

**NOVEL BIOLOGICAL ACTIVITIES OF THE E-PEPTIDE
OF FISH PRO-IGF-I**

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EXTENDED ABSTRACT ONLY- DO NOT CITE

E-peptides of pro-insulin-like growth factor-I (pro-IGF-I) are proteolytically cleaved from the pro-hormone after translation, and have long been regarded as biologically inactive. Tian *et al.* (1999) recently demonstrated that recombinant rainbow trout pro-IGF-I E-peptides (rtEa-2-, rtEa-3- and rtEa-4-peptide), like hIGF-I, exhibited a dose-dependent mitogenic activity in several non-transformed mammalian cell lines.

We showed recently that treatment of established human and fish cancer cells (MCF-7; HT-29; HepG2, ZR-75-1, SK-N-F1 and HC) and retroviral transformed human embryonic kidney cells (293GP) with recombinant rtEa-2- or rtEa-4-, but not rtEa-3-peptide, resulted in a dose-dependent induction of morphological change and enhanced cell attachment Chen *et al.*, 2002).

The Ea-4-peptide-induced morphological changes were sensitive to treatment with α -amanitin or cycloheximide, known inhibitors of RNA and protein synthesis. The *in vitro* colony formation activity of established human tumor cells (HT-29 and MDA-MB-231) was greatly reduced or diminished by treatment with the rtEa-4-peptide.

Both morphological change and reduction of colony formation activity in MDA-MB-231 cells were also observed following transfection with an Ea-4 transgene

construct. Furthermore, the invasive activity of HT1080 cells, known invasive cancer cells, was reduced 3-4 fold by treatment with the rEa-4-peptide.

Results of transgenic studies revealed that transfer of a transgene expressing rEa4-peptide into newly fertilized eggs of medaka resulted development of heart and blood vessel abnormality. These results suggest that rEa4 might possess antiangiogenic activity. The antiangiogenic activity of rEa4-peptide was further confirmed by an assay conducted on chicken Chorio-Allantoic Membrane (CAM).

As shown in Figures 1 and 2, rEa4-peptide caused a dose-response dependent reduction of blood vessel density development on the chicken CAM. These results suggest that Ea-4-peptide of rainbow trout pro-IGF-I possesses novel biological activities controlling malignant properties of cancer cells *in vitro*. These novel biological activities include: mitogenic activity in NIH3T3 cells, induction of morphological changes in cancer cells, reduction of colony formation and invasive activities and antiangiogenic activity

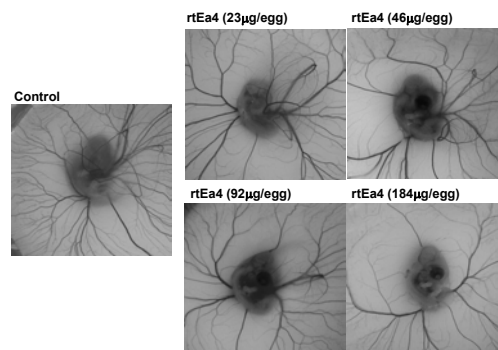


Figure 1: Effect of rEa4-peptide on blood vessel density of the Chorio-Allatoic Membrane of chicken embryos. Day 3 chicken embryos were placed in egg-cups, treated with various amounts of rEa4-peptide, and then incubated at 37°C for an additional four days. At the end of incubation, the embryos were photographed with a digital camera.

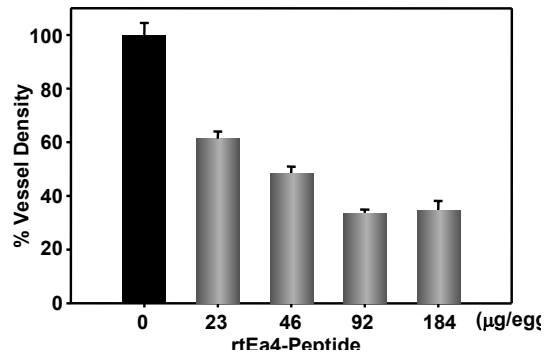


Figure 2: Dose-dependent effect of rtEa4-peptide on blood vessel density development on the chicken Chorio-Allantoic Membrane.

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References

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