

# **Paganin single-distance phase retrieval reconstruction in NRecon**

**Method note  
MCT-133**

## 1. Introduction

NRecon version 2.0 now includes phase contrast for enhanced contrast from non-absorption interactions, using the single-distance phase-retrieval algorithm described by David Paganin *et al.* (2002)<sup>1</sup>. X-ray phase contrast imaging makes use of the phenomena of refraction and diffraction of X-rays at internal boundaries of a sample and is thus complementary to conventional absorption contrast. Phase contrast was initially developed for synchrotron X-ray imaging, but with the phase retrieval method described here, lab instruments equipped with a microfocus X-ray source (focal spot size < 5 – 10 micron) can also usefully employ phase contrast effects.

The phase retrieval method can provide enhanced contrast in a wide range of materials and objects (although not all), limited by the extent of edge scattering in the X-ray images. It works well with low density objects containing air spaces such as polymers, plastics, carbon fiber and non-mineralised biological tissue. It is not limited to such samples, also giving good results for medium density bone, geological or other materials. High resolution generally is needed to detect edge scattering.

Please note that the Paganin phase retrieval method is not a reconstruction method but a method of processing 2D X-ray projection images. By applying it to all the projection images in a dataset, the projections thus transformed are reconstructed in NRecon in the same way as a regular scan (with most recon parameters unchanged).

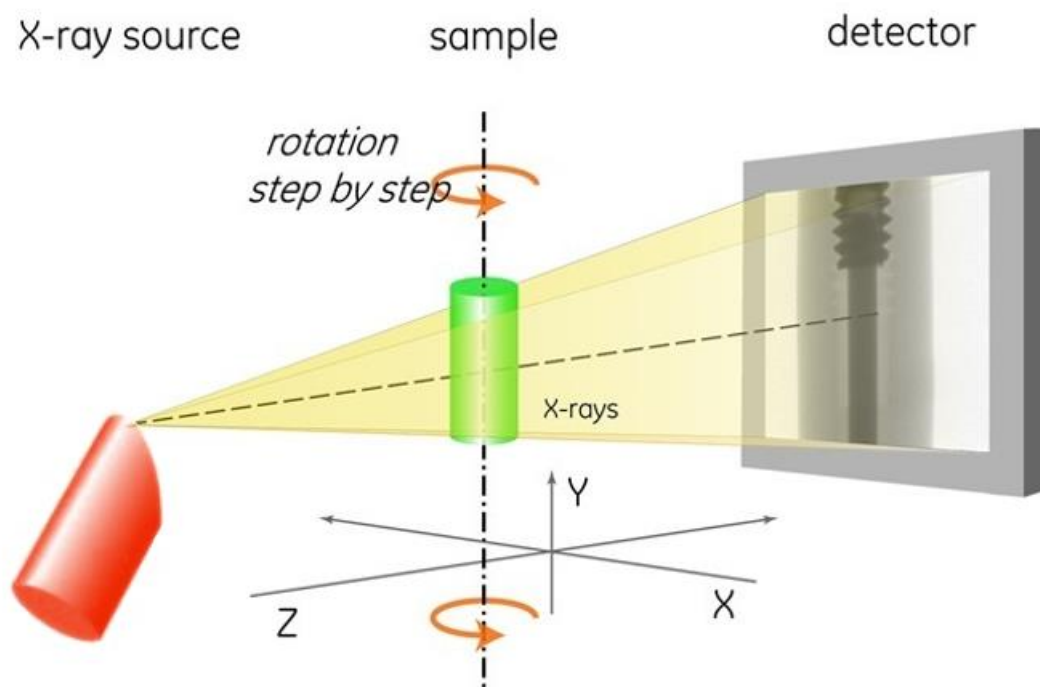
### 1.1. Background

Before we get to phase contrast and retrieval, we will make a brief recap of X-ray microCT essentials. This will help to provide context for later.

Tense cinematic moments in modern movies are often created by orbiting the camera around the subject. The film is taken from a camera rotating in a circle facing the hero in the circle center. Think Keanu Reeves (“Neo”) in The Matrix, dodging bullets on the roof of a high-rise building. In the same way in X-ray CT, projection images are taken

while the angle of the camera from the sample rotates, by either the sample on a stage or the opposed camera and source rotating in a circle. The geometry of X-ray CT acquisition is shown in figure 1.

X-ray micro computed tomography (microCT) is based on “attenuation” which means literally “weakening”. The scanned object between the source and camera causes the detected flux of X-rays emitted by the source to be reduced with some photons not making it to the camera, because they have interacted with the sample. The shadow projection image that is the raw material of tomographic reconstruction is one where the dark areas are the interesting features that convey the important data – areas where photons are “missing”, rather than bright areas.



**Figure 1.** Whether the X-ray source and camera rotate on a gantry around the sample, or the sample rotates on a stage, the resulting acquisition geometry in X-ray CT is one where multiple projection images are taken with incrementally rotating angle of the sample around the vertical Y axis.

Thus, CT imaging is based on a negative signal – absence of X-rays – that would otherwise reach the camera if nothing but air filled the space between source and camera<sup>i</sup>. This negative signal means that tomographic reconstruction is what we call mathematically an “inverse problem” – how to figure out what is not there, rather than what is (Tikhonov 1943)<sup>2</sup>.

## 1.2. Absorption CT

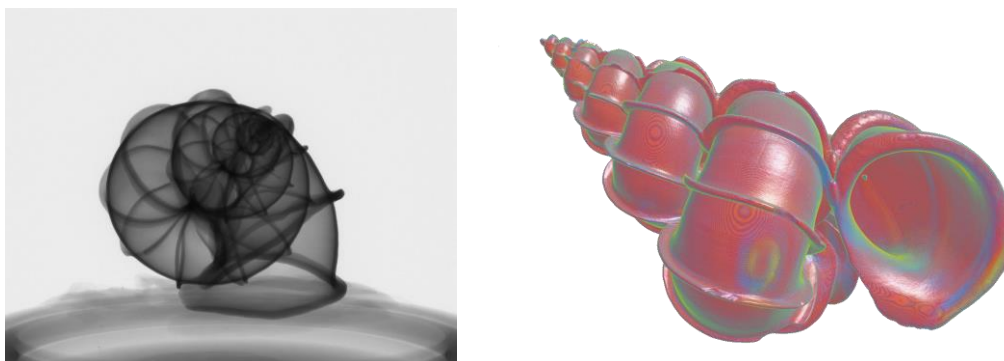
We call this conventional method “absorption” CT, since the no-show of X-rays at the camera is assumed to result from absorption in the sample, somewhere between the source and detector. The two analytical processes of back-projection and convolution together reconstruct, from multiple projections around the scan orbit, a 3D map of the magnitude of X-ray depletion. This mathematical detective work figures out exactly where all the missing X-ray photons disappeared. The result is our 3D absorption map (see figure 2).

