# Dogs Never Get Prion Diseases. The Entropic Landscape Analysis of Prion Proteins Answers Why.

Kentaro ONIZUKA
PsiPhiFoldings Co. Ltd
onizuka.kentaro@jp.panasonic.com

**Abstract**

The Entropic Landscape Analysis was applied to the prion protein sequences of various mammals in order to detect potential sites of variants that would be responsible for the susceptibility of prion disease infection. Among familiar mammals, canines including dogs have been demonstrating strong resistance to prion diseases. Among the canine specific substitutions the entropic landscape analysis pinpoints the substitutions Asn104Gly and Ser107Asn having the biggest impact to the conformational transition and stability. Although they must be further corroborated by experiments in vivo et vitro, the results are demonstrating that the entropic landscape analysis is useful enough to screen substitutions and polymorphisms potentially relevant to conformational stability and transition because the calculation time for the analysis is as long as a few seconds, and the analysis can be done without knowing the 3D structures.

## 1 Introduction

Given a protein sequence of amino-residues, the entropy of the sequence fragments of any length and position within the given sequence can be estimated without knowing the 3D structure of the protein, and the set of calculated entropies over all possible fragments constitutes the Entropic Landscape of the given protein sequence[3]. Although the method for the entropic landscape analysis was originally devised to predict protein folding pathways which would be in turn informative for de-novo protein structure prediction [2], the studies to which the entropic landscape analysis could be applied would not be confined solely to the folding pathway prediction.

The entropy of a sequence fragment itself is closely related to the conformational stability of the fragment, and would suggest whether the fragment is flexible or rigid in the native conformation[1][3]. When the analysis is applied to the variants of a particular protein, the entropic landscape could pinpoint which variant would be the most relevant to the structural stability and which variant the most significant for the protein to transform its structure.

One of the most famous cases of protein structure transformation is regarding prion diseases (TSE). Most prion diseases are considered to be caused by the prion structure's transition from $\alpha$ helix-rich conformation to $\beta$ sheet-rich in brain tissues deteriorating the brain and nerve system functions. Although the best-known prion disease is BSE after its outbreak in the UK, prion diseases are indeed not confined to humans and cattle. Various livestocks and wild mammals are reported to be pron to the diseases. There are, however, some exceptions among familiar species: canine mammals including dogs (canis familialis) have never been reported to get prion diseases[7]. After prion's amino-residue sequences of various animals were determined, couple of studies have been published about the canine prion's difference from that of other species. Among various variants of mammalian prion sequence, the canine prion has some unique substitutions which are suspected to be responsible for canine resistance to prion diseases[4, 5].

In this study, the entropic landscape analysis was applied to the prion protein sequences of various mammals, in order to pinpoint which variant(s) could be responsible for the canine resistance to prion diseases.



# 2 Results

The alignments of whole length mammalian prion sequences are shown in the following pages (split into five fragments), where intra-canine variants are marked with `+`, and canine specific substitutions to other mammalian sequences are marked with `*`. Because the representative dog prion sequence "`O46501` Canis Familialis (dog)" is very different from other canine prion sequences, it was eliminated from further analysis. The canine specific substitutions are Asn104Gly, Ser107Asn, Asn163Asp/Glu, His181Arg, and Ser241Pro. To see the difference between entropic landscape of canine prion sequences and those of other mammals, a special prion sequence of *virtual canine ancestor*(VCA) was introduced which is identical to canine sequences except for the canine specific substitutions which are set to normal mammalian types, thus 104th residue is Asn, 107th Ser, 163th Asn, 181st His, and 241st Ser. For the two intra-canine variants (Gly101Ser and Asp163Glu), two canine sequence were used, where one is O46593 Dog (Ser for 101st and Glu for 163d), and the other is the canine consensus sequences (Gly for 101st and Aspfor 163d). The intra-canine variant Gly101Ser of the sequence of VCA is set for Gly because it is Gly in bovine sequence, where that of human and sheep is Ser, and is Asn in sequence of mouse, rat and rabbit.

