Ministry of Health of Ukraine
Ukrainian Medical Stomatological Academy

It is approved
on meeting of department of
medical informatics, medical and biological physics
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Methodical instructions
for students' self-preparation work at preparation for a practical lesson
at home and at the classroom

Subject matter	**Medical and biological physics**
The unit	2. Bases of medical physics
Theme of lecture:	**Induced irradiation. Dimensioning of erythrocytes**
Year	1
Faculty	Medical
Speciality	Medicine

Poltava - 2020

The topic significance:

The wave nature of light limits image sharpness (or resolving limit). Due to diffraction it is impossible to resolve a details, which size is less then wave length, therefore it is essential limit of an optical microscopy opportunity. But, as diffraction picture is completely determined by characteristics of those objects, which have created this picture, it can be used for analysis of those characteristics.

Analysis of this picture can give the information, both about size, and about arrangement of these objects in space. Firstly, it is diffraction using for the small objects sizes definition, secondary - for the analysis of object structure (for example, X-ray structure analysis). These methods are widely used in biophysical researches.

Specific targets:

- To have general knowledge of the topic studied;
- To understand, to remember and to use the knowledge received;
- To form the professional experience by reviewing, training and authorizing it;
- Study of diffraction of a laser irradiation on monodisperse particles (erytrocites).
- Measurement of erytrocite diameter.
- To be able to carry out laboratory and experimental work.

Basic knowledge, experience, skills necessary for studying the topic in connection with other subjects:

Disciplines	Obtainable skills
Previous (providing disciplines): physics	To know concepts: electromagnetic field, wave spreading, optic appearances – diffraction, interference, atom energetic levels.
The subsequent disciplines:	To know appearances: atom excitations,

Biochemistry	transmissions between energetic level, spontaneous and forced irradiation, laser structure and principle of operation.

Materials for the before-class self-preparation work:
List of main term, parameters, characteristics, which student have to learn at preparation to class:

Term	Definition
Laser	Device used forced irradiation of light for it amplification
Stimulated emission	It is a process happens if a photon is hit to the electron in an excited state.
Diffraction	Bending of waves around small obstacles and the spreading of waves passed through small openings.

Theoretical questions to class:
1. Describe the laser structure. Explain the laser work principle.
2. The phenomenon of diffraction and it's meaning.
3. What is diffraction grating?
4. The interference phenomenon and it's meaning.

Practice work executed at class:
Erythrocyte size determining with the lazer irradiation diffraction using.
– to put a glass with erythrocytes in a support;
– to measure distance from a glass with erythrocytes up to the screen *L;*
– to switch on the laser and achieve precise a picture of diffraction;
– to measure distance from the diffraction picture center (of a zero maximum) up to first and second minima and first maximum (***l_1, l_2, l_3***);
– determine radius of erythrocytes corresponds to the formulas:

for the first minimum: $r_1 = \frac{0,61 \cdot \lambda \cdot \sqrt{L^2 + l_1^2}}{l_1}$;

for the first maximum: $r_2 = \frac{0,81 \cdot \lambda \cdot \sqrt{L^2 + l_2^2}}{l_2}$;

for the second minimum: $r3 = \frac{1,12 \cdot \lambda \cdot \sqrt{L^2 + l_3^2}}{l_3}$.

- to calculate average value of erythrocyte radius;
- to put results into the first row of the table.
- to change distance *L* between the glass and the screen, repeate experiment, fill next table row.
- To calculate total average value of erythrocyte radius

Table

N	L,m	l_1, m	r_1, m	l_2, m	r_2, m	l_3, m	r_3, m	Average r, m
1								
2								
3								

Make the conclusion.

The contents of the topic:

Installation is optic bench with devices: helium-neon gas laser; blood smear on object-plate disposed in holder; screen; ruler.

Helium-neon laser is source of radiation with wavelength λ = $632{,}8\cdot10^{-9}$ m.

Diffraction method of small object size measurement has high precision, higher than microscopic or many others.

Lasers generate electromagnetic waves with set of specific characteristics: coherence; directionality; monochromaticity; focusability; intensity or irradiance.