Two phenomena account for what we call absorption; first photoelectric absorption, where the X-ray photon is effectively stopped, and which happens predominantly with lower X-ray photon energies and with atoms of higher atomic number (Z). The second is Compton scattering occurring at higher photon energies. Although the X-ray is not

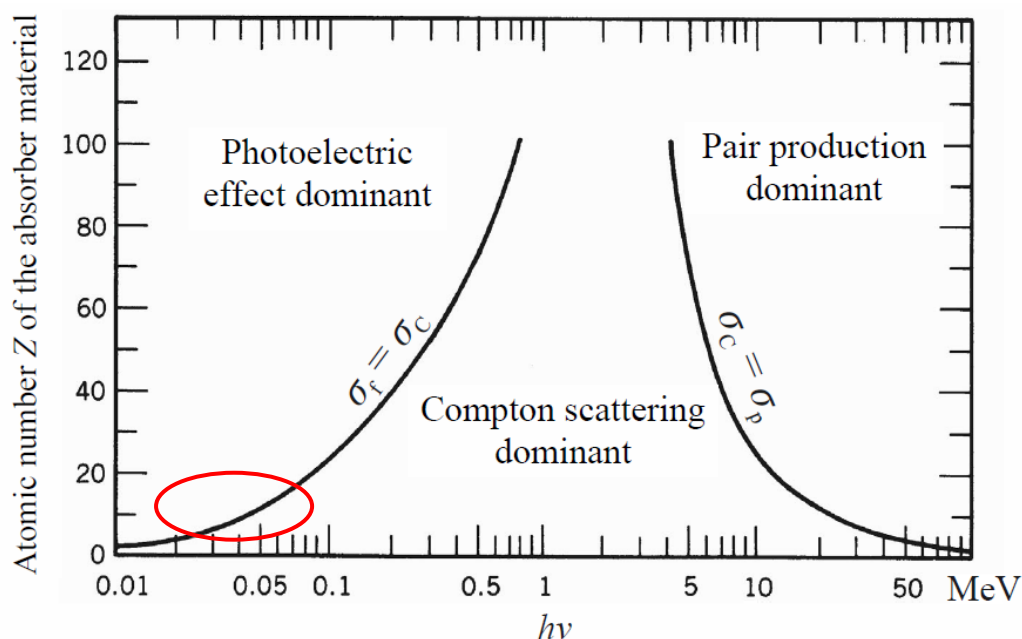
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<sup>i</sup> Please note that for physical accuracy it is incorrect to describe light as “travelling” between A and B and taking time to do so. This is a common narrative device which can help to explain things, but technically it’s wrong. X-rays are a kind of light (a mass-less field) and travel at light-speed, thus they *experience no passing of time*. Take a star like the red giant Betelgeuse (left shoulder of Orion) 700 lightyears from us. Was the light we see from it emitted 700 years ago? No. It took no time to reach us. But the consequence of relativity is that time at the star is 700 years different from time at our end of the photon. Einstein’s insight is that time and distance are one. Photons don’t “travel” since that involves time, and photons don’t do time.

stopped but deflected, from the point of view of imaging the photon is effectively removed from the beam path.



**Figure 2.** Absorption based CT maps the depletion or attenuation of X-rays in the projection image (left) into a 3D map of intensity of attenuation which allows 3D reconstruction of objects as shown in this scan of an *Epitonium* sea snail (right) volume rendering of Feldkamp backprojection-reconstruction.



**Figure 3.** The interaction modes of x-rays with matter depending on atomic number  $Z$  and the x-ray photon energy  $h\nu$ . The red oval indicates the region of microCT imaging of biological tissue. Biological soft tissue elements C, N and O ( $Z=6,7,8$ ) are near the boundary of equality of photoelectric absorption and Compton scattering, at photon energies used in microCT. Since the sensitivity of absorption to  $Z$  is higher in photoelectric absorption than in Compton scattering, in practice contrast within biological tissue is improved by decreasing x-ray photon energy. (From *The Atomic Nucleus*, Evans 1955<sup>3</sup>.)

Figure 3 shows the “phase space” of predominant X-ray interaction mode relative to two parameters, the X-ray photon energy (horizontal x axis, log) and the element atomic number  $Z$  (vertical y axis; Evans 1955)<sup>3</sup>. In microCT a lot of the absorption occurs near the bottom left of this diagram close to the boundary of the regions where photoelectric absorption and Compton scattering predominate.

### 1.3. Phase contrast CT

However, absorption is not the only thing that can happen to an X-ray photon between the source and the camera. Other interactions with a sample that can take place include small angle scattering (different from Compton scattering) and refraction with change to a photon’s wavetrain phase. Diffraction with associated bright and dark layered fringes can also occur. These other, non-absorption interactions provide the basis of a different way of obtaining image data, and collectively are referred to as “phase contrast”. This is directly equivalent to the phase contrast imaging that is performed with visible light microscopes. Phase interactions of X-rays are of smaller magnitude than those that occur with visible light, but they are significant enough for X-ray phase contrast methods to exploit these phenomena.

There are several different non-absorption based methods using different interactions that fall into the broad category of “phase contrast”, and it is not the purpose of this method note to give a full theoretical background to them all. A review of X-ray phase contrast techniques and their historic development is provided by Mayo *et al.* 2012<sup>4</sup> who explain the need for coherence of X-rays for refraction-based phase contrast to work effectively. Coherence means the X-ray wavetrains are in “in step” with each other, in the same phase. This requirement leads to a stringent technical limitation to the application of these X-ray phase contrast methods in practice. Synchrotron facilities, where particles are accelerated in large circular structures with diameters of up to several kilometers, allow the generation of controlled beams of X-rays which are both monochromatic (all of the same photon energy) and also coherent. However, laboratory X-ray sources, which generate X-rays from accelerated electrons impinging

on a metal (e.g. tungsten) target<sup>ii</sup>, cannot produce such controlled X-ray fluxes. These are the X-ray sources used in laboratory micro- and nanoCT systems. The X-rays from laboratory sources are polychromatic and non-coherent – although some attempts have been made to create partly monochromatic and coherent X-rays in large laboratory setups. A review of the principles of phase contrast tomography with coherent synchrotron radiation is provided by Cloetens *et al.* 2002<sup>5</sup>. They show that combining coherent X-ray tomography with different source-detector distances generates refraction-based interference allowing effective imaging of structures in the absence of X-ray absorption contrast; this technique is sometimes called “holotomography”. The authors show the application of this form of phase contrast to imaging the tissues of the cabbage-like plant *Arabidopsis*. Another type of phase contrast is a method using finely spaced gratings in front of the source and detector, and employs diffractive effects to add a degree of coherence to the polychromatic X-rays emitted by laboratory sources. This is the Talbot-Lau interferometer method<sup>6</sup> of which there are several variants. The need for high spatial precision in the gratings can limit the range of magnification possible, and this somewhat restricts the applicability of the Talbot-Lau approach, in the laboratory at least. Generally, there are two types of phase contrast method, the gratings approach (e.g. Talbot-Lau) and “free propagation” methods that use no grating but rely on distance from reflecting-diffracting surfaces to generate interference effects.

#### **1.4. The Paganin phase retrieval method**

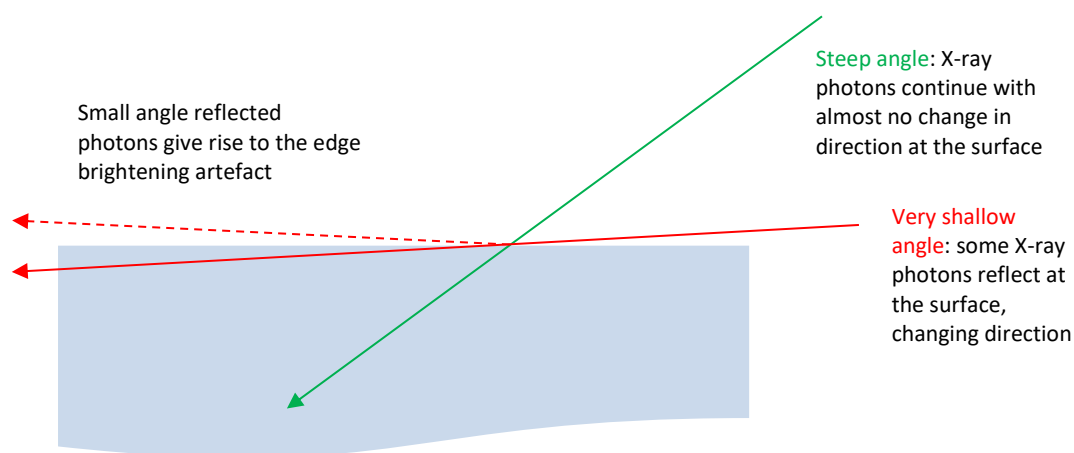
This requirement for coherence and monochromaticity would seem to exclude laboratory X-ray microCT systems from carrying out free propagation based phase contrast, and limit the technique to those with access to large synchrotron facilities.

