Molecular features of interaction between VEGF-A and anti-angiogenic molecules: a biophysical computational study

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Background: Anti-VEGF agents are used to inhibit primary tumor and metastasis growth [1], both in the adjuvant, and more recently, in neoadjuvant settings [2]. Recently, anti-angiogenic agents have been used to treat ocular pathological conditions such as age-related macular degeneration (AMD) and diabetic macular edema (DME) [3, 4]. Diabetic retinopathy is the leading cause of vision loss of working-age adults, and DME is the most frequent cause of vision loss related to diabetes. AMD is a progressive neurodegenerative and multifactorial disease that impairs the visual field. Currently, three VEGF inhibitors are commonly used to treat the retinal disorders characterized by neovessels formation: ranibizumab (Lucentis), aflibercept (Eylea) and bevacizumab (Avastin), this latter used as off-label.
Purpose: To analyze at atomistic level the energetic features of well characterized anti-angiogenic agents by means of computational approaches.
Methods: We carried out molecular modeling of aflibercept binding domain (VEGFR1d2_R2d3). We further modeled loops of ranibizumab and Fab-bevacizumab. Anti-VEGFA/VEGFA complexes have been predicted through protein-protein docking simulations by means of Pydock software. Three replicas of all-atom molecular dynamics simulations of VEGFR1d2_R2d3, ranibizumab and Fab-bevacizumab and corresponding complexes bound to VEGFA were carried out by means of GROMACS software. Binding energies of anti-angiogenic/VEGFA complexes have been predicted with MM-PBSA calculations through usage of the g_mmpbsa tool.
Results: Protein-protein binding is strongly influenced by ionic strength, thus it is reported that greater electrostatic stabilization energy of a complex is associated to faster K_{on} [5]. Papadopoulos et al. (2012) [6] reported a higher K_{on}, upon binding with VEGFA for aflibercept compared to ranibizumab and bevacizumab. We confirmed that VEGFR1d2_R2d3/VEGFA is stabilized by electrostatic energy. Furthermore, hydrophobic effect is one of the driving forces of protein-protein binding. Apolar desolvation energy accounts to hydrophobic effect, indeed, we found that the apolar contribution to desolvation energy was correlated (R^2 =0.81, p=0.001) with experimental KD of analyzed complexes. The low experimental K_{off} of ranibizumab has been explained with higher number of stable contacts, H-bonds and lower conformational fluctuation compared to VEGFR1d2_R2d3 and Fab-bevacizumab bound to VEGFA. Data about aflibercept binding domain bound to PlGF1 were included in the analysis. The g_mmpbsa tool helped the quantitative evaluation of contribution of residues at protein interface, 'hot-spots' [7, 8].
Conclusions: Molecular modeling approaches are feasible to evaluate binding features of VEGFA in complex with binding domains of anti-angiogenic drugs; in general the approach hereby used would help the characterization of protein-protein complexes. Furthermore, this validated approach may be useful to develop more effective anti-VEGF agents.

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