Workshop 3 – Diagnosis and management of lysosomal storage disorders

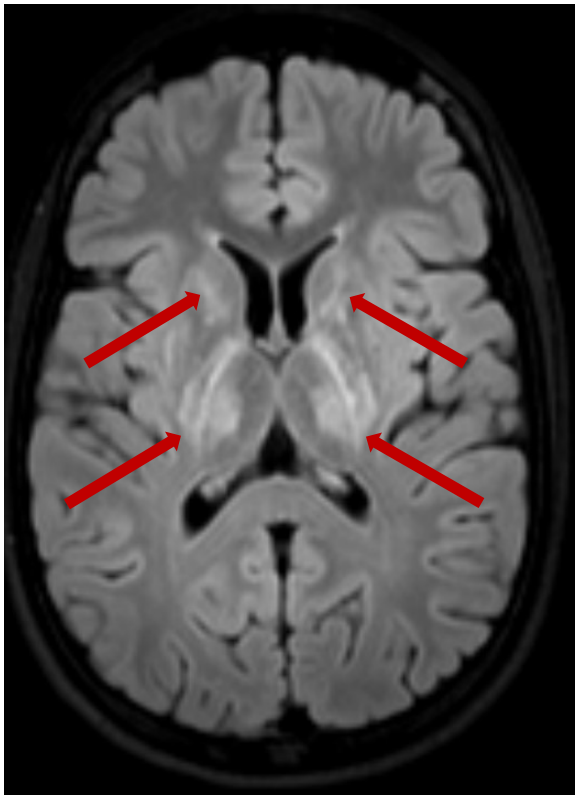
SSIEM Academy 2024, Amsterdam

The teenager with school problems

Patient

5 years	Gastroenteritis
	Serum ALT 146 U/L (0 – 40)
	Serum AST 113 U/L (0 – 40)
11 years	First symptoms: learning difficulties, dyscalculia
14 years	Increasing problems at school
	Child neurologist: no clear diagnosis, wait-and-watch
	(no testing performed)

Radiology



Brain MRI at age 14.5 y:

increased signal intensity in basal ganglia,

capsula interna and thalamus

What are your differential diagnoses?

(Start with treatable diseases!)

What tests would you consider?

Copper metabolism

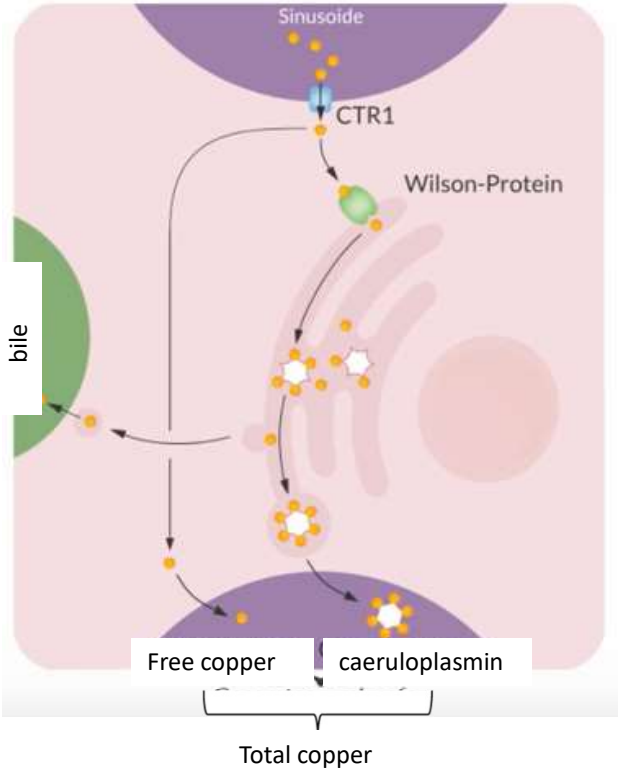
Copper in blood	6.0 µmol/L (12.6-24.3)
Caeruloplasmin	0.13 g/L (0.11-0.5)
Copper in 24 h urine	7.91 µmol/d (< 0.93)

→ What is the likely diagnosis?

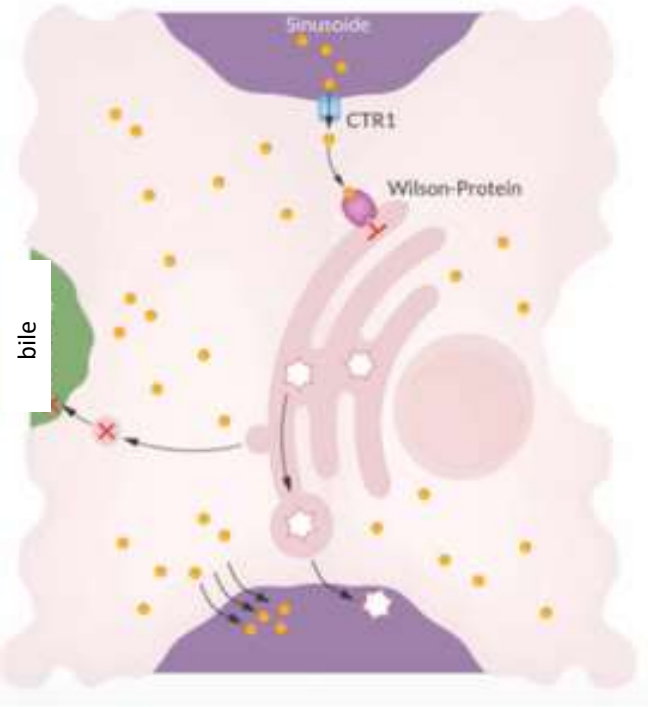
→ Diagnosis of Wilson disease confirmed biochemically

