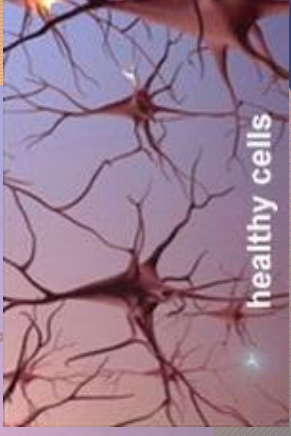
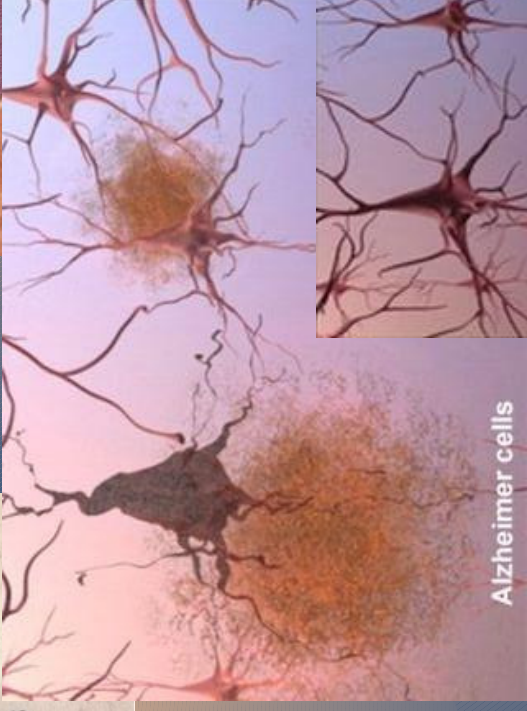
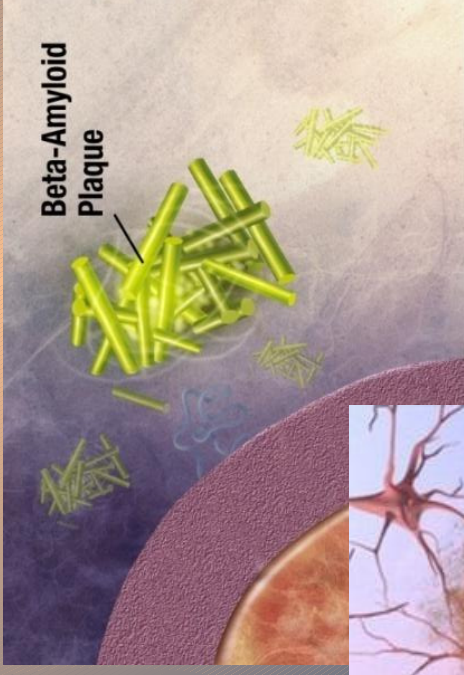
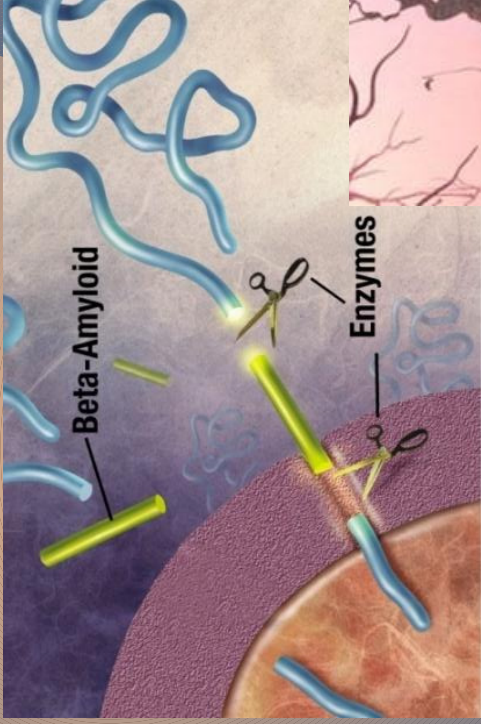


