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# ELECTROLYTIC AGGREGATION IN SOLUTIONS WITH QUANTUM DOTS AND GOLD NANOPARTICLES MODIFIED WITH OLIGONUCLEOTIDES

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*Aim*. To investigate electrolytic aggregation of different nano-objects in solutions with quantum dots (QDs) and Au nanoparticles (NPs) modified by oligonucleotides as well as the effect of aggregates on the photoluminescence (PL) of QDs.

*Methods*. Au NPs and AgInS2/ZnS QDs were modified by oligonucleotides. Two types of QDs that differ in size and stabilizing ligand were used. PL and optical absorption of nano-objects in water and SSC buffer solutions were studied.

*Results*. The transfer of modified by oligonucleotides QDs from water to a buffer solution and the addition of Au NP modified by oligonucleotides to the solution caused quenching of the QD PL intensity. The PL quenching was observed for the QDs of two types and increased during the incubation of solutions, but didn't depend on its multiplicity. An aggregation of Au-DP occurred only in buffer solutions with QDs of one type and increased with multiplicity of the buffer solution.

*Conclusion*. It is found that the electrolytic aggregation of Au NPs modified by oligonucleotides in buffer solutions with QDs depends on the QD type and didn't affect the quenching of the PL intensity of the QDs.

***Key words:*** quantum dots, Au nanoparticles, photoluminescence, electrolytic aggregation.

Recently, gold nanoparticles (Au NPs) have become an important component widely used in optical bio-sensors due to their unique chemical and physical properties, as well as high biocompatibility [1]. In particular, when Au NPs are irradiated with visible light, the phenomenon of surface plasmon resonance (SPR) can be observed. It is caused by the resonant excitation of oscillations of free electrons on the surface of Au NPs (surface plasmon) when absorbing the energy of an incident electromagnetic wave [2]. In the optical absorption spectra of Au NPs, an SPR peak is observed. The intensity, spectral position, and half-width of SPR peak depend on the size and shape of the nanoparticle, as well as on its local dielectric environment. The interaction between metal NPs and luminescent semiconductor crystals of nanometer size, the so-called quantum dots (QDs), attracts considerable attention of researchers due to wide range of their potential applications extending from sensors to quantum information processing [3]. This interaction can lead to the enhancement of the QD photoluminescence (PL) intensity as well as to its reduction. PL enhancement occurs due to the excitation of surface plasmons in metal NPs and energy transfer to QDs [4], and quenching of PL is caused by the non-radiative transfer of energy from QDs to NPs [5]. In the last case, the so-called fluorescence (Foster) resonance energy transfer (FRET) takes place — the process of energy transfer from a fluorescent donor to a lower energy acceptor through a distance-dependent dipole-dipole

interaction [6, 7]. It is believed that quenching of QD PL in the presence of Au NP is an effect that acts at a short distance and weakens with distance much faster than the electromagnetic field of the SPR [4]. In the vast majority of fluorescent biosensors, the same effect of PL quenching by Au NPs is used, and the detection of oligonucleotides in vitro or in vivo occurs by recording the change in the PL intensity of QDs during formation the QD-Au NP conjugate or its decay [3]. However, it is known that Au NPs are inherent in electrolytic aggregation in salt buffer solutions [8], which can affect the PL of QDs. Therefore, in this paper the effect of electrolytic aggregation of Au NPs on the optical characteristics of QDs modified by thiolated oligonucleotides was investigated.

## Materials and Methods

In this paper, water-soluble Au NPs with a diameter of 13 nm, stabilized by citrate, and aqueous solutions of QDs of two types: (1) QD-88 — a fraction of $AgInS_2/ZnS$ QDs with a diameter of ≈3 nm, stabilized with mercaptoacetic acid, (2) QD-92 – unfractionated preparation with QD $AgInS_2/ZnS$ with a diameter of 2–3 nm, stabilized with glutathione were used. In both types, the stabilizing ligands formed a negative charge on the QD surface. QDs and Au NPs were modified by oligonucleotides. The 23-base oligonucleotide GCTGAAGGGCTTTTGAACT (hereafter MP-SH), in which a sequence of four thymidines, 6 methylene groups, and a sulfhydryl group covalently connected to the 3′-end, was used for QD modification; and the 26-base oligonucleotide TGGCTGAGTGGACGATGA (hereinafter HS-DP), in which a sequence with eight thymidines, 6 methylene groups, and a sulfhydryl group covalently connected to the 5′-end, was used to modify Au NPs. The surface of Au NPs was additionally covered with lipoic acid (LA) and mercaptohexanol (MCH). Citrate on the surface of Au NPs is replaced by sulphurous compounds (LA, MCH, HS-DP), what makes the NPs more stable in a saline solution, preserving their water-soluble properties, and creating a negative charge on their surface [8, 9]. All solutions were prepared in deionized water and in SSC buffer solutions with different multiplicities (0.1×SSC, 0.25×SSC, 0.5×SSC).