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1-59                 ___________________________________________________________
                     _________1_________2_________3__ _______4_________5_________
                     12345678901234567890123456789012 34567890123456789012345678

A1YVW4 Dog           MVKSHIGGWILLLFVATWSDVGLCKKRPKPGG WNTGGGSRYPGQGSPGGNRYPPQGGGG
O46593 Dog           MVKSHIGGWILLLFVATWSDVGLCKKRPKPGG WNTGGGSRYPGQGSPGGNRYPPQGGGG
Q1W2J9 Dog           MVKSHIGGWILLLFVATWSDVGLCKKRPKPGG WNTGGGSRYPGQGSPGGNRYPPQGGGG
A5JUM5 Red fox       MVKSHIGGWILLLFVATWSDVGLCKKRPKPGG WNTGGGSRYPGQGSPGGNRYPPQGGGG
B0FYL5 Arctic fox    MVKSHIGGWILLLFVATWSDVGLCKKRPKPGG WNTGGGSRYPGQGSPGGNRYPPQGGGG
B7SKY3 Kit Fox       MVKSHIGGWILLLFVATWSDVGLCKKRPKPGG WNTGGGSRYPGQGSPGGNRYPPQGGGG
B7SKW7 Gray wolf     MVKSHIGGWILLLFVATWSDVGLCKKRPKPGG WNTGGGSRYPGQGSPGGNRYPPQGGGG
B7SKX7 Raccoon dog   MVKSHIGGWILLLFVATWSDVGLCKKRPKPGG WNTGGGSRYPGQGSPGGNRYPPQGGGG

O18754 Cat           MVKSHIGSWILVLFVAMWSDVGLCKKRPKPGGGWNTGG SRYPGQGSPGGNRYPPQGGGG
P04156 Human         MA  NLGCWMLVLFVATWSDLGLCKKRPKPGG WNTGG SRYPGQGSPGGNRYPPQGGGG
P04925 Mouse         MA  NLGYWLLALFVTMWTDVGLCKKRPKPGG WNTGG SRYPGQGSPGGNRYPPQGG T
P13852 Rat           MA  NLGYWLLALFVTTCTDVGLCKKRPKPGG WNTGG SRYPGQGSPGGNRYPPQSGGT
Q95211 Rabbit        MA  HLGYWMLLLFVATWSDVGLCKKRPKPGGGWNTGG SRYPGQSSPGGNRYPPQGGG
P49927 Pig           MVKSHIGGWILVLFVAAWSDIGLCKKRPKPGGGWNTGG SRYPGQGSPGGNRYPPQGGGG
P10279 Bovine        MVKSHIGSWILVLFVAMWSDVGLCKKRPKPGGGWNTGG SRYPGQGSPGGNRYPPQGGGG
P23907 Sheep         MVKSHIGSWILVLFVAMWSDVGLCKKRPKPGGGWNTGG SRYPGQGSPGGNRYPPQGGGG

