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# PART 1

## AUTOANTIBODIES IN SYSTEMIC AUTOIMMUNE DISEASES

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### Alanyl-tRNA (tRNA<sup>Ala</sup>) Synthetase Antibodies

#### ➤ PL-12 antibodies

### Alpha-Enolase Antibodies

#### Autoantigen

The ubiquitous glycolytic enzyme  $\alpha$ -enolase is 82% homologous with the other two isoforms,  $\beta$ - and  $\gamma$ -enolase, and homologous with tau, a soluble lens protein of the crystalline family.

#### Detection Methods

Enzyme immunoassay (EIA) or Western blot (WB) using purified  $\alpha$ -enolase.

#### Clinical Relevance

- **Parameter of nephropathy:**
  - Detected in 69% of patients with primary membranous nephropathy and in 58% of those with the secondary form (patients with ➤ SLE and ➤ rheumatoid arthritis)

- Detected in 27% of patients with SLE; associated with active renal disease.
- Found in 32% of patients with mixed ➤ cryoglobulinemia with nephritis, but not in cryoglobulinemia without nephritis.
- **Prognostic marker for ➤ systemic sclerosis?**  
Detected in 30% of scleroderma patients and associated with severity of disease (pulmonary and vascular involvement).
- Alpha-enolase was identified as a target autoantigen in sera of **endometriosis** patients.
- Autoantibodies are commonly (69%) associated with paraneoplastic retinal degeneration (**cancer-associated retinopathy**).
- ANCA-associated diseases (vasculitis, chronic-inflammatory bowel disease): Alpha-enolase is a cytoplasmic enzyme of neutrophils.
- Low titers of alpha-enolase antibodies are found in autoimmune liver diseases, e.g., in 27% of patients with primary sclerosing cholangitis, in 29% with primary biliary cirrhosis (PBC), and in 30–60% with autoimmune hepatitis. The presence of these antibodies in patients with hepatitis is associated with rapidly progressive disease.

### Indications

None currently established. It remains to be determined whether testing for these autoantibodies will provide any clinical advantages for diagnosis of mixed cryoglobulinemia (renal involvement), retinopathies (paraneoplasia?) or other diseases.

**Comments:** Alpha-enolase antibodies are a heterogeneous group of autoantibodies that recognize various epitopes of  $\alpha$ -enolase proteins (and some of  $\beta$ - and  $\gamma$ -enolase proteins) and perhaps some related proteins as well (e.g., the lens protein tau). Their relationship to ➤ ANCA and ➤ centrosome antibodies (centrosome antibody positive sera detected purified  $\alpha$ - and  $\gamma$ -enolase [Rattner et al. 1991] ) still needs to be clarified. Further investigations are therefore necessary for differentiation and, thus, for improved clinical utility of  $\alpha$ -enolase antibodies.

## Alpha-Fodrin Antibodies

### Autoantigen

The 120 kDa alpha-fodrin binds to beta-fodrin to form a heterodimer - abundant proteins of eukaryotic cell membrane cytoskeleton.

### Detection Methods

WB or EIA using the recombinant antigen; commercial assay kits are now available.

### Clinical Relevance

Detected in adults with primary (55-75%) and secondary (40-60%) ➤ **Sjögren's syndrome**; rarely detected in SLE without Sjögren's syndrome (2-7%) and healthy blood donors (0.6-1.9%). Other up to now unpublished studies found lower prevalence and specificity, however. IgA antibodies seem to be more sensitive and more specific for Sjögren's syndrome than IgG antibodies [Witte et al. 2000]. Associated with hypergammaglobulinemia, ➤ rheumatoid factor, ➤ La/SS-B and ➤ beta-fodrin antibodies. Not associated with *erythema annulare*, photosensitivity, vasculitis or renal involvement. Alpha-fodrin antibodies can be detected in up to 100% in patients with juvenile onset of Sjögren's syndrome, in some cases before ➤ Ro/SS-A or La/SS-B antibody become positive [Kobayashi et al., 2001]. They are also detectable at higher frequencies in patients with ➤ rheumatoid arthritis [Witte et al. 2000], ➤ juvenile idiopathic arthritis and juvenile SLE [Takahashi et al 2001].

### Indications

None currently established and early data requires corroboration. May gain significance in diagnosis of Sjögren's syndrome, especially at early stages or in subclinical or Ro/SS-A antibody negative forms. The relevance for juvenile systemic autoimmune diseases remains to be established.

