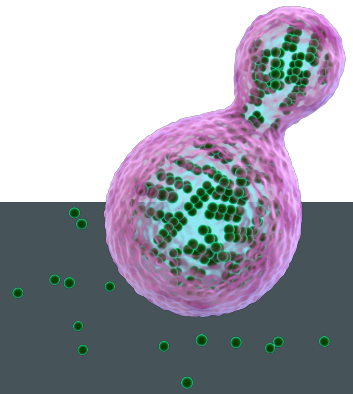
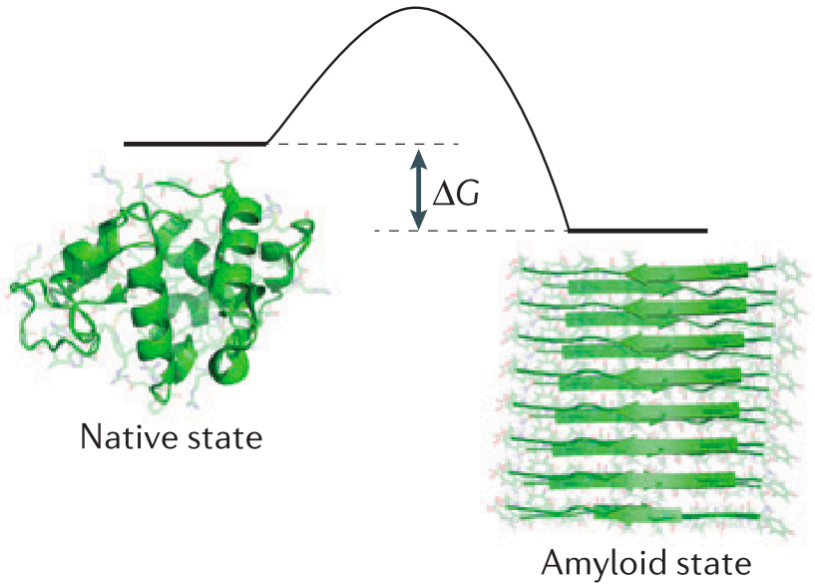
Novel Yeast-Based Platform for Studying Inborn Error of Metabolism Disorders

Shon Levkovich

Supervisors: Prof. Ehud Gazit and Dr. Dana Laor
Department of Molecular Microbiology and Biotechnology
Tel-Aviv University



The amyloid state as an energetic minima and its association with protein misfolding diseases

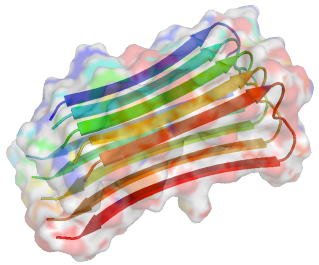


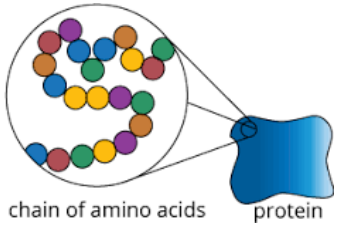
Protein Misfolding → Amyloid aggregation → Apoptosis