60-109               ____________________________________        ____1_________
                     6_________7_________8_________9_____        ____0_________
                     01234567890123456789012345678901234        67890123456789
                                                                      +
A1YVW4 Dog           WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHGQWGKPNKP
O46593 Dog           WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHSQWGKPNKP
Q1W2J9 Dog           WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHSQWGKPNKP
A5JUM5 Red fox       WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHGQWGKPNKP
B0FYL5 Arctic fox    WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHGQWGKPNKP
B7SKY3 Kit Fox       WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHGQWGKPNKP
B7SKW7 Gray wolf     WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHGQWGKPNKP
B7SKX7 Raccoon dog   WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHGQWGKPNKP
                                                                        *  *
O18754 Cat           WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ         GGSHSQWNKPSKP
P04156 Human         WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGG WGQ        GGGTHSQWNKPSKP
P04925 Mouse         WGQPHGGGWGQPHGGSWGQPHGGSWGQPHGGG WGQ        GGGTHNQWNKPSKP
P13852 Rat           WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGG WSQ        GGGTHNQWNKPSKP
Q95211 Rabbit        WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGG WGQ         GGTHNQWGKPSKP
P49927 Pig           WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ        GGGSHGQWNKPSKP
P10279 Bovine        WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGG WGQPHGGGGWGQGGTHGQWNKPSKP
P23907 Sheep         WGQPHGGGWGQPHGGGWGQPHGGGWGQPHGGGGWGQ         GGSHSQWNKPSKP
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110-169                 ____________________________________________________________
                        1_________2_________3_________4_________5_________6_________
                        01234567890123456789012345678901234567890123456789012345678
                                                                                +
A1YVW4 Dog              KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPEQVYYRP
O46593 Dog              KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPEQVYYRP
Q1W2J9 Dog              KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPDQVYYRP
A5JUM5 Red fox          KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPDQVYYRP
B0FYL5 Arctic fox       KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPDQVYYRP
B7SKY3 Kit Fox          KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPDQVYYRP
B7SKW7 Gray wolf        KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPDQVYYRP
B7SKX7 Raccoon dog      KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPDQVYYRP
                                                                              *
O18754 Cat              KTNMKHMAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPNQVYYRP
P04156 Human            KTNMKHMAGAAAAGAVVGGLGGYMLGSAMSRPIIHFGSDYEDRYYRENMHRYPNQVYYRP
P04925 Mouse            KTNLKHVAGAAAAGAVVGGLGGYMLGSAMSRPMIHFGNDWEDRYYRENMYRYPNQVYYRP
P13852 Rat              KTNLKHVAGAAAAGAVVGGLGGYMLGSAMSRPMLHFGNDWEDRYYRENMYRYPNQVYYRP
Q95211 Rabbit           KTSMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPNQVYYRP
P49927 Pig              KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGSDYEDRYYRENMYRYPNQVYYRP
P10279 Bovine           KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGSDYEDRYYRENMHRYPNQVYYRP
P23907 Sheep            KTNMKHVAGAAAAGAVVGGLGGYMLGSAMSRPLIHFGNDYEDRYYRENMYRYPNQVYYRP

170-229                 ______________________________ 2______________________________
                        7_________8_________9_________0_________1_________2_________
                        01234567890123456789012345678901234567890123456789012345678

A1YVW4 Dog              VDQYSNQNNFVRDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
O46593 Dog              VDQYSNQNNFVRDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
Q1W2J9 Dog              VDQYSNQNNFVRDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
A5JUM5 Red fox          VDQYSNQNNFVRDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
B0FYL5 Arctic fox       VDQYSNQNNFVRDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
B7SKY3 Kit Fox          VDQYSNQNNFVRDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
B7SKW7 Gray wolf        VDQYSNQNNFVRDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
B7SKX7 Raccoon dog      VDQYSNQNNFVRDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
                                  *
O18754 Cat              VDQYSNQNNFVHDCVNITVKQHTVTTTTKGENFTETDMKIMERVVEQMCVTQYQKESEAY
P04156 Human            MDEYSNQNNFVHDCVNITIKQHTVTTTTKGENFTETDVKMMERVVEQMCITQYERESQAY
P04925 Mouse            VDQYSNQNNFVHDCVNITIKQHTVTTTTKGENFTETDVKMMERVVEQMCVTQYQKESQAY
P13852 Rat              VDQYSNQNNFVHDCVNITIKQHTVTTTTKGENFTETDVKMMERVVEQMCVTQYQKESQAY
Q95211 Rabbit           VDQYSNQNSFVHDCVNITVKQHTVTTTTKGENFTETDIKIMERVVEQMCITQYQQESQAA
P49927 Pig              VDQYSNQNSFVHDCVNITVKQHTVTTTTKGENFTETDVKMIERVVEQMCITQYQKEYEAY
P10279 Bovine           VDQYSNQNNFVHDCVNITVKEHTVTTTTKGENFTETDIKMMERVVEQMCITQYQRESQAY
P23907 Sheep            VDRYSNQNNFVHDCVNITVKQHTVTTTTKGENFTETDIKIMERVVEQMCITQYQRESQAY
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230-257                  __ ____ ______________________
                         3_ ____ ____4_________5_______
                         01 2345 6789012345678901234567