## Aminoacyl-tRNA Synthetase Antibodies

### Synonym:

Anti-synthetase antibodies

**Collective term** for all autoantibodies directed against aminoacyl-tRNA synthetases. The following ones have been described so far:

- Histidyl-tRNA synthetase antibodies (➤ Jo-1 antibodies),
- Threonyl-tRNA synthetase antibodies (➤ PL-7 antibodies),
- Alanyl-tRNA synthetase antibodies (➤ PL-12 antibodies),
- Isoleucyl-tRNA synthetase antibodies (OJ antibodies),
- Glycyl-tRNA synthetase antibodies (EJ antibodies),
- Lysyl-tRNA synthetase antibodies (SC antibodies),
- AsparaginyI-tRNA synthetase antibodies (KS antibodies),
- Leucyl-tRNA synthetase antibodies,
- GlutaminyI-tRNA synthetase antibodies.

### Autoantigens

Aminoacyl-tRNA synthetases. These cytoplasmic enzymes catalyze the binding of a specific amino acid to the corresponding transfer RNA (tRNA) molecule. For instance, the amino acid histidine is bound to tRNA<sup>His</sup> with the aid of histidyl-tRNA synthetase, leading to the formation of histidyl-tRNA complexes.

### Detection Methods

- Indirect immunofluorescence (IIF) using HEp-2 cells: some sera with this reactivity produce diffuse or fine granular staining of the cytoplasm.

**NOTE:** The staining pattern is not seen with all sera and when it is seen, it is suggestive of but not specific for anti-synthetase antibodies.

- Immunodiffusion (Ouchterlony technique) or counter-current immunoelectrophoresis (CIE) using soluble antigens from rabbit thymus or human spleen.

**NOTE:** This technique has a low sensitivity and does not detect all aminoacyl-tRNA synthetase antibodies.

- WB using HeLa or MOLT4 cell extracts or special aminoacyl-tRNA synthetase preparations.
- NOTE:** False-negative results are possible.
- EIA using biochemically purified or recombinant aminoacyl-tRNA synthetases.
  - Immunoprecipitation (IP), e.g., radioimmunoprecipitation of tRNA from <sup>32</sup>P-labeled cell extracts (HeLa S3 cell lineage).

**Comments:** Immunodiffusion or CIE (Jo-1, PL-7, PL-12 antibodies), EIA and WB (Jo-1 antibodies) are currently used in routine laboratory testing.

### Clinical Relevance

Aminoacyl-tRNA synthetase antibodies are specific markers for idiopathic (autoimmune) forms of myositis, especially ➤ **polymyositis**. The diseases associated with these autoantibodies usually have similar symptoms and are therefore referred to as ➤ anti-Jo-1 or **anti-synthetase syndrome**. Fibrosing alveolitis is one of the most conspicuous clinical feature in these patients.

Except for Jo-1 antibodies, the individual aminoacyl-tRNA synthetase antibodies are rarely found in idiopathic myositis (< 1-3%). These antibodies generally do not cross-react or co-exist with others.

At least one of the aforementioned aminoacyl-tRNA synthetase antibodies is found in 60 to 80% of all patients with myositis and interstitial lung disease.

### Indications

1. Diagnosis/differential diagnosis of myositis.
2. Clarification of suspected diagnosis of fibrosing alveolitis.

**Comments:** Even when the available tests for Jo-1, PL-7 and PL-12 antibodies are not positive, the detection of immunofluorescence patterns typical of aminoacyl-tRNA synthetase antibodies may still be a valuable diagnostic indicator in patients with suspected idiopathic myositis, especially in those with concomitant pulmonary fibrosis. In these cases, the occurrence of other aminoacyl-tRNA synthetase antibodies or ➤ KJ antibodies is possible.

## Annexin V Antibodies

### Autoantigen

Annexin V, a  $\text{Ca}^{2+}$ -dependent, phospholipid-binding protein that is abundant in cells exposed to blood, eg., platelets, trophoblast (mainly syncytiotrophoblast) and endothelial cells. Annexin V inhibits procoagulant reaction and is probably involved in multiple cellular functions, e.g., transmembrane  $\text{Ca}^{2+}$  channel activity, inhibition of phospholipase A2, regulation of membrane integrity.

### Detection Methods

EIA using annexin V immobilized on  $\gamma$ -irradiated polystyrene plates.

