How to evaluate vitamin and iron deficiency in practice?

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B12 & folate deficiency in cancer
Requirements: 2-5 µg/day (meat, fish, egg, milk)
Absorption requires intrinsic factor (IF) secreted by gastric parietal cells for absorption in terminal ileum
Storage: 2-5 mg (liver) → enough for 2-4 yrs
VITAMIN B12
Vitamin B12 : metabolism & deficiency

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- Deficiency
  - Inappropriate diet: malnutrition, vegetarian diet
  - Defective IF
    * Biermer = pernicious anemia (autoimmune gastric atrophy)
    * Gastrectomy
  - Malabsorption: caeliac disease, tropical sprue, Crohn, ileal resection
  - Competition
    * Bacteria: blind loop syndrome (diverticulae or localized inflam.)
    * Fish tapeworm (Diphyllobothrium latum)

→ 3-7% of cancer patients
VITAMIN B12
Vitamin B12 deficiency: diagnosis

**Vitamin B12 in serum**: 200-500 pg/ml

- **Stage 1**: Serum B12 normal to < 200 pg/ml
- **Stage 2 (1-2 yrs)**: Serum B12 < 150 pg/ml → Macrocytosis, hypersegmented neutrophils
- **Stage 3 (> 3 yrs)**: Serum B12 < 100 pg/ml → Megaloblastic anemia, anisopoikilocytosis, N retics → ↑ LDH & bilirubin, ↑ SeFe & Tsat → Thrombocytopenia, leucopenia → Hunter’s glossitis & neuropathy (spinal cord degeneration)
VITAMIN B12
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  → Megaloblastic anemia, anisopoikilocytosis, N retics
  → ↑ LDH & bilirubin, ↑ SeFe & Tsat
  → Thrombocytopenia, leucopenia
  → Hunter’s glossitis & neuropathy (spinal cord degeneration)

**Biermer**
- Gastric parietal cell antibodies: 80-90% but nonspecific
- Intrinsic factor antibodies: 55% but specific
- Abnormal Schilling absorption test corrected by IF
Requirements: 100-200 µg/day (liver, nuts, vegetables)

Absorption in proximal jejunum

Storage: 10-15 mg (RBC) → enough for 3-4 mo
### FOLATE

**Folate : metabolism & deficiency**

- **Requirements**: 100-200 µg/day (liver, nuts, vegetables)
- **Absorption** in proximal jejunum
- **Storage**: 10-15 mg (RBC) → enough for 3-4 mo

### Deficiency
- Poor nutrition: poverty, age, alcoholism
- Malabsorption
  - Caeliac disease, tropical sprue, Crohn
  - Drugs: phenytoin, barbiturates, valproate, OC, nitrofurantoin
- Increased needs: pregnancy, hyperplastic erythropoiesis, dialysis, exfoliative dermatitis
- Altered usage
  - Alcoholism
  - Anti-folate drugs: MTX, trimetoprim, pyrimethamin, pentamidine

→ 0.5-1% of cancer patients
FOLATE
Folate deficiency : diagnosis

<table>
<thead>
<tr>
<th>Folate in serum</th>
<th>5-18 ng/ml</th>
</tr>
</thead>
<tbody>
<tr>
<td>Folate in RBC</td>
<td>125-600 ng/ml</td>
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</table>

- **Stage 1 (3 wks)**
  → serum folate ↓

- **Stage 2 (7 wks)**
  → Hypersegmented neutrophils

- **Stage 3 (16 wks)**
  → RBC folate ↓
  → Macrocytosis

- **Stage 4 (20 wks)** : as B12 deficiency without glossitis/neuropathy
  → Megaloblastic anemia, anisopoikilocytosis, N retics
  → ↑ LDH & bilirubin, ↑ SeFe & Tsat
  → Thrombocytopenia, leucopenia
B12 OR FOLATE DEFICIENCY

Treatment

- **Vitamin B12** (give also 5 mg/d folate)
  - IM: hydroxocobalamin > cyanocobalamin
    * 1 mg / d x 5, 1 mg / wk x 5, 1 mg / mo if necessary (Biermer…)
  - PO (if mild deficiency): 0.5-1 mg/d apart from meals

- **Folate** (not without verifying that B12 is normal)
  - PO: 10-20 mg/d x 2 mo + 5 mg/d thereafter if necessary

- **Rest, O₂**: if severe anemia
- **Monitor K**: possible hypokalemia
- **Avoid transfusions**: worsened leucopenia/thrombocytopenia
- **Treat underlying cause**
Iron deficiency in cancer
IRON METABOLISM

Internal iron exchanges

- Erythroid marrow: 25 mg
- Circulating RBC
- Liver: 3 mg → 3 mg
- Macrophages (RE cells): 25 mg
- Plasma: Fe
- Intestinal absorption: 1 mg

