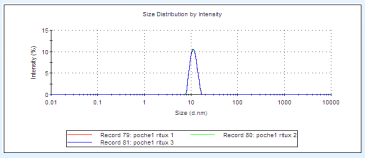


Figure 3: A monodisperse repartition was observed (polydispersity indexes were inferior to 0.1) with only one population md = 11.34 ± 0.03 nm in DLS.

Table 1: Just a slight increase of optical densities (0.0019 to 0.004) is observed, nevertheless optical densities remained low.

		350nm
Rituximab	Bag 1	0,0019 ± 0,0001
	Bag 2	0,0027 ± 0,0006
	Bag 3	0,0035 ± 0,0001
	Bag 4	0,0035 ± 0,0005
	Bag 5	0,0043 ± 0,0002

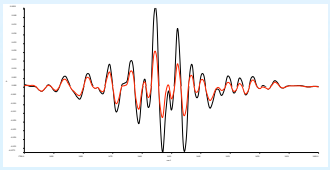


Figure 4: Derivated spectra for bags 1 (red) and 2 (black)

No modification of the FT-IR spectra was observed. Similarity coefficients were close to 1. Even after 8 passages, no modification was observed.

In presence of air, significant modifications were found after 4 cycles.

Table 2: For bags with air, an increase in optical densities was also observed (0.0053 to 0.0074).

		350nm
Rituximab	Bag 6	0,0053 ± 0,0002
	Bag 7	0,0062 ± 0,0002
	Bag 8	0,0057 ± 0,0001
	Bag 9	0,0063 ± 0,0002
	Bag 10	0,0074 ± 0,0004

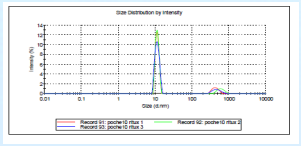


Figure 5: 2 populations were found by DLS with a polydispersity index of about 0.24 and with the presence of a second population md= 499 ± 91.54 nm in DLS.

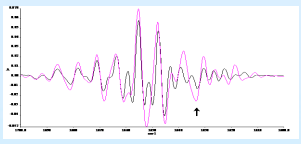


Figure 6: Derivated spectra for bags 6 (black) and 10 (pink)

We observed important modifications between spectra without a route and spectra after 8 routes for bags with head space. (similarity coefficient<0.9)

Conclusion

In practice these preliminary results encourage us to use a pneumatic system for transport of diluted rituximab without air (and probably other monoclonal antibodies).

However, the presence of air into the bags must be avoided because aggregation during the pneumatic transportation is strongly dependant on the presence of air/liquid interfaces.