

Abstract:

Nanodiamonds represent a member of vast group of carbon nanomaterials. They are characterized by chemical stability, biocompatibility, low toxicity and possibility of surface chemical modification that enables further conjugation of biomolecules, namely proteins. The most important property of nanodiamonds is easily detectable fluorescence resistant towards photobleaching. This luminescence is produced by so called (N-V) centers that can be formed by irradiation of nanodiamonds using high energy beams. Fluorescence of nanodiamonds is useful for construction of fluorescent labels and probes, cellular targeting and internalization, controlled drug delivery as well as enzyme immobilization. Two main connection modes are suitable for biomolecules' attachment: non-covalent and covalent. The first one has been utilized for decoration of nanodiamonds by various proteins, e.g. lysozyme, cytochrome c, neurotoxin or antigen. The second possibility has been demonstrated also on various proteins and, furthermore, on glycoproteins, oligonucleotides, vitamins or growth factor. During the immobilization of biomolecule on the nanoparticle surface it is crucial to keep its function, for example the catalytic activity in enzymes. For such purposes, the oxime ligation (connection of aldehyde presented on biomolecule and aminoxy group bound on nanodiamond surface) represent a convenient pathway, because of reasonable effectivity of reaction at mild conditions and well defined way of attachment. In this bachelor thesis, the spectrophotometric procedure for aminoxy group quantitation on nanodiamonds using pyridoxal-5'-phosphate was developed. The conjugation of model proteins by covalent and also by non-covalent bonds is further demonstrated.

(In czech)