Copper metabolism

healthy cell

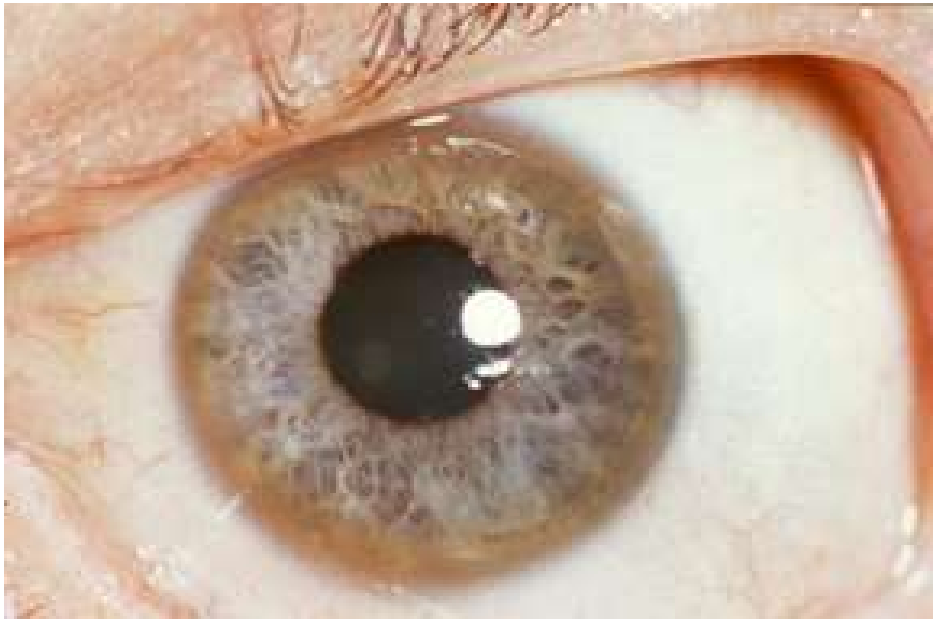


Cell with Wilsons‘s disease



Which other investigations would you perform?

Kayser-Fleischer ring



Genetic confirmation

Sanger sequencing of the *ATP7B* gene:

Compound heterozygosity for

c.3207C>A; p.His1069Gln and c.2128G>A; p.Gly710Ser

How would you treat this patient?

Treatment

→ Therapy started with trientine 4 HCl (1 x 600 mg/d p.o.)

After single dose of trientine

Copper in 24 h urine 22.92 µmol/d (< 0.93)

No regular monitoring of transaminases and parameters of copper metabolism performed

Course under therapy

Crisis: 8 months after diagnosis
sudden onset of movement disorder:
severe dystonia, hyperactivity, dysphagia, ataxia

Copper in blood	1.3 µmol/L (12.6-24.3)
Caeruloplasmin	0.11 g/L (0.11-0.5)
Copper in 24 h urine	11.18 µmol/d (< 0.93)

Cause of crisis after start of trientine?

Neurological deterioration after therapy initiation

Systematic Review: Wiggelinkhuizen et al, Aliment Pharm Ther, 2009

582 articles screened

13 articles selected (12 cohort studies, 1 randomised study)

Total patients:	234
Patients with neurological Wilson:	82
Patients with initial deterioration of neurological signs or symptoms after start of therapy :	7 (6 on D-penicillamin, 1 on zinc)

Initial neurological worsening is thought to reflect the **mobilization of hepatic copper** by D-penicillamine **into the circulation and its redistribution in the brain** (Brewer et al.; Arch Neurol1987;44: 490–3)

Further course after crisis

Therapy:	fast tapering of trientine within 2 weeks
	start with zinc (3 x 25 mg/d)
	inpatient rehabilitation for 6 months
Further course:	partial recovery over more than 3 years
	mild ataxia & dystonia
	(almost) normal speech

Discussion/ Take home messages

- Caeruloplasmin levels normal in 10-15% of patients

- Lifelong adherence to medical treatment is crucial!

- Therapy:
  - chelating agents (D-penicillamine/trientine)
    cave: common side effects, bioavailability of trientine is reduced by 50% when taken together with a meal!
  - zink: blockage of intestinal copper reabsorption

- Dietary restrictions differ between countries (Sturm et al. 2016)

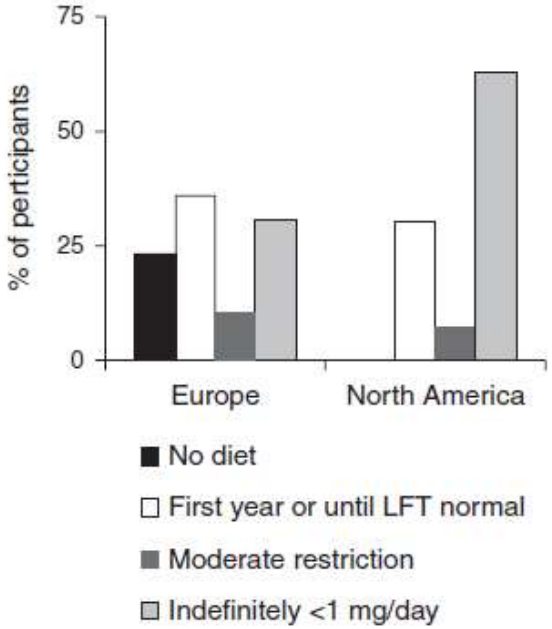


FIGURE 1. Restriction of dietary copper intake. Participants were asked which advice is given to their patients with WD on the restriction of dietary copper. Data are presented as percentage of participants per continent that selected the option as described in the legend. In total, 39 participants from Europe and 43 from Northern America answered this question. LFT = liver function tests; WD = Wilson disease.