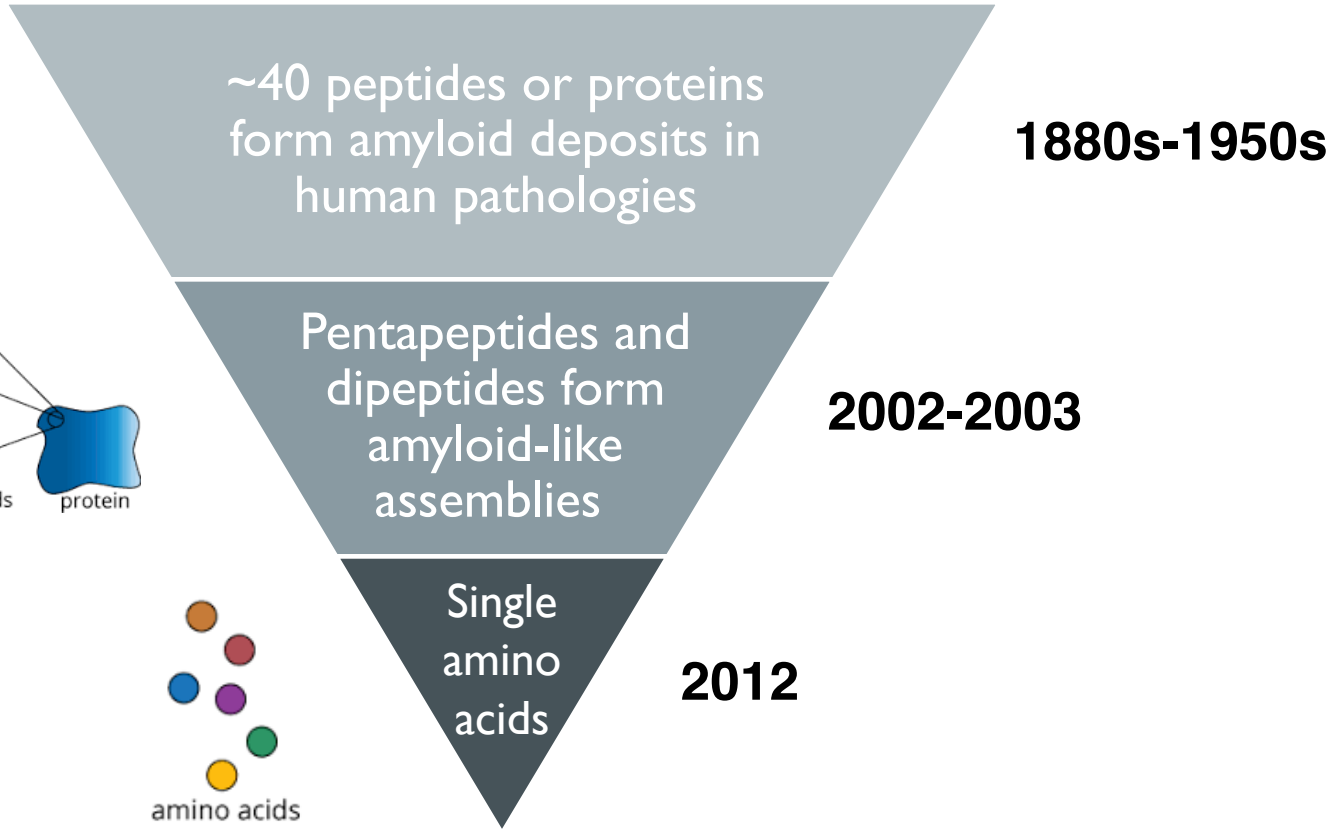
Disease	Aggregating protein or peptide
Neurodegenerative diseases	
Alzheimer's disease	Amyloid-β peptide
Spongiform encephalopathies	Prion protein or its fragments
Parkinson's disease	α-synuclein
Amyotrophic lateral sclerosis	Superoxide dismutase 1
Huntington's disease	Huntingtin fragments
Familial amyloidotic polyneuropathy	Transthyretin mutants
Non-neuropathic systemic amyloidosis	
Amyloid light chain (AL) amyloidosis	Immunoglobulin (Ig) light chains or its fragments
Amyloid A (AA) amyloidosis	Serum amyloid A1 protein fragments
Senile systemic amyloidosis	Wild-type transthyretin
Haemodialysis-related amyloidosis	β_2-microglobulin
Lysozyme amyloidosis	Lysozyme mutants
Non-neuropathic localized amyloidosis	
Apolipoprotein A1 (Apo A-1) amyloidosis	Apo A-1 fragments
Type II diabetes	Amylin

Knowles et al., *Nat. Rev. Mol. Cell Biol. (2014)*

Reductionist studies on amyloid fibrils



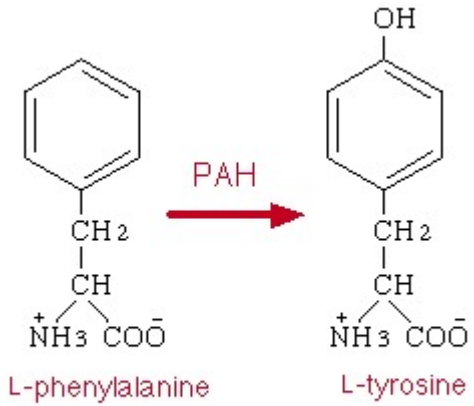


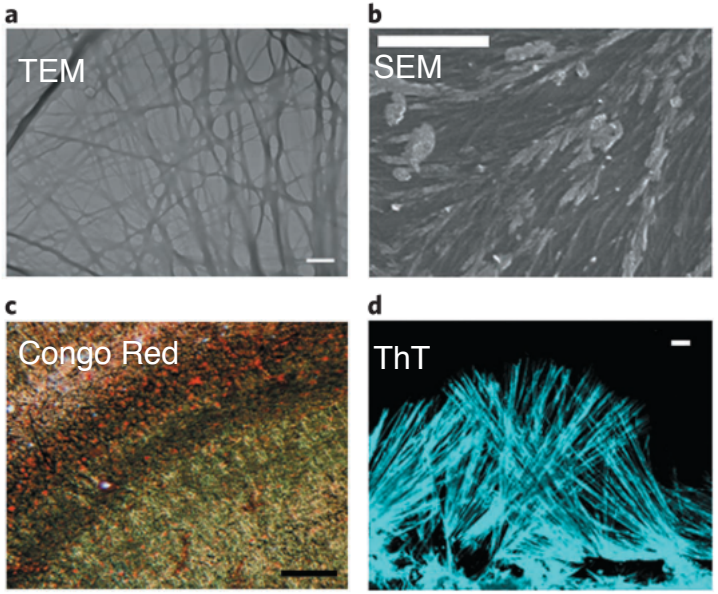


Phenylalanine assembly into toxic fibrils suggests amyloid etiology in phenylketonuria