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<sup>ii</sup> Emitting X-rays by characteristic (photoelectric) emission and Brehmsstrahlung interactions.

However, there is one phase contrast phenomenon that does happen readily in laboratory microCT systems even with noncoherent laboratory sources, and without the need of any gratings; this is small angle scattering.

Figure 4 below illustrates what happens when X-rays moving through a low attenuation medium like air strike a surface of a higher density solid. Basically, some of them bounce. This only happens at very shallow angles of incidence – less than one degree. (Figure 4 exaggerates the angle a little, just for visual clarity.)

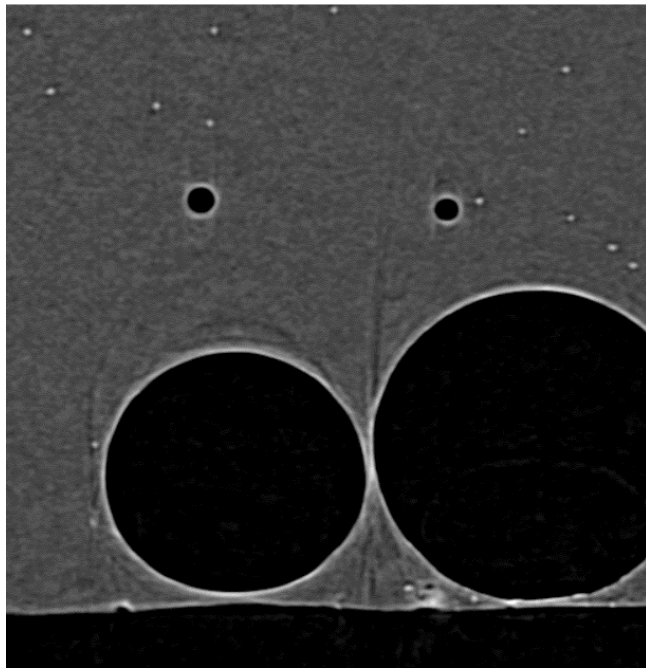


**Figure 4.** A diagram of small angle X-ray reflection at surfaces.

What is the consequence of this small angle scattering? Remember the above discussion about X-ray tomography being an inverse problem, that X-ray attenuating density is inferred from the absence of X-rays passing through a volume? Small angle scattering in a different but analogous way removes X-rays from the beam transects passing along surfaces, so the reconstruction algorithm “sees” a region that depletes X-rays, and thus assigns to that region a high attenuation, or a bright greyscale. The algorithm “thinks” that there is more absorption at the surface, although the X-rays are actually being depleted by scattering. Thus you get the artefact of a bright, dense layer at a surface, when no such layer exists – the material is uniform. An example of this is shown in figure 5 below, a scan at 1.9 micron voxel size by the SkyScan 1172



desktop microCT, of a low density polymer containing air bubbles and fibers. The bright layers on the surface of the bubbles are clear examples of edge enhancement caused by small angle scattering. This did not require a synchrotron, or even submicron resolution in a powerful nanoCT scanner with an open transmission source. The phenomenon is widespread in regular microCT imaging and is most prominent at surfaces with air – although it does occur at interfaces of materials not including air also.



**Figure 5.** A microCT cross-section (voxel size 1.9um) of plastic containing air bubbles (black circles) and fibers (white dots) scanned in the SkyScan 1172. The bubbles appear to be bounded by a bright dense layer, but this is an artefact of small angle scattering. This edge enhancement is the basis of the Paganin phase retrieval method.

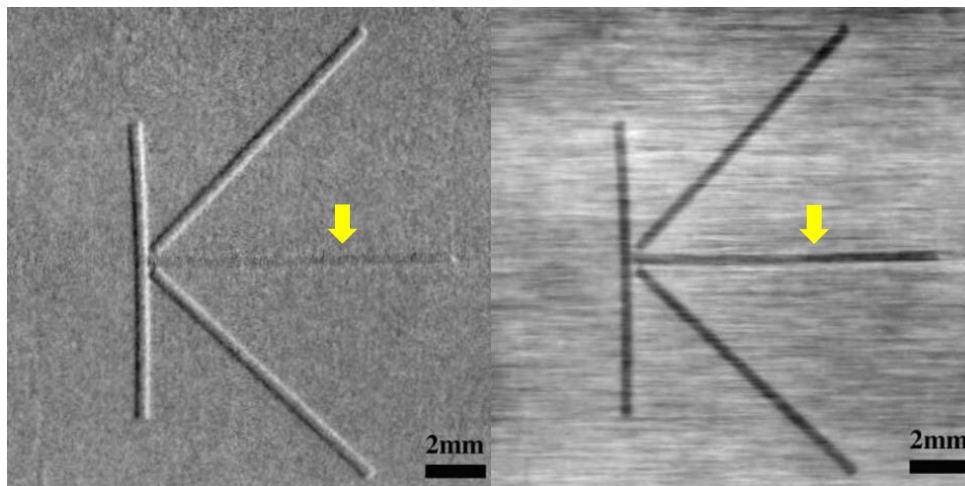
#### *1.4.1. Converting edge scattering into enhanced contrast*

This edge brightening can be looked at in two ways. On the plus side, it represents a form of sharpening, making surfaces more visible. It actually increases spatial resolution, and is one of the reasons why it is worthwhile to go down to pixel sizes below the spot size of the X-ray source in microCT imaging, where theory of penumbral blurring would predict that no gain in resolution would occur. But we find

that apparent resolution and detail detectability do in fact increase. This is in part down to small angle scattering. Indeed this edge enhancement is itself referred to as a type of phase contrast.

But the minus side of the small angle scattering is that the edge enhancement, while possibly useful for visualising features, is not real but an artefact. The enhancement implies the existence of an X-ray dense layer at surfaces that is not in reality there. Furthermore, Paganin and colleagues realized that the edge enhancement phenomenon was causing a loss of information about material contrast in X-ray images; so they set out to try to get this information back.

The key insight of the Paganin phase retrieval (PR) method is that it is possible to “retrieve” the image signal associated with the edge scattering and edge depletion-brightening, and convert this signal into enhanced image contrast between materials. How this is done is set out in the papers by David Paganin and colleagues (Paganin *et al.* 2002<sup>1</sup>, 2004<sup>7</sup>, 2008<sup>8</sup>) who established the eponymous Paganin phase retrieval method. The method was initially applied to 2D images from microscopy and radiography, but by applying the method to a set of tomographic projection images, it works for 3D CT also. In their 2004 paper<sup>7</sup> an illustration is given of how in a 2D clinical radiogram (figure 6) diffraction gives a directional shadow effect. This results in elements in certain directions – such as the horizontal element in figure 6 – being poorly visualised. However by what Paganin describes as “diffraction enhanced imaging” (DEI) the image of the horizontal element is retrieved, so that it becomes as clearly resolved as the other elements at different angles.



Original X-ray projection

Phase-retrieved projection

**Figure 6.** A clinical X-ray radiograph of a phantom object in 2D shows that the original image (left) has a directional component associated with diffraction. Thus the horizontal element is poorly visualised. In the phase retrieved (“diffraction enhanced”) image to the right, the horizontal element is as well visualised as all the other elements<sup>7</sup>.