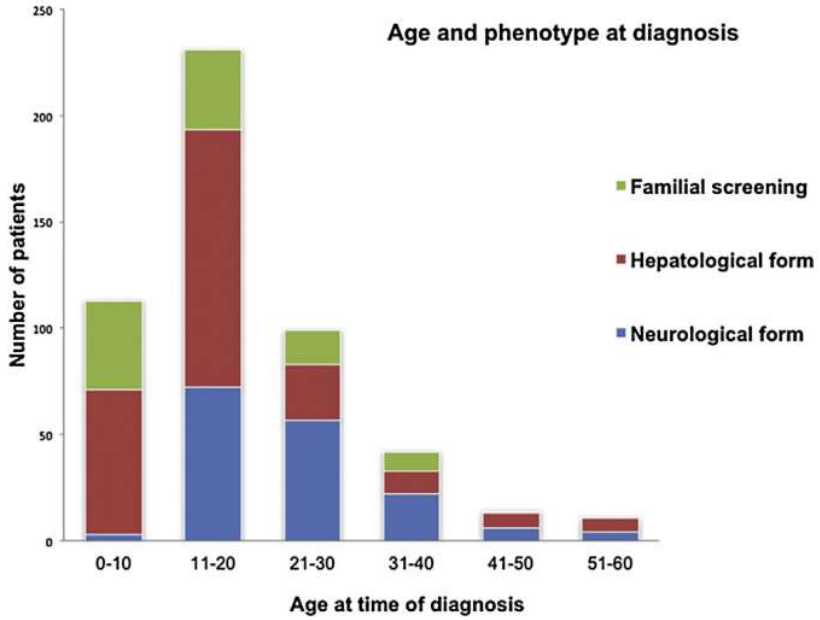
Sturm E et al. 2016: Controversies and Variation in Diagnosing and Treating Children With Wilson Disease: Results of an International Survey. J Pediatr Gastroenterol Nutr. 2016 Jul;63:82-7

Different presentations

Often neurological or psychiatric disease:

- Parkinson
- Ataxia
- Dystonia (choreoathetosis, dysphagia, hypersalivation)
- Stereotypic movements

- Mood disorders
- Behaviour and personality problems
- Psychosis and schizophrenia



Poujois A, Woimant F. Wilson's disease: A 2017 update. Clin Res Hepatol Gastroenterol. 2018;42:512-520.

Table 2 Biochemical and histological tests for Wilson disease

Test	Value in WD	Pitfalls/comments
Plasma caeruloplasmin	<200 mg/L (in 85%–90% of cases).	Elevated by hepatic or other inflammation. Low in other conditions, for example, malnutrition, aceruloplasminaemia and protein losing enteropathy, infants <1 year. WD heterozygotes may have plasma ceruloplasmin <200 mg/L.
Serum copper	Low, normal or high.	Poor diagnostic value in WD.
Free serum copper (or non-ceruloplasmin bound copper)	>7 (µM).	Estimated from serum copper and serum ceruloplasmin levels but is dependent on adequacy of the methods used measuring both and as such unreliable in diagnosing WD.
Urine copper (prepenicillamine)	>1.25 µmol/24 hours. >40 µg/24 hours in asymptomatic WD. >100 µg/L in symptomatic WD.	May be increased in acute hepatitis but usually much higher in WD.
Urine copper (postpenicillamine)*	>25 µmol/24 hours. >1600 µg/24 hours.	Indicated in symptomatic children if prepenicillamine test is normal or doubtful poor sensitivity in presymptomatic siblings.
Liver histology	Macro or micro vesiculars teatosis, portal or lobular inflammation, fibrosis, Mallory denk bodies, copper staining, canalicular cholestasis glycogenated hepatocyte nuclei.	Negative copper staining in liver biopsy does not exclude WD.
Liver copper	>250 µg/g dry weight (normal <55).	Higher values can be found in newborns and prolonged cholestasis due to other liver diseases (eg, sclerosing cholangitis). In patients with WD with cirrhosis, this value can be <250 µg/g, which can be confusing.
MRI (T1 and T2 weighted)	Detect atrophic changes and changes of the putamen. Giant panda's sign.	
Ophthalmic examination	Looking for KF rings and other eye abnormalities.	

*Penicillamine challenge is undertaken by giving two doses of penicillamine 500 mg at each time, at the beginning and 12 hours following commencement of urine collection.
KF, Kayser-Fleischer.

