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ZAPORIZHZHIA STATE MEDICAL UNIVERSITY

Biological Chemistry Department

Biochemistry of vitamins

*a manual for «Biological chemistry» discipline
for teachers*

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and it is recommended for the use in educational process for foreign students.*

Reviewers:

Prihodko O. B., Head of Department of Medical Biology, Parasitology and Genetics. Dr. Hab, assoc. professor

Belenichev I. F., Head of Department of Pharmacology and Medicinal Preparations, Dr. Hab., professor

Authors:

Aleksandrova K. V.

Rudko N. P.

Aleksandrova K. V.

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Manual was made in accordance with the program of biological chemistry for training of students of higher educational institutions of III-IV levels of accreditation for specialty «Medicine».

This manual is recommended to use for teachers that works with students of International Department (the second year of study).

Александрова К. В.

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LESSON ON TOPIC: “THE ROLE OF WATER-SOLUBLE VITAMINS IN THE METABOLISM OF HUMANS. VITAMIN SIMILAR SUBSTANCES”

1. RELEVANCE OF TOPIC

Vitamins are essential dietary factors that enter the body in small amounts, but have strongly pronounced effect on metabolism and body functions as a whole. Most water-soluble vitamins used in humans as coenzymes in biochemical reactions and therefore their deficiency leads to the development of diseases.

Nowadays vitamins are widely used for prevention and treatment of pathological conditions that determines the importance of studying the subject for students of medical university.

2. LEARNING OBJECTIVES FOR LESSON

To learn theoretical material about classification, biochemical properties and mechanism of action of water-soluble vitamins and vitamin similar substances:

1. Consolidate knowledge of theoretical material on "Water-soluble vitamins and vitamin-like substance," paying special attention to chemical structure, properties and biological role of these compounds.

2. To acquaint students with qualitative reactions to vitamins C and B₁.

3. To acquaint students with the quantitative determination of vitamin C.

To offer students:

a) determine the quantitative value of vitamin C in various foods (potatoes, cabbage, apple) and compare the results;

b) to carry out quantitative determination of vitamin C in the urine.

Necessary to know:

1. Vitamins: classification, common notions about their use in humans. Vitamin-like substances. Antivitamins. Hypervitaminosis, hypovitaminosis and avitaminosis. The reasons of vitamins deficiency. Vitamins occurrence in Nature. The interaction of vitamins in an organism.

2. Thiamine (B_1). Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
3. Riboflavin (B_2). Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
4. Pyridoxine (B_6). Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
5. Cobalamin (B_{12}). Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
6. Pantothenic acid (B_5). Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
7. Folic acid (B_9). Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
8. Biotin. Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
9. Vitamin C. Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency and
10. Vitamin PP, B_3 (nicotinic acid, nicotine amide). Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
11. Vitamin P (Rutin). Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
12. Vitamin-similar substances (Lipoic acid, Carnitine, CoQ) and antivitamin: their functions in humans.

Necessary to be able to do:

1. Quantitative determination of ascorbic acid.
2. Qualitative reactions for vitamins C and B_1 .

3. A FLOW CHART OF THE PRACTICAL TRAINING

Stage	Time (min)	Educational materials		Location
		Content of the plan point	Equipment	
1. Organizational moment	5 min	Check the presence of students	Register	Classroom
2. Colloquium on the outstanding questions at home or essential for the explanation of the practical part of classes	15 min		Methodical recommendations for practical training	Classroom
2. Control of incoming knowledge	15 min	Variants of test tasks	Printed materials	Classroom
3. Students' individual work with the table "Characteristics of water-soluble vitamins and their functions" and discussion of the results	20 min	Textbooks Methodical recommendations for practical training	Empty table	Classroom
4. Carrying out of laboratory works on practical training theme: To divide student group on 3 subgroups: Subgroup №1: Carry out the determination of ascorbic acid concentration in vegetables (a potato, cabbage). Subgroup №2: Carry out the determination of ascorbic acid content in the urine. Subgroup №3: Carry out the qualitative reactions for vitamins C and B ₁ . Each subgroup makes its laboratory task, but records results of all tasks.	30 min	Biochemistry laboratory manual. Module 2. Conjugated protein metabolism. Molecular biology. The biochemistry of cell-to-cell interrelations	Equipment and reagent for laboratory work	Classroom, biochemical laboratory
5. Discussion of results of laboratory works and their explanation	10 min	Oral consideration		Classroom
6. Control of the final level of knowledge	15 min	MCQs	Printed materials	Classroom
7. Discussion of student's work during practical training	15 min	Oral questioning of students about results of laboratory works and topic of the lesson	A list of references to help students to eliminate the disadvantages of studying topics	Classroom
8. Rating of all types of students works during the practical training and motivation for the next practical training	10 min	Check and sign of protocols, recommendations for next lesson topic and tasks for independent work		Classroom

4. LEARNING CONTENT

4.1. Organizing time

To acquaint students with the goals and plan of the practical lesson. To note the value of studying topics, widely used to treat by various vitamins. Check readiness of students for practical classes (presence of "Biochemistry laboratory manual Module 2. Conjugated protein metabolism. Molecular biology. The biochemistry of cell-to-cell interrelations. For independent work at home and in class, preparation for licensing examination "Krok 1" for students of second year study of international faculty" with marked MCQs.

4.2. Interview with issues that have not been cleared up at home. In an interview with the students:

1. Find incomprehensible to students' questions and be sure to consider:
 - 1) the concept and definition of the class "Vitamins";
 - 2) classification of vitamins, general description of water-soluble vitamins;
 - 3) the structure, properties and biological role in the mechanisms of metabolism of vitamin C;
 - 4) structure, coenzyme form, its properties, biological role in the mechanisms of metabolism of vitamin B₁;
 - 5) clinical and diagnostic value of determination of vitamin C in urine as well as the practical part of classes linked to qualitative determination of vitamins C, B₁ and quantitative determination of vitamin C.
2. Mark the importance of understanding the mechanisms of participation of vitamins in metabolic processes:
 - knowledge of the active (coenzyme) form of water-soluble vitamins;
 - types of reactions catalyzed by enzymes, which are composed of certain coenzymes;
 - processes that contain these reactions to explain the causes of pathological conditions at hypo- and avitaminosis.
3. Pay attention to the multi-use of vitamin C in humans, vitamin C is involved in redox processes in the synthesis of steroid hormones, bile acids,

catecholamines, as a cofactor hydroxylase enzymes that convert proline into hydroxyproline, promotes the absorption of iron, activating pepsinogen . Lack of vitamin C leads to disruption of these processes.

- 4.3. Check the entry-level knowledge of the biochemistry of water-soluble vitamins and vitamin-like substance. Teachers should check the elementary students' knowledge on the structure, properties, daily needs, sources, symptoms of deficiency of vitamins and vitamin-like substance. In order to clarify compliance with the initial level of knowledge required, students are offered to perform the proposed tasks:

Variant 1

Give the variation of vitamin number with letters of its derivative and of its name of enzyme/process or function

Vitamin	Derivative from vitamin	Name of enzyme/ process or function
1. B ₁	A. NAD ⁺	α. Acetyl-CoA carboxylase
2. B ₂	B. CoASH	β. NADH-dehydrogenase from respiratory chain
3. B ₃	C. Carboxybiotin	γ. Activation of High Fatty Acids
4. B ₅	D. FMN	δ. Hydroxylation of proline residues in collagen
5. B ₆	E. Pyridoxal phosphate	ε. Synthesis of nucleotides
6. B ₉	F. TPP	ζ. Alanine aminotransferase
7. B ₁₂	G. CoQH ₂	η. Methylmalonic acid conversion to succinyl-CoA
8. C	H. Tetrahydrofolate derivative	θ. Isocitrate dehydrogenase of Krebs cycle
9. H	I. Deoxyadenosylcobalamin	ι. Transketolase of Pentose phosphate cycle
10. PP	J. Derivative is absent	κ. The control of calcium ions content in the blood

Variant 2

Vitamin	Derivative from vitamin	Name of enzyme/ process or function
1. B ₉	A. Tetrahydrofolate derivative	α. Antioxidant
2. B ₆	B. Pyridoxal phosphate	β. Methylmalonic acid conversion to succinyl-CoA
3. B ₃	C. CoQH ₂	γ. Glutamate dehydrogenase
4. B ₅	D. TPP	δ. Synthesis of nucleotides
5. B ₂	E. CoASH	ε. Acetyl-CoA carboxylase
6. B ₁	F. FAD	ζ. The control of calcium ions content in the blood
7. PP	G. Carboxybiotin	η. Succinate dehydrogenase
8. H	H. NADP ⁺	θ. Activation of High Fatty Acids
9. C	I. Deoxyadenosylcobalamin	ι. Pyruvate dehydrogenase
10. B ₁₂	J. Derivative is absent	κ. Aspartate aminotransferase

The example of how to get up the answer for task:

1F α; 2 A θ; 3 B η ...and so on.

4.4. Independent work with the table "Characteristics of water-soluble vitamins and their functions".

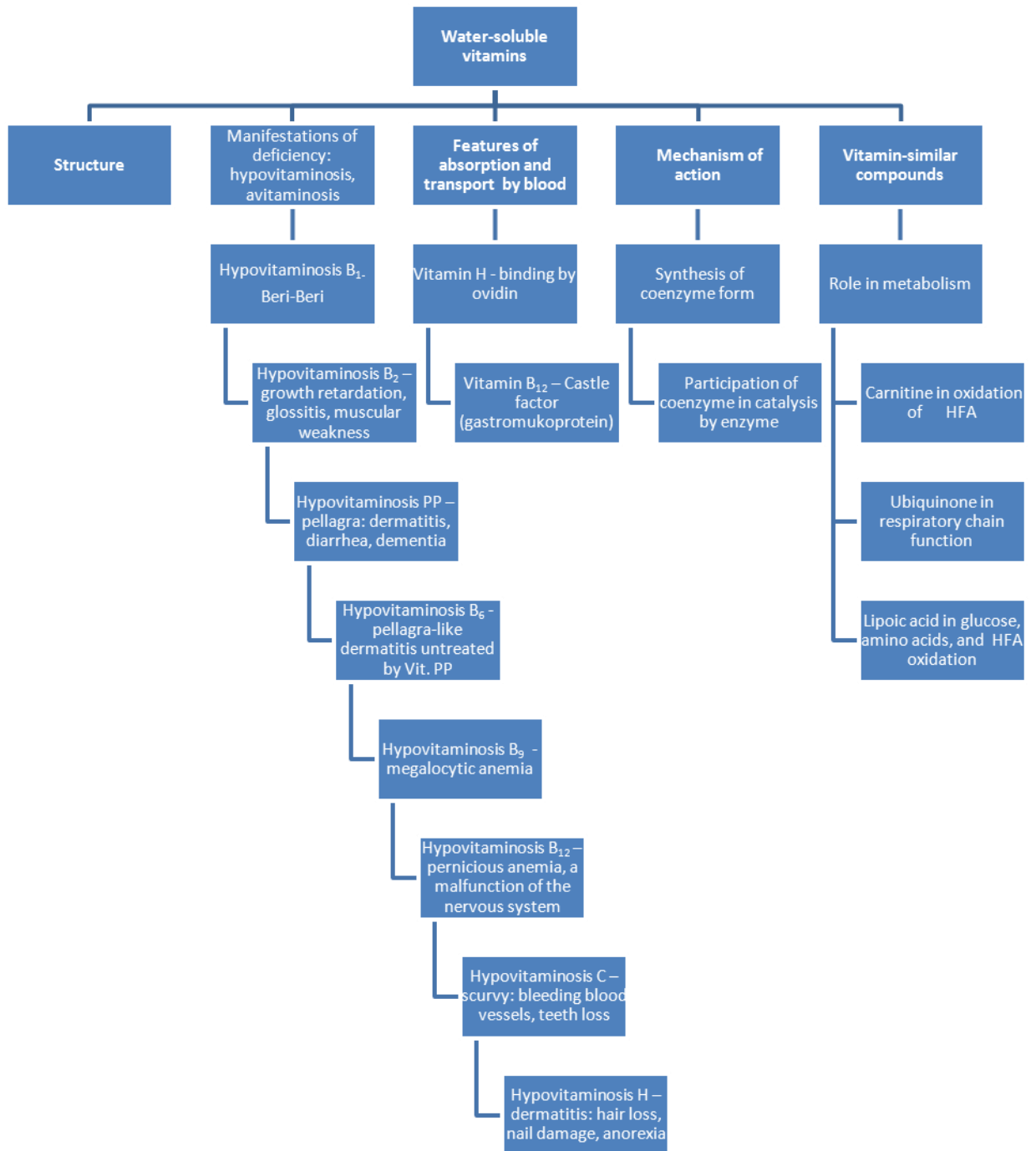
Student should pay special attention to the biochemical aspects of water-soluble vitamins participating in metabolic processes. Students are encouraged to fill in the table:

Table 1. Vitamin functions and manifestations of hypo- and avitaminoses

Vitamin	Functions	Hypovitaminosis symptomes
B ₁		
B ₂		
B ₃ (PP)		
B ₅		
B ₆		
B ₇ (H)		
B ₉		
B ₁₂		
C		

After the self-study students report the results to prove to the logical end of table. The teacher summarizes students' independent work. At the interview the teacher should monitor the use of all concepts and terms that occur in a graph of logical structure of theme:

4.4 GRAPH OF LOGICAL STRUCTURE OF THEME



4.5. Laboratory work performance.

The students begin to perform the laboratory works:

- 1) "Quantitative determination of vitamin C in plant foods: potatoes, apples, cabbage";
- 2) "Quantitative determination of vitamin C in the urine";
- 3) "Qualitative reactions to vitamin C and B₁";

Group is divided into three subgroups. The first subgroup performs laboratory work number 1 (vitamin C in potatoes) and number 2; second - laboratory work number 1 (vitamin C apple) and the number 3; third - laboratory work number 1 (vitamin C in cabbage) and the number 3.

Results of determination of indicators are recorded into the protocols of each student in each work and self-made conclusions on the results are recorded too.

4.5. LABORATORY WORK ALGORITHM

1. Quantitative determination of ascorbic acid.

THE PRINCIPLE OF THE METHOD:

The quantitative determination of vitamin C is based on its capacity to restore 2,6-dichlorophenolindophenole (DCIP). While vitamin C is in titrated solution, the poured DCIP will become colourless due to the formation of the restored form. As soon as all the quantity of vitamin C being in the solution is oxidized the titrated solution becomes pink due to the formation of the DCIP restorative form.

1.1. The determination of ascorbic acid concentration in vegetables (a potato, cabbage).

THE COURSE OF THE WORK:

Crush 5 g of potato (cabbage or other product) with a scalpel and pound it in a mortar, add 3 drops of 10 % hydrochloric acid solution and gradually 15 ml of distilled water. The mass received pour into a flask for titration. Titrate by 0.001 N DCIP solution up to the appearance of pink coloring, which will not disappear within 30 sec.

The calculation will be carried out according to the formula:

$$0.088 \cdot A \cdot 100$$

X = ----- mg %, where

5

X - the content of vitamin C, mg %;

0.088 - the equivalent of ascorbic acid, which is titrated by 0,001N DCIP solution;

A - the quantity of DCIP (ml), used for titration;

100 - recalculation at 100 g of the product;

5 - quantity (g) of the product taken for the analysis.

Compare the received results with the content of vitamin C in foodstuff: in potatoes (6-20 mg %), cabbage (20-50 mg %), apples (20-40 mg%), lemons (40-55 mg%), needles (150-250 mg %), onions (30 mg %), parsley (150 mg %), cauliflower (70 mg %).

