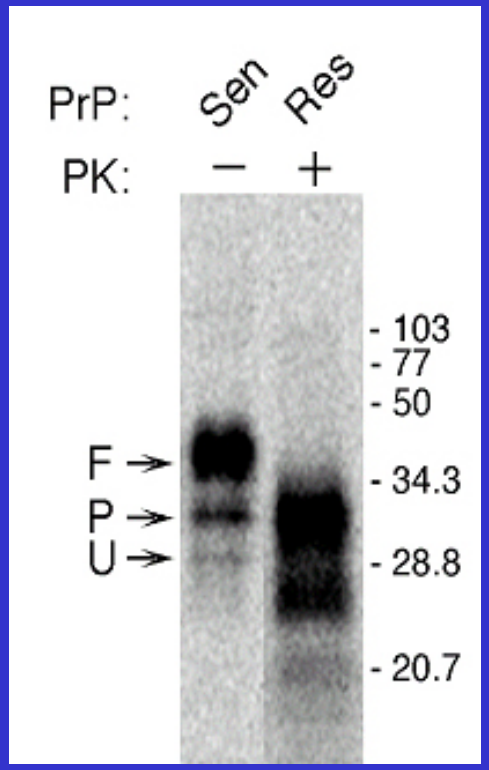
Prion Protein (PrP) and TSE Disease

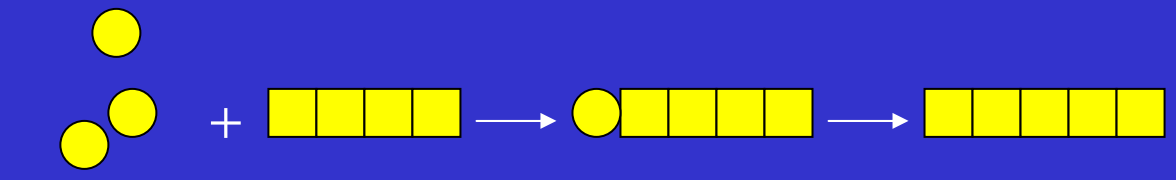


Normal PrP (PrP-sen)

- Proteinase K sensitive
- Detergent soluble
- Expressed in many different tissues
- Primarily alpha-helix/ loop structure

Abnormal PrP (PrP-res)

- Proteinase K resistant
- Detergent insoluble, aggregated
- TSE specific (CNS, LRS)
- Mostly beta-sheet structure



