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CoralCT: a platform for standardized analyses of growth parameters in coral skeletal cores

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Abstract

We present CoralCT, a software application for analysis of annual extension, density, and calcification in coral skeletal cores. CoralCT can be used to analyze computed tomography (CT) scans or X-ray images of skeletal cores through a process in which observers interact with images of a core to define the locations of annual density bands. The application streamlines this process by organizing the observer-defined banding patterns and automatically measuring growth parameters. Analyses can be conducted in 2- or 3-dimensional spaces, and observers have the option to utilize an automatic band detection feature. CoralCT is linked to a server that stores both the raw CT and X-ray image data, as well as output growth rate data for hundreds of cores. Overall, this server-based system enables broad collaborations on coral core analysis with standardized methods—and crucially—creates a pathway for implementing multi-observer analysis. We assess the method by comparing multiple techniques for measuring annual extension and density, including a corallite-tracing approach, medical imaging software, 2-dimensional versus 3-dimensional analysis, and between multiple observers. We recommend that CoralCT be used not only as a measurement tool, but also as a platform for data archiving and conducting open, transparent, collaborative science.
1. Introduction

Coral reef ecosystems are formed over thousands of years as calcium carbonate (CaCO$_3$) shells and skeletons of reef-dwelling organisms become cemented together. Scleractinian corals contribute much of the CaCO$_3$ building blocks on shallow tropical reefs, and are thus considered ecosystem engineers. In addition to harboring some of the highest concentrations of species in the marine realm, coral reefs provide ecosystem goods and services valued at billions of dollars annually, mainly through shoreline protection, tourism, and supporting fisheries (Costanza et al., 2014). On shorter, decadal timescales, the production of CaCO$_3$ by corals is also essential for reefs to keep pace with rising sea levels and to construct the 3-dimensional habitat that fosters biodiversity on reefs (Alvarez-Filip et al., 2009).

Coral calcification is under threat from ongoing ocean warming and acidification. As anthropogenic CO$_2$ emissions are absorbed into the ocean, the seawater carbonate system shifts in a predictable manner toward lower pH and lower aragonite saturation state ($\Omega_{Ar}$) (Caldeira and Wickett, 2003; Sabine, 2014). Short-term laboratory experiments have demonstrated that coral calcification declines under reduced $\Omega_{Ar}$ (Chan and Connolly, 2013), and extrapolations of the laboratory results to reef communities suggest that the impacts to calcification are substantial enough to limit the capacity of most of the world’s reefs to keep pace with rising sea levels throughout the remainder of this century (Cornwall et al., 2021). An outstanding question, however, is to what extent adaptive responses will enable corals to fare better as ocean acidification proceeds over the coming decades, relative to the responses inferred from experiments that are often weeks-to-months and only occasionally longer than one year (Comeau et al., 2019; McLachlan et al., 2022). Additionally, anthropogenic CO$_2$ causes warming via the greenhouse effect, and both laboratory (e.g., Schoepf et al., 2021) and field studies (e.g., Cantin et al., 2010;
DeCarlo et al., 2017) point to impacts on coral calcification under rising temperatures. Together, ocean warming and acidification place reefs at risk of transitioning from net calcifying to net eroding due in large part to the expected declines in coral calcification (Cornwall et al., 2021; Eyre et al., 2018). Arguably, the largest source of uncertainty in these projections is not based on uncertainties in future emissions or climate sensitivity, but rather arises from our lack of knowledge in whether long-term coral responses differ from the short-term responses observed in the laboratory.

Coral skeletal cores are the best available tool to probe long-term responses of corals to ocean warming and acidification. Massive coral colonies can live for centuries, or even up to a millennium in rare instances (Soong et al., 1999). Additionally, alternating patterns of high- and low-density bands formed on a yearly basis enable annually-resolved chronologies to be developed from skeletal cores (Knutson et al., 1972). Most crucially, the density bands can also be used to measure annual coral growth rates. Skeletal cores enable measurement of different annual growth parameters: extension is the length of skeleton accreted along the growth axis in one year (cm yr\(^{-1}\)), density is the mass of skeleton per volume of growth (g cm\(^{-3}\)), and calcification is the mass of CaCO\(_3\) formed per area per time (the product of extension and density, in g cm\(^{-2}\) yr\(^{-1}\)). While these characteristics make skeletal cores promising archives of the long-term coral responses to environmental change, their utility in advancing knowledge in this field is only as good as the tools applied to extract accurate information.