RESULTS:

1.2. Determination of ascorbic acid content in the urine.

THE COURSE OF THE WORK:

Pour 10 ml of the urine into a flask for titration and add 10 ml of distilled water, then add 20 drops of 10 % hydrochloric acid. Titrate from the tube 0.001N of DCIP solution up to permanent pink coloring. Calculate daily excretion of vitamin C according to the formula:

$$X = \frac{0.088 \cdot A \cdot B}{C}, \text{ where}$$

X - daily excretion of vitamin C, in mg;

0.088 -the equivalent of an ascorbic acid, which is titrated with 1 ml 0.001 N DCIP solution

A – the volume of the indicator spent for titration;

B - daily average volume of the urine: men - 1500ml, women -1200 ml;

C - urine volume taken for titration.

RESULTS:

The significance of vitamin C determination in the blood plasma and urine:

In norm the content of vitamin C in the urine is 20-30 mg / daily.

It is very important to define this index during the stage: the latent form of vitamin C Hypovitaminosis. The patient drinks the ascorbic acid - glucose solution, containing a daily norm of vitamin C (correlated with the patient's age). In 2-3 hours later the vitamin C concentration is determined in the patient's urine. If the result correlates with normal value, you can say about the latent form of vitamin C Hypovitaminosis.

2. Qualitative reactions for vitamins C and B₁.

2.1. Qualitative reaction for vitamin C.

THE PRINCIPLE OF THE METHOD:

The qualitative revealing of vitamin C is based on its capacity to restore $K_3[Fe(CN)_6]$ and the methylenic blue.

THE COURSE OF THE WORK:

a) Add 1 drop of 10 % NaOH solution into 5 drops of 1% vitamin C solution, some grains of $K_3[Fe(CN)_6]$ and mix. Then add 3 drops of 10 % hydrochloric acid solution and 1 drop of 1 % $FeCl_3$ solution. The residue $Fe_4[Fe(CN)_6]_3$ drops out. For the control test: repeat the course of the work, using distilled water instead of vitamin C.

RESULTS:

b) Add 1 ml of methylenic blue into 1 ml of 1 % ascorbic acid solution. Put the test tube in a boiling water bath. With time you can see the decolorizing of the mixture.

RESULTS:

2.2. Qualitative reaction for vitamin B₁.

THE PRINCIPLE OF THE METHOD:

Thiamine is oxidized by action of K₃[Fe(CN)₆] solution to thiochrome. There is a fluorescence of thiochrome solution under UV-rays.

THE COURSE OF THE WORK:

Pour into a test tube: 1 drop of 5% thiamine solution, 5-10 drops of 10% NaOH solution, 1-2 drops of K₃[Fe(CN)₆] solution and shake. Then use a fluoroscope to see the blue fluorescence of obtained thiochrome solution.

RESULTS:

4.6. Final control of knowledge on the topic.

After summing up the individual work the teacher is necessary to control the final level of students' knowledge respectively theoretical questions for the class:

1. Vitamins and their role in metabolism. Provitamins. Classification of vitamins. The features of absorption for water-soluble vitamins in the gastro-intestinal tract.
2. Vitamins deficiency: exogenous and endogenous reasons of it. Avitaminosis (examples).
3. Water-soluble vitamins (H, B₁, B₂, PP (B₃), B₅, B₆, B₉, B₁₂): structure, sources of reception, daily requirement, biological role.
4. Vitamins C and P: structure, mechanisms of function in humans, daily requirement, and clinical symptoms of their deficiency.
5. Vitamin-similar substances (CoQ, carnitine, lipoic acid): structure and function in humans.

4. 6. TESTS

1. Choose the correct definition of vitamin:
 - A. Essential food proteins
 - B. Food factors that cannot be synthesized in human organism

- C. Essential biologic amines
 - D. Organic compounds, containing amino group
 - E. Essential energy sources
2. Choose the vitamin, whose oxidation results in blue fluorescing product under UV-light:
- A. Pyridoxine
 - B. Rutin
 - C. Thiamine
 - D. Folic acid
 - E. Ascorbic acid
3. Choose the vitamin that contains the isoalloxazine fragment in its structure:
- A. Thiamine
 - B. Riboflavin
 - C. Pyridoxine
 - D. Ubiquinone
 - E. Naphthoquinone
4. Point out the vitamin, whose deficiency leads to pellagra:
- A. Vitamin P
 - B. Vitamin A
 - C. Vitamin C
 - D. Vitamin B₃
 - E. Vitamin B₂
5. Name the metabolic pathway which is in need for vitamins B₁, B₂, B₃, B₅ supplement at the same time:
- A. Pentose Phosphate Cycle
 - B. Glycolysis
 - C. Urea Cycle
 - D. Krebs Cycle
 - E. Glycogenesis

6. Find out the enzyme name whose activity is depended on the presence of vitamin B₂:
- A. Pyruvate carboxylase
 - B. Succinate dehydrogenase
 - C. Malate dehydrogenase
 - D. Isocitrate dehydrogenase
 - E. Citrate synthase
7. The avitaminosis of ascorbic acid is named as:
- A. Cushing`s syndrome
 - B. Addison`s disease
 - C. Kwashiorkor
 - D. Hemolytic anemia
 - E. Scurvy
8. Propose the main food product to promote the intake of vitamin C:
- A. Parsley
 - B. Black currant
 - C. Beef
 - D. Milk
 - E. Butter
9. Find out the vitamin whose deficiency is associated with damaged transamination of amino acids:
- A. Pyridoxine
 - B. Rutin
 - C. Thiamine
 - D. Folic acid
 - E. Ascorbic acid
10. The glycolysis duration is in need for one vitamin, only. Name it:
- A. Pyridoxal phosphate
 - B. Riboflavin
 - C. Thiamine

- D. Nicotinic acid
- E. Ascorbic acid

TESTS FOR PREPARATION TO «KROK-1» EXAMINATION

1. Examination of a patient with frequent hemorrhages from internals and mucous membranes revealed proline and lysine being a part of collagen fibers. What vitamin absence caused disturbance of their hydroxylation?
 - A. Vitamin A
 - B. Thiamine
 - C. Vitamin K
 - D. Vitamin E
 - E. Vitamin C
2. A woman who has been keeping to a clean-rice diet for a long time was diagnosed with polyneuritis (beriberi). What vitamin deficit results in development of this disease?
 - A. Folic acid
 - B. Thiamine
 - C. Ascorbic acid
 - D. Riboflavin
 - E. Pyridoxine
3. Most participants of Magellan expedition to America died from avitaminosis. This disease declared itself by general weakness, subcutaneous hemorrhages, falling of teeth, gingival hemorrhages. What is the name of this avitaminosis?
 - A. Biermer's anemia
 - B. Polyneuritis (beriberi)
 - C. Pellagra
 - D. Rachitis
 - E. Scurvy

4. The structural analogue of vitamin B₂ is administered (acrichine) in a case of enterobiasis. The disorder of which enzyme synthesis is caused by this medicine in microorganisms?
- A. NAD-dependent dehydrogenases
 - B. Cytochrome oxidases
 - C. FAD-dependent dehydrogenases
 - D. Peptidases
 - E. Aminotransferases
5. Pyruvate concentration in the patient's urine has increased 10 times from normal amount. What vitamin deficiency can be the reason of this change:
- A. Vitamin B₆
 - B. Vitamin A
 - C. Vitamin E
 - D. Vitamin C
 - E. Vitamin B₁
6. Hydroxylation of endogenous substrates and xenobiotics requires a donor of protons. Which of the following vitamins can play this role?
- A. Vitamin C
 - B. Vitamin E
 - C. Vitamin P
 - D. Vitamin A
 - E. Vitamin B₆
7. A 10-year-old girl often experiences acute respiratory infections with multiple hemorrhages in the places of clothes friction. Hypovitaminosis of what vitamin is in this girl organism?
- A. A
 - B. B₂
 - C. B₁
 - D. B₆
 - E. C

8. A 9-month-old infant is fed with artificial formulas with unbalanced vitamin B₆ concentration. The infant presents with pellagra dermatitis, convulsions, anaemia. Convulsions development might be caused by the disturbed formation of:
- A. Dopamine
 - B. Histamine
 - C. Serotonin
 - D. DOPA
 - E. GABA
9. In clinical practice tuberculosis is treated with isoniazid preparation – that is an antivitamin able to penetrate into the tuberculosis bacillus. Tuberculostatic effect is induced by the interference with replication processes and oxidation-reduction reactions due to the buildup of pseudo-coenzyme:
- A. FMN
 - B. NAD
 - C. CoQ
 - D. FAD
 - E. TDP
10. Some infection diseases caused by bacteria are treated with sulfanilamides, which block the synthesis of bacteria growth factor. What is the mechanism of their action?
- A. They inhibit the absorption of folic acid
 - B. They are allosteric enzyme inhibitors
 - C. They are allosteric enzymes
 - D. They are anti-vitamins of para-amino benzoic acid
 - E. They are involved in red-ox processes
11. A 20-year old male patient complains of general weakness, rapid fatigability, irritability, decreased performance, bleeding gums, petechiae on the skin. What vitamin deficiency may be caused of these changes?
- A. Riboflavin

- B. Ascorbic acid
- C. Retinol
- D. Thiamine
- E. Folic acid

12. A number of disorders can be diagnosed by evaluation activity of blood transaminases. What vitamin is one of cofactors for these enzymes?

- A. B₆
- B. B₁
- C. B₅
- D. B₂
- E. B₈

4.7. Discussion of practical classes

A teacher sums up all kinds of work for each student at the end of class. Student work is positively evaluated in the case of all types of work are done. The teacher recommends students literature to help eliminate the disadvantages of studying the topic in the case of negative results of two tests.

At the end of class the teacher should be sure to consider the amount of student preparation for the next lesson, emphasize the most important issues for home preparation.

5. INDIVIDUAL INDEPENDENT STUDENT WORK:

It can be recommended for students to prepare essay on the topic:

1. The antioxidant function of vitamins.

6. MATERIAL PROVISION:

Equipment:

1. Test tubes - 3 pcs.
2. Flasks for titration - 4 pieces (250 mL)
3. Pipettes: - 2 pcs. (5 ml); 2 pcs. (1 ml); eye dropper – 5 pcs.

4. Flasks for reagents (10 pcs.)
5. Burette (1 pc).
6. Funnel - 1 pc.
7. Mortar and pestle - 1 pc.
8. Water bath - 1 piece
9. Fluoroscope

Material for the study:

1. Potato
2. Apple
3. Cabbage
4. Urine
5. Solutions: 1% vitamin C and 5% vitamin B₁

Reagents:

2. Solutions:
 - 10% hydrochloric acid
 - 0,001 N 2, 6-dichlorophenolindophenole sodium (DCIP)
 - 10% NaOH
 - 1% FeCl₃
 - 5% K₃[Fe(CN)₆]
3. Methylene blue
4. Grains of K₃[Fe(CN)₆]

7. REFERENCES TO PREPARE FOR CLASS

Basic:

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LESSON ON TOPIC: “THE ROLE OF FAT-SOLUBLE VITAMINS IN THE METABOLIC PROCESSES OF HUMANS. ANTIVITAMINS”

1. RELEVANCE OF TOPIC

Vitamins of the groups A, D, E, K, and vitamin-like substances unsaturated higher fatty acids (vitamin F) are related to fat-soluble vitamins. Fat-soluble vitamins due to long hydrocarbon radicals in the structure of molecules interact well with hydrophobic solvents. Therefore most of these vitamins are components of biomembranes, under which they perform specific biological functions. For example vitamins E, A, carotinoids are powerful bioantioxidants.

Fat-soluble vitamins are different in comparison with water soluble vitamins by absorption. Absorption of the above mentioned vitamins in the small intestine occurs only if there are lipid digestion products, and bile acids produced by the liver. This knowledge gives understanding of the causes of vitamin A deficiency symptoms in the case, for example, bile duct obstruction. On the other hand, unlike the water-soluble vitamins, excessive intake of fat-soluble vitamins (especially A, D, K) is dangerous for the human body, as these compounds can accumulate in the tissue depot and cause toxic effects so-called state of hypervitaminosis.

Determination of the fat-soluble vitamin content in pharmaceuticals, biological fluids, tissue homogenates is performed by qualitative reactions and quantitative methods using spectrophotometry, fluorometry, etc. This has a practical application in the assessment of certain states and hypovitaminosis and hypervitaminosis.

Understanding the mechanism of action and metabolism of fat-soluble vitamins and antivitamins necessary future doctors, because nowadays fat-soluble vitamins and antivitamins widely used for prevention and treatment of various disorders of metabolic processes.

2. LEARNING OBJECTIVES FOR LESSON:

To learn theoretical material about mechanism of functioning, synthesis, transport in blood and influence on the metabolism of fat-soluble vitamins.

Consolidate knowledge of theoretical material on topic "The role of fat-soluble vitamins in the metabolic processes of humans. Antivitamins", paying special attention to chemical structure, properties and biological role of these compounds.

Necessary to know:

1. Fat-soluble vitamins: classification, common notions about their use in humans. Antivitamins. Hypervitaminosis, hypovitaminosis and avitaminosis for fat-soluble vitamins. The reasons of fat-soluble vitamins deficiency and their occurrence in nature. The interaction of the vitamins in an organism.
2. Vitamin A. Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency in adults and children.
3. Vitamin D. Its structure, biological role, sources of reception in an organism, daily need, signs of insufficiency in adults and children.
4. Vitamin E. Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
5. Vitamin K. Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.
6. Vitamin F. Its structure, biological role, sources of reception in organism, daily need, signs of insufficiency.

3. A FLOW CHART OF THE PRACTICAL TRAINING

Stage	Time (min)	Educational materials		Location
		Content of the plan point	Equipment	
1. Organizational moment	5 min	Check the presence of students	Register	Classroom
2. Colloquium on the outstanding by students questions at home but necessary for the performance of individual tasks	20 min		Methodical recommendations for practical training	Classroom
3. Control of incoming knowledge	15 min	Variants of test tasks	Printed materials	Classroom
4. Independent work of students: - abstractive students report on the topics "Antioxidant function of vitamins ", and "Features of vitamin provision for children" and their discussion; - individual tasks	45 min	Textbooks Methodical recommendations for practical training	Empty table	Classroom
5. Control of the final level of knowledge	20 min	MCQs	Printed materials	Classroom
6. Discussion of student's work during practical training	20 min	Oral questioning of students on topic of the lesson	A list of references to help students to eliminate the disadvantages of studying topics	Classroom
7. Rating of all types of students works during the practical training and motivation for the next practical training	10 min	Check and sign of protocols, recommendations for next lesson topic and tasks for independent work		Classroom

4. LEARNING CONTENT

4.1. Organizing time

To acquaint students with the goals and plan of the practical lesson. To note the value of studying topics, widely used to treat by various vitamins. Check readiness of students for practical classes (presence of “Biochemistry laboratory manual Module 2. Conjugated protein metabolism. Molecular biology. The biochemistry of cell-to-cell interrelations. For independent work at home and in class, preparation for licensing examination “Krok 1” for students of second year study of international faculty” with marked MCQs.

4.2. Interview with issues that have not been cleared up at home. In an interview with the students:

1. Find incomprehensible to students' questions and be sure to consider:

1) general properties of fat-soluble vitamins: features of absorption in the gastrointestinal tract, blood transport, the possibility of accumulation in membranes and development of hypervitaminosis;

2) the structure, properties and biological role, mechanisms of functioning in metabolic processes for vitamins A, D, K, E and F;

2. Mark the importance of understanding the mechanisms of participation of fat-soluble vitamins in metabolic processes. Pay student's attention on understanding the mechanism of antivitamins action that enables their use in treatment of diseases.

4.3. Check the initial level of knowledge on role of fat-soluble vitamins in metabolism and mechanism of antivitamin action.