A1YVW4 Dog               YQ RGAS AILFSPPPVILLISLLILLIVG
O46593 Dog               YQ RGAS AILFSPPPVILLISLLILLIVG
Q1W2J9 Dog               YQ RGAS AILFSPPPVILLISLLILLIVG
A5JUM5 Red fox           YQ RGAS AILFSPPPVILLISLLILLIVG
B0FYL5 Arctic fox        YQ RGAS AILFSPPPVILLISLLILLIVG
B7SKY3 Kit Fox           YQ RGAS AILFSPPPVILLISLLILLIVG
B7SKW7 Gray wolf         YQ RGAS AILFSPPPVILLISLLILLIVG
B7SKX7 Raccoon dog       YQ RGAS AILFSPPPVILLISLLILLIVG
                                    *
O18754 Cat               YQ RRAS AILFSSPPVILLISFLIFLIVG
P04156 Human             YQ RGSS MVLFSSPPVILLISFLIFLIVG
P04925 Mouse             YDGRRSSSTVLFSSPPVILLISFLIFLIVG
P13852 Rat               YDGRRSS AVLFSSPPVILLISFLIFLIVG
Q95211 Rabbit            YQ RAAG VLLFSSPPVILLISFLIFLIVG
P49927 Pig               AQ RGAS VILFSSPPVILLISFLLFLIVG
P10279 Bovine            YQ RGAS VILFSSPPVILLISFLIFLIVG
P23907 Sheep             YQ RGAS VILFSSPPVILLISFLIFLIVG
```

Fig 1 shows the entropic landscapes of VCA vs two canine variant 101Gly163Asp and 101Ser163Glu. When entropies of VCA agree wtih those of two canine variants, the curves are blue in colour, while entropies of both variants have the same value which is different from that of VCA, green curves are visible. If two variants disagree with each other and both variants have different values from those of VCA, the red curve becomes visible. Hence red curves are visible around 100-110 (corresponding to 101Gly/Ser and Asn104Gly, Ser107Asn) and 160-170 (corresponding to Asn163Asp/Glu). For other substitutions, His181Arg and Ser241Pro, green curves are visible but not red ones.

The parameter $k$ indicates the length of the sequential fragments whose entropy is calculated. When $k = 4$ the length of the fragment is $k + 1 = 5$.

The entropic landscape of the prion protein sequence by absolute method shows that the N-terminal half has high entropy while the C-terminal half has low entropy. The entropic landscapes by the net and cross methods are completely opposit to that by the absolute method in this respect. These results agree with the 3D structure by NMR, which shows the N-termal half is disordered. In general, sequential regions with low entropy by the absolute method are stable in the final fold usually stabilized by abundant hydrogen-bonds and hydrophobic interactions exerted from surrounding structures. On the contrally those regions with low entropy by the net or cross method usually have strong desire to form particular conformations by themselves, which in some cases leads those sequential regions to end up being disordered, forming loop, or in case of short regions making turn structures, because they want to form irregular structures.

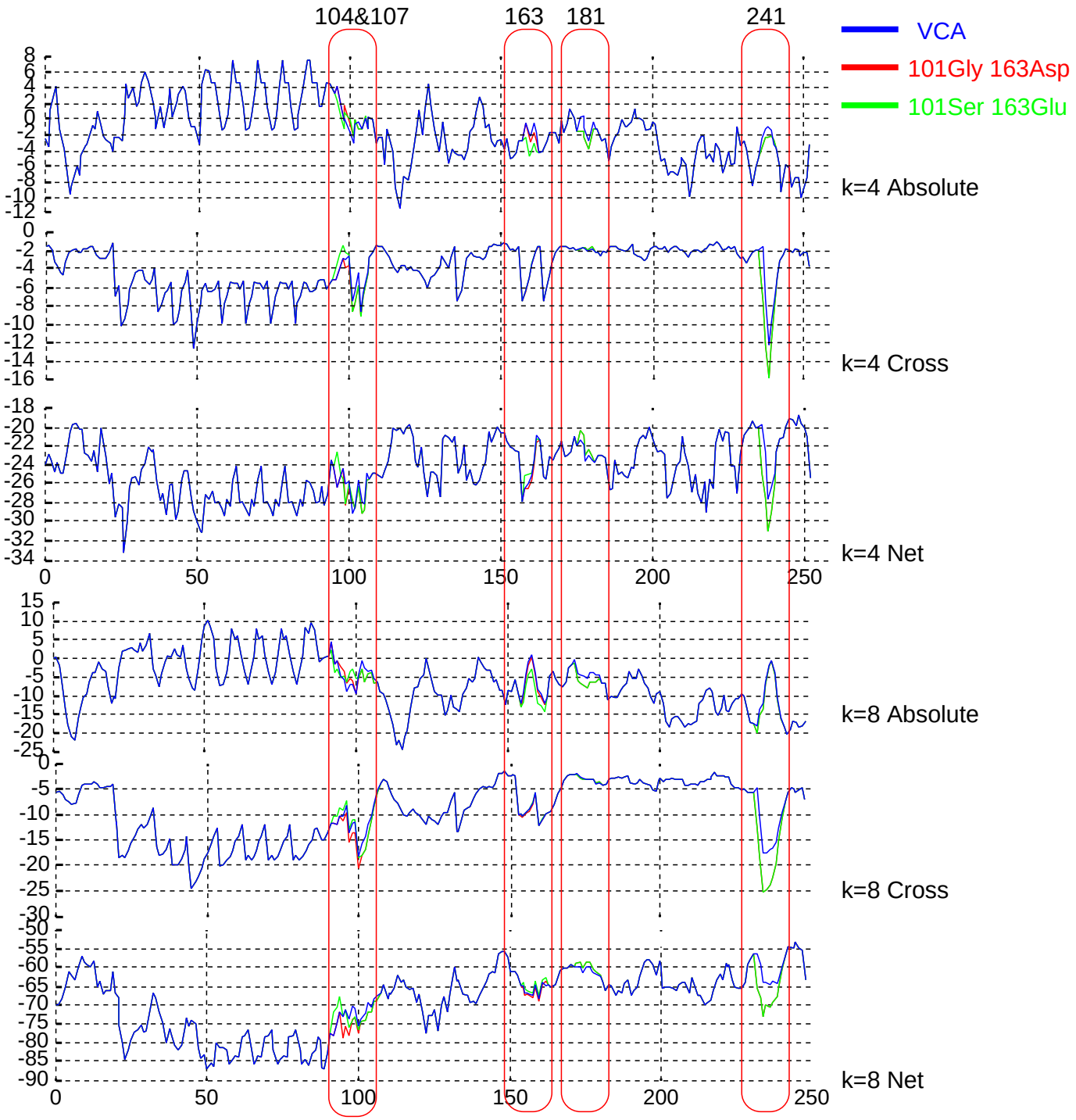


Figure 1: The Entropic Landscapes of Prions Canine (Red/Green) vs. Virtual Canine Ancestor (Blue).