Skeletal cores have been used to investigate coral responses to environmental variability since the 1970s (Buddemeier et al., 1974; Hudson, 1981; Hudson et al., 1976). Their utility has progressed over time as new tools enabled more information to be extracted from cores. The early studies physically cut cores and imaged the density-banding patterns with 2-dimensional X-
radiographs (X-rays) and/or 1-dimensional gamma densitometry (De’ath et al., 2009; Lough, 2008). Conversely, computed tomography (CT) produces 3-dimensional datasets of density variations without the need for physical slicing. Bosscher (1993) first applied CT to coral skeleton blocks, and Cantin et al. (2010) later applied CT to reconstruct growth rates in skeletal cores. While CT allowed researchers to digitally cut 2-dimensional slices oriented along the axis of growth (Cantin et al., 2010), measurements made on a single digital slice are still inherently 2-dimensional (i.e., lacking components of growth into or out of the screen). Coral colonies grow in complex 3-dimensional orientations, and thus capturing 3-dimensional growth with CT scans may be beneficial compared to 2-dimensional slices that exclude growth directed in-to or out-of the slice. More recently, it has become possible to measure coral annual growth rates in 3 dimensions with automated tracking of the growth direction throughout an entire core (DeCarlo and Cohen, 2016).

Here, we describe CoralCT, a computer application for analyzing coral growth parameters in CT scans or X-rays of skeletal cores. CoralCT includes a graphical user interface for observers to interpret density banding patterns, and automated analysis of identified bands in 2-dimensions or 3-dimensions. To improve standardization in the field of coral core analysis, CoralCT is linked to a server that stores CT scans or X-ray images, enabling broad participation in interpreting cores and analyzing growth data.

2. Materials and Procedures

2.1 Materials

CT scanning is the best method for visualizing the density banding in coral skeletons due to the 3-dimensional nature of the resulting images. A CT scan is composed of a series of 2-dimensional axial images that can be assembled into a complete 3-dimensional reconstruction of
the sample. The CT image files are in the format of Digital Imaging and Communications in Medicine (DICOM, saved as .dcm or .dicom files). Each DICOM image contains metadata regarding the pixel spacing in each direction (i.e., a conversion between numbers of pixels and distance) and the slice location (i.e., the image’s position in along the z-axis in the series of images). Typically, the x- and y- pixel spacings within an image are equal, as is the spacing among images in a series, but in some cases the horizontal resolution within an image may be different from the vertical resolution in the image series. In any case, as long as these metadata are known, the 2-dimensional images can be assembled into a 3-dimensional reconstruction with measurable and accurate length dimensions. Once assembled, the pixels that compose the 2-dimensional images are referred to as voxels in the 3-dimensional image. The information contained within each voxel is a measure of density on the Hounsfield Unit (HU) scale. This scale was developed for the medical community and has a typical relation to absolute density such that -1000 HU is air (~0 g cm\(^{-3}\)) and 0 HU is water (1 g cm\(^{-3}\)). However, the crystalline and porous nature of coral skeletons causes a departure from the typical relationship. For example, DeCarlo et al. (2015) created a calibration between HU and coral skeleton cylinders of known bulk and reported a relationship of HU = density (g cm\(^{-3}\)) x 1485.5 - 768.9.