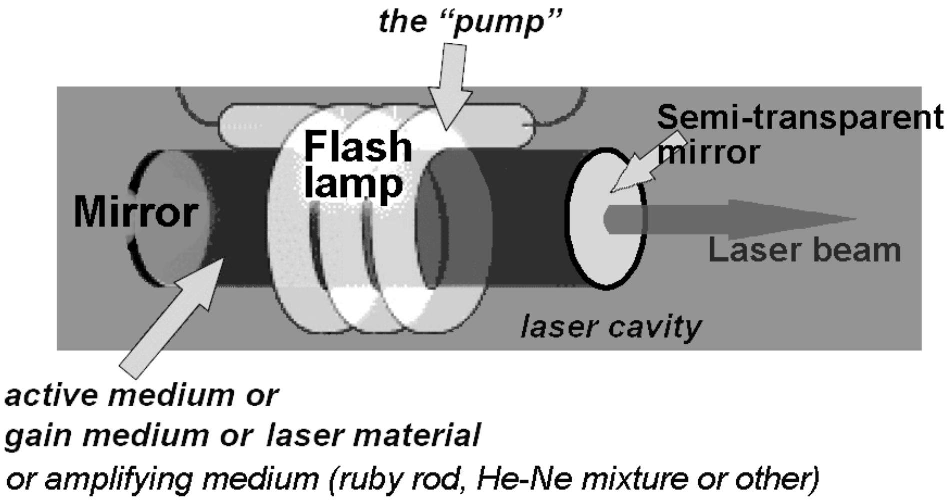

Fig.1. Construction scheme of laser.

There are three radiative transitions that are important in semiconductor lasers and occur between the conduction and valence bands of the material. A schematic diagram of the transitions is shown on fig.3.

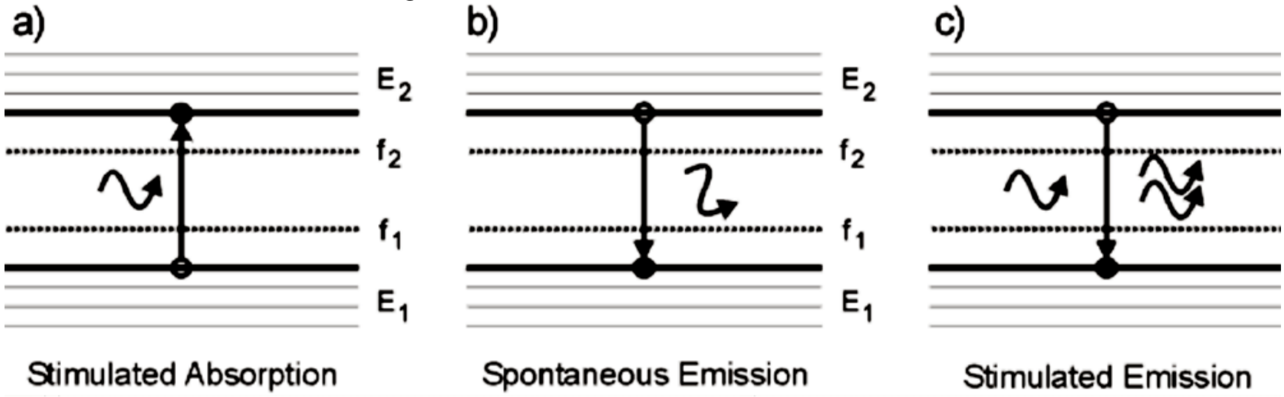

Fig.2. Energy transitions of electrons in laser active medium.

In the first process, an electron in the valence band gains energy by absorbing a photon, exciting it to a higher energy level within the conduction band. The energy gained by the electron is equal to the energy of the photon.

$\hbar\omega = E_2 - E_1$.

The spontaneous emission process begins when an electron is in an excited state in the conduction band. The electron can fall back into the valence band, releasing the excess energy in the form of a photon with an energy given by $\hbar\omega$.

The transition probability of an excited particle falling into a vacant lower state is value A_{21}. It describes spontaneous emission rate by way:

$R_{spon} = A_{21}p_2(1-p_1)$, where p_2 and p_1 are the occupation probabilities of the upper and lower states respectively.

Stimulated emission is a process happens if a photon is hit to the electron in an excited state. It can cause electron to pass to a lower energy level, releasing a photon of the same energy. The emitted photon has the same direction and phase as the incident photon.

When high quantity of medium atoms stay on excited energy level, then spontaneous radiation of some atom can to involve chain radiation reaction. But only waves propagated along rod axis can amplify due to mirrors – fully reflecting of one end and semitransparent on work (radiating) end.