PL spectra were excited by the 411 nm line of a continuous laser and recorded by a BLACK-Comet C-SR-50 spectrometer. PL intensity was also measured at a fixed wavelength of ~590 nm using a scanning spectrofluorometer “Synergy HT”. Optical absorption spectra were recorded with a NanoDrop 2000 microvolume spectrophotometer, and optical absorption intensity at fixed wavelengths (~540 nm, ~620 nm, ~690 nm) was measured using a TitertekMultiskan MCC/340 photometric reader.

## Results and Discussion

In the PL spectra of aqueous solutions of QDs of both types (Fig. 1, *a*, *b*), a broad PL band is observed in the yellow-red spectral region. The band is due to emission of defects in QDs [10]. QD88 are characterized by a longer wavelength position of the PL band maximum (~630 nm) compared to QD92 (~587 nm), which is consistent with bigger QD

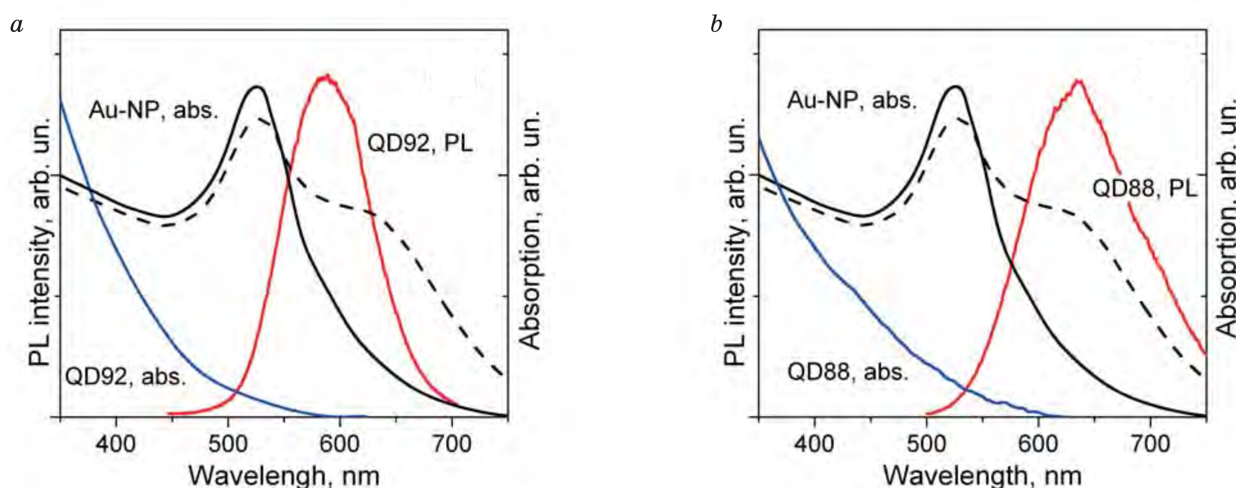


*Fig. 1.* **Optical absorption and photoluminescence spectra of QD92 (*a*) and QD88 (*b*) in water and optical absorption spectra of Au NPs in water (solid lines) and partially aggregated Au NPs (dashed lines)**

size in the case of QD88. The optical absorption spectra of QD solutions show an edge extended into the long-wavelength region without a well-defined exciton absorption peak. The extended edge in the QD absorption spectra of $A_1A_3B_6$ compounds is usually explained by scatter of QDs in size and shape, and/or optical transitions via the levels of defects in the QD band gap [11]. In the absorption spectra of solutions with Au NPs, a clear SPR maximum at ~510 nm is observed (Fig. 1). In the case of Au NP aggregation, the intensity of the peak at 510 nm decreased and a new peak at 640 nm appeared, due to SPR in the aggregated NPs.