PrP-sen PrP-res

Accumulation and disease

Susceptibility of different cell types to TSE infection

Susceptible Cell Lines

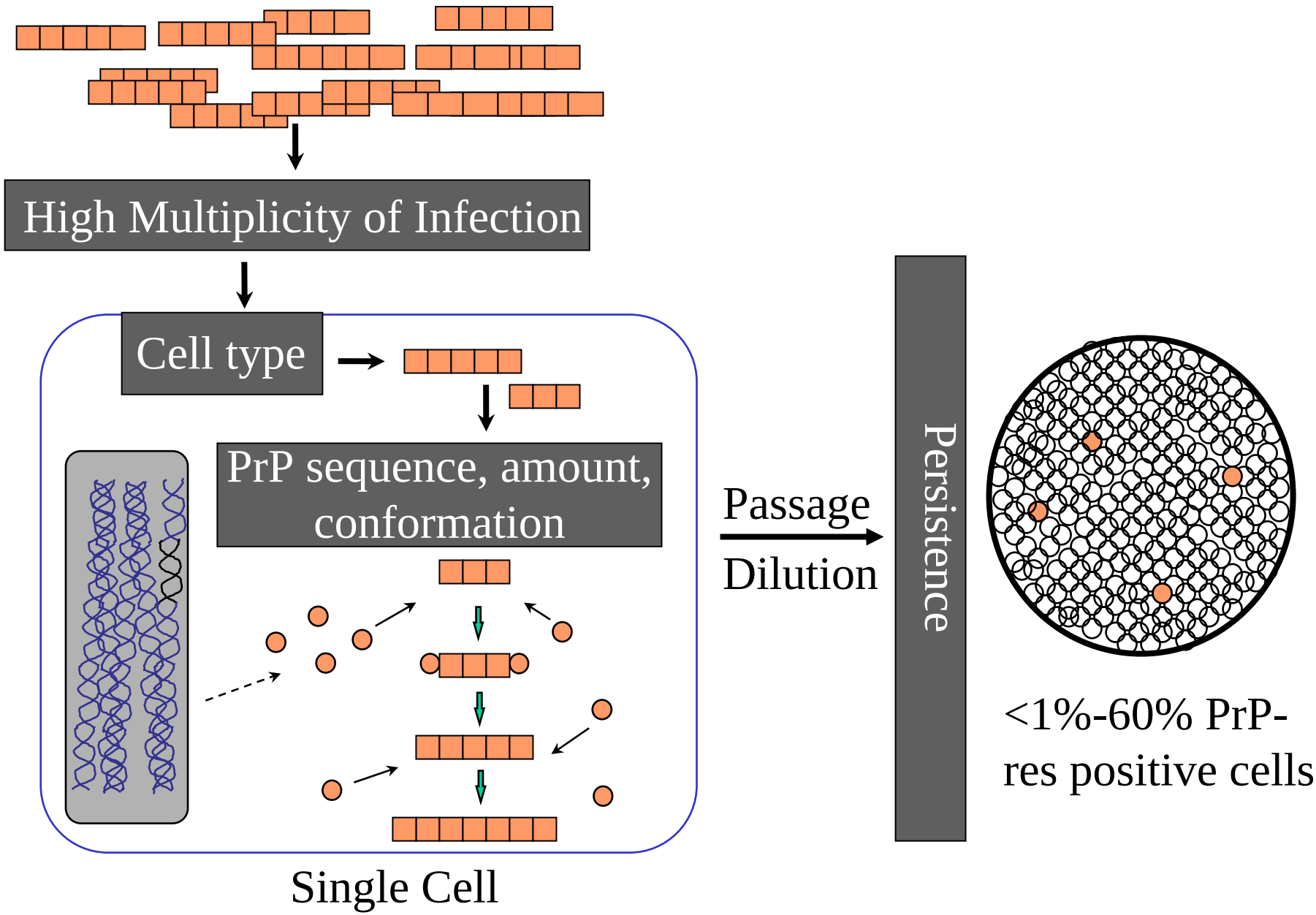
- Neuronal Cells
  - Mouse neuroblastoma (N2A, MNB)
  - Mouse hypothalamic (GT-1)
  - Rat pheochromocytoma (PC-12)
  - Hamster or mouse brain cells
- Non-Neuronal Cells
  - Mouse L-fibroblasts
  - Rabbit kidney epithelial (modified)
  - Mouse Schwann (MSC-80)

Non-Susceptible Cell Lines

- Neuronal Cells
  - Human neuroblastoma (p12-3)
  - Human glioma (HJC-15)
- Non-Neuronal Cells
  - Human embryo lung fibroblasts (WI-38, MDC-5)
  - Human embryo brain fibroblasts
  - Human embryo kidney
  - Chinese Hamster Ovary
  - AGMK (BSC-1)

→ So far no human or bovine cell lines

TSE infection of tissue culture cells is often inefficient



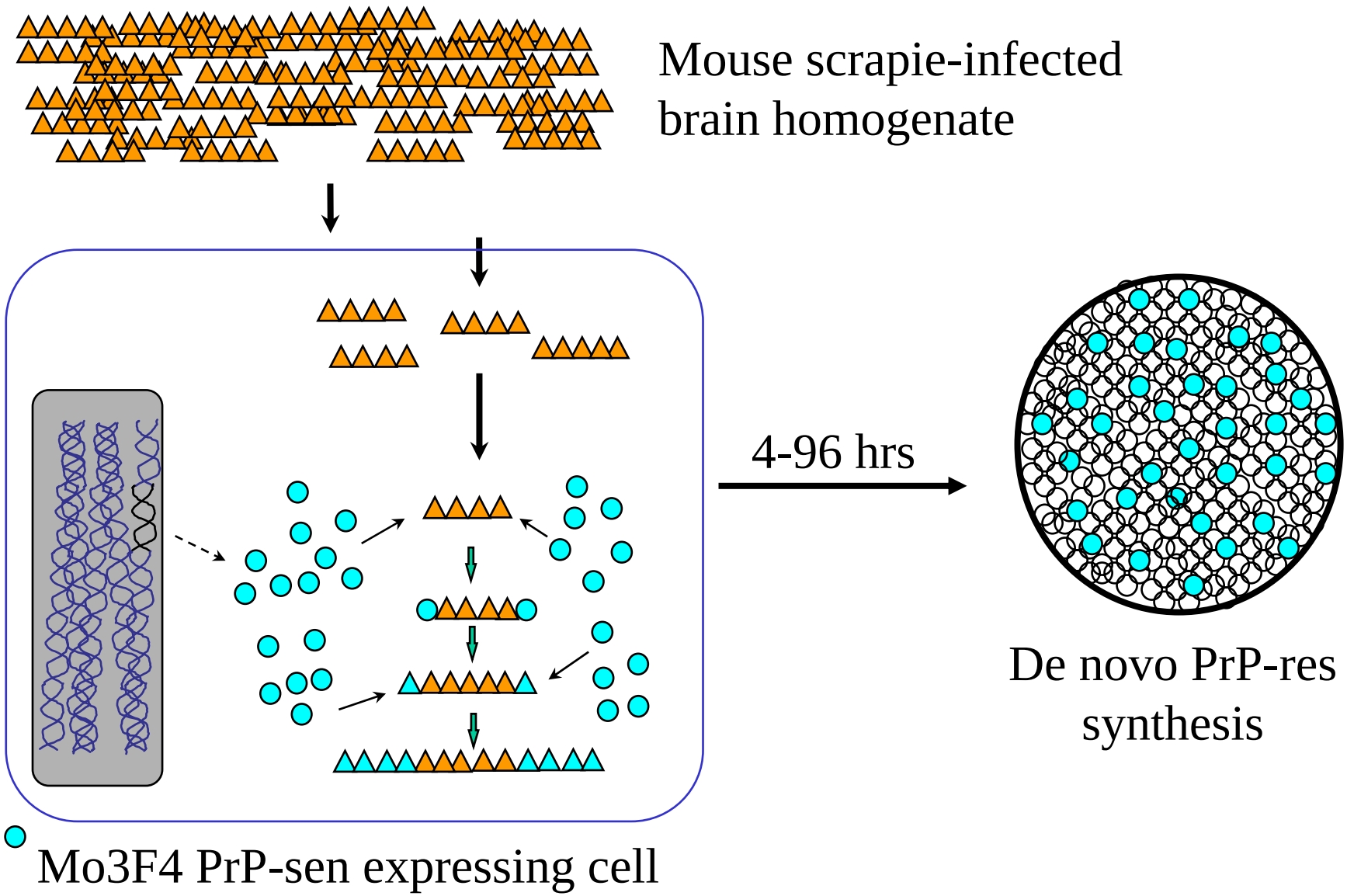
Possible factors influencing susceptibility of tissue culture cells to TSE infection