Wilson disease score

Typical clinical symptoms and signs	
KF rings	
Present	2
Absent	0
Neurologic symptoms**	
Severe	2
Mild	1
Absent	0
Serum ceruloplasmin	
Normal (>0.2 g/L)	0
0.1-0.2 g/L	1
<0.1 g/L	2
Coombs-negative hemolytic anemia	
Present	1
Absent	0

Other tests	
Liver copper (in the absence of cholestasis)	
>5x ULN (>4 µmol/g)	2
0.8-4 µmol/g	1
Normal (<0.8 µmol/g)	-1
Rhodanine-positive granules*	1
Urinary copper (in the absence of acute hepatitis)	
Normal	0
1-2x ULN	1
>2x ULN	2
Normal, but >5x ULN after D-penicillamine	2
Mutation analysis	
On both chromosomes detected	4
On 1 chromosome detected	1
No mutations detected	0

TOTAL SCORE	Evaluation:
4 or more	Diagnosis established
3	Diagnosis possible, more tests needed
2 or less	Diagnosis very unlikely

*If no quantitative liver copper available, **or typical abnormalities at brain magnetic resonance imaging. KF, Kayser–Fleischer; ULN, upper limit of normal.

EASL Clinical Practice Guidelines: Wilson's disease. J Hepatol. 2012;56:671-85

Boy with fine, brittle hair

Background

- First, male child of unrelated parents
- Born at 37 weeks by C-section due to fetal distress, 2.4 kg
- Discharged home at 3 days
- No developmental concerns

Presentation

- 4 months – presentation to local pediatrician
  - Loss of head control and tone
  - Deterioration in motor skills
  - Seizure-like episodes
  - EEG showed hypsarrhythmia
  - Managed with vigabatrin
- Referred to Neurology

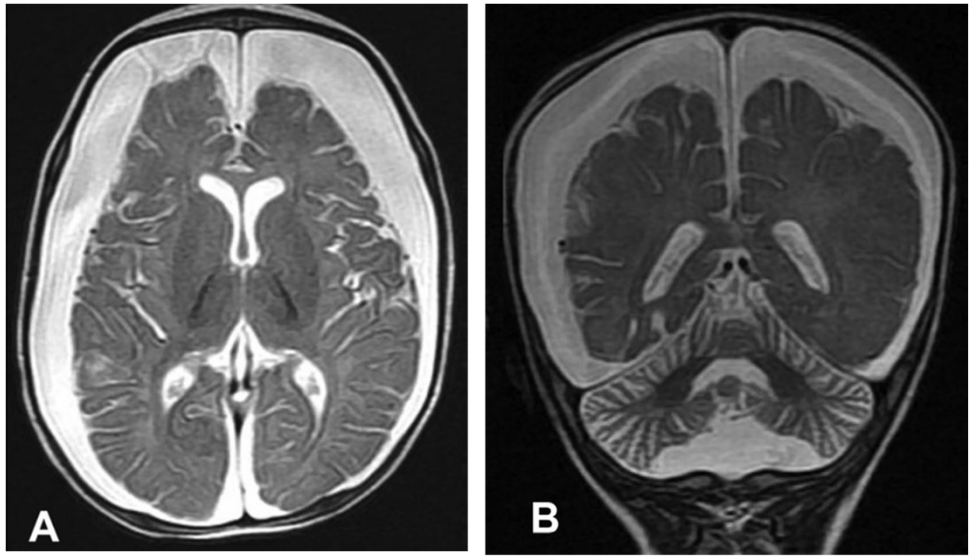
Examination

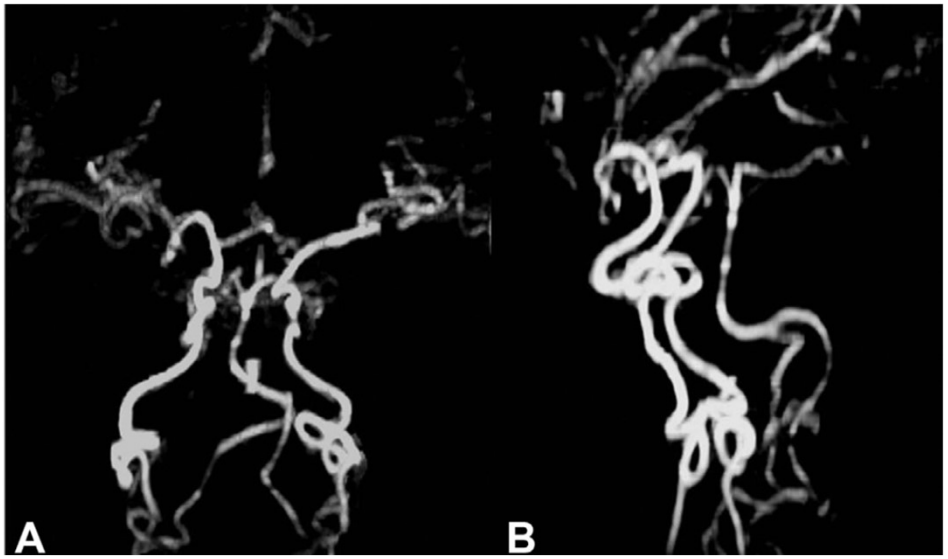
- Height, weight and head circumference on 2nd – 9th centile
- Sparse, fine hair
- Loose skin
- Hypotonic
- No organomegaly

Image from Iran J Pediatr 2007; 17: 388-392

What investigations would you recommend?