### Clinical Relevance

Annexin V antibodies have been detected in patients with ➤ **antiphospholipid syndrome (APS)** and ➤ connective tissue diseases, especially in ➤ systemic lupus erythematosus and ➤ rheumatoid arthritis. Patients with annexin V antibodies have clinical features of APS even in the absence of ➤ phospholipid antibodies [Kaburaki et al. 1997]. These autoantibodies are closely related to **fetal loss**. They are found in 36% of patients with habitual abortions and 20% of preeclampsia [Matsuda et al. 1994]. The prevalence and levels of annexin V antibodies are significantly higher in patients with SLE and fetal loss as well as venous thrombosis than those without these complications. A multivariate logistic regression analysis of risk factors reveals that annexin V antibody is a significant risk factor for fetal loss in SLE patients: OR 5.9, 95% CI 1.4-14.8 [Nojima et al. 2001].

The actions of annexin V antibodies may affect embryo implantation and pregnancy outcome. In pregnant BALB/c mice the infusion of annexin V antibodies lead to the induction of fetal loss, placental thrombosis and tissue necrosis. It has been shown that these autoantibodies can induce endothelial cell and syncytiotrophoblast apoptosis and the inhibition of trophoblast gonadotropin secretion [di Simone et al 2001].

### Indications

None currently established. The determination may be useful in fetal loss risk assessment in patients with phospholipid antibodies and in patients with connective tissue diseases, especially SLE patients.

## Antineutrophil Cytoplasmic Antibodies (ANCA)

**Collective term** for all autoantibodies directed against cytoplasmic antigens of neutrophils. These include:

- Azurocidin antibodies,
- BPI antibodies,
- cANCA (➤ proteinase 3 antibodies),
- pANCA (➤ myeloperoxidase antibodies).

Autoantibodies against cytoplasmic antigens of human neutrophils were discovered in patients with segmental necrotizing glomerulonephritis and systemic vasculitis in the early 1980's [Davies 1982, Hall 1984]. These antibodies initially referred to as anticytoplasmic antibodies (ACPAs) were soon found to be specific for ➤ Wegener's granulomatosis (WG) [van der Woude 1985]. Investigators later demonstrated that the antibody titers correlate with disease activity. In addition to the classical ACPA (later named cANCA), a second primary immunofluorescence pattern (pANCA) was found in patients with necrotizing glomerulonephritis and ➤ microscopic polyangitis (MPA) [Falk 1988]. Myeloperoxidase (MPO) was identified as the target antigen of MPA-specific ANCA, and proteinase 3 (PR3) of WG-specific ANCA [Falk 1988, Jenne 1990].

### Autoantigens

Enzymes of the azurophile and specific granules of neutrophil granulocytes, in part also present in monocytes and endothelial cells:

| ANCA<br>target antigen | Immunofluorescence pattern on<br>ethanol fixed human neutrophils |
|------------------------|------------------------------------------------------------------|
|------------------------|------------------------------------------------------------------|

|                 |                          |
|-----------------|--------------------------|
| Proteinase 3    | cANCA, vary rare pANCA   |
| Myeloperoxidase | pANCA, vary rare cANCA   |
| Elastase        | pANCA                    |
| Cathepsin G     | pANCA                    |
| Azurocidin      | pANCA                    |
| Lactoferrin     | pANCA                    |
| Lysozyme        | pANCA                    |
| BPI             | atypical ANCA oder pANCA |

Ubiquitous proteins such as  $\alpha$ -enolase, HMG proteins, histone 1, lamin B1, and EEA-1 were also identified as target antigens of ➤ neutrophil-reacting autoantibodies.

### Detection Methods

- IIF using ethanol-fixed human neutrophils produces different cytoplasmic staining patterns, e.g., granular cytoplasmic (➤ cANCA), perinuclear (➤ pANCA), and atypical (aANCA or xANCA).
- EIAs using highly purified or recombinant antigens (➤ myeloperoxidase antibodies, ➤ proteinase 3 antibodies).

### Clinical Relevance

- cANCA (specific for proteinase 3) and pANCA (specific for myeloperoxidase) are markers of systemic vasculitides, which are therefore referred to as ➤ **ANCA-associated vasculitides**.
- Apart from these classical ANCA, ➤ neutrophil-specific antibodies (NSA) with perinuclear or nuclear patterns (atypical ANCA; xANCA; GS-ANA) are found in ➤ rheumatoid arthritis, ➤ Felty's syndrome, chronic inflammatory bowel disease, primary sclerosing cholangitis, and autoimmune hepatitis type 1. These autoantibodies target many different autoantigens

except proteinase 3 and myeloperoxidase (see autoantigens) and should be clearly distinguished from cANCA or pANCA [Wiik 2002].

