

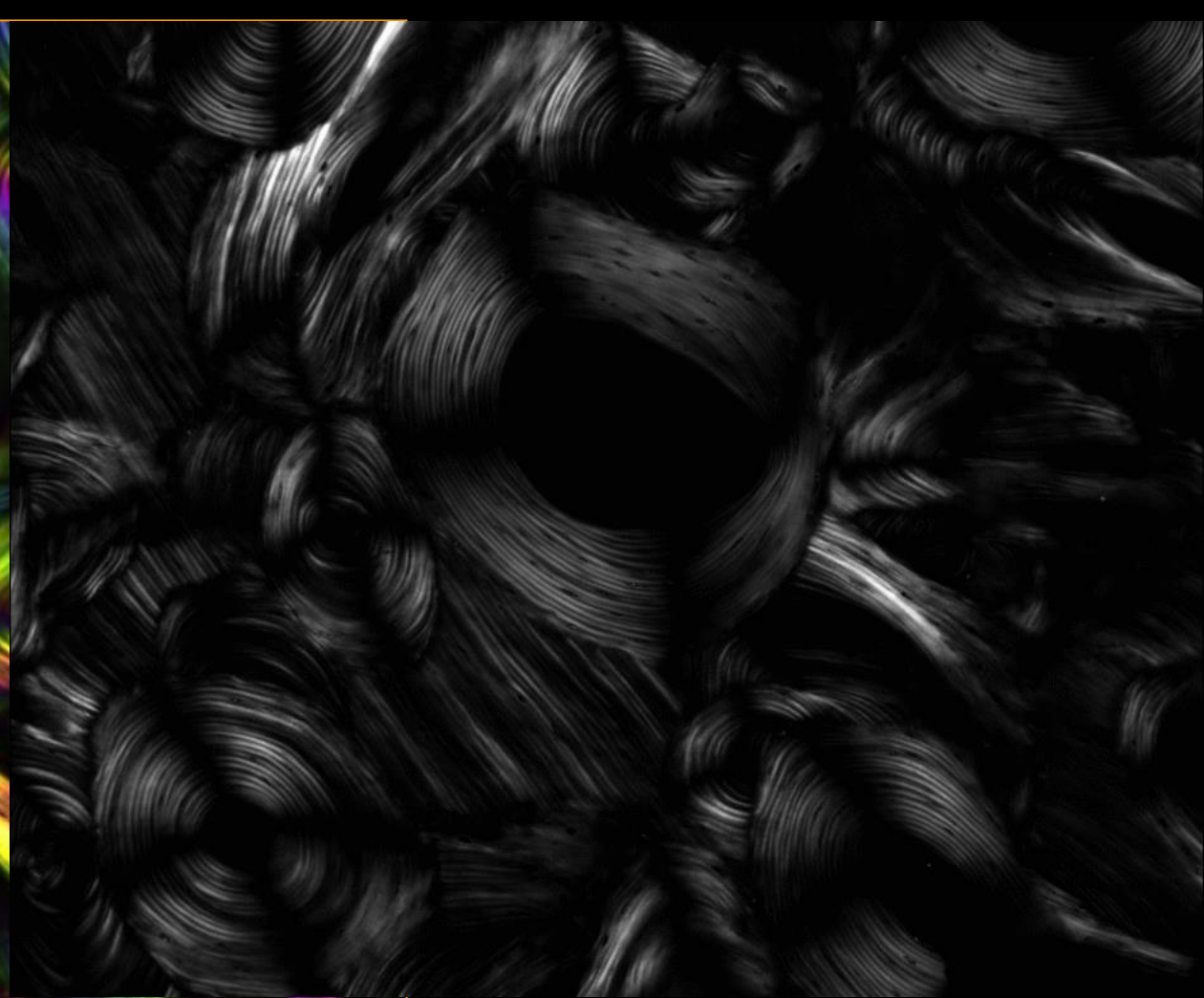
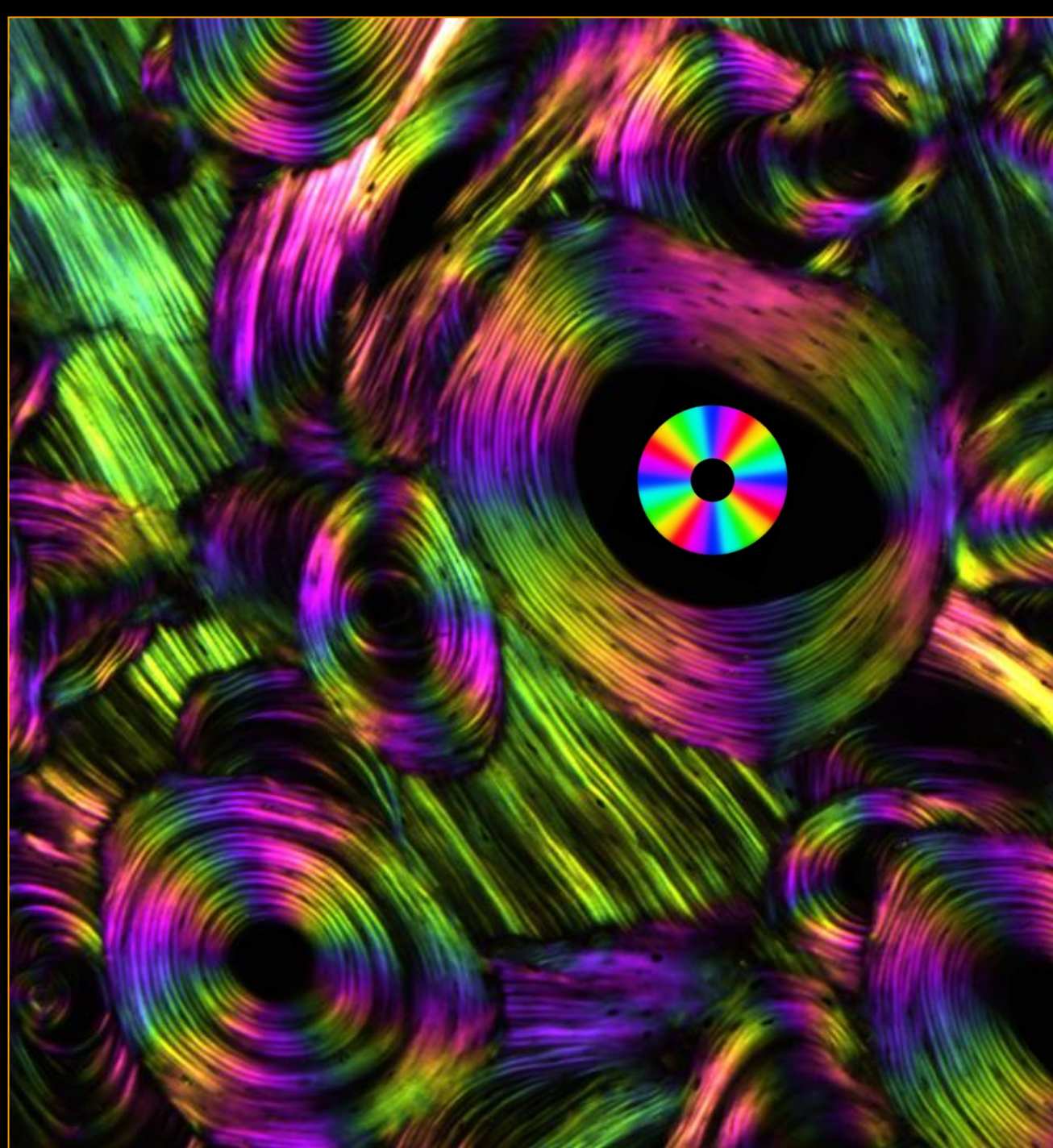
# New quantitative method for increasing information content in polarised light imaging of bone tissue.