MRI





T2: subdural hygromas, symmetric cerebral and cerebellar atrophy

MR angiography (time-of-flight): tortuous extra- / intracranial vessels

DD: shaken baby syndrome

Laboratory Investigations

Test	Result	Reference range
Plasma amino acids	Normal	
DBS acylcarnitines	Normal	
Urine organic acids	Normal	
Transferrin isoforms	Normal	
Biotinidase	Normal	
Serum copper	2.2 µmol/L	10 - 30
Serum caeruloplasmin	< 0.1 g/L	0.23 – 0.51

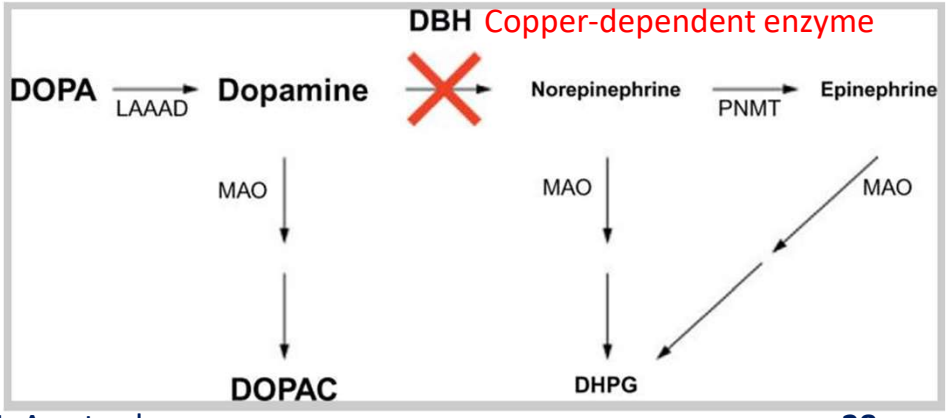
What is the likely diagnosis?

Follow-up

- Genetic testing
  - Hemizygous c.3548 G>A (p.Gly1183Asp) variant in *ATP7A*
  - Defect in copper transport protein ATP7A
  - Confirms diagnosis of Menkes disease
- Analysis of plasma catecholamine ratios
  - Dopamine/norepinephrine
  - DOPAC/DHPG

Is it helpful in this case?

How do you manage the patient?



Management

- Copper-histidine s.c. injections (50-150 µg/kg/d), daily or twice daily
  - Unlikely to improve outcome, esp. if late treatment start
- Anti-convulsant drugs
- Supportive therapy

Adult Aspects

- Comprising ~10% of the patients
- Occipital horn syndrome: juvenile-adult onset, normal IQ, occipital “horns”, bladder diverticula causing recurrent urinary tract infections

- X-linked distal spinal muscular atrophy-3: adult onset, normal caeruloplasmin, hypomorphic missense variants

- Acquired copper deficiency after bariatric surgery

Take home message

- X-linked disorder
- Initial hypotonia replaced by spasticity in older patients
- Intracranial tortuosity and bladder diverticula are common among all subtypes
- Copper and caeruloplasmin levels are physiologically low in first 3 months of life, thereafter, caeruloplasmin is more sensitive than copper (normal in 16%)
- Dopamine/norepinephrine ratio is quite specific
- About 1/3 of patients have *de novo* mutations
- Treatment should not be started after 1 month of age
- Often poor outcome (>40% mortality in childhood) despite early therapy in null mutations

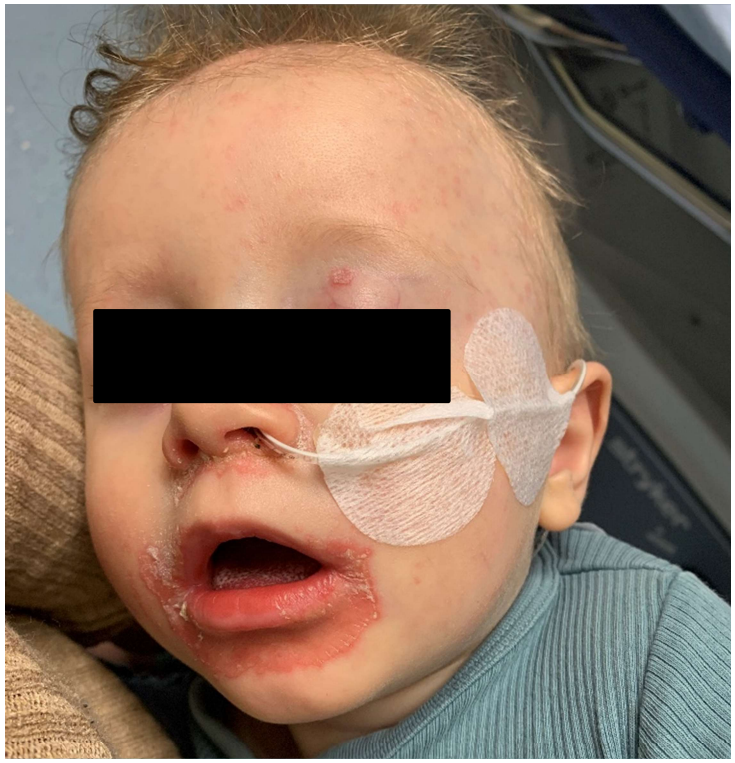
See review: De Feyter S et al, *JIMD*, 2023

