Aspen simulation

# Gel filtration(GF)

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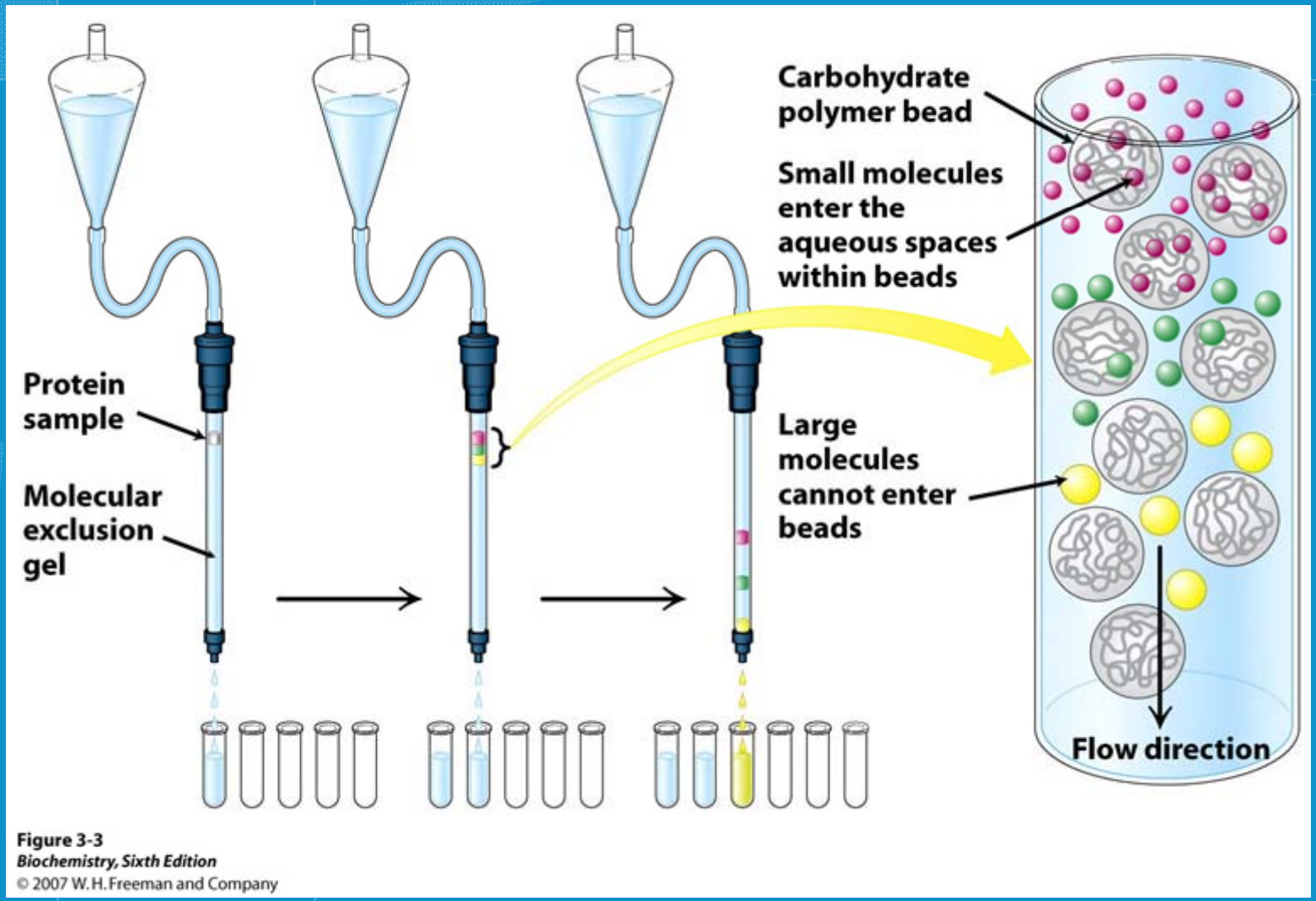
# GF (Gel Filteration)

- 겔을 운반체(캐리어)로 사용하여 측정하고자 하는 고분자용액을 주입하면 분자량이 작은 분자는 겔이 이루고 있는 그물에 잘 흡착되고, 분자량이 커질수록 잘 흡착되지 않는다.

- 유출시간의 차이에 의해서 고분자의 분자량 분포 측정이나, 분자량에 따른 분별에 사용된다.


Carbohydrate polymer bead
Small molecules enter the aqueous spaces within beads
Protein sample
Molecular exclusion gel
Large molecules cannot enter beads
Flow direction
Figure 3-3
Biochemistry, Sixth Edition
© 2007 W.H. Freeman and Company