2.2. Procedure

2.2.1. Initial organization of CT scan data

When an observer chooses a core to load, CoralCT reads all the .dcm files within the provided data folder. The 2-dimensional images are read into successive layers of a 3-dimensional matrix. Once all image files have been loaded, CoralCT sorts the stack of images based on the metadata parameter “SliceLocation” (or “InstanceNumber” if SliceLocation does not exist in the
metadata). From the 3-dimensional matrix, a 2-dimensional “slice” of the scan needs to be displayed for an observer to begin visualizing the core. A slice need not be one voxel in depth, but rather it can have a thickness and is then projected onto a 2-dimensional screen by either taking the minimum, mean, or maximum of HU within the depth dimension of the slice (Fig. 1). Initially, the slice is placed in the center of the scan at 0° rotation, with a thickness of the number of voxels closest to 3 mm, and displayed as a mean projection.

Figure 1. Representation of (a) mean and (b) minimum projections in a coral skeletal core CT scan. In each of (a) and (b), a slice from the real CT image is projected on the right, and a schematic of how projected pixels are displayed is shown on the left. In each example, a number of voxels going into the page are represented on the front face (i.e., the 2-D projection), and the voxels have the exact same values in both (a) and (b). In (a), the voxels across depth into-the-screen are averaged together, while in (b), the minimum voxel in this depth is projected as the CT slice. In CT images, darker (lighter) shading indicates lower (higher) density.
2.2.2. Observer identification of annual density banding

The first step is for the observer to adjust the appearance of the CT image to best visualize the annual density banding pattern. This can include adjusting the (1) brightness, (2) contrast, (3) projection mode, (4) slice thickness, (5) slice location, and (6) slice rotation. The interface for adjusting these parameters in CoralCT is shown in Figure 2. Overall, the goal of adjusting the image is to view projection(s) of the core that best enable the observer to identify and trace annual density bands. As described in “Growth rate analyses” below, the parameters chosen for band identification do not affect the subsequent calculation of skeletal density. Brightness and contrast affect how HU are colored (in grayscale) on the screen, and they can be adjusted to optimize density band appearance. Generally, density bands become most clearly visible when brightness and contrast are adjusted such that the darkest pixels in low density bands are nearly black and some of the brightest pixels in thecal walls are nearly white. As shown in Figure 1, the projection mode affects how the depth of the slice is compressed into a 2-dimensional image. Typically, a mean projection works best, but in some cases a minimum projection helps sharpen the visibility of low density bands (Fig. 1). The slice thickness defines how many voxels in depth contribute to the projection displayed on the screen. This is illustrated by the red box in the axial view on the lower right of Figure 2. Increasing the thickness while in mean projection can improve the resolution of density bands due to greater replication of voxels in the averaging, however, if the bands are tilted in-to or out-of the screen, then increasing slice thickness will make the image blurrier. Decreasing the slice thickness will produce a sharper image, but it will also be noisier due to fewer voxels in the averaging such that the bands may become less visible. Slice location and slice rotation together define from where in the core the slice is drawn. Slice location moves the projection in-to or out-of the screen (i.e., moves the red box up or down in the axial view at the
bottom right of Fig. 2), while slice rotation enables the projection to be drawn at different angles (see magenta text in axial view of Fig. 2). Some cores, especially ones with tilted or wavy bands, may be difficult to interpret in some slice positions because the direction of growth can be partially in-to or out-of the screen. Adjusting slice location and rotation can help draw the projection where the annual bands are most visible. Additionally, changing the slice location and rotation are essential for mapping the annual bands in 3-dimensions, as described below.

**Figure 2.** Default display of a coral skeletal core in the CoralCT app. Sliders found on the right side of the screen, such as “contrast” and “slice location”, can be used to adjust the cores appearance to increase visual clarity of annual density bands.
Observers begin interacting with a core by first adjusting the image settings described above, and then zooming in near the top of the core. A zoom button appears when an observer moves their mouse over the CT image. Typically, an observer should zoom to see the top ~10 annual density bands. Visual interpretation of the banding is most effective when several bands are visible so that the regular banding pattern is apparent (Fig. 3). For example, between bands 8 and 9 of the core shown in Figure 3, there are thin bands that are distinct in both intensity and spacing relative to the regular marked annual bands, and thus these are clearly intra-annual bands that should not be identified. This may be less obvious if an observer was zoomed in so far as to only see the area between bands 8 and 9, without the ability to view the intra-annual banding within the context of the regular banding pattern of this core. Intra-annual banding may arise from sub-seasonal variability in temperature or light, or for some corals due to dissepiment formation that occurs on a monthly basis (DeCarlo and Cohen, 2017).