Iron turnover in mg per day;
1 mg iron is lost per day
IRON METABOLISM

Regulation by hepcidin

Hypoxia
Erythropoiesis
Iron deficiency

Liver

Inflammation
Iron overload

Nicolas, Blood Cells Mol Dis 29:327, 2002
IRON DEFICIENCY
Absolute vs functional

**Absolute iron deficiency**
- no iron stores: ferritin < 20 µg/L in N individual
- < 100 µg/L in infl/cancer patient

**Functional iron deficiency**
- iron stores present but ID in erythroid bone marrow
  a) Iron sequestration in macrophages
  - Inflammation (ACD, anemia of chronic disease)
  b) Increased iron requirements
  - EPO therapy
FUNCTIONAL IRON DEFICIENCY
ACD (cancer, inflammation)

• ID erythropoiesis despite normal-increased iron stores
  – Impaired iron absorption (1 mg/d)
  – Inhibition of storage iron release (25 mg/d)
• Mediator = hepcidin
• 40% of cancer patients
FUNCTIONAL IRON DEFICIENCY

ACD (cancer, inflammation)

- ID erythropoiesis despite normal-increased iron stores
  - Impaired iron absorption (1 mg/d)
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FID

- Hypochromic microcytic anemia if long duration
- Normochromic normocytic anemia if short duration
- ↓ SeFe & TSat
- N-↑ Ferritin

ASH slide bank
ID in cancer
Diagnostic parameters
IRON PARAMETERS

Storage iron: serum ferritin

- Erythroid marrow
- Circulating RBC
- Liver
- Plasma
- Macrophages (RE cells)
FERRITIN
Serum ferritin

- Represents iron stores (macrophages and hepatocytes) \(1 \mu g/L = 120 \mu g/kg\) storage iron
- Low ferritin < 20 \(\mu g/L\) (12-30 according to assay) 100% specific for iron deficiency
- Normal range varies with age and sex

ASH slide bank
• **Conditions with falsely elevated serum ferritin**
  – Inflammation (including cancer)
    • Lower limit = 100 (40–120) µg/L
    • Lower levels define absolute ID in cancer patients
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FERRITIN
Serum ferritin interpretation

• Conditions with falsely elevated serum ferritin
  – Inflammation (including cancer)
    • Lower limit = 100 (40–120) µg/L
    • Lower levels define absolute ID in cancer patients
  – Some forms of cancer (e.g. neuroblastoma)
  – Renal failure (lower limit 40–100 µg/L)
  – Liver damage
  – Hyperthyroidism
  – Poorly controlled diabetes mellitus (ferritin glycosylation)
  – Hyperferritin-cataract syndrome
  – Benign hyperferritinemia
FERRITIN
Serum ferritin interpretation

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Absolute ID = no iron stores

ferritin < 20 µg/L in normal individual
< 100 µg/L in cancer patient
IRON PARAMETERS

Plasma iron : transferrin saturation

- Circulating RBC
- Erythroid marrow
- Liver
- Macrophages (RE cells)
- Plasma
TRANSFERRIN SATURATION

Normal

SeFe  8-30 µmol/L
Tsat  20-45 %

Red blood cells
Senescent RBCs taken-up by macrophages

Macrophages
Recycling and storage of iron from red blood cells

Plasma Transferrin
Dynamic equilibrium of transferrin saturation

Marrow
Sufficient iron available for erythropoiesis
TRANSFERRIN SATURATION

Normal

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Red blood cells
Senescent RBCs taken-up by macrophages

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Sufficient iron available for erythropoiesis
TRANSFERRIN SATURATION
(Functional) iron deficiency

Absolute ID

Empty iron stores

Red blood cells	Macrophages	Plasma transferrin	Marrow
TRANSFERRIN SATURATION
(Functional) iron deficiency

Absolute ID

Functional ID
(Inflammation/cancer)

Empty iron stores

Blocked iron release

Red blood cells  Macrophages  Plasma transferrin  Marrow
TRANSFERRIN SATURATION
(Functional) iron deficiency

Absolute ID

Functional ID (Inflammation/cancer)

Functional ID (EPO therapy)

Empty iron stores

Blocked iron release

Iron need exceeds delivery

TSat < 20%
%HYPO > 5%
CHr < 28 pg

Red blood cells
Macrophages
Plasma transferrin
Marrow
IRON PARAMETERS
Erythroid marrow: sTfR

Erythroid marrow → Circulating RBC → Liver → Plasma → Macrophages (RE cells)
SOLUBLE TRANSFERRIN RECEPTOR

ACD vs. ID vs. combined ACD+ID

sTfR 3-7 µg/l

Normal

Infl (ACD)  ID  ACD + ID

sTfR (mg/L)
IRON PARAMETERS

RBC iron: % HYPO and CHr
Hypochromic erythrocytes

- Normal: < 5%
- ID erythropoiesis: > 5–10%

→ long term indicator of ID erythropoiesis
(RBC lifespan: 120 d)

% Hypo 24.4%

Brugnara, Blood 83:3100, 1994
**Hypochromic erythrocytes**
- Normal: < 5%
- ID erythropoiesis: > 5–10%
  → long term indicator of ID erythropoiesis (RBC lifespan: 120 d)

