Discrimination of Classical and Atypical BSE by a Distinct Immunohistochemical PrP^{Sc} Profile

Fast et al. 2023, Pathogens, 12:353

C. Fast, C. Graham, M. Kaatz, K. Santiago-Mateo, T. Kaatz, K. MacPherson, A. Balkema-Buschmann, U. Ziegler, M.H. Groschup, S. Czub

Rome 2.10.2023

> Vet Rec. 1987 Oct 31;121(18):419-20. doi: 10.1136/vr.121.18.419.
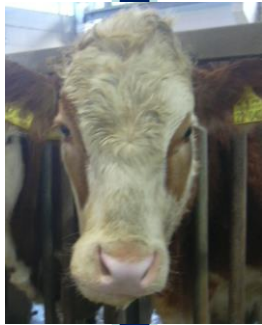
A novel progressive spongiform encephalopathy in cattle

G A Wells [1], A C Scott, C T Johnson, R F Gunning, R D Hancock, M Jeffrey, M Dawson, R Bradley

Affiliations + expand

PMID: 3424605   DOI: 10.1136/vr.121.18.419

1987

PNAS

Identification of a second bovine amyloidotic spongiform encephalopathy: Molecular similarities with sporadic Creutzfeldt–Jakob disease

Cristina Casalone*[†], Gianluigi Zanusso[†‡], Pierluigi Acutis*, Sergio Ferrari[‡], Lorenzo Capucci[§], Fabrizio Tagliavini[¶], Salvatore Monaco[‡∥], and Maria Caramelli*

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Edited by Stanley B. Prusiner, University of California, San Francisco, CA, and approved December 23, 2003 (received for revie
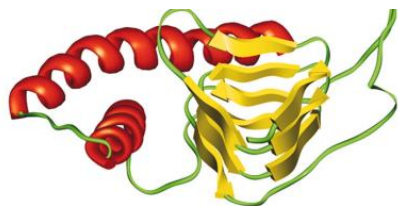
2004

EMBO reports

scientific report

Distinct molecular phenotypes in bovine prion diseases

Anne-Gaëlle Biacabe[1], Jean-Louis Laplanche[2], Stephen Ryder[3] & Thierry Baron[1+]

[1]AFSSA-Lyon, Unité 'Virologie-ATNC', Lyon, France, [2]UPRES EA 3621, Faculté des Sciences Pharma   Université Paris, Paris, France, and [3]Neuropathology Unit, Department of Pathology, Veterinary Labora
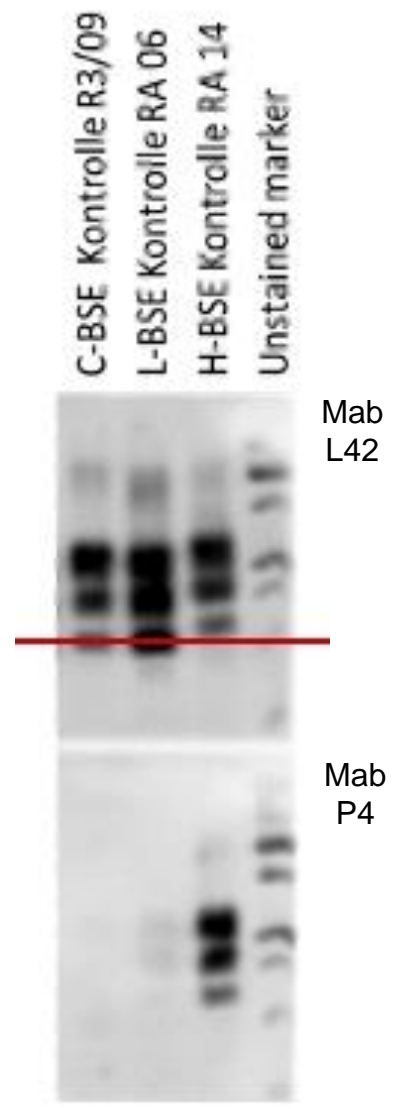
2004

Discrimination of BSE-types

PrP^{Sc} molecular signature of H- and L-type BSE differed from C-BSE in terms of protease-resistant fragment size and glycoform pattern

Proteinase

BSE PrP^{Sc}

H-type PrP^{Sc}

mab P4

mab L42

C-BSE Kontrolle R3/09

L-BSE Kontrolle RA 06

H-BSE Kontrolle RA 14

Unstained marker

Mab L42

Mab P4

➡ **Discriminatory Immunoblot**

	C-Type	L-Type	H-Type
2x glycos band	Most distinct band	<40%, comparable with unglycosylated band	Most distinct band
P4/L42	L42 >>> P4	L42 >>> P4	P4 > L42
MW unglyco-sylated band	Medium	Low	High

Aim of the study:

Establishing immunohistochemistry as an additional tool to discriminate BSE types

Fast et al. 2023

Literature Review

Numerous, in parts contrasting results

BUT

Indications for discriminatory potential in certain areas

Systematic examination necessary!

