Pediatric Congenital Hematologic
Disorders Research Center
مرکزتحقیقات بیماری های خونی مادرزادی کودکان

Neutropenia

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TABLE III. Classification of Neutropenias (EO, 8.4, A)

- Isolated neutropenias
  - Severe congenital neutropenias (SCN)
    - with known genetic lesion
      - ELA2 (autos dom, sporadic)
      - HAX 1 (autos rec.) can be associated to neurologic symptoms
    - without known genetic lesion
  - Cyclic neutropenia (CyN) (ELA2, autos dom, sporadic)
  - Autoimmune neutropenia (AIN)
  - Neonatal allo-immune neutropenia
  - Post-infectious neutropenia
  - Drug-related neutropenia
  - Familial benign/ethnical neutropenia
  - Idiopathic neutropenia (IN)
- Neutropenias associated to other pathological condition
  - Associated to mitochondrial diseases:
    - Shwachman–Bodian–Diamond syndrome
    - Pearson syndrome
  - Associated to congenital organ malformations:
    - G6PC3 gene mutation
    - Blackfan–Diamond syndrome
  - Associated to metabolic diseases:
    - Glicogenosis Ib
    - Organic-acidosis
    - Tyrosinemia
    - Barth syndrome
    - Gaucher disease

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Associated to immunodeficit:
- Hyper IgM
- Hypoagammaglobulinemia X-linked
- Common variable immunodeficiency
- Isolated IgA deficiency
- Reticular dysgenesia
- Dubowitz syndrome
- WHIM's syndrome
- Cohen syndrome
- X-linked neutropenia
- GFI1 deficiency

Associated to immunodeficit with hypopigmentation:
- Griscelli syndrome (type 2)
- Chediack–Higaschi syndrome
- Hermansky–Pudlak syndrome (type 2)
- P14 deficiency

Associated to autoimmune diseases:
- SLE
- Rheumatoid arthritis or Felty syndrome
- Scleroderma
- Sjogren syndrome
- Autoimmune lymphoproliferative syndrome (ALPS)
- Celiac disease
- Primitive biliary cirrhosis
- Crohn disease
- Associated to activation of C5

Associated to nutritional deficiencies:
- Vitamin B12 deficiency
- Folate deficiency
- Copper deficiency

Associated to intrinsic or extrinsic marrow failure:
- Aplastic anemia
- Myelodysplastic syndromes

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- Associated to intrinsic or extrinsic marrow failure:
  - Aplastic anemia
  - Myelodysplastic syndromes
  - Primitive or secondary macrophage activation
  - Fanconi anemia
  - Dyskeratosis congenita
  - Hair-cartilage hypoplasia
  - Myelofibrosis
  - Osteopetrosis
  - Marrow infiltration
- Associated to myelo-lymphoproliferative disorders:
  - Acute myeloid leukemia
  - Acute lymphoblastic leukemia
  - Chronic myeloid leukemia
  - Juvenile myelo monocytic leukemia
  - Lymphomas
  - Chronic lymphoblastic leukemia
  - LGL syndrome
- Associated to hypersplenism (± anemia, ±thrombocytopenia)
- Associated to sequestration in infectious foci

followed by start of treatment. The same was considered appe
priate also for patients with moderate neutropenia (EO, 9, A).

In case of a personal history of assumption of medicatio
known to be associated with neutropenia or in case of recurren
of neutropenia after drug exposure and its regression after dr
withdrawal [11,12] a diagnosis of drug-related neutropenia look
appropriate (V, EO, 8.1, B). The list of drugs associated wi
neutropenia reported in Table V was considered by the pan
exhaustive (V, EO, 9, A).

In Blacks of South African extraction [13] (II), in Americ
Mexicans [14] (I), in Afro Caribbean [15] (I), in Yemenite Jew
ancestries and in some Arabic ethnicity [13–16] (II) an AN
between 0.5 and 1.0×10^9/L mainly if not associated to infe
tions and found also in the parents, was considered to allow t
diagnosis of the Ethnic neutropenia which is considered a va
ation from the normal [17] (V, EO, 8.7, B).

