CJD Pathology and Diagnostic Markers

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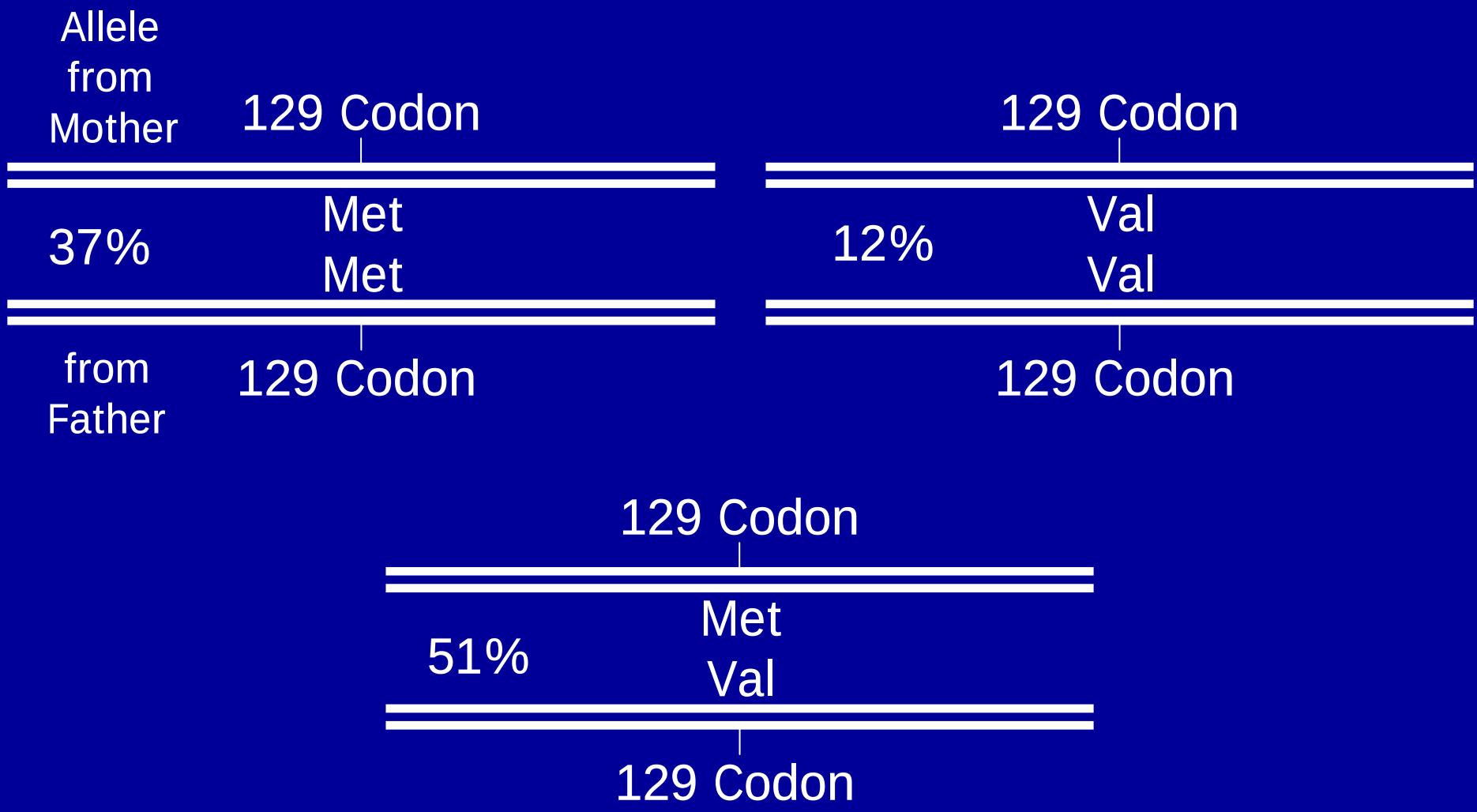
National Prion Disease Pathology Surveillance Center

Human prion diseases: Classification

Form	Phenotype
Sporadic	CJD Typical M/M (M/V) 1 Early V/V 1 Long M/M2 Kuru plaques M/V 2h Ataxic V/V 2 FI
Inherited	Creutzfeldt-Jakob disease (CJD) Fatal Familial Insomnia (FFI) Gerstmann-Sträussler-Scheinker (GSS) Mixed or undefined
Acquired by infection	Kuru Iatrogenic CJD (iCJD) Variant of CJD (vCJD)

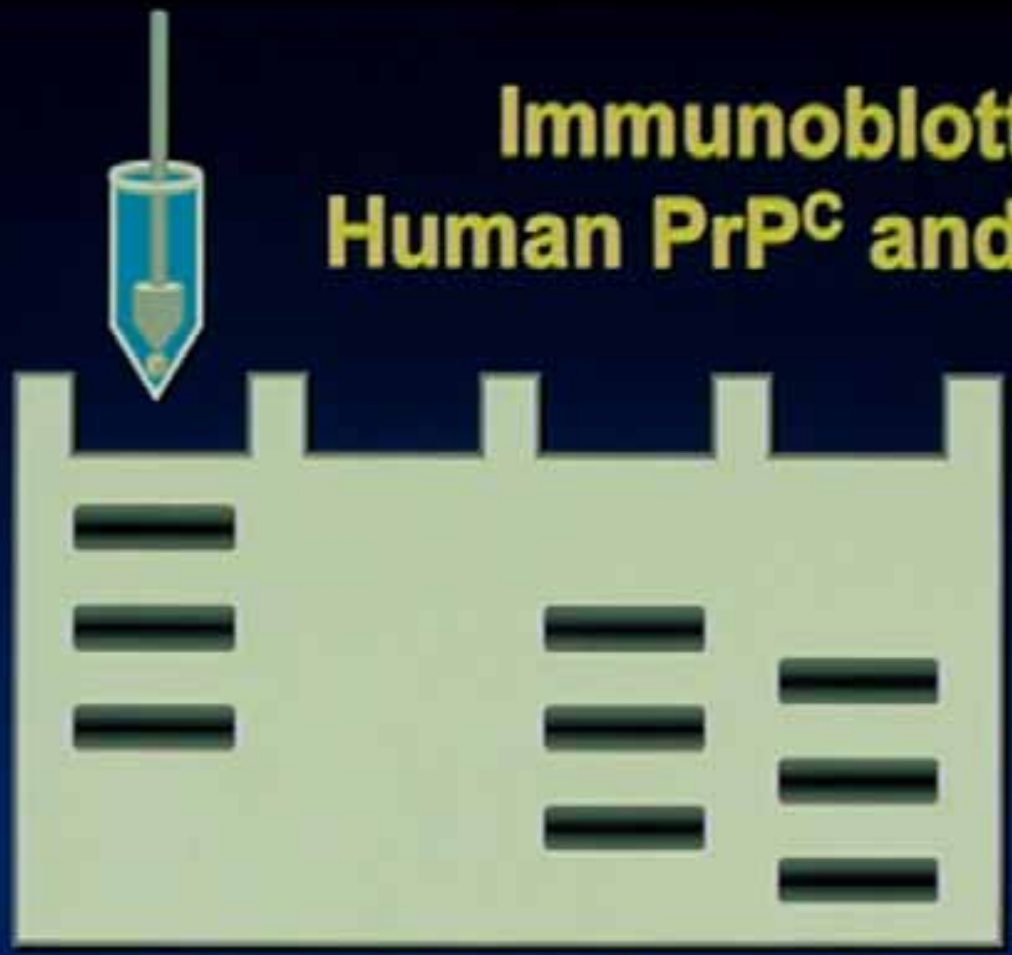
Human Prion Protein Gene
Codon 129 Polymorphism