BSE Type	Animal ID	Incubation Time (mpi)	Lesions (H&E)	Obex/PrP^{Sc} Deposition
C-Type Canada	25015	26	++	+++
	25022	27	++	+++
	25023	27	++	+++
	25032	26	+	+++
	25034	24	++	+++
	29024	20	+	++
	29026	18	(+)	++
H-Type Canada	29018	18	+	++
	29033	17	+	++
L-Type Canada	29012	18	++	++
	29030	17	++	+
H-Type Germany	RA10	12	(+)	+
	RA13	15	++	+++
	RA14	14	+	++
	RA15	16	+	+++
	RA16	16	++	+++
L-Type Germany	RA02	17	+++	+++
	RA03	16	++	++
	RA04	16	+++	+++
	RA05	11	(+)	(+)
	RA06	14	++	+

Material available

<u>i.c. inoculation</u>

<u>Inocula:</u>
1ml of 10% brain homogenate

<u>Inoculation Site:</u>
(Rostral) Midbrain

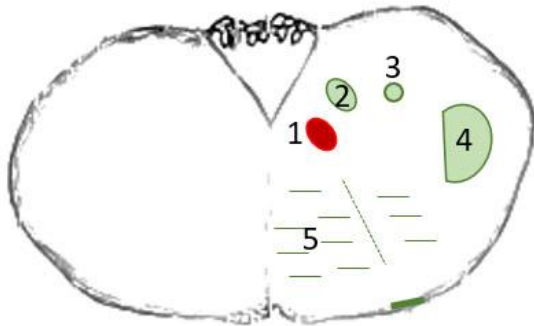| BSE Type	Animal ID	Incubation Time (mpi)	Lesions (H&E)	Obex/PrP^{Sc} Deposition
C-Type Canada	25015	26	++	+++
	25022	27	++	+++
	25023	27	++	+++
	25032	26	+	+++
	25034	24	++	+++
	29024	20	+	++
	29026	18	(+)	++
H-Type Canada	29018	18	+	++
	29033	17	+	++
L-Type Canada	29012	18	++	++
	29030	17	++	+
H-Type Germany	RA10	12	(+)	+
	RA13	15	++	+++
	RA14	14	+	++
	RA15	16	+	+++
	RA16	16	++	+++
L-Type Germany	RA02	17	+++	+++
	RA03	16	++	++
	RA04	16	+++	+++
	RA05	11	(+)	(+)
	RA06	14	++	+
Oral C-Type Germany	IT18	50	+++	+++
	IT23	36	++	+++
	IT49	36	+++	+++

Material available

i.c. inoculation

Inocula:
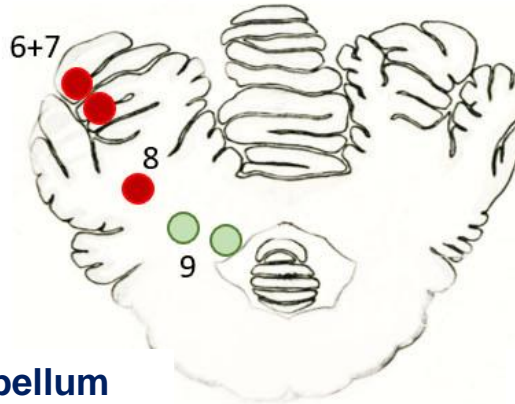1ml of 10% brain homogenate

Inoculation Site:
(Rostral) Midbrain
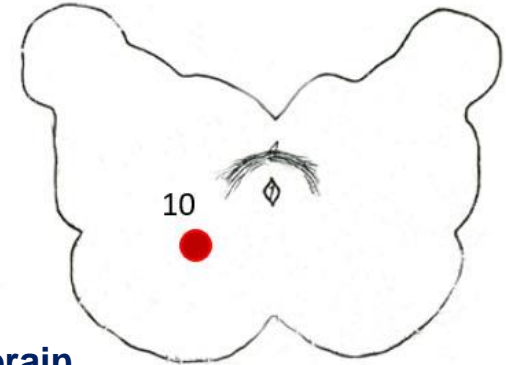
Oral inoculation
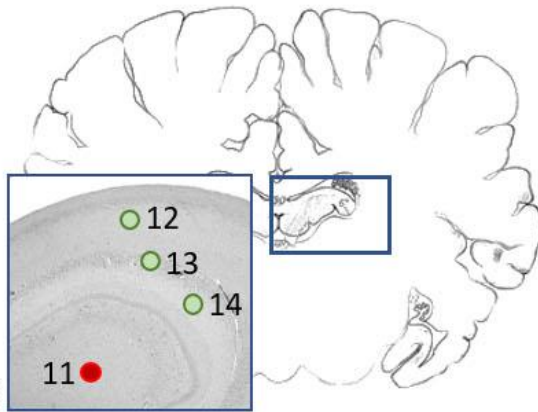100 ml 10% brain homogenate