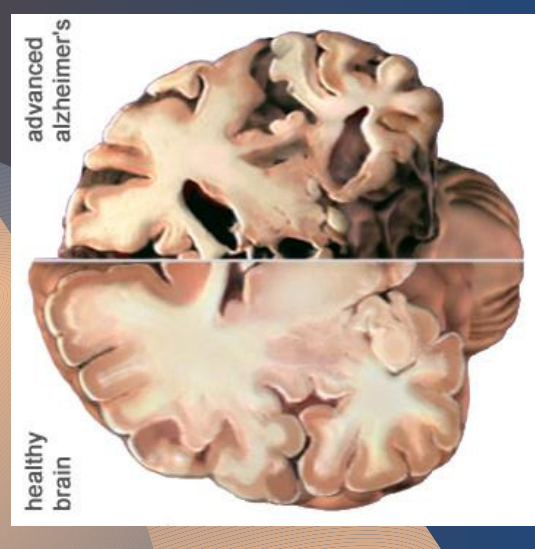
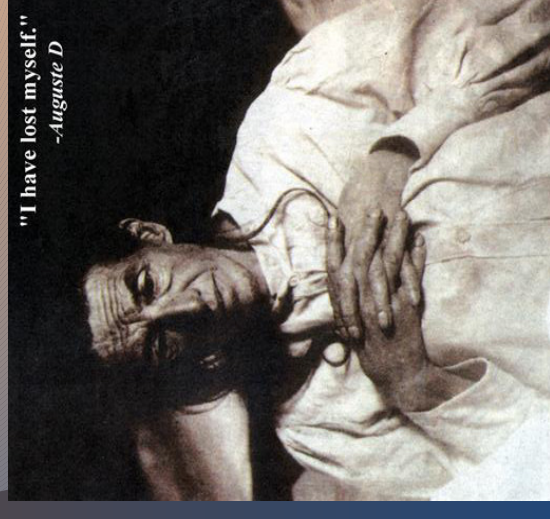
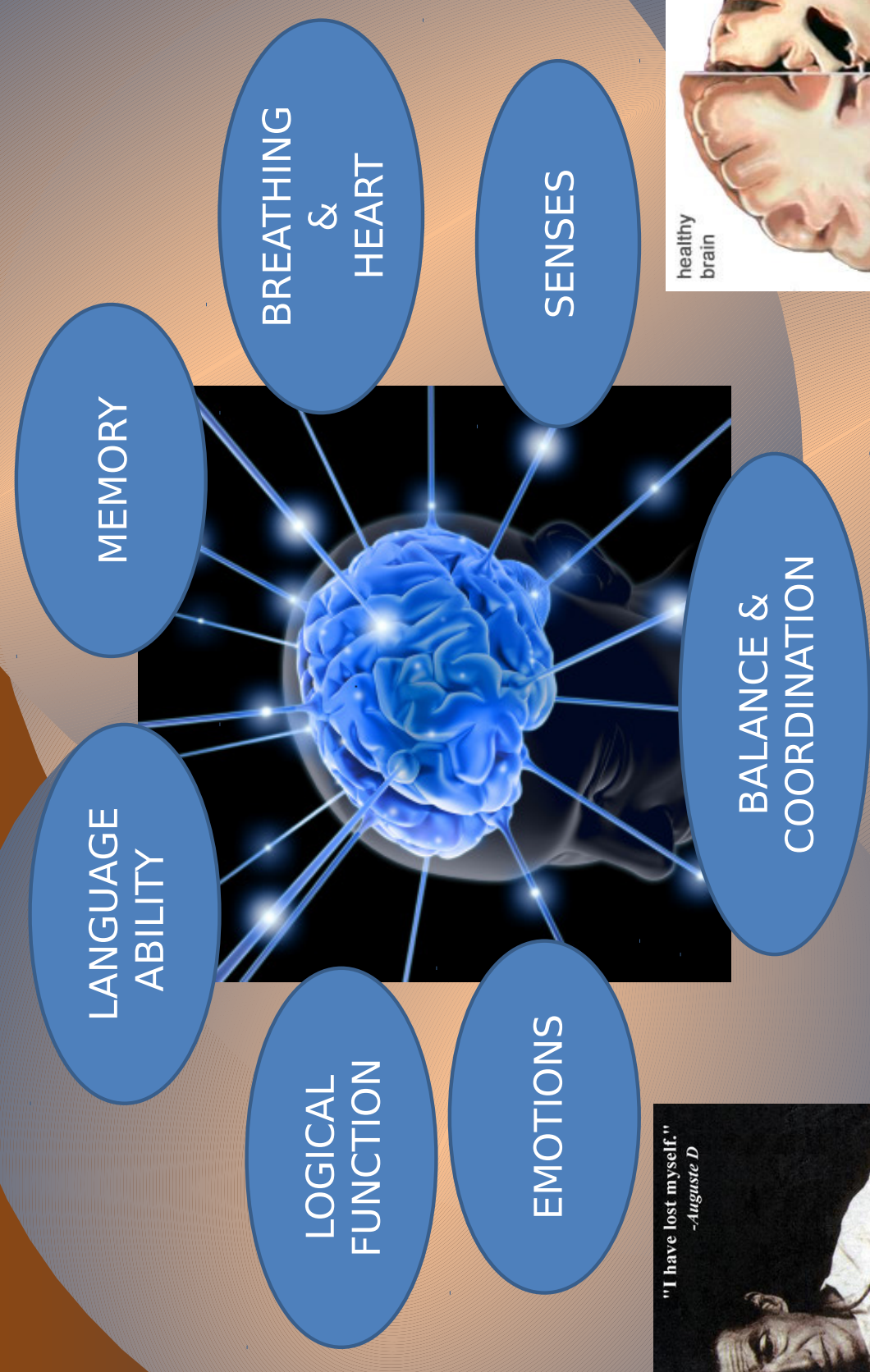


„Insight into beta amyloid protein”

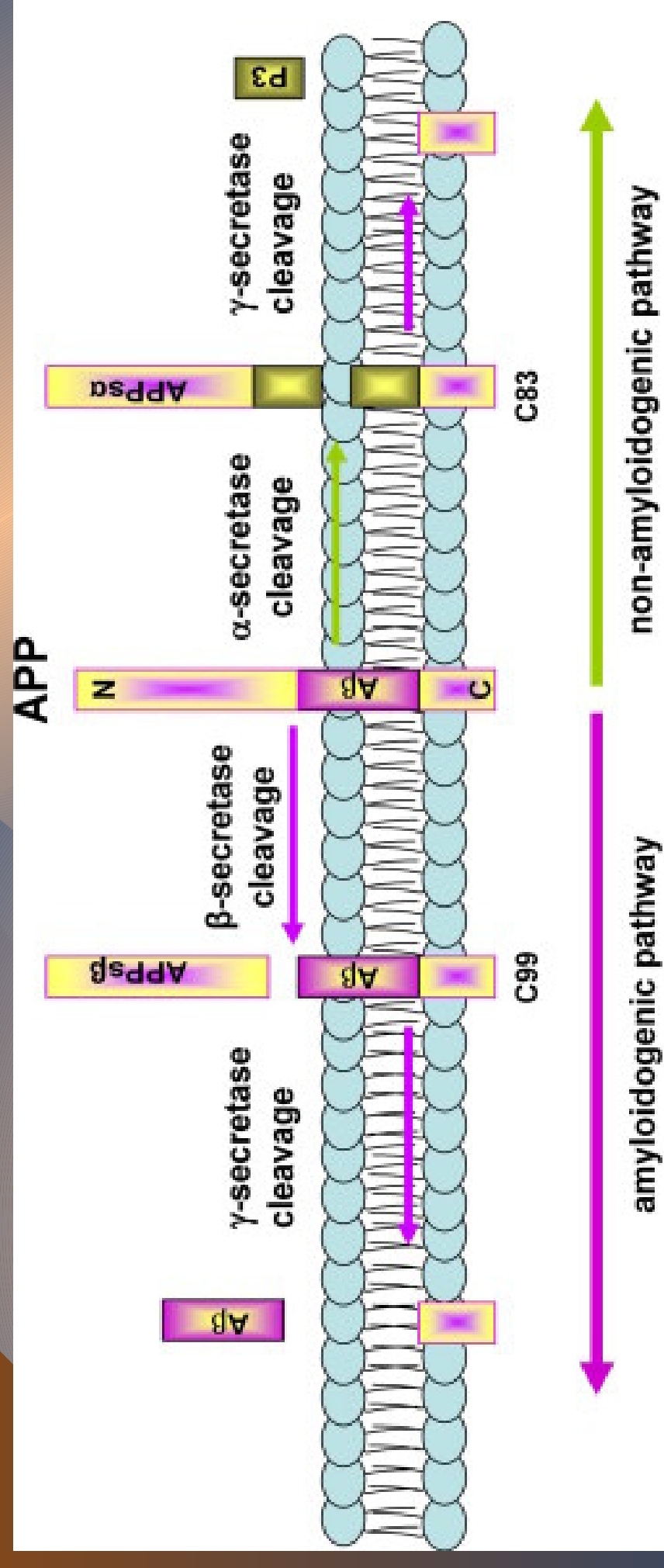
by Magdalena Korsak



Beta amyloid and Alzheimer's disease



A β is the main component of amyloid plaques - deposits found in the brains of patients with Alzheimer's disease