- ANCA are also found in cystic fibrosis (➤ BPI antibodies).

### Indications

1. Suspicion of ANCA-associated vasculitis.
2. Differential diagnosis of nephritis.
3. Suspicion of chronic inflammatory bowel disease.
4. Suspicion of primary sclerosing cholangitis.
5. Hemorrhagic alveolitis.

## Antinuclear Antibodies (ANA)

**Collective term** for all autoantibodies directed against conserved (e.g., those occurring in all cell types) nuclear antigens. These include:

- Centromere antibodies,
- Coilin antibodies,
- DEK antibodies,
- dsDNA antibodies,
- Histone antibodies,
- IFI 16 antibodies,
- La/SS-B antibodies,
- Mi-2 antibodies,
- Nucleolar antibodies,
- Nucleosome antibodies,
- PCNA antibodies,
- Ro/SS-A antibodies,
- Scl-70 antibodies,
- Sm antibodies,
- ssDNA antibodies,
- U1-RNP antibodies.

In addition, various other proteins of different nuclear compartments (coiled bodies, PML bodies, nuclear matrix, nuclear membrane/nuclear pore complexes) may be targets of antinuclear antibodies.

In the narrower sense, ANA are those autoantibodies that cause nuclear staining in tissue specimens (e.g., rat liver) or human tumor cell monolayer (e.g., HEp-2 cells) used in IIF.

### Autoantigens

These (non-organ-specific) antigens are ubiquitous in cell nuclei. They may be located in chromatin or in the nucleolus, nucleoplasm, nuclear matrix, or nuclear membrane. The major target antigens of antinuclear antibodies include double-stranded deoxyribonucleic acid (dsDNA) and DNA-associated proteins (histone, centromere proteins) as well as cytobiologically important enzymes (e.g., splicing enzymes, RNA polymerases, DNA topoisomerase I). Nuclear compartments and autoantigens include:

| Localisation                 | Antigenes                                                                                       |
|------------------------------|-------------------------------------------------------------------------------------------------|
| Chromatin                    | dsDNA, ssDNA, histones, high mobility group (HMG) proteins                                      |
| Centromere                   | centromere-associated proteins (CENP-A, -B, -C)                                                 |
| Nucleolus                    | nucleolar RNA or nucleolar proteins (fibrillarin, RNA polymerase, PM-Scl antigen)               |
| Nucleoplasm/<br>Nucleomatrix | proteins of U-snRNP (Sm- and U1-RNP antigen) or Y-snRNP complexes (Ro/SS-A and La/SS-B antigen) |
| Coiled bodies                | p80 coilin                                                                                      |
| PML bodies                   | Sp100                                                                                           |
| Nuclear membrane             | lamins, lamin-B receptor, gp210                                                                 |

### Detection Methods

- IIF using monolayer of human tumor cells: nuclear labeling. The nuclear pattern varies according to the localization of the nuclear antigen.

**NOTE:** HEp-2, a human laryngeal carcinoma cell line, is most commonly used. All diagnostically relevant antinuclear antibody specificities (dsDNA, histone, Sm, U1-RNP, Ro/SS-A, Scl-70, centromere and antinucleolar antibodies) should test positive by this technique.

- EIAs using whole nuclei, nuclear extracts, or mixtures of purified or recombinant nuclear proteins.

**NOTE:** The results of different EIAs may vary depending on the type of antigen mixture used (nuclear extracts, mixtures of purified and/or recombinant antigens). A good assay should allow the detection of all diagnostically relevant ANA specificities.

### Clinical Relevance

Antinuclear antibodies are a typical finding in systemic inflammatory rheumatic diseases (➤ **connective tissue diseases**, especially ➤ systemic lupus erythematosus, ➤ Sjögren's syndrome, ➤ mixed connective tissue disease, and ➤ systemic sclerosis). Nonetheless, ANA occur in variable frequencies in almost all other autoimmune diseases as well as tumors. ANA are rarely found (usually in low titers) in healthy subjects, and their frequency of detection rises with age.

The presence of ANAs is indicative of autoimmune hepatitis in patients with chronic liver disease, and of an increased risk of chronic iridocyclitis (anterior uveitis) in those with ➤ juvenile idiopathic arthritis.