Teachers should check the elementary students' knowledge on structure, properties, daily requirement, sources, symptoms of deficiency of fat-soluble vitamins A, D, E, K and definition for antivitamin. In order to clarify compliance with the initial level of knowledge required students the teacher offers the following tasks:

Reading a statement to evaluate the accuracy of it and in the event of incorrect statements suggest replacement words for a true statement.

Variant 1

1. Beta carotene and other provitamin A carotenoids are cleaved in the intestinal mucosa by carotene dioxygenase yielding retinaldehyde.
2. Vitamin E is not strictly a vitamin since it can be synthesized in the skin.
3. There has been provided evidence that vitamin E is needed as a stimulator for the hepatic synthesis of at least four protein enzymes involved in the complicated process of blood coagulation.
4. γ -carboxylation of glutamic acid residue in the protein molecule proceeds post-translationally with the vitamin-A-mediated participation of γ -glutamyl-carboxylase.
5. Vitamin E and selenium act synergistically in their actions against lipid peroxides.
6. Impaired fat absorption leads to fat-soluble vitamins deficiency because they are found dissolved in the fat of the diet and are liberated and absorbed during fat digestion.
7. Retinoic acid functions as the prosthetic group of the light-sensitive opsin proteins.
8. The main function of vitamin D is in the regulation of calcium absorption and homeostasis.
9. Calcitriol acts to reduce its own synthesis by inducing the 1-hydroxylase and repressing the 24-hydroxylase in the kidney.
10. The capacity of dicoumarol to increase blood coagulation is currently made use of in the treatment of human diseases with increased blood coagulability.

Variant 2

1. Vitamin K preparations have found applications in medical practice – occasionally they are used to prevent spontaneous abortions in human females.
2. Vitamin K has fertility biological activity.

3. Vitamin K is a monobasic unsaturated cyclic alcohol, whose structural basis is constituted by a fused ring system of cyclopentanoperhydrophenanthrene. It is related to steroids.
4. Vitamins belonging to the D group are polyisoprenoid-substituted naphthoquinones.
5. Vitamin D is the most active and, probably, most important natural fat-soluble antioxidants capable of preventing polyunsaturated fatty acids from oxidation.
6. Fat-soluble vitamins are transported in the blood by proteins: first of all by ceruloplasmin and transferrin.
7. Vitamin E is a cyclic unsaturated monobasic alcohol, composed of a six-membered ring, two isoprene residues and a primary carbinol group
8. In thromboses and thrombophlebitis dicoumarol can be effective in the dissolution of blood clots.
9. A most important function of vitamin K is control of cell differentiation and turnover.
10. More prolonged deficiency of vitamin D leads to xerophthalmia.

Variant 3

1. Vitamin A has antirachitic biological activity.
2. Vitamin A in excess can lead to contraction of blood vessels, high blood pressure and calcinosis – the calcification of soft tissues.
3. Vitamin E has antixerophthalmic biological activity.
4. All retinols are derivatives of tocol. Various retinols differ from each other in the number and position of methyl groups in the aromatic ring.
5. Vitamin D has antihemorrhagic biological activity.
6. The biological role of vitamin D is thus seen in the prevention of biomembrane lipids from autooxidation.
7. Sterilization of the large intestine by antibiotics can result in deficiency of vitamin D when dietary intake is limited.

8. Vitamin D is metabolized to the active metabolite, calcitriol, in liver and kidney.
9. α -tocopherol acts as cofactor of the carboxylase.
10. The naturally occurring fat-soluble vitamins are absorbed only in the presence of trypsin and chymotrypsin.

Keys:

Variant 1

N	Incorrect statement contains	Correct statement must contains
1	True	
2	E	D
3	E	K
4	A	K
5	True	
6	True	
7	Retinoic acid	Retinaldehyde
8	True	
9	Inducing the 1-hydroxylase Repressing the 24-hydroxylase	Inducing the 24-hydroxylase Repressing the 1-hydroxylase
10	To increase	To decrease

Variant 2

N	Incorrect statement contains	Correct statement must contains
1	K	E
2	K	E
3	K	D
4	D	K

5	D	E
6	Proteins: Ceruloplasmin & transferrin	Lipoproteins: ChM& VLDL
7	E	A
8	True	
9	K	A
10	D	A

Variant 3

N	Incorrect statement contains	Correct statement must contains
1	A	D
2	A	D
3	E	A
4	A	E
5	D	K
6	D	E
7	D	K
8	calciol	calcitriol
9	E	K
10	Trypsin&chymotrypsin	Bile salts

4.4. Independent work with the table "Characteristics of fat-soluble vitamins and their functions".

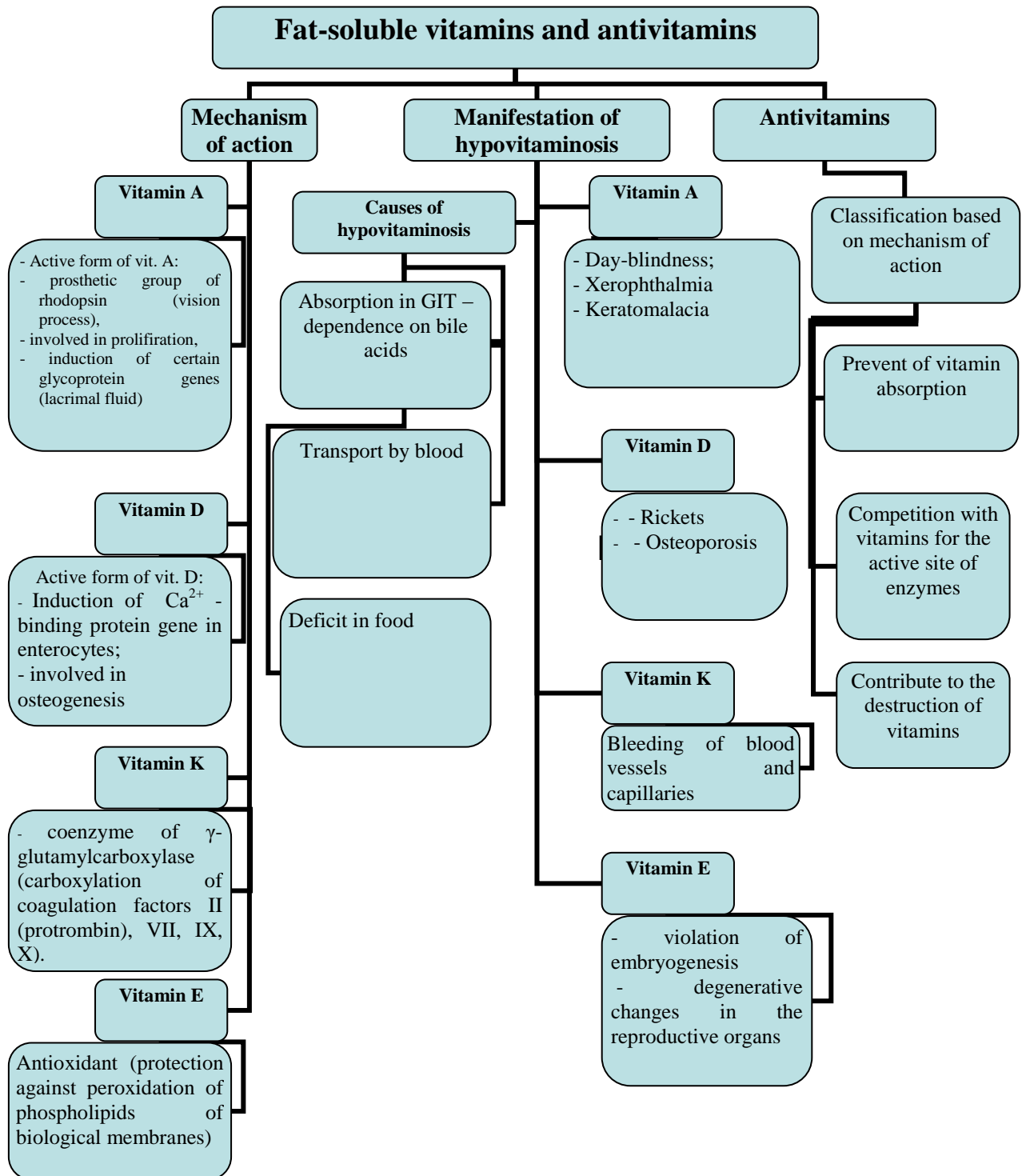
Student should pay special attention to the biochemical aspects of fat-soluble vitamins participating in metabolic processes. Students are encouraged to fill in the table:

Table 1. Vitamin functions and manifestations of hypo- and avitaminoses

Vitamin		Functions	Hypo- and hepervitaminosis symptomes
A			
D			
E			
K			
F			

After the self-study students report the results to prove to the logical end of table. The teacher summarizes students' independent work. At the interview the teacher should monitor the use of all concepts and terms that occur in a graph of logical structure of theme:

4.4. GRAPH OF LOGICAL STRUCTURE OF THEME



4.5. Final control of knowledge on the topic.

After summing up the individual work the teacher is necessary to control the final level of students' knowledge respectively theoretical questions for the class:

1. The features of fat-soluble vitamins absorption in the gastro-intestinal tract.
2. Hypervitaminosis states for fat-soluble vitamins.
3. A group of vitamin A (retinol, retinal, retinoic acid) and β -carotenes: structure, sources of reception, daily requirement, biological role; clinical symptoms of deficiency
4. A group of vitamin E (tocopherols): structure, sources of reception, daily requirement, biological role; clinical symptoms of deficiency.
5. A group of vitamin D (D_2 , D_3 , calcitriols): structure, sources of reception, daily requirement, biological role; clinical symptoms of deficiency in children (1-3 years of age) and in adults.
6. A group of vitamin K (naphtoquinones): structure, sources of reception, daily requirement, biological role; clinical symptoms of deficiency.
7. Vitamin F (a complex of unsaturated high fatty acids): structure, sources of reception, daily requirement, biological role; clinical symptoms of deficiency.
8. Antivitamins: the examples of their mechanism of action.
9. Interaction of vitamins. Combined vitamin drugs (multi-vitamins) in the prophylactic and treatment of some diseases.

4.5. TESTS

1. Point out the vitamin, which is soluble in lipids:
 - A. Vitamin C
 - B. Vitamin B₁
 - C. Vitamin PP
 - D. Vitamin K
 - E. Vitamin H
2. Choose the vitamin, whose precursor is named as b-carotene:
 - A. Vitamin C
 - B. Vitamin D
 - C. Vitamin A

- D. Vitamin B₁₂
 - E. Vitamin P
3. Choose the vitamin, whose molecule structure is unsaturated cyclic alcohol (one hydroxide-group only):
- A. Vitamin K
 - B. Vitamin F
 - C. Vitamin B₅
 - D. Vitamin D₂
 - E. Vitamin H
4. Choose the vitamin, whose antivitamin is named as Dicoumarol:
- A. Vitamin A
 - B. Vitamin B₆
 - C. Vitamin C
 - D. Vitamin D
 - E. Vitamin K
5. Choose the vitamin, whose deficiency leads to osteomalacia at adults:
- A. Vitamin C
 - B. Vitamin E
 - C. Vitamin D
 - D. Vitamin K
 - E. Vitamin PP
6. Choose the vitamin, which is a powerful natural antioxidant:
- A. Retinal
 - B. Tocopherol
 - C. Ergocalciferol
 - D. Riboflavin
 - E. Pyridoxine
7. Name the blood plasma index whose low value will prove the deficiency of vitamin K in patient:

- A. Urea
- B. Albumins
- C. Immunoglobulin G
- D. Prothrombin
- E. C-reactive protein

8. Name the active form of vitamin whose level in the blood is depended on the secretion rate of parathyroid hormone:

- A. Ascorbic acid
- B. Calcitriol
- C. Thiamine
- D. Tocopherol
- E. Naphtoquinone

9. Find out the fat-soluble vitamin whose function is hormone-similar one:

- A. Vitamin C
- B. Vitamin E
- C. Vitamin D
- D. Vitamin K
- E. Vitamin PP

10. Vitamin A group contains substance whose function is associated mainly with stimulation of proliferation and differentiation processes in tissues. Name it:

- A. Cholecalciferol
- B. Pantothenic acid
- C. Retinoic acid
- D. Nicotinic acid
- E. Nicotine amide

ANSWERS TO TEST-TASKS FOR SELF-CONTROL:

1	2	3	4	5	6	7	8	9	10
D	C	D	E	C	B	D	B	D	C

14. TESTS FOR PREPARATION TO «KROK-1» EXAMINATION

1. A patient suffers from vision impairment – hemeralopia (night blindness). What vitamin preparation should be administered the patient in order to restore his vision?
 - A. Pyridoxine
 - B. Retinol acetate
 - C. Vicasol
 - D. Thiamine chloride
 - E. Tocopherol acetate
2. There is disturbed process of Ca^{2+} absorption through intestinal wall after the removal of gall bladder in patient. What vitamin will stimulate this process?
 - A. K
 - B. C
 - C. D_3
 - D. PP
 - E. B_{12}
3. A 6 y.o child was administered vicasol to prevent postoperative bleeding. Vicasol is a synthetic analogue of vitamin K. Name post-translation changes of blood coagulation factors that will be activated by vicasol:
 - A. Carboxylation of glutamic acid residues
 - B. Polymerization
 - C. Partial proteolysis
 - D. Glycosylation
 - E. Phosphorylation of serine radicals
4. A patient who was previously ill with mastectomy as a result of breast cancer was prescribed radiation therapy. What vitamin preparation has marked radioprotective action caused by antioxidant activity?
 - A. Tocopherol acetate
 - B. Riboflavin

- C. Folic acid
 - D. Ergocalciferol
 - E. Thiamine chloride
5. There is an inhibited coagulation in the patients with bile ducts obstruction, bleeding due to the low level of absorption of vitamin. What vitamin is in deficiency?
- A. K
 - B. E
 - C. D
 - D. A
 - E. Carotene
4. A 2-year-old child has got intestinal dysbacteriosis, which results in hemorrhagic syndrome. What is the most likely cause of hemorrhage of the child?
- A. Activation of tissue thromboplastin
 - B. PP hypovitaminosis
 - C. Fibrinogen deficiency
 - D. Vitamin K insufficiency
 - E. Hypocalcemia
7. During examination of an 11-month-old infant a pediatrician revealed osteoectasia of the lower extremities and delayed mineralization of cranial bones. Such pathology is usually provoked by the deficit of the following vitamin:
- A. Thiamin
 - B. Riboflavin
 - C. Bioflavonoids
 - D. Pantothenic acid
 - E. Cholecalciferol
8. A patient presents with twilight vision impairment. Which of the following vitamins should be administered?
- A. Cyanocobalamin

- B. Ascorbic acid
 - C. Nicotinic acid
 - D. Retinol acetate
 - E. Pyridoxine hydrochloride
9. After the disease a 16-year-old boy is presenting with decreased function of protein synthesis in the liver as a result of vitamin *K* deficiency. This may cause disorder of:
- A. Erythropoietin production
 - B. Erythrocyte sedimentation rate
 - C. Blood coagulation
 - D. Osmotic blood pressure
 - E. Anticoagulant production
10. A patient suffers from vision impairment – hemeralopia (night blindness). What vitamin preparation should be administered the patient in order to restore his vision?
- A. Pyridoxine
 - B. Retinol acetate
 - C. Vicasol
 - D. Thiamine chloride
 - E. Tocopherol acetate
11. A 6 y.o child was administered vicasol to prevent postoperative bleeding. Vicasol is a synthetic analogue of vitamin K. Name post-translation changes of blood coagulation factors that will be activated by vicasol:
- A. Carboxylation of glutamic acid residues
 - B. Polymerization
 - C. Partial proteolysis
 - D. Glycosylation
 - E. Phosphorylation of serine radicals