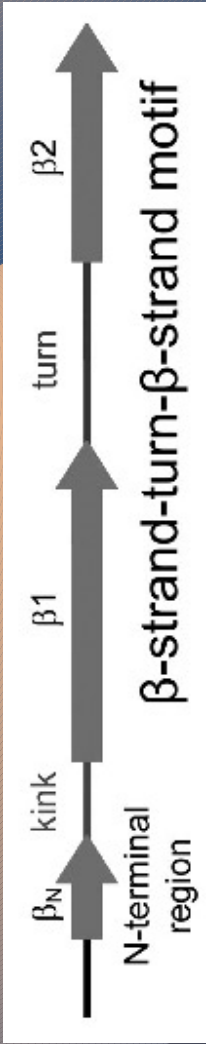


Abeta fibrils characterization

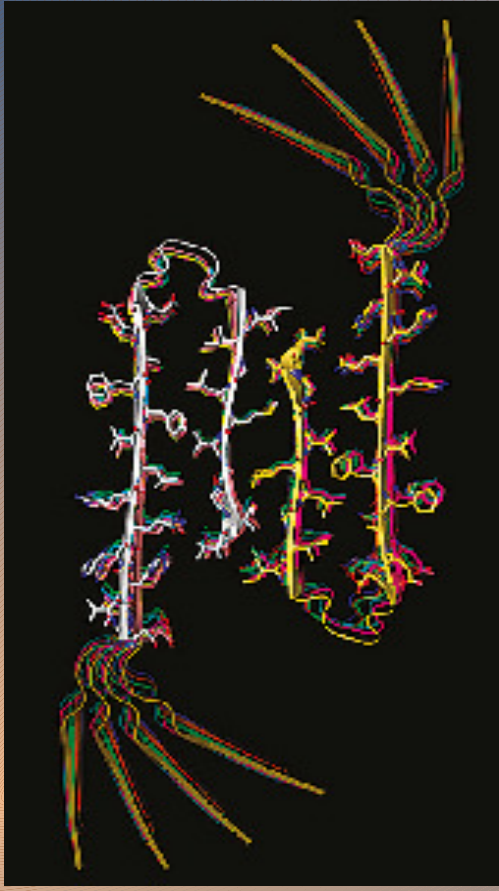
β -sheets rich structures , >100 nm in length , 8-12 nm in diameter

amyloid fibrils are polymorphic - structural variations include changes in symmetry and in the nature of β -sheet structures

Abeta40 fibrils

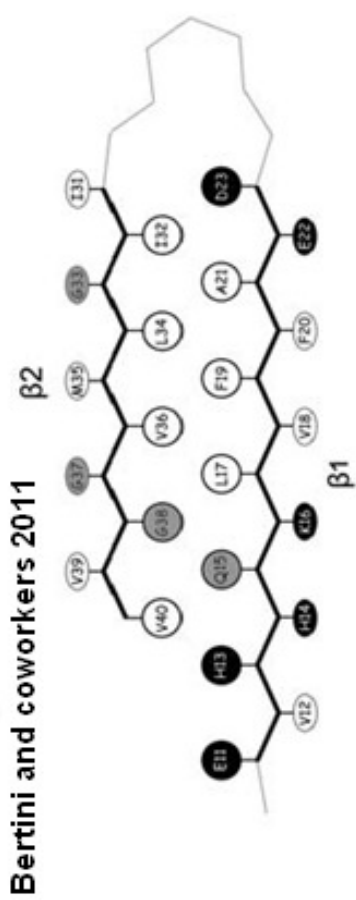


N-terminal part of A β 40 peptides can adopt distinct conformations and thereby also contribute to the structural diversity



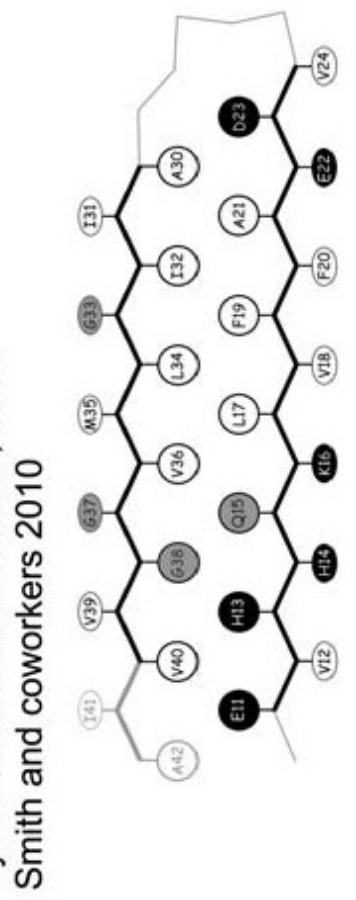
I. Bertini, L. Gonnelli, C. Luchinat, J. Mao, A. Nesi, JACS, 2011