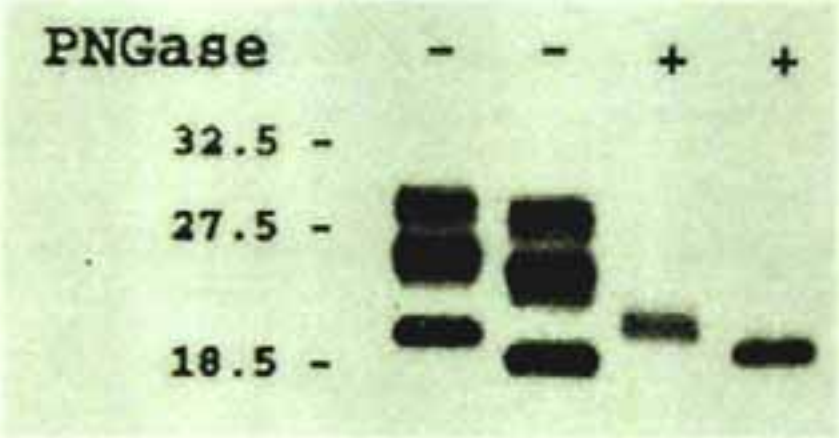
Prevalence of the Met/Val Polymorphism at Codon 129 of the Prion Protein Gene in a Normal Caucasian Population

Met/Met	37%
Met/Val	51%
Val/Val	12%
Met allele	62%
Val allele	38%

Immunoblotting of Human PrP^{C} and PrP^{Sc}



PrP^{Sc} Type 1 and 2
Size of PK-resistant fragment



82 231

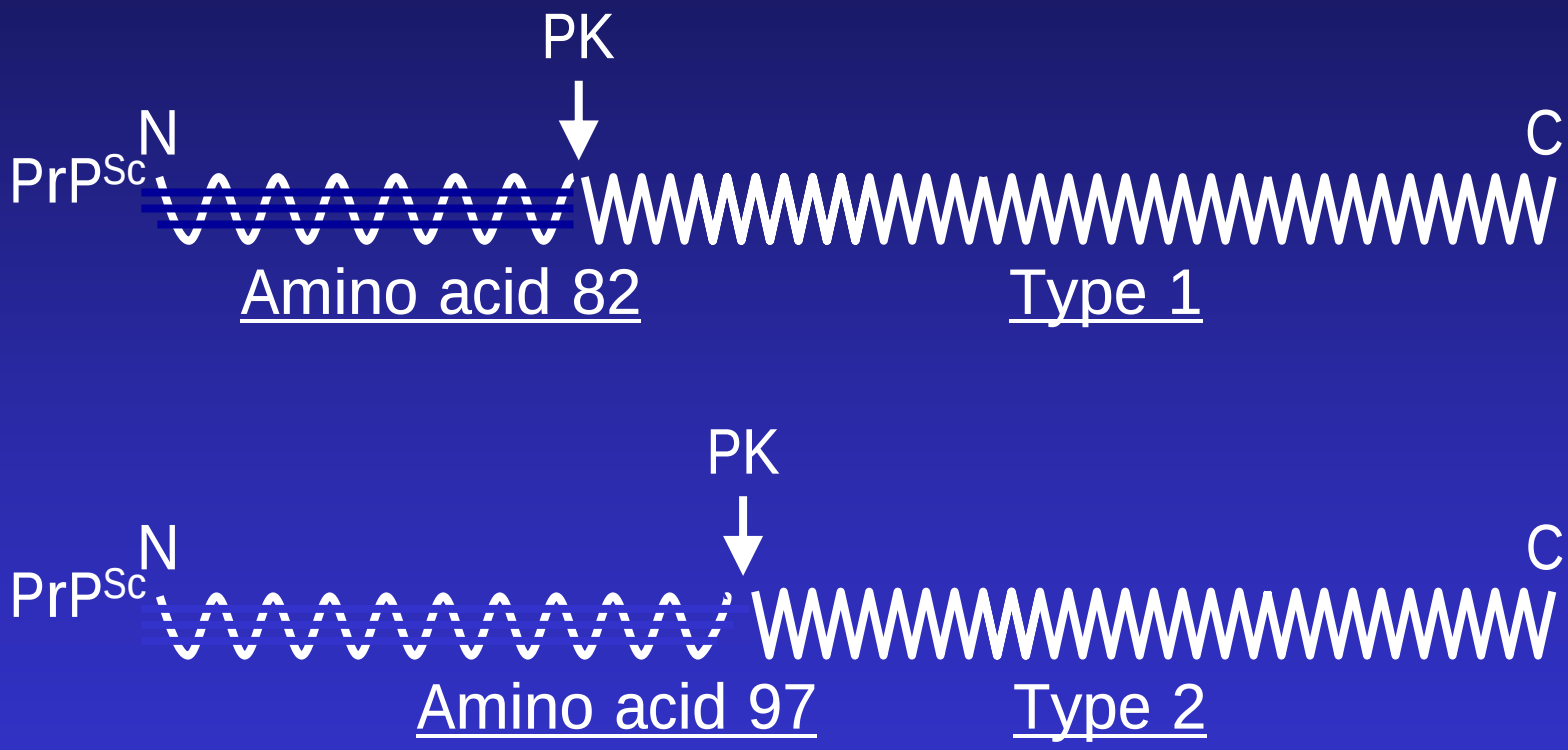
Type 1 GQPHGGGWGOGGGTHSQWNKP...S

97 231

Type 2 SQWNKP...S

PrPSc Type

How they are detected



The cleavage of the PrPSc by PK occurs at different sites because of the different conformation of the two PrP^{Sc} species

The Meaning of Generating PrP^{Sc} fragments of different sizes following digestion with proteases

The formation of PrP^{Sc} fragments of different sizes following treatment of PrP^{Sc} with a proteolytic enzyme indicates that the original PrP^{Sc} have different conformations and offer distinct cleavage sites to the proteolytic enzyme.

Subtypes of sCJD

Typical or myoclonic

Amyotrophic

Visual or Heidenhain

Thalamic

Ataxic or cerebellar

Corticostriatal

Panencephalopathic

Long Duration

The six possible combinations of codon 129 alleles and PrP^{Sc} types correlated with disease phenotypes in patients with sporadic prion diseases

Codon 129	PrP^{Sc} Type
Met/Met	1
Met/Met	2
Met/Val	1
Met/Val	2
Val/Val	1
Val/Val	2

Classification of Sporadic Prion Diseases

Variant	Previous Classificat.	Case (%)	Onset (yrs)/ Durat.(mo)	Distinctive Features
			sCJD	
M/M 1 **M/V 1**	**Myoclonic** **Heidenhain**	**71**	**63.2 / 3.9**	**Typical CJD clinically and pathologically. Typical EEG (83%). "Synaptic" pattern of immunostain**
V/V 1	**Unavailable**	**1**	**46.0 / 15.3**	**Early onset. No typical EEG. Cerebell spared. Weak "synaptic" immunostain**
M/M 2 Cortical	**Long duration**	**2**	**60.3 / 15.7**	**No typical EEG. Coarse spongiosis and immonostain. Cerebellum spared**
M/V 2	**Cerebellar or ataxic**	**8**	**60.3 / 17.0**	**Ataxia at onset. Rarely typical EEG. Kuru plaques. No cerebell. atrophy**
V/V 2	**Cerebellar or ataxic**	**16**	**60.3 / 6.6**	**As M/V 2 but no kuru plaques and cerebell. atrophy. Plaque-like present.**
			sFI	
M/M 2	**Sporadic fatal insomnia**	**2**	**60.3 / 14.0**	**Clinically and pathologically indistinguishable from FFI**

Cases of sporadic CJD examined in the six molecular subtypes and sensitivity of 14-3-3 protein test

Sporadic CJD	14-3-3 Test Sensitivity	nPositive/ nTotal
PrP^{Sc} type 1	96%	45/47
MM1	95%	40/42
MV1	100%	2/2
VV1	100%	3/3
PrP^{Sc} type 2	80%	24/30
VV2	82%	14/17
MV2	80%	4/5
MM2[b]	75%	6/8
Overall	**90%**	**69/77**

[a]According to Parchi et al. (1999).