12. A patient who was previously ill with mastectomy as a result of breast cancer was prescribed radiation therapy. What vitamin preparation has marked radioprotective action caused by antioxidant activity?
- A. Tocopherol acetate
 - B. Riboflavin
 - C. Folic acid
 - D. Ergocalciferol
 - E. Thiamine chloride
13. There is an inhibited coagulation in the patients with bile ducts obstruction, bleeding due to the low level of absorption of vitamin. What vitamin is in deficiency?
- A. K
 - B. E
 - C. D
 - D. A
 - E. Carotene
14. A 2-year-old child has got intestinal dysbacteriosis, which results in hemorrhagic syndrome. What is the most likely cause of hemorrhage of the child?
- A. Activation of tissue thromboplastin
 - B. PP hypovitaminosis
 - C. Fibrinogen deficiency
 - D. Vitamin K insufficiency
 - E. Hypocalcemia
15. Deficiency of which of the following vitamins can lead to anemia?
- A. Folic acid
 - B. Vitamin B₁₂
 - C. Vitamin C
 - D. Vitamin E
 - E. All of the above
16. Choose the most active form of vitamin D₃:

- A. 25-Hydroxycholecalciferol
- B. 25-Hydroxyergocalciferol
- C. 24, 25-Dihydroxycholecalciferol
- D. 1, 25-Dihydroxycholecalciferol
- E. Calcidiol

17. What reaction is in need of vitamin K?

- A. Gamma-carboxylation
- B. Oxidation
- C. Methylation
- D. Hydroxylation
- E. Alpha-decarboxylation

18. All of the following are antioxidants except one. Choose it.

- A. Tocopherol
- B. Beta-Carotene
- C. L-Ascorbic acid
- D. Cholecalciferol
- E. Retinol

19. During examination of an 11-month-old infant a pediatrician revealed osteoectasia of the lower extremities and delayed mineralization of cranial bones. Such pathology is usually provoked by the deficit of the following vitamin:

- A. Thiamine
- B. Riboflavin
- C. Bioflavonoids
- D. Pantothenic acid
- E. Cholecalciferol

20. A patient presents with twilight vision impairment. Which of the following vitamins should be administered?

- A. Cyanocobalamin
- B. Ascorbic acid

- C. Nicotinic acid
- D. Retinol acetate
- E. Pyridoxine hydrochloride

21. In clinical practice tuberculosis is treated with isoniazid preparation – that is an antivitamin able to penetrate into the tuberculosis bacillus. Tuberculostatic effect is induced by the interference with replication processes and oxidation-reduction reactions due to the buildup of pseudo-coenzyme:

- A. FMN
- B. NAD
- C. CoQ
- D. FAD
- E. TPP

22. Some infectious diseases caused by bacteria are treated with sulfanilamides, which block the synthesis of bacteria growth factor. What is the mechanism of their action?

- A. They inhibit the absorption of folic acid
- B. They are allosteric enzyme inhibitors
- C. They are allosteric enzymes
- D. They are anti-vitamins of para-amino benzoic acid
- E. They are involved in red-ox processes

23. A patient complains of photoreception disorder and frequent acute viral diseases. He has been prescribed a vitamin that affects photoreception processes by producing rhodopsin, the photosensitive pigment. What vitamin is it?

- A. Cyanocobalamin
- B. Tocopherol acetate
- C. Pyridoxine hydrochloride
- D. Thiamine
- E. Retinol acetate

24. A 6-year-old child suffers from delayed growth, disrupted ossification processes, decalcification of teeth. What can be the cause?

- A. Vitamin D deficiency
 - B. Hyperthyroidism
 - C. Vitamin C deficiency
 - D. Decreased glucagon production
 - E. Insulin deficiency
25. A patient, who has been suffering for a long time from intestine disbacteriosis, has increased hemorrhaging caused by disruption of posttranslational modification of blood coagulation factors II, VII, IX and X in the liver. What vitamin deficiency is the cause of this condition?
- A. K
 - B. B₁₂
 - C. B₉
 - D. C
 - E. P
26. During regular check-up a child is detected with interrupted mineralization of bones. What vitamin deficiency can be the cause?
- A. Calciferol
 - B. Riboflavin
 - C. Tocopherol
 - D. Folic acid
 - E. Cobalamin
27. Choose uncharacteristic symptom of vitamin A deficiency in humans among the listed ones below:
- A. Growth retardation
 - B. Malformation of the long bones
 - C. Loss of body mass
 - D. Affected mucous epithelium and eyes
 - E. Night blindness

Answers to test-tasks for self-control:

1	2	3	4	5	6	7	8	9	10
E	C	A	A	A	D	E	D	C	B
11	12	13	14	15	16	17	18	19	20
A	A	A	D	E	C	A	D	E	D
21	22	23	24	25	26	27			
B	D	E	A	A	A	B			

4.6. Discussion of practical classes

A teacher sums up all kinds of work for each student at the end of class. Student work is positively evaluated in the case of all types of work are done. The teacher recommends students literature to help eliminate the disadvantages of studying the topic in the case of negative results of two tests.

At the end of class the teacher should be sure to consider the amount of student preparation for the next lesson, emphasize the most important issues for home preparation.

5. INDIVIDUAL INDEPENDENT STUDENT WORK:

It can be recommended for students to prepare essay on the topic:

1. Antioxidant function of vitamins
2. Features of vitamin provision for children.

6. MATERIAL PROVISION:

Equipment:

1. Monitor for a slide show

7. THEORETICAL MATERIAL FOR LESSONs:

- THE ROLE OF WATER-SOLUBLE VITAMINS IN THE METABOLISM OF HUMANS. VITAMIN SIMILAR SUBSTANCES”
- THE ROLE OF FAT-SOLUBLE VITAMINS IN THE METABOLIC PROCESSES OF HUMANS. ANTIVITAMINS

Vitamins are a group of organic nutrients of various nature required in small quantities for multiple biochemical reactions for the growth, survival and reproduction of the organism, and which, generally, cannot be synthesized by the body and must therefore be supplied by the diet. The most prominent function of the vitamins is to serve as coenzymes (or prosthetic group) for enzymatic reactions.

The discovery of the vitamins began with experiments performed by Hopkins at the beginning of the twentieth century; he fed rats on a defined diet providing the then known nutrients: fats, proteins, carbohydrates, and mineral salts. The animals failed to grow, but the addition of a small amount of milk to the diet both permitted the animals to maintain normal growth and restored growth to the animals that had previously been fed the defined diet. He suggested that milk contained one or more “accessory growth factors” – essential nutrients present in small amounts, because the addition of only a small amount of milk to the diet was sufficient to maintain normal growth and development. The first of the accessory food factors to be isolated and identified was found to be chemically an amine; therefore, in 1912, Funk coined the term *vitamine*, from the Latin *vita* for “life” and amine, for the prominent chemical reactive group. Although subsequent accessory growth factors were not found to be amines, the name has been retained—with the loss of the final “-e” to avoid chemical confusion. The decision as to whether the word should correctly be pronounced “vitamin” or “veitamin” depends in large part on which system of Latin pronunciation one learned – the Oxford English Dictionary permits both. During the first half of the twentieth century, vitamin deficiency diseases were common in developed and developing countries. At the beginning of the twenty-first century, they are generally rare, although

vitamin A deficiency is a major public health problem throughout the developing world, and there is evidence of widespread subclinical deficiencies of vitamins B₂ and B₆. In addition, refugee and displaced populations are at risk of multiple B vitamin deficiencies, because the cereal foods used in emergency rations are not usually fortified with micronutrients.

Vitamins are grouped together according to the following **general biological characteristics**:

1. Vitamins are not synthesized by the body and must come from food. An exception are vitamin B₃ (PP), which active form NADH (NADPH) can be synthesized from tryptophan and vitamin D₃ (cholecalciferol), synthesized from 7-dehydrocholesterol in the skin. Amount of those ones and vitamins partially synthesized by intestinal microflora (B₁, B₂, B₃, B₅, B₆, K, and others) is normally not sufficient to cover the body's need them.

2. Vitamins are not plastic material. Exception is vitamin F.

3. Vitamins are not an energy source. Exception is vitamin F.

4. Vitamins are essential for all vital processes and biologically active already in small quantities.

5. They influence biochemical processes in all tissues and organs, i.e. they are not specific to organs.

6. They can be used for medicinal purposes as a non-specific tools in high doses for: diabetes mellitus - B₁, B₂, B₆; colds and infectious diseases - vitamin C; bronchial asthma - vitamin PP; gastrointestinal ulcers - vitamin-like substance U and nicotinic acid; in hypercholesterolemia - nicotinic acid.

Since only a few vitamins can be stored (A, D, E, B₁₂), a lack of vitamins quickly leads to **deficiency diseases (hypovitaminosis or avitaminosis)**. These often affect the skin, blood cells, and nervous system. The causes of vitamin deficiencies can be treated by improving nutrition and by administration vitamins in tablet form. An overdose of vitamins leads to **hypervitaminosis state** only, with toxic symptoms, in the case of vitamins A and D. Normally, excess vitamins are rapidly excreted with the urine.

Lack of vitamins leads to the development of pathological processes in the form of specific hypo- and avitaminosis. Widespread hidden forms of vitamin deficiency have not severe external manifestations and symptoms, but have a negative impact on performance, the overall tone of the body and its resistance to various adverse factors.

Avitaminosis is a disease that develops in the absence of a particular vitamin. Currently avitaminosis are not commonly found, but hypovitaminoses are observed with vitamin deficiency in the body. Numerous examples you can see in the table 1.

Table 1. **Vitamin functions and manifestations of hypo- and avitaminoses**

Vitamin		Functions	Hypovitaminosis symptoms
B ₁	Thiamin	Functional part of coenzyme TPP in pyruvate and α -ketoglutarate dehydrogenases, transketolase; poorly defined function in nerve conduction	Peripheral nerve damage (polyneuritis beriberi) or central nervous system lesions (Wernicke-Korsakoff syndrome) Concentration of pyruvate is increased in the patient's blood, the most of which is excreted with urine
B ₂	Riboflavin	Functional part of coenzymes FAD, FMN in oxidation-reduction reactions	Epithelial, mucosa, cutaneous, corneal lesions: lesions of corner of mouth, lips, and tongue; seborrheic dermatitis
B ₃ (PP)	Niacin, nicotinic acid, nicotinamide	Functional part of coenzymes NAD ⁺ , NADP ⁺ in oxidation-reduction reactions	Pellagra: photosensitive dermatitis, glossitis (tongue inflammation), alopecia (hair loss), edema (swelling), diarrhea, depressive psychosis, aggression, ataxia (lack of coordination), dementia, weakness
B ₅	Pantothenic acid	Functional part of coenzyme CoA (universal acyl carrier in Krebs cycle, fatty and other carboxylic acid metabolism) and phosphopantetheine (acyl carrier protein in fatty acid synthesis)	Numbness in the toes, burning sensation in the feet, the defeat of mucous membranes of internal organs, early graying, hair loss, various disorders of the skin: the development of small cracks in the corners of the mouth, the appearance of white patches on various parts of the body. There may also be depressed mood, fatigue.
B ₆	Pyridoxine, pyridoxal, pyridoxamine	Functional part of coenzyme PLP in transamination and decarboxylation of amino acids and glycogen phosphorylase	Dermatitis of the eyes, nose, and mouth. There is mental confusion, glossitis and peripheral neuropathy, convulsions (due to lack of inhibitory neurotransmitter GABA)
B ₇ (H)	Biotin	Coenzyme in carboxylation reactions in gluconeogenesis and fatty	Seborrheic dermatitis, anemia, depression, hair loss, high blood sugar levels, inflammation or pallor of the skin and

		acid synthesis	mucous membranes, insomnia, loss of appetite, muscle aches, nausea, sore tongue, dry skin, high blood cholesterol
B ₉	Folic acid	Functional part of coenzyme THFA in transfer of one-carbon fragments	Megaloblastic anemia: red tongue, anemia, lethargy, fatigue, insomnia, anxiety, digestive disorders, growth retardation, breathing difficulties, memory problems. Deficiency during pregnancy is associated with neural tube defects
B ₁₂	Cobalamin	Functional part of coenzymes adenosylcobalamin (Methylmalonyl Co A mutase) and methylcobalamin (Methionine synthase) in transfer of one-carbon fragments and metabolism of folic acid	Vitamin B ₁₂ -deficiency anemia (in other words pernicious anemia or Addison–Biermer anemia) is one of many types of megaloblastic anemias with degeneration of the spinal cord, anemia, fatigue, depression, low-grade fevers, diarrhea, weight loss, neuropathic pain, glossitis (swollen, red and smooth appearance of the tongue), angular cheilitis (sores at the corner of the mouth) Possible manifestations are also hypochromic anemia, splitting hair and loss of hair, increased nail bottling and taste alteration
C	Ascorbic acid	It serves as a donor of protons in hydroxylation reaction for: - collagen synthesis (prolyl- and lysyl residues are hydroxylated by prolyl 3(4)-hydroxylase and lysyl 5-hydroxylase respectively; - catecholamines and steroid hormone synthesis; It has properties of antioxidant; enhances absorption of iron	Scurvy: general weakness, subcutaneous hemorrhages (frequent hemorrhages from internals and mucous membranes), gingival hemorrhages, loss of teeth, formation of spots on the skin, spongy gums, yellow skin, fever, neuropathy Multiple hemorrhages in the places of clothes friction are possible if a person often experiences acute respiratory infections
A	Retinol	Functional part of visual pigments (rhodopsins and iodopsins) in the retina; regulation of gene expression and cell differentiation; β-carotene (provitamin A) is an antioxidant	Vision impairment hemeralopia (night blindness), xerophthalmia; keratinization of skin
D	Calciferol	Stimulation of Ca ²⁺ absorption through intestinal wall, maintenance of calcium balance and mobilization of bone mineral	Rickets = poor mineralization of bone; osteomalacia = bone demineralization Osteoectasia of the lower extremities and delayed mineralization of cranial bones are onerved in infants
E	Tocopherols	Antioxidant, especially in cell membranes	Extremely rare is a serious neurologic dysfunction

K	Phyllo-quinone, mena-quinones	Coenzyme in formation of γ -carboxyglutamate residues in structure of: - factors II (prothrombin), VII, IX, X, XIV, protein S (blood coagulation system); - bone matrix proteins	Impaired blood clotting, hemorrhagic disease, osteoporosis and coronary heart disease Intestinal dysbacteriosis occurs hemorrhagic syndrome
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External causes for hypovitaminosis

1. Lack of the vitamin in the diet or presence of food factors hindering the absorption of vitamin. For example, use of large amounts of raw eggs (they contain protein avidin binds vitamin H (biotin)) as a result may develop a state of hypovitaminosis H.

2. Do not take into account the need for a particular vitamin. For example, in protein-free diet is increasing demand for vitamin PP (with normal diet it may be partially synthesized from tryptophan). If a person consumes much protein, it can increase the need for vitamin B₆ and reduce the need for vitamin PP.

3. Social reasons: urbanization, power and extremely high purity of canned food; antivitamin presence in food. People are not enough exposed to sunlight in large cities - so it can be hypovitaminosis D. In such cases, the medicine uses ultraviolet radiation in the form of different physical treatments, which activate the synthesis of vitamin D₃ from 7-dehydrocholesterol in the skin cells.

Internal causes of hypovitaminosis

1. Physiological increased need for vitamins, for example, during pregnancy, with heavy physical labor.

2. Long-term severe infectious diseases, as well as during the recovery period.

3. Disturbance of vitamin absorption in some diseases of the digestive tract, for example impaired absorption of fat-soluble vitamins is observed at cholelithiasis; vitamin B₁₂ is done with atrophy of the gastric mucosa and a deficiency of Castle intrinsic factor. Another case if a person who hadn't been consuming fats but had been getting enough carbohydrates and proteins for long

time revealed dermatitis, poor wound healing, vision impairment. Lack of vitamins A, D, E, K, F (linoleic, linolenic, arachidonic acids) is probable cause of the metabolic disorder.