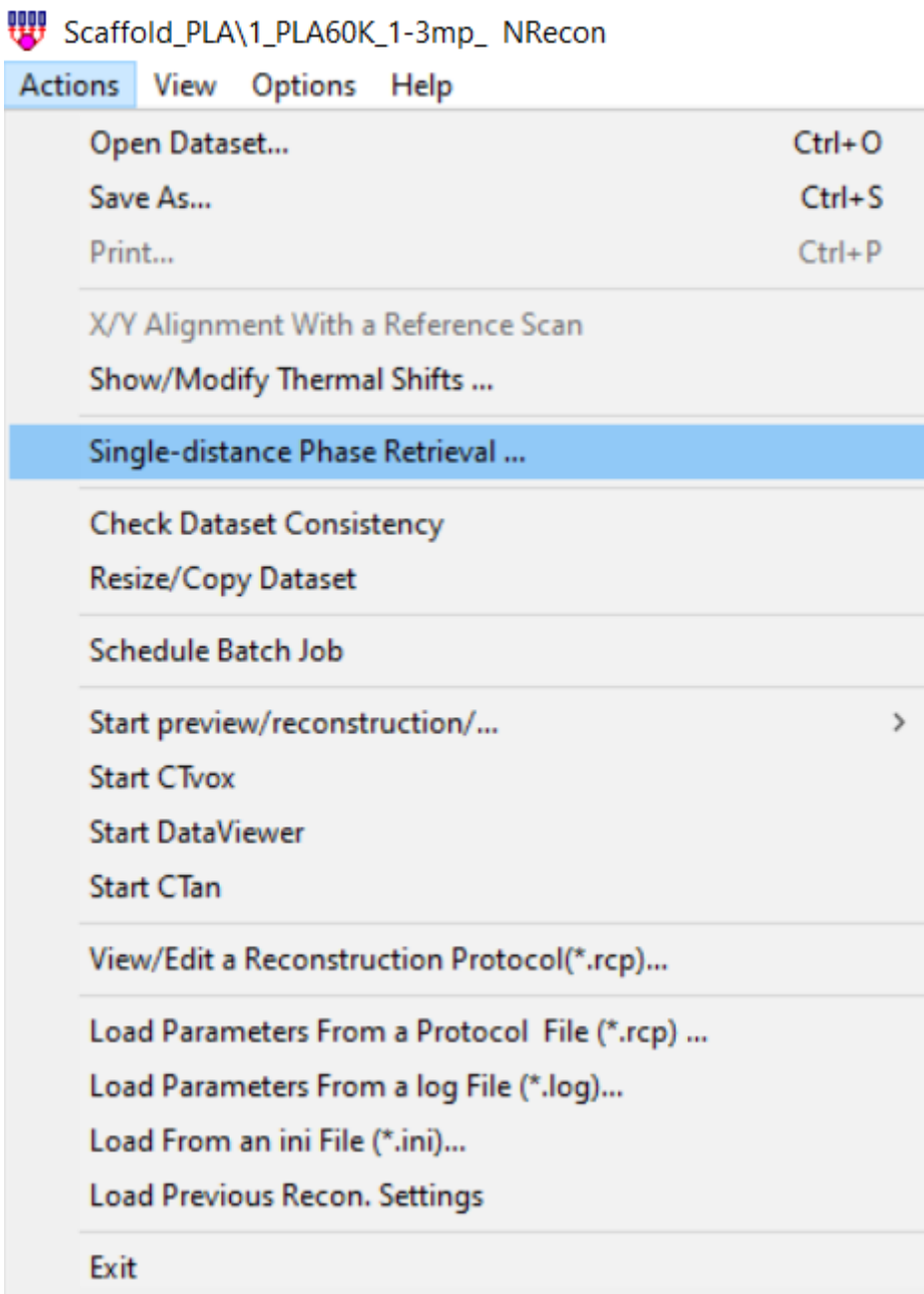
An image processing method derived from Paganin’s “diffraction enhanced imaging” (DEI) is applied in microCT to allow phase retrieval of CT projection images.

To repeat from above, this phase retrieval is not a different CT reconstruction algorithm. It is a method applied to individual 2D X-ray projection (and other) images. By applying it to a full set of scan projections, micro and nanoCT can be performed with phase retrieval. In this method note we will see how to do that.

Paganin phase retrieval (PR) has become an important alternative method for reconstructing microCT datasets, following PR processing of projection images, allowing material contrast to be significantly enhanced in certain cases. (It is not applicable to all scans, however.) Some of you might have encountered this method in the form of the popular ImageJ plugin “Ankaphase”. The new PR function in Bruker NRecon software that will be described here, does the same thing as Ankaphase – it writes a copy of the projection dataset with PR processing; except the NRecon version is a lot more user-friendly with fewer adjustments to make (many parameters are silently read from the scan log file). This makes it accessible for all microCT users.

## 2. Method

First load a projection dataset into NRecon. Then select from the Actions menu the item “Single-distance Phase Retrieval”. The window shown below will open. There are several parameters displayed but essentially only one that you need to adjust – that is the delta / beta ( $\delta/\beta$ ) ratio.



Here is the phase retrieval (PR) window:

Single-distance phase retrieval ✕

Parameters

Delta/Beta  X-ray(KeV):   
☒ Image restoration Gauss width  Stabiliser

Output options

☒ Auto range Margin(%)   
Range (min/max)

Time	Prj	Description
Prj	---	Show original projections
08:50:34,Thu	0	Delta/Beta:700.0, Xray: 29.5, Gauss: 0.1, Stabiliser: 0.5.

Preview done. Found [Min, Max] = [6.9, 8.8]. Result in subfolder PR.

### 2.1. The phase retrieval parameters

- Delta/Beta ratio.** (Or  $\delta/\beta$  ratio.) This is the most important (and to start with, the only) parameter that you need to adjust. It is thus described in its own section below.
- X-ray energy (keV).** There is a cell for average X-ray photon energy, but this is greyed out since NRecon approximates the mean photon energy as half the applied voltage. This parameter does not influence the result very much in practice. No action needed.
- Image restoration.** This is an algorithm that reduces noise. Generally, the tick box “Image restoration” should be ticked. The two parameters to the

left of this option, “Gauss width” and “Stabiliser”, relate to the Image Restoration process.

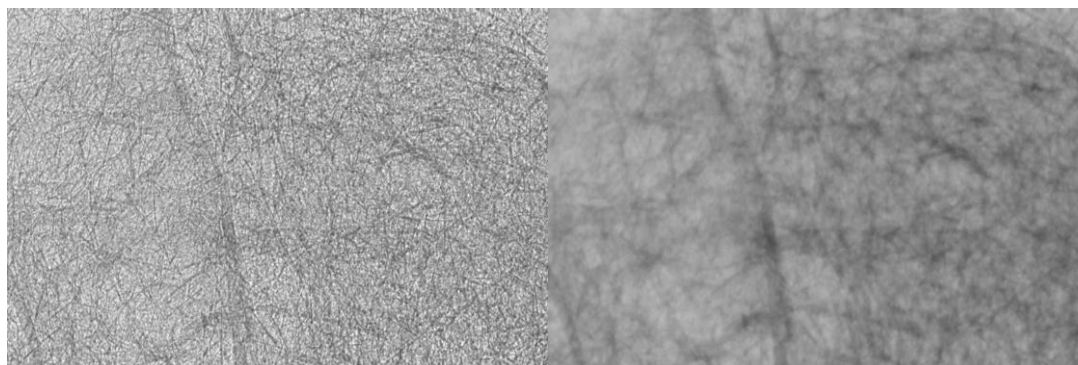
- **Gauss width.** The width of the Image Restoration smoothing kernel. Smaller values give a sharper edge. 0.1 is a good starting value.
- **Stabiliser.** This is added arbitrarily to avoid division by zero in the deconvolution calculations. Although in synchrotrons values of stabiliser of 0.1-0.2 are recommended, we find that in laboratory CT systems a value of 0.5 generally works well and is the default.