Lihi Adler-Abramovich[1], Lilach Vaks[1], Ohad Carny[1], Dorit Trudler[2,3], Andrea Magno[4], Amedeo Caflisch[4], Dan Frenkel[2,3] & Ehud Gazit[1]*

Phenylketonuria (PKU) is characterized by phenylalanine accumulation and progressive mental retardation caused by an unknown mechanism. We demonstrate that at pathological concentrations, phenylalanine self-assembles into fibrils with amyloid-like morphology and well-ordered electron diffraction. These assemblies are specifically recognized by antibodies, show cytotoxicity that can be neutralized by the antibodies and are present in the hippocampus of model mice and in parietal cortex brain tissue from individuals with PKU. This is, to our knowledge, the first demonstration that a single amino acid can form amyloid-like deposits, suggesting a new amyloidosis-like etiology for PKU.





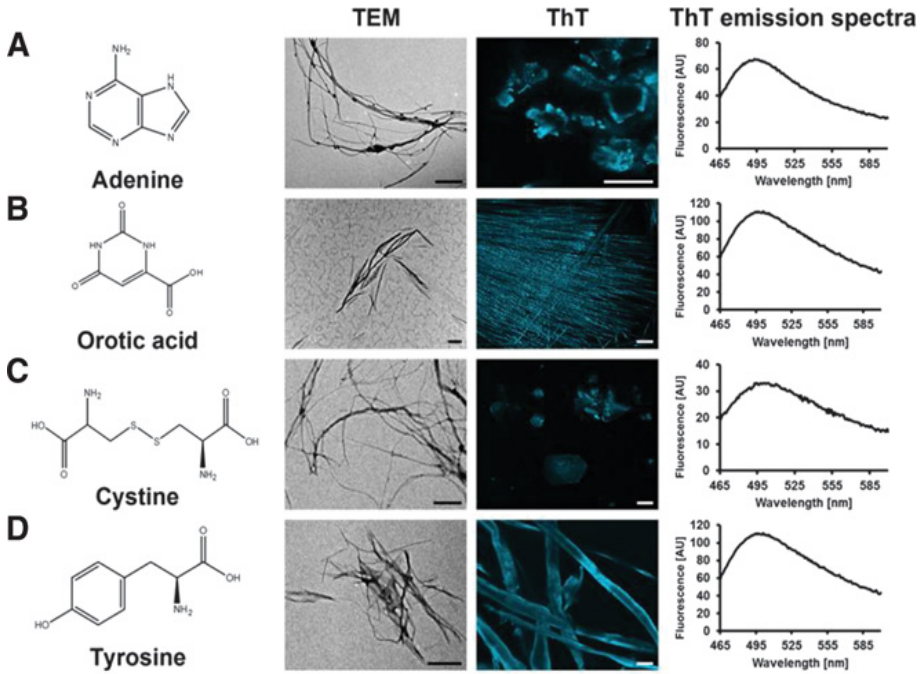
Adler-Abramovich *et al. Nature Chem. Biol.* (2012)