[b]All MM2 subjects belong to the MM2-cortical subtype (Parchi et al, 1999)

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Pathogenic mutations and polymorphisms of the human prion protein

Pathogenic Mutations and Phenotypes

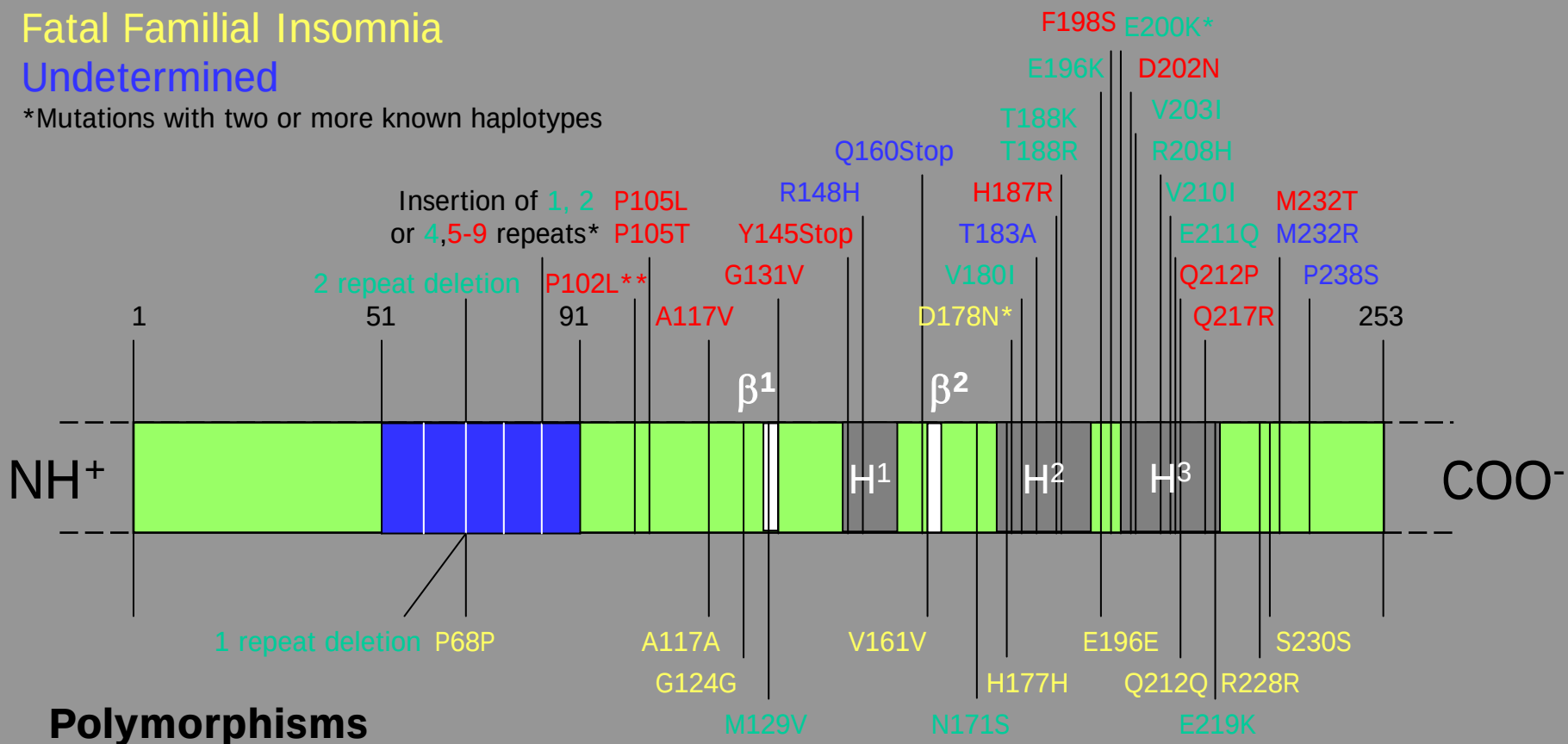
Gerstmann-Sträussler-Scheinker Disease

Creutzfeldt-Jakob Disease

Fatal Familial Insomnia

Undetermined

*Mutations with two or more known haplotypes



Polymorphisms

Variant

Silent

Human prion diseases: Classification

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Acquired by infection	Kuru Iatrogenic CJD (iCJD) Variant of CJD (vCJD) Others?

National Prion Surveillance Centers
Why we need them

Animal to human transmission

BSE → vCJD 170[&] cases (UK154;FR9;IRL2; IT1;CA1*;US1*;JP1*;SA1)

