Mapping and Quantification of Vascular Branching in Plants, Animals and Humans by VESGEN Software

Humans face daunting challenges in the successful exploration and colonization of space, including adverse alterations in gravity and radiation. The Earth-determined biology of humans, animals and plants is significantly modified in such extraterrestrial environments. One physiological requirement shared by humans with larger plants and animals is a complex, highly branching vascular system that is dynamically responsive to cellular metabolism, immunological protection and specialized cellular/tissue function.

The VESsel GENeration (VESGEN) Analysis has been developed as a mature beta version, pre-release research software for mapping and quantification of the fractal-based complexity of vascular branching. Alterations in vascular branching pattern can provide informative read-outs of altered vascular regulation. Originally developed for biomedical applications in angiogenesis, VESGEN 2D has provided novel insights into the cytokine, transgenic and therapeutic regulation of angiogenesis, lymphangiogenesis and other microvascular remodeling phenomena. Vascular trees, networks and tree-network composites are mapped and quantified. Applications include disease progression from clinical ophthalmic images of the human retina; experimental regulation of vascular remodeling in the mouse retina; avian and mouse coronary vasculature, and other experimental models in vivo. We envision that altered branching in the leaves of plants studied on ISS such as Arabidopsis thaliana can also be analyzed.

(Supported by NASA GRC IR&D04-54 and 2010 TTP Fund, NIH EY-01759 & NSF Center of Excellence, University of Washington Engineered Biomaterials)
VESGEN
Innovative Research Discovery Tool

Mapping and Quantification of Vascular Branching in Plants, Animals and Humans by VESGEN Software

Patricia A. Parsons-Wingerter, Mary B. Vickerman and Patricia A. Keith

Point of Contact: Patricia Parsons-Wingerter PhD
Biomedical Research Engineer, patricia.a.parsons-wingarter@nasa.gov
216-433-8796

Glenn Research Center
VESGEN Patent Pending at Lewis Field
Vascular Alterations, Immunosuppression & Bone Loss: NASA-defined risk categories for human space exploration
VESGEN 2D
APPLICATIONS

Vascular Trees
Human Retina
Avian CAM, Yolksac and Murine/Avian Coronary Vessels

Plant Leaf Venation such as in Arabidopsis thaliana?

Vascular Networks
Mouse Postnatal Retina and Intestinal Inflammation
CAM Lymphatic Vessels

Vascular Tree-Network Composites
Normal and Abnormal Embryonic Coronary Vessels

Glenn Research Center
VESGEN Patent Pending
at Lewis Field
Main panel
• Image specification
• Algorithm selection
• Process initiation

Panel to specify vessel type

VESGEN Patent Pending
Mapping and Quantification of Microvascular Remodeling and Angiogenesis by VESGEN

1. Cells in Vitro
2. Blood Vessels
3. Avian CAM
4. Transgenic Mouse
5. Humans
VESGEN Hypothesis: ‘Signature’ Vascular Patterns

FGF-2 as a Simple Stimulator
(Fibroblast Growth Factor-2)

Arterio Scler Thromb Vasc Biol 20 2000

VEGF as a Complexity Factor
(Vascular Endothelial Growth Factor-2)

Microvascular Research 72(3) 2006

TGF-β1 as a Simple Inhibitor
But Complex Potentiator
(Transforming Growth Factor-β1)

Microvascular Research 59(2) 2000
**Long-Term Hypothesis**

Vascular pattern provides an integrative read-out of dominant molecular regulators in complex signaling pathways of angiogenesis and microvascular remodeling.

**VESsel GENeration (VESGEN) Analysis Software**
Vessel Number Density, $N_v$
Vessel Length Density, $L_v$
Vessel Diameter, $D_v$
Fractal Dimension, $D_f$
Branchpoint + Endpoint Densities, $Br_v + E_v$
Clinical Steroid (TA) Treatment in CAM Vascular Tree

IF BIODIVERSITY WERE AN OLYMPIC SPORT, life on land would take home the gold and the sea might not even enter a team. Given the vastness of the oceans and the length of time life has thrived there, you might expect marine species to outnumber terrestrial ones. Yet, microbes aside, upward of nine in ten species crowd into the 30% of Earth’s surface that’s dry.