Modification of the QDs with MP-SH oligonucleotides didn't change the spectral position of the PL peak and led to a slight increase of PL intensity in the case of QD88. At the same time, transfer of QDs modified with oligonucleotides (MP-QD) into a buffer solution resulted in a slight decrease in PL intensity (Fig. 2). It turned out that the addition of Au NPs modified with oligonucleotides (Au-DP) to the solutions with MP-QD88 or MP-QD92 also results in a decrease in the PL intensity of QD. The degree of PL intensity quenching with Au NPs depended on the type of QD solutions and increased with an increase of incubation time (Fig. 2).

The PL intensity decreased in all QD solutions after 24 h of incubation compared to 1 hour in the same solution. In particular, for MP-QD88 the PL intensity of solutions without Au-DP (Fig. 2a,c) decreased by 11%, 22.0%, 29.6% and 27.8% in water, 0.1×, 0.25× and 0.5×SSC, respectively; and for solutions with Au-DP by 27.9%, 43.0%, 39.5% and 40.7%, respectively. So, the decrease of MP-QD88 PL occurs more intensively in the presence of Au-DP. Similarly, the PL decrease in MP-QD92 solutions (Fig. 2, *b, d*) was also more noticeable in the presence of Au-DP, although the absolute indexes were smaller. In particular, the PL intensity of MP-QD92 without Au-DP