4. Intestinal dysbacteriosis. It has the meaning as some vitamins are synthesized by the intestinal microflora (these vitamins are B₃, B₆, B₇ (H), B₉, B₁₂, and K).

5. Cirrhosis. The liver is the major depot of many vitamins, particularly fat-soluble (especially high hepatic reserves of fat soluble vitamins A, D), but also certain water-soluble, such as B₉, B₁₂, etc. In case of vitamin consumption increase and reducing their dietary intake, which is usually the case, for example, in alcoholism, megaloblastic anemia is developed in a short time as a characteristic sign of hypovitaminosis B₉. Patients with cirrhosis may experience blurred vision in the twilight due to malabsorption of vitamin A in the intestine and its reduced deposit in the liver.

6. Genetic defects of some enzymatic systems. For example, vitamin D-resistant rickets occurs in children lack the enzymes involved in the formation of the active form of vitamin D - calcitriol (1, 25-dihydroxycholecalciferol).

CLASSIFICATION AND NOMENCLATURE OF THE VITAMINS

In addition to systematic chemical nomenclature, the vitamins have an apparently illogical system of accepted trivial names arising from the history of their discovery. For several vitamins, a number of chemically related compounds show the same biological activity, because they are either converted to the same final active metabolite or have sufficient structural similarity to have the same activity. Different chemical compounds that show the same biological activity are collectively known as vitamers. Where one or more compounds have biological activity, in addition to individual names there is also an approved generic descriptor to be used for all related compounds that show the same biological activity.

When it was realized that milk contained more than one accessory food factor, they were named A (which was lipid-soluble and found in the cream) and B (which was water-soluble and found in the whey). This division into fat- and water-soluble vitamins is still used, although there is little chemical or nutritional reason for this, apart from some similarities in dietary sources of fat-soluble or water-soluble vitamins. Water-soluble derivatives of vitamins A and K and fat-soluble derivatives of several of the B vitamins and vitamin C have been developed for therapeutic use and as food additives.

As the discovery of the vitamins progressed, it was realized that “Factor B” consisted of a number of chemically and physiologically distinct compounds. Before they were identified chemically, they were given a logical series of alphanumeric names: B₁, B₂, and so forth.

Table 2. Classification of vitamins. Group B.

Alphanumeric name of vitamin	Chemical and other names of vitamin
Vitamin B ₁	thiamine
Vitamin B ₂	riboflavin
Vitamin B ₃	niacin, niacinamide, niacin, niacinamide, RR
Vitamin B ₄	choline
Vitamin B ₅	pantothenic acid
Vitamin B ₆	pyridoxine, pyridoxal
Vitamin B ₇	biotin, vitamin H
Vitamin B ₈	inositol, myo-inositol, vitamin U
Vitamin B ₉	folic acid, foliatsin, vitamin B _C , M
Vitamin B ₁₀	para-aminobenzoic acid, PABA, vitamin H ₁
Vitamin B ₁₁	L-carnitine, vitamin T, vitamin D
Vitamin B ₁₂	cyanocobalamin, cobalamin
Vitamin B ₁₃	orotic acid
Vitamin B ₁₄	pyrrolo-quinoline quinone, metoksantin, coenzyme of

	PQQ (Pyrroloquinoline Quinone)
Vitamin B ₁₅	pangamic acid, sometimes referred to as vitamin B ₁₆
Vitamin B ₁₆	sometimes pangamic acid - B ₁₅ , and sometimes cyanocobalamin - B ₁₂
Vitamin B ₁₇ (misnomer)	L-citral, letral, letril, amygdalin (The structure - a 2glucose + mandelonitrile)

And such vitamins or vitamin similar compounds as:

Vitamin C - ascorbic acid;

Vitamin P - bioflavonoids: quercetin, rutin, myricetin, apigenin, hesperin, hesperidin, luteolin, catechin, eriodictyol, cyaniding and others;

Vitamin N - lipoic acid;

Vitamin U (ulcus - ulcer) - a derivative of methionine-methionine-methyl sulfonium (pharmacology known as "metiosulfoniya chloride")

As can be seen from Table 2, a number of compounds were assigned vitamin status, and were later shown either not to be vitamins, or to be compounds that had already been identified and given other names.

For a compound to be considered a vitamin, it must be shown to be a dietary essential. Its elimination from the diet must result in a more-or-less clearly defined deficiency disease, and restoration must cure or prevent that deficiency disease. Demonstrating that a compound has pharmacological actions, and possibly cures a disease, does not classify that compound as a vitamin, even if it is a naturally occurring compound that is found in foods. Equally, demonstrating that a compound has a physiological function as a coenzyme or hormone does not classify that compound as a vitamin. It is necessary to demonstrate that endogenous synthesis of the compound is inadequate to meet physiological requirements in the absence of a dietary source of the compound. There is some evidence that premature infants and patients maintained on long-term total parenteral nutrition may be unable to meet their requirements for carnitine, choline,

and taurine unless they are provided in the diet, and these are sometimes regarded as “marginal compounds,” for which there is no evidence to estimate requirements.

The rigorous criteria outlined here would exclude niacin and vitamin D from the list of vitamins, because under normal conditions endogenous synthesis does indeed meet requirements. Nevertheless, they are considered to be vitamins, even if only on the grounds that each was discovered as the result of investigations into once common deficiency diseases, pellagra and rickets.

In addition to the marginal compounds like carnitine choline, there are a number of compounds present in foods of plant origin that are considered to be beneficial, in that they have actions that may prevent the development of atherosclerosis and some cancers, although there is no evidence that they are dietary essentials, and they are not generally considered as nutrients.

GROUP I. FAT-SOLUBLE VITAMINS

Group I is Fat-soluble vitamins: A (retinol), D (calciferol), E (tocopherol), K (naphthoquinone), F (polyunsaturated fatty acid: linoleic, linolenic, arachidonic).

VITAMIN A

Vitamin A includes two vitamers: retinol and dehydroretinol but group of vitamin A consists and their biologically active molecules retinal (retinaldehyde) and retinoic acid.

Beta-carotene, which consists of two molecules of retinal linked at their aldehyde ends, is also referred to as the provitamin form of vitamin A.

Ingested β -carotene is cleaved in the lumen of the intestine by beta-carotene dioxygenase to yield retinal. Retinal is right here reduced to retinol by retinaldehyde reductase, an NADPH requiring enzyme within the intestines. Retinol is esterified to palmitic or stearic acids and delivered to the blood via chylomicrons. The uptake of chylomicron remnants by the liver results in delivery of retinol to this organ for storage as a lipid esters. Transport of retinol from the liver to extrahepatic tissues occurs by binding of retinol to retinol binding protein

(RBP). the retinol-RBP complex is then transported to the cell surface within the Golgi and secreted. Within extrahepatic tissues retinol is bound to cellular retinol binding protein (CRBP). Plasma transport of retinoic acid is accomplished by binding to albumin. One protein else is capable to transport of vitamin A: it is transthyretin. Transthyretin (TTR) is a serum and cerebrospinal fluid carrier of the thyroid hormone thyroxine (T_4) and retinol-binding protein bound to retinol. This is how transthyretin gained its name, **transports thyroxine and retinol**. The liver secretes transthyretin into the blood, and the choroid plexus secretes TTR into the cerebrospinal fluid.

TTR was originally called *prealbumin* (or thyroxine-binding prealbumin) because it ran faster than albumin on electrophoresis gels.

Gene Control Exerted by Retinol and Retinoic Acid

Within cells both retinol and retinoic acid bind to specific receptor proteins. Following binding, the receptor-vitamin complex interacts with specific sequences in several genes involved in growth and differentiation and affects expression of these genes. In this capacity retinol and retinoic acid are considered hormones of the steroid/thyroid hormone superfamily of proteins. Vitamin D also acts in a similar capacity. Several genes whose patterns of expression are altered by retinoic acid are involved in the earliest processes of embryogenesis including the differentiation of the three germ layers, organogenesis and limb development.

Vision and the Role of Vitamin A

Photoreception in the eye is the function of two specialized cell types located in the retina; the rod and cone cells. Both rod and cone cells contain a photoreceptor pigment in their membranes. The photosensitive compound of most mammalian eyes is a protein called opsin to which is covalently coupled an aldehyde of vitamin A. The opsin of rod cells is called scotopsin. The photoreceptor of rod cells is specifically called rhodopsin or visual purple. This compound is a complex between scotopsin and the 11-*cis*-retinal (also called 11-

cis-retinene) form of vitamin A. Rhodopsin is a serpentine receptor imbedded in the membrane of the rod cell. Coupling of 11-*cis*-retinal occurs at three of the transmembrane domains of rhodopsin. Intracellularly, rhodopsin is coupled to a specific G-protein called transducin.

When the rhodopsin is exposed to light it is bleached releasing the 11-*cis*-retinal from opsin. Absorption of photons by 11-*cis*-retinal triggers a series of conformational changes on the way to conversion all-*trans*-retinal. One important conformational intermediate is metarhodopsin II. The release of opsin results in a conformational change in the photoreceptor. This conformational change activates transducin, leading to an increased GTP-binding by the α -subunit of transducin. Binding of GTP releases the α -subunit from the inhibitory β - and γ -subunits. The GTP-activated α -subunit in turn activates an associated phosphodiesterase; an enzyme that hydrolyzes cGMP to GMP. cGMP is required to maintain the Na^+ channels of the rod cell in the open conformation. The drop in cGMP concentration results in complete closure of the Na^+ channels. Metarhodopsin II appears to be responsible for initiating the closure of the channels. The closing of the channels leads to hyperpolarization of the rod cell with concomitant propagation of nerve impulses to the brain.

Additional Role of Retinol

Retinol also functions in the synthesis of certain glycoproteins and mucopolysaccharides necessary for mucous production and normal growth regulation. This is accomplished by phosphorylation of retinol to retinyl phosphate which then functions similarly to dolichol phosphate.

Clinical Significances of Vitamin A Deficiency

Vitamin A is stored in the liver and deficiency of the vitamin occurs only after prolonged lack of dietary intake. The earliest symptoms of vitamin A deficiency are night blindness. Additional early symptoms include follicular hyperkeratinosis, increased susceptibility to infection and cancer and anemia

equivalent to iron deficient anemia. Prolonged lack of vitamin A leads to deterioration of the eye tissue through progressive keratinization of the cornea, a condition known as xerophthalmia.

The increased risk of cancer in vitamin deficiency is thought to be the result of a depletion in beta-carotene. β -carotene is a very effective antioxidant and is suspected to reduce the risk of cancers known to be initiated by the production of free radicals. Of particular interest is the potential benefit of increased beta-carotene intake to reduce the risk of lung cancer in smokers. However, caution needs to be taken when increasing the intake of any of the lipid soluble vitamins. Excess accumulation of vitamin A in the liver can lead to toxicity which manifests as bone pain, hepatosplenomegaly, nausea and diarrhea.

VITAMIN D

Vitamin D is a steroid hormone that functions to regulate specific gene expression following interaction with its intracellular receptor. The biologically active form of the hormone is 1,25-dihydroxy vitamin D₃ (1,25-(OH)₂D₃, also termed calcitriol). Calcitriol functions primarily to regulate calcium and phosphorous homeostasis.

Active calcitriol is derived from ergosterol (produced in plants) and from 7-dehydrocholesterol (produced in the skin). Ergocalciferol (vitamin D₂) is formed by UV (ultraviolet) irradiation of ergosterol. In the skin 7-dehydrocholesterol is converted to cholecalciferol (vitamin D₃) following UV irradiation.

Vitamin D₂ and D₃ are processed to D₂-calcitriol and D₃-calcitriol, respectively, by the same enzymatic pathways in the body. Cholecalciferol (or ergocalciferol) are absorbed from the intestine and transported to the liver bound to a specific vitamin D-binding protein. In the liver cholecalciferol is hydroxylated at the 25 position by a specific D₃-25-hydroxylase generating 25-hydroxy-D₃ [25-(OH)D₃] which is the major circulating form of vitamin D. Conversion of 25-(OH)D₃ to its biologically active form, calcitriol, occurs through the activity of a specific D₃-1-hydroxylase present in the proximal convoluted tubules of the kidneys, and in bone and placenta. 25-(OH)D₃ can also be hydroxylated at the 24

position by a specific D_3 -24-hydroxylase in the kidneys, intestine, placenta and cartilage.

Calcitriol functions in concert with parathyroid hormone (PTH) and calcitonin to regulate serum calcium and phosphorous levels. PTH is released in response to low serum calcium and induces the production of calcitriol. In contrast, reduced levels of PTH stimulate synthesis of the inactive $24,25-(OH)_2D_3$. In the intestinal epithelium, calcitriol functions as a steroid hormone in inducing the expression of calbindin D_{28K} , a protein involved in intestinal calcium absorption. The increased absorption of calcium ions requires concomitant absorption of a negatively charged counter ion to maintain electrical neutrality. The predominant counter ion is Pi . When plasma calcium levels fall the major sites of action of calcitriol and PTH are bone where they stimulate bone resorption and the kidneys where they inhibit calcium excretion by stimulating reabsorption by the distal tubules. The role of calcitonin in calcium homeostasis is to decrease elevated serum calcium levels by inhibiting bone resorption.

Clinical Significance of Vitamin D Deficiency

As a result of the addition of vitamin D to milk, deficiencies in this vitamin are rare in this country. The main symptom of vitamin D deficiency in children is **rickets** and in adults is **osteomalacia**. Rickets is characterized improper mineralization during the development of the bones resulting in soft bones. Osteomalacia is characterized by demineralization of previously formed bone leading to increased softness and susceptibility to fracture.

VITAMIN E

Vitamin E is a mixture of several related compounds known as tocopherols. The α -tocopherol molecule is the most potent of the tocopherols.

α -Tocopherol is the main source found in supplements and in the European diet, where the main dietary sources are olive and sunflower oils, while γ -

tocopherol is the most common form in the American diet due to a higher intake of soybean and corn oil.

Tocotrienols, which are related compounds, also have vitamin E activity. All of these various derivatives with vitamin activity may correctly be referred to as "vitamin E". Tocopherols and tocotrienols are fat-soluble antioxidants but also seem to have many other functions in the body.

Vitamin E is absorbed from the intestines packaged in chylomicrons. It is delivered to the tissues via chylomicron transport and then to the liver through chylomicron remnant uptake. The liver can export vitamin E in VLDLs. Due to its lipophilic nature, vitamin E accumulates in cellular membranes, fat deposits and other circulating lipoproteins. The major site of vitamin E storage is in adipose tissue.

The major function of vitamin E is to act as a natural antioxidant by scavenging free radicals and molecular oxygen. In particular vitamin E is important for preventing peroxidation of polyunsaturated membrane fatty acids. The vitamins E and C are interrelated in their antioxidant capabilities. Active α -tocopherol can be regenerated by interaction with vitamin C following scavenge of a peroxy free radical. Alternatively, α -tocopherol can scavenge two peroxy free radicals and then be conjugated to glucuronate for excretion in the bile.

Clinical significances of Vitamin E Deficiency

No major disease states have been found to be associated with vitamin E deficiency due to adequate levels in the average American diet. The major symptom of vitamin E deficiency in humans is an increase in red blood cell fragility. Since vitamin E is absorbed from the intestines in chylomicrons, any fat malabsorption diseases can lead to deficiencies in vitamin E intake. Neurological disorders have been associated with vitamin E deficiencies associated with fat malabsorptive disorders. Increased intake of vitamin E is recommended in premature infants fed formulas that are low in the vitamin as well as in persons consuming a diet high in polyunsaturated fatty acids. Polyunsaturated fatty acids

tend to form free radicals upon exposure to oxygen and this may lead to an increased risk of certain cancers.