## 2.2. *The phase retrieval output options*

- **Autorange.** It is suggested that “Autorange” is selected with a margin of 10%. However whatever is selected regarding range, please note that after your first preview reconstruction with the PR transformed projection dataset, the first thing you will have to do is make a substantial change to the contrast limits at the NRecon output tab, since the units of PR are very different from attenuation coefficient used in standard absorption reconstruction. The numbers are much higher.
- **Preview.** This will show you in the image window the result of PR transformation of the single current projection image with the selected phase retrieval parameters. (It does not show a reconstructed cross-section.)
- **Retrieve.** Once you are ready to run with the chosen parameters to a full PR reconstruction, click on “Retrieve”. Then you will need to wait for several minutes – depending on the projection dataset volume – while a PR transformed copy of the projection images is written to a subfolder called “PR”. Please note the implications of this regarding disc space.

Clicking “preview” will – after a short pause – show the single current projection image after PR with the chosen parameters ( $\delta/\beta$  ratio). Note that this PR projection will appear smoothed, even blurry. Figure 7 compares a standard absorption projection

with its PR counterpart using  $\delta/\beta$  of 700. You will see later however that apparent blurriness in the PR image does not necessarily mean loss of detail in the reconstructed PR image. The apparent blurriness increases with larger values of  $\delta/\beta$ . Sometimes high numbers are needed to achieve the full PR effect (refer to figure 12).



**Figure 7.** A projection image (left) of a biomaterial scaffold (SkyScan 2214), and the corresponding phase retrieval (PR) projection (right) using  $\delta/\beta$  of 700.

### ***2.3. The main adjustment in PR is the “delta / beta ratio”***

Beta and delta relate to the mathematical core of the Paganin phase retrieval method – and the maths here gets serious. They are the imaginary and real numbers (!) of the estimated complex-valued X-ray index of refraction (Paganin *et al.* 2002, 2004)<sup>1, 7</sup>. There are references (websites) where values of beta and delta for various materials are published (see links to CXRO, XOP in references<sup>9, 10</sup>). However these values were obtained in synchrotron imaging and are context sensitive. Therefore the approach recommended is to use trial and error for the delta/beta ratio within a recommended range. Generally you should try values from 100 to 1000. Since the magnitude of the  $\delta/\beta$  ratio determines the strength of removal of edge enhancement, then the guide as to what number to try is the degree of edge brightening visible in the absorption reconstruction of the scan. Samples with surfaces with air at very high resolution showing the most pronounced edge brightening would require higher values up to about 1000 (or sometimes even higher), while samples either scanned at lower resolution or with less sharply contrasting edges and with less edge enhancement

visible, would work better with lower values down to about 100. If in doubt, start at 500.

Note that clicking “Preview” will only preview the current projection image. You must retrieve the whole projection dataset and then load the PR dataset, before you can make a cross-section preview and reconstruction. Therefore trial and error with different delta / beta values can take a while!

## **2.4. *Reconstructing the PR-transformed projection dataset***

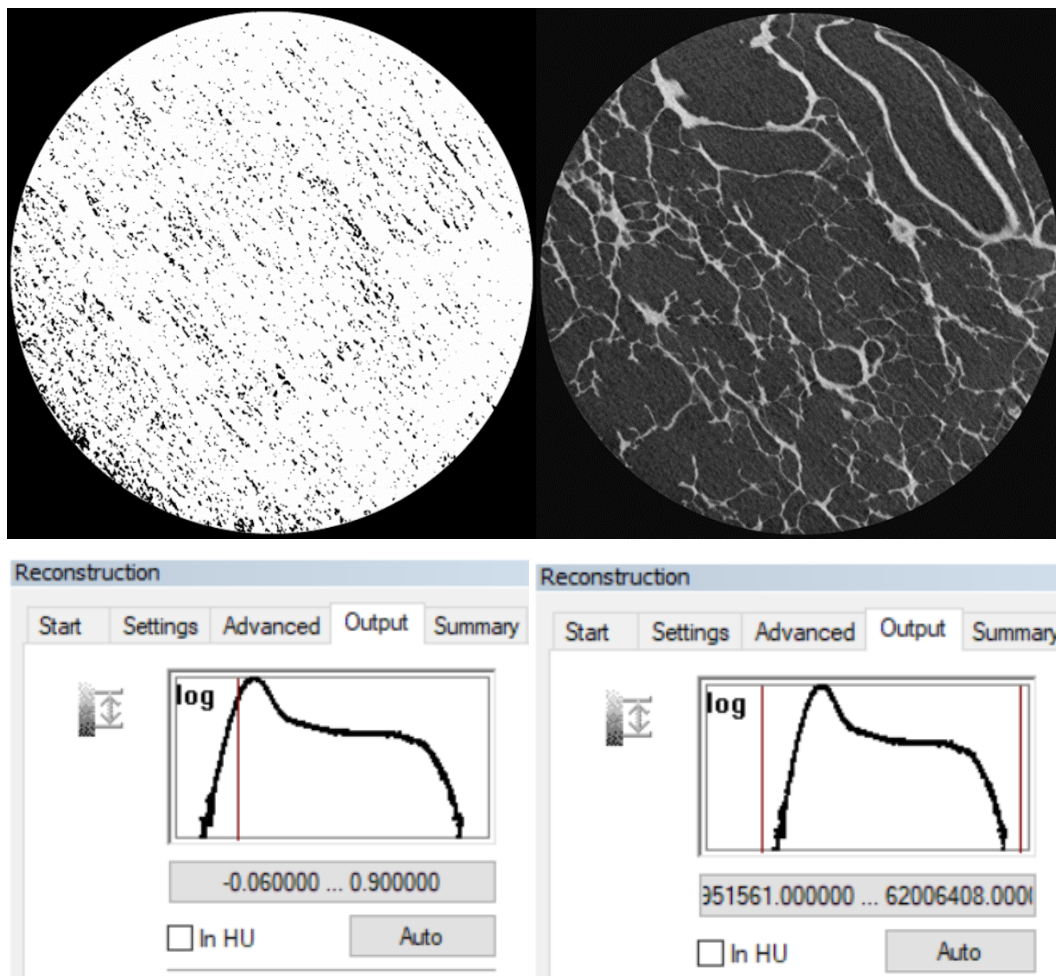
This description will assume that the reader already has some experience with standard reconstruction in Bruker NRecon software. (If not – please check out the help notes in NRecon, they’re quite extensive, and also the method note MN62, a complete overview of reconstructing with NRecon.)

First load the dataset created in the subfolder called “PR” into NRecon. Most of the reconstruction parameters don’t need to be changed from the ones you already optimized for the standard, absorption reconstruction. For instance, post-alignment correction, ring reduction, beam hardening, do not need to be re-visited. However if smoothing was applied, it might be better to set this to zero, since the PR method involves a significant smoothing (in the Fourier domain) and this means generally that no smoothing is needed for PR reconstructions.

But one setting will need to be changed – a lot. That is the histogram contrast range at the output tab. This is illustrated in figure 8. The first image that you see on running preview will probably look like a binary image – black and white, with no grey contrast visible. You might think that something has gone wrong – but it hasn’t. You just need to adjust the minimum and maximum limits – the vertical red lines on the “attenuation” histogram at the output page. You might find that both red lines are stuck together and need to be separated. You should place the contrast limits just above and below the new histogram range as shown in figure 8.