After confirmation of the neutropenia, the experts consider
appropriate to perform the panel of first line investigations
Table VI (EO, 8.2, B), Figure 1. This panel was intended as
package aiming to confirm/exclude the commonest causes
neutropenia and to direct further diagnostic steps in case a fir
diagnosis was not achieved. If history, clinical findings and fir
level investigations suggested a form associated to other path
logical conditions (Table III), the panel reckoned appropriate
proceed to further more targeted analyses as indicated by patien
history and clinical-laboratory data (Fig. 2) (EO, 8.2, B).

If neutropenia was found to be associated to bone abnorma
ities of the chest and upper and lower limbs, hepatomegaly, dia
rhea, anemia and/or thrombocytopenia and to consistent
modified first level investigations (electrolytes changes and met
bolic acidosis) diseases like Shwachman–Diamond, Pearson
syndrome and even Blackfan–Diamond syndrome should ha
been taken in account (Table III). In these cases the panel rec
oned appropriate to proceed to further more targeted analys
(genomic DNA mutation study for Shwachman–Diamond, mit
chondrial DNA analysis for Pearson's syndrome, erythrocy
ADA and mutation search for Blackfan–Diamond syndrom
[18–21] aiming to make a firm diagnosis of these diseas
(Table VII) (EO, 8.2, B). In the case of signs of nutriti
deficiency and consistent first level investigation (low IgG seru
level and altered liver function tests), measurement of seru
levels of Vit. B12, transcobalamin, folate, and copper was reco
mended [17] (EO, 8.4, A).

In the case of early, severe and recurrent infections associat

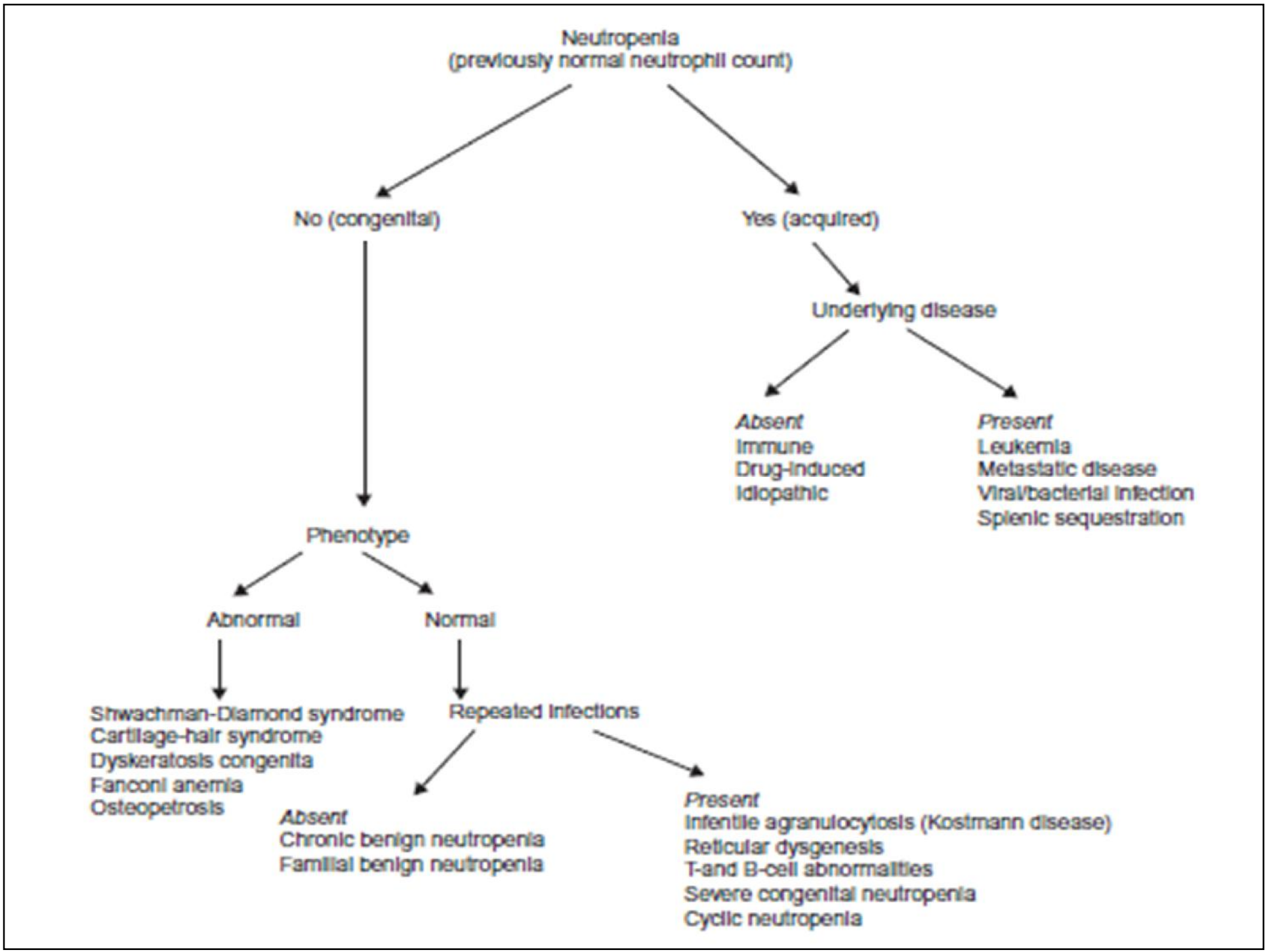
Neutropenia
(previously normal neutrophil count)
No (congenital)
Yes (acquired)
Underlying disease
Absent
Immune
Drug-induced
Idiopathic
Present
Leukemia
Metastatic disease
Viral/bacterial infection
Splenic sequestration
Phenotype
Abnormal
Normal
Shwachman-Diamond syndrome
Cartilage-hair syndrome
Dyskeratosis congenita
Fanconi anemia
Osteopetrosis
Repeated infections
Absent
Chronic benign neutropenia
Familial benign neutropenia
Present
Infantile agranulocytosis (Kostmann disease)
Reticular dysgenesis
T-and B-cell abnormalities
Severe congenital neutropenia
Cyclic neutropenia