- Cell type (neuronal vs non-neuronal)
  - Ability of cell to support PrP-res formation
- Strain and species of TSE infectivity
- PrP-sen expression

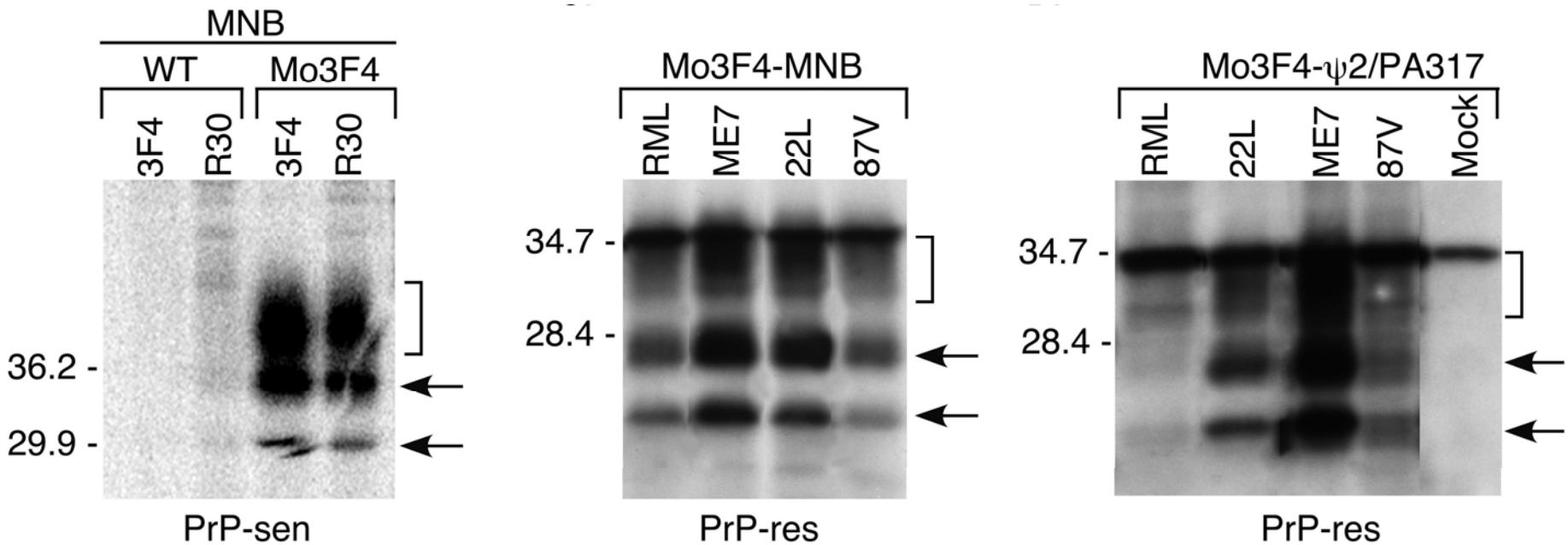
a) Expression level

b) Amino acid sequence (species specificity)