[그림. 1] GF의 원리

# BSA & NaCl Properties

## BSA (Bovine serum albumin)

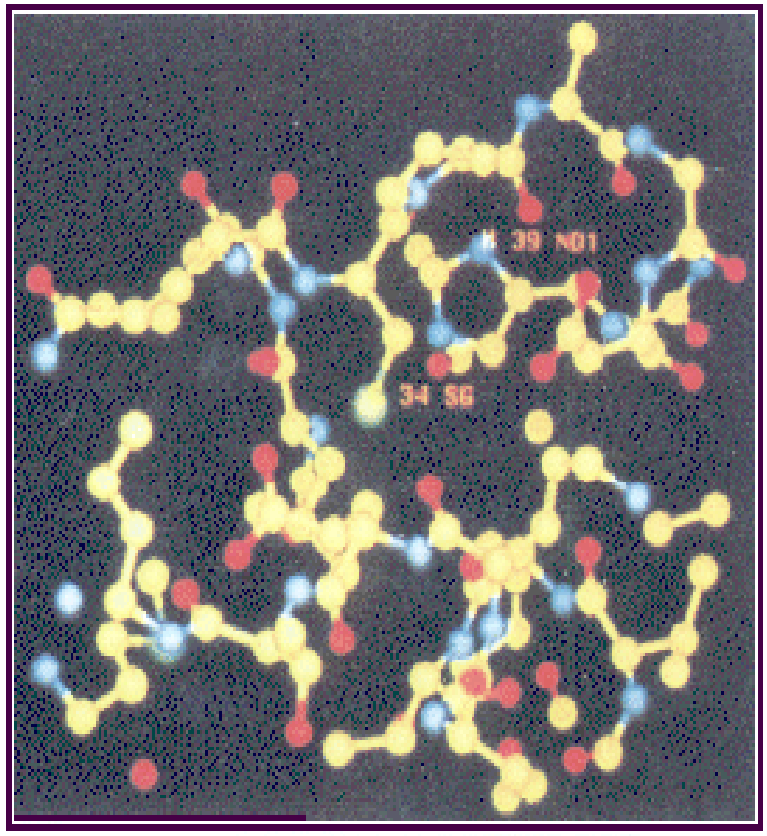
- 소 혈청 알부민
- 분자량 : 약 66.4 kDa
- 종류 : 단백질 (대부분의 동물에 많이 존재)
- 용도

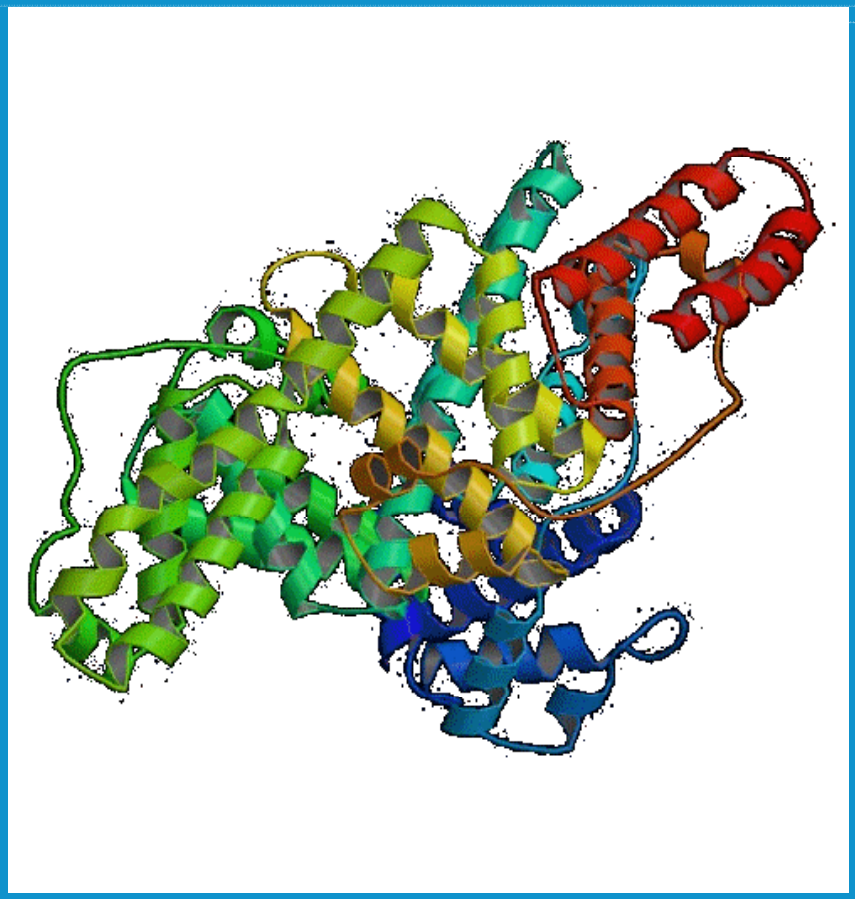
- 세포 배양시에 세포의 영양분
- 단백질 정량에서 검정곡선을 얻기 위한 표준물
- 생화학적 실험에서 특정 항체를 검출 하고자 하는 단백질이 붙여주기 전에 nonspecific binding에 항체가 달라 붙는 비특이적 결합을 막아 주기 위해 사용