Bertini and coworkers 2011



Tycko and coworkers 2006, 2008

Smith and coworkers 2010



Goals of the researchers

UNDERSTANDING OF THE MECHANISM OF BETA

AMYLOID GENERATION

IDENTIFICATION OF TOXIC ABETA SPECIES

ELUCIDATION OF STRUCTURAL AND BIOLOGICAL

PROPERTIES OF THE DIFFERENT AGGREGATION

STATES



Purification of A β M40 and A β M42 peptides

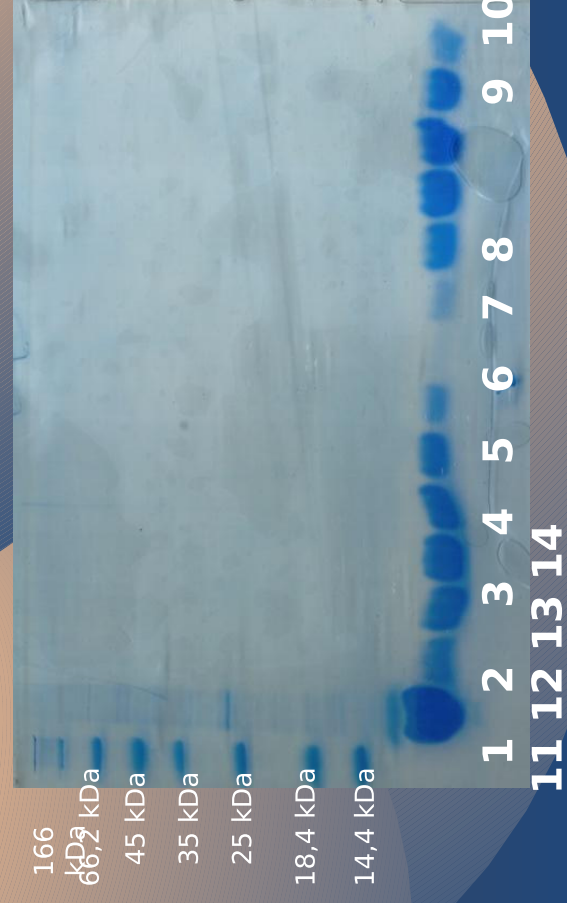
2 PURIFICATION STEPS:



Anion exchange chromatography



Gel filtration



1 - marker

2 - load sample

3-14 - purified A β protein

Fibrils mixtures A β M40 / A β M42

- double labelled A β M40 and unlabelled A β M42
- molar ratio 7:3 (A β M40 : A β M42)
- incubated at 37 °C under shaking (950 rpm) for 5