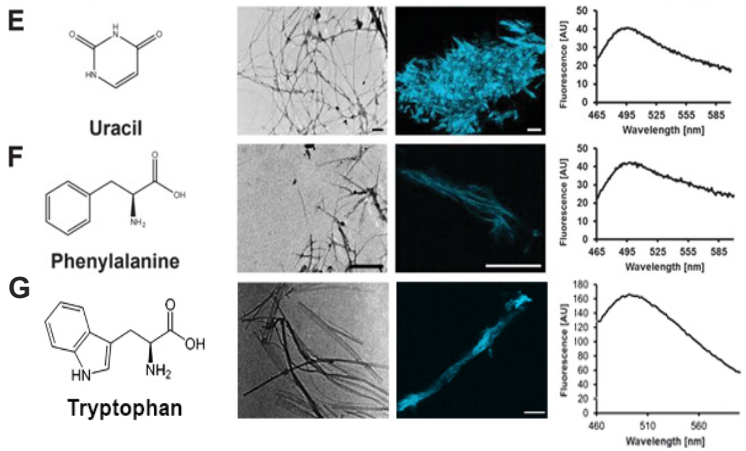
RESEARCH ARTICLE

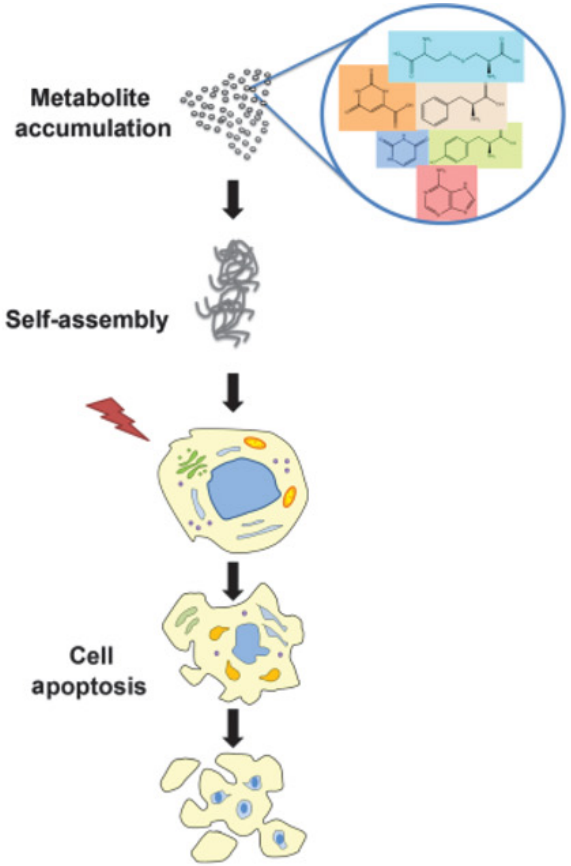
BIOLOGICAL CHEMISTRY

Extension of the generic amyloid hypothesis to nonproteinaceous metabolite assemblies

Shira Shaham-Niv,[1] Lihi Adler-Abramovich,[1,2] Lee Schnaider,[1] Ehud Gazit[1,3]*

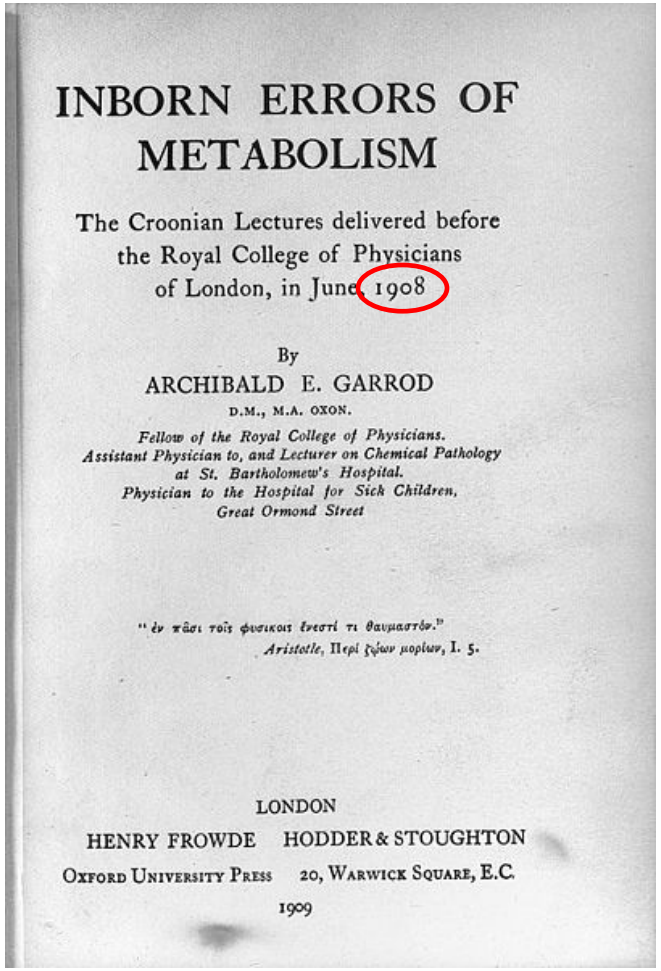






→ A new paradigm for the etiology of inborn error of metabolism disorders

Shaham-Niv *et al. Science Advances.* (2015)



INBORN ERRORS OF METABOLISM

The Croonian Lectures delivered before the Royal College of Physicians of London, in June, 1908

By

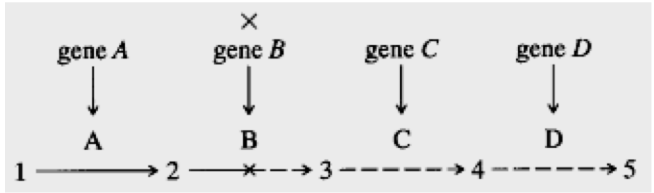
ARCHIBALD E. GARROD

D.M., M.A. OXON.

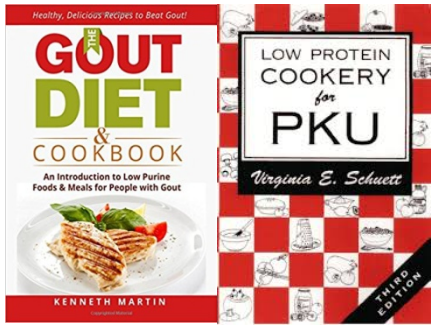
Fellow of the Royal College of Physicians.
Assistant Physician to, and Lecturer on Chemical Pathology at St. Bartholomew's Hospital.
Physician to the Hospital for Sick Children, Great Ormond Street

"ἐν πᾶσι τοῖς φυσικοῖς ἔνεστί τι θαυμαστόν."
Aristotle, Περὶ ζῴων μορίων, I. 5.