## NaCl

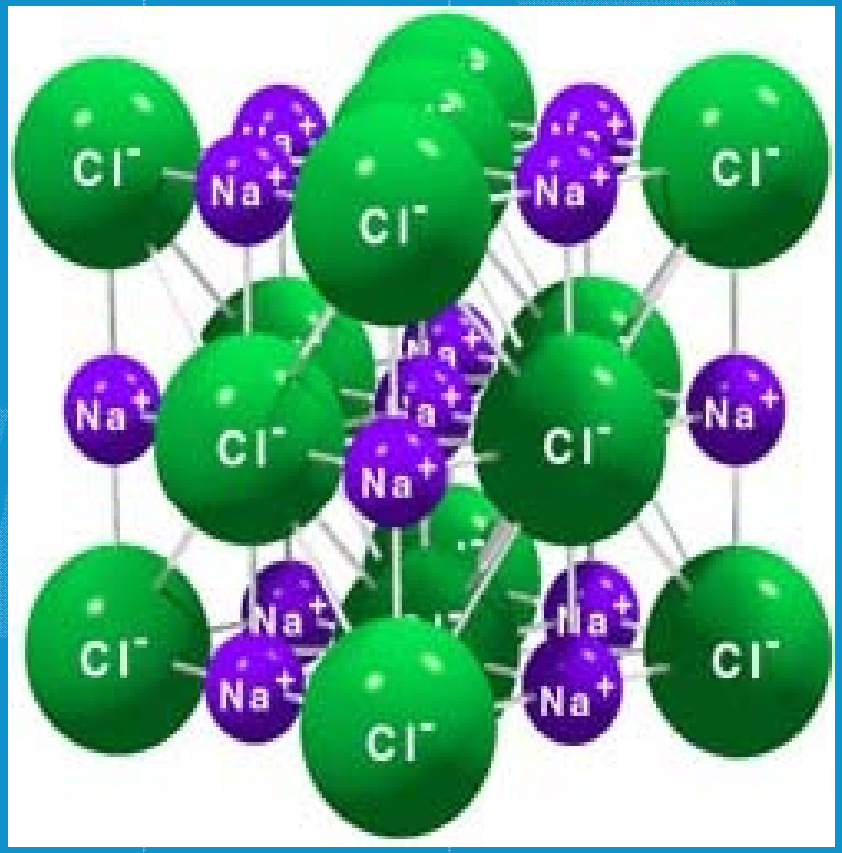
- 소금
- 분자량 : 58.44 Da
- 용도

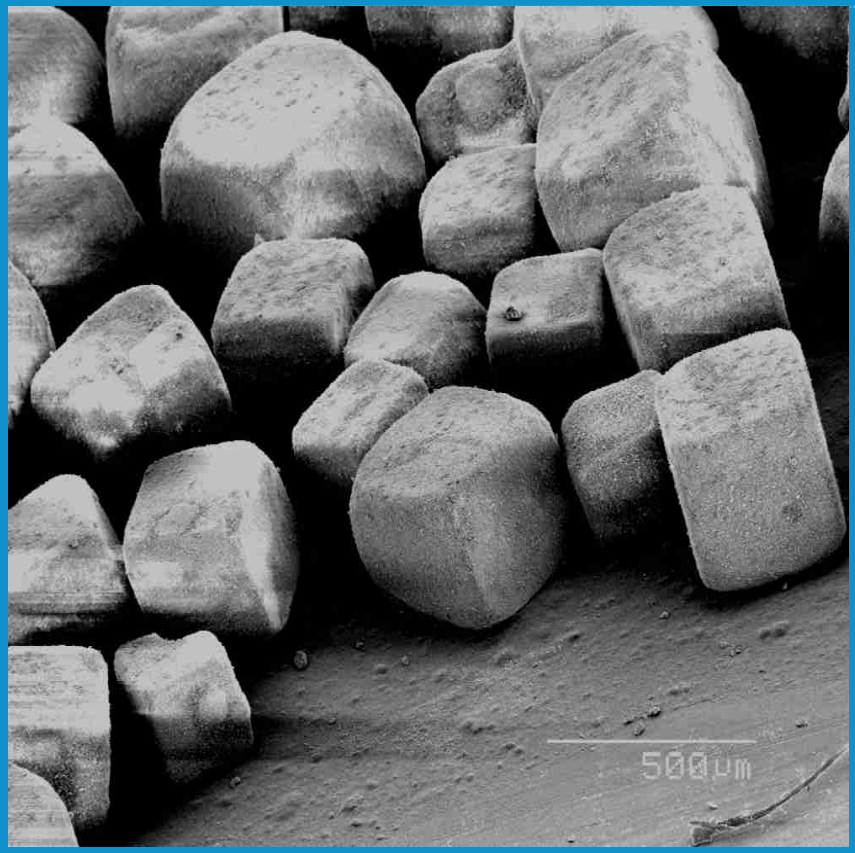
- 조미, 염장 등의 일상 생활과 공업 방면에서 사용
- 동물의 체내 삼투압의 유지