17 Regions examined:
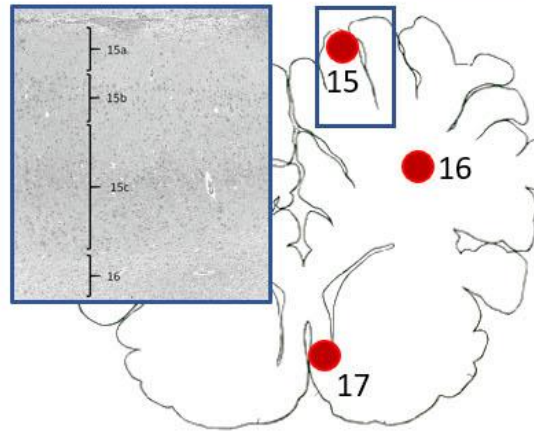

Brainstem

Cerebellum

Mid brain

Hippocampus

Frontal Cortex and Septum

Lesion Profile

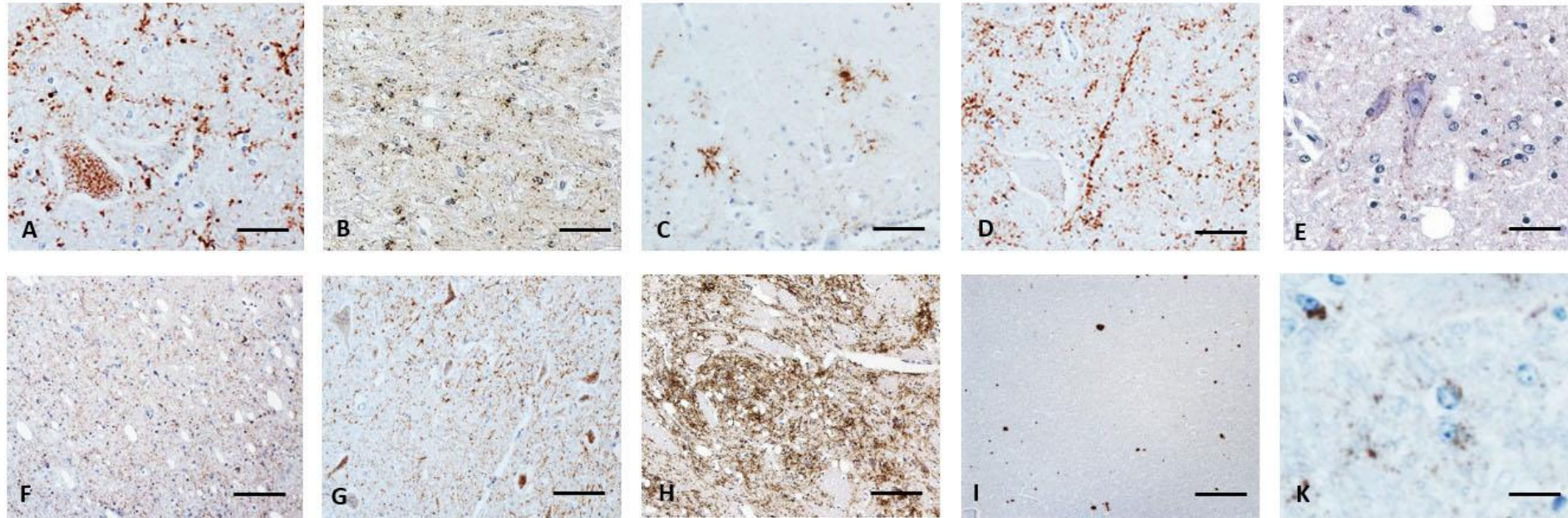**PrP^Sc Profile**

11 PrP^{Sc} Reaction Pattern

➡ **mab F99**

(A) intraneuronal (ITNR);
(B) intramicroglial (ITMG);
(C) stellate (STEL);
(D) linear (LIN);
(E) perineuronal (PNER);
(F) fine particulate (f-PART);
(G) coarse pariculate (c-PART);
(H) coalescing (COL);
(I) plaque-like (PL)
(J) Intra-astrocytic (ITAS);
Submengeal (SMENG) is not shown

Lesion Profile

Lesion profile (H.&E.)

Highly accordant, but for Red nucleus in C-type

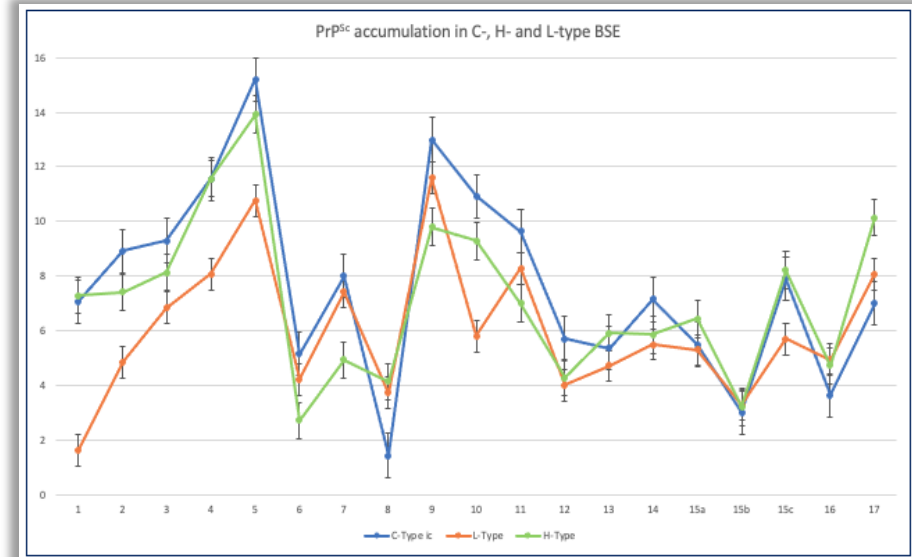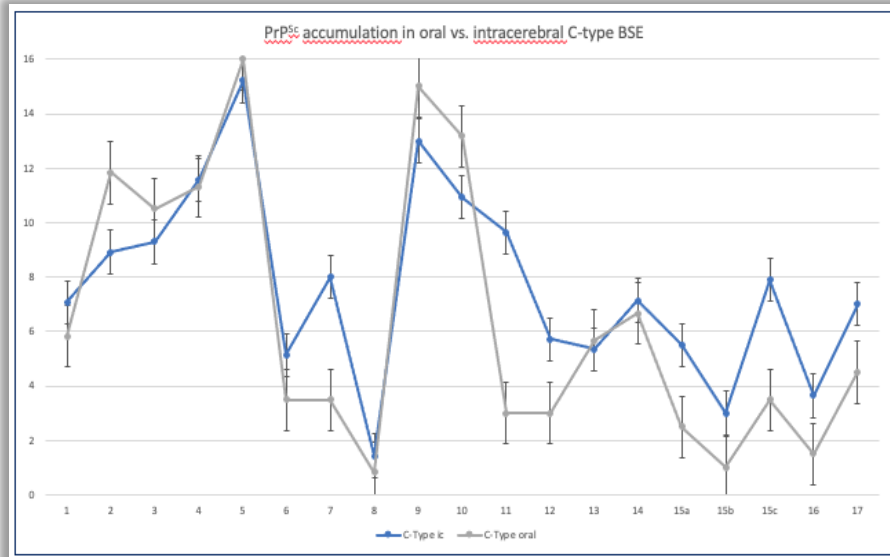**Frontal Cortex distinctly involved in Atypical BSE, but not in C-type**