The difference between landscapes of canine sequence and that of other mammals represented by the VCA sequence is obvious. The sites for the canine specific substitutions have different entropies from those of VCA's. Among those it is clear that the entropies around 100 to 110 corresponding to the substitutions, Asn104Gly and Ser107Asn, have the biggest difference in the landscapes by any methods, particularly in the landscape by the net method, when substitution Ser241Pro is put aside because the site is within the C-terminal region that is removed before folding. This result alone could lead us to conclude that the substitions Asn104Gly and Ser107Asn have the biggest impact to the conformational stability and transitability. The substition Asn163Asp/Glu should be eliminated from the list of suspected responsible sites for canine resistance to prion-diseases because in the case of Asn163Glu, the difference in entropy from the normal type (VCA) is just slight. The substitution His181Arg could have bigger impact than either Asn104Gly or Ser107Asn alone. But it cannot beat the combination of the two substitutions.



# 3 Discussion

The entropic landscape analysis is a very cheep method compared to other experimental or computational methods to screen potential significant sites of regional stability and transformation of protein structures. It took just less than three hours to pinpoint two substitutions (Asn104Gly and Ser107Asn) seemingly having biggest impact to conformational stability and transitability among the canine prion sequence. And the most of that three hours was spent to get prion sequences via the Internet, and to modify the sequence files into suitable forms that can be fed to the entropic landscape calculation system. The net time for calculating the entropic landscape was as long as 5 to 10 seconds for each sequence, using a PC with a single 2GHz Pentium processor (8 years old, powered by Linux). And again most of that 5 to 10 seconds was spent to take and load the entropy table as input from HD to RAM. The entropy table had been prepared before the sequence analysis. It takes less than an hour to compile over 1500 PDB entry files, into statistical data, and it takes a few minutes to prepare the entropy table from the statistical data.

The result from the entropic landscape analysis that two substitutions, Asn104Gly and Ser107Asn, of the dog prion sequence would be responsible for the canine resistance to prion diseases is implicitly supported both by computational and experimental studies. In a computational study, the molecular dynamics simulation of human prion protein for the wild type and mutant with Pro102Leu substitution (which is equivalent to Pro106Leu in canine sequence) showed that the mutant human prion is more prone to the transition into $\beta$-sheet rich structure although the site is located within the disordered region[8]. For experimental studies, rabbits' substitution Asn104Gly (numbering in the canine prion sequence) was introduced to the mouse prion sequence, which showed resistance to prion transmission, though the authors concluded that the substitution alone would play part of the role in rabbits' resistance to prion transmission[9].