[그림. 2] BSA의 분자 구조와 3차원 리본 구조




[그림. 3] NaCl의 화학 구조와 현미경으로 본 NaCl

# Column


HiTrap Desalting Column 5ml

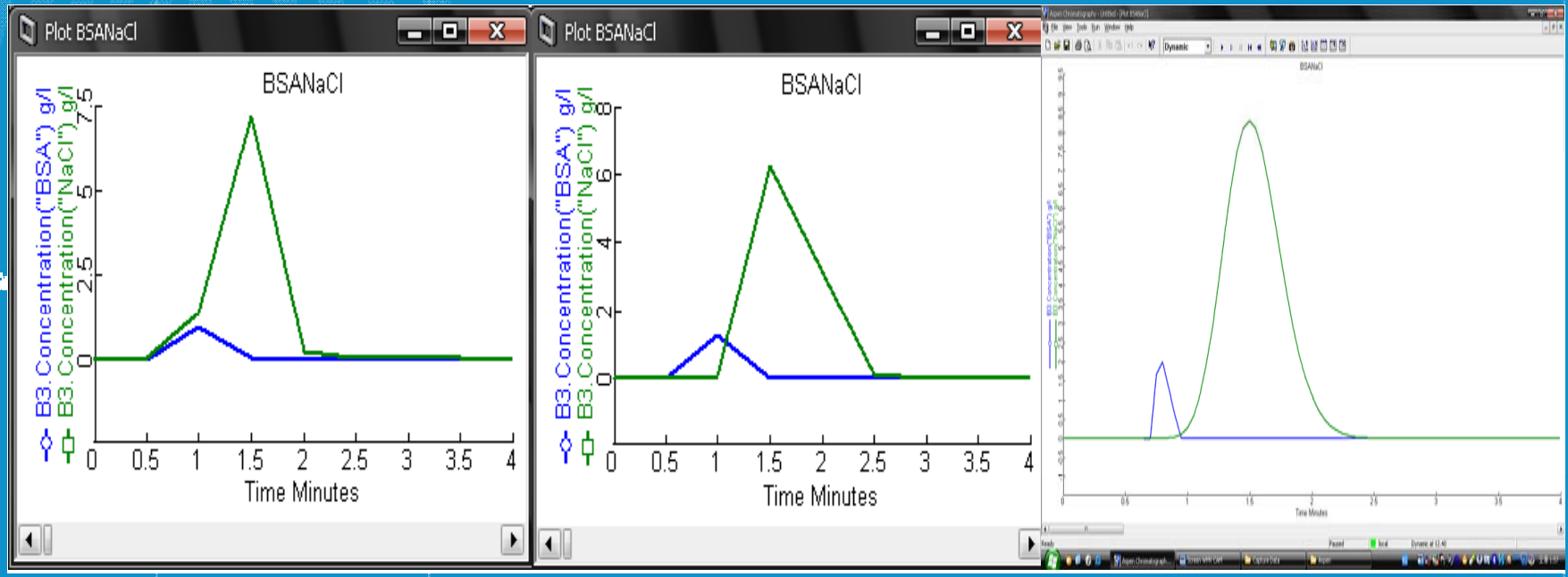
| Column volume | 5 ml |
|---|---|
| Medium | Sephadex™ G-25 Superfine |
| Column dimensions | **1.6 × 2.5 cm** |
| Sample volume | 0.25-1.5 ml |
| Max. flow rate ($H_2O$ at 25°C) | 15 ml/min |
| Recommended flow rate | 1-10 ml/min |
| Max. pressure | 3 bar (43 psi, 0.3 MPa) |

# Experimental Conditions

- Column : HiTrap Desalting 5ml
- Sample : Bovine serum albumin (BSA) 2mg/ml
  0.5 M sodium chloride (NaCl)
  0.05 M sodium phosphate
  pH 7.0
- Buffer : 0.05 M sodium phosphate
  0.15 M sodium chloride
  pH 7.0
- Sample volume : 0.8 ml
- Flow : 5 ml/mim

# IP Change

| | 1차 | 2차 | 3차 (최종값) |
|---|---|---|---|
| BSA IP1 | 1.0 | 0.5 | 0.1 |
| NaCl IP1 | 2.0 | 3.0 | 2.5 |
| BSA IP2 | 0.0 | 0.0 | 0.0 |
| NaCl IP2 | 0.0 | 0.0 | 0.0 |



# Simulation Video

http://blog.naver.com/rapy1004/150075292689

# Simulation Video - IP change

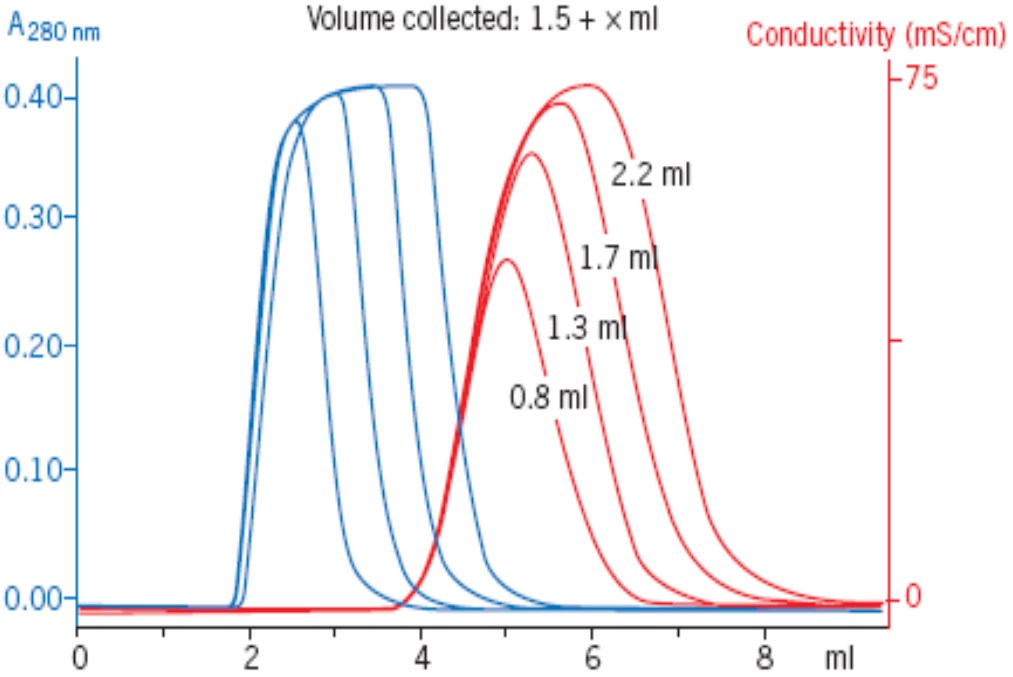
http://blog.naver.com/rapy1004/150075491308