Energetic pumping of medium is realized by electrical discharge (as in helium-neon gas laser), electric current (in semiconductor laser diode), chemical reaction, flashlamp (as in ruby solid-state laser).

Diffraction manifests itself in the apparent bending of waves around small obstacles and the spreading of waves passed through small openings.

The first proof that light had wave characteristics was furnished in 1800 by the experiment on the interference of light from a double slit by Thomas Young (1773-1825). The experiment is now referred to as *Young's double-slit experiment.*

Young's double-slit experiment is shown in fig.3. Light from the source passes through the narrow slit in the first screen and is diffracted. When light from the first slit reaches the second set of slits, it is again diffracted as if there was a point source at each of the secondary slits. The waves from each slit now propagate toward the screen where they interfere, or superimpose, with each other.

Under the Fraunhofer conditions, the light curve of a multiple slit arrangement will be the interference pattern multiplied by the single slit diffraction envelope. This assumes that all the slits are identical.

Where the superimposed waves are in phase on the screen, there is constructive interference and a brightness of image rises.

"In phase" means that difference between the two waves is a whole number of wavelengths.

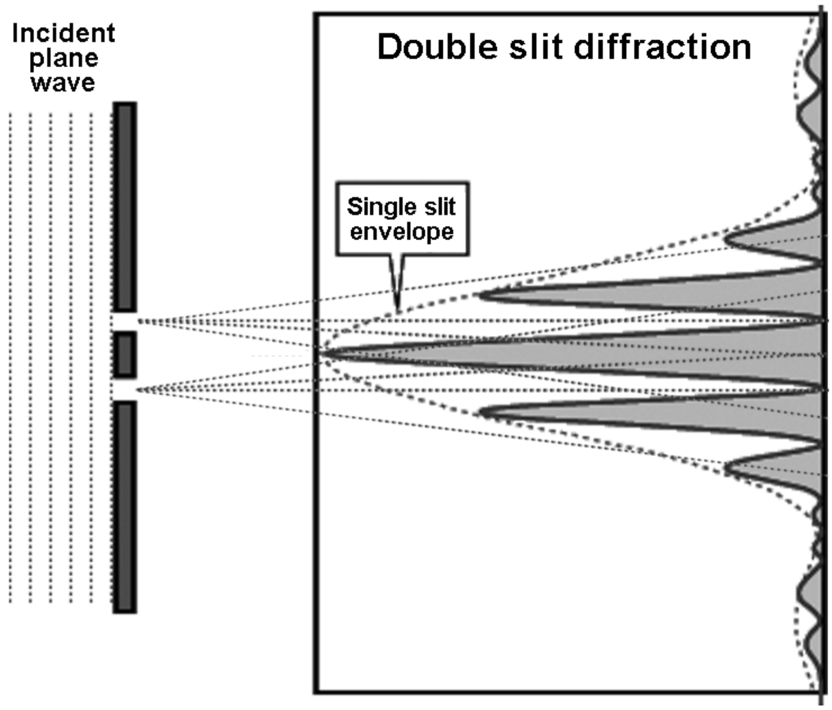

Fig.3. Young's double-slit experiment.

Hence, the observed distribution of light, that is, bright and dark rings, can be explained by the processes of diffraction and interference, both wave manifestations of light.