Once the annual banding pattern is understood, an observer begins identifying bands by clicking the “Identify next band” button. The text, “Next band will be band X” below this button indicates which band will be created or edited. If an observer wishes to work on a band other than the one indicated by this text, they may use the “Jump to band” input box and click the “Apply” button. Figure 3 shows the appearance during band identification mode. In this mode, an observer clicks multiple times on the selected band, and the application adds a white circle indicating each mouse click (as shown for band 11 in Fig. 3). The main rule when deciding which part of a band to click on is to be consistent. Observers may click on low-density (dark) or high-density (light) bands, the application will work with either approach, but generally the low-density bands are more distinct. For example, in Fig. 3, each band has a clear and relatively thin dark (low-density) band, but the light (high-density) band is not as consistent (e.g., between bands 9 and 10 there is a
fade from light to dark more than a distinct light band). Generally, clicking in the middle, or darkest part, of the low-density band is best for maintaining consistency. In Fig. 3, the bands have been identified as the dark, or most distinct, part of the low-density band rather than the “middle” of it (e.g., see bands 2 and 10). For these reasons, it is best to visually scan through at least 10 bands to gain confidence in understanding the banding pattern and where in the bands to click for consistency, before beginning to click on the first band. Observers do not necessarily need to click across the entire width of the core. In some cases, the banding may become less clear on one side of the image, or there may be pieces of core missing. The greater width of the core that is identified, the more of the CT scan is used in the growth-rate analysis, but there is no specific requirement to click across a certain width of each image.
**Figure 3.** Band-identification mode. The yellow digits indicate band numbers, and they are plotted at the center of where the existing 3-dimensionally interpolated bands cross into this slice. For clarity, the interpolated bands (which would be displayed as yellow lines) have been hidden in this image. On this core, the observer has identified bands as the darkest part of low-density bands. The observer has chosen to add tie-points to band 11, and the added tie-points are plotted as white circles. In the axial image toward the right, the current slice location is displayed by the red box, and white dots indicate the distribution of tie-points for the present band (band 11 in this case). The vertical streak of solid white in the axial view is the result of accepting an automatic band detection (see Section 2.2.3 below).

An observer must define band 1 at the top of the core, and subsequent bands must sequentially increase farther down the core. The bands must be defined to an extent that a line drawn orthogonal to the surface of one band will intersect the next band above it (see Growth rate analyses below for more details). At a minimum, this can be accomplished by defining bands on one slice, in which case the growth rate analysis will be 2-dimensional. Typically, observers will define the bands on multiple slices so that the bands—and growth rate analysis—are 3-dimensional. To do this, observers must define bands in at least one other slice at a different location in the core. This can be done either by adjusting the slice location or the slice rotation. Adjusting the slice rotation is generally the best approach because any new rotation will have intersection points of bands from the previous slice, enabling the observer to easily keep track of which density band corresponds with which numbered band (*i.e.*, yellow numbers in Fig. 3). However, for some cores, especially those with tilted bands, the annual banding pattern may not be clearly visible at all rotations. In these cases, it may be necessary to adjust the slice location.
These approaches are not mutually exclusive; an observer can change the slice location and rotation together or change one parameter at a time. In all cases, the observer is simply adding new tie-points to the 3-dimensional surfaces defining each density band. Similarly, it is not necessary to complete all the bands on one slice before moving to a new slice location or rotation. Bands can be completed in any order, and new points can always be added to a band that was previously traced on the same slice or a new slice.

