IN SILICO SEARCH FOR POTENTIAL AMYLOIDOGENIC AND PRION-LIKE PLANT PROTEINS INDUCED BY X-RAY IRRADIATION

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Background of the research field

Amyloid proteins and their infectious subclass, prions, are best known in the context of incurable pathologies in humans and mammals [1]. Importantly, a nano-amount of a prion protein is sufficient for infectious activity (a "seed" aggregate may consist of just a few protein molecules) [2]. In 2016, amyloid prion-like proteins (PrLPs) were discovered in plants. Unlike prions, they are not associated with pathogenesis and can perform a variety of useful functions, such as seed storage and protection, maintenance of plant homeostasis under stress conditions, production of antioxidants, etc. [3]. These prion-like proteins have significant potential applications in agriculture, medicine, and new technologies and materials.

Background of the present work

Previous experiment [4]:

Pea seeds (*Pisum sativum* L.)

Effects of X-ray irradiation:

1. Slowed germination of irradiated seeds, but
2. accelerated growth, maturation and aging of plants grown from these seeds;
3. **An increased amount of proteins in a plant grown from irradiated seeds;**
4. **(Hypothesis!) An increased amount of amyloid proteins in a plant grown from irradiated seeds**

Objectives

• to check the hypothesis that irradiation of plant seeds gives rise to amyloidogenesis in plants grown from these seeds
• to check whether the proteins of plants grown from irradiated seeds have prion-like properties

Objects and methodology

A particularly large accumulation was found for 29 proteins, the objects of our study.

***In silico* screening with the PLAAC (Prion-Like Amino Acid Composition) program was used to find out whether there are any potential amyloid and prion-like proteins in the sample under study.**

Results and discussion

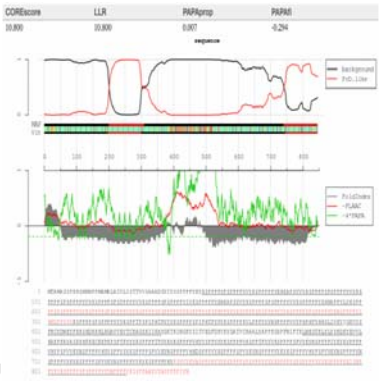
In silico detection of prion-like properties

Input: PLAAC → **Output:**

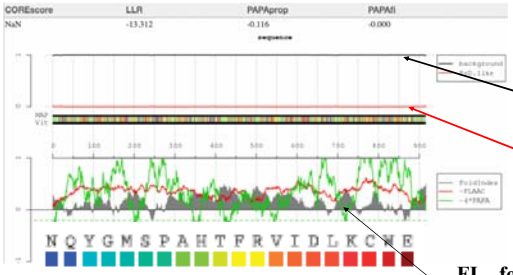
Primary protein structure

MDLNGNGNGNGILKVVDNVEESDVSFVRIIWVN
HSGQYKCRAIPRKYFYETVTKNGVALPPVTMV
RTSLLEKPAPGSGVDFVGEARLTPDLSTIKTIP
WCKQDEMVLGDLNVKPGQASEHCPRETFRR
VTKILKDEFDLIISTGFEVEFYLLKSIIREGKQEW
VEIDSSPYCCTSAFDVACPILREIASALQSIGIQIE
QLHVETGKGQFEVVLKHTICNKAVDNLVYTREI
IKAIARKHGLLATFVPKYNLDDAGSACHAHLSL
WQNGKNVFMASDESSKYGISTLGKEFMAGILY
HLSSILSFITPLPISYERLQSTTWGTYLFWGNEN
KEAPLRAASSFGTPDGLKSNFEIRLIDGCAN
PYFGSAIIAAGIDGLRRHLTLPEPIDIHPDLET
LERIPKSLSESLEALHKAKFLEEFFGNKLLTAIKA
IQKVEIDQYSKNKDAYKQLIHRY

Example: Extensin 2 domain-containing protein (involved in cell wall formation)



Comparison of results for potential non-prion and prion proteins



Non-prion-like protein

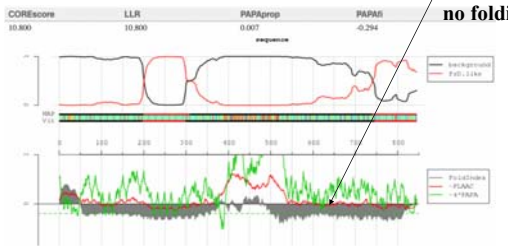
Probability of a non-prion state

Probability of the prion state

FI – folding index;
FI>0: folding/structural order

FI<0: no folding/structural disorder

Prion-likeness is mainly governed by the frequency of glutamine (Q) and asparagine (N) residues.



Prion-like protein

Criteria for classifying a protein as potential prion-like

1. **COREscore > 10 (chosen threshold)**
2. **There are domains with a highly probable prion state.**
3. **By the color scheme of amino acid composition (for initial qualitative assessment only)**

4 of the 29 proteins were found to be potential prion-like

Protein No.	Protein ID	Protein name	Functional group	CORE score
5.	Psat6g050240.1	Extensin 2 domain-containing protein	Cell wall biogenesis	10.800
24.	Psat5g239760.1	multiple organellar RNA editing factor (MORF)	RNA editing	30.702
25.	Psat1g122920.1	Catalytic component 1 of Argonaute RISC (AGO1)	RNA interference	22.683
29.	Psat7g158440.1	Ole e 1-like protein	vesicular trafficking	17.114

Proteins were found to be internally disordered in prion-like domains.

Conclusions

• Using the bioinformatic algorithms implemented in the PLAAC (Prion-Like Amino Acid Composition) program, 4 proteins from the sample of 29 proteins were found to be potential amyloidogenic and prion-like; ➔
• It was confirmed that irradiation of plant seeds gives rise to amyloidogenesis in plants grown from these seeds;
• It was established that prion-like properties are inherent in internally disordered proteins with a negative folding index value.

References **[1].** *Prusiner S.B.* Prions // PNAS.–1998.-**95**, N 23.-P. 13363-13383. **[2].** *Silveira J.R., Raymond G.J., Hughson A.G. et al.* The most infectious prion protein particles // Nature.-2005.-**437**, N 7056.-P. 257-261. **[3].** *Chakrabortee S., Kayatekin C., Newby G.A. et al.* Luminidependens (LD) is an Arabidopsis protein with prion behavior // PNAS.-2016.-**113**, N 21.-P. 6065-6070 **[4].** *Kryvokhyzha M., Litvinov S., Danchenko M. et al.* How does ionizing radiation affect amyloidogenesis in plants? // Int. J. Radiat. Biol.-2024.-**100**, N 6.-P. 922-933.