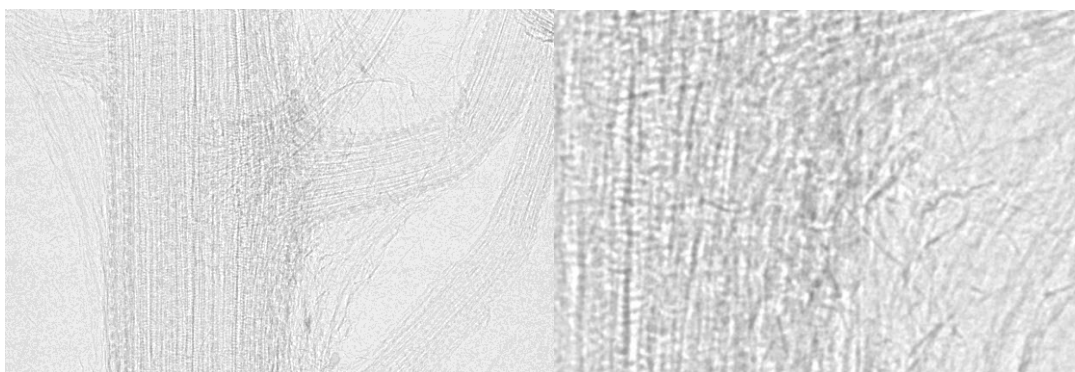
Note also that NRecon records in the scan log file the details of the PR reconstruction, including the  $\delta/\beta$  ratio, in between the acquisition and reconstruction sections.



**Figure 8.** The first preview of a PR projection dataset (upper left) will show a black and white image, with the histogram contrast limits (red lines) stuck together (lower left). This is because the attenuation numbers for PR are much larger than the usual attenuation numbers (normally less than 1). This is corrected (right) by moving the contrast limits to just above and below the new PR histogram range, as shown.

That's it! Once you have a right-looking preview, run the reconstruction in the normal way. You are now ready to get started with phase retrieval (PR) reconstructions of your scans. The kind of scans that are likely to give the most useful result from PR are:

- Scans at high resolution in systems with spot size 5 microns or less, and with either CCD cameras or scientific CMOS cameras with high pixel density (physical camera pixel 20 microns or less).
- Samples with air surfaces throughout
- Samples with low to medium density, up to about the X-ray density of bone or teeth
- Samples where you see in the projection image the characteristic white areas at object surfaces that indicate that edge small angle scattering is taking place; figure 9 shows this in the case of the scan of a plant root (SkyScan 2214).



**Figure 9.** White patches at object surfaces in scan projection images – such as in this scan of a small plant root – indicate the surface scattering edge enhancement that provide the basis for useful phase retrieval.

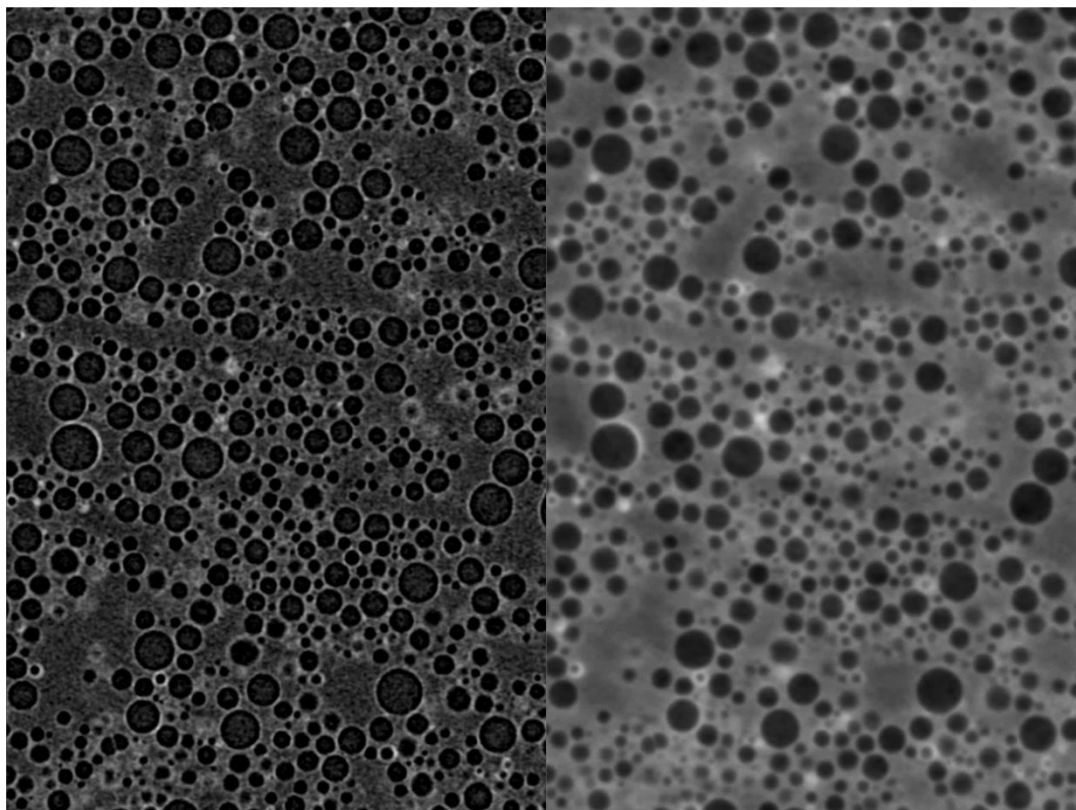
## **2.5. *Hint for scanning with the intention of PR reconstruction: reduce camera signal intensity a little***

As we have just seen, the edge scattering-enhancement phenomenon, which is the basis of PR, results in white looking patches at object surfaces in the scan projection images (figure 8). In some cases this local brightening of the projection image can push the local signal intensity close to or even above saturation. As we know from our microCT scan essentials – that must be bad. Therefore, when scanning with the intention of phase retrieval, it is useful to reduce slightly the camera signal strength. Remember when we are setting up a scan mode, that we look at an empty image of air only, with flat field removed, and using the profile line (right button click on image)

we adjust exposure time to get an average profile intensity of about 60%? For scans with PR as the intention, you should aim instead for 50% mean intensity – or even 45% if the edge enhancement effect is very strong. You can compensate for this loss of signal strength by increasing frame averaging.

### 3. Results

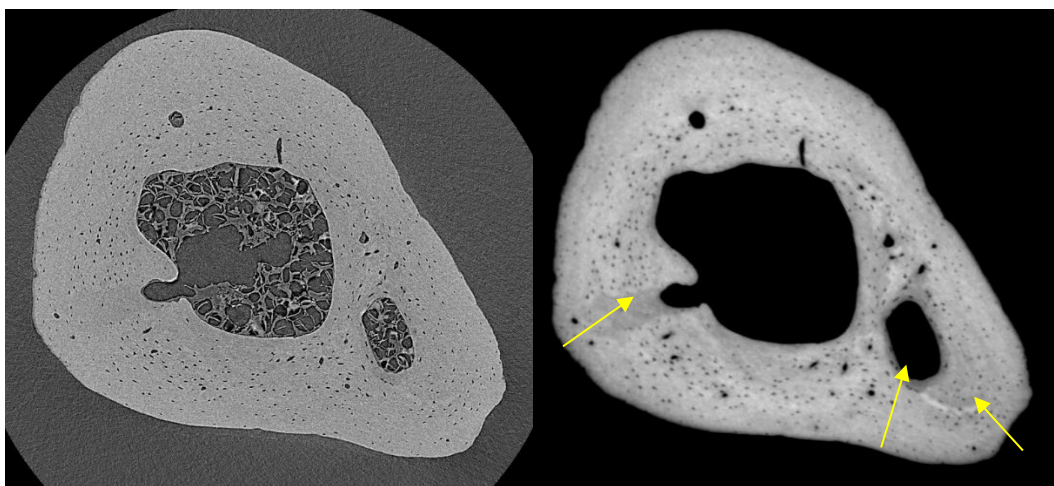
In the first example, a porous polymeric material containing embedded mineral is shown from absorption and phase retrieval reconstructions (figure 10). In the absorption image the spherical bubbles are well resolved and the edge enhancement seems to accentuate the porosity. However the background matrix appears fragmented and not solid. By contrast in the PR image the matrix is smoothly and solidly reconstructed and local densities and large scale structure are more clearly visible. The removal of edge brightening thus enhances visualisation of the material.