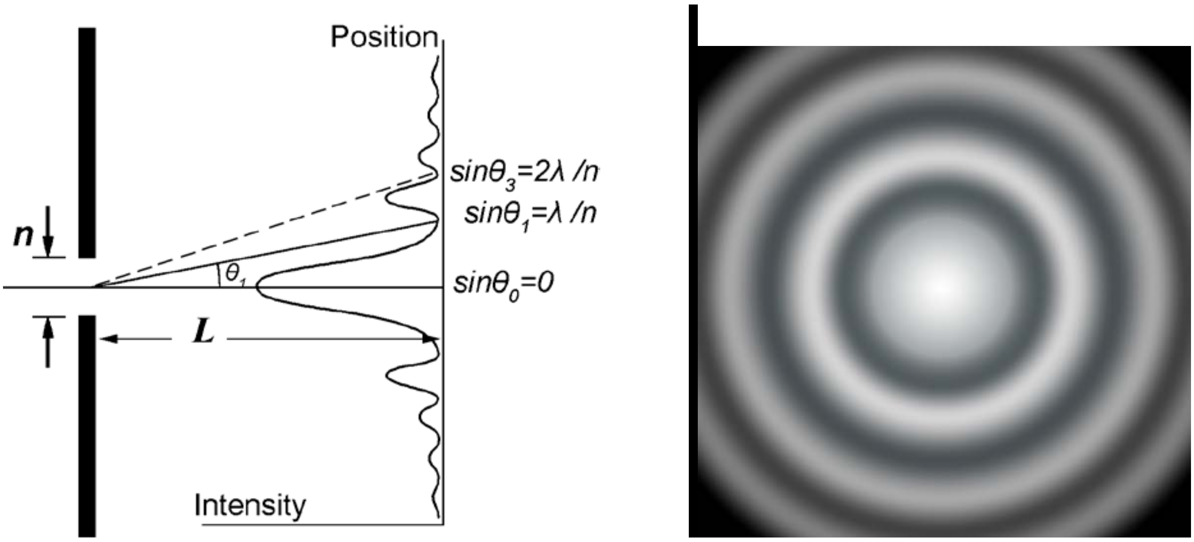

Fig.4. Diffraction picture of small opening.

A large number of parallel, closely spaced slits is a diffraction grating. The condition for maximum intensity is the same as that for the double slit or multiple slits, but with a large number of slits the intensity maximum is very sharp and narrow, providing the resolution for spectroscopic applications.

If there is a need to separate light of different wavelengths with high resolution, then a diffraction grating is most often the tool of choice. This "super prism" property of the diffraction grating leads to application for measuring atomic spectra in both laboratory instruments and telescopes.

The wave properties of light lead to interference, but certain conditions of coherence must be met for these interference effects to be readily visible. Thin films the optical properties of thin films arise from interference and reflection. The basic conditions for interference depend upon whether the reflections involve 180° phase changes.

A diffraction grating is the tool of choice for separating the colors in incident light. The condition for maximum intensity is the same as that for a double slit. However angular separation of the maxima is generally much greater because the slit spacing is o small for a diffraction grating (fig.3).

In experiment monoshromatic laser beam falls on round particle (erythrocyte) with radius ***r***, and forms diffraction pattern on screen placed at distance ***L*** from particle. At relative large distance diffracted rays form beams, which are practically parallel. Diffraction pattern on screen is periodical distribution of brightness in the form of rings – diffraction maxima and minima (fig.4, 5).

