INTRODUCTION

It is well established that macular degeneration is a multifactorial disease related to age, genetics and a host of environmental factors such as light exposure and smoking. Choroidal ischaemia is also well accepted as a causative factor, but its severity and prevalence have not been well documented because of the difficulty in imaging the choroid due to light absorption by the overlying retinal pigment epithelium (RPE).

While reduced function of the RPE and Bruch’s membrane–choriocapillaris complex has also been described,1 7 8 Since these microvascular and regional inflammatory changes are difficult to detect in vivo, clinicians and researchers have sought imaging characteristics predictive of AMD onset or severity. Researchers have used fluorescein angiography, indocyanine green angiography, pulsatile ocular blood flow tonometry and Doppler flowmetry.9-13 Observed anatomic modifications could be accompanied by or even preceded by localised ischaemia.1 If microarchitectural choroidal changes that occur concomitantly with early clinical signs of AMD could be detected and documented, then applying early therapy could possibly arrest or interdict AMD progression.

Although advances in optical coherence tomography (OCT)12 13 allow imaging of retinal structures in great detail, adequate visualisation of the choroid has been challenging.14-17 Although enhanced depth imaging (EDI)-OCT can obtain measurements of choroidal thickness, it does not provide data regarding choroidal function. Ultrasonography circumvents the RPE’s optical barrier, and while resolution at even as high a frequency as 20 MHz (75 μm axially) is modest in comparison with OCT, ultrasound echo waveforms are affected by the size and spatial distribution of subresolvable scattering elements.18 Regional vascular and other changes in the choroidal interstitium would be expected to significantly alter the reflective interfaces within the ultrasound beam, leading to a complex alteration of the waveform that can be detected using wavelet analysis techniques.

We hypothesise that information from wavelet statistical models may serve as imaging surrogates for vessel diameter, vessel density and extraluminal fluid in the choroid. In this study, our primary objective was to determine if significant differences exist among wavelet parameters of eyes without AMD, with dry AMD and with wet AMD.

METHODS

This study was performed under a protocol approved by the Institutional Review Board of Weill Cornell Medical College. Written informed consent was obtained from the subjects.

This is a prospective case series of 69 eyes of 52 patients. Patients with AMD and age-matched patients without AMD were recruited in an academic retina practice (DJC) for fundus photography, OCT and indocyanine green (ICG) (in some patients) and high-resolution ultrasound in all patients. OCT and rare ICG were only performed as part of treatment decision-making in wet and dry
AMD, and not at all in the normal group. These data were not used for correlation with wavelet data, but instead were used clinically to aid in confirming the distinction between wet and dry AMD. We performed 20 MHz ultrasound scans on each eye using a B-scan immersion technique with a prototype instrument of our own design. Normal saline was used as the coupling medium, and the eyelids were held open with a Barraquer wire lid speculum. Phase-resolved (radiofrequency) echo data were digitised at a sample rate of 250 MHz with 8-bit precision.

We used a wavelet analysis technique which produces a time–frequency representation of the echo data. This allows addition of the property of shift invariance and gives superior identification of subresolvable information within the waveforms.

The data analysis began with segmentation of the macular retinal–choroidal area from the denoised B-scan using semiautomated methods. Independent components analysis (ICA) was then performed on echo data from the segmented region to obtain waveforms consisting of speckle and baseband components. Wavelet analysis was then applied to the ICA-processed data and summary statistics were calculated. A multivariate statistical technique was then used to identify wavelet coefficients and statistical structures differentiating eyes without AMD from those with dry AMD and those with wet AMD.

**RESULTS**

In all, 69 eyes of 52 patients were studied. A total of 18 did not have AMD, 23 had dry AMD and 28 had wet AMD.

The multivariate model showed statistically significant differences for individual wavelet coefficients and wavelet covariance parameters between groups. One measure, interscale phase consistency, a parameter sensitive to tissue interfaces, separated the three subject groups by itself (figure 1). The optimal multivariate model included three wavelet parameters related to persistence and clustering of coefficients between scales and adjacent lateral B-scan line segments as well as the rate that coefficient magnitude decays over the scales. The model demonstrated excellent separation of the three classes and a volume under the multiclass receiver operating surface of 0.892±0.17 (figure 2).

When applied to local echo data, the model can be used to produce wavelet images by performing successive and overlapping wavelet analyses centred on each pixel position using a sliding window. Such synthetic or hybrid ultrasound images show enhancement of contrast between fluid and tissue boundaries due to the differences in microarchitectural properties, as shown in figure 3. We term such images wavelet augmented ultrasound (WAU images) to differentiate them from conventional reflectance amplitude images. In figure 3, the hybrid image is superimposed on an ultrasound mid-band fit backscatter image which enhances boundary planes of the choroid, and like EDI-OCT studies, indicates thinning and irregularity of thickness in dry AMD eyes. These images demonstrate the data from which the interscale phase classifiers were derived.