Overlay assay for de novo PrP-res formation

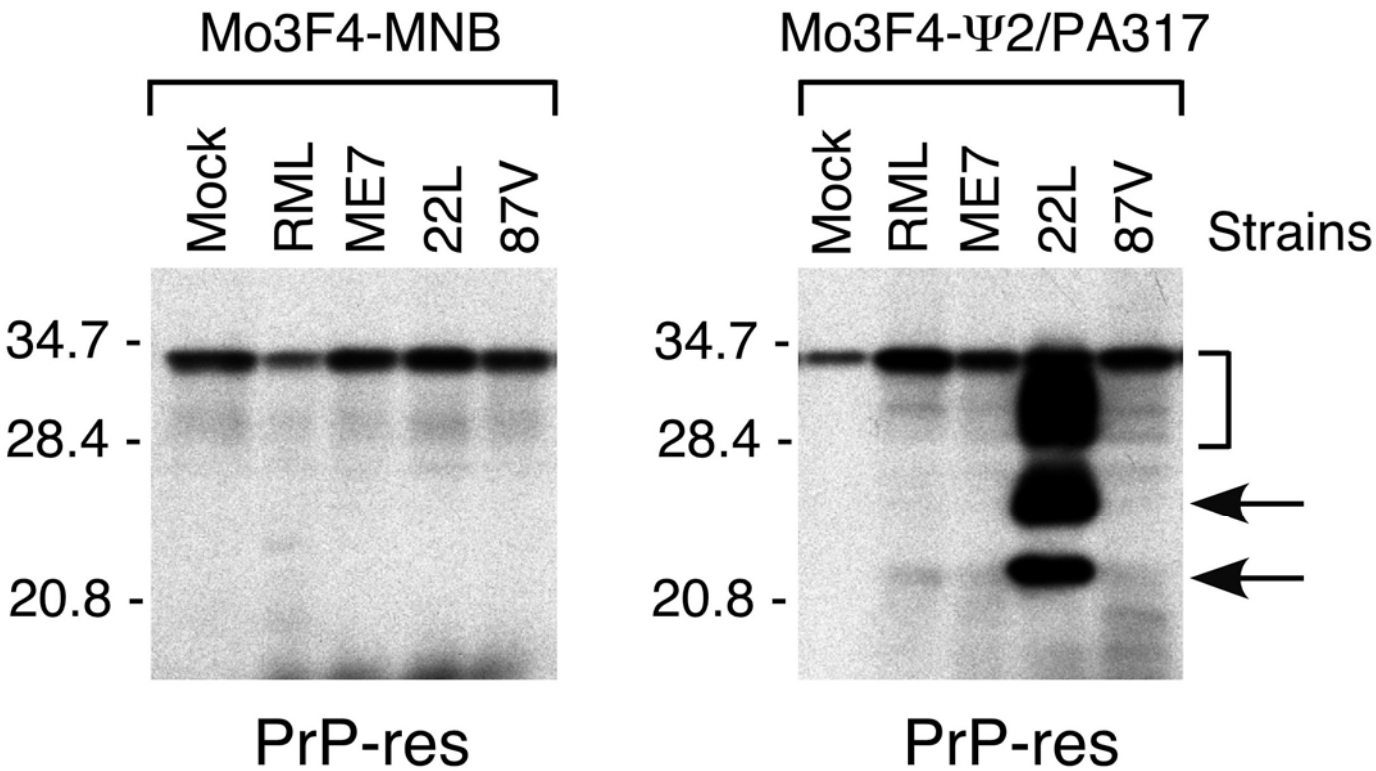


Both neuronal and non-neuronal cells can support PrP-res formation



J. Biol. Chem. 279:29218 (2004)

Acute PrP-res formation does not necessarily lead to persistent PrP-res formation