LONDON
HENRY FROWDE HODDER & STOUGHTON
OXFORD UNIVERSITY PRESS 20, WARWICK SQUARE, E.C.
1909



Metabolite	Disorder
Adenine	Adenine phosphoribosyltransferase deficiency
Arginine	Argininemia
Cystine	Cystinuria, Cystinosis
Glycine	Non-ketotic hyperglycinemia, D-Glyceric academia, Iminoglycinuria
Homogentisic acid	Alkaptonuria
Isoleucine	Maple syrup urine disease
Leucine	Maple syrup urine disease
Lysine	Saccharopinuria
Methionine	Hypermethioninemia
N-acetyl aspartate	Canavan disease
Orotic acid	Ornithine transcarbamylase deficiency
Phenylalanine	Phenylketonuria
Proline	Hyperprolinemia, Iminoglycinuria
Tryptophan	Hypertryptophanemia , Hartnup disease
Tyrosine	Tyrosinemia
Uracil	Ornithine transcarbamylase deficiency, Dihydropyrimidine dehydrogenase deficiency
Uric acid	Gout, Lesch-Nyhan syndrome
Valine	Isobutyryl-CoA dehydrogenase deficiency, Maple syrup urine disease



The awesome power of yeast: modeling protein misfolding disorders

Aggregation of huntingtin in yeast varies with the length of the polyglutamine expansion and the expression of chaperone proteins

Sylvia Krobitsch and Susan Lindquist*

***PNAS**, 2000*

Functional Links Between Aβ Toxicity, Endocytic Trafficking, and Alzheimer's Disease Risk Factors in Yeast

Sebastian Treusch,[1,2] Shusei Hamamichi,[1,3] Jessica L. Goodman,[1] Kent E. S. Matlack,[1,2] Chee Yeun Chung,[1] Valeriya Baru,[1,2] Joshua M. Shulman,[4,5] Antonio Parrado,[6] Brooke J. Bevis,[1] Julie S. Valastyan,[1,2] Haesun Han,[1] Malin Lindhagen-Persson,[7] Eric M. Reiman,[8,9] Denis A. Evans,[10] David A. Bennett,[11] Anders Olofsson,[7] Philip L. DeJager,[4,5] Rudolph E. Tanzi,[6] Kim A. Caldwell,[3] Guy A. Caldwell,[3] Susan Lindquist[1,2*]

***Science**, 2011*

Yeast Reveal a "Druggable" Rsp5/Nedd4 Network that Ameliorates α-Synuclein Toxicity in Neurons

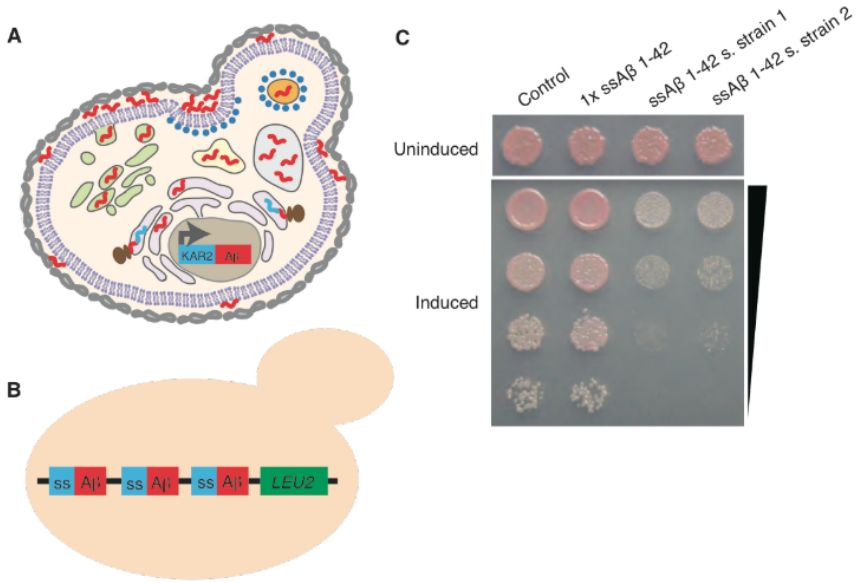
Daniel F. Tardiff,[1] Nathan T. Jui,[2] Vikram Khurana,[1,3] Mitali A. Tambe,[4] Michelle L. Thompson,[5*] Chee Yeun Chung,[1] Hari B. Kamadurai,[6] Hyoung Tae Kim,[7] Alex K. Lancaster,[1†] Kim A. Caldwell,[5] Guy A. Caldwell,[5] Jean-Christophe Rochet,[4] Stephen L. Buchwald,[2] Susan Lindquist[1,8‡]