The site of residues 104-107 where canine substitutions are located has various variants. For most of the species, `NKPS` is dominant but canine mammals have `GKPN`. If the prion-disease resistance affected the evolution of mammals, the natural selection would have favoured the

prion disease resistance more for carnivores than for herbivores because carnivores feeding on animals must have been, in theory, more exposed to prion transmission. In this respect, canine resistance to prion diseases is natural because they are originally carnivores. Then cats should also show the resistance because they are also carnivores. Although the sequence of cat (felis silvestris catus) has the dominant type NKPS, some big cat sequences have unique variants. The Siberian tigers' (panthera tigris altaica) sequence has NQPS, the lions' (panthera leo), the mountain tigers' (puma concolor) and Cheetahs' (acinonyx jabatus) have GKPS which is identical to the rabbits'. In case of rabbits, their GKPS is suspected to be responsible for their resistance to prion diseases as mentioned above[9], though some minks who are known to be susceptible to prion diseases also have GKPS.

Prion diseases have a lot to do with longevity because it takes a long time for abnormal prion to accumulate in brain tissues. Therefore among carnivores, big carnivores which feed on big mammals (which should be old enough to grow big) are more exposed to prion transmission. Common cats which feed on small animals and thus must have been less exposed to abnormal prion transmission may have normal type NKPS sequence while big cats like lions and tigers could have been more favoured to be resistant to prion diseases. Although the responsibility of GKPS for the prion disease resistance is questionable when the case of minks is considered, small difference in prion disease susceptibility might have affected the natural selection of the lineage, thus natural is that lions and tigers have GKPS, though rabbits' resistance remains mysterious.

For other variants, some primates (particularly macaques and baboons) have HKPS, and mandrills have HKPN in their prion sequence, whose susceptibility to prion disease infection is unknown.

As a conclusion, I propose following hypotheses.

1) Canine mammals' strong resistance to prion diseases is mainly played out by GKPN at the site from 104th through 107th, where both of two substitutions Asn104Gly and Ser107Asn are cooperatively reinforce the resistance. 2) In case of rabbit, GKPS plays a certain role in the species' resistance to prion transmission, but other rabbit specific substitutions cooperatively reinforce the resistance. 3) Herbivores show less resistance to prion diseases probably because they are less exposed to prion transmission, while 4) carnivores developed prion-disease resistance independently through the evolution of their lineage, among which canines developed almost perfect prion disease resistance. 5) a mutation at the site from 104th to 107th except for 106th seems to be neutral in other respects than prion-disease resistance, because various variants are found int the prion sequence of broad range of mammals.