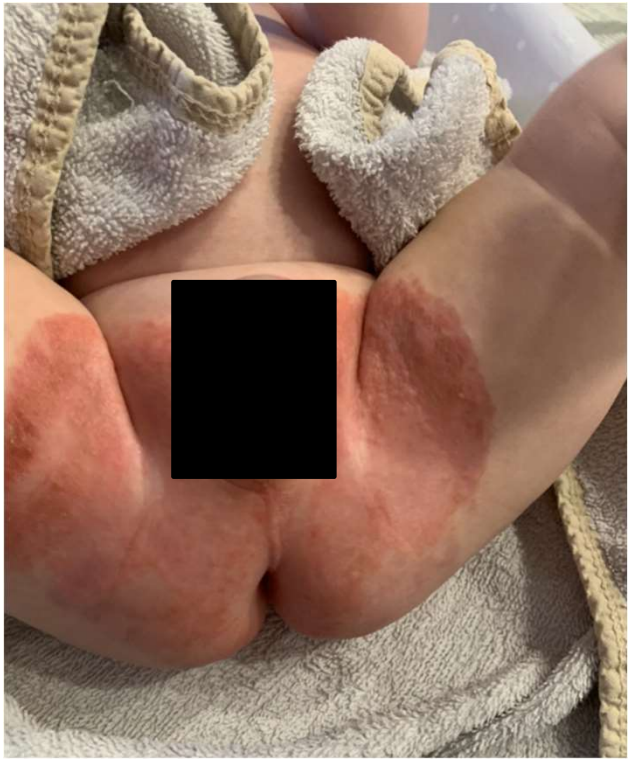
Telemedicine for perioral dermatitis

Case report

- 8 months old boy, fully breast fed
- Normal development, but weak for past weeks
- Since age 6 months: eczema lower lip, extending to entire perioral region and cheeks → telemedicine
- Amoxicillin for 6 days → progression, switch to trimethoprime-sulfamethoxazole (penicillin-resistant Staph aureus)
- At age 9 months: perianal region also affected, diarrhoea, impaired feeding, hair loss
- Father: psoriasis

Patient at 8 months





Images: courtesy of parents

What are your differential diagnoses?

- Infectious dermatitis
- Acrodermatitis enteropathica
- Atopic dermatitis
- Psoriasis
- Langerhans cell histiocytosis

What investigations would you order?

- Blood count, CRP, albumin, AST, ALT, B12: normal
- Alkaline Phosphatase 111 U/L (141-460)
- Zinc 2.1 µmol/L (6.5-17)
- Hair: «kinky» hair aspect on dermatoscopy

What is your suspected diagnosis?

- Acrodermatitis enteropathica

How would you confirm the diagnosis?

Genetic testing (trio exome):

- *SLC39A4:* c.1203G>A; p.Trp401*, homozygous

Treatment

- Zinc per oral (2-3 mg/kg/d elemental zinc); comes in different preparations: zinc chloride, zinc sulfate (may cause gastric upset)
- Rapid improvement of skin lesions and diarrhoea after few days

How would you monitor?

- Zinc, Alk Phosphatase, copper

Why monitor copper?

Background

- Zinc: essential mineral present in most cells, stimulates the activity of about 300 enzymes
- Zinc has main roles in: immune system, wound healing, taste & smell, DNA synthesis
- In plasma, 75% of zinc is bound to albumin
- Dietary zinc is mainly (>70%) absorbed in duodenum
- Uptake through SLC39A4 at the luminal side of enterocytes

Acquired acrodermatitis enteropathica

- Anorexia nervosa, bulimia nervosa
- Crohn's disease
- Celiac disease
- Food allergy
- Intestinal parasitic infestations

Other IEM associated with similar skin lesions?

Take home message

- Think of zinc if dermatitis
- Decreased Alkaline Phosphatase can indicate this condition
- Symptoms of zinc deficiency usually develop after weaning
- A single mutation in *SLC30A2* can reduce breast milk zinc, thus if mother is a carrier, symptoms may start during breast feeding
- Cave: therapeutic zinc can suppress copper uptake

Dietary iron deficiency?

Workshop 2, Case 4

History

- *4 year old girl*
  - *Presents with lethargy and pallor*
  - *Noted to consume large amount of cow's milk*
  - *Full blood count: Hb 9 g/dL (12-16), MCV 70 fL (79-99)*

What's the likely cause and how would you treat?

- **Cow's milk-induced iron deficiency:**
  - Decreased iron absorption
  - Inhibition of non-heme iron absorption by calcium and casein
  - Low iron content in cow's milk
  - Satiety impacting iron-rich food consumption
  - Occult gastrointestinal bleeding

- **Oral iron supplementation**

History

At 10 years age:

- *Seen by General Practitioner*
  - *Complaining of being "tired all the time"*
  - *Acute febrile illness – diagnosed with tonsillitis*

- *Full blood count: Hb 10 g/dL (12-16), MCV 78 fL (79-99)*
- *Ferritin 200 µg/mL (9.5-75)*

Why is the ferritin high?

- *acute phase reactant*

- *Given further courses of iron supplementation*

History

At 16 years:

- *Represents to GP with 2 week history of polyuria polydipsia*
  - *Random glucose 15 mmol/L*
  - *Haemoglobin A1c glycosylated (HbA1c) 50 mmol/mL (20-42)*

- *Diagnosed **diabetes** and treated with insulin*
  - *Islet cell antibodies negative*

- *Persistent microcytic anaemia*
  - *Ferritin 350 µg/mL*
  - *Continued oral iron supplementation*

History

At 25 years

- *Persistent asthenia, weight loss, abdominal pain*
- *Emerging concerns about cognitive impairment*
- *Noted to have bradykinesia, finger-nose ataxia*

    - *Hb 10.9 g/dL (12-16)*
    - *Iron 15 µg/dL (37-170)*
    - *Ferritin 455 ng/mL (10-290)*
    - *Transferrin saturation 8.5% (20-55)*
    - *Blood Film: anisopoikilocytosis*