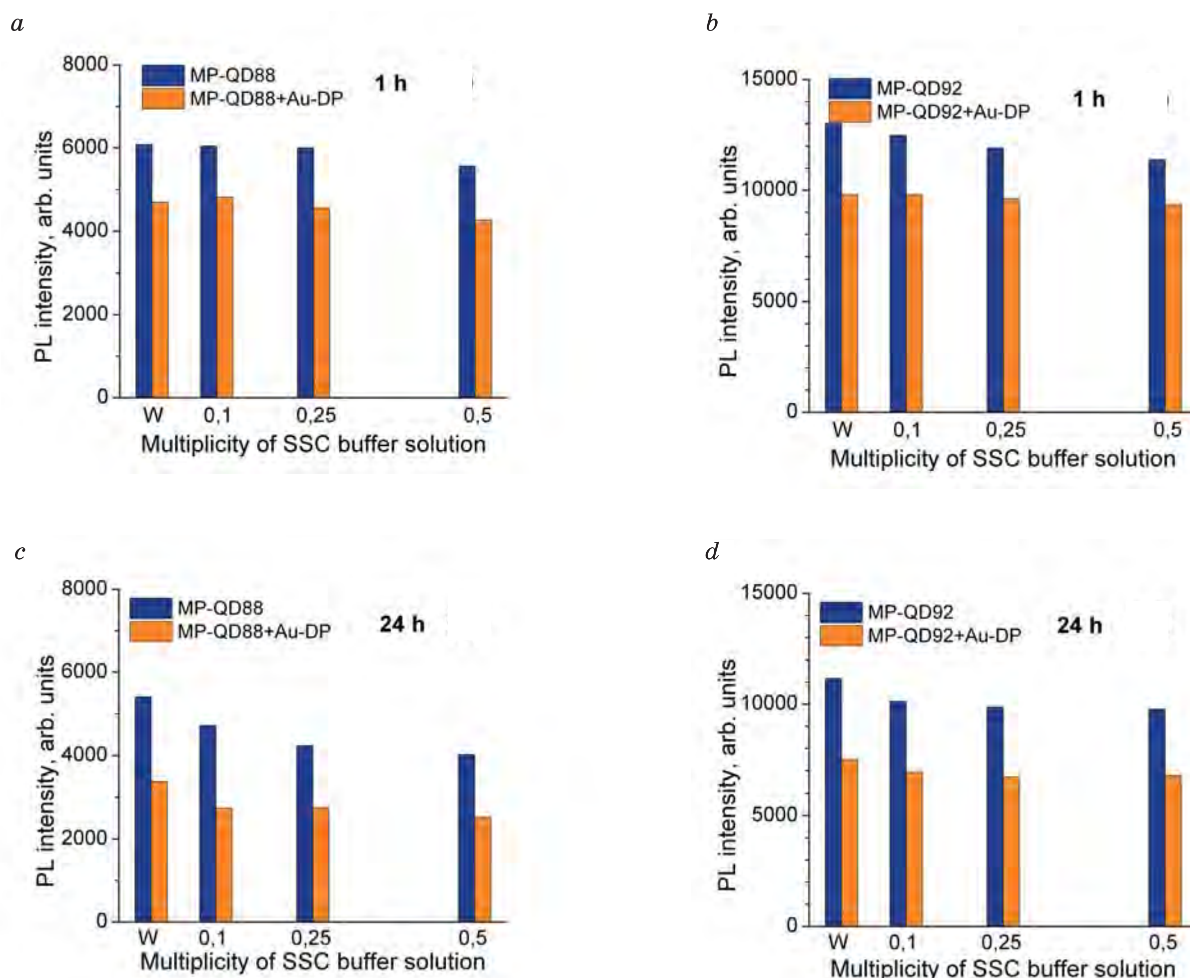


*Fig. 2.* **PL intensity of mixture of MP-QD88 (*a, c*) and MP-QD92 (*b, d*) with Au-DP in water (w) and in SSC buffer solutions of different multiplicity (0.1, 0.25, 0.5) detected 1 h (*a, b*) and 24 h (*c, d*) after preparation**

decreases in water and 0.1×, 0.25× and 0.5× SSC by 14.3%, 18.9%, 17.2% and 14.1%, respectively, and in the presence of Au-DP by 23.3%, 29.2%, 30.2% and 27.5%, respectively. As can be seen, the decrease in PL intensity during 24 h for a mixture of QDs and Au NPs is bigger in buffer solutions as compared to water. At the same time, there is no clear dependence of the degree of PL quenching the on the multiplicity of the buffer solution.

To detect the electrolytic aggregation of Au NPs and to study its influence on the PL of QDs, the optical absorption of QD and Au NP solutions was measured at the wavelengths corresponding to the SPR peak of unaggregated (~540 nm) and aggregated Au NPs (~620 and 690 nm) (Fig. 3 ). It has been turned out that the formation of aggregates depends on the type of QDs and the multiplicity of the buffer solution. In particular, in samples with MP-QD88 and Au-DP, an increase in absorbance at 620 nm and 690 nm, due to the appearance of Au NPs aggregates, was observed in the solutions with a multiplicity of 0.25× and 0.5×SSC one hour after the preparation and continued to increase during the 24 h of incubation (Fig. 3, *a, c*). Aggregation of Au NPs was not observed in water and in 0.1×SSC solution. Therefore, for MP-QD88, an increase in the concentration of the buffer solution promotes formation of Au NP aggregates. At the same time, in all solutions with MP-QD92 and Au-DP, the aggregation of Au NPs during 24 h was not recorded (Fig. 3, *b, d*).

As the analysis of the obtained results shows, in the buffer solutions of 0.5×SSC an obvious aggregation of Au NPs takes place for MP-QD88 and no sign of aggregation is found for MP-QD92. At the same time, for both types of QDs, the PL intensity decreases during 24 h. Similarly, for MP-QD88, a comparable decrease in PL intensity occurs in all buffer solutions, although aggregation is recorded