PrP^{Sc} Profile
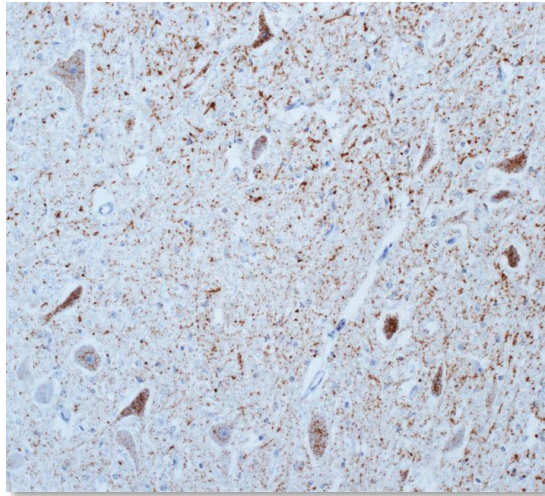
PrP^{Sc} profile in general

The extent of the different PrP^{Sc} reaction patterns in each brain region was determined for each animal, and the following scores were used: weak (score 1.0), mild (2.0), moderate (3.0), and severe (4.0). These scores were then totalled to determine the total amount of PrP^{Sc} accumulation for each brain region for each animal. For the graphs the average of all animals per group (C-, H-, and L-type and orally infected C-type) was determined for each brain region.
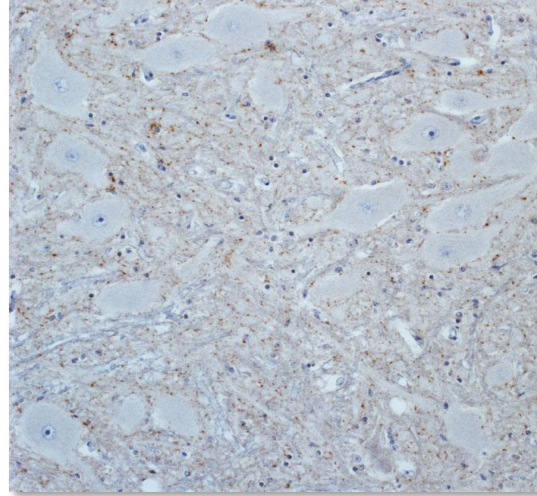
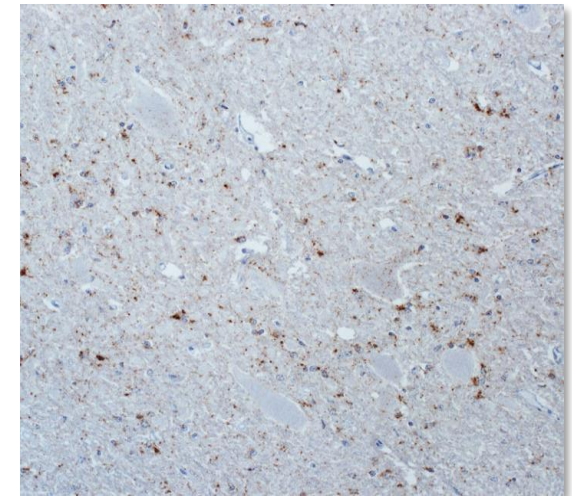**Overall PrP^{Sc} profiles showed a high similarity, independently of BSE type and route of infection**
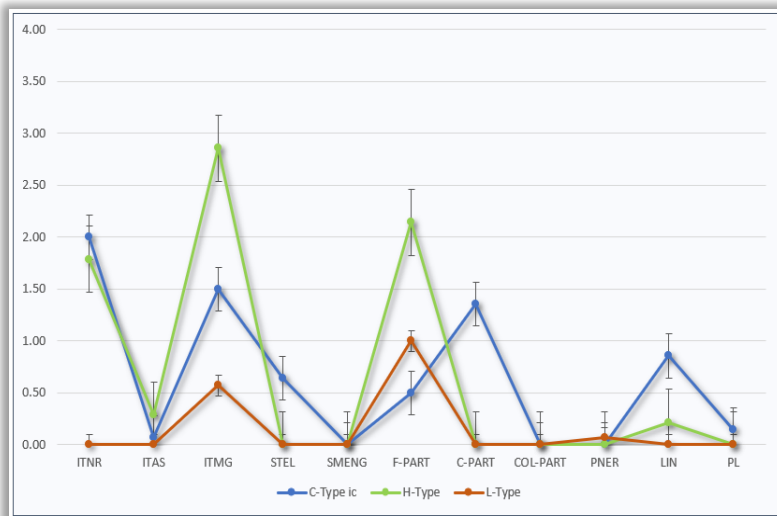
PrP^{Sc} profile / Nc. Hypoglossus (obex)