→ Fibroblast cells are susceptible to TSE infection

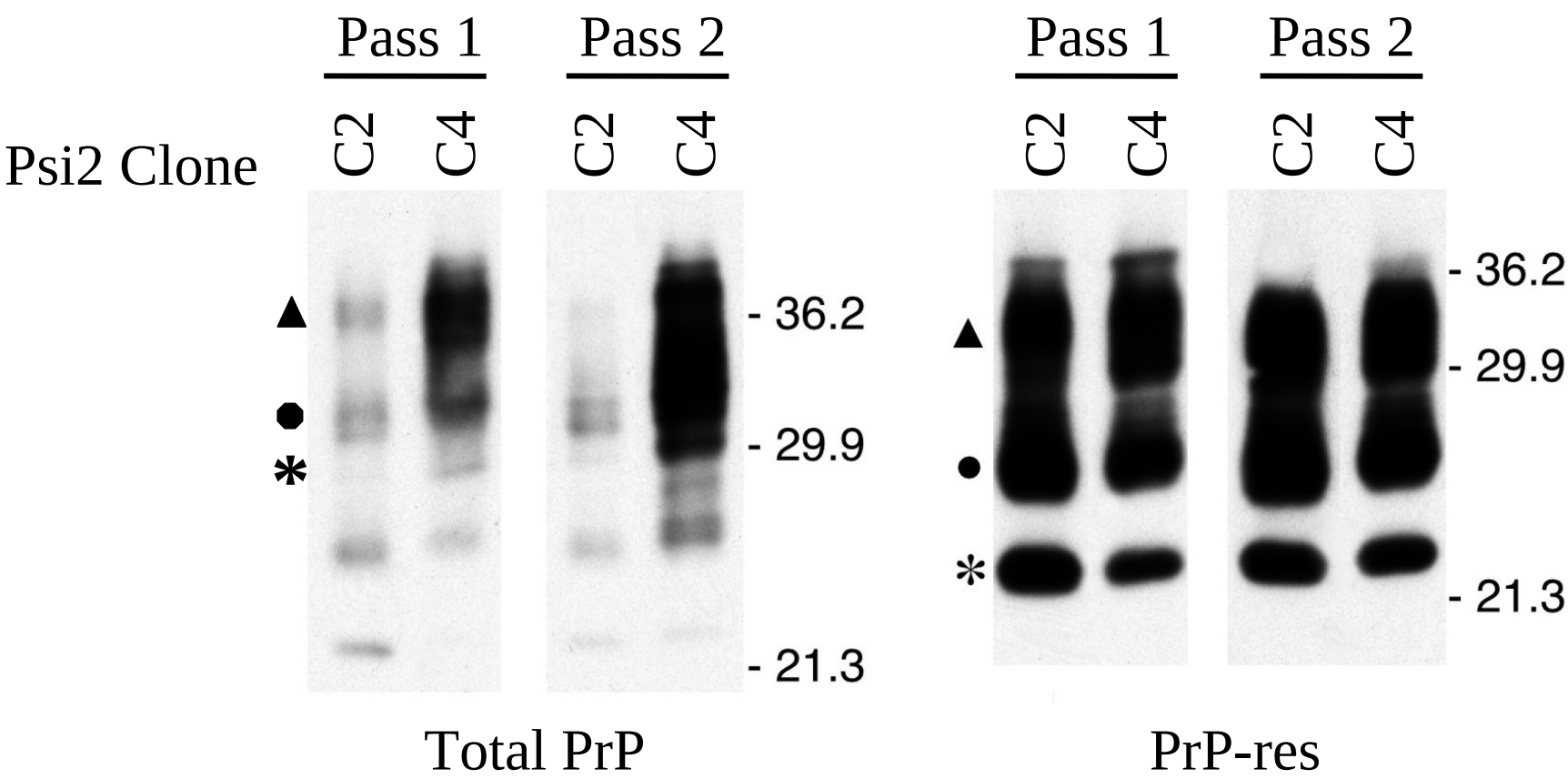
Mouse bioassay of cells exposed to TSE infectivity

Mouse Cell Type

Strain	Mo3F4-MNB	Mo3F4-Psi2/PA317
22L	0/6 (>755dpi)	8/8 (285±40dpi)
RML	0/4 (>755dpi)	1/7 (624, >753dpi)
ME7	0/7 (>755dpi)	0/5 (>753)
87V	0/4 (>755)	0/6 (>755)