It wasn’t always that way, say Richard Grosberg and Geerat Vermeij. These researchers from the University of California (UC), Davis, have been studying land and ocean features to understand how evolution proceeds in these two realms. At a recent meeting,* they argued that the difference in diversity is a recent phenomenon.

Back in the Devonian period, 400 million years ago, the seas were home to an abundance of species, perhaps even more than on land. But about 110 million years ago, land plants went through a burst of speciation, so did the pollinators, fungi, and herbivores associated with them. These relationships made “rare” species possible, as plants acquired help in dispersing their pollen and seeds, resulting in relatively low population densities for individual species. Quickly, their numbers left marine biodiversity behind. The trigger for this terrestrial explosion, Grosberg and Vermeij say, was the evolution of a more efficient way in which land plants use water.

“This is an excellent and thoughtful paper addressing an issue in biodiversity that has rarely been tackled,” says Michael Benton, a paleontologist at the University of Bristol in the United Kingdom. Jeremy Jackson, a marine ecologist at the Scripps Institution of Oceanography in San Diego, California, calls it “a very big-picture paper . . . it’s the kind of paper that you think about forever.”

A physical phenomenon?
Grosberg started thinking about these issues when he was preparing a series of talks for the 200th anniversary of Charles Darwin’s birth.

“To me, the interesting question is why are there so many fewer species in the sea than on the land,” says Grosberg.

The difference is striking. In 1994, Robert May of the University of Oxford in the United Kingdom concluded that 85% of the world’s macroscopic species lived on land, based on the existing record of species across the globe. A 2009 study by Benton found landdwellers to be even more common, accounting for 95% to 98% of the world’s multicelled species. “Both recognized that the estimates were ballpark, simply because we don’t actually know how

*The Society for Integrative and Comparative Biology meeting was held 3 to 7 January in Seattle, Washington.
Vein pattern development in adult leaves of *Arabidopsis thaliana*


VESGEN
Research Tool for Mapping and Quantification of Vascular Branching Pattern in Genetically Engineered A. thalia on ISS

**Results**

- Fractal Dimension: $D_f = 1.32$
- Area, Length, Branchpoint Densities:
  - $A_V(\text{LARGE}) = 0.195$
  - $A_V(\text{SMALL}) = 0.013$
- Average Vessel Diameter ($\mu$m):
  - $D_V(\text{LARGE}) = 11.8 \mu$m
  - $D_V(\text{SMALL}) = 7.0 \mu$m

**Conclusions**

- Less mature branching
- Increased vascular complexity—especially density of smaller vessels
VASCULAR NETWORKS IN TRANSGENIC MOUSE RETINA

Fluorescence Microscopy

VESGEN Network Output

Distance Mapping

Colorized Skeleton

Mouse Retina

Control (P15)

TA-Treated


VESGEN Patent Pending
CORONARY VESSEL NETWORK-TO-TREE TRANSITIONS

Vickerman et al, VESGEN Review, Anatomical Record A 292(3), 2009
P Parsons-Wingerter et al IOVS 51(1) 2010 & M Vickerman et al Anat Rec 292(3) 2009
Surprising Oscillation of Angiogenesis with Vascular Dropout during Progression of Diabetic Retinopathy

Integrative Systems Biology:
‘Fingerprint’ or ‘Signature’ Mapping of Dominant Vascular Patterns Induced by VEGF and Other Angiogenesis Regulators
Fractal-Based VESGEN for Multi-Parametric Pattern Analysis according to Branching Generation

Research Discovery Tool for
Variety of plant, animal and human applications on ISS?
Acknowledgements

**NASA Glenn Research Center**
Mary Vickerman MS, Patricia Keith MS, Mark Wernet PhD, Terri McKay BS, Dan Gedeon, Alan Hylton MS, Daniela Ribita BS, Harry Olar BS, Camille Everhart, Dedra Whitfield

**Cleveland Clinic Foundation**
**Cole Eye Institute**
Peter Kaiser MD, Jonathan Sears MD, Quteba Ebrahem MD

**Lerner Research Institute**
Paul DiCorleto PhD, Unni Chandrasekharan PhD, Ron Midura PhD

**University of New Mexico School of Medicine/Ohio Aerospace Institute**
Krishnan Radhakrishnan MD PhD

**University Hospitals, Case Western Reserve University**
Steven Fisher MD, Hong-Bin Liu PhD
Michiko Watanabe PhD, Ganga Karunamuni BS, Monica Montano PhD