CWD → CJD**?** 26 cases of prion d. in hunters

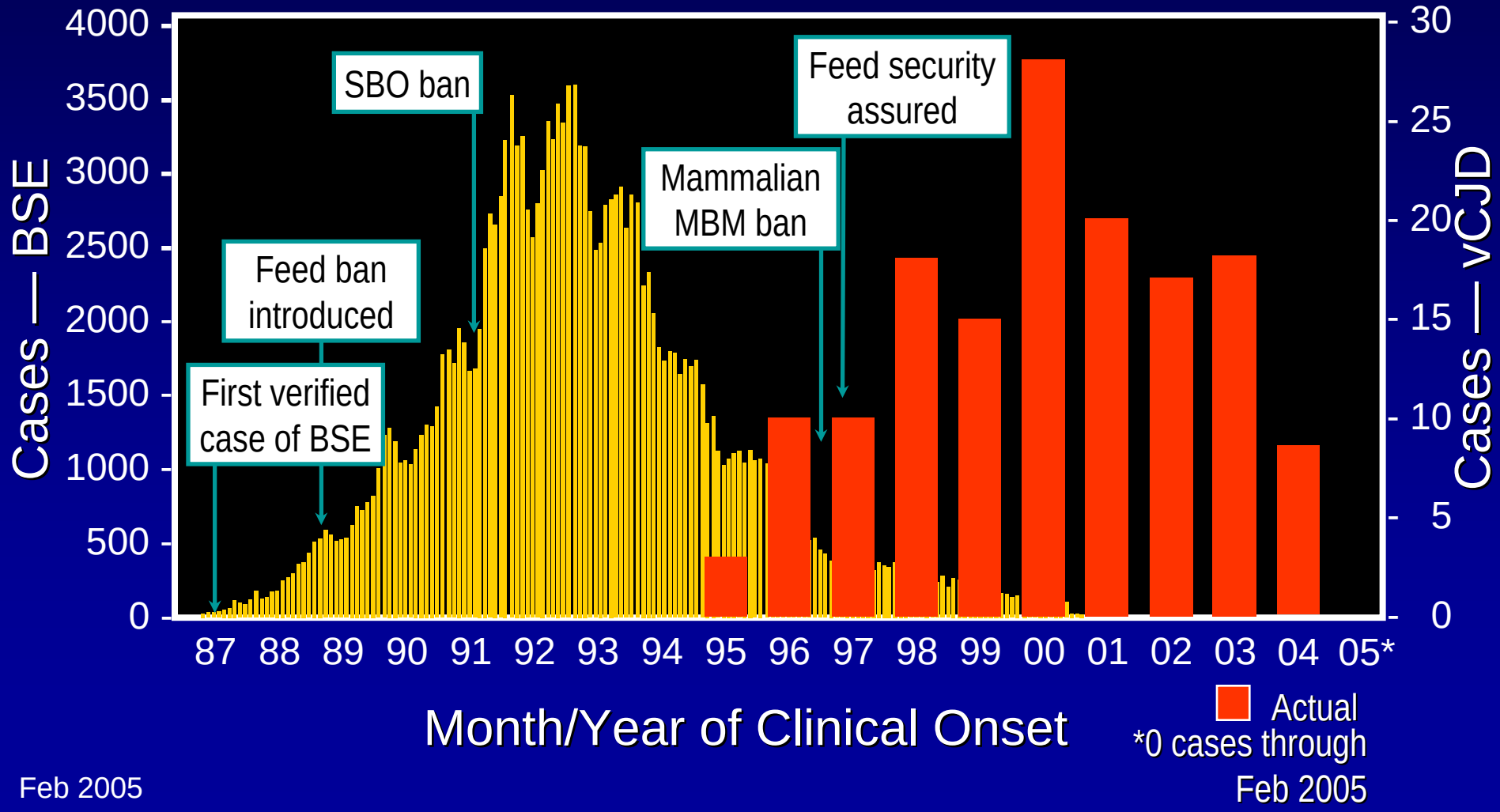
Human to human transmission

iCJD >300 cases worldwide

[*] Probably acquired in UK

[&]As of February 22, 2005

BSE and vCJD Epidemics
Time Course



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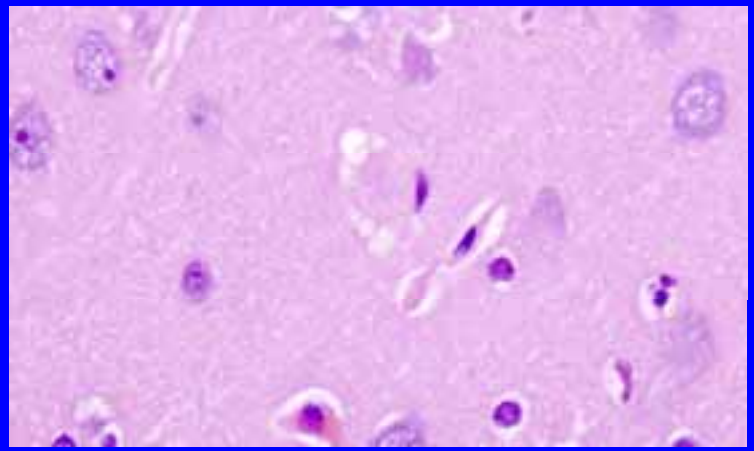
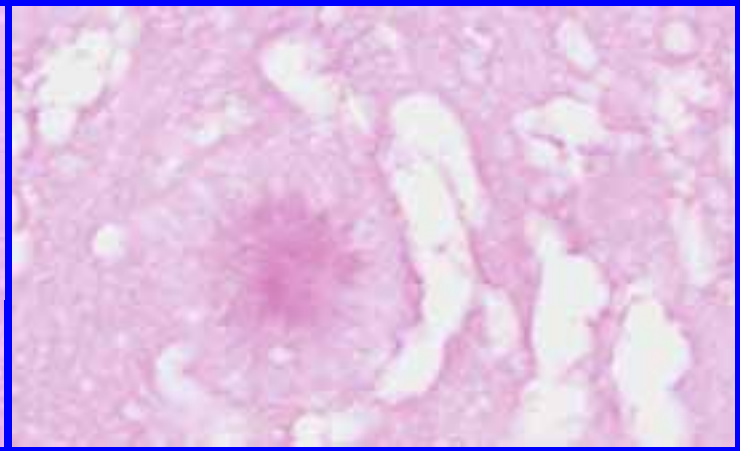
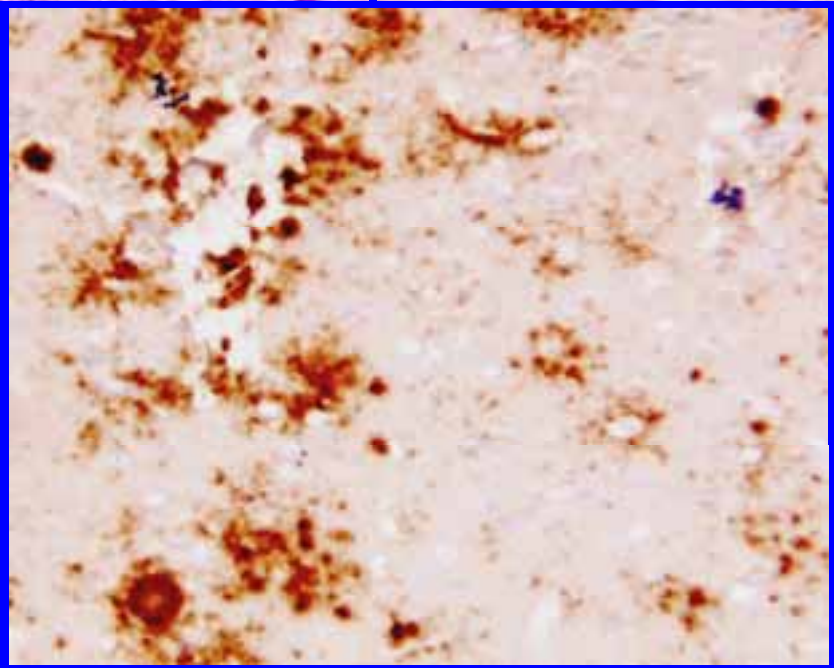
Variant Creutzfeldt-Jakob Disease Biopsy