If defining bands in 3-dimensions, observers must add enough tie-points to each band such that the interpolated bands effectively represent each density band. On slices in which an observer has defined bands, this will inherently be the case, but when the observer switches to a new slice location or rotation, the interpolated bands should all be good fits before the observer completes the core. A typical approach may proceed as follows. An observer defines all the bands on the first slice at rotation 0°. Then, the observer rotates the core 90° for the second slice, leveraging the intersection points from the first slice to ensure band numbers are assigned correctly (e.g., yellow numbers in Fig. 3). Next, the observer rotates to 45° and defines the bands before finally rotating to 135° and defining the bands yet again. When rotating to 45° and 135°, the initial interpolation of the bands as displayed may appear a poor representation of the underlying density bands, but this will improve as more tie-points are added. These 4 slices are a general rule of thumb for adding enough tie-points for reasonable 3-dimensional interpolations. However, the observer should rotate, or adjust the slice location, so that a new slice is displayed that was not previously used for defining bands. If the interpolated bands are a good representation of the underlying density banding (see Fig. 4), ideally on multiple new slices without adding tie-points, then the observer can reasonably conclude analysis of the core.
Figure 4. Finalizing band identifications. Similar to Fig. 3, this image shows an example of nearing the completion of band identification. The observer has defined bands across multiple slices to generate a representative distribution of tie-points (white points in axial cross-section toward lower right). In this image, the observer has switched to a new slice rotation that has not previously been used to add tie-points (i.e., notice lack of white dots within red box in the axial cross-section). Additionally, the observer has switched from showing interpolated clicks to showing smoothed bands (orange lines) because these smoothed bands are what defines banding for the growth rate analysis (see Section 2.2.4). If these smoothed bands provide a reasonable representation of the underlying density banding, the core can be considered complete. In this case, several of the bands (e.g. 1, 4, 6, and 11) could benefit from the addition of tie-points toward the right of the image—this makes sense considering in the axial cross-section the right side of the
slice includes the area of the core more sparse of tie-points, leaving some of the interpolated and smoothed bands (orange lines) needing additional tie points in that region.

CoralCT includes a variety of tools for editing identified bands. While adding tie-points in band-identification mode, an observer can click “Redo band” (Fig. 3) if mistakes were made. This will cross-out all the tie-points for the current band added on the current slice, and allow an observer to continue adding new tie-points. From the default view (Figs. 2 and 4), an observer can redo, add, or delete bands. Redoing a band means that all existing tie-points are deleted across all slices for that band, but the band’s position is retained (i.e., the numbers of all other bands do not change). Adding a band means that all bands below (i.e., higher number bands) the added band are increased by one band number. For example, if an observer adds a band below band 4, then bands 1-3 are unchanged, previous band 4 becomes 5, previous band 5 becomes 6, etc., and a blank band 4 now requires tie-points. Deleting a band decreases the band number of all bands below the deleted one. For example, if an observer deletes band 7, then previous band 8 becomes 7, previous band 9 becomes 8, etc.

2.2.3. Band auto-detection

CoralCT includes an optional feature that can automatically define low-density bands (Fig. 5). This function works by first filtering the slice image to remove small-scale (i.e., pixel-to-pixel) variations in density. An observer may adjust the settings of this filtering step by using the “Band-detection settings” button, so there is not a specific set method for this filtering step, but it is similar to the smoothing described below in Section 2.2.4 (Growth-rate analysis). Next, the auto-detection feature requires a starting point: this is either an observer’s first mouse click on the band if it is a
new band that has not been previously defined, or the starting point is the mid-point of the
intersection of the interpolated band if this band has previously been defined in at least one other
slice. The intensity of the banding pattern is defined by calculating the density range in a search of
pixels within 3.5 mm upward and downward of the starting point. Then, the app searches both to
the left and to the right of the starting point, and at each pixel-step the minimum density in the
filtered image is located vertically, within 1 mm of the previous position of the auto-detected band.
Finally, the intensity of the band at this new location is again calculated as the density range in a
search of pixels within 3.5 mm upward and downward of this newly identified low-density point.
If this band intensity is less than half that of the intensity at the starting point, the new point is
rejected and searching in its direction (left or right) ceases. Qualitatively, this process extends the
low-density band identified at the starting point horizontally as long as the band extends with
similar intensity. The filtering step is designed to prevent the auto-detection from following small-
scale variations such as low-density centers of corallites (see Fig. 6), which are the skeletal tubes
constructed by each coral polyp. In practice, the auto-detection can work reasonably well on cores
with clear density banding patterns, especially cores from the genus *Porites*. The auto-detection
feature may not produce good results on other genera that have larger corallites than *Porites*, or on
cores with cracks or especially curvy bands. Overall, the auto-detection feature is intended to
enable faster processing of cores with clear density bands. It is not meant to supersede an
observer’s judgement or to be used in all cases without visual confirmation that the result is
appropriate. An observer always has the option to reject the proposed automatic-band detection or
to disable this entire feature.
Figure 5. Example results of an automatically detected band. In this example, the starting point was based on the first mouse click by an observer, and the magenta circles indicate the position of the automatically detected band. The stepwise changes in vertical position are due to the resolution of this CT scan, but the filtering step in the growth-rate analysis will produce a smooth band regardless. An observer could also choose to reject the auto-detected bands and either try again (with a different starting point) or click manually. But in this case, the auto-detected band fits well with the underlying density banding and can be accepted.