C-type

L-type

H-type

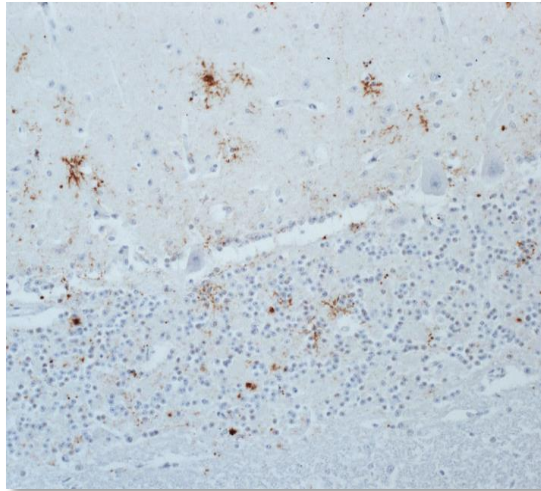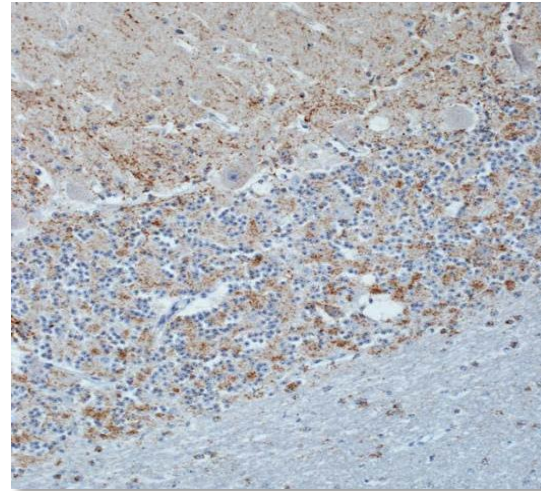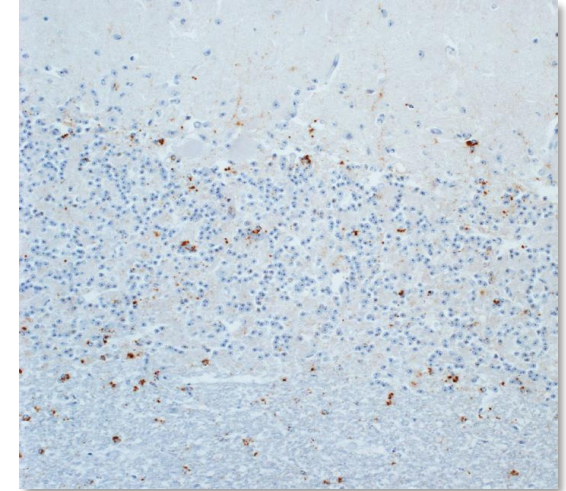**L-type:**
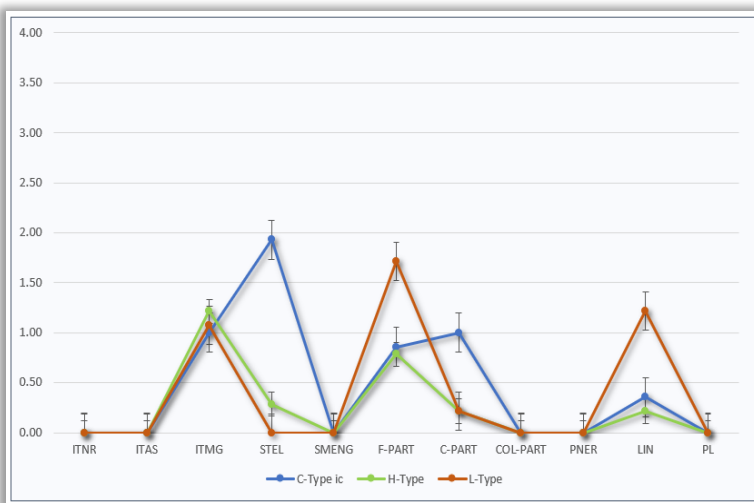**No ITNR staining reaction**

H-type:
Distinct ITMG reaction pattern

C-type
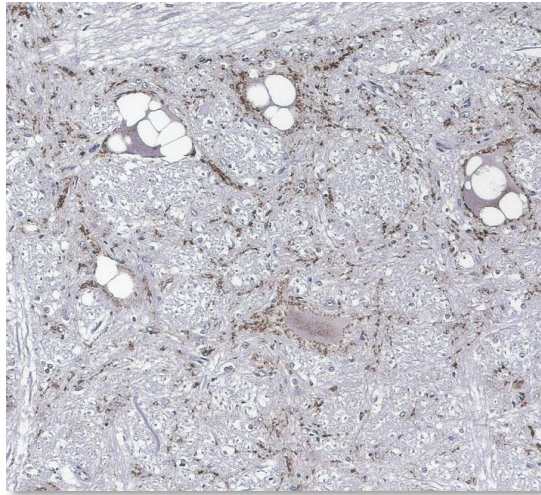**Mixed pattern and distinct coarse-PART**