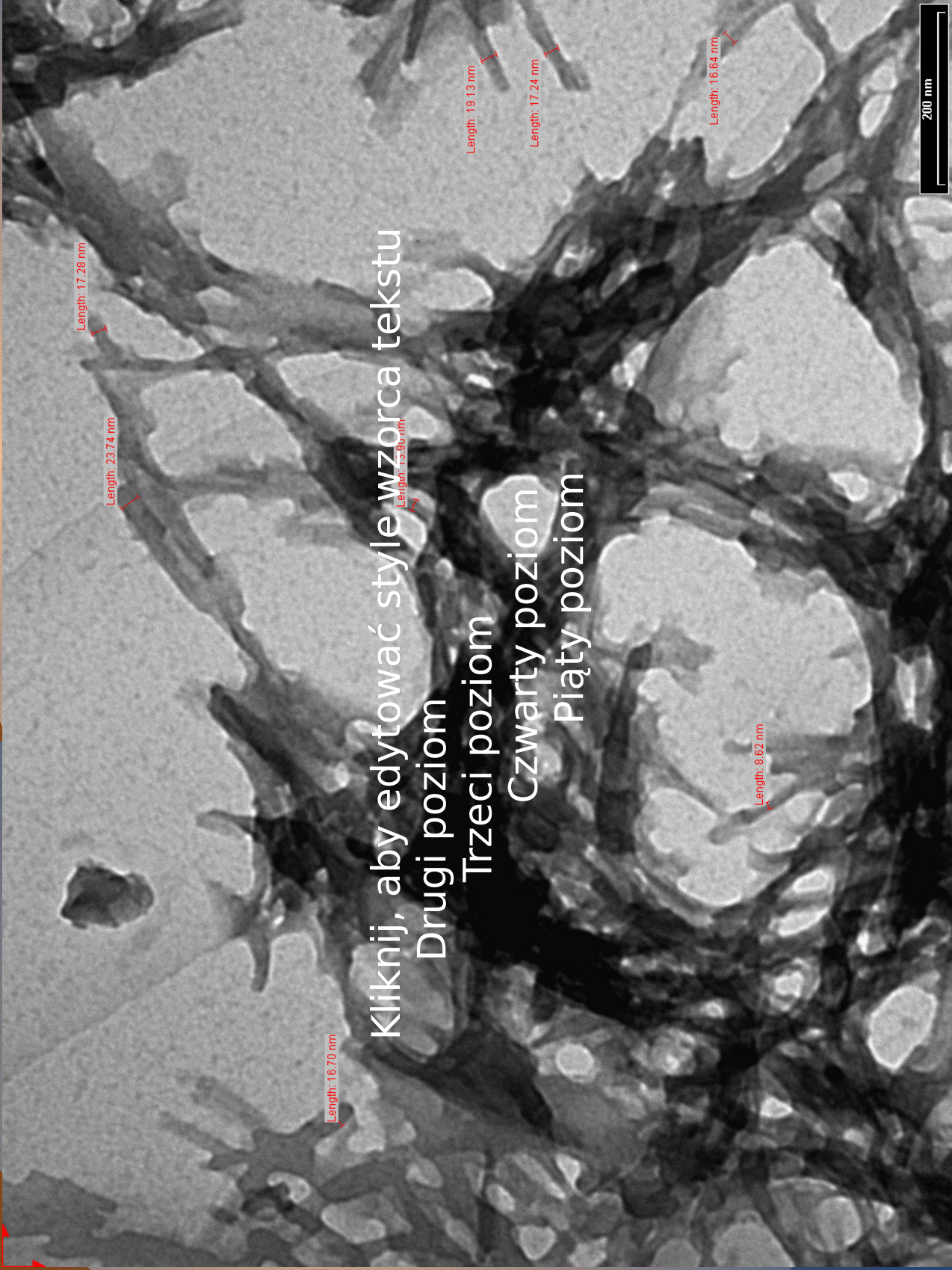
weeks

- collected by the overnight ultracentrifugation at

60000 rpm

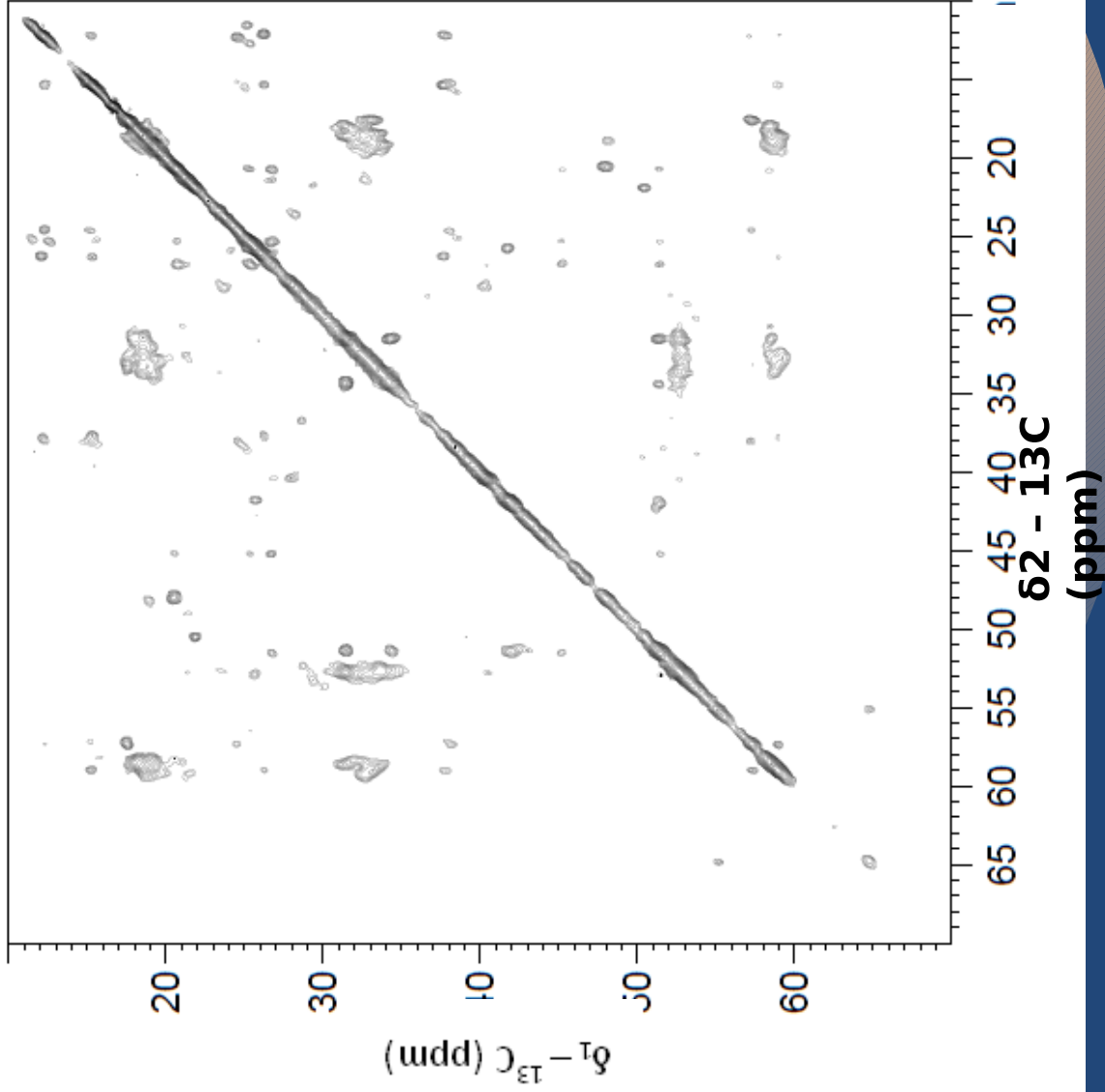
Fibrils mixtures AβM40 /AβM42

Transmission electron microscopy image



Mature fibrils SS NMR - SHANGHAI

Fibrils mixtures AβM40 /AβM42



^{13}C - ^{13}C correlation

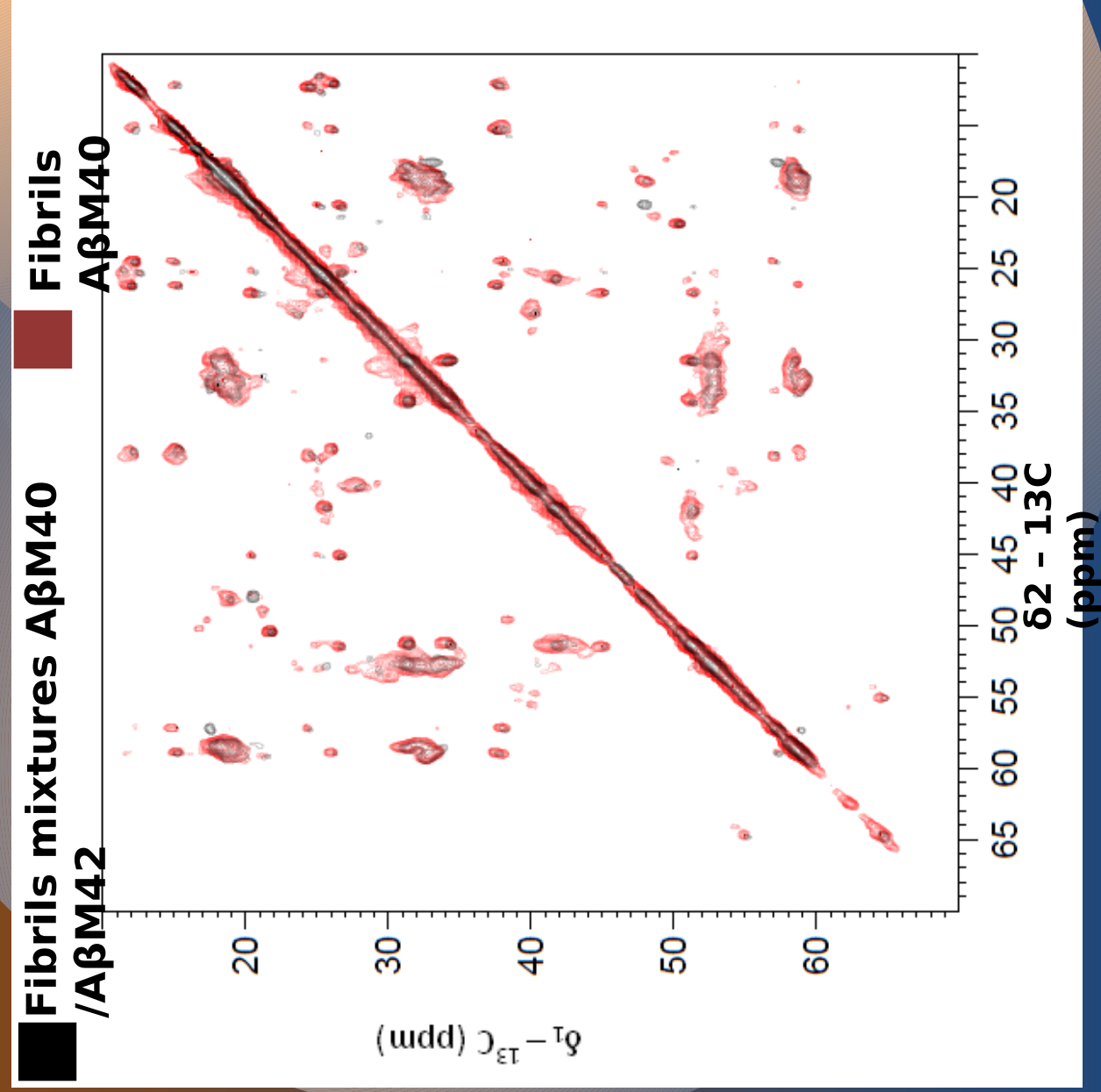
Magnetic field:
700 MHz (16.4 T)