2.2.4. Growth rate analyses

Once an observer finishes adding tie-points to all density bands in a core, the CoralCT app automatically completes the remaining analysis of growth rate parameters. First, the 3-dimensional
image stack must be classified as the core versus surrounding air. A key distinction in this step is that the air within pore spaces inside the skeleton must be classified as core because the pore spaces affect the bulk skeletal density. In this sense, the term “porosity” is technically the most appropriate, but we will continue to use the term skeletal density to be consistent with previous literature, noting that this refers to the bulk density of skeleton and air within the core. To accomplish this, we identify a threshold HU that separates skeleton from air, but we first filter the CT images to effectively blend together skeleton and interior pore spaces (Fig. 6).

**Figure 6.** Image filtering process. An axial slice (a) is smoothed by passing over it a round filter with width of 4 mm and standard deviation of 20 mm (b). As the filter moves over the raw image, each new pixel in the smoothed image is calculated as a weighted average of neighboring pixels, with weights defined by the filter. Panel b shows the relative weights (colors) and the red line indicates the single pixel that would be calculated from these relative weights and the HU of the neighboring 4x4 mm area. Corallite centers are visible in (a) and (b) as low-density areas approximately 1 mm in width for the genus *Porites*. The smoothed image (c) does not contain the small-scale density variations associated with corallites that are visible in the raw images, and this smoothed image can be used to create a binary map of pixels inside versus outside of the core.
This smoothing step is applied sequentially to all axial slices throughout the entire CT scan. Each smoothed image is converted to a binary image (within versus outside of core) using Otsu’s method (Otsu, 1975), which identifies a threshold between groups such that the variance in pixel values within each group is minimized. The binary image is then used to crop the raw data (Fig. 7), and the subsequent analyses described below work from this cropped image. In other words, the filtering step is used to crop the area representing the core, but all growth-rate calculations are based on the HU of the original CT scan data, not the smoothed images. This process is effective at identifying the circular shape of the core in each axial slice, but also in excluding where there are substantial pieces of core missing, such as in cracks between different pieces of core (Fig. 7). As a result, analyses can proceed across cracks between core pieces—as long as they are fit together such that extension measurements are unaffected—because the density calculations will be based only on voxels that represent skeleton or pore space, not including air gaps within a crack.
Figure 7. Raw images (first column) are filtered (second column) before conversion to binary (third column). The binary images are used to crop the raw data (fourth column). The series of images shown here are axial slices displayed at 2 mm intervals across a break between core pieces.
The cropping process effectively retains only pixels with core data, excluding areas around the core, even across the break between core pieces that are held together by electrical tape.

The second main step in the growth-rate analysis is to interpolate and smooth the user-defined density bands (Fig. 8). For each band, an observer should have identified tie-points in multiple slices. When viewed in an axial orientation, the tie points for each band can be plotted spatially in the horizontal direction, and the value of each time point indicates the vertical position within the CT scan (Fig. 8a). These tie-points can then be used to interpolate a continuous 3-dimensional surface representing that density band (Fig. 8b). Finally, the interpolated surface is smoothed (Fig. 8c), using the same process as for the core detection shown in Figs. 6-7. These steps are repeated for all density bands defined by an observer.