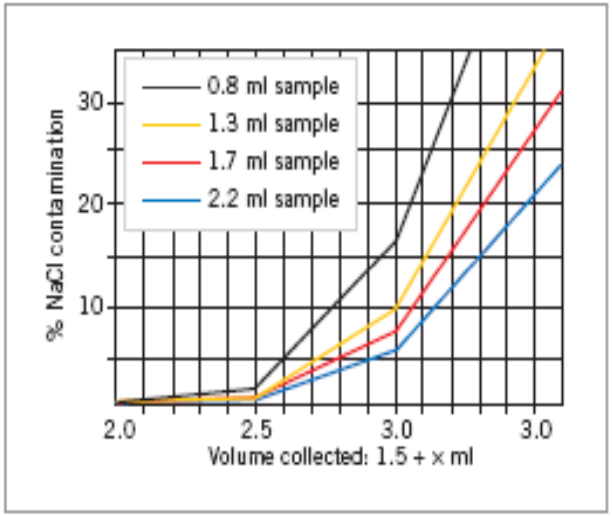
# Aspen Simulation Variables

- Sample Volume
  - 0.8, 1.3, 1.7, 2.2 ml
    - Column Volume : 5 ml
    - Flow : 5 ml/min
- Flow
  - 1.7, 3.3, 6.7, 10.0, 16.7, 20.0 ml/min
    - Column Volume : 5 ml
    - Sample Volume : 0.8 ml
- Scale up
  - HiTrap Desalting 1x5 ml, 3x5 ml, 5x5 ml
    - Sample : 28% x $V_t$ (1.4, 4.3, 7.1 ml respectively)

# Sample Volume

*Column*: HiTrap Desalting 5 ml
*Sample*: Bovine serum albumin, 2 mg/ml in 0.5 M NaCl, 0.05 M sodium phosphate, pH 7.0
*Buffer*: 0.05 M sodium phosphate, 0.15 M NaCl, pH 7.0
*Sample volume*: 0.8, 1.3, 1.7, 2.2 ml
*Flow*: 5 ml/min