Dimension of rotor:
3.2 mm (~14 mg of
fibrils)

14 kHz spinning

100 kHz ^1H decoupling

Mature fibrils SS NMR SHANGHAI / DARR



^{13}C - ^{13}C correlation

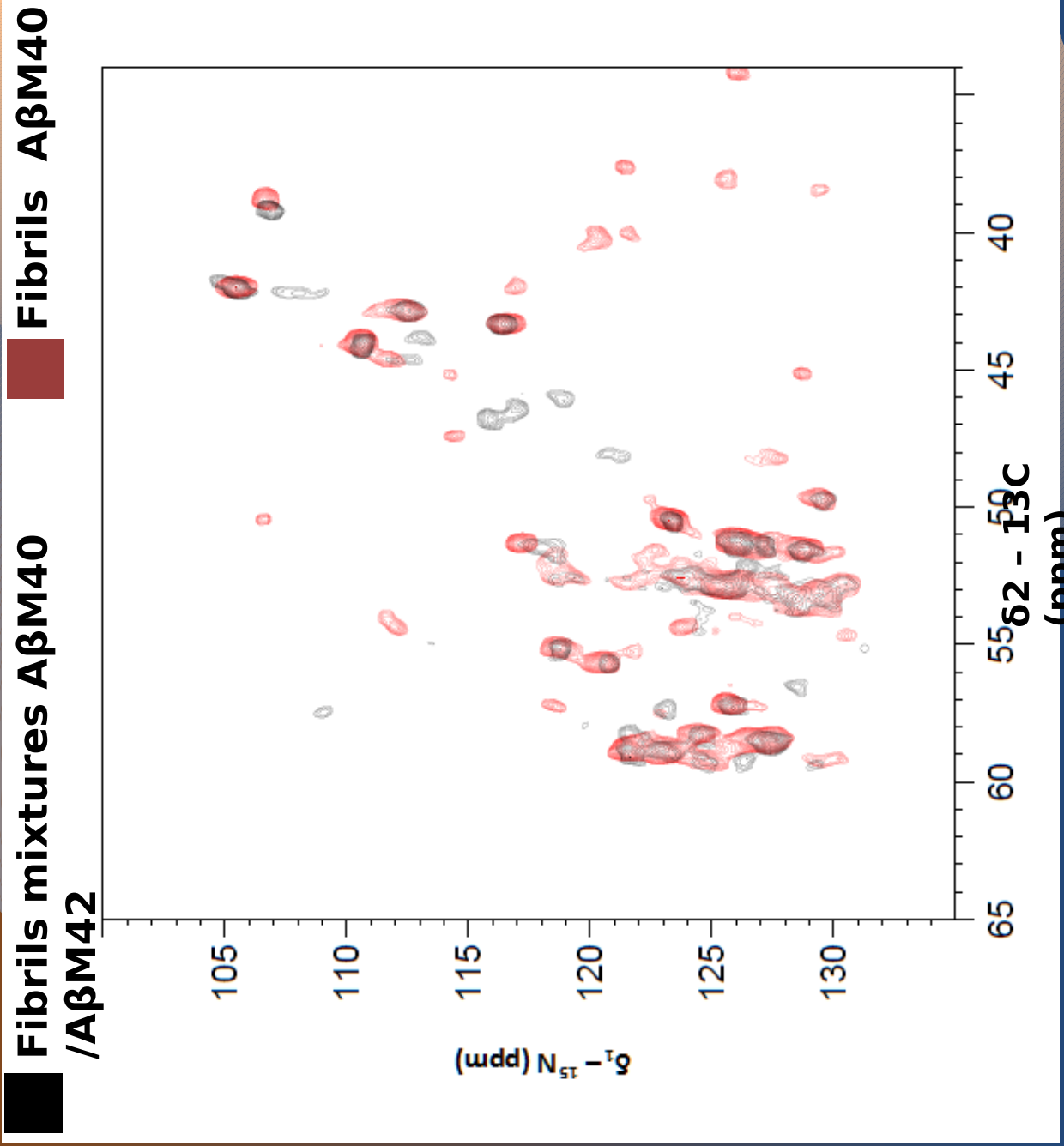
Magnetic field:
700 MHz (16.4 T)

Dimension of rotor:
3.2 mm (~14 mg of
fibrils)