***Science**, 2013*

Translocon Declogger Ste24 Protects against IAPP Oligomer-Induced Proteotoxicity

Can Kayatekin,[1,11,*] Audra Amasino,[2] Giorgio Gaglia,[1] Jason Flannick,[3,4] Julia M. Bonner,[1] Saranna Fanning,[1] Priyanka Narayan,[1] M. Inmaculada Barrasa,[1] David Pincus,[1] Dirk Landgraf,[1] Justin Nelson,[5] William R. Hesse,[2] Michael Costanzo,[6] AMP T2D-GENES Consortium, Chad L. Myers,[7] Charles Boone,[6] Jose C. Florez,[3,4,8,9] and Susan Lindquist[1,2,10]

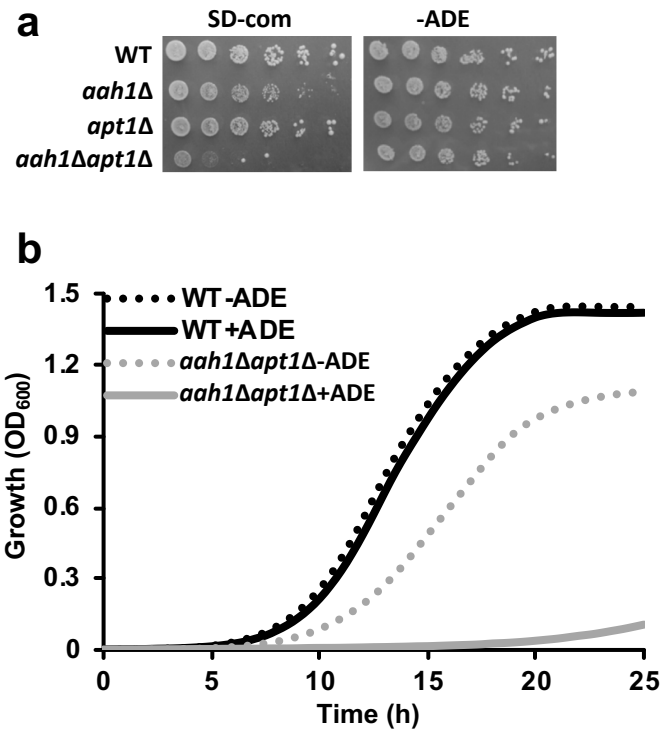
***Cell**, 2018*

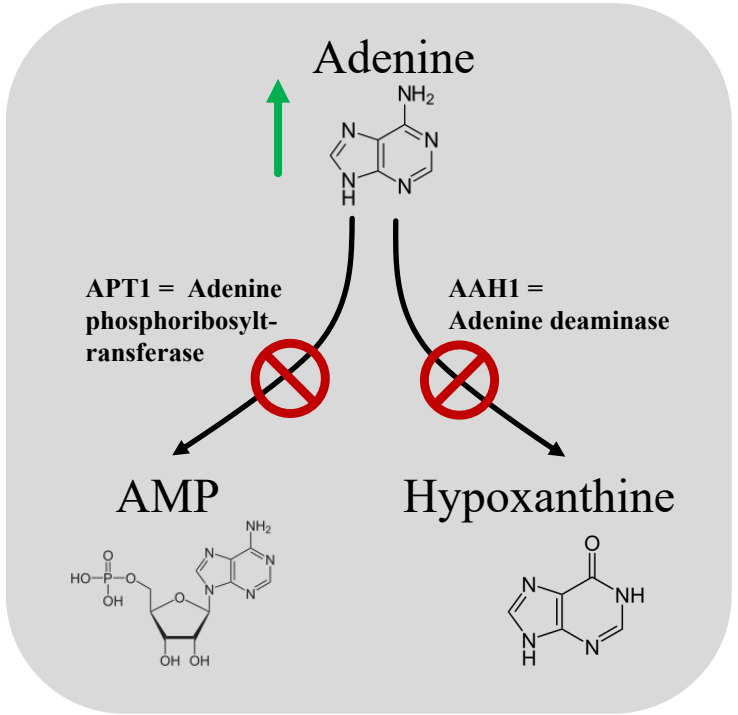


Modeling Inborn Errors of Metabolism in yeast

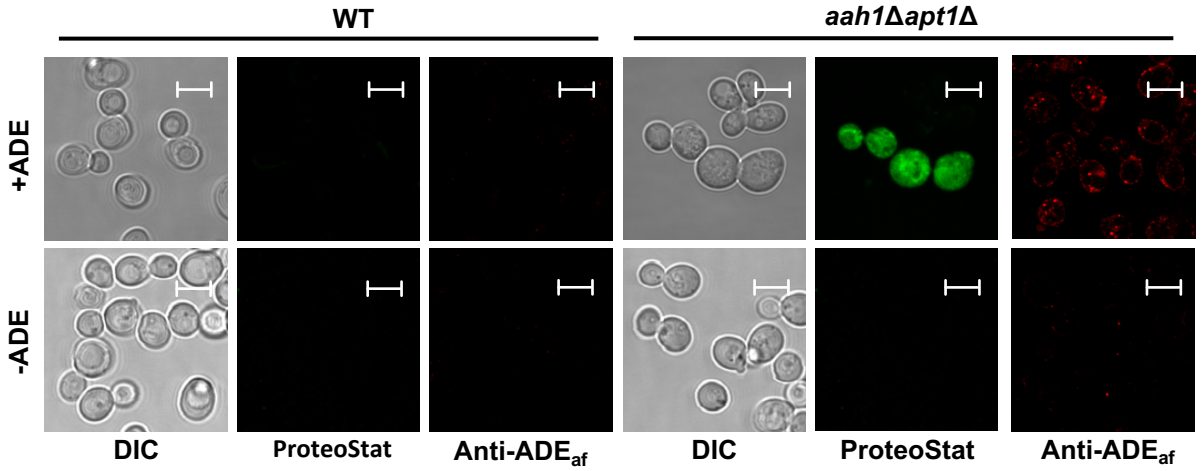