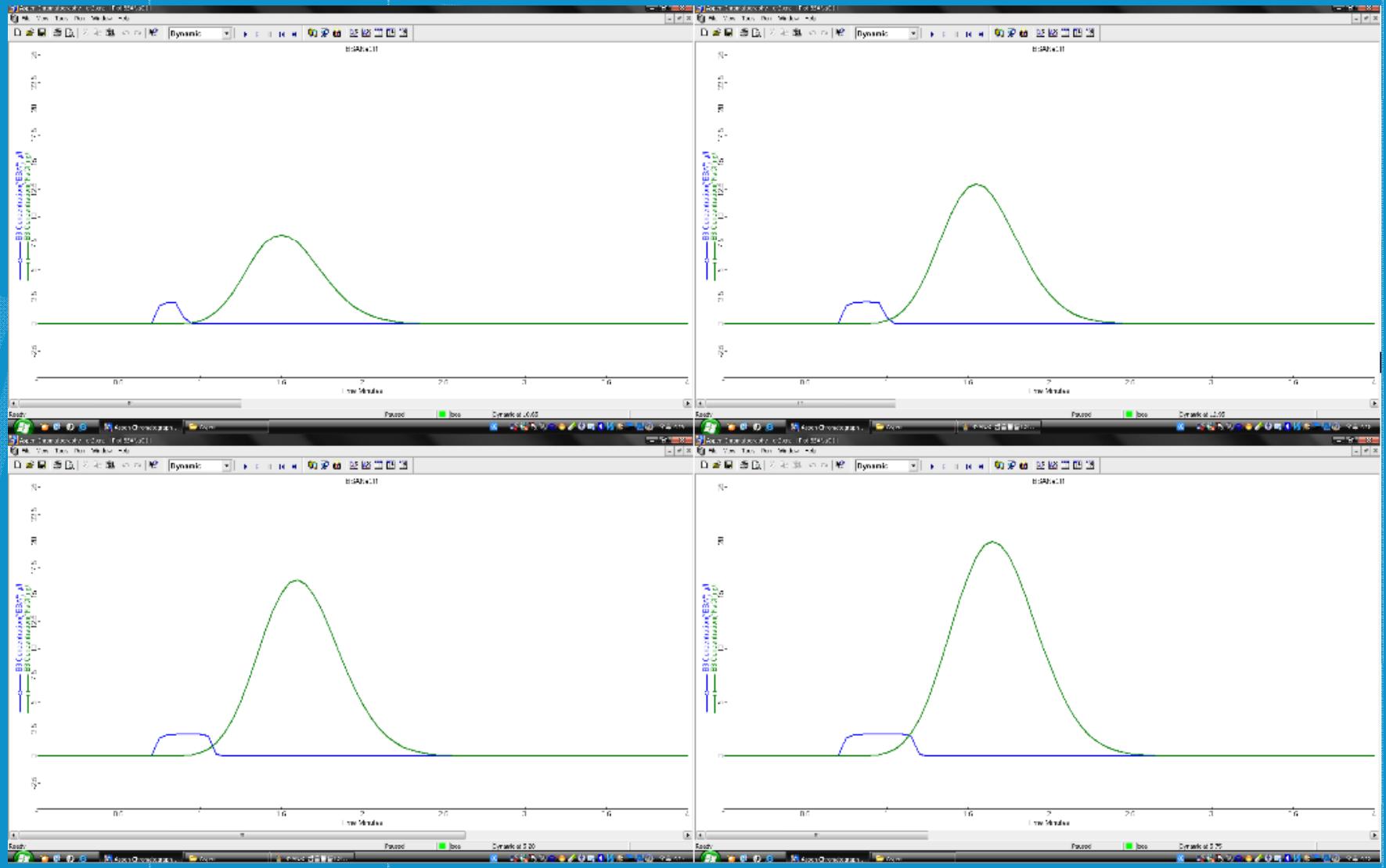


# Simulation Video - Sample Volume

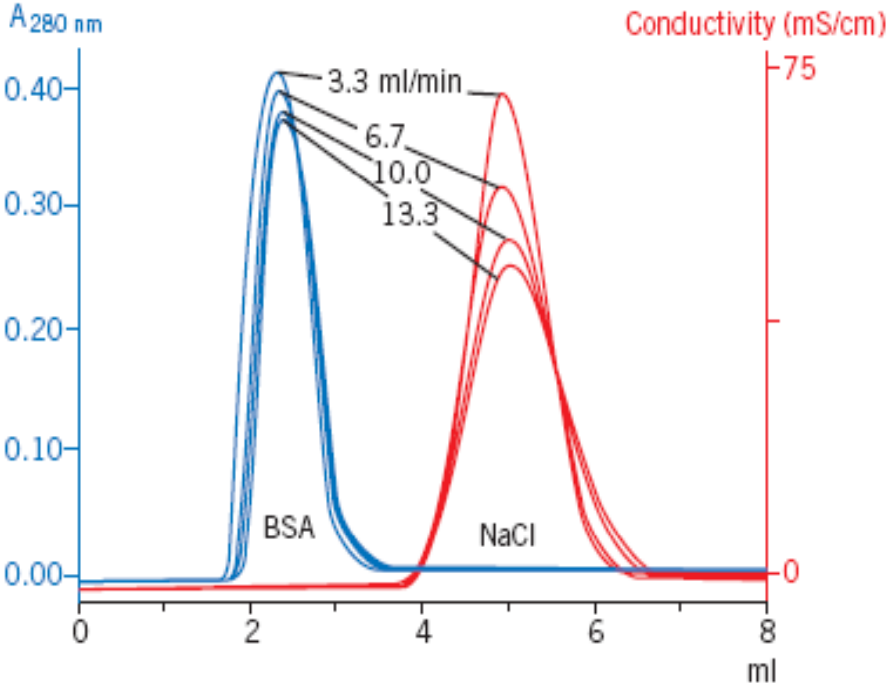
http://blog.naver.com/rapy1004/150075491442

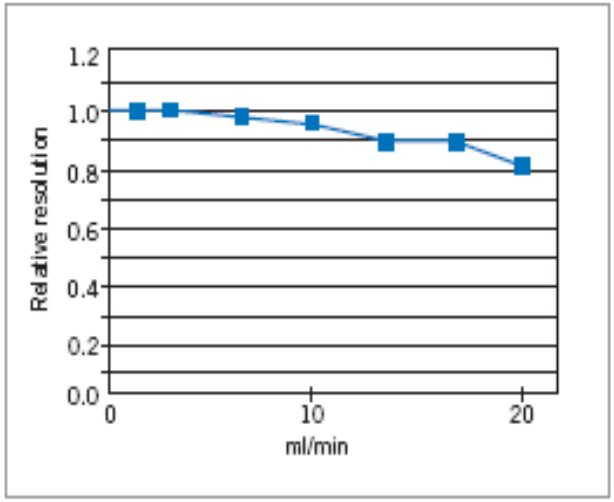
# Sample volume change



# Flow

*Column*: HiTrap Desalting 5 ml
*Sample*: Bovine serum albumin, 2 mg/ml in 0.5 M NaCl, 0.05 M sodium phosphate, pH 7.0
*Buffer*: 0.05 M sodium phosphate, 0.15 M NaCl, pH 7.0
*Sample volume*: 0.8 ml
*Flow*: 1.7, 3.3, 6.7, 10.0, 13.3, 16.7, 20.0 ml/min

