Glutamate decarboxylase-derived IDDM autoantigens displayed on self-assembled protein nanoparticles

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The recombinant protein nanoparticle (rPNP) formed self-assembled spherical supramolecules with the size comparable to native one. We tried to express the GAD65 C-terminal fragments, i.e., 448-585 ($GAD65_{448-585}$), 487-585 ($GAD65_{487-585}$), and 512-585($GAD65_{512-585}$) amino acid fragments, using PNP as N-terminus fusion expression partner in E.coli. All of recombinant fusion proteins also formed nanoparticles due probably to the self-assembly function of the fused rPNP. The antigenic epitopes within $GAD65_{448-585}$, $GAD65_{487-585}$, and $GAD65_{512-585}$ against insulin-dependent diabetes mellitus marker were localized at the surface of the spherical PNP so that anti-GAD65 Ab could recognize them. The PNP seem to provide advantages over other inorganic nanoparticles through the bacterial synthesis, the active capture probes can be located at the PNP with constant orientation/conformation via covalent cross-linking. Also the PNP have uniform particle size. Thus, the rPNP can be used as a three-dimensional and nanometer-scale probe structure that is a key component in ultra-sensitive protein chip for detecting protein-small molecule interactions and protein-protein interactions.