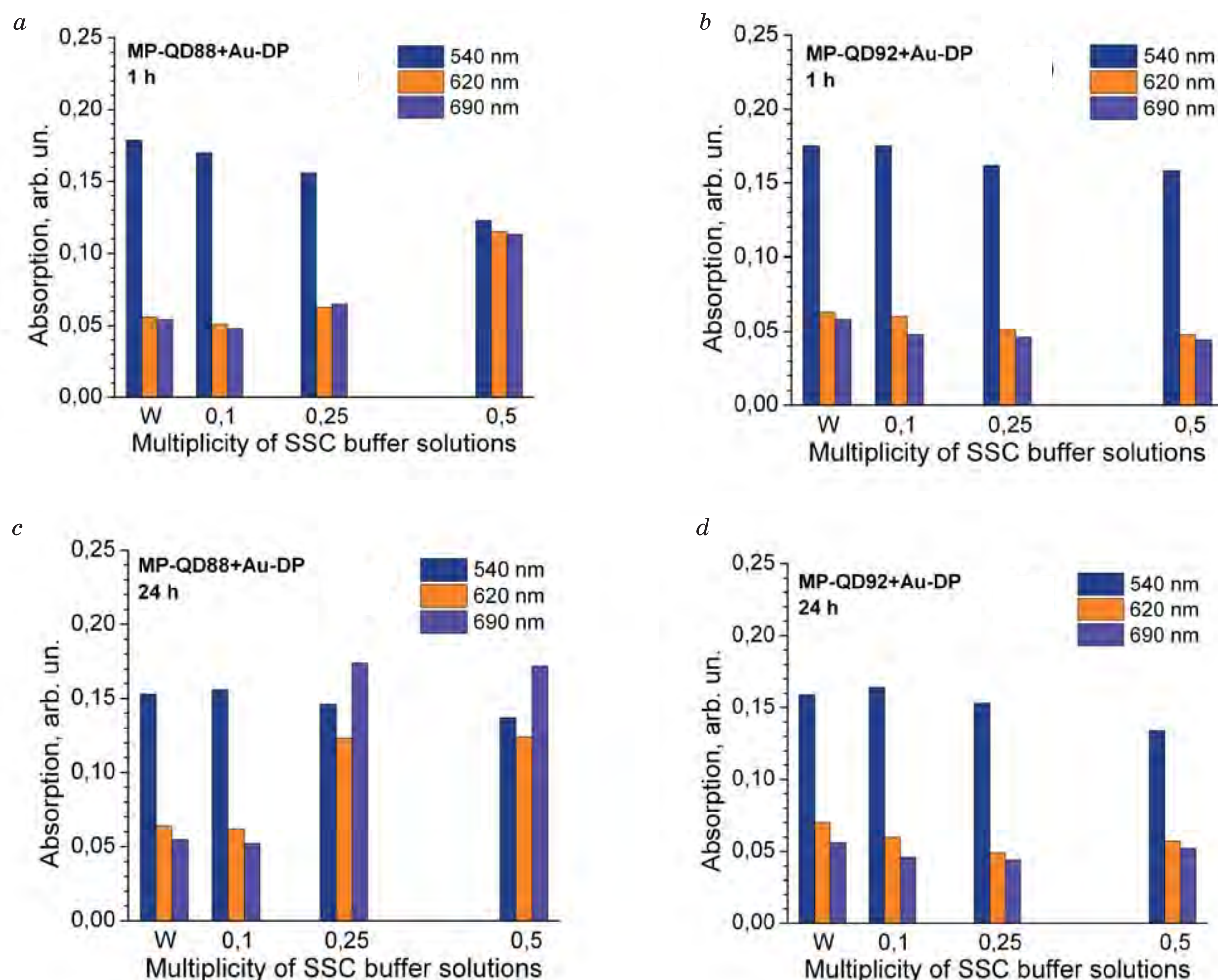


*Fig. 3.* **Optical absorption of MP-QD88 (*a, c*) and MP-QD92 (*b, d*) with Au-DP in water (W) and in SSC buffer solutions of different multiplicity 1 h (*a, b*) and 24 h (*c, d*) after preparation**

only for solutions with a multiplicity of 0.25× and 0.5×SSC. It can be concluded that the formation of electrolytic aggregates of Au NPs in solutions of QDs and Au NPs modified by oligonucleotides does not affect the decrease of PL intensity.

## Conclusion

It is found that the transfer of modified by oligonucleotides $AgInS_2/ZnS$ QDs from water to SSC buffer solution results in a decrease in PL intensity. The addition of Au NP modified by oligonucleotides to QD buffer solutions also causes a decrease in the PL intensity of QDs, but the magnitude of the effect is larger. It is found that the degree of PL intensity quenching depends on the type of QDs' stabilizing ligand (glutathione and mercaptocytic acid) and increases with the incubation time of the solutions from 1 h to 24 h. At the same time, the multiplicity of the buffer solution in the range from 0.1 to 0.5 has weak effect on the change in PL intensity.

It is found that the electrolytic aggregation of Au NPs modified by oligonucleotides in buffer solutions with QDs depends on the QD type. In particular, no signs of aggregation of Au NPs were observed within 24 h in buffer solutions with QDs (∅~2-3 nm) stabilized with glutathione and modified by oligonucleotides. At the same time, the aggregation of Au NPs in solutions with QDs (∅~3 nm) stabilized by mercaptoacetic acid and modified by oligonucleotides, was observed already 1 h after solution incubation. In this case, the degree of aggregation increased with the increase of the concentration (multiplicity) of buffer solution. It is found that the changes in the aggregate state of modified by oligonucleotides Au NPs in SSC buffer solution do not affect the decrease in PL intensity of QDs.

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