- *Liver function tests: ALT 55 U/L (0-35), AST 98 U/L (0-50)*

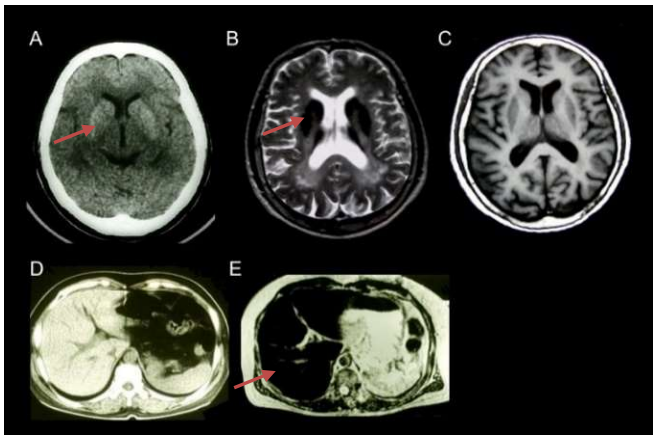
What further tests would you consider?

History

- *Copper 7.6 µmol/L (12.6-26.8)*
- *Caeruloplasmin <2.0 mg/dL (17-65)*

- *Urine copper excretion low/normal*
- *Haemoglobinopathy screen normal*
- *MRI liver: iron overload*
- *MRI brain: iron accumulation in basal ganglia, red nucleus*

What is the diagnosis?



CT **MRI**

Aceruloplasminaemia

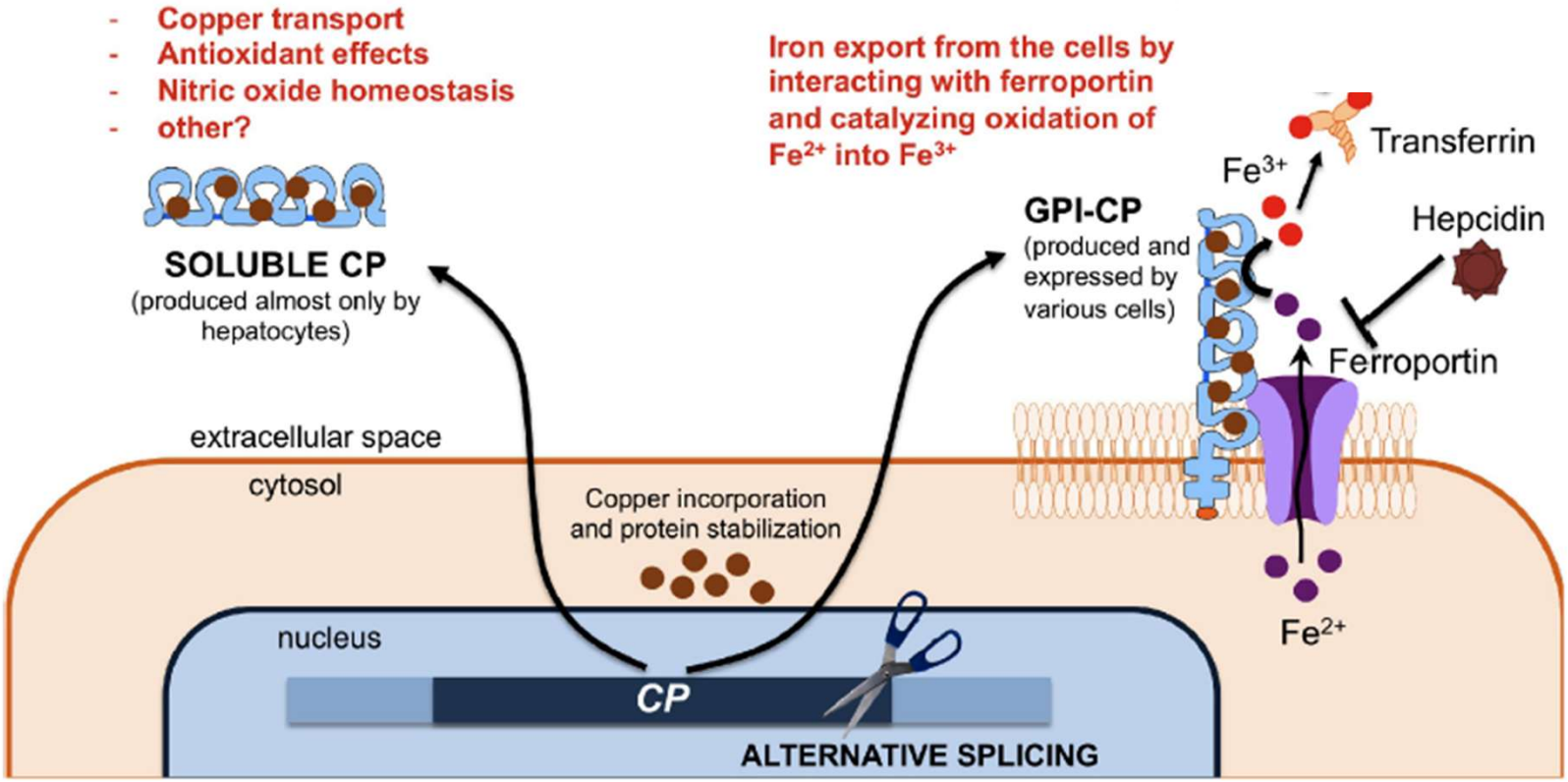
- ***CP* gene:**
  - Biallelic pathogenic variants