**DISCUSSION**

As demonstrated here, WAU tissue characterisation allowed distinction of the dry, wet, and non-AMD choroid. Our methodology used digitised raw ultrasound data which differs from standard ultrasound imaging by retaining phase information present in ultrasound waveforms, facilitating both Fourier and Wavelet signal processing. Wavelet analysis is advantageous compared with Fourier analysis as it localises functions in both time and frequency instead of frequency only and adds the property of shift invariance. These properties produce superior...
microvascular anomalies.36 The ISCP clearly separated the supply or other causes such as in
pared with the normal eye. This indicates choroidal ischaemia in
boundary contrast (ie, decreased
phase consistency (ISCP)) that allows differentiation of wet, dry
AMD.

In dry AMD, the wavelet descriptors showed a reduction in
boundary contrast (ie, decreased fluid to tissue ratio) when com-
pared with the normal eye. This indicates choroidal ischaemia in
dry AMD. In wet AMD, descriptors showed an increased bound-
ary enhancement which could be caused by an enhanced blood
supply or other causes such as inflammation or intrachoroidal
microvascular anomalies.16 The ISCP clearly separated the
normal, dry AMD and wet AMD eyes in this small series, based
on the boundary ratios of fluid and tissue. The dry AMD separa-
tion can only be caused by ischaemia. The increased ratio of fluid
to tissue in wet AMD could have many causes, including inflam-
mation as suggested by Hageman et al6 or intrachoroidal micro-
vascular anomalies as suggested by Fukushima et al18 and at this
time can only be speculative. Future studies may be able to clarify
the cause of the increased fluid to tissue ratio in wet AMD.

The genesis of RPE degeneration and choroidal neovasculari-
sation in AMD is multifactorial, and is an open question. Is it
primarily genetic or oxidative stress or ageing of the RPE itself
or is it thickening of Bruch’s membrane? Could all be related to
ischaemia in the choriocapillaris and Sattler’s layer of small
arterioles? While the results of this study may not conclusively
answer the question of causation of all AMD, and do not

Figure 3 20 MHz ultrasound images of normal (top) and age-related
macular degeneration (AMD) (bottom) eyes after semiautomated
segmentation of retina (R), choroid (C) and sclera (S) as indicated by
coloration. Wavelet processing was applied to the echo data
encompassed by boxed regions. Note enhancement of druse in
wavelet-processed area of AMD image. The choroid is more irregular
in thickness in AMD than in normal eyes, as demonstrated here. These
‘hybrid’ or wavelet augmented ultrasound images demonstrate the use
of classifiers such as interscale phase consistency.

performance compared with Fourier methods in extraction of
subresolvable information from echo waveforms.34 We con-
ducted wavelet analysis of 20 MHz ultrasound data to deter-
mine and select descriptors most effective in distinguishing
between normal eyes and wet and dry forms of AMD. Wavelet
parameters were found to have significant classification power
and may serve as biomarkers for assessing choroidal change
in AMD.

Spectral-domain OCT allows choroidal thickness determin-
ation, as does ultrasound,35 and its representation of the chor-
oidal microvasculature such in EDI-OCT12 is excellent. While
OCT can now allow some visualisation of choroidal vasculature,
no comparable analysis based on Wavelet or Fourier transforms
has to-date been applied to OCT images of the choroid to dif-
ferrate wet, dry and non-AMD eyes. The methods described
here may be applicable to such data as well. Our technique
permits identification of a descriptor or classifier (ie, interscale
phase consistency (ISCP)) that allows differentiation of wet, dry
and non-AMD eyes based on choroidal ultrasound backscatter.

In dry AMD, the wavelet descriptors showed a reduction in
boundary contrast (ie, decreased fluid to tissue ratio) when com-
pared with the normal eye. This indicates choroidal ischaemia in
dry AMD. In wet AMD, descriptors showed an increased bound-
ary enhancement which could be caused by an enhanced blood
supply or other causes such as inflammation or intrachoroidal
microvascular anomalies.16 The ISCP clearly separated the
normal, dry AMD and wet AMD eyes in this small series, based
on the boundary ratios of fluid and tissue. The dry AMD separa-
tion can only be caused by ischaemia. The increased ratio of fluid
to tissue in wet AMD could have many causes, including inflam-
mation as suggested by Hageman et al6 or intrachoroidal micro-
vascular anomalies as suggested by Fukushima et al18 and at this
time can only be speculative. Future studies may be able to clarify
the cause of the increased fluid to tissue ratio in wet AMD.

The genesis of RPE degeneration and choroidal neovasculari-
sation in AMD is multifactorial, and is an open question. Is it
primarily genetic or oxidative stress or ageing of the RPE itself
or is it thickening of Bruch’s membrane? Could all be related to
ischaemia in the choriocapillaris and Sattler’s layer of small
arterioles? While the results of this study may not conclusively
answer the question of causation of all AMD, and do not
dispute genetics, oxidative stress or other factors, these results
do confirm a principal correlation, if not causation, of dry
AMD by choroidal dysfunction.