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TABLE IV. Initial Evaluation for Patients WHO Have Neutropenia (EO, 8.7, A)

Family history

Ascertain ethnic origin, occurrence of other neutropenia cases, consanguinity

Personal history

Ask for occurrence of viral or bacterial infections and drug assumption during pregnancy and neonatal period

Investigate number, type, site, and recurrence of infections. Ask specifically for occurrence of gingivitis, periodontitis, skin infections, abscesses, otomastoiditis, and pneumonias and for type, administration way, duration of treatment and response to antibiotics

Drug history

Ask for type and duration of drug assumption, particularly those indicated as to be associated with occurrence of neutropenia (Table V)

Physical examination

Focus on weight, stature, psychomotor development, somatic dysmorphisms, signs of infections (skin, mouth), hearth function, liver, and spleen size, presence of enlarged lymphonodes, joints, neurological symptoms, symptoms compatible with autoimmune, metabolic, gastrointestinal, nutritional diseases

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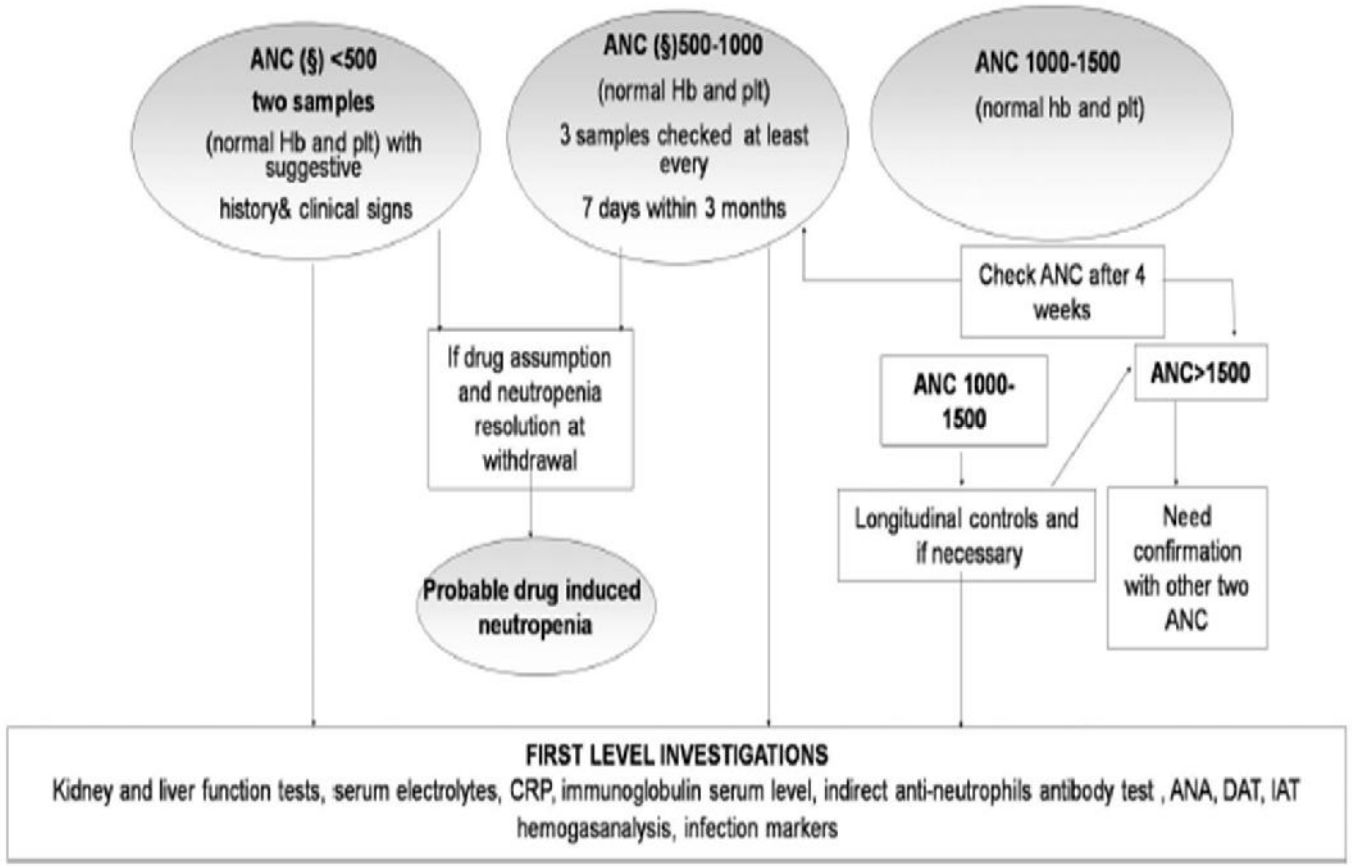

TABLE VI. First Level Investigations (EO, 8.2, B)

Kidney and liver function tests
Serum electrolytes
Venous blood pH
C-reactive protein (CRP)
Immunoglobulin serum level
Indirect anti-neutrophil antibodies (4 tests over 4–6 months) by flow cytometry analysis
Viral (serology or DNA/RNA) and bacterial investigations
Direct and indirect antiglobulin test
Antibodies against nucleus (ANA) test