Fibril formation and therapeutic targeting of amyloid-like structures in a yeast model of adenine accumulation

Dana Laor[1], Dorin Sade[1], Shira Shaham-Niv[1], Dor Zaguri[1], Myra Gartner[1], Vasantha Basavalingappa[1], Avi Raveh[2], Edward Pichinuk[2], Hamutal Engel[2], Keita Iwasaki[3], Tatsuyuki Yamamoto[3,4], Hemanth Noothalapati[4] & Ehud Gazit[1,2,5]

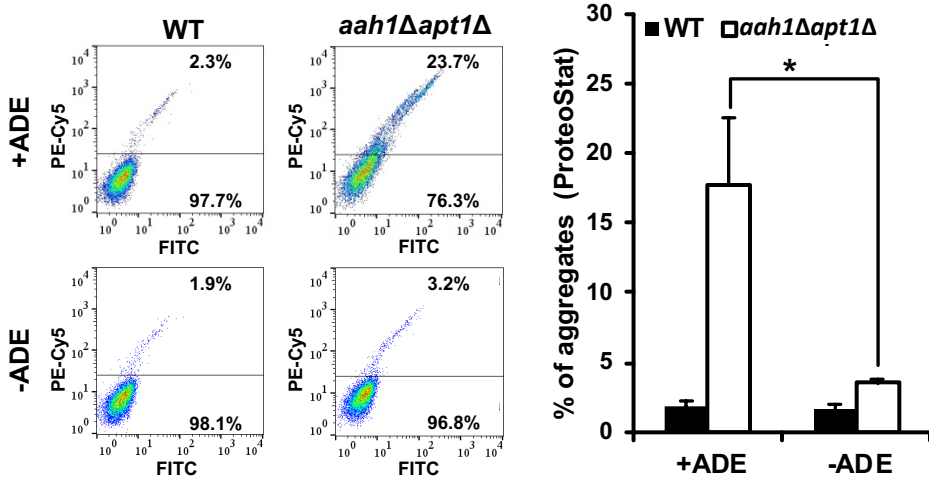


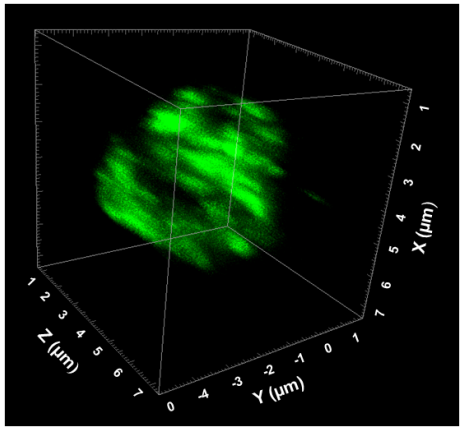


Laor et al., *Nat. Commun, 2019*



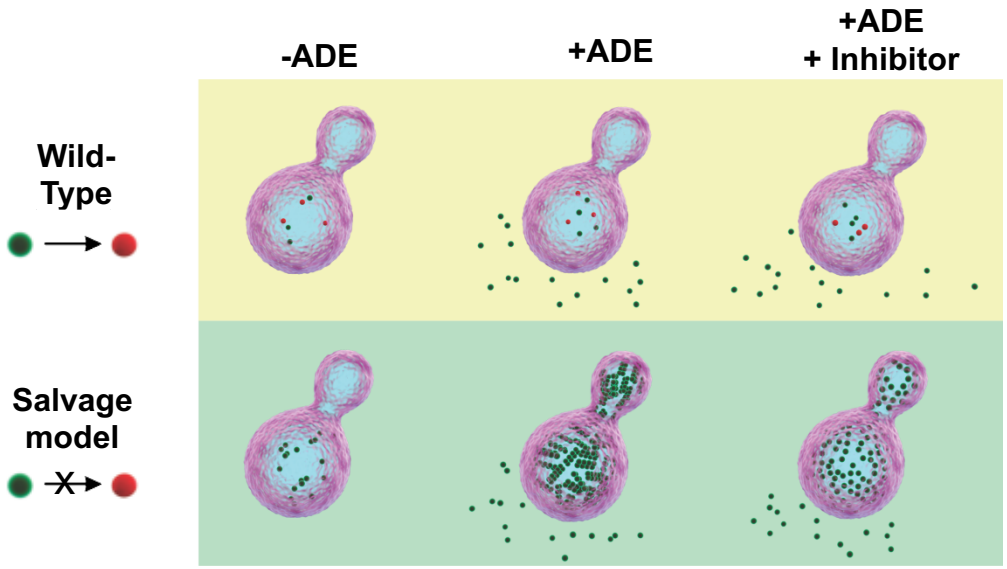
- *In vivo* formation of **adenine amyloid-like** assemblies using an amyloid-specific fluorescent dye and antibodies against adenine fibrils structures

