Diagnosis of alpha1-antitrypsin deficiency

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• Clinical Recognition of AATD

• Lab Diagnosis of AATD: an interplay of 4 biochemical methods

• AATD Diagnostic Algorithms

• Specific Issues
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ATS/ERS Statement: Standards for the Diagnosis and Management of Individuals with AATD

Type A recommendation for diagnostic testing:
- Symptomatic adults with COPD, or asthma with airflow obstruction incompletely reversible
- Individuals with unexplained liver disease
- Asymptomatic individuals with persistent obstruction on PFTs with identifiable risk factors
- Adults with necrotizing panniculitis

Type A recommendation for predispositional testing:
- Siblings of an AATD individual
Type B recommendation for **diagnostic** testing:

- Adults with bronchiectasis without evident etiology
- Adolescents with persistent airflow obstruction
- Asymptomatic individuals with persistent obstruction on PFTs and no risk factors
- Adults with C-ANCA positive vasculitis

Type B recommendation for **predispositional** testing:

- Individuals with family history of COPD or liver disease not known to be attributed to AATD
- Distant relatives of an AATD individual
- Offspring or parents of an AATD individual
- Siblings, offspring, parents, distant relative of an individual heterozygous for AATD allele
Targeted Screening

Suggestions for Clinical Recognition of AATD

- Early onset emphysema (age 45 or less)
- Emphysema in the absence of a recognized risk factor
- Emphysema with prominent basilar hyperlucency
- Unexplained Bx
- Unexplained liver disease
- Necrotizing panniculitis
- C-ANCA +ve vasculitis
- Family history of: emphysema, Bx, liver disease, panniculitis
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Protein electrophoresis of serum from a patient with AAT deficiency

α₁-band missing

Serum of AATD patient
Serum of healthy control

Modified from Eriksson Chest 1989 (95)

α₁-band missing

Courtesy Talecris Slide Kit 2008
Possible phylogenetic tree of the AAT gene
Thorax 2004
SERPINA1 product: protein (AAT) quantitative determination

Serum Alpha1-antitrypsin protein concentration

- (Rocket immunoelectrophoresis)
- (Radial immunodiffusion)
- Nephelometry
SERPINA1 product: protein (AAT) qualitative determination

Isoelectric focusing (IEF)

("phenotype")

Anode

Cathode

M1 M2  M1 S  M1 I  M1 M1  M1 Z  Z Z

F

M

S

Z
SERPINA1 gene:
Z and S mutation rapid analysis

Genotyping
SERPINA1 gene: sequence analysis

Sequence analysis

PI*M/Mprocida

PI*M/Mmalton

PI*M/Plowell

C insertion

Deletion
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Evaluation of an Integrative Diagnostic Algorithm for the Identification of People at Risk for $\alpha_1$-Antitrypsin Deficiency

Joshua A. Bornhorst, PhD, Melinda Procter, Cindy Meadows, Edward R. Ashwood, MD, and Rong Mao, MD

Am J Clin Pathol 2007;128:482-490
DOI: 10.1309/4dKBCFO8E9D1B8

From the Departments of Pathology, University of Arkansas for Medical Sciences, Little Rock, and University of Utah, Salt Lake City; and ARUP Institute for Clinical and Experimental Pathology, Salt Lake City.
Diagnosis of α-1-Antitrypsin Deficiency: An Algorithm of Quantification, Genotyping, and Phenotyping

Melissa R. Snyder, Jerry A. Katzmann, Malinda L. Butz, Ping Yang, D. Brian Dawson, Kevin C. Halling, W. Edward Hightsmith, and Stephen N. Thibodeau

Clinical Chemistry 52:12
2234–2242 (2006)

– Division of Clinical Biochemistry & Immunology, Department of Laboratory Medicine & Pathology.
– Division of Hematology, Department of Internal Medicine.
– Division of Laboratory Genetics, Department of Laboratory Medicine & Pathology.
– Department of Health Sciences Research, Mayo Clinic College of Medicine, Rochester, MN.
Identification of individuals with alpha-1-antitrypsin deficiency by a targeted screening program

Robert Bals\textsuperscript{a,*}, Rembert Koczulla\textsuperscript{a}, Viktor Kotke\textsuperscript{a}, Juergen Andress\textsuperscript{a}, Karlheinz Blackert\textsuperscript{b}, Claus Vogelmeier\textsuperscript{a}

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Sample collection (DBS)

- AAT protein level: nephelometer
- S and Z variant genotype: SexAI/Hpy99I RFLP

AAT > 113 mg/dl

- S Z genotype: negative
- Phenotype (IEF)

AAT < 113 mg/dl

- S Z genotype: positive
- Phenotype (IEF)
  - Sequencing
  - PI*ZZ or PI*SS?
    - yes
    - AAT < 70 mg/dl?
      - yes
      - Sequencing
      - no
    - no

Homozygous for deficient allele?

1 = normal
2 = intermediate AATD
3 = severe AATD (compound heterozygous)
4 = severe AATD (homozygous)

Ferrarotti et al, *Transl Res* 2007;150:267
*Erratum 2008:151:232*
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Look at the discordance!

A: AAT plasma level
Patient D: 41.2 mg/dL
Patient E: 22 mg/dL

B: Genotyping

C: Phenotyping

D: Sequencing patient D (exon 5)
Dried blood spot (DBS) is not serum/plasma
Validation of a simple, rapid method to measure $\alpha_1$-AAT in human dried blood spots

Gorrini M et al

Clin Chem 2006;52:899
Validation of a simple, rapid method to measure α1-AT in human dried blood spots

Gorrini M et al
Clin Chem 2006;52:899
RELATIONSHIP BETWEEN C-REACTIVE PROTEIN AND ALPHA₁-ANTITRYPsin LEVELS DETERMINED IN DRIED BLOOD SPOT FLUID

\[ y = 0.9753x + 0.0085 \]

\[ R^2 = 0.9927 \]
Stratification of AAT levels in a series of PI*MZ individuals according to C Reactive Protein level

![Graph showing the stratification of AAT levels. The horizontal line at 113 mg/dL separates points above for CRP > 0.8 mg/dL and below for CRP ≤ 0.8 mg/dL.](image-url)