Our hypothesis, based on the results of this study and the
findings of other investigators, is that ischaemia, primarily of
Sattler’s layer of the choroid, exists in all or nearly all cases of
dry AMD. This smooth muscle endothelial dysfunction of the
terminal arterioles would reduce the production of nitric oxide,
a messenger molecule produced by the endothelium of the arterioles and choriocapillaris. This arteriolar damage may in
turn be caused by parasympathetic neuronal deterioration37 38
as in erectile dysfunction and other arteriolar smooth muscle
autonomic nerve-related diseases. Reiner and others39–42 have
described the autonomic nerve relationship in the choroid in
animal models. Fluid transfer between the choriocapillaris and
the interstitial space of the retina would be based on the gradi-
ent of hydrostatic and osmotic pressure. Flower and cowor-
kers43 have proposed that fluid transfer into the extracellular
space around the RPE and removal of waste products may be
governed by Starling’s Law. These waste products (eg, drusen)
and resultant toxic effects give rise to inflammation,15 27 vascular
endothelial growth factor production and ultimately wet AMD.
Our hypothesis would thus support a parasympathetic neuronal
control of the terminal arterioles as a causative factor in reduced
perfusion of the extracelluar region surrounding the RPE.
While other causes such as genetic predilection and/or oxidative
stress are certainly present, RPE deterioration in AMD could
thus be at least partly an effect rather than the sole direct cause
of dry AMD, and ultimately wet AMD. The concept of para-
sympathetic choroidal perfusion regulation would relate
pharmacological treatment of choroidal dysfunction to that of
e erectile dysfunction, another parasympathetic dystrophy for
which a rich pharmacological data base is available.

Based on our results of ultrasonographic evidence of chor-
oidal ischaemia in AMD and the results of EDI-OCT,44 45 we,
like others,4 46 feel that pharmacological agents such as sildena-
fil, tadalafl, niacin or other agents that increase choroidal perfu-
sion could have a beneficial effect on delaying or interdicting
AMD. Choroidal perfusion measurements with swept scan ultra-
sound and OCT measurements of thickness offer a means of
measuring and monitoring pharmacological therapy.34 45

Ultimately, longitudinal studies, including swept scan quanti-
tative measurement of choroidal perfusion coupled with OCT,
will be required to determine if reversing or moderating chor-
oidal ischaemia will stabilise or interdict AMD. Such studies will
also provide increased insight into the sequence of RPE change
relative to the other parts of the tunica ryschiana and more
definitively determine not only the role of ischaemia as a
primary causative factor of AMD but allow us to determine the
role of pharmacological agents that could have a beneficial
effect on the development and progression of AMD.

Contributors  Design and conduct of the study: DJC, RHS and RVPC; collection,
management, analysis, and interpretation of the data: DJC, RHS, MUR, HOL and AAX; and preparation and review of the manuscript: DJC, RHS, MUR and HOL.

Funding  This study was supported in part by NIH grant 1R01EY002038, the Dyson
Foundation, the St. Giles Foundation and Research to Prevent Blindness.

Competing interests  None.

Ethics approval  The study and data accumulation were carried out with
prospective approval from the Well Cornell Medical Center Institutional Review
Board (IRB) (#0508008050) and was conducted in compliance with the tenets of
the Declaration of Helsinki. Informed Consent for the research was obtained from
the patients and the study is in accordance with HIPAA regulations.

Provenance and peer review  Not commissioned; externally peer reviewed.

Open Access This is an Open Access article distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 3.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited and the use is non-commercial. See: http://creativecommons.org/licenses/by-nc/3.0/

REFERENCES


Age-related macular degeneration: choroidal ischaemia?

D Jackson Coleman, Ronald H Silverman, Mark J Rondeau, Harriet O Lloyd, Aziz A Khanifar and R V Paul Chan

Br J Ophthalmol 2013 97: 1020-1023 originally published online June 5, 2013
doi: 10.1136/bjophthalmol-2013-303143

Updated information and services can be found at:
http://bjo.bmj.com/content/97/8/1020

These include:

References
This article cites 40 articles, 8 of which you can access for free at:
http://bjo.bmj.com/content/97/8/1020#BIBL

Open Access
This is an Open Access article distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 3.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited and the use is non-commercial. See: http://creativecommons.org/licenses/by-nc/3.0/

Email alerting service
Receive free email alerts when new articles cite this article. Sign up in the box at the top right corner of the online article.

Topic Collections
Articles on similar topics can be found in the following collections

Open access (252)

Notes

To request permissions go to:
http://group.bmj.com/group/rights-licensing/permissions

To order reprints go to:
http://journals.bmj.com/cgi/reprintform

To subscribe to BMJ go to:
http://group.bmj.com/subscribe/