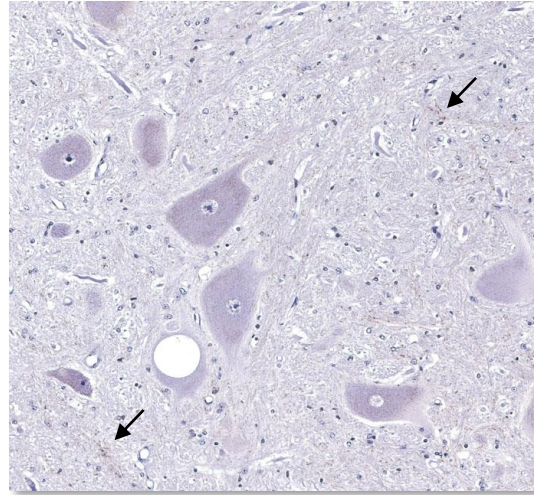# PrP^{Sc} profile / Cerebellum (Molecular + Granular Layer)

C-type

L-type

H-type

L-type:
Diffuse fine PrP^{Sc} deposition

H-type:
Distinct ITMG reaction pattern
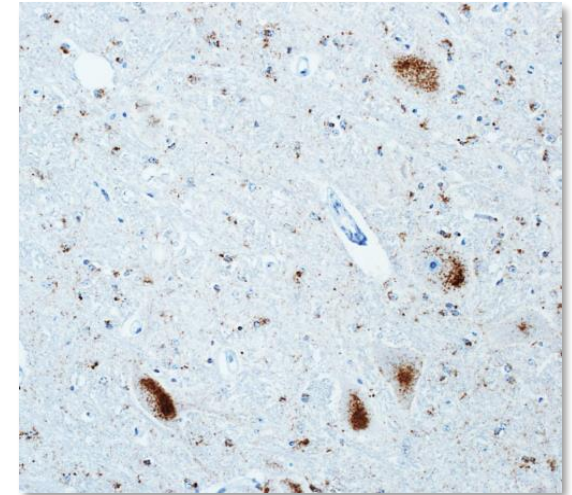
C-type
Prominent stellate staining reaction