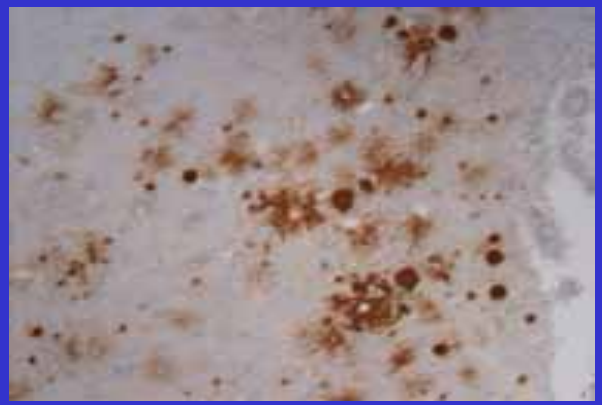






PrP immunostaining in variant CJD, sporadic CJD and normal brain

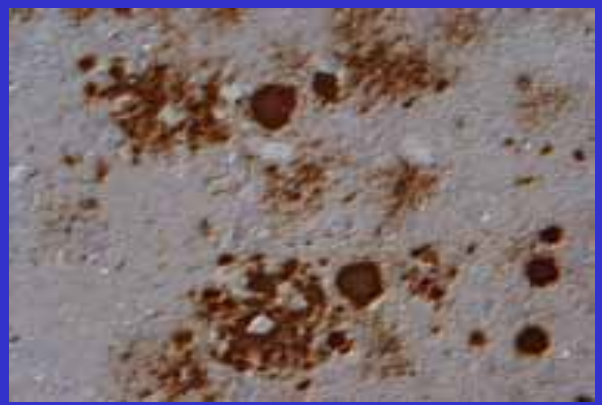
vCJD 20X Mag



sCJD 20X Mag

Normal 20X Mag

vCJD 40X Mag

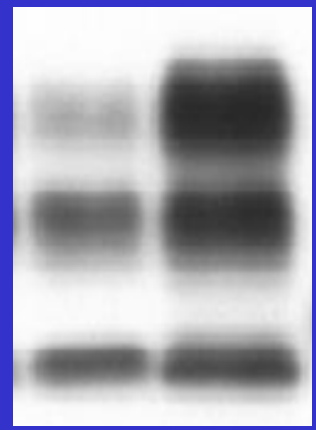


sCJD 40X Mag

Normal 40X Mag

Immunoblot of sporadic and variant Creutzfeldt-Jakob Disease

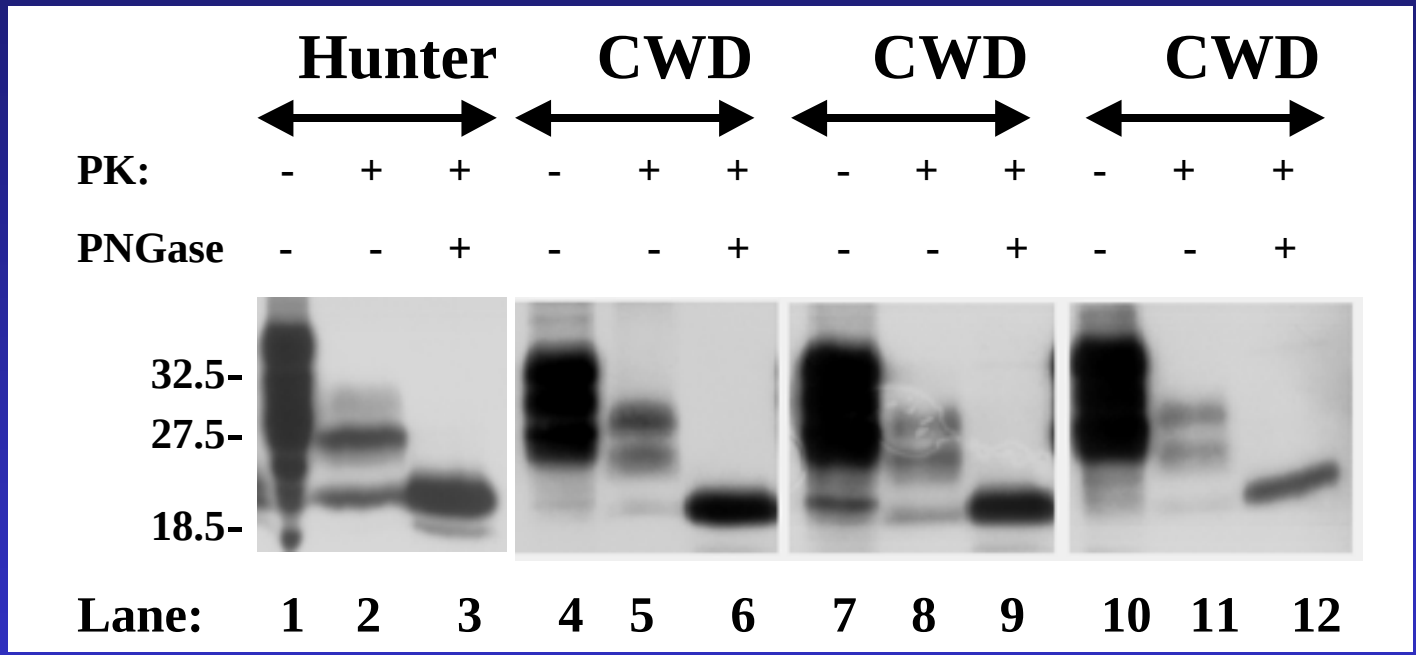
Proteinase K-treated PrP^{Sc}



Immunohistochemical Staining of PrP^{Sc} in CWD

Brain tissue

Comparison of PrP^{Sc} in brains of a hunter with CJD and elk with CWD

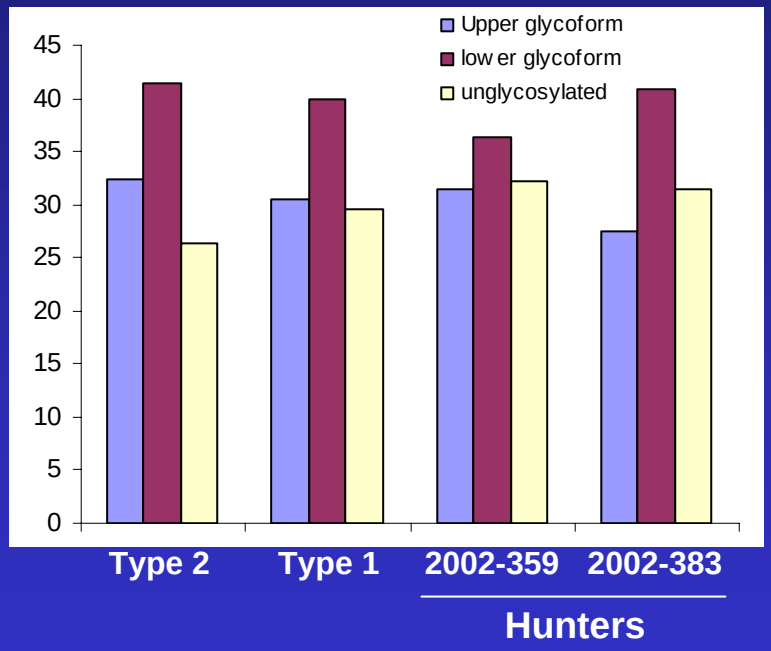


Species	# of Cases	PrP^{Sc} Type
Elk	9	1
Mule Deer	7	1

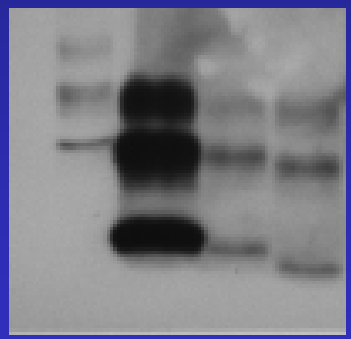
Summary of CJD Cases Investigated for a Possible Causal Association with CWD

- Number of cases: 26
- Age range: 25-80 (average age: 54)
- Final diagnosis:
  - sCJD: 22 cases
    - M/M 1: 9 cases
    - M/M 2: 1 case
    - M/V 1: 1 case
    - V/V 1: 2 cases
    - V/V 2: 5 cases
    - Unknown: 4 cases
  - fCJD: 1 case (E200K mutation)
  - GSS: 3 cases (all P102L mutation)

Ratios of the PrP^{Sc} glycoforms in sCJD and in two cases of CJD in hunters



	Di-	Mono	Unglycos
Type 2	32.31	41.37	26.32
Type 1	30.54	39.93	29.53
02-359	31.45	36.32	32.22
02-383	27.57	40.92	31.53



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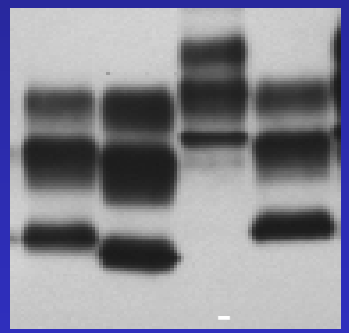
2002-359