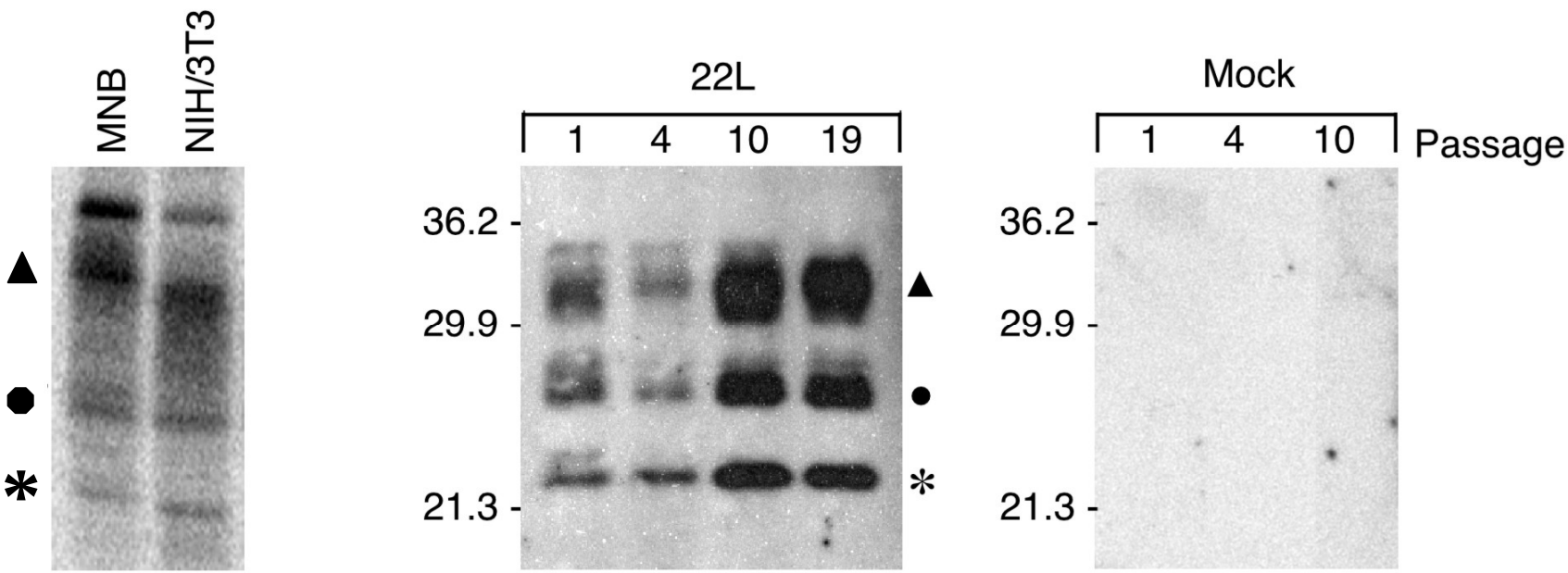
J. Biol. Chem. 279:29218 (2004)

PrP-sen expression level does not predict the susceptibility of a cell to TSE infection