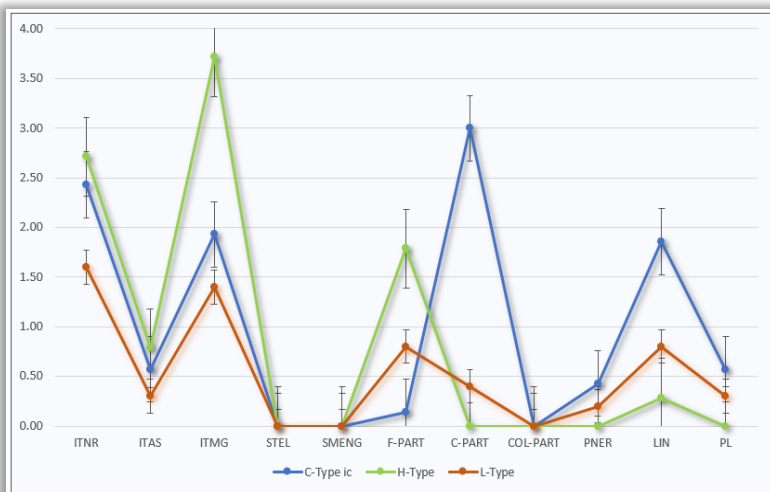# PrP^{Sc} profile / Red nucleus

C-type

L-type

H-type

L-type:
Multifocal fine PART

H-type:
Distinct ITMG reaction pattern
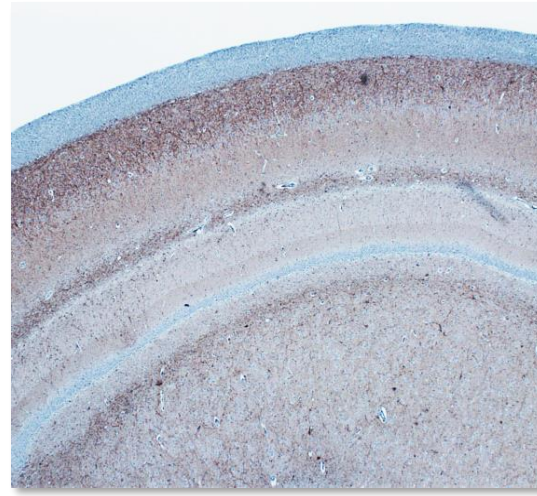
C-type
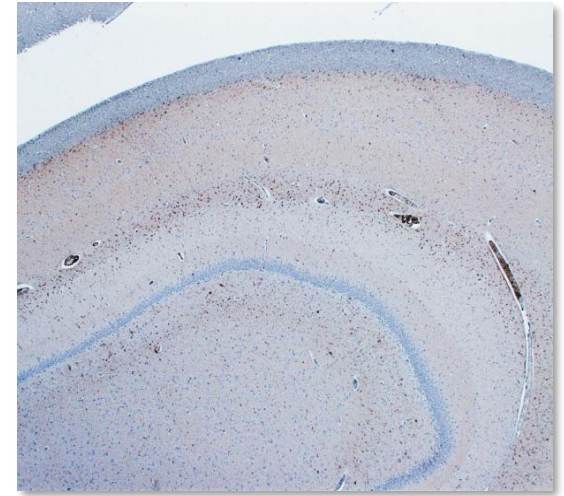**Highly variable staining reaction**
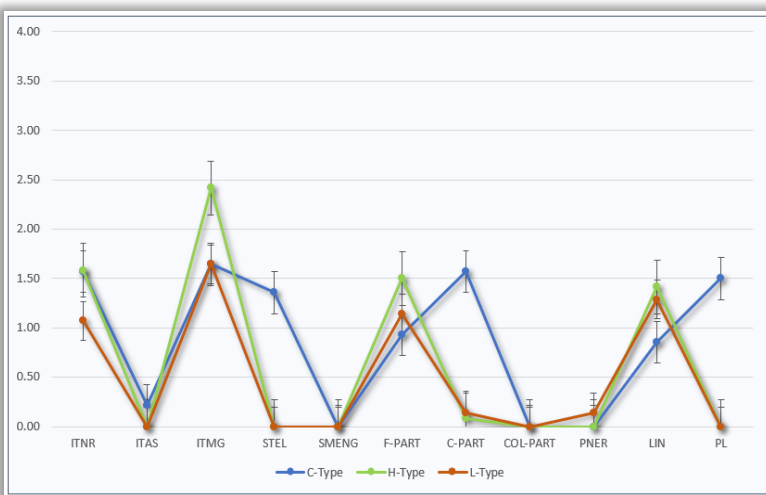
PrP^{Sc} profile / Hippocampus (Hilus)
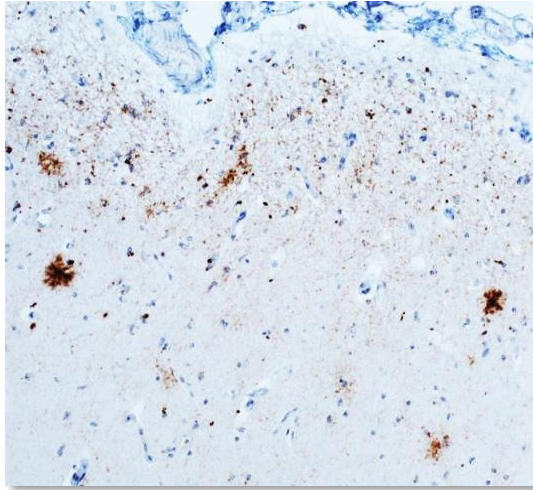

C-type

L-type

H-type

L-type / H-type:
Mainly in peripheral parts of hilus
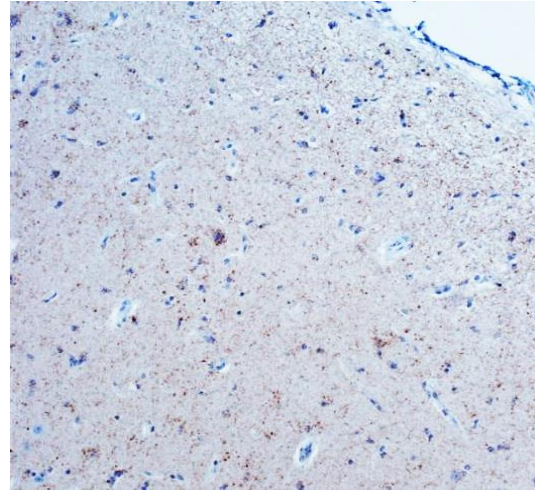
C-type
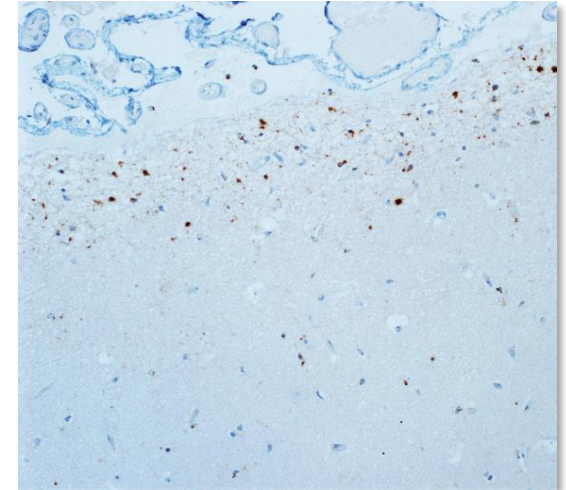**Predominant in central parts of hilus**

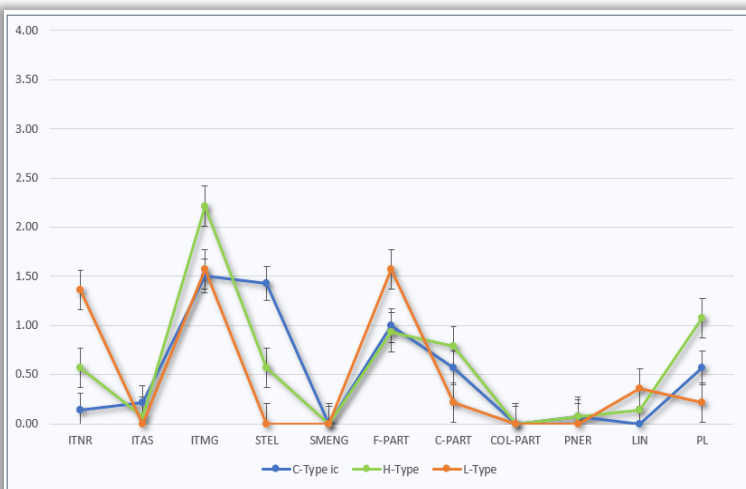# PrP^Sc profile / Cerebrum (Molecular Layer)

C-type

L-type

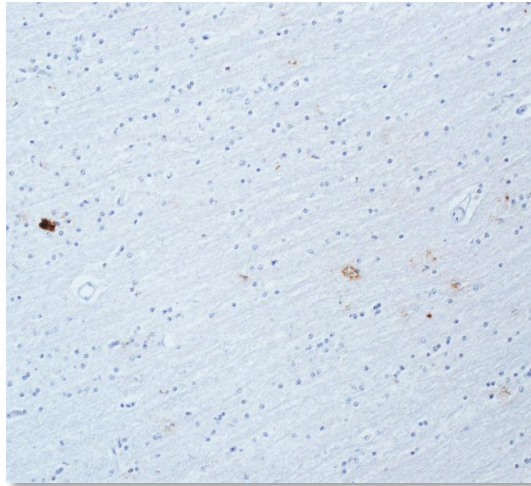**H-type**

L-type:
Diffuse fine PART

H-type:
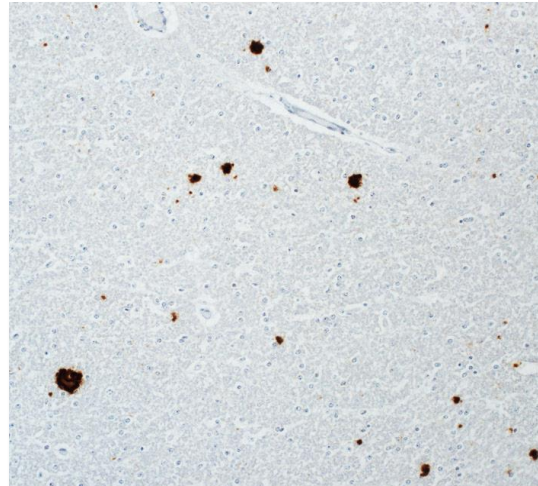Distinct ITMG, mostly tape-like

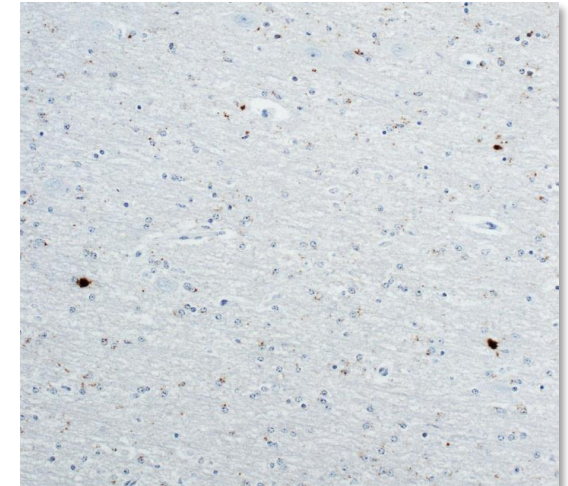**C-type**
Tape-like pattern, multifocal STELL

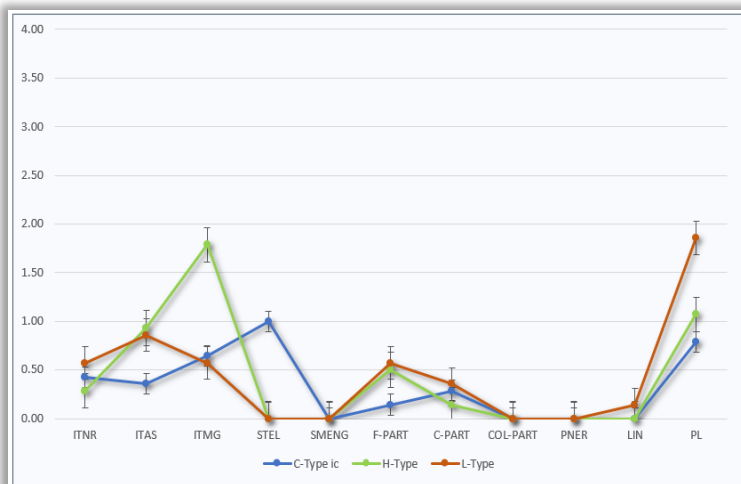# PrP^{Sc} profile / Cerebrum (White matter)

C-type

L-type

H-type

L-type:
Predominant PL pattern

H-type:
Distinct ITMG

C-type
Multifocal STELL PrP^{Sc} accumulation

Take Home Message

Lesion and PrP^{Sc} profile with slight differences between oral and i.c. C-type infected animals

➡ *Results of intracerebral studies are comparable to field cases*

High similarity of the general PrP^{Sc} profile independently of the BSE type and the route of infection

➡ *Depending on on consistent PrP^{C} distribuion in cattle brain?*

Qualitative differences of the cellular reaction pattern in a selection of neuro-anatomical structures

➡ *Nc. Hypoglossus; Cerebellum (Molecular-/Granular layer); Red nucleus; Hippocampus (Hilus); Septal nucleus (data no shown); Cerebrum (Molecular layer, White matter)*

Characteristic cellular PrP^{Sc} pattern for each BSE type throughout the brain

➡ *C-type highly variable; L-type diffuse fine PART; H-type distinct ITMG*

However!

Field cases are usually autolytic

In most field cases only brain stem and (with some luck) cerebellum are available

?

It remains unclear if this approach works with routine submissions!

Do you have formalinfixed/paraffin embedded material available and, *if yes*, are you willing to share?