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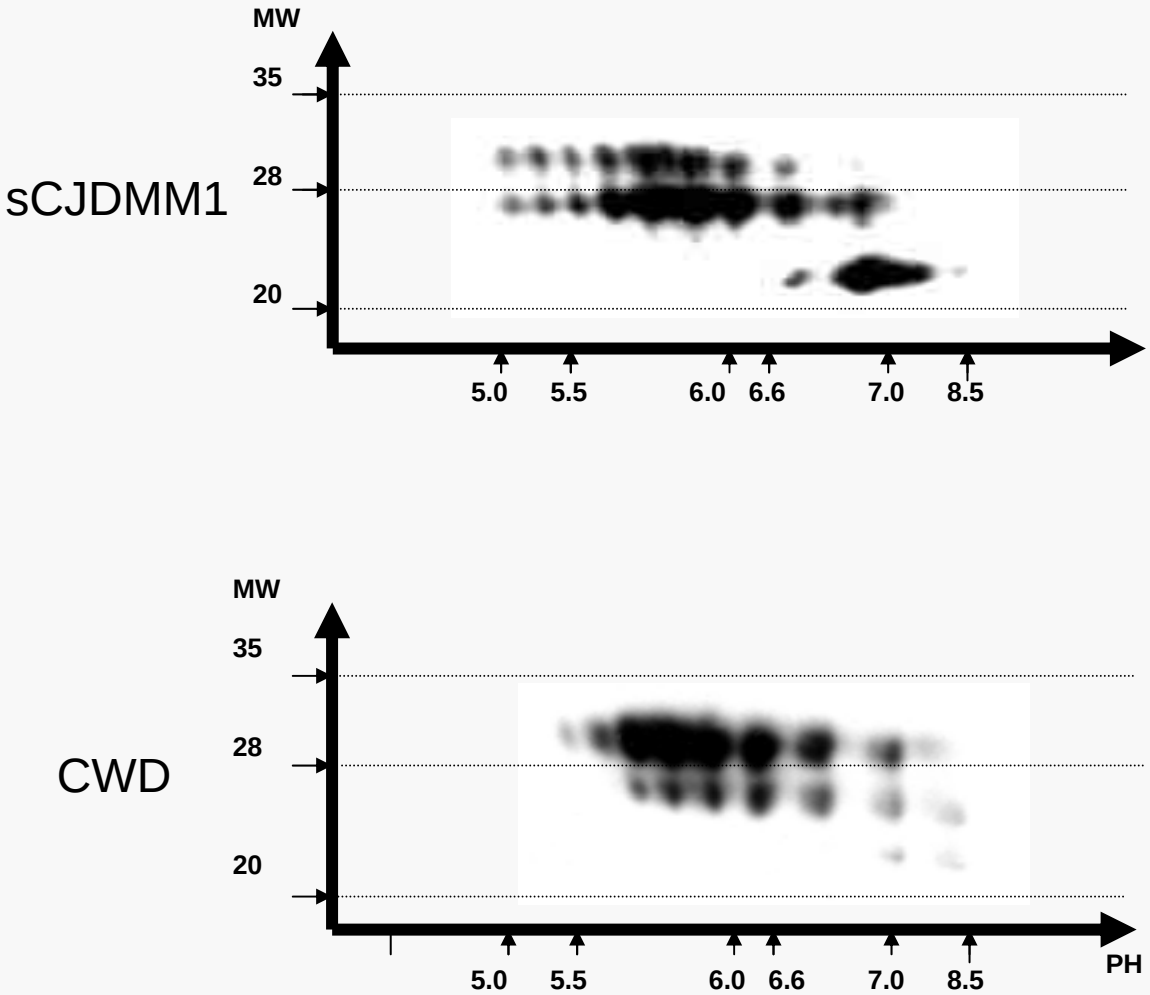
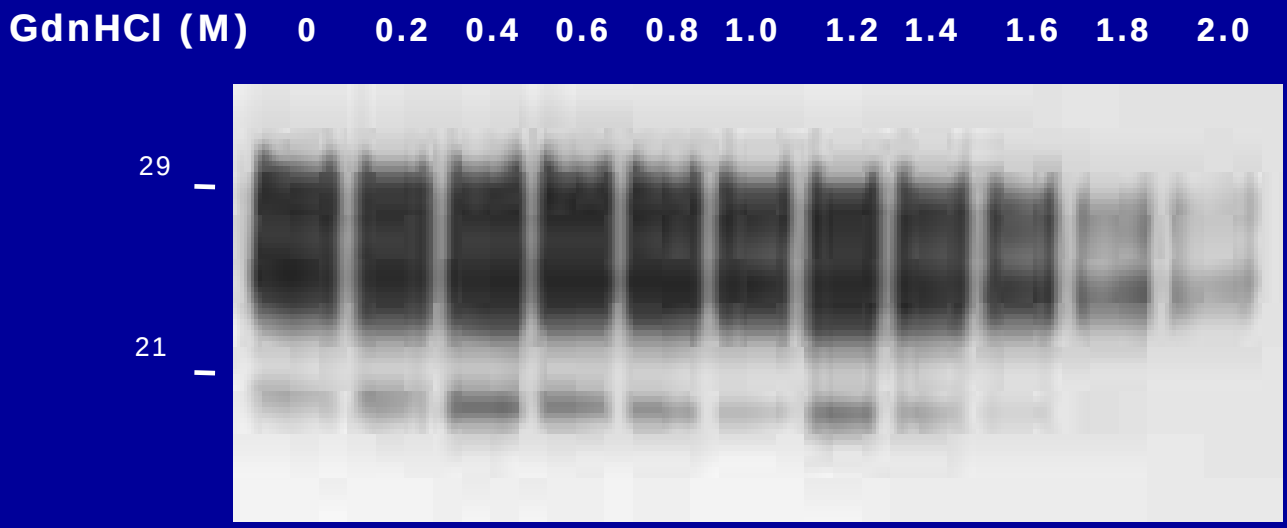
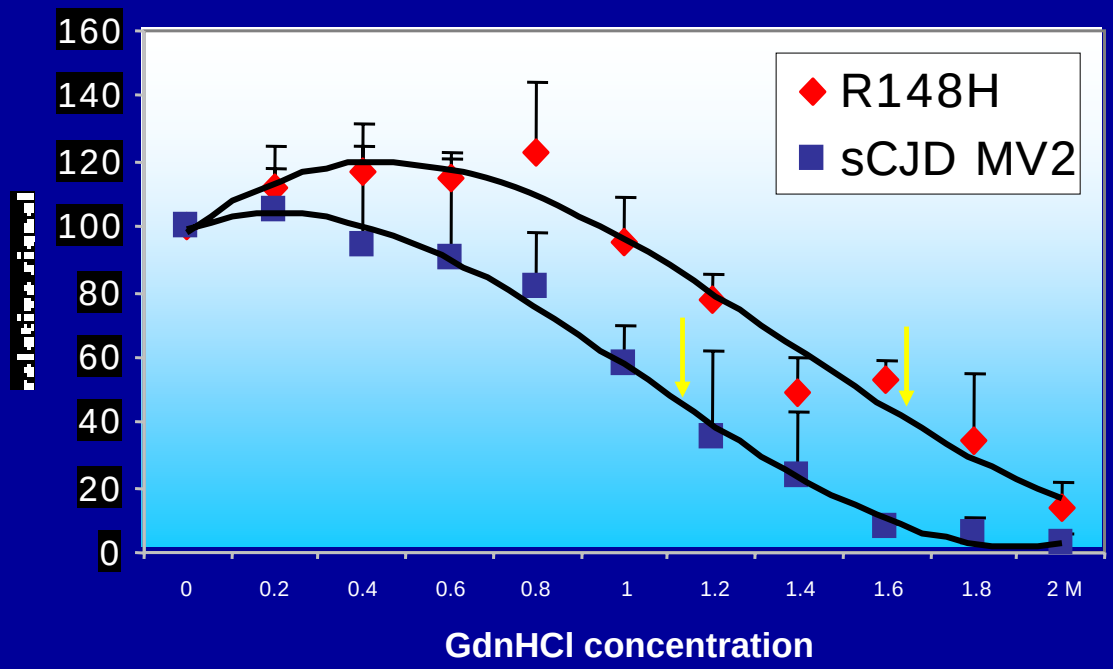
2002-383