14 kHz spinning

100 kHz ^1H decoupling

Mature fibrils SS NMR - NCA



15N - 13C correlation

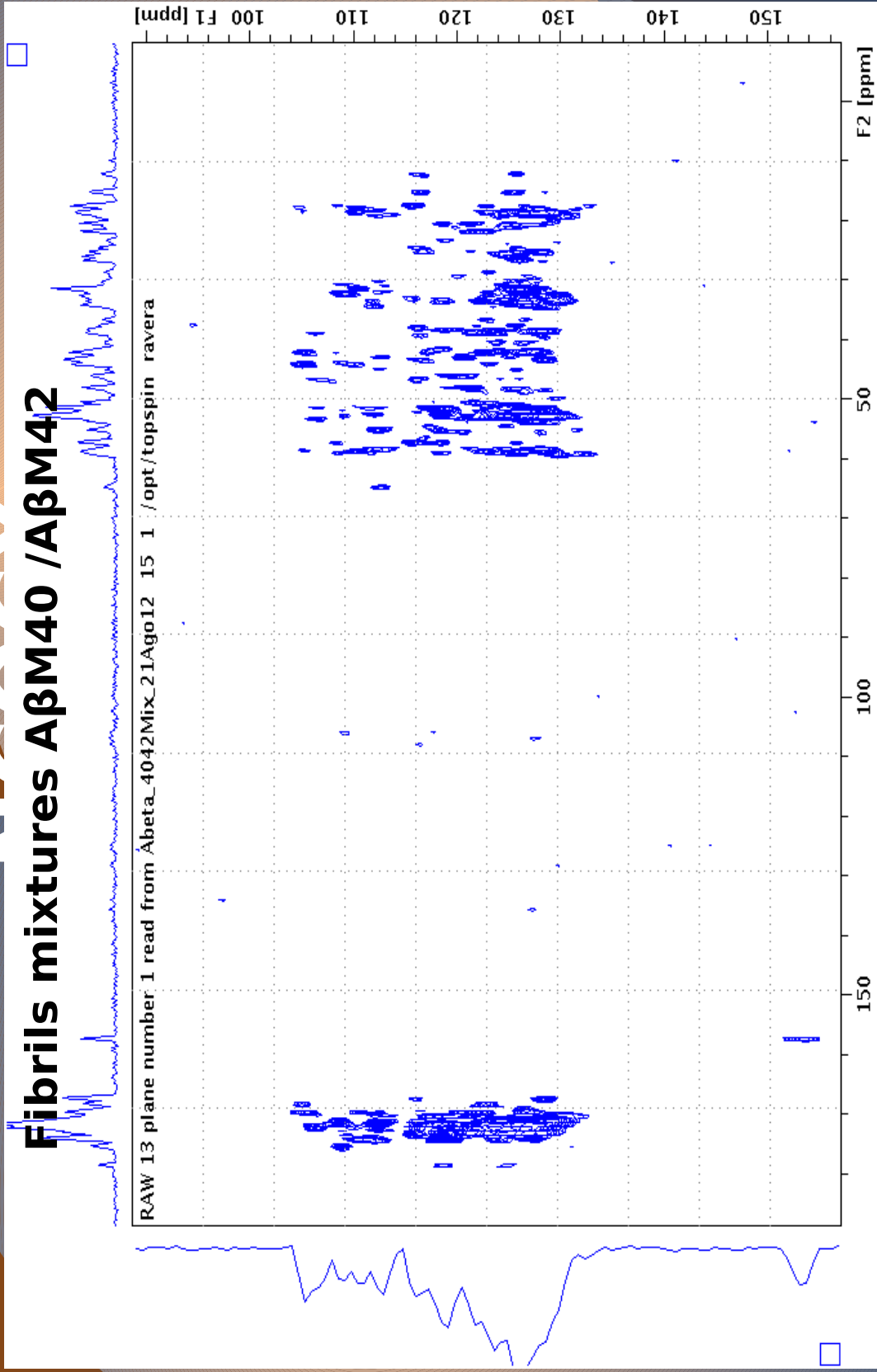
Magnetic field:
700 MHz (16.4 T)

Dimension of rotor:
3.2 mm (~14 mg of
fibrils)

14 kHz spinning

100 kHz 1H decoupling

Mature fibrils SS NMR -



15N - 13C correlation, 14 kHz spinning, 100 ms DARR mixing

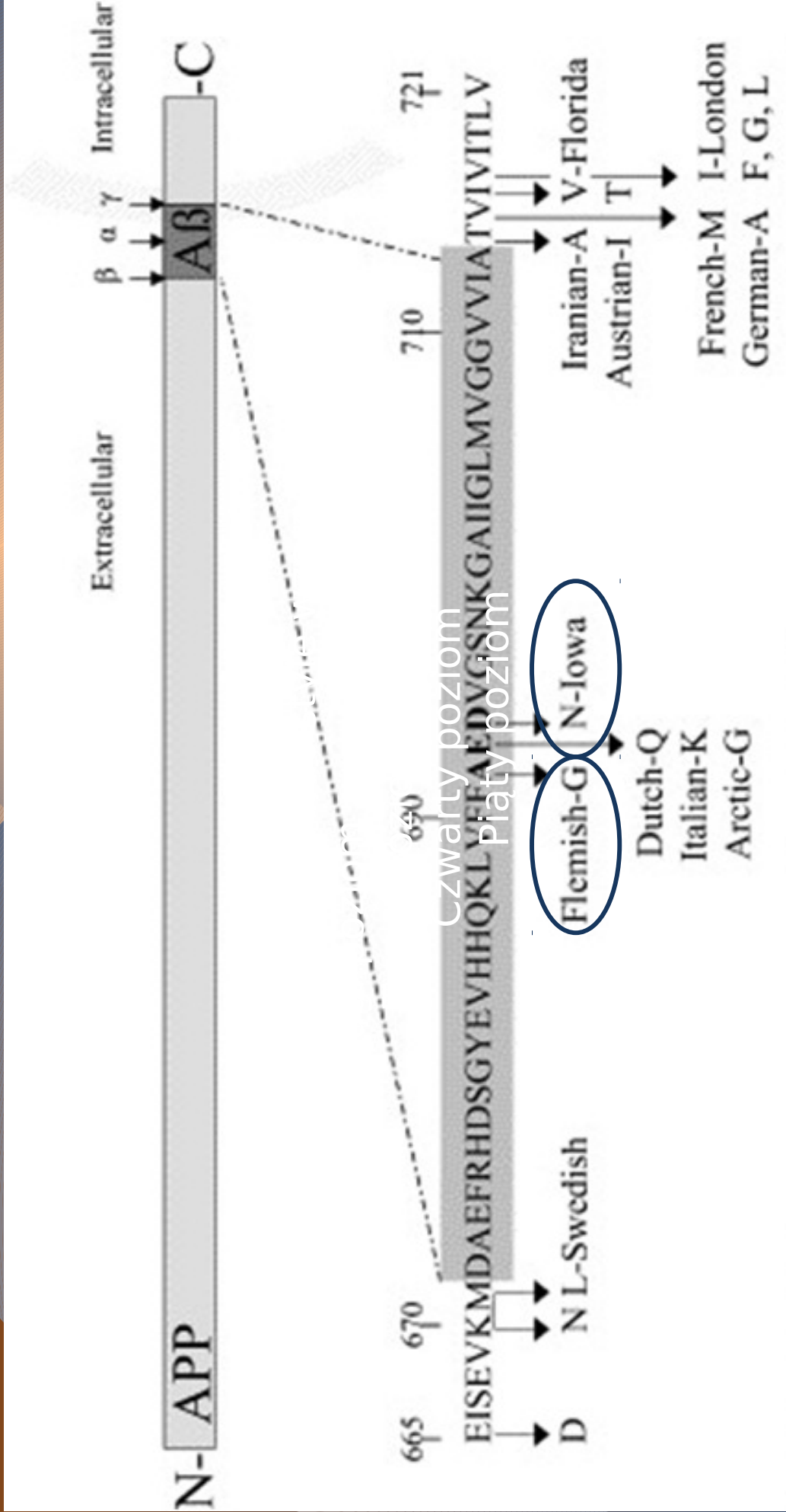
Magnetic field: 850 MHz (20 T), Dimension of rotor : 3.2 mm (~ 14 mm of fibrils)