**Figure 10.** A scan (0.72  $\mu\text{m}$  voxel, SkyScan 2214) of a porous polymeric material containing embedded mineral, reconstructed from standard absorption projections (left) and phase retrieved projections (right). PR improves the visualisation of the matrix surrounding the

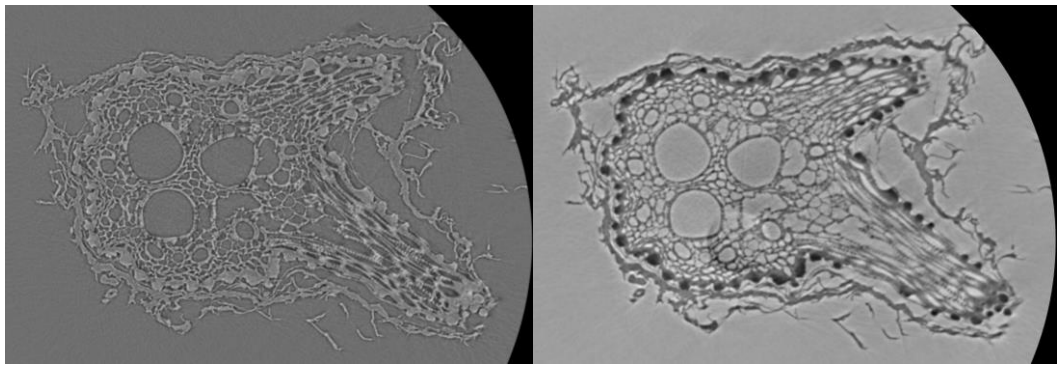
pores and density variation therein, while reducing edge brightening around the pores. Courtesy of Christopher Hansen, Mechanical Engineering, University of Massachusetts Lowell.

The following are the results of phase retrieval reconstruction of a number of further samples, ranging from very low density up to small bones. In figure 11 it is notable that PR can enhance visualisation of fine spatial variations in bone mineralization density. One can speculate that this may be due to edge scattering at porous surfaces within mineralized bone that are too small for spatial resolution. This and the other examples below are discussed in their figure captions.

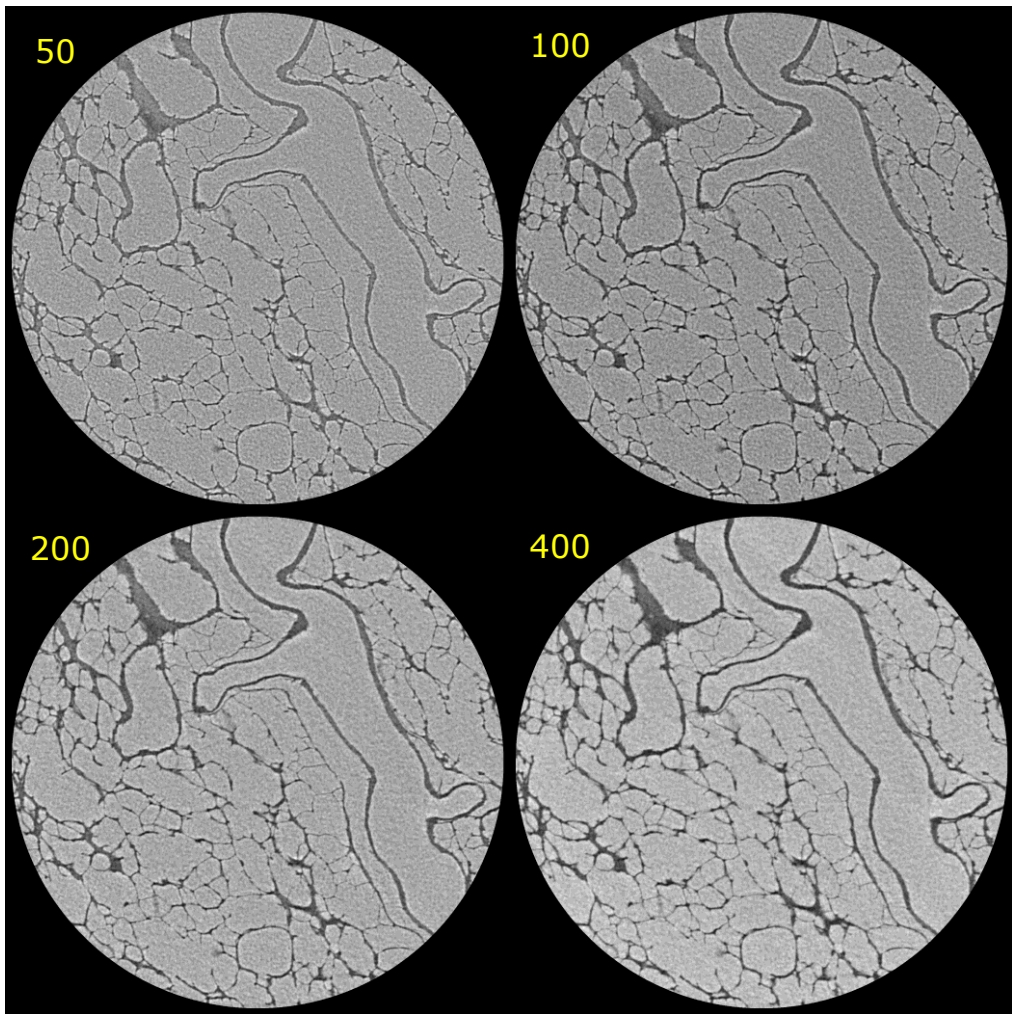


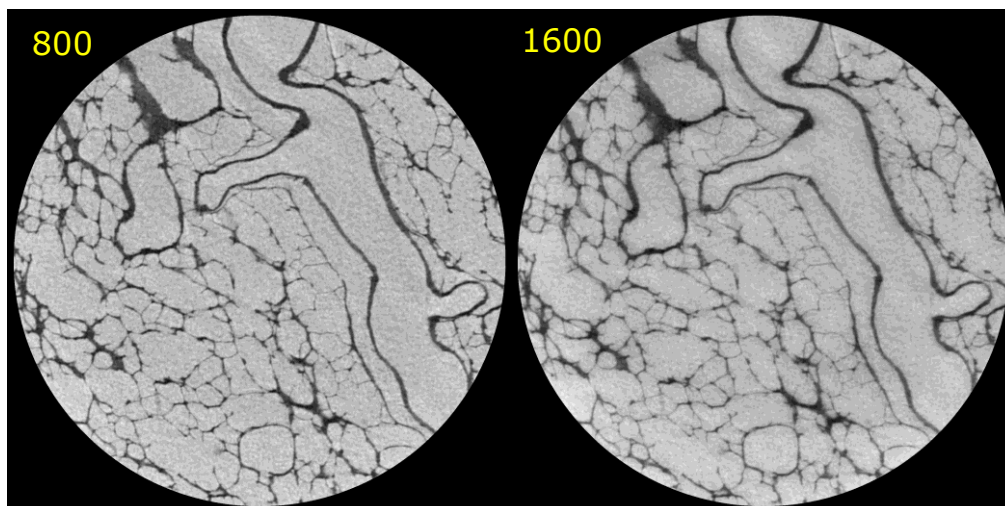
**Figure 11.** A mouse femur scanned at 0.5  $\mu\text{m}$  voxel in the SkyScan 2214: absorption (left) and phase retrieval ( $\delta/\beta$  500) reconstruction (right). Regions of both higher and lower than average mineralization (arrows) are more clearly visible in the PR images. Courtesy of Dr Claire Clarkin, Biological Sciences, Southampton University, UK.



