1509: Cytokine Shifts and Expression of VEGF-A, VEGF-C and VEGF-D in Clinical Age-Related Macular Degeneration

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PURPOSE

Approximately 45% of subjects undergoing treatment for wet AMD develop some resistance to anti-VEGF-A monotherapy. VEGF-C and VEGF-D are members of the VEGF family that bind to VEGFR-2 and VEGFR-3 and are critical mediators of blood and lymphatic vessel growth and vascular permeability. Recently, we have demonstrated that blockade of VEGF-C and VEGF-D with a soluble form of VEGFR-3 (VGX-300 or OPT-302) inhibits lesion formation and vascular leakage in the laser induced mouse model of CNV, and that this activity is comparable to the activity of the VEGF-A inhibitor aflibercept. Furthermore, an additive effect of combined VEGF-A, VEGF-C and VEGF-D inhibition was also observed in this model, indicating that more complete blockade of the VEGF pathway may be more clinically effective and may shut-down mechanisms of resistance to VEGF-A inhibitors. In addition, previous studies have shown that normal primary retinal pigment epithelial cells express VEGF-C and that VEGF-C has a role in the development of retinal vascularisation.. Here we investigated the circulating levels and tissue localization of VEGF-C, VEGF-D and VEGFR-3 in pathogenesis of AMD.

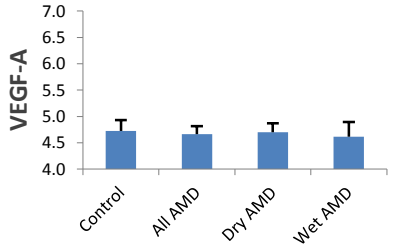
Methodology

Plasma samples were collected from 174 subjects with various stages of AMD and analyzed by ELISA for VEGF-A, VEGF-C and VEGF-D. Samples included non-AMD controls, dry and neovascular AMD. Data was analyzed both by non-parametric (Mann-Whitney) and parametric analysis of transformed data.

Paraffin-embedded ocular tissue were evaluated for stage of AMD and subjected to immunohistochemistry for expression of VEGF-C and its cognate receptor VEGFR-3. Some samples were also immunostained for VEGF-A expression.

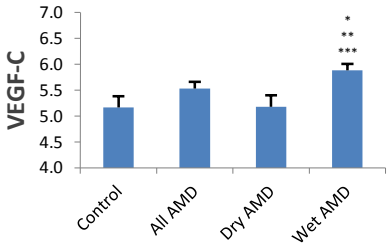
RESULTS

Mean Plasma Levels of VEGF-A



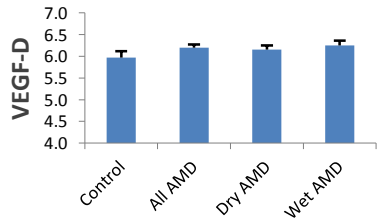
No significant difference in VEGF-A plasma levels was observed between groups (mean pg/ml ± SD).

Mean Plasma Levels of VEGF-C



VEGF-C is significantly elevated in wet AMD.
**** p <0.01 for wet AMD group versus Controls; ** p = 0.05 for wet AMD versus All AMD; *** p = 0.01 for wet AMD versus dry AMD (mean pg/ml ± SD).***

Mean Plasma levels of VEGF-D



No significant difference in VEGF-D plasma levels was observed between groups (mean pg/ml ± SD).

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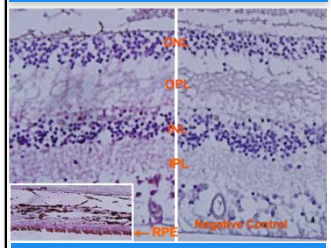
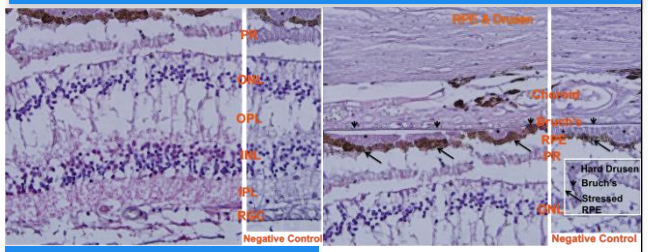
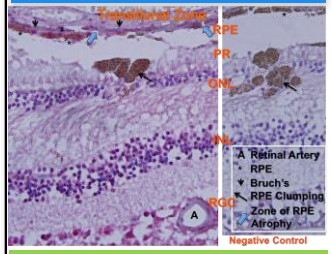
VEGF-C: No AMD



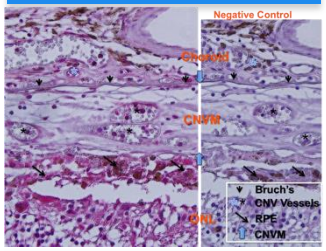
Intermediate Dry AMD



Geographic Atrophy



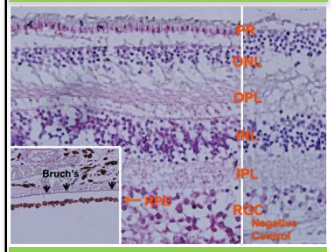
Neovascular AMD



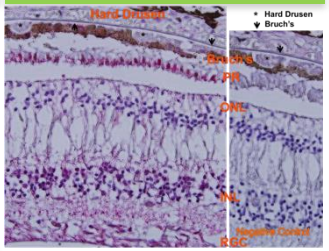
OBSERVATIONS:

1. VEGF-C & VEGFR-3 are expressed in most layers of the retina and RPE. In RPE their expression is polarized to the Bruch's side.
2. In dry AMD and GA, VEGF-C & VEGFR-3 are increasingly expressed in RPE around drusen & are associated with disease severity.
3. In wet AMD, VEGF-C & VEGFR-3 are expressed in RPE and endothelial cells associated with the CNV.

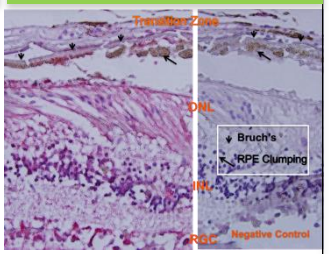
VEGFR-3: No AMD



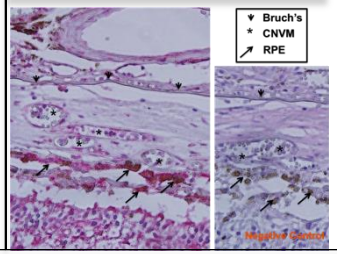
Intermediate Dry AMD



Geographic Atrophy



Neovascular AMD



CONCLUSIONS

1. VEGF-C is significantly elevated in the plasma of wet AMD patients compared to healthy volunteers and patients with dry AMD.
2. VEGF-C is expressed in the RPE of healthy eyes. VEGF-C levels in the retina increase with disease severity and were highest in neovascular membranes.
3. Inhibition of VEGF-C may be an effective therapeutic strategy for wet AMD, particularly in subjects who are resistant to anti-VEGF-A monotherapy.

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