J. Infect Dis. 189:431 (2004)

NIH 3T3 mouse fibroblast cells can be persistently infected with mouse scrapie

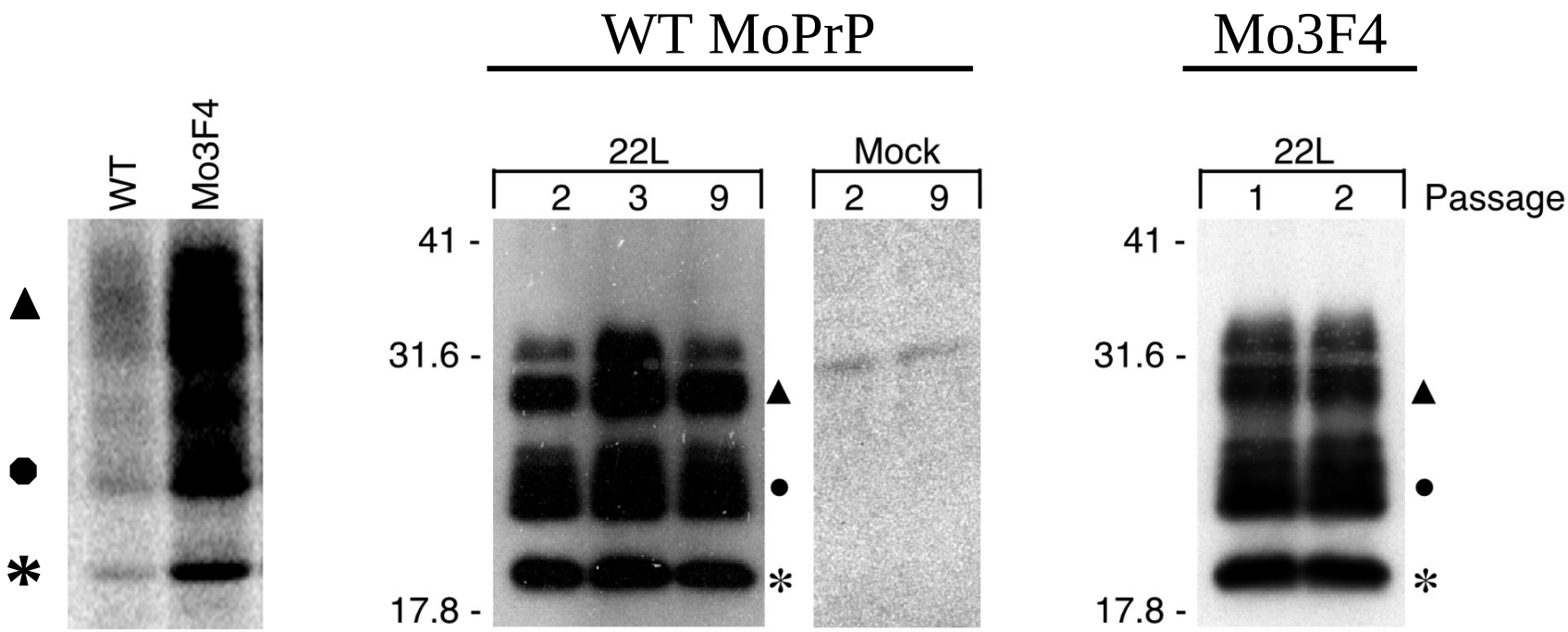


PrP-sen

PrP-res

J. Infect Dis. 189:431 (2004)

Mouse L929 fibroblast cells can be persistently infected with mouse scrapie

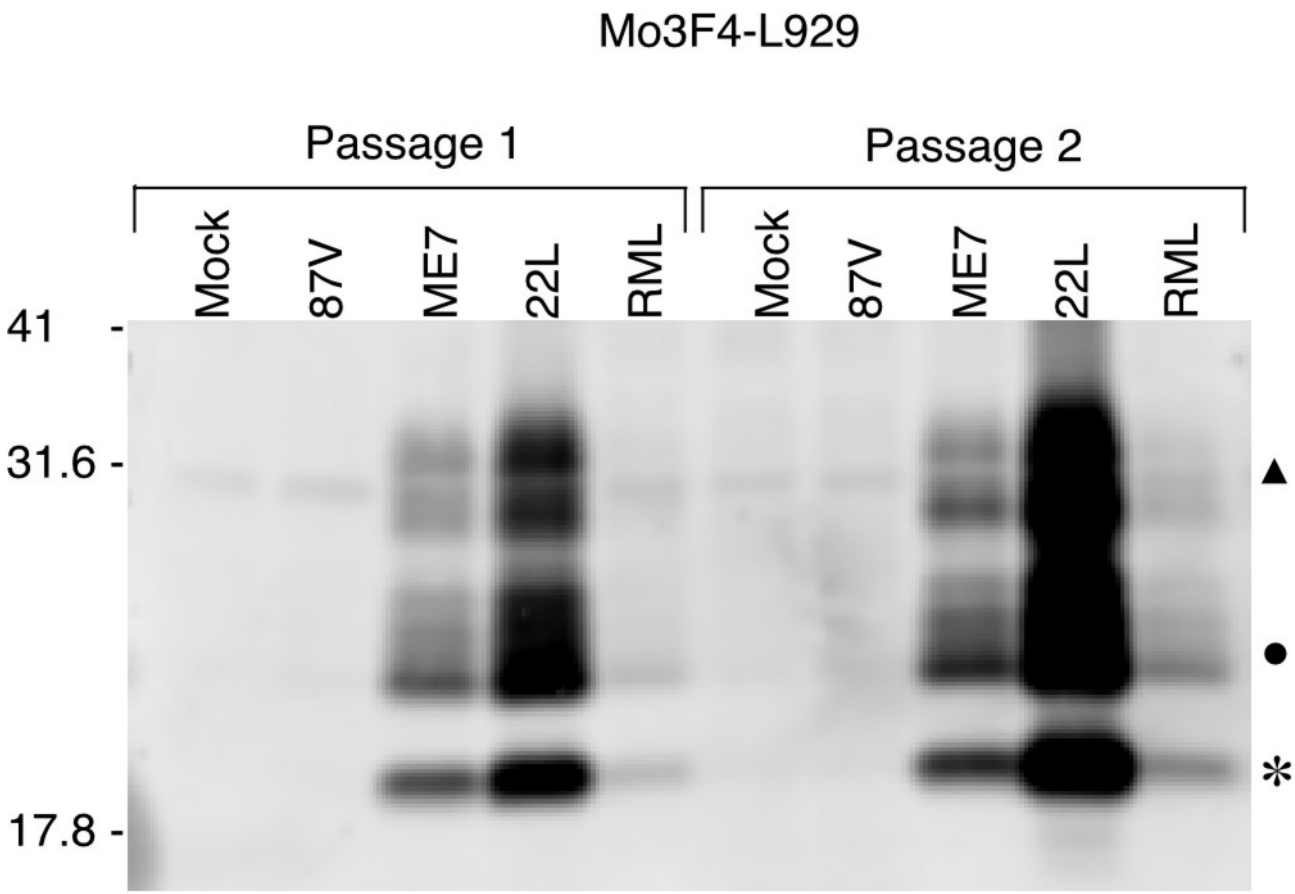


PrP-sen

PrP-res

J. Infect Dis. 189:431 (2004)

Multiple mouse scrapie strains can persistently infect mouse L929 fibroblasts



PrP-res

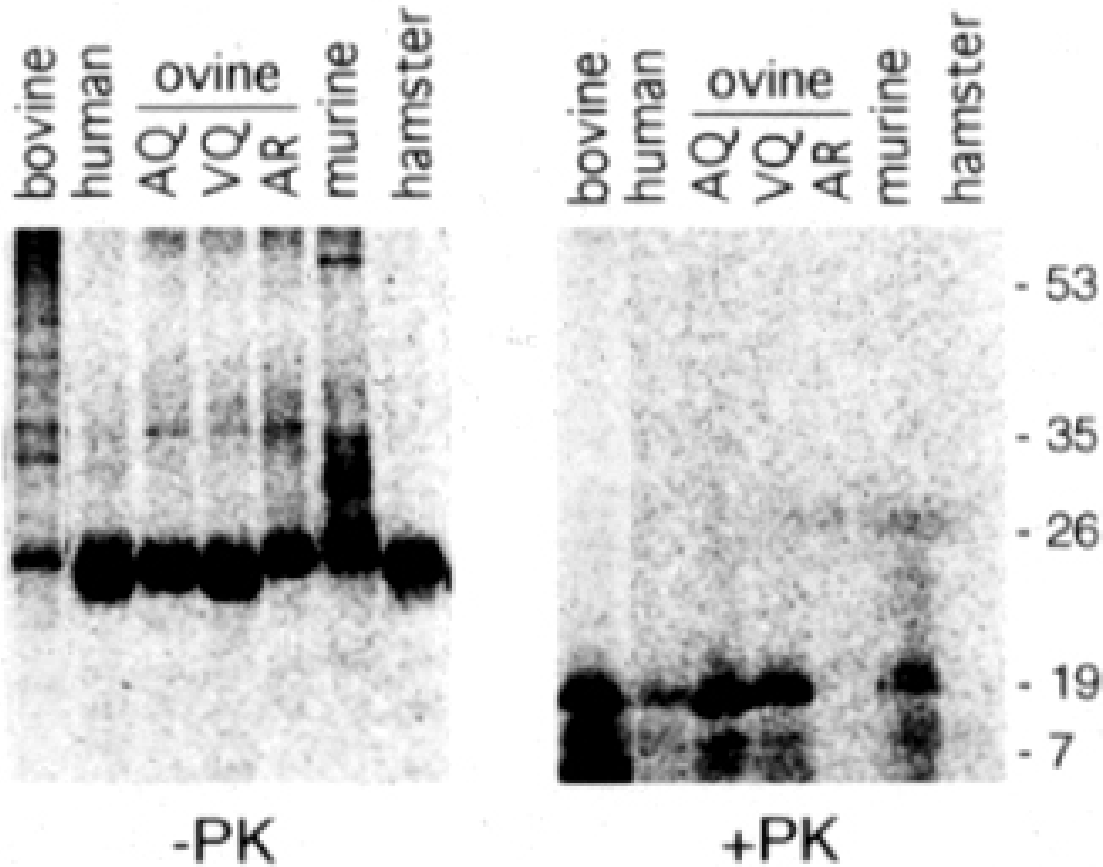
J. Infect Dis. 189:431 (2004)

Conclusions

- Cannot predict susceptibility to TSE infection based upon:
  - Acute PrP-res formation
  - Expression level of PrP-sen
  - Cell type
    - Neuronal vs non-neuronal
    - Cell passage history
  - Strain of TSE agent

- Factors that likely influence susceptibility to TSE infection:
  - PrP-sen must be expressed
  - PrP-sen amino acid sequence homology with PrP-res [Villette et al. PNAS 98:4055 (2001)]

PrP^{BSE}-induced conversions of various species' [^{35}S] PrP-sen molecules



Nature 388:285 (1997)

Implications

- Without direct assessment of susceptibility to TSE infection, it is largely invalid to assume that a particular cell type will be resistant to a TSE agent

- To assay the susceptibility of a given cell type to TSE infection:
  - Expose cells to TSE agent of interest at both high and low multiplicities of infection
  - Assay cells for PrP-res formation over multiple passages
  - If PrP-res is not detected:
    - Assay cells for infectivity using the appropriate in vivo bioassay

Acknowledgements

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