# 4 Methods and Meterials

The calculation method of entropic landscape has been detailed in my previous postings[2, 3] to the Nature Precedings. The methods specific to this study are listed below.

- $C^{\alpha}$-$C^{\alpha}$ interaction is considered dependent on the residue-types of both residues. Thus the method used $\rho^{ab}_{C^{\alpha}C^{\alpha}}(k, r)$.

- Since other atoms are considered sequence independent, the interaction between atoms that are not $C^{\alpha}$ was not taken into account. When one of the two atoms in interacting is CA, they are taken into account. Thus the method used $\rho^{ax}_{C^{\alpha}v}(k,r)$ and $\rho^{xb}_{uC^{\alpha}}(k,r)$, where $u$ and $v$ are not $C^{\alpha}$.

- Consequently, $\rho^{a}(\phi,\psi)$ nor $\rho^{x}(\phi,\psi)$ is not used in this study, but $\rho^{ab}(\psi,\omega,\psi)$ and $\rho^{xx}(\psi,\omega,\psi)$ are used.

These modifications to the methods of the previous study are mainly due to the stability of the results by the entropic landscape analysis. The absolute entropy is fairly stable for any choice of sequence-dependent atoms, while net and cross entropies are significantly affected by the choice. By seeing the relationship between experimentally revealed folding pathways and the entropic landscapes by net and cross method, the choice was exploited as above.

The computer software for this study, "PPF_eLandscape" package is commercially available.

The prion amino-acid sequences are obtained from UniProt web site.

- Canine prion sequences
  - O46501 Canis familialis (Dog)
    This sequence is very different from other canine mammals', rather closer to pig's.
  - A1YVW4 Canis familialis (Dog)
  - O46593 Canis familialis (Dog)
  - Q1W2J9 Canis familialis (Dog)
    Pekingese Dog sequence[4].
  - A5JUM5 Vulpes vulpes (Red fox)
  - B0FYL5 Vulpes lagops (Arctic fox)
  - B7SKY3 Vulpes velox (Kit fox)
  - B7SKW7 Canis lupus (Gray wolf)
  - B7SKX7 Nyctereutes procyonoides (Raccoon dog)
- O18754 Cat
- P04156 Human
- P04925 Mouse
- P13852 Rat
- Q95211 Rabbit
- P49927 Pig
- P10279 Bovine
- P23907 Sheep

# 5 Acknowledgements

I was very lucky when I was invited to give a talk about entropic landscape and found Sekijima gave a talk about his MD simulation of human prion protein before I made the presentation. I came up with the application of entropic analysis to prion sequences after I heard his talk two weeks ago. I also thank Akiyama who chaired the closed meeting of IPAB, and invited me to have an opportunity to give the talk.

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