ONGOING WORKS

Generation and characterization of different mutants of A β

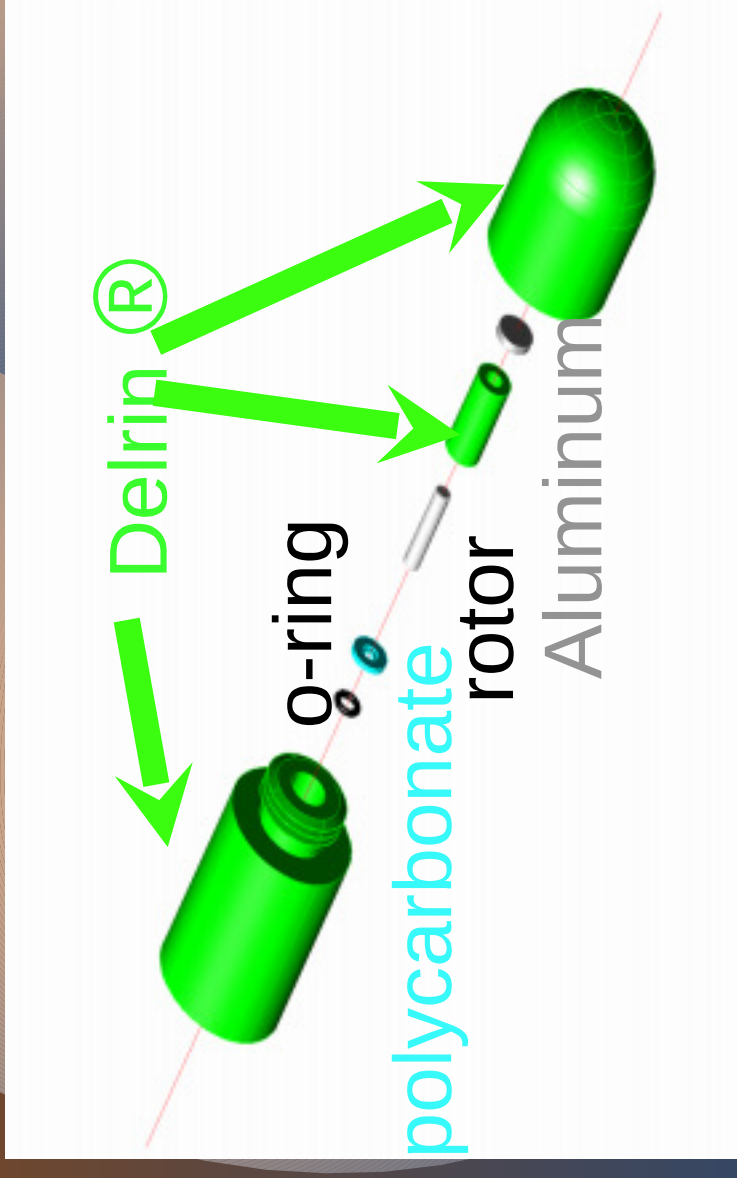
Isolation and characterization of A β oligomers and protofibrils by sedimented solute NMR

Mutations in familial Alzheimer's disease



Irie K, Murakami K, Masuda Y, Morimoto A, Ohgashi H, Ohashi R, Takegoshi K, Nagao M, Shimizu T, Shirasawa T, *Journal of bioscience and bioengineering*

Isolation and characterization of A β M40 oligomers and protofibrils for SS NMR and solution NMR studies by sedimented solute NMR



UC-induced sedimentation

1. Large volume
2. Very simple setup of experiment
3. Allows for fractional separation

MAS-induced sediment

I.Bertini, F.Engelke, L.Gonnelli, B.Knott, C.Luchinat,
D.Osen, E.Ravera, J. Biomol. NMR, in press

Future goals

Biophysical and structural characterization of mutants

- A22G
- D24N

Expression and purification of various A β peptides

- A β M40
- A β M42

Generation of fibrils for SS NMR studies

Acknowledgments



Prof. Ivano Bertini



Prof. Claudio Luchinat

Gianluca Gallo

Enrico Ravera



folded



An abstract graphic featuring a dark blue background with a vertical orange-to-blue gradient. Three large, overlapping circles are positioned on the left side. The top circle is a light blue-grey, the middle one is a darker blue, and the bottom one is a light orange. The text 'Thank you for your kind attention' is written in white, bold, sans-serif font, centered within the circles.

**Thank you for your
kind attention**