VITAMIN K

The K vitamins exist naturally as K₁ (phylloquinone) in green vegetables and K₂ (menaquinone) produced by intestinal bacteria and K₃ is synthetic menadione (vicasol). When administered, vitamin K₃ is alkylated to one of the vitamin K₂ forms of menaquinone.

The major function of the K vitamins is in the maintenance of normal levels of the blood clotting proteins, factors II, VII, IX, X and protein C and protein S, which are synthesized in the liver as inactive precursor proteins. Conversion from inactive to active clotting factor requires a posttranslational modification of specific glutamate residues. This modification is a carboxylation and the enzyme responsible requires vitamin K as a cofactor. The resultant modified protein residues are λ -carboxyglutamate. This process is most clearly understood for factor II, also called prothrombin. Prothrombin is modified preprothrombin. The λ -carboxyglutamate residues are effective calcium ion chelators. Upon chelation of calcium, prothrombin interacts with phospholipids in membranes and is proteolysed to thrombin through the action of activated factor X (Xa).

During the carboxylation reaction reduced hydroquinone form of vitamin K is converted to a 2,3-epoxide form. The regeneration of the hydroquinone form requires an uncharacterized reductase. This latter reaction is the site of action of the dicoumarol based anticoagulants such as warfarin.

The isoprene-derived molecule whose structure is shown here is known alternately as Coumarin and warfarin. By the former name, it is a widely prescribed anticoagulant. By the latter name, it is a component of rodent poisons. How can the same chemical species be used for such disparate purposes? The key to both uses lies in its ability to act as an antagonist of vitamin K in the body. Vitamin K stimulates the carboxylation of glutamate residues on certain proteins, including some proteins in the bloodclotting cascade. Carboxylation of these coagulation

factors is catalyzed by a carboxylase that requires the reduced form of vitamin K (vitamin KH₂), molecular oxygen, and carbon dioxide. KH₂ is oxidized to vitamin K epoxide, which is recycled to KH₂ by the enzymes vitamin K epoxide reductase (1) and vitamin K reductase (2, 3). Coumarin/warfarin exerts its anticoagulant effect by inhibiting vitamin K epoxide reductase and possibly also vitamin K reductase. This inhibition depletes vitamin KH₂ and reduces the activity of the carboxylase. Coumarin/warfarin, given at a typical dosage of 4 to 5 mg/day, prevents the deleterious formation in the bloodstream of small blood clots and thus reduces the risk of heart attacks and strokes for individuals whose arteries contain sclerotic plaques. Taken in much larger doses, as for example in rodent poisons, Coumarin/warfarin can cause massive hemorrhages and death.

Clinical significance of Vitamin K Deficiency

Naturally occurring vitamin K is absorbed from the intestines only in the presence of bile salts and other lipids through interaction with chylomicrons. Therefore, fat malabsorptive diseases can result in vitamin K deficiency. The synthetic vitamin K₃ is water soluble and absorbed irrespective of the presence of intestinal lipids and bile. Since the vitamin K₂ form is synthesized by intestinal bacteria, deficiency of the vitamin in adults is rare. However, long term antibiotic treatment can lead to deficiency in adults. The intestine of newborn infants is sterile, therefore, vitamin K deficiency in infants is possible if lacking from the early diet. The primary symptom of a deficiency in infants is a hemorrhagic syndrome.

GROUP II. WATER-SOLUBLE VITAMINS

Group II is water-soluble vitamins:

- Group B: B₁ (thiamine), B₂ (riboflavin), B₃ or PP (nicotinamide, niacin), B₅ (pantothenic acid), B₆ (pyridoxine), B₇ or H (biotin), B₉ or Bc (folic acid), B₁₂ (cyanocobalamin) ;
- Vitamin C (ascorbic acid);
- Vitamin P (rutin and other bioflavonoids).

Water-soluble vitamins are usually functioning as precursors of coenzymes and prosthetic groups of enzymes. For example, coenzyme form of:

- Vitamin B₁ is TPP (thiamine pyrophosphate) (trade name - cocarboxylase);
- Vitamin B₂ is FMN (flavin mononucleotide) and FAD (flavin adenine dinucleotide);
- Vitamin B₃ is NAD⁺ (nicotinamide adenine dinucleotide) or NADP⁺ (nicotinamide adenine dinucleotide phosphate);
- Vitamin B₅ is Coenzyme A (coenzyme of acylation);
- Vitamin B₆ is PLP (pyridoxal phosphate);
- Vitamin B₉ is THFA (tetrahydrofolic acid);
- Vitamin B₁₂ is adenosylcobalamin and methylcobalamin.

Holoenzymes containing coenzymes (as its non-protein part) which are often vitamin derivatives perform multiple functions. For example, the first enzyme in gluconeogenesis pyruvate carboxylase uses biotin for carboxylation of pyruvate; but the transformation of the pyruvate to acetyl-CoA by pyruvate dehydrogenase complex requires five coenzymes: TPP, lipoic acid, CoA, FAD, NAD⁺. Since TPP is involved in this conversion first, pyruvate accumulation in cells of the nervous system (primarily) and then increase in pyruvate content in the blood and urine of patients in the case of vitamin B₁ deficiencies becomes obvious.

VITAMIN B₁

Vitamin B₁ is also known as thiamine.

Thiamine is derived from a substituted pyrimidine and a thiazole which are coupled by a methylene bridge. Thiamine is rapidly converted to its active form, **Thiamine Pyrophosphate (TPP)**, in the brain and liver by a specific enzymes, thiamine diphosphotransferase.

TPP is necessary as a cofactor for the pyruvate and α -ketoglutarate dehydrogenase catalyzed reactions as well as the transketolase catalyzed reactions of the pentose phosphate pathway. A deficiency in thiamine intake leads to a severely reduced capacity of cells to generate energy as a result of its role in these reactions.

The dietary requirement for thiamine is proportional to the caloric intake of the diet and ranges from 1.0 - 1.5 mg/day for normal adults. If the carbohydrate content of the diet is excessive then an in thiamin intake will be required.

Clinical Significances of Thiamine Deficiency

The earliest symptoms of thiamine deficiency include constipation, appetite suppression, nausea as well as mental depression, peripheral neuropathy and fatigue. Chronic thiamine deficiency leads to more severe neurological symptoms including ataxia, mental confusion and loss of eye coordination. Other clinical symptoms of prolonged thiamine deficiency are related to cardiovascular and musculature defects.

The severe thiamine deficiency disease known as Beriberi, is the result of a diet that is carbohydrate rich and thiamine deficient. An additional thiamine deficiency related disease is known as Wernicke-Korsakoff syndrome. This disease is most commonly found in chronic alcoholics due to their poor dietetic lifestyles.

VITAMIN B₂

Vitamin B₂ is also known as riboflavin.

Riboflavin is the precursor for the coenzymes, flavin mononucleotide (FMN) and flavin adenine dinucleotide (FAD). The enzymes that require FMN or FAD as cofactors are termed flavoproteins. Several flavoproteins also contain

metal ions and are termed metalloflavoproteins. Both classes of enzymes are involved in a wide range of redox reactions, e.g. succinate dehydrogenase and xanthine oxidase. During the course of the enzymatic reactions involving the flavoproteins the reduced forms of FMN and FAD are formed, FMNH₂ and FADH₂, respectively. The normal daily requirement for riboflavin is 1.2 - 1.7 mg/day for normal adults.

Clinical Significances of Riboflavin Deficiency

Riboflavin deficiencies are rare due to the presence of adequate amounts of the vitamin in eggs, milk, meat and cereals. Riboflavin deficiency is often seen in chronic alcoholics due to their poor dietetic habits.

Symptoms associated with riboflavin deficiency include, glossitis, seborrhea, angular stomatitis, cheilosis and photophobia. Riboflavin decomposes when exposed to visible light. This characteristic can lead to riboflavin deficiencies in newborns treated for hyperbilirubinemia by phototherapy.

VITAMIN B₃.

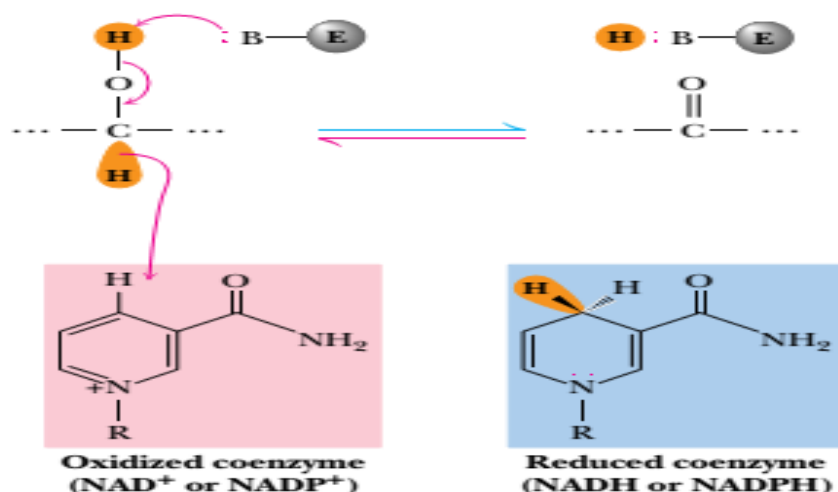
Vitamin B₃ is also known as niacin (nicotinic acid and nicotinamide).

Both nicotinic acid and nicotinamide can serve as the dietary source of vitamin B₃. Niacin is required for the synthesis of the active forms of vitamin B₃, nicotinamide adenine dinucleotide (NAD⁺) and nicotinamide adenine dinucleotide phosphate (NADP⁺). Both NAD⁺ and NADP⁺ function as cofactors for numerous dehydrogenase, e.g., lactate and malate dehydrogenases.

Niacin is not a true vitamin in the strictest definition since it can be derived from the amino acid tryptophan. However, the ability to utilize tryptophan for niacin synthesis is inefficient (60 mg of tryptophan are required to synthesize 1 mg of niacin). Also, synthesis of niacin from tryptophan requires vitamins B₁, B₂ and B₆ which would be limiting in themselves on a marginal diet.

Examination of the structures of NADH and NADPH reveals that the 4-position of the nicotinamide ring is pro-chiral, meaning that while this carbon is not chiral, it would be if either of its hydrogens were replaced by something else.

As shown in following figure the hydrogen “projecting” out of the page toward you is the “pro-R” hydrogen because, if a deuterium is substituted at this position, the molecule would have the R-configuration. Substitution of the other hydrogen would yield an S-configuration.



An interesting aspect of the enzymes that require nicotinamide coenzymes is that they are stereospecific and with draw hydrogen from either the pro-R or the pro-S position selectively. This stereospecificity arises from the fact that enzymes (and the active sites of enzymes) are inherently asymmetric structures. These same enzymes are stereospecific with respect to the substrates as well.

The NAD- and NADP-dependent dehydrogenases catalyze at least six different types of reactions: simple hydride transfer, deamination of an amino acid to form an α -keto acid, oxidation of α -hydroxy acids followed by decarboxylation of the α -keto acid intermediate, oxidation of aldehydes, reduction of isolated double bonds, and the oxidation of carbon–nitrogen bonds (as with dihydrofolate reductase).

The recommended daily requirement for niacin is 13 - 19 niacin equivalents (NE) per day for a normal adult. One NE is equivalent to 1 mg of free niacin).

Clinical Significances of Niacin and Nicotinic Acid

A diet deficient in niacin (as well as tryptophan) leads to glossitis of the tongue, dermatitis, weight loss, diarrhea, depression and dementia. The severe symptoms, depression, dermatitis and diarrhea, are associated with the condition known as **pellagra**.

Pellagra is a disease characterized by dermatitis, diarrhea, and dementia, has been known for centuries. It was once prevalent in the southern part of the United States and is still a common problem in some parts of Spain, Italy, and Romania. Pellagra was once thought to be an infectious disease, but Joseph Goldberger showed early in this century that it could be cured by dietary actions. Soon thereafter, it was found that brewer's yeast would prevent pellagra in humans. Studies of a similar disease in dogs, called black tongue, eventually led to the identification of nicotinic acid as the relevant dietary factor. Elvehjem and his colleagues at the University of Wisconsin in 1937 isolated nicotinamide from liver, and showed that it and nicotinic acid could prevent and cure black tongue in dogs. That same year, nicotinamide and nicotinic acid were both shown to be able to cure pellagra in humans. Interestingly, plants and many animals can synthesize nicotinic acid from tryptophan and other precursors, and nicotinic acid is thus not a true vitamin for these species. However, if dietary intake of tryptophan is low, nicotinic acid is required for optimal health. Nicotinic acid, which is beneficial to humans and animals, is structurally related to nicotine, a highly toxic tobacco alkaloid. In order to avoid confusion of nicotinic acid and nicotinamide with nicotine itself, niacin was adopted as a common name for nicotinic acid. Cowgill, at Yale University, suggested the name from the letters of three words—nicotinic, acid, and vitamin.

Several physiological conditions (e.g. Hartnup disease and malignant carcinoid syndrome) as well as certain drug therapies (e.g. isoniazid) can lead to niacin deficiency. In Hartnup disease tryptophan absorption is impaired and in malignant carcinoid syndrome tryptophan metabolism is altered resulting in excess

serotonin synthesis. Isoniazid (the hydrazide derivative of isonicotinic acid) is the primary drug for chemotherapy of tuberculosis.

Nicotinic acid (but not nicotinamide) when administered in pharmacological doses of 2 - 4 g/day lowers plasma cholesterol levels and has been shown to be a useful therapeutic for hypercholesterolemia. The major action of nicotinic acid in this capacity is a reduction in fatty acid mobilization from adipose tissue. Although nicotinic acid therapy lowers blood cholesterol it also causes a depletion of glycogen stores and fat reserves in skeletal and cardiac muscle. Additionally, there is an elevation in blood glucose and uric acid production. For these reasons nicotinic acid therapy is not recommended for diabetics or persons who suffer from gout.

VITAMIN B₅.

Vitamin B₅ is also known as pantothenic acid.

Pantothenic acid is formed from β -alanine and pantoic acid. Pantothenate is required for synthesis of coenzyme A, CoA and is a component of the acyl carrier protein (ACP) domain of fatty acid synthase. Pantothenate is, therefore, required for the metabolism of carbohydrate via the TCA cycle and all fats and proteins. At least 70 enzymes have been identified as requiring CoA or ACP derivatives for their function.

Deficiency of pantothenic acid is extremely rare due to its widespread distribution in whole grain cereals, legumes and meat. Symptoms of pantothenate deficiency are difficult to assess since they are subtle and resemble those of other B vitamin deficiencies.