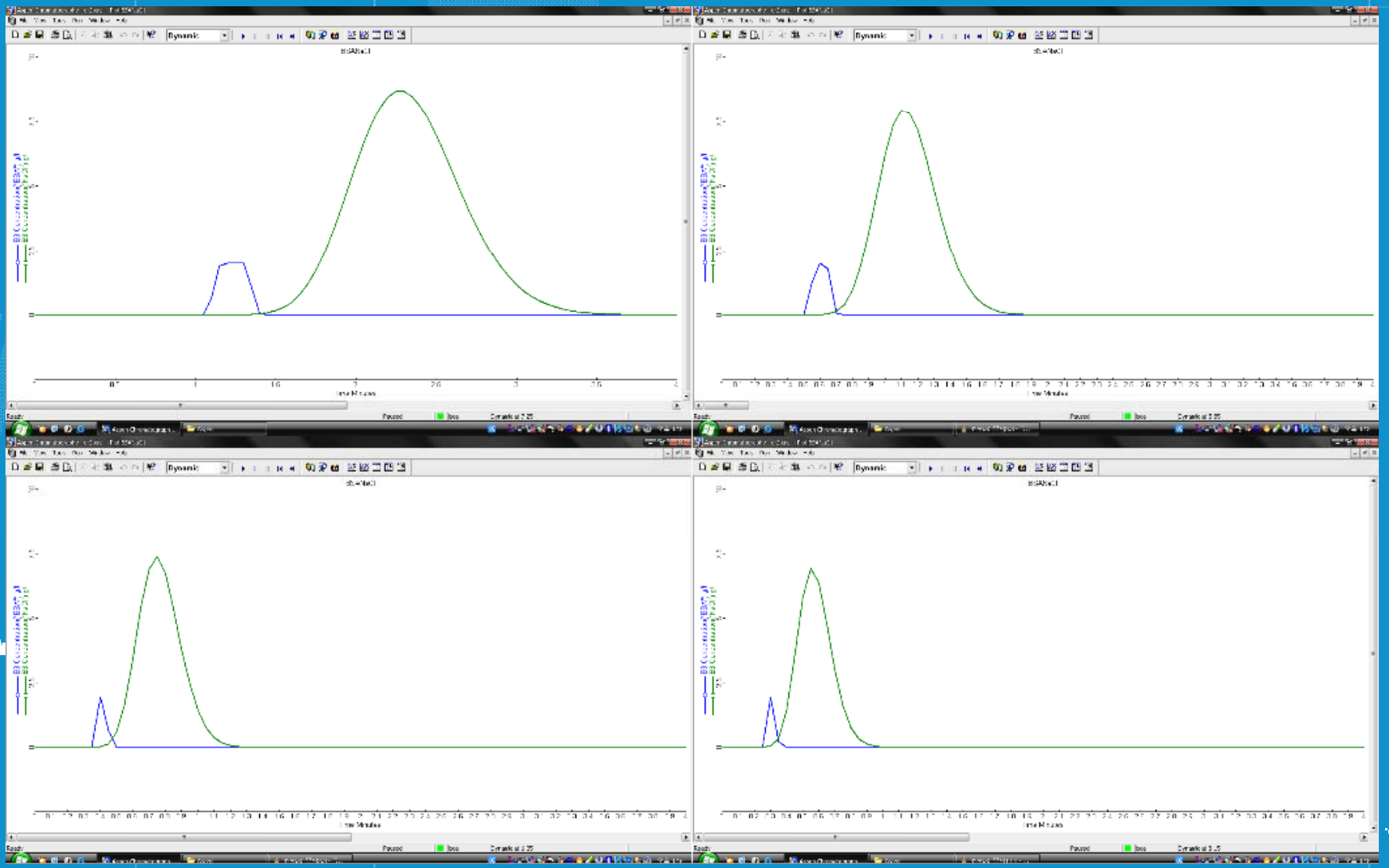


# Simulation Video - Flow

http://blog.naver.com/rapy1004/150075491634

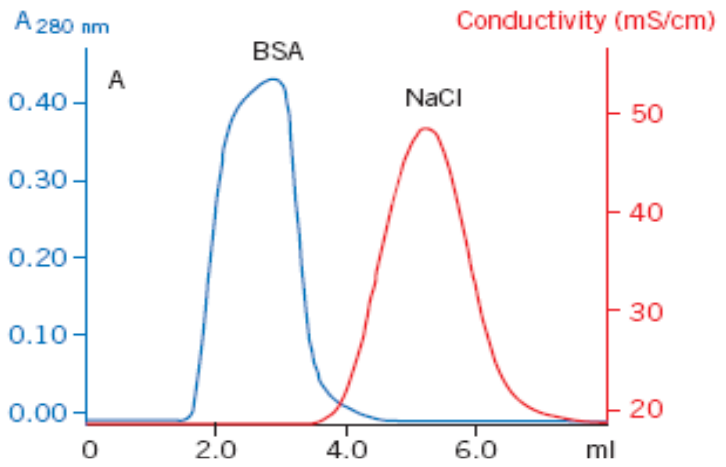
# Flow rate change



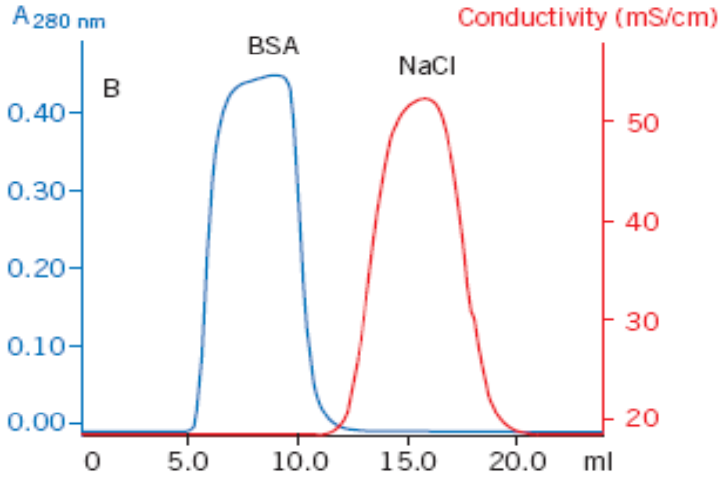
# Scale up

*Column:* HiTrap Desalting, 1 × 5 ml, 3 × 5 ml, 5 × 5 ml
*Sample:* 2 mg/ml BSA in 50 mM sodium phosphate, 0.5 M sodium chloride, pH 7.0
*Sample volume:* 28% × $V_t$ (1.4, 4.3 and 7.1 ml respectively)
*Buffer:* 50 mM sodium phosphate, 0.15 M sodium chloride, pH 7.0
*Flow:* 5 ml/min