### Indications

1. Screening of patients suspected of having an inflammatory rheumatic disease.
2. Suspicion of drug-induced lupus.
3. Suspicion of autoimmune hepatitis.
4. Differential diagnosis/prognosis of juvenile idiopathic arthritis

**Comments:** Depending on the diagnostic query and observed immunofluorescence pattern, positive ANA test result should be followed up by assays to determine the type of ANA specificity.

## Asparaginyl-tRNA (tRNA<sup>Asp</sup>) Synthetase Antibodies

**Synonym** (named after prototype patient):

KS antibody

➤ **Aminoacyl-tRNA synthetase antibodies**

## Azurocidin Antibodies

**Synonym:**

CAP37 antibody

➤ **ANCA**

### Autoantigen

Azurocidin, a cationic antimicrobial protein (CAP37) found in the azurophil granules of neutrophils.

### Detection Methods

- IIF using ethanol-fixed human neutrophils reveals a typical pANCA pattern in monospecific antisera.
- EIA using biochemically purified azurocidin.

**Comments:** The positive reactivities observed in the EIA do not correlate with the pANCA pattern! Azurocidin antibodies are detected in roughly 8% of all pANCA-positive sera, but also in about 3% of cANCA-positive and 8% of ANCA-negative sera analyzed by EIA [Yang 1996].

### Clinical Relevance

- Azurocidin antibodies are most commonly found in patients with systemic vasculitis. They are not associated with pauci-immune glomerulonephritis.
- Azurocidin antibodies are often detected in combination with other ANCA specificities (e.g., ➤ MPO, ➤ BPI and lactoferrin antibodies). Hydralazine-induced vasculitis was diagnosed in 4 out of 6 patients with co-expression of azurocidin, MPO and lactoferrin antibodies [Zhao 1996].

### Indications

None currently established because too few studies are available. Moreover, the frequent co-expression of other ANCA specificities hampers the interpretation of test results. Further investigations are therefore needed to determine whether azurocidin antibodies can be useful in diagnosing systemic vasculitis.

## Beta-Fodrin Antibodies

### Autoantigen

Beta-Fodrin, a membrane skeleton protein associated with ion channels and pumps. Forms together with alpha-fodrin the fodrin heterodimer, an abundant protein of eukaryotic cell membrane cytoskeleton. The immunoreactive amino-terminal region shows no homology to alpha-fodrin.

### Detection Methods

EIA or WB using the amino-terminal fragment of beta-fodrin. The whole protein shows no reactivity!

### Clinical Relevance

Frequently detected in patients with ➤ **Sjögren's syndrome** (70%), but infrequently found in patients with other ➤ connective tissue diseases without Sjögren's syndrome (12%) or healthy controls (4%). Beta-fodrin antibodies

more frequently occurred together with ➤ alpha-fodrin antibodies than with ➤ Ro/SS-A or ➤ La/SS-B antibodies. The cleavage and altered distribution of beta-fodrin in glandular epithelial cells may induce autoantibody production and glandular destruction in Sjögren's syndrome patients [Kuwana et al. 2001].

### Indications

None established because only one study is currently available [Kuwana et al. 2001]. May gain significance in diagnosis of Sjögren's syndrome.

## Beta-2 Glycoprotein I ( $\beta$ 2-GPI) Antibodies

### Synonym:

Apolipoprotein H antibodies

**Introduction/History:** Harris and colleagues [1983] introduced a radioimmunoassay for detection of cardiolipin antibodies which was soon replaced by an EIA [Loizou et. al. 1985]. In the course of numerous clinical trials, investigators found that the detection of cardiolipin antibodies and other phospholipid antibodies (aPL) was enhanced by a cofactor, which was identified by several independent groups as  $\beta$ 2-glycoprotein I (apolipoprotein H) [McNeil 1990; Matsuura 1990]. This cofactor has characteristic binding affinities for cardiolipin and other phospholipids (PL). By binding to anionic (negatively charged) phospholipids, it induces a conformational change in the  $\beta$ 2-glycoprotein I molecule with the aid of a cryptic epitope responsible for aPL binding. These aPL are associated with the well-known manifestations of the ➤ antiphospholipid syndrome (APS), i.e. thrombophilia, recurrent spontaneous abortion, neurologic complications, etc. On the other hand, the transient occurrence of aPL independent of  $\beta$ 2-glycoprotein I ( $\beta$ 2-GPI) is observed in AIDS and in a number of infectious diseases such as syphilis, malaria, leprosy, borreliosis, tuberculosis, rubella, mumps and infections induced by streptococci, mycoplasma, salmonella and coli bacteria. Consequently, these aPL are not specifically associated with the clinical picture of APS.