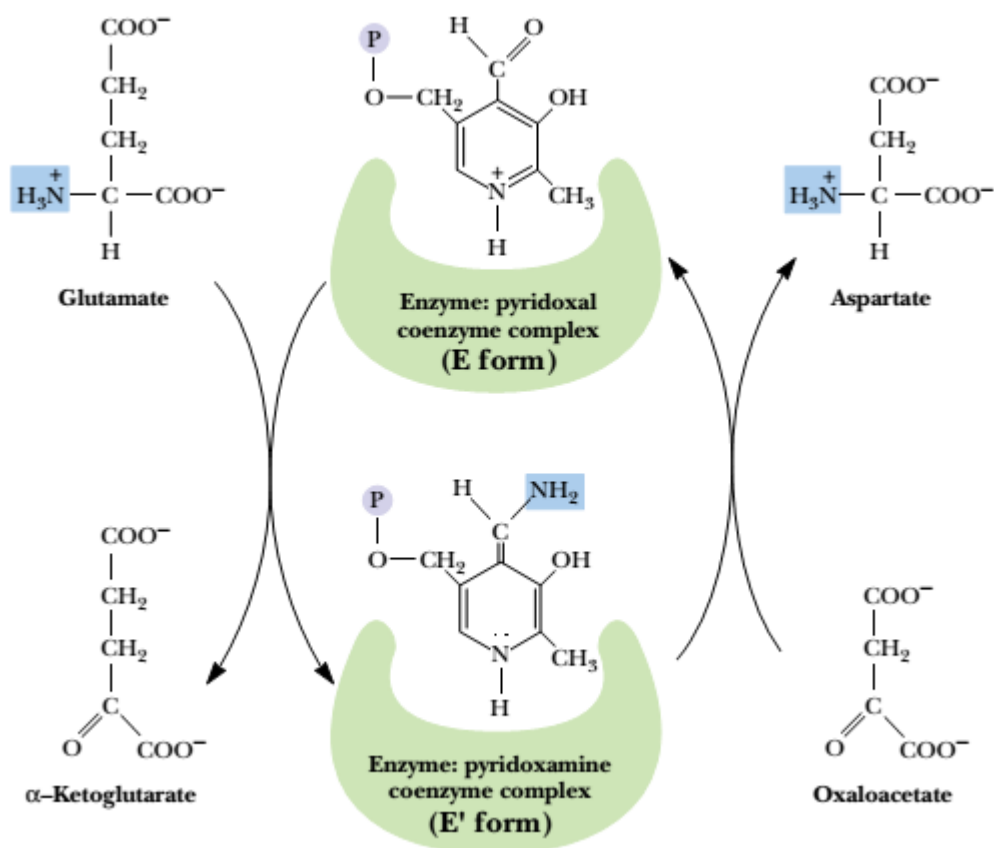
VITAMIN B₆

Vitamin B₆. are collectively known as pyridoxal, pyridoxamine and pyridoxine.

All three compounds are efficiently converted to the biologically active form of vitamin B₆, pyridoxal phosphate. This conversion is catalyzed by the ATP requiring enzyme, pyridoxal kinase.

Pyridoxal phosphate functions as a cofactor in enzymes involved in transamination reactions required for the synthesis and catabolism of the amino acids as well as in glycogenolysis as a cofactor for glycogen phosphorylase.

A specific example would be glutamate : aspartate aminotransferase. It is a pyridoxal phosphate – dependent enzyme. Glutamate : aspartate aminotransferase is an enzyme conforming to a double-displacement bisubstrate mechanism. The pyridoxal serves as the -NH₂ acceptor from glutamate to form pyridoxamine. Pyridoxamine is then the amino donor to oxaloacetate to form aspartate and regenerate the pyridoxal coenzyme form.



The requirement for vitamin B₆ in the diet is proportional to the level of protein consumption ranging from 1.4 - 2.0 mg/day for a normal adult. During

pregnancy and lactation the requirement for vitamin B₆ increases approximately 0.6 mg/day.

Deficiencies of vitamin B₆ are rare and usually are related to an overall deficiency of all the B-complex vitamins. Isoniazid (see niacin deficiencies above) and penicillamine (used to treat rheumatoid arthritis and cystinurias) are two drugs that complex with pyridoxal and pyridoxal phosphate resulting in a deficiency in this vitamin.

VITAMIN B₇

Vitamin B₇ is known as biotin.

Biotin is the cofactor required of enzymes that are involved in carboxylation reactions, e.g. acetyl-CoA carboxylase and pyruvate carboxylase. Biotin is found in numerous foods and also is synthesized by intestinal bacteria and as such deficiencies of the vitamin are rare. Deficiencies are generally seen only after long antibiotic therapies which deplete the intestinal fauna or following excessive consumption of raw eggs. The latter is due to the affinity of the egg white protein, avidin, for biotin preventing intestinal absorption of the biotin.

VITAMIN B₉

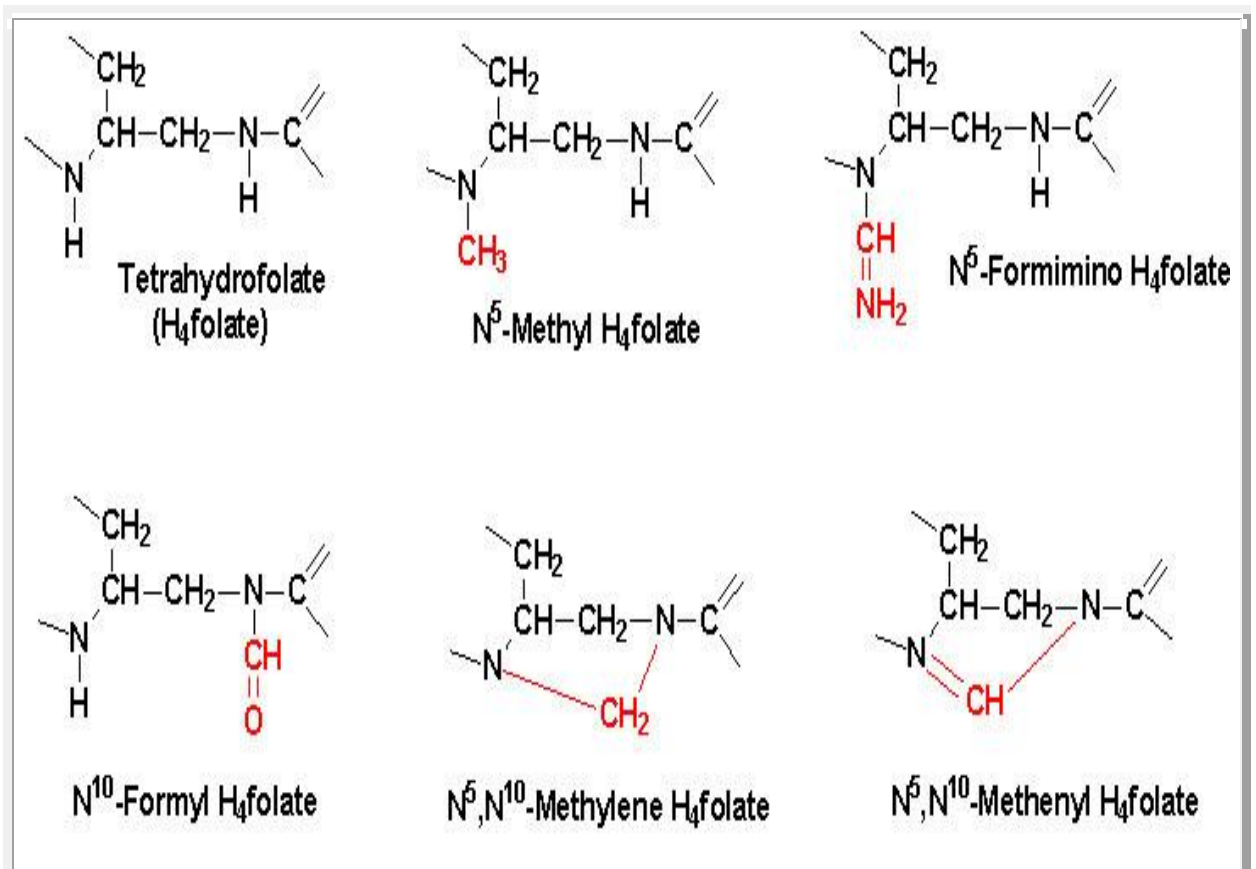
Vitamin B₉ is known as folic acid.

Folic acid is a conjugated molecule consisting of a pteridine ring structure linked to para-aminobenzoic acid (PABA) that forms pteronic acid. Folic acid itself is then generated through the conjugation of glutamic acid residues to pteronic acid. Folic acid is obtained primarily from yeasts and leafy vegetables as well as animal liver. Animal cannot synthesize PABA nor attach glutamate residues to pteronic acid, thus, requiring folate intake in the diet.

When stored in the liver or ingested folic acid exists in a polyglutamate form. Intestinal mucosal cells remove some of the glutamate residues through the action of the lysosomal enzyme, conjugase. The removal of glutamate residues makes folate less negatively charged (from the polyglutamic acids) and therefore

more capable of passing through the basal laminal membrane of the epithelial cells of the intestine and into the bloodstream. Folic acid is reduced within cells (principally the liver where it is stored) to tetrahydrofolate (THF also H₄folate) through the action of dihydrofolate reductase (DHFR), an NADPH-requiring enzyme.

The function of THF derivatives is to carry and transfer various forms of one carbon units during biosynthetic reactions. The one carbon units are either methyl, methylene, methenyl, formyl or formimino groups.



Active center of tetrahydrofolate (THF). Note that the N⁵ position is the site of attachment of methyl groups, the N¹⁰ the site for attachment of formyl and formimino groups and that both N⁵ and N¹⁰ bridge the methylene and methenyl groups.

These one carbon transfer reactions are required in the biosynthesis of serine, methionine, glycine, choline and the purine nucleotides and dTMP.

The ability to acquire choline and amino acids from the diet and to salvage the purine nucleotides makes the role of N⁵,N¹⁰-methylene-THF in dTMP synthesis the most metabolically significant function for this vitamin. The role of vitamin B₁₂ and N⁵-methyl-THF in the conversion of homocysteine to methionine also can have a significant impact on the ability of cells to regenerate needed THF.

Clinical Significance of Folate Deficiency

Folate deficiency results in complications nearly identical to those described for vitamin B₁₂ deficiency. The most pronounced effect of folate deficiency on cellular processes is upon DNA synthesis. This is due to an impairment in dTMP synthesis which leads to cell cycle arrest in S-phase of rapidly proliferating cells, in particular hematopoietic cells. The result is megaloblastic anemia as for vitamin B₁₂ deficiency. The inability to synthesize DNA during erythrocyte maturation leads to abnormally large erythrocytes termed macrocytic anemia.

Folate deficiencies are rare due to the adequate presence of folate in food. Poor dietary habits as those of chronic alcoholics can lead to folate deficiency. The predominant causes of folate deficiency in non-alcoholics are impaired absorption or metabolism or an increased demand for the vitamin. The predominant condition requiring an increase in the daily intake of folate is pregnancy. This is due to an increased number of rapidly proliferating cells present in the blood. The need for folate will nearly double by the third trimester of pregnancy. Certain drugs such as anticonvulsants and oral contraceptives can impair the absorption of folate. Anticonvulsants also increase the rate of folate metabolism.

VITAMIN B₁₂

Vitamin B₁₂ is known as cobalamin.

Cobalamin is more commonly known as vitamin B₁₂. Vitamin B₁₂ is composed of a complex tetrapyrrol ring structure (corrin ring) and a cobalt ion in the center. Vitamin B₁₂ is synthesized exclusively by microorganisms and is found

in the liver of animals bound to protein as methylcobalamin or 5'-deoxyadenosylcobalamin. The vitamin must be hydrolyzed from protein in order to be active. Hydrolysis occurs in the stomach by gastric acids or the intestines by trypsin digestion following consumption of animal meat. The vitamin is then bound by intrinsic factor, a protein secreted by parietal cells of the stomach, and carried to the ileum where it is absorbed. Following absorption the vitamin is transported to the liver in the blood bound to transcobalamin II.

There are only two clinically significant reactions in the body that require vitamin B₁₂ as a cofactor. During the catabolism of fatty acids with an odd number of carbon atoms and the amino acids valine, isoleucine and threonine the resultant propionyl-CoA is converted to succinyl-CoA for oxidation in the TCA cycle. One of the enzymes in this pathway, methylmalonyl-CoA mutase, requires vitamin B₁₂ as a cofactor in the conversion of methylmalonyl-CoA to succinyl-CoA. The 5'-deoxyadenosine derivative of cobalamin is required for this reaction.

The second reaction requiring vitamin B₁₂ catalyzes the conversion of homocysteine to methionine and is catalyzed by methionine synthase. This reaction results in the transfer of the methyl group from N⁵-methyltetrahydrofolate to hydroxycobalamin generating tetrahydrofolate (THF) and methylcobalamin during the process of the conversion.

Clinical Significances of B₁₂ Deficiency

The liver can store up to six years worth of vitamin B₁₂, hence deficiencies in this vitamin are rare. Pernicious anemia is a megaloblastic anemia resulting from vitamin B₁₂ deficiency that develops as a result a lack of intrinsic factor in the stomach leading to malabsorption of the vitamin. The anemia results from impaired DNA synthesis due to a block in purine and thymidine biosynthesis. The block in nucleotide biosynthesis is a consequence of the effect of vitamin B₁₂ on folate metabolism. When vitamin B₁₂ is deficient essentially all of the folate becomes trapped as the N⁵-methylTHF derivative as a result of the loss of functional methionine synthase. This trapping prevents the synthesis of other THF

derivatives required for the purine and thymidine nucleotide biosynthesis pathways.

Neurological complications also are associated with vitamin B₁₂ deficiency and result from a progressive demyelination of nerve cells. The demyelination is thought to result from the increase in methylmalonyl-CoA that result from vitamin B₁₂ deficiency. Methylmalonyl-CoA is a competitive inhibitor of malonyl-CoA in fatty acid biosynthesis as well as being able to substitute for malonyl-CoA in any fatty acid biosynthesis that may occur. Since the myelin sheath is in continual flux the methylmalonyl-CoA-induced inhibition of fatty acid synthesis results in the eventual destruction of the sheath. The incorporation methylmalonyl-CoA into fatty acid biosynthesis results in branched-chain fatty acids being produced that may severely alter the architecture of the normal membrane structure of nerve cells

VITAMIN C

Vitamin C is more commonly known as ascorbic acid. Ascorbic acid is derived from glucose via the uronic acid pathway. The enzyme L-gulonolactone oxidase responsible for the conversion of gulonolactone to ascorbic acid is absent in primates making ascorbic acid required in the diet.

The active form of vitamin C is ascorbate acid itself. The main function of ascorbate is as a reducing agent in a number of different reactions. Vitamin C has the potential to reduce cytochromes a and c of the respiratory chain as well as molecular oxygen. The most important reaction requiring ascorbate as a cofactor is the hydroxylation of proline residues in collagen. Vitamin C is, therefore, required for the maintenance of normal connective tissue as well as for wound healing since synthesis of connective tissue is the first event in wound tissue remodeling. Vitamin C also is necessary for bone remodeling due to the presence of collagen in the organic matrix of bones.

Scurvy results from a dietary vitamin C deficiency and involves the inability to form collagen fibrils properly. This is the result of reduced activity of prolyl hydroxylase, which is vitamin C-dependent. Scurvy leads to lesions in the skin and

blood vessels, and, in its advanced stages, it can lead to grotesque disfiguration and eventual death. Although rare in the modern world, it was a disease well known to sea-faring explorers in earlier times who did not appreciate the importance of fresh fruits and vegetables in the diet.

Hydroxylation of proline residues is catalyzed by prolyl hydroxylase. The reaction requires α -ketoglutarate and ascorbic acid (fig 1).