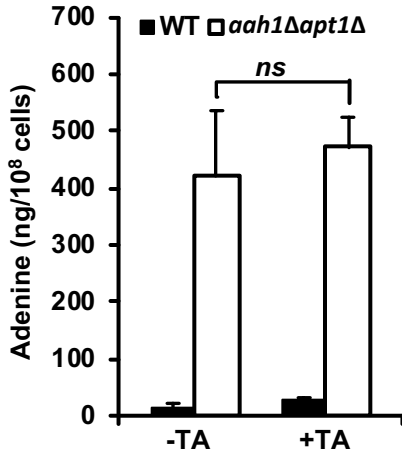




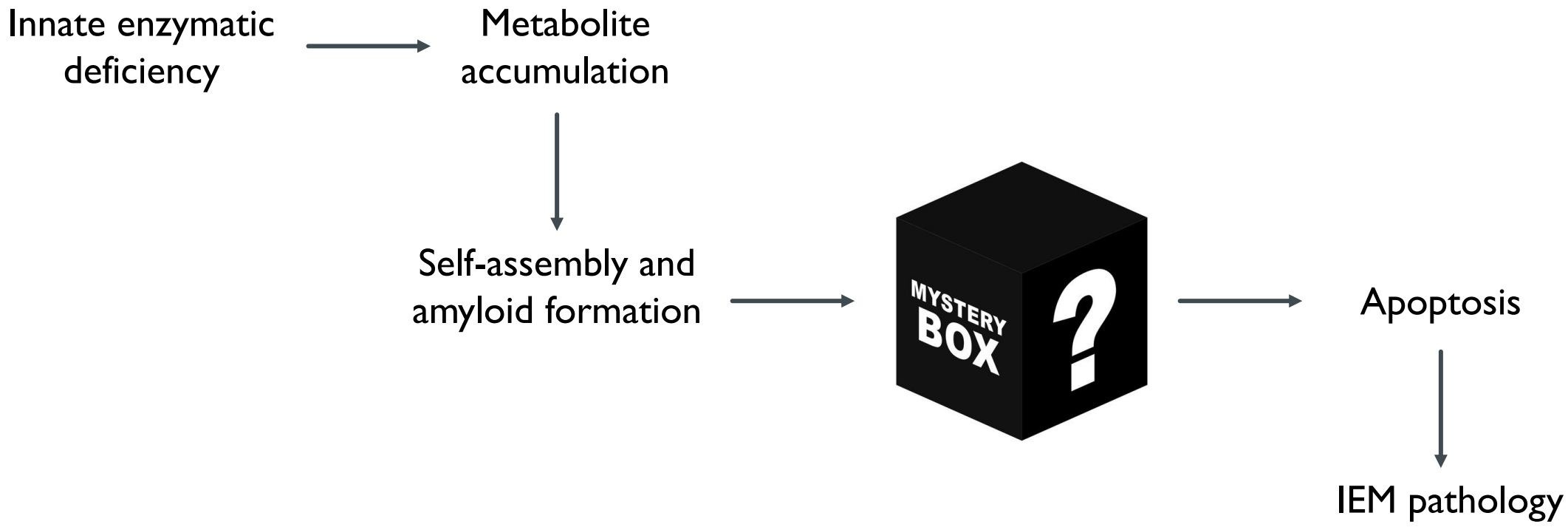
Z-stack 3D reconstruction

Polyphenol amyloid inhibitor rescues phenotype without changing intracellular adenine concentration





The mysterious pathophysiology of IEMs: how do metabolite amyloids lead to apoptosis?



Acknowledgments

Prof. Ehud Gazit
Dr. Dana Laor
The Gazit Lab Members

Prof. Shay Ben-Aroya
Faculty of Life Sciences
Bar-Ilan University

Prof. Martin Kupiec
Dr. Adi Cohen
Batya Lifshitz
Lihi Gershon

Dr. Eddy Pichinuk
The BCDD Staff