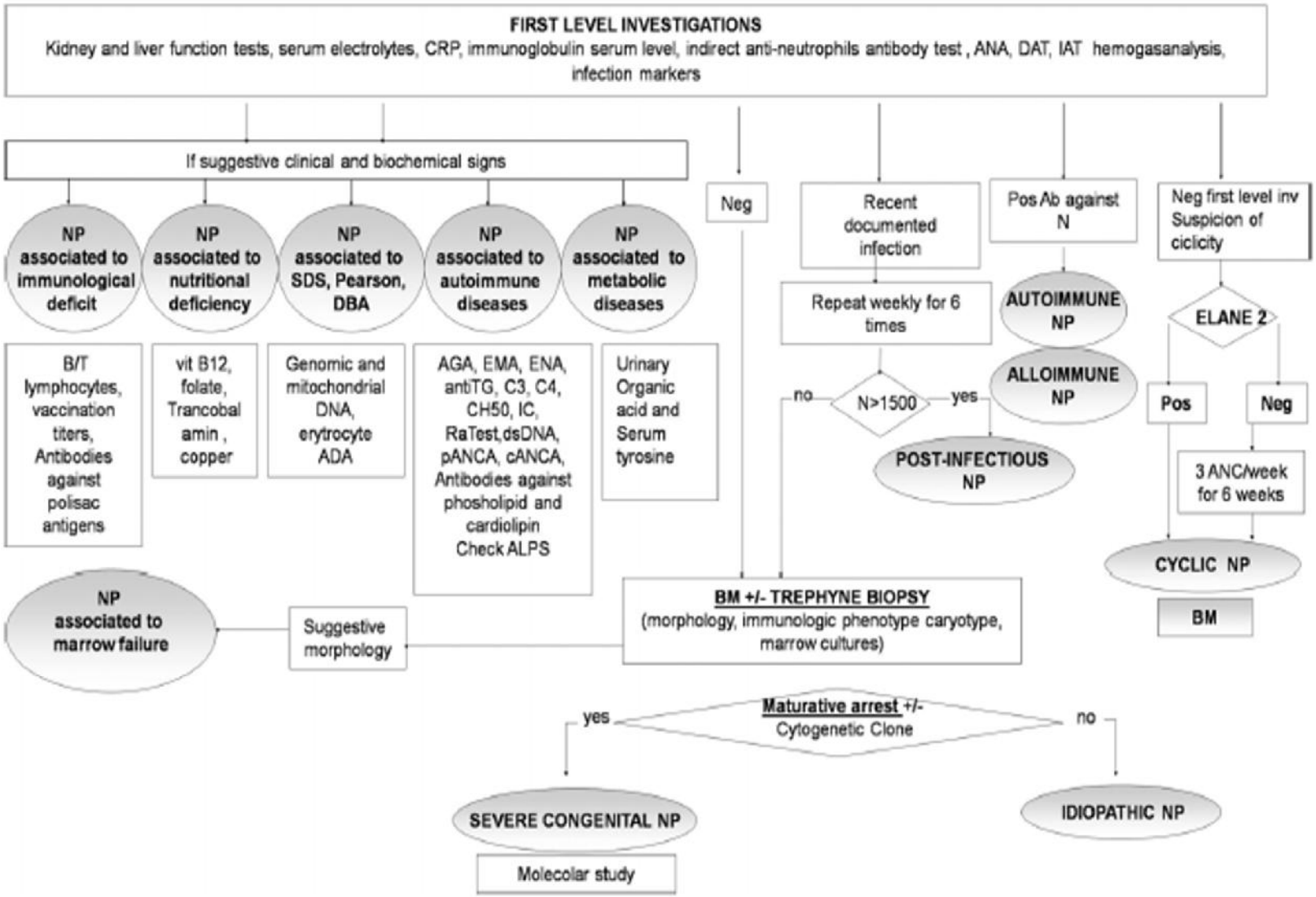

Fig. 2. Advanced level investigations. BM, bone marrow; ADA, serum adenosine deaminase; SDS, Shwachman–Diamond syndrome; DBA, Diamond–Blackfan anemia; AGA, antiglobulin antibodies; EMA, antiendomysium antibodies; ENA, antibodies against extractable nuclear antigen; antiTG, antibodies against thyroglobulin; ALPS, autoimmune lymphoproliferative syndrome.