**Hb content of reticulocytes (CHr)**
- Normal: 28-32 pg
- ID erythropoiesis: < 28 pg
  → short term indicator of ID erythropoiesis (Retic lifespan: 4 d)

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Brugnara, *Blood* 83:3100, 1994
Hypochromic erythrocytes
- Normal: < 5%
- ID erythropoiesis: > 5–10% → long term indicator of ID erythropoiesis (RBC lifespan: 120 d)

Hb content of reticulocytes (CHr)
- Normal: 28-32 pg
- ID erythropoiesis: < 28 pg → short term indicator of ID erythropoiesis (Retic lifespan: 4 d)
IRON METABOLISM

Hepcidin

- Erythroid marrow
- Circulating RBC
- Plasma
- Liver
- Intestinal absorption
- Macrophages (RE cells)
HEPCIDIN

Serum hepcidin

NL = normal, CRP = inflammation, MM = multiple myeloma, ACKD & PCKD = adult & pediatric CKD (not dialyzed), IDA = ID anemia,

Ganz et al. Blood 2008;112:4292
IRON DEFICIENCY

Cancer: absolute vs functional ID

Low Tsat

Absolute ID
Low

Ferritin

Functional ID
N-High
IRON DEFICIENCY

Cancer: absolute vs functional ID

Absolute ID
- Low
- Ferritin
- Low Tsat
- Depleted body iron stores
  - Low serum ferritin (< 100 ng/ml)
  - TSAT < 20%

Functional ID
- N-High
IRON DEFICIENCY
Cancer: absolute vs functional ID

Absolute ID
- Low serum ferritin (< 100 ng/ml)
- TSAT < 20%

Functional ID
- Depleted body iron stores
- Normal or high ferritin (> 100 ng/ml)
- TSAT < 20%

Functional iron deficiency
- Inadequate iron supply to meet demand despite normal or abundant iron stores
- TSAT < 20%
IRON DEFICIENCY IN CANCER

ID in cancer

Absolute ID (no iron stores)
Ferritin < 100 ng/mL
Low hepcidin

Functional ID (iron stores +/-++)
Ferritin ≥ 100 ng/mL
High hepcidin

TSAT <20%

At initial diagnosis (40%)
Monthly during FU

Microcytic hypochromic A

IV iron

Microcytic hypochromic A (chronic)
Normocytic normochromic A (acute)

ESA + IV iron

IDA

ACD
FUNCTIONAL ID

Overcoming hepcidin block with IV iron

Hentze et al, Cell 142:24, 2010
Delaby et al, Blood 106: 3979, 2005
Beaumont, Haematologica 95:1233, 2010
FUNCTIONAL ID

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Hentze et al, Cell 142:24, 2010
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FUNCTIONAL ID

Overcoming hepcidin block with IV iron

IV iron → ↑ Hepcidin → RBC

Ferroportin

Tf

Hentze et al, Cell 142:24, 2010
Delaby et al, Blood 106: 3979, 2005
Beaumont, Haematologica 95:1233, 2010
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Conclusions
<table>
<thead>
<tr>
<th>Anemic cancer patient</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Step 1</strong></td>
</tr>
<tr>
<td><strong>Step 2</strong></td>
</tr>
<tr>
<td><strong>W&amp;W</strong></td>
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<td><strong>W&amp;W</strong></td>
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<tr>
<td><strong>W&amp;W</strong></td>
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<tr>
<td><strong>Iron</strong></td>
</tr>
<tr>
<td><strong>Folate</strong></td>
</tr>
<tr>
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</tr>
<tr>
<td><strong>B12</strong></td>
</tr>
</tbody>
</table>
Anemic cancer patient

Step 1
Absolute hematonic deficiency?

- Folate: Serum folate < 5 ng/ml (1% cancer patients)
  - Folate (PO)

- Iron: Serum ferritin < 100 ng/ml (15% cancer patients)
  - Iron (IV)

- B12: Serum B12 < 200 pg/ml (5% cancer patients)
  - B12 (PO / IV)
Anemic cancer patient

Step 1
Absolute hematinic deficiency?

Folate
↓ Serum folate < 5 ng/ml
1% cancer patients

Folate (PO)

Step 2
ESA therapy (EORTC guidelines)?

No

Iron
↓ Serum ferritin < 100 ng/ml
15% cancer patients

Iron (IV)

Yes

B12
↓ Serum B12 < 200 pg/ml
5% cancer patients

B12 (PO / IV)

ESA (SC)

W&W

No

Yes
Anemic cancer patient

Step 1
Absolute hematinic deficiency?

Folate
↓ Serum folate < 5 ng/ml
1% cancer patients

Iron
↓ Serum ferritin < 100 ng/ml
15% cancer patients

B12
↓ Serum B12 < 200 pg/ml
5% cancer patients

Folate (PO)

Iron (IV)

B12 (PO / IV)

Step 2
ESA therapy (EORTC guidelines)?

No
W&W

Yes
ESA (SC)

Iron (IV)

Step 3: functional ID?
TSat < 20% (or HYPO < 5% or CHr < 28 pg)
Before & during treatment

No
W&W

Yes
Iron (IV)
THANK YOU !