Two-Dimensional Immunoblots of PK-resistant PrPSc in Human sCJD MM1 and Elk CWD



Conformation stability assay





Conclusions

The threat to public health and the complexity of prion diseases made it necessary to establish Prion Surveillance Centers to:

1. Timely identify cases due to exogenous infection in order to limit spread
2. monitor all cases of prion disease to limit possible sources of infection
3. Collect and store tissues for research

National Prion Disease Pathology Surveillance Center

Supported by the
Centers for Disease Control & Prevention (CDC)
Sponsored by the
American Association of Neuropathologists (AANP)

National Prion Disease Pathology Surveillance Center

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Dr. Mark Cohen *Histopathology*
Dr. Clive Hamlin *Biosafety & Quality Control*
Dr. Robert Petersen *Consultant Genetics*
Dr. Man-Sun Sy *Consultant Immunotesting*
Ms. Carrie Harris *Center Manager*

Jan 2005

National Prion Disease Pathology Surveillance Center
Cases Examined

Year	Referrals	Prion D. Total	Sporadic	Familial	Iatrogenic	vCJD
1997	104	60	54	6	0	0
1998	94	51	44	6	1	0
1999	114	74	65	9	0	0
2000	169	111	97	12	2	0
2001	247	154	138	16	0	0
2002	265[1]	151	127[1]	22	1	1[2]
2003	284[3]	191[4]	142	45	1	0
2004	351[5]	192[6]	138	19	0	0
Total	1628	984	805	135	5	1

[1] Includes 2 inconclusive
[2] Acquired in United Kingdom
[3] Includes 1 inconclusive
[4] Includes 3 type unknown
[5] Includes 8 pending
[6] Includes 6 type unknown, 26 type pending

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CSF Examined

Year	Referrals	14-3-3 Positive	14-3-3 Negative
1997	13	5	8
1998	34	11	23
1999	74	27	47
2000	130	64	76
2001	202	95	107
2002	313	107	206
2003	292	81	77
2004	514	125	182
Total	1,572	515	726

Cases examined by the NPDPSC (n=561) classified according to Parchi et al 1999

Phenotype	Variant	Distribution (%) Center	Parchi
Creutzfeldt-Jakob disease	M/M & M/V1 Classical	324 (58%)	71%
	V/V1 Early onset	24 (4%)	1%
	M/M2 Coarse spongiosis, long duration, cortical pathol.	48 (9%)	2%
	M/V2 Ataxia, kuru plaques, long duration, subcort. pathol.	75 (13%)	17%
	V/V2 Ataxia, plaque-like, short duration, subcort. pathol.	83 (15%)	7%
Fatal Insomnia	M/M2 Similar to FFI	7 (1%)	2%

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Mutations Identified
1997 – January 2005

Mutation	No	Mutation	No
E200K-129M/V	60	G114V-129M	1
D178N-129M/V	13	G94S-129V[3]	1
Insert-129M/V	13	A118V[3]	1
A117V-129V[1]	11	R148H-129M	1
P102L-129M/V	12	N171S-129V	1
V210I-129M[2]	9	T188K-129V	1
F198S-129V[1,3]	6	Q217R-129M/V	1
T183A-129M/V	2	2Rep.Del-129M	1
H187R-129M/V	2	H208R – 129M	1

Contributed by: 1 B. Ghetti, 2 J. Mastrianni and 3 S. DeArmond

Note: New mutations are underlined.

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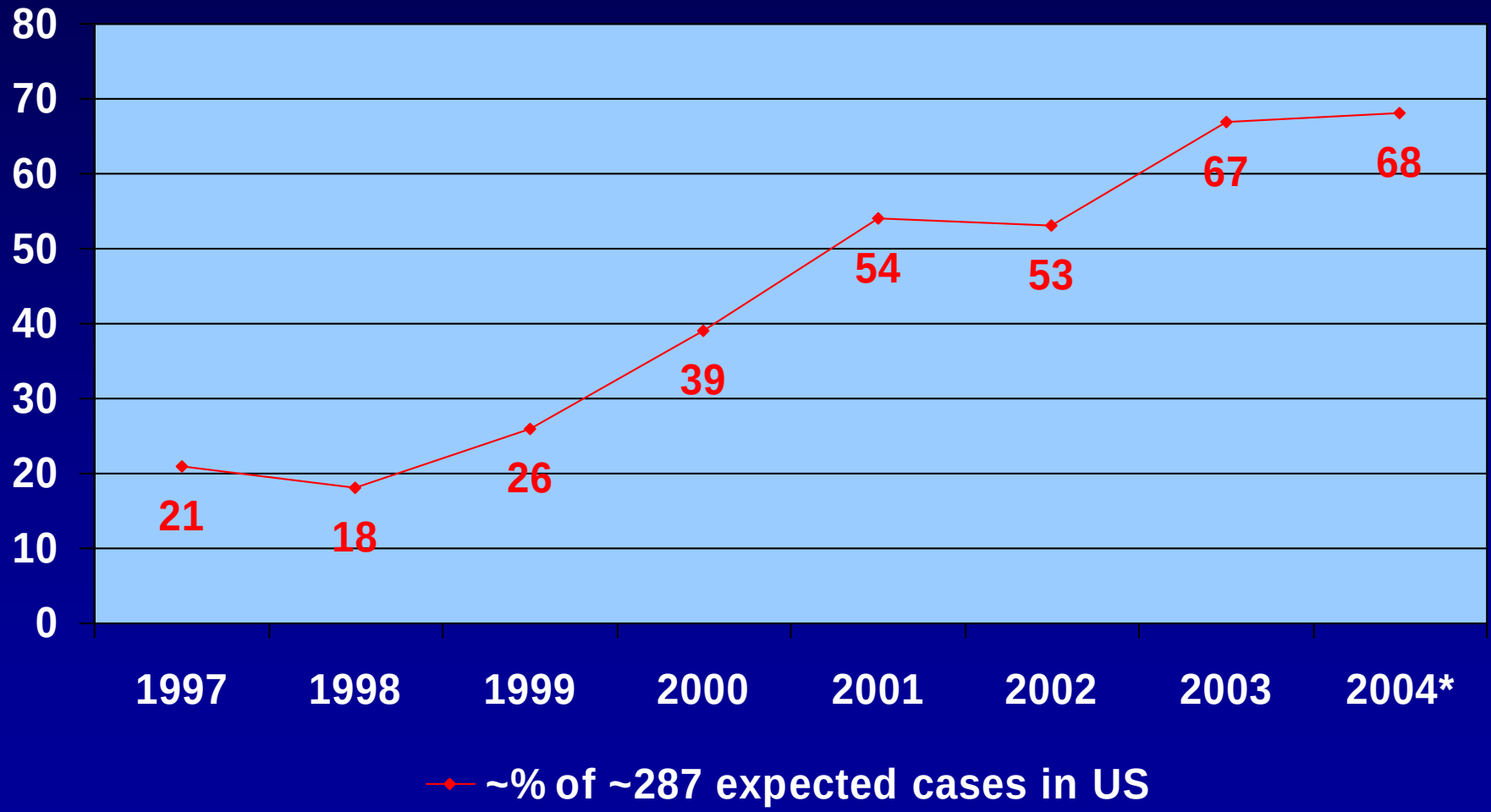
National Prion Disease Pathology Surveillance Center
Cases Examined as Percent of Cases Expected