1. History of drug ingestion, toxin exposure, infectious history
2. Physical examination—nature of infectious lesions, growth and development, presence of anomalies, presence of enlarged lymph nodes or hepatosplenomegaly
3. Familial: absolute granulocyte count in family members
4. Blood count: CBC with differential and platelet count, absolute granulocyte count and reticulocyte count; CBC and differential three times per week for 6–8 weeks (to exclude cyclic neutropenia)
5. Bone marrow
   a. Maturation characteristics of myeloid series; ? reduction in mature granulocytes
   b. Maturation and number of megakaryocytes and erythroid precursors
   c. Karyotype (to identify myelodysplasia or acute myelocytic leukemia) and FISH studies for chromosome 7 and 5q
   d. Electron microscopy (subcellular morphology, congenital dysgranulopoiesis)
6. Detection of antineutrophil antibodies (see text for details)
   a. Granulocyte immunofluorescence test (GIFT)
   b. Granulocyte indirect immunofluorescence test (GIIFT)
   c. Granulocyte agglutination test (GAT)
   d. Enzyme linked immunoassay (ELISA)
   e. Monoclonal antibody specific immobization of granulocyte antigens (MAIGA)

7. Immunologic tests:
   a. Immune globulins (IgA, IgG, IgM, IgE)
   b. Cellular immunity (skin-test activity, purified protein derivative (PPD), lymphocyte subsets; suppressor T-cell assay)
   c. Antinuclear antibodies, C_3, C_4, CH_{50}
8. Evidence of metabolic disease
   a. Plasma and urine aminoacid screening
   b. Serum vitamin B_{12}, folic acid and copper
9. Evidence of pancreatic disease
   a. Exocrine pancreatic function: stool fat, pancreatic enzyme assays, CT scan of pancreas for pancreatic lipomatosis, serum levels of trypsinogen and isoamylase
10. Chromosomal breakage analysis (Fanconi anemia)
11. Radiographic bone survey (cartilage-hair hypoplasia, Shwachman-Diamond syndrome, Fanconi anemia)
12. Serum muramidase (ineffective myelopoiesis)

13. Flow cytometry for CD59 (or other GPI linked protein) (paroxysmal nocturnal hemoglobinuria) This study is much more specific and reliable than the Sucrose hemolysis test or HAM test that had been used in the past to make this diagnosis
14. Bone density studies (14% of patients with chronic neutropenia show nonclinical osteoporosis or osteopenia)
15. Many gene mutation analyses are commercially available including: Neutrophil elastase (ELA-2) (SCN and cyclic neutropenia), GFI-1 (SCN), WAS (X-linked neutropenia), SBDS (Shwachman-Diamond), HAX 1, TAZ (Barth syndrome), Fanconi family of genes, LYST (Chediak Higashi syndrome) and others that are continually being discovered. Molecular diagnostic studies have made diagnosing many of these entities more accurate. In the past, physicians had to rely on interpretation of colony-forming unit (CFU) assays and colony-stimulating activity (CSA) assays to try to distinguish between these different entities

Other investigations that are rarely used today, but may prove useful in making a diagnosis in a particular patient are listed below:

1. Estimate of marginating granulocyte reserve pool
   Epinephrine stimulation tests (0.1 ml 1:1,000 epinephrine SC)
   (1) Absolute granulocyte counts at 5, 10, 15 and 30 minutes
   (2) Normal: double base count
2. Estimate of bone marrow granulocyte reserve pool
   a. Cortisone stimulation tests (5 mg/kg IV)
      (1) Absolute granulocyte counts hourly for 6 hours
      (2) Normal: Increase of more than 2,000 neutrophils/mm^3
   b. Typhoid stimulation tests (0.5-ml vaccine SC)
      (1) Absolute granulocyte count at 3, 6, 12 and 24 hours
      (2) Normal: threefold to fourfold increase
3. Rebuck skin window (to assess leukocyte migration and chemotaxis)
   Normal: at 3 hours, neutrophils; at 6 hours, mixed neutrophils and monocytes; at 24 hours, monocytes