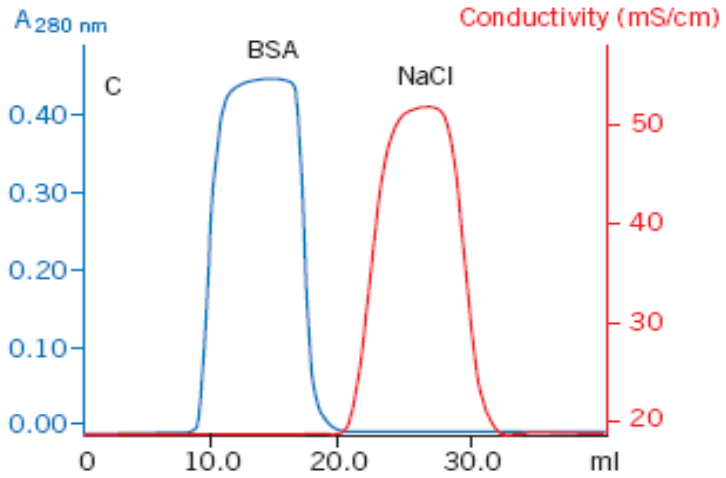
HiTrap Desalting 1 × 5 ml




HiTrap Desalting 3 × 5 ml in series



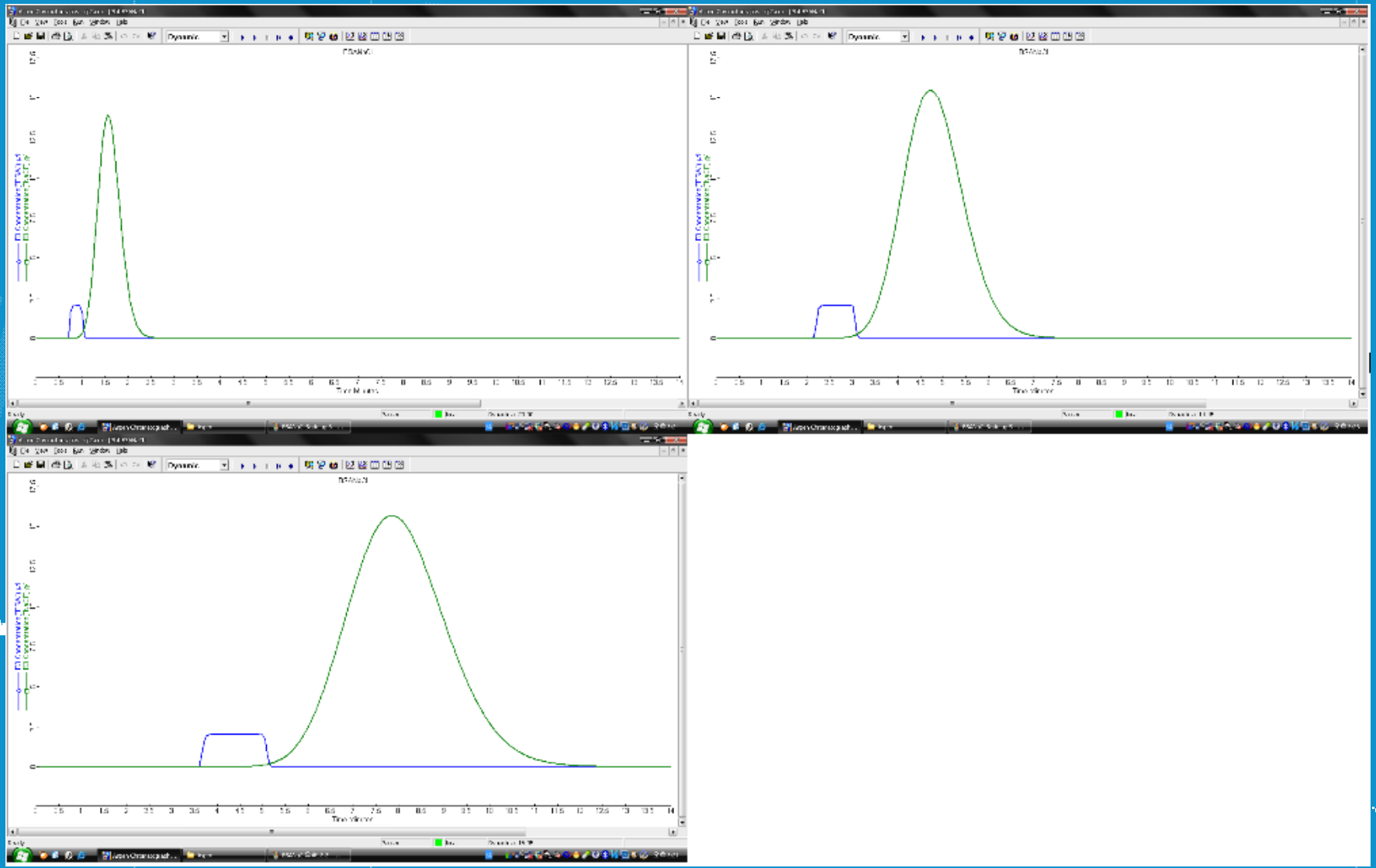

HiTrap Desalting 5 × 5 ml in series



# Simulation – Scaleup

http://blog.naver.com/rapy1004/150075491739

# Scale up



# Conclusions

- 체류시간 및 용출시간과 관련된 변수는 IP, Column Volume, Flow
- 그래프 넓이 및 높이와 관련된 변수는 Sample Volume