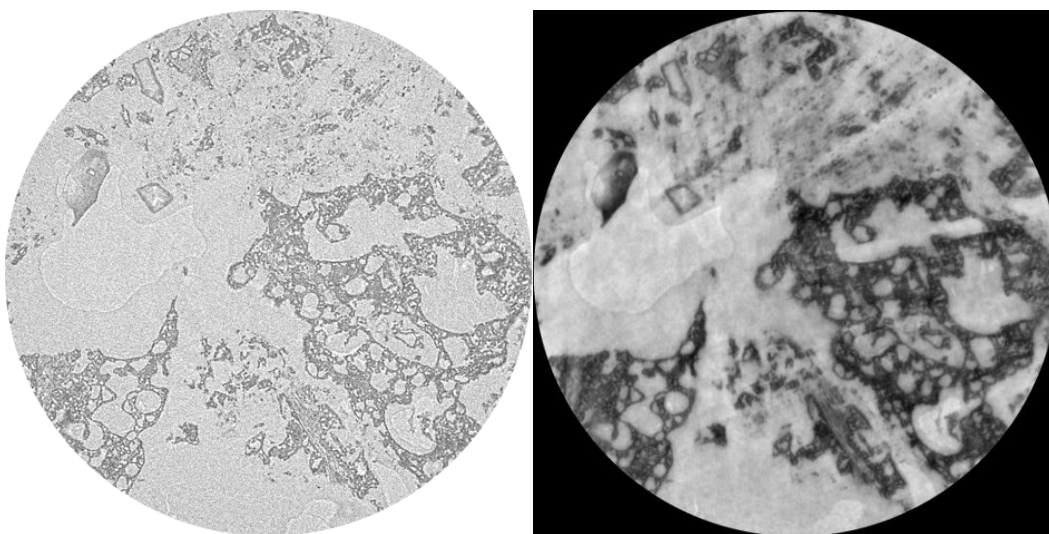


**Figure 12.** A root of *Sorghum bicolor* scanned at 0.17  $\mu\text{m}$  voxel size in the SkyScan 2214. Left: absorption reconstruction showing significant edge enhancement. Right: the phase retrieval reconstruction ( $\delta/\beta$  700). The higher density of the cell layer just under the surface is visible only in the PR image. Courtesy of Dr Rivka Elbaum, Faculty of Agriculture, Hebrew University of Jerusalem, Israel.

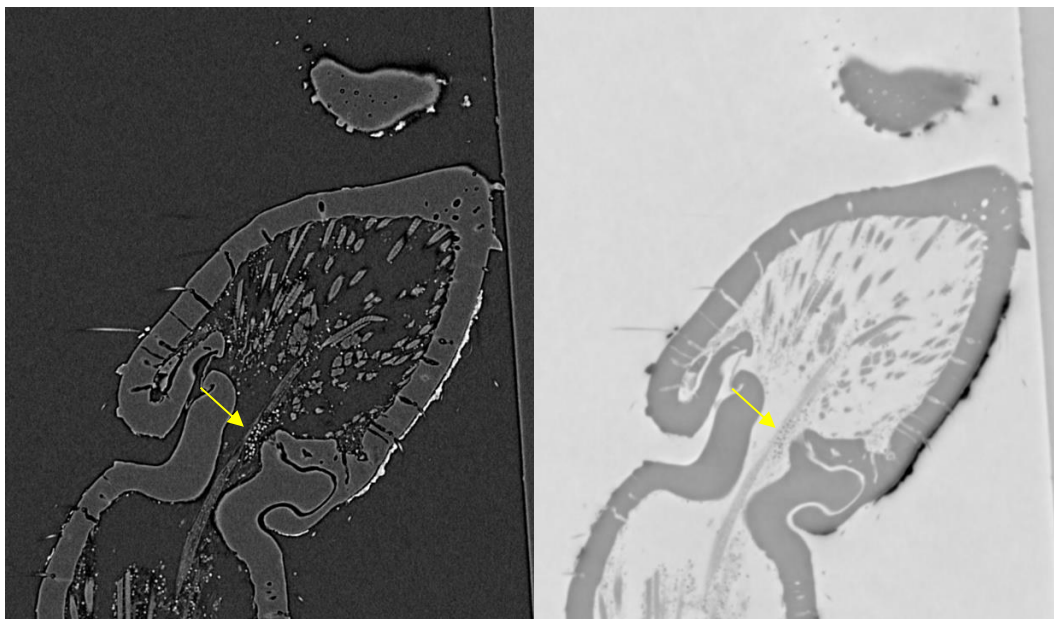




**Figure 13.** A lung sample ( $\text{OsO}_4$  stained) scanned in the SkyScan 2214 ( $0.65\ \mu\text{m}$  voxel) and reconstructed from PR processed projections. The result of  $\delta/\beta$  values from 50 up to 1600 are shown. With low values the surface edge enhancement is still visible, not being completely removed until a ratio value of 800. A value of 1600 is clearly excessive, leading to smearing artefacts. With values up to 800, the finest structures stand out increasingly well from background noise. Courtesy of Fabian Westhauser, Department of Medicine, Heidelberg University.



**Figure 14.** A wax embedded biomaterial scaffold sample, scanned with a voxel size of  $0.85\ \mu\text{m}$  in the SkyScan 2214. Again, on the left is the absorption and on the right the phase retrieval ( $\delta/\beta$  700) reconstructions. It is notable how the PR enhances regional contrast – such as of the air bubble top left, and shows more clearly the low density matrix in which small particles in the top right are embedded. Courtesy of Fabian Westhauser, Department of Medicine, Heidelberg University.



**Figure 15.** A spider leg (row 3), scanned in the SkyScan 2214, with voxel size 0.5  $\mu\text{m}$ . Left absorption and right phase retrieval ( $\delta/\beta$  500) reconstructions. The arrow indicates a nerve running between two leg segments which is more clearly and continuously visible in the PR image than the absorption. In the absorption image the edge enhancement – while giving the impression of sharpness – disrupts visualisation of tissues and regional densities. Courtesy of Dr. Ligia Rosario Benavides Silva from Museum of Comparative Zoology at Harvard University.

## 4. Conclusion

The phase retrieval method can provide enhanced contrast and material / tissue visualisation in a range of objects, limited by the extent of edge brightening from small angle scattering in the X-ray projection images. These include low-density polymers, plants, arthropods and other low-density materials including biomaterial scaffolds. Albers *et al.* (2018)<sup>11</sup> show how phase retrieval can enhance tissue contrast in embedded soft biological tissues using a desktop scanner (the SkyScan 1272). However more dense materials such as bone can also benefit from phase retrieval to enhance visualisation of contrasting structure and micro-density, based on extensive internal porosity in biological mineralised tissues.

Now it's over to you – to try out the PR method on some of your scans to see where it can be usefully employed to provide enhanced contrast and structural information.



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- <sup>10</sup> XOP: <http://www.esrf.eu/UsersAndScience/Experiments/TBS/SciSoft/xop2.3>, the web site of XOP (X-Ray Oriented Programs), a software suite for X-ray optical calculations including a database of X-ray optical properties (DABAX). XOP is maintained by two synchrotron light laboratories, the European Synchrotron Radiation Facility (ESRF) in Grenoble, France, and the Advanced Photon Source (APS) in Argonne, Illinois, USA.
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