**Figure 8.** Interpolation and smoothing of observer-defined annual density bands. (a) Tie points plotted as an axial view and colored by vertical position (relative to the lowest tie-point). (b) 2-dimensional interpolation of the band vertical position from tie-points (black), producing a 3-dimensional band surface. (c) Smoothed band after passing an image filter over it.
Annual growth parameters are measured between successive pairs of observer-defined density bands. This means that for \( n \) density bands defined by an observer, there will be \( n-1 \) annual measurements of each growth parameter (density, extension, and calcification). Extension is measured as the distance between successive density bands, with the growth direction orthogonal to the surface of each band. To implement this, CoralCT simulates 5,000 corallites extending from the lower band to the upper band (Fig. 9). The corallites are placed randomly on the lower band, and each corallite extends as a vector in a direction orthogonal to the surface of the band at its starting point. This process simulates corallites in coral skeletons, which are likewise oriented perpendicular to density bands (e.g., see Fig. 1). Because the corallites must connect from the lower band to the upper band, they can only be measured where the two bands overlap (see Fig. 9b,c). Extension rate (cm yr\(^{-1}\)) is measured as the mean of the central 90% of corallite lengths (\( i.e., \) the lowest 5% and highest 5% are excluded). The shortest and longest corallites are excluded because the distribution of their lengths will not necessarily always be normally distributed, and there is more potential for anomalously long corallites if relatively small bumps in an observer-defined band artificially lower the angle of corallite propagation. The observer-defined bands are smoothed (as described above) to alleviate this issue, and the removal of the outer 10% of corallite lengths is a second step in excluding these outliers. Because each individual corallite is a separate measurement of linear extension rate, we can also take the standard deviation of corallite lengths as a measure of uncertainty in each year’s extension rate.
Figure 9. Measurement of annual linear extension. (a) Illustration of simulated corallites (black lines) between two successive observer-defined density bands (colors). The gray and black boxes correspond to panels b and c, respectively. (b,c) Two-dimensional views of corallites drawn orthogonally from the lower band (red) to the upper band (blue). In (a), only 100 corallites are plotted for clarity (compared to the 5,000 used in calculations), and in (b,c) a corallite is plotted every 1 mm for illustrative purposes.

Density is calculated from the HU of voxels between successive pairs of density bands. In other words, CoralCT identifies each voxel that is located between each pair of density bands, and averages the HU of all of those voxels. HU are converted to density (g cm$^{-3}$) using the equation in DeCarlo et al. (2015) as a default, but this equation can also be revised if core-specific density calibrations exist (see Section 2.2.7 on metadata). Each voxel is not a separate measurement of
annual density because there are known differences in density within each band, especially due to
corallite centers versus thecal walls. Thus, there is only a single measurement of annual density
(the mean of all voxels between bands), and it is not possible to estimate an uncertainty of density
per year.

Calcification (g cm\(^{-2}\) yr\(^{-1}\)) is simply the product of annual extension and annual density. In
CoralCT, the uncertainty of annual extension is propagated to uncertainty of annual calcification
rate, but there is also an unquantified uncertainty of density that, in theory, makes calcification
uncertainty greater than as presented by CoralCT.

2.2.5. Analysis in 2-dimensions, including X-ray images

CoralCT works under the same framework in 2- or 3-dimensions. The steps described
above are for the intended purpose of 3-dimensional analysis. However, if an observer only
identifies density bands on one slice and then processes growth rates, CoralCT will treat the growth
rate analysis in a similar manner but without the 3\(^{rd}\) dimension (in-to or out-of the screen). In other
words, the linear extension would be defined similar to the schematics shown in Figure 9b,c rather
than Figure 9a. Additionally, density would be calculated from pixels between the 2-dimensional
bands.

The flexibility of CoralCT in being able