Galli and other investigators first reported antibodies exclusively directed against  $\beta$ 2-glycoprotein I in the absence of phospholipids in 1990. Using a radioactive or plasma irradiation technique, Matsuura and colleagues introduced negatively charged groups into the surface of the polystyrene plates

used in EIAs.  $\beta$ 2-GPI consequently undergoes a conformational change that leads to the expression of a neoepitope. However, Koike's group [1998] demonstrated by phage display that the antibodies directed against the cardiolipin/ $\beta$ 2-glycoprotein I complex are not identical to  $\beta$ 2-glycoprotein I antibodies.

In 1994, Blank and colleagues succeeded in inducing APS in mice immunized with  $\beta$ 2-GPI. In this experimental model, APS was characterized by the co-occurrence of antibodies directed against both cardiolipin and  $\beta$ 2-GPI. A number of research groups later demonstrated that  $\beta$ 2-GPI antibodies occur in patients with primary and secondary APS. The  $\beta$ 2-GPI antibodies appear to be a diagnostic marker for thrombotic events in these groups of patients.

### Autoantigen

$\beta$ 2-glycoprotein I consists of 326 amino acids and has a molecular weight of  $\sim 50$  kDa. Approximately 19% of the molecule is comprised of carbohydrates. The plasma concentration of  $\beta$ 2-GPI is approx. 200 mg/l.  $\beta$ 2-GPI inhibits the intrinsic pathway of blood coagulation.

### Detection Methods

EIA using human  $\beta$ 2-GPI immobilized on  $\gamma$ -irradiated polystyrene plates.

### Clinical Relevance

- Diagnostic marker of  $\blacktriangleright$  **antiphospholipid syndrome (APS)**.  $\beta$ 2-GPI antibodies are associated with arterial or venous thrombosis and recurrent spontaneous abortion.  $\beta$ 2-GPI antibodies are reported to have a higher specificity but lower sensitivity than  $\blacktriangleright$  cardiolipin antibodies, but they are not yet included in the classification criteria for APS.
- $\beta$ 2-GPI antibodies can be of the IgG, IgM or IgA isotype; those of the IgA isotype seems to be most prevalent. No confirmed data on the possible association of the isotypes to specific disease manifestations are currently available. However,  $\beta$ 2-GPI antibodies, especially those of the IgG isotype, are associated with arterial and venous thrombosis.
- The prevalence and levels of  $\beta$ 2-GPI antibodies are significantly higher in patients with SLE and arterial or venous thrombosis than those without these complications. A multivariate logistic regression analysis of risk factors reveals that  $\beta$ 2-GPI antibody is a significant risk factor for arterial thrombosis in SLE patients [Nojima et al. 2001].
- $\beta$ 2-GPI antibodies are seldom detectable in infectious diseases.

### Indications

1. Suspicion of primary APS (PAPS): Patients under 45 years of age with recurrent and inexplicable deep venous or arterial thrombosis without the associated risk factors or history of recurrent spontaneous abortion are especially suspicious for PAPS.
2. Suspicion of secondary APS (SAPS) in patients with vascular complications (thrombosis, vasculopathies) secondary to ➤ SLE or other forms of ➤ connective tissue diseases.
3. Risk assessment for thrombophilia and spontaneous abortion in patients at risk for APS (connective tissue diseases, especially SLE).

**Comments:** Patients with clinical signs of APS should always be tested for ➤ cardiolipin antibodies or ➤ lupus anticoagulant when  $\beta$ 2-GPI antibodies have been detected (see Appendix VI).

The isolated occurrence of  $\beta$ 2-GPI antibodies in cardiolipin-negative patients is possible. Conversely, APS patients positive for cardiolipin antibodies may be negative for  $\beta$ 2-GPI antibodies. Parallel screening for cardiolipin and  $\beta$ 2-GPI antibodies is therefore recommended when APS is suspected.

### BPI Antibodies

**Synonyms:**

CAP57 antibodies  
BPI-ANCA

➤ ANCA

### Autoantigen

BPI (bactericidal permeability-increasing protein), a cationic antimicrobial protein (CAP57) found in the azurophil granules of neutrophils.