Highly Sensitive Detection for Proteins Using Graphene Oxide-aptamer Based Sensors

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The detection of proteins by using bare graphene oxide (GO) to quench the fluorescence of fluorescein-labeled protein binding aptamers have been reported.[1-3] However, the proteins can be adsorbed on the surface of bare GO to prevent the sensitivity of detection from further being improved. This nonspecific adsorption has seldom been taken into account in previous reports. In order to solve this problem, we described a detection method for proteins using thrombin as an example in this study. Fig. S1 showed the change of fluorescence intensity in the presence of FAM-modified aptamer under the different conditions. The fluorescence intensity in the presence of FAM-modified aptamer showed strong fluorescence intensity (Fig. S1, curve a). After GO was added into the FAM-modified aptamer solution, most of the fluorescence was quenched (Fig. S1, curve b). It showed GO had the high quenching efficiency for FAM. FAM-modified aptamer in the GO solution showed significant fluorescence restoration with the addition of 20 nM thrombin (Fig. S1, curve c). In our study, we applied PEG to coat the surface of GO. Before this, optimizing the conditions of GO-aptamer based sensor were indispensable. Fig. S2 showed different fluorescence intensities of FAM-modified aptamer with the change of GO concentrations. The fluorescence intensity of FAM-modified aptamer trended to a minimum value at 40 μg/mL. Therefore, 40 μg /mL GO was used for the following experiments. Fig. S3 showed the fluorescence quenching of FAM-modified aptamer in the presence of GO. The intensity of fluorescence was fast decreased after the FAM-modified aptamer was adsorbed on the surface of GO. It reached the equilibrium in 30 min. The release of the thrombin–aptamer complex from GO also needs 30 min (Fig. S3). Fig. S4 displayed the fluorescence response of the GO-based aptamer with thrombin alone and further with Guanidine·HCl. The result showed this assay rapidly reacted with the molecules that can denature thrombin. Fig. S5 showed the GO-aptamer based sensor exhibited smaller fluorescence increase to the nonspecific binding of other proteins compared with thrombin. This demonstrated that GO-aptamer based sensor responded selectively toward thrombin over other tested proteins.

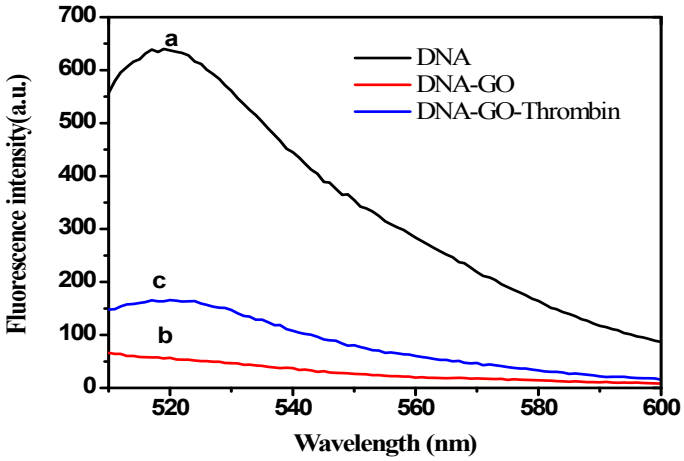

Fig. S1 The fluorescence intensity of FAM-modified aptamer (20 nM) under the following conditions: (a) FMA-modified aptamer in the solution; (b) FAM-modified aptamer + GO (40 μg/ml); and (c) FMA-modified aptamer+GO (40 μg/ml)+ thrombin (20 nM).

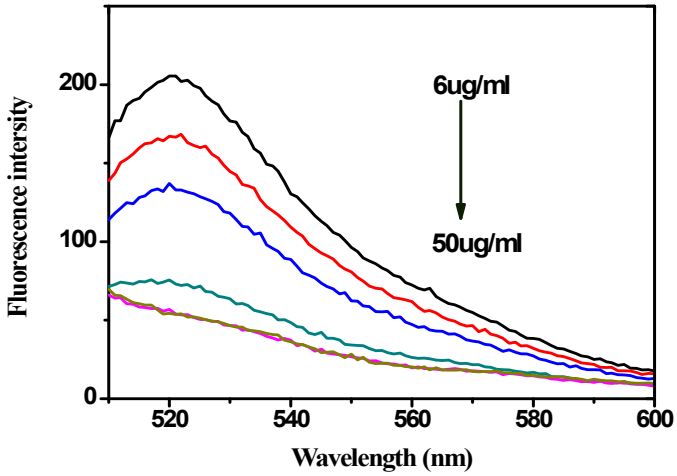

Fig. S2 The fluorescence intensity of FAM-modified aptamer (20 nM) in the presence of various concentrations of GO (6, 10, 20, 30, 40, 50 μg/mL).

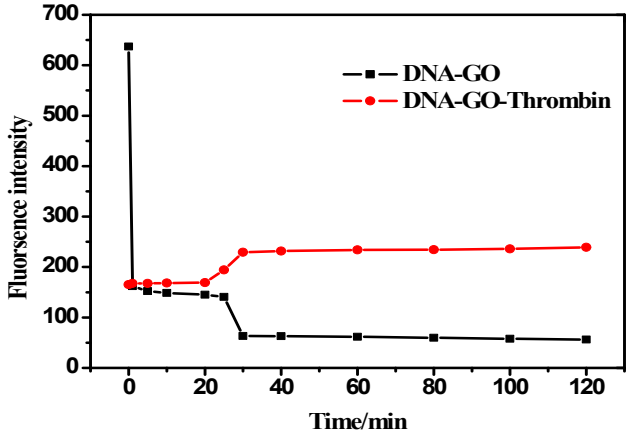

Fig. S3 The fluorescence intensity of FAM-modified aptamer (20 nM) under the following conditions: (a) in GO (40 μg/mL) with the change of time; and (b) in GO (40 μg/mL) by thrombin (20 nM) with the change of time.

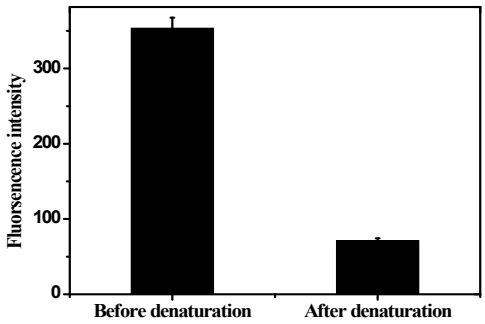

Fig. S4 The change of fluorescence intensity upon sequentially alternating treatment with thrombin (60 nM) and Guanidine·HCl (6 M).

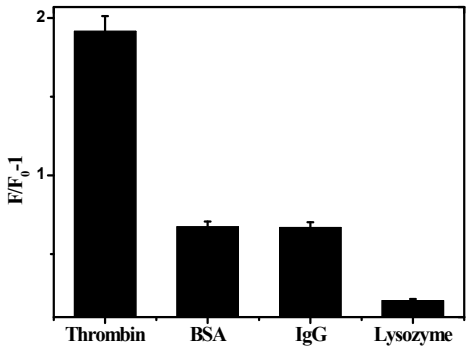

Fig. S5 The fluorescence intensity of FAM-modified aptamer (20 nM)-GO (40 ug/mL) in the presence of other proteins (BSA, Ig G, and lysozyme) at the concentrations of 20 nM.

References

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