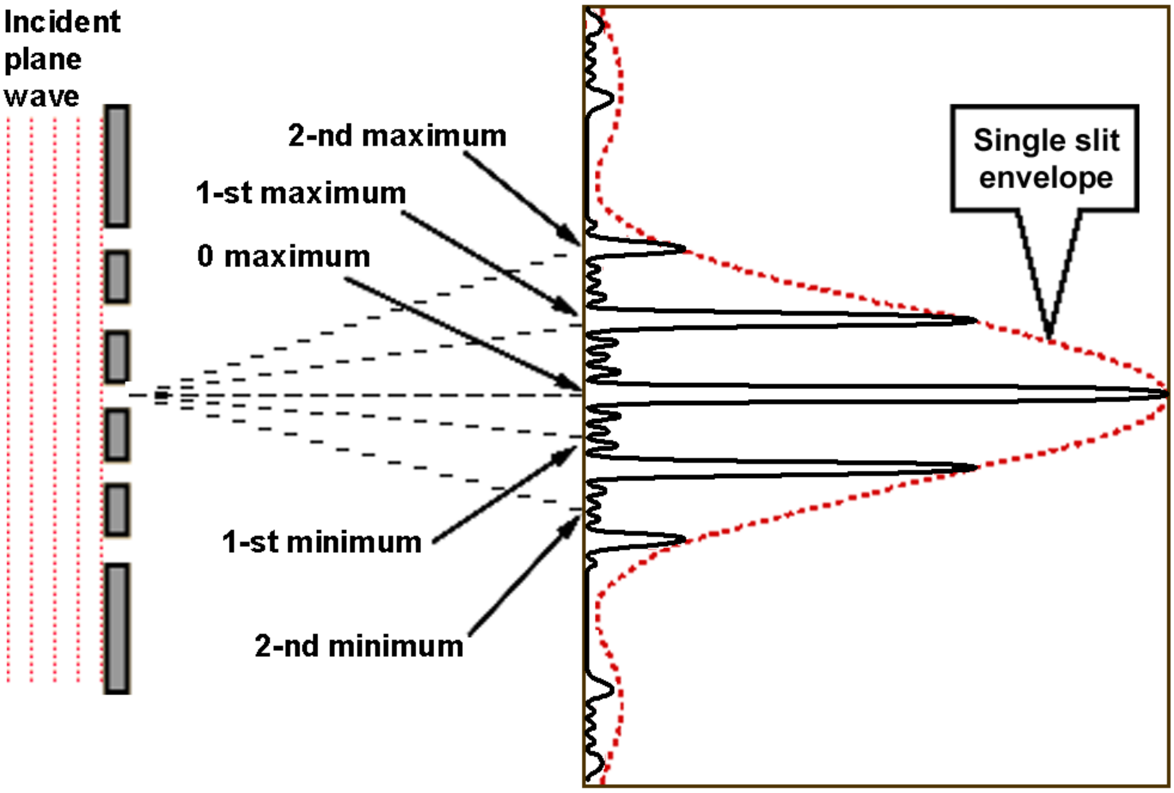

Fig.5. Five slit diffraction. Similar picture will be in case of blood smear.

Conditions of maxima: $r \cdot sin\varphi_0 = 0$,

$r \cdot sin\varphi_2 = 0,81 \cdot \lambda$,

$r \cdot sin\varphi_4 = 1,33 \cdot \lambda$,

Conditions of minima: $r \cdot sin\varphi_1 = 0,61 \cdot \lambda$,

$r \cdot sin\varphi_3 = 1,12 \cdot \lambda$,

$r \cdot sin\varphi_5 = 1,62 \cdot \lambda$,

where φ – angles of ray diffraction, λ – vawe length.

Obtained diffraction pattern of one erythrocyte can be extremely weak an background of direct undiffracted light. In blood smear large quantity of erythrocytes multiply diffraction pattern.

At normal falling of rays on blood smear relation of vawe length, slit size between erythrocytes and angle of rays declination is:

$$n\lambda = ds \cdot sin\varphi,$$

whence

$$d = n\lambda / s \cdot sin\varphi.$$

It gives possibility to calculate diffraction angle by the schematic diffraction pattern (fig.6.):

$$d = \frac{n\lambda \cdot \sqrt{L^2 + l_1^2}}{l_1},$$

whence

$$sin\varphi = \frac{l}{\sqrt{L^2 + l_1^2}}.$$

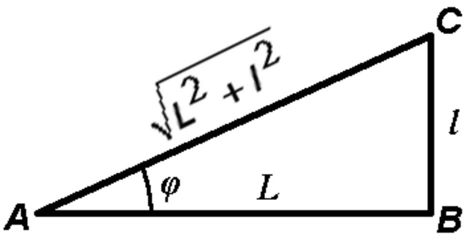

Fig. 6.

Determine radius of erythrocytes corresponds to the formulas:

for the first minimum: $r_1 = \dfrac{0{,}61 \cdot \lambda \cdot \sqrt{L^2 + l_1^2}}{l_1}$;

for the first maximum: $r_2 = \dfrac{0{,}81 \cdot \lambda \cdot \sqrt{L^2 + l_2^2}}{l_2}$;

for the second minimum: $r3 = \dfrac{1{,}12 \cdot \lambda \cdot \sqrt{L^2 + l_3^2}}{l_3}$;

for the second maximum: $r_4 = \dfrac{1{,}33 \cdot \lambda \cdot \sqrt{L^2 + l_4^2}}{l4}$;

for the third minimum: $r_5 = \dfrac{1{,}62 \cdot \lambda \cdot \sqrt{L^2 + l_5^2}}{l5}$.

Self-control material:

B. Test tasks

***1*. What device used in Young's double-slit experiment?**

a) diffraction grating;
b) observation slit;
c) double-slit;
d) diffraction cell.

2. When is maximum of brightness observed after diffraction of laser irradiation on the thin layer of small objects?

a) in the center of screen;
b) in the first maximum ring;
c) in the second maximum ring.

3. What phenomenon observed in Young's double-slit experiment?

a) reflection;
b) refraction;
c) diffraction;
d) interference.

4. What is the difference between coherent and incoherent light?

a) coherent light have only one frequency waves;
b) incoherent light have only one frequency waves;
c) coherent light have coincident phases of waves;
d) incoherent light have coincident phases of waves;
e) coherent light have less biological effect than incoherent light at same power.

5. Does a fluorescent light lamp produce coherent or incoherent light?

a) yes;
b) only temporally coherent;
c) only spatially coherent;

d) no.

Literature recommended

Main sources.

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