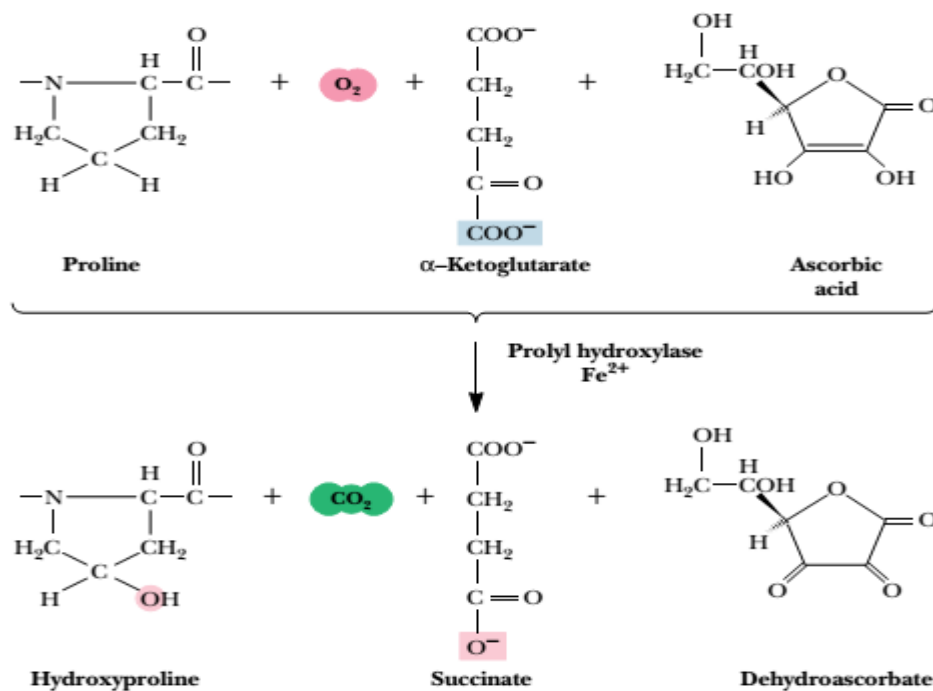


Figure 1. Hydroxylation of proline residues is catalyzed by prolyl hydroxylase. (The reaction requires α -ketoglutarate and ascorbic acid (vitamin C).

Several other metabolic reactions require vitamin C as a cofactor. These include the catabolism of tyrosine and the synthesis of epinephrine from tyrosine and the synthesis of the bile acids. It is also believed that vitamin C is involved in the process of steroidogenesis since the adrenal cortex contains high levels of vitamin C which are depleted upon adrenocorticotrophic hormone (ACTH) stimulation of the gland.

Deficiency in vitamin C leads to the disease **scurvy** due to the role of the vitamin in the post-translational modification of collagens. Scurvy is characterized

by easily bruised skin, muscle fatigue, soft swollen gums, decreased wound healing and hemorrhaging, osteoporosis, and anemia. Vitamin C is readily absorbed and so the primary cause of vitamin C deficiency is poor diet and/or an increased requirement. The primary physiological state leading to an increased requirement for vitamin C is severe stress (or trauma). This is due to a rapid depletion in the adrenal stores of the vitamin. The reason for the decrease in adrenal vitamin C levels is unclear but may be due either to redistribution of the vitamin to areas that need it or an overall increased utilization.

GROUP III. VITAMIN-LIKE SUBSTANCES

Group III is vitamin-like substances. They are separated:

- **fat-soluble:** Coenzyme Q (ubiquinone),
- **water-soluble** vitamins: B₄ (choline), B₈ (inositol), B_T or B₁₁ (carnitine), B₁₃ (orotic acid), B₁₅ (pangamic acid), U (S-methylmethionine), N (lipoic acid).

Most water-soluble vitamins must be supplied regularly with food, as they are quickly removed or destroyed in the body. Fat-soluble vitamins can be deposited in the body. Furthermore, they are poorly excreted, therefore, hypervitaminosis as diseases associated with high doses of fat-soluble vitamin intoxication of organism are observed. Such diseases are described for vitamins A and D.

Choline

Choline appears to be an essential nutrient for a number of animals and microorganisms that cannot synthesize adequate quantities to satisfy their requirements. Choline is a constituent of an important class of lipids called phospholipids, which form structural elements of cell membranes; it is a component of the acetylcholine molecule, which is important in nerve function. Choline also serves as a source of methyl groups ($-CH_3$ groups) that are required in various metabolic processes. The effects of a dietary deficiency of choline itself can be alleviated by other dietary compounds that can be changed into choline. Choline also functions in the transport of fats from the liver; for this reason, it may

be called a lipotropic factor. A deficiency of choline in the rat results in an accumulation of fat in the liver. Choline-deficiency symptoms vary among species; it is not known if choline is an essential nutrient for humans since a dietary deficiency has not been demonstrated.

***Myo*-inositol**

The biological significance of *myo*-inositol has not yet been established with certainty. It is present in large amounts—principally as a constituent of phospholipids—in humans. Inositol is a carbohydrate that closely resembles glucose in structure; inositol can be converted to phytic acid, which is found in grains and forms an insoluble (and thus unabsorbable) calcium salt in the intestines of mammals. Inositol has not been established as an essential nutrient for humans; however, it is a required factor for the growth of some yeasts and fungi.

Para-aminobenzoic acid

Para-aminobenzoic acid (PABA) is required for the growth of several types of microorganisms; however, a dietary requirement by vertebrates has not been shown. The antimicrobial sulfa drugs (sulfanilamide and related compounds) inhibit the growth of bacteria by competing with PABA for a position in a coenzyme that is necessary for bacterial reproduction. Although a structural unit of folic acid, PABA is not considered a vitamin.

Carnitine

Carnitine is essential for the growth of mealworms. The role of carnitine in all organisms is associated with the transfer of fatty acids from the bloodstream to active sites of fatty acid oxidation within muscle cells. Carnitine, therefore, regulates the rate of oxidation of these acids; this function may afford means by which a cell can rapidly shift its metabolic patterns (e.g., from fat synthesis to fat breakdown). Synthesis of carnitine occurs in insects and in higher animals; therefore, it is not considered a true vitamin.

Lipoic acid

Lipoic acid has a coenzyme function similar to that of thiamin. Although it is apparently an essential nutrient for some microorganisms, no deficiency in mammals has been observed; therefore, lipoic acid is not considered a true vitamin.

Bioflavonoids

The bioflavonoids once were thought to prevent scurvy and were designated as vitamin Pc, but additional evidence refuted this claim.

HEALTH EFFECTS OF VITAMINS AND ANTIVITAMINS

Currently, vitamins and antivitamins widely used to prevent and treat a variety of disorders of metabolism. For example:

- vitamin K or menadione, or vicasol (both are synthetic water-soluble analogue of vitamin K) are prescribed to stimulate the synthesis (specifically post-translational γ -carboxylation of glutamic acid residues) such enzymes of coagulation system as factors II (prothrombin), VII, IX and X in the liver. They are usually used after long-term antibiotic treatment (if there is increased bleeding with small injuries, increase in blood clotting time) and in the preoperative period;

- vitamin K antagonist (antivitamin K) dicumarol reduces the efficiency of the blood coagulation promoting blood thinning thereby it use for the treatment of blood clotting diseases, in particular, thrombosis, thrombophlebitis;

- Vitamin A and its derivatives like retinol acetate are used for treating of vitamin A deficiency. For example, they can be administered a patient in order to restore his vision if the patient suffers from vision impairment hemeralopia (night blindness, twilight vision impairment), age-related glaucoma, cataracts etc. Vitamin A drug is also used for skin conditions including acne, eczema, psoriasis, cold sores, wounds, burns, sunburn. It is also used for gastrointestinal ulcers, gum disease, urinary tract infections, diseases of the nervous system;

- drug isoniazid which is antivitamin nicotinic acid and pyridoxine is used In the treatment of patients with pulmonary tuberculosis;

- the structural analogue of vitamin B₂ acrichine is formerly widely used as an antimalarial drug but superseded by chloroquine in recent years. It has also been used as an anthelmintic (in enterobiasis) and in the treatment of giardiasis and malignant effusions. The mechanism action of the drug is based on preventing of microorganism FAD(FMN)-dependent dehydrogenases;

- Ascorutinum is recommended to use as a more effective drug in comparison with ascorbic acid for patients with reduced immunity and frequent colds. Vitamin C (Ascorbic acid) is involved in the hydroxylation of prolyl- and lysyl residues by prolyl 3(4)-hydroxylase and lysyl 5-hydroxylase during collagen synthesis. Effect of the vitamin C is enhanced by vitamin P, which stabilizes the ground substance of fibrous connective tissue in way of hyaluronidase inhibition. Ascorutinum can be recommended in case of bleeding gums, petechial hemorrhages;

- Sulfonamide drugs are folic acid antivitamin. They are structurally resemble paraaminobenzoic acid and due to this similarity it is displaced from its complex with the enzyme synthesizing folic acid. This leads to the inhibition of bacterial growth. This mechanism of action of sulfonamides allows their use as antibacterial agents;

- Pregnant women with a history of several miscarriages is assigned the therapy including α -tocopherol (vitamin E) vitamin supplements using, It contributes to the childbearing. Furthermore, tocopherol acetate, vitamin preparation is usually given in the course of radiation therapy, since this substance has a distinct radioprotective membrane stabilizing action due to its antioxidant activity;

- Derivatives of pyridoxine (vitamin B₆) are used as neurotrophic agents for the correction of mental retardation in childre; in cases of mental disorders in adults; as neuroprotective agents in rehabilitation of patients with stroke and other pathological conditions. The positive effects of pyridoxine is explained by its use as a precursor of PLP that is prosthetic group of the enzyme glutamate

decarboxylase in neurons. The enzyme carries out inhibitory neurotransmitter GABA formation.

- Cabbage and potato juices rich in vitamin U are recommended to drink for patient with duodenal ulcer after the therapy course. Whether taken as a supplement or from foods, vitamin U has been shown to be able to treat a variety of gastrointestinal conditions, including ulcerative colitis, acid reflux, and peptic ulcers. It may also be able to treat skin lesions, improve the symptoms of diabetes, and strengthen the immune system. Some studies show that it can also help prevent liver damage by protecting the organ from the effects of high doses of acetaminophen. Additionally, it may be able to reduce allergies and sensitivities to cigarette smoke and improve cholesterol levels.

The aforementioned examples are only a small part of the use of vitamins and their derivatives in medicine.

Therefore, knowledge of the biochemical basis of vitaminology is of great importance for future doctors.

METHODS USED IN VITAMIN RESEARCH

Determination of vitamin requirements

If a specific factor in food is suspected of being essential for the growth of an organism (either by growth failure or some other clinical symptoms that are alleviated by adding a specific food to the diet) a systematic series of procedures is used to characterize the factor.

The active factor is isolated from specific foods and purified; then its chemical structure is determined, and it is synthesized in the laboratory. Structural determination and synthesis, which may be achieved only after long and intensive research, must be completed before the function and the quantitative requirements of the factor can be established accurately. Established organic and analytical chemical procedures are used to determine the structure of the factor and to synthesize it.

Biological studies may be performed to determine functions, effects of deprivation, and quantitative requirements of the factor in various organisms. The development in an organism of a deficiency either by dietary deprivation of the vitamin or by administration of a specific antagonist or compound that prevents the normal function of the vitamin (antivitamin) often is the method used. The obvious effects (e.g., night blindness, anemia, dermatitis) of the deficiency are noted. Less obvious effects may be discovered after microscopic examination of tissue and bone structures. Changes in concentrations of metabolites or in enzymatic activity in tissues, blood, or excretory products are examined by numerous biochemical techniques. The response of an animal to a specific vitamin of which it has been deprived usually confirms the deficiency symptoms for that vitamin. Effects of deprivation of a vitamin sometimes indicate its general physiological function, as well as its function at the cellular level. Biochemical function often is studied by observing the response of tissue enzymes (removed from a deficient host animal) after a purified vitamin preparation is added. The functions of most of the known vitamins have been reasonably well defined; however, the mechanism of action has not yet been established for some.

The procedure for determining the amount of a vitamin required by an organism is less difficult for microorganisms than for higher forms; in microorganisms, the aim is to establish the smallest amount of a vitamin that produces maximal rate of multiplication of the organisms when it is added to the culture medium. Among vertebrates, particularly humans, a number of procedures are used together to provide estimates of the vitamin requirement. These procedures include determinations of: the amount of a vitamin required to cure a deficiency that has been developed under controlled, standard conditions; the smallest amount required to prevent the appearance of clinical or biochemical symptoms of the deficiency; the amount required to saturate body tissues (i.e., to cause “spillover” of the vitamin in the urine; valid only with the water-soluble vitamins); the amount necessary to produce maximum blood levels of the vitamin plus some tissue storage (applicable only to the fat-soluble vitamins, particularly

vitamin A); the amount required to produce maximum activity of an enzyme system if the vitamin has a coenzyme function; the actual rate of utilization, and hence the requirement, in healthy individuals (as indicated by measuring the excreted breakdown products of radioisotope-labeled vitamins).

The above procedures are practical only with small groups of animals or human subjects and thus are not entirely representative of larger populations of a particular species. A less precise, but more representative, method used among human populations involves comparing levels of dietary intake of a vitamin in a population that shows no deficiency symptoms with levels of intake of the vitamin in a population that reveals clinical or biochemical symptoms. The data for dietary intakes and incidence of deficiency symptoms are obtained by surveys of representative segments of a population.

Determination of vitamin sources

A quantitative analysis of the vitamin content of foodstuffs is important in order to identify dietary sources of specific vitamins (and other nutrients as well). Three methods commonly used to determine vitamin content are described below.

Physicochemical methods

The amount of vitamin in a foodstuff can be established by studying the physical or chemical characteristics of the vitamin—e.g., a chemically reactive group on the vitamin molecule, fluorescence, absorption of light at a wavelength characteristic of the vitamin, or radioisotope dilution techniques. These methods are accurate and can detect very small amounts of the vitamin. Biologically inactive derivatives of several vitamins have been found, however, and may interfere with such determinations; in addition, these procedures also may not distinguish between bound (i.e., unavailable) and available forms of a vitamin in a food.

Microbiological assay

Microbiological assay is applicable only to the B vitamins. The rate of growth of a species of microorganism that requires a vitamin is measured in growth media that contain various known quantities of a foodstuff preparation

containing unknown amounts of the vitamin. The response (measured as rate of growth) to the unknown amounts of vitamin is compared with that obtained from a known quantity of the pure vitamin. Depending on the way in which the food sample was prepared, the procedure may indicate the availability of the vitamin in the food sample to the microorganism.

Animal assay

All of the vitamins, with the exception of vitamin B₁₂, can be estimated by the animal-assay technique. One advantage of this method is that animals respond only to the biologically active forms of the vitamins. On the other hand, many other interfering and complicating factors may arise; therefore, experiments must be rigidly standardized and controlled. Simultaneous estimates usually are made using a pure standard vitamin preparation as a reference and the unknown food whose vitamin content is being sought; each test is repeated using two or more different amounts of both standard and unknown in the assays listed below.

In a growth assay, the rat, chick, dog (used specifically for niacin), and guinea pig (used specifically for vitamin C) usually are used. One criterion used in a vitamin assay is increase in body weight in response to different amounts of a specific vitamin in the diet. There are two types of growth assay. In a prophylactic growth assay, the increase in weight of young animals given different amounts of the vitamin is measured. In a curative growth assay, weight increase is measured in animals first deprived of a vitamin and then given various quantities of it. The curative growth assay tends to provide more consistent results than the prophylactic technique.

In a reaction time assay, an animal is first deprived of a vitamin until a specific deficiency symptom appears; then the animal is given a known amount of a food extract containing the vitamin, and the deficiency symptom disappears within a day or two. The time required for the reappearance of the specific symptoms when the animal again is deprived of the vitamin provides a measure of the amount of vitamin given originally. The graded response assay, which may be prophylactic or curative, depends on a characteristic response that varies in degree

with the vitamin dosage. An example of this technique is an assay for vitamin D in which the measured ash content of a leg bone of a rat or chick is used to reflect the amount of bone calcification that occurred as a result of administration of a specific amount of vitamin D. In an all-or-none assay, the degree of response cannot be measured; an arbitrary level is selected to separate positive responses from negative ones. The percent of positively reacting animals provides a measure of response; i.e., vitamin E can be measured by obtaining the percent of fertility in successfully mated female rats.

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