- Undetectable caeruloplasmin, low serum copper, normal urine copper excretion
- Iron deficiency microcytic anaemia, raised ferritin, low transferrin saturation
- Tissue iron accumulation
  - Pancreas: diabetes
  - Brain: movement disorder & dementia
  - Liver: chronic liver disease
  - Retina: retinal degeneration
  - Thyroid
  - Cardiac

Ceruloplasmin:

Ubiquitous GPI-anchored membrane form

- brain astrocytes, glial, hepatocytes, macrophages, pancreatic, retinal
- Role in iron export from cells
- Mutation --> intracellular iron accumulation --> tissue pathology

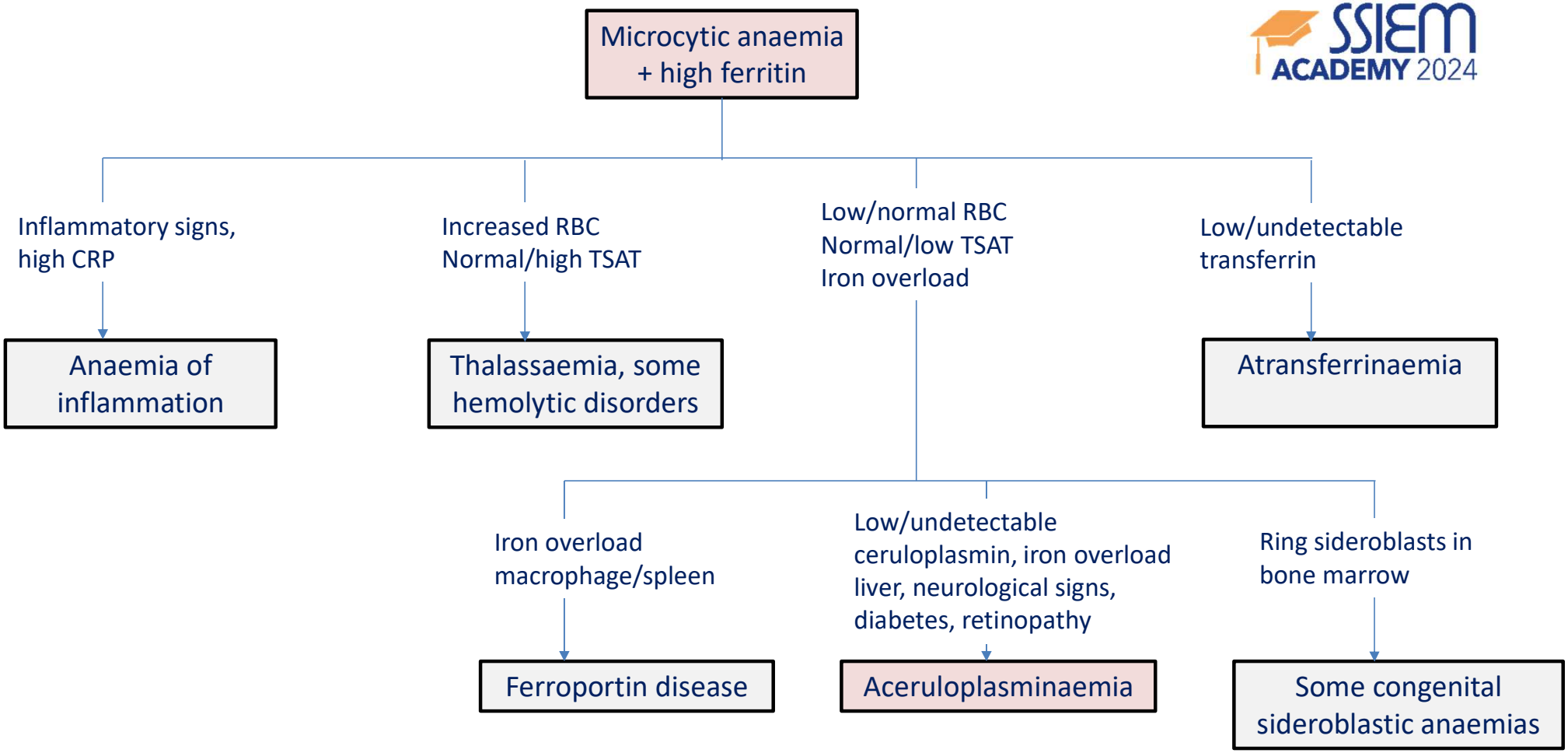


Marchi G, Busti F, Lira Zidanes A, Castagna A and Girelli D (2019) Aceruloplasminemia: A Severe Neurodegenerative Disorder Deserving an Early Diagnosis. Front. Neurosci. 13:325.

Aceruloplasminaemia

- **Treatment options**
  - Iron chelation (desferrioxamine) to lower total iron body stores
  - Phlebotomy?
  - Ceruloplasmin replacement?
  - Fresh frozen plasma?
  - Zinc supplementation?

  - Unclear impact on neurological aspects of disease



Adapted from Int J Mol Sci 2020;21:2374

Take home message

- Iron deficiency microcytic anaemia with persistently elevated ferritin
  - requires investigation
  - Can be early feature of **Aceruloplasminaemia**

- **Aceruloplasminaemia**
  - Multisystem iron overload due to aberrant iron metabolism (failure of cellular iron export)
  - Potentially treatable