1. Admit to hospital for persistent fever over 101°F and ANC <500 or patient is toxic appearing
2. Obtain appropriate cultures (blood, throat, urine, infected area) and sensitivity
3. Administer parenteral antibiotics (see Chapter 31)
   a. If an organism is isolated, 10–14 days intravenous treatment is required
   b. If no organism is isolated, antibiotic is continued until afebrile and neutropenia is resolved
4. Whenever possible, patient should be in a single-patient room. If not available, the patient sharing the room should be "infection-free"
5. Staff, family members and visitors should observe strict hand-washing procedures. Visitors should be free from colds or other infections
6. Wash skin carefully with a povodine or chlorhexidine-containing solution before all skin puncture procedures
7. Minimize manipulation of skin, oral mucosa, perineum and rectum; rectal temperatures and enemas are contraindicated
8. Treat mouth ulcerations and gingivitis with appropriate systemic antibiotics if secondary bacterial infection is found and 3% hydrogen peroxide–1% alum mouthwash which usually produces symptomatic relief. Use a soft toothbrush for brushing
9. Administer G-CSF[b] for treatment of Kostmann disease, Shwachman-Diamond syndrome, other congenital neutropenias and severe neutropenia following chemotherapy (the starting dose is 5 μg/kg SC with dose modification according to the patient's absolute neutrophil count)

TABLE V. Drugs Implicated in Causing Neutropenia [11] (V, EO, 9, A)

Analgesics and non-steroidal antinfiammatory drugs	Acetaminophen, acetylsalicylic acid, aminopyrine, benoxaprofen, diclofenac, diflunisal, dipyrone, fenoprofen, indomethacin, ibuprofen, naproxen, phenylbutazone, piroxicam, sulindac, tenoxicam, tolmetin
Antipsychotics, hypnosedatives, and antidepressants	Amoxapine, chlomipramine, chlorpromazine, chlordiazepoxide, clozapine, diazepam, fluoxetine, haloperidolo, levopromazine, imipramine, indalpin, meprobamate, mianserin, olanzapine, phenothiazine, risperidone, tiapride, ziprasidone
Antiepileptic drugs	Carbamazepine, ethosuximide, phenitoin, trimethadione, valproate acid (valproate sodium)
Antithiroid drugs	Carbimazole, methimazole, potassium perchlorate, potassium thiocyanate di potassio, propylthiouracil
Cardiovascular drugs	Acetylsalicylic acid, amiodarone, aprindine, bepridil, captopril, coumarins, dipyrisamole, digoxin, flurbiprofen, furosemide, hydralazine, lisinopril, methyldopa, nifedipine, phenidione, procainamide, propafenone, propranonol, quinidina, ramipril, spironlactone, thiazide diuretics, ticlopidine, vesnarinone
Antinfective agents	Abacavir, acyclovir, amodiaquine, atovaquone, cephalosporins, chloramphenicol, chloroguanine, chloroquina, ciprofloxacin, clindamyicin, dapsone, ethambutol, flucytosine, fusidic acid, gentamicin, hydroxychloroquine, isoniazid, levamizole, linezolid, macrolids, mebendazole, mepacrine, metronizadole, minocycline, nitrofurantoin, norfloxacin, novobiocin, penicillins, pyrimethamine, quinine, rifampicin, streptomycin, terbinafine, tetracycline, thioacetazone, tinidazole, cotrimoxazole, vancomycin, zidovudine
Miscellaneous drugs	Acetazolamide, acetylcysteine, allopurinol, aminoglutethimide, arsenic compoounds, benzafibrate, brompheniramine, calcium dobesilate, chlorpheniramine, cimetidine, colchicine, dapsone, deferiprone, famotidine, flutamide, gold, glucocordicoids, hydroxychloroquine, mesalazine, metapyrilene, methazolamide, metoclopramide, levodopa, olanzapine, omeprazole, oral hyoglycemic agents (glibenclamide), mercurial diuretics, penicillamine, ranitidine, riluzole, sulfasalazine, sulfonamides, tamoxifene, thenalidine, tetinoid, tripelennamine