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In linearly polarised light (LPL), birefringent structures appear brightest if they lie both in the plane of the section and at  $45^\circ/135^\circ$  to the axes of the crossed polarising filter elements, but dark if perpendicular to the section plane or parallel to either polarizer or analyser, preventing measurement of the whole scene at once because nothing can be resolved in the dark sectors of the 'Maltese cross'. This may be solved using circularly polarised light (CPL), when dip with respect to the section-plane may be quantified for plane parallel sections and we can use pseudocolour to produce dip maps. CPL, however, does not differentiate between in-plane orientations. We provide a new solution by combining numbers of PLM images to map orientations in 3D. We have automated the coupled rotations of polarising and analysing filters at, for example,  $3^\circ$ ,  $5^\circ$ ,  $7.5^\circ$ ,  $10^\circ$  or  $15^\circ$  intervals through a range of  $90^\circ$  - with digital LPL images recorded at each orientation - and exploit digital processing. For in-plane orientation mapping display we use the colour circle sequence Red, Yellow, Green, Cyan, Blue, Magenta, where colour shows the orientation with 4 repeat cycles in  $360^\circ$ . Brightness is proportional to the cosine of the dip/strike angle with respect to section plane, being brightest in plane, and black when perpendicular to that plane, i.e., parallel to the optic axis. The dip value can be displayed in a pseudocoloured version of the sum of the separate monochrome LPL images using a Look-Up-Table with six  $15^\circ$  vertical orientation classes. The new method is powerful, label free and best used with unstained sections, but most stains do not interfere too much. Thus it may be used for much archival material. It proves to be excellent for undecalcified, uncalcified and decalcified tissue sections.



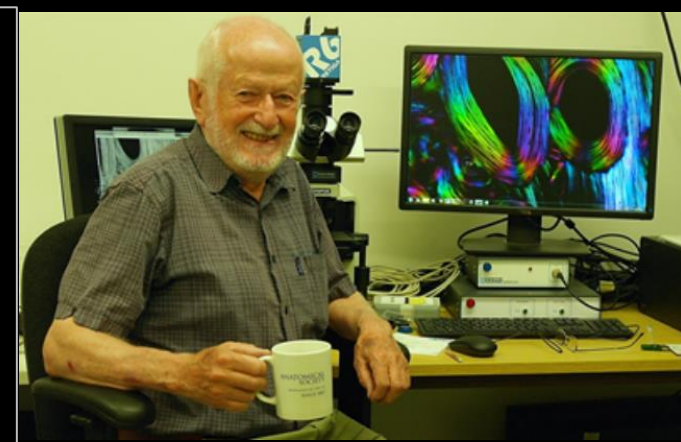


100  $\mu\text{m}$  Annular saw 'GS'  
Human Femur TS<sub>110 10x 09</sub>  
6 images at 15°  
Field width 1220  $\mu\text{m}$



albatross section 95 4/0.13.  
max 16.5 x 12 mm

21 images each taken with 12  
rotations of crossed polars  
outer ring RYGBM 100%  
middle sum 75%  
inner 6 classes 15 degree  
orientations, 50%  
innermost, as above  
but at 30%

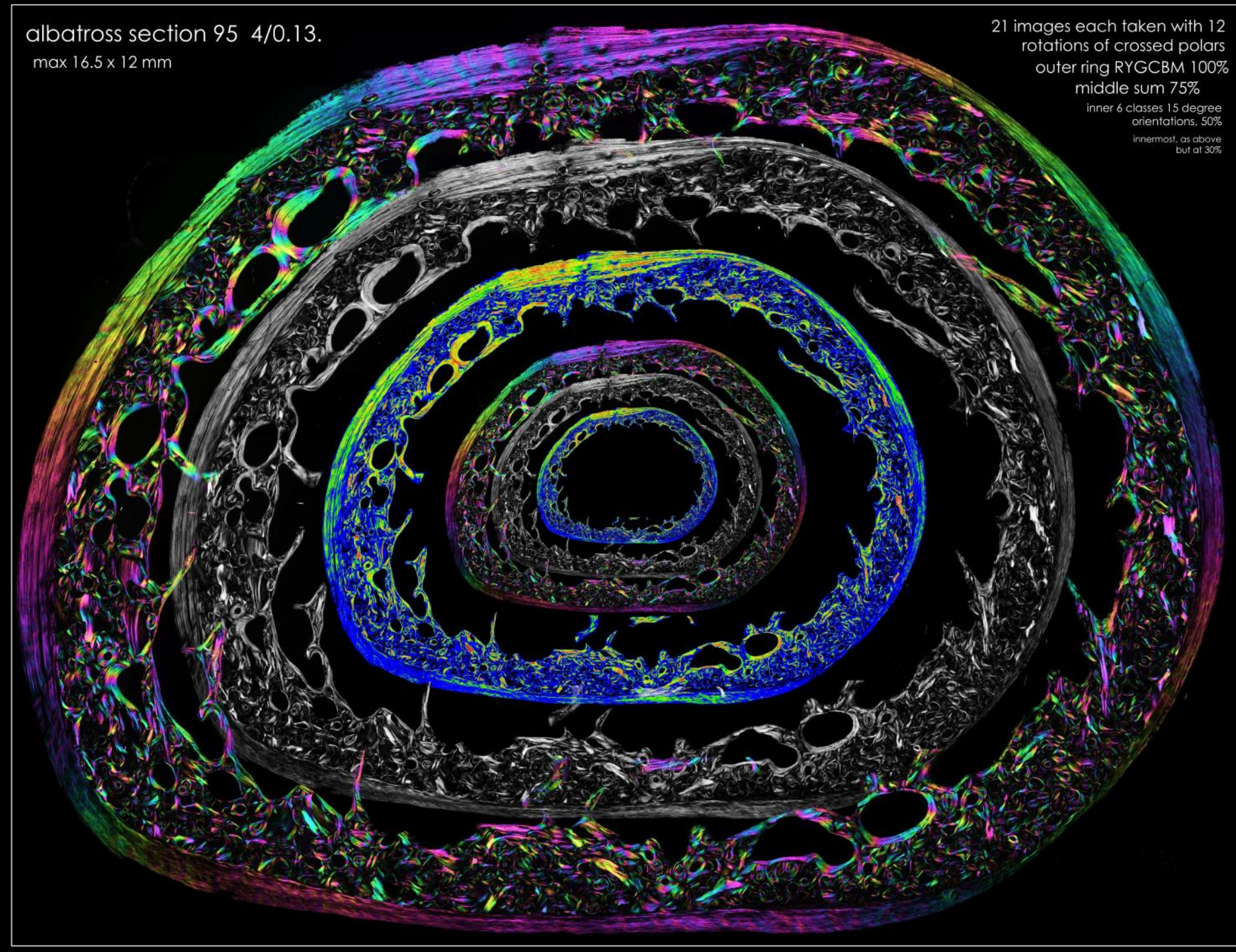


The kit is in the background  
behind the 'Senior Author'.

The one time in our lives that  
we really wanted a poster and  
we get stuck with this horrid  
video presentation business!!!!

And we actually made the  
poster in February before the  
dread Covid-19 made all the  
headlines. This is it. Huge.

The nested 'Russian dolls' show  
the same data set with different  
look up tables and scales.





21 images each taken with 12  
rotations of crossed polars  
outer ring RYGCBM 100%  
middle sum 75%

Snip from  
the top right  
hand corner

inner 6 classes 15 degree  
orientations, 50%  
innermost, as above  
but at 30%

albatross section 95 4/0.13.  
max 16.5 x 12 mm

TS humerus,  
preserved and  
transported in  
diesel oil by BAS:  
most other  
liquids freeze in  
the Antarctic