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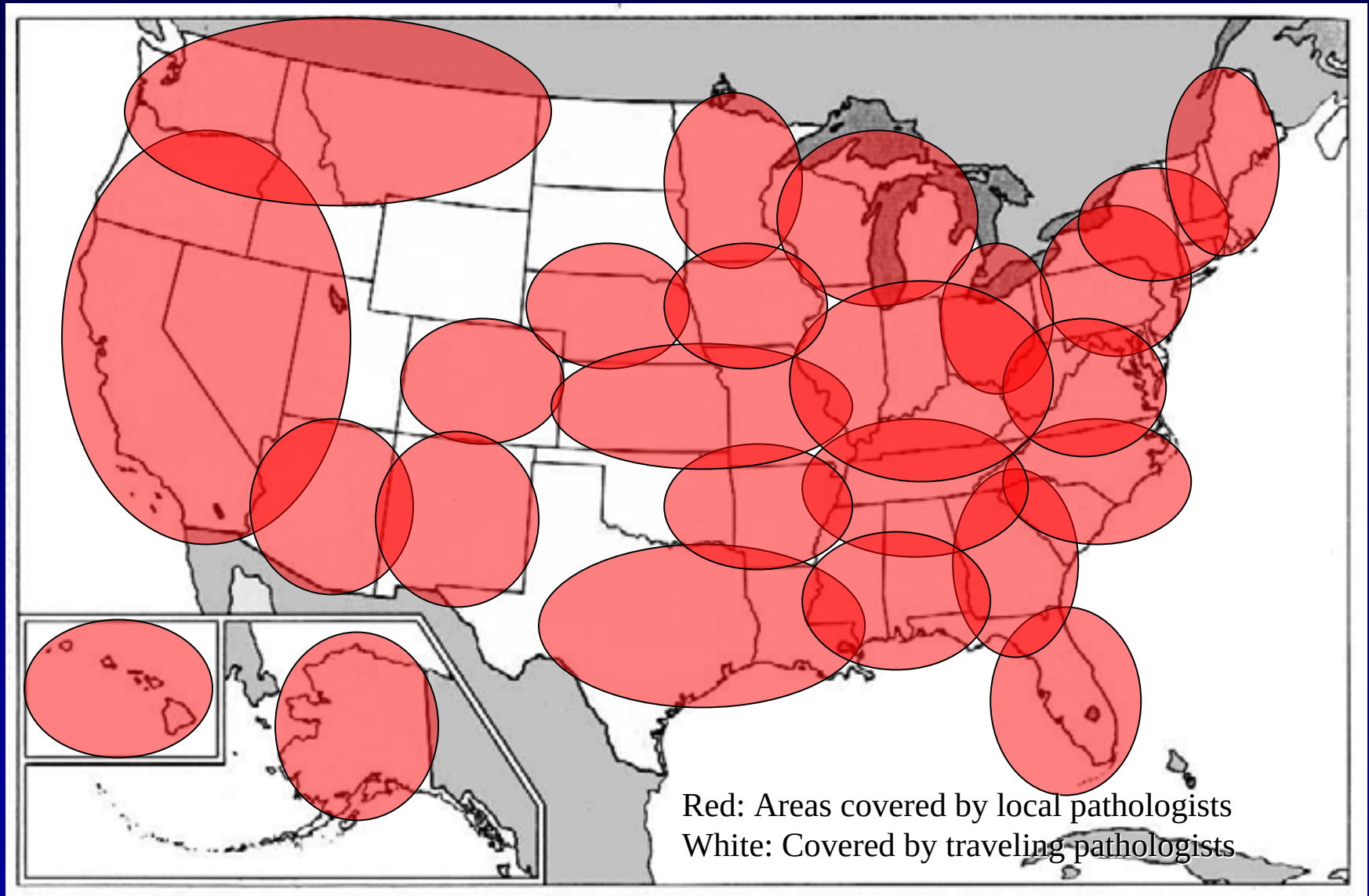
*2004 figures are preliminary.

Goals of Human Prion Surveillance

Overall goal is to timely detect all possible sources of prion infection by:

1. Monitoring the incidence of all prion diseases
2. Characterize and classify as accurately as possible all prion diseases to identify sporadic, familial, and transmitted cases
3. Deal with “atypical cases”
4. Update and develop techniques

National Autopsy Network of the Surveillance Center



We have established a network of institutions that perform reimbursed CJD autopsies on request. But we need the help of the State Health Departments to make the network more effective.

Cases/Million Received by the NPDPSC by State (2003-2004)

- 0.0 to 0.1 cases per million population: Idaho, Nevada, South Dakota, Vermont, West Virginia
- 0.2 to 0.3 cases per million population: Alabama, Illinois, Indiana, Louisiana, North Carolina, Utah
- 0.4 to 0.5 cases per million population: Arkansas, Georgia, Iowa, Kentucky, Mississippi, Rhode Island, South Carolina, Tennessee, Texas
- 0.6 to 0.7 cases per million population: Arizona, Connecticut, Delaware, Florida, Kansas, Maryland, Michigan, Minnesota, Nebraska, New Jersey, Oklahoma, Oregon, Pennsylvania
- 0.8 to 0.9 cases per million population: Alaska, California, Colorado, Hawaii, Massachusetts, Missouri, New Hampshire, New York, North Dakota, Ohio, Virginia, Wisconsin
- 1.0 or more cases per million population: Maine, Montana, New Mexico, Washington, Wyoming

*States where CJD is reportable are printed in yellow. States which have sent letters encouraging neurologists to report cases to the NPDPSC are underlined.

State of origin was unavailable in 30 out of 383 cases.

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NPDPSC Cases/Million According to High and Low Referral States (2003-2004)

- 0.0 to 0.5 cases per million population: Total 20 States; 11 (55%) with CJD reportable; 3 (15%) with mailing
- 0.6 to 1.0 or more cases per million population: Total 30 States; 20 (67%) with CJD reportable; 10 (33%) with mailing

National Prion Disease Pathology Surveillance Center

Corrective Measures

- We need to increase reporting by raising the level of awareness especially of the Neurologists
- The Departments of Health of 44 States in collaboration with the NPDPSC have written (or agreed to write) to the Neurologists urging them to report all cases of suspected prion disease at the time of diagnosis so they can be followed and autopsied
- These initiatives may be more effective in the States where prion diseases are reportable
- We also need national clinical diagnostic criteria for probable and possible prion disease

Surveillance Center

I wish to thank...

CDC Drs. Lawrence Schoemberger and Ermias Belay

AANP Past-President Dr. Bernardino Ghetti

AAN Drs. Stanley Fahn, Raymond Roos,

Sami Harik, Richard Johnson

NIH

CJD Foundation Florence Kranitz, Mayra Lichter

CJD Insight

The Department of Health of many States, especially NYS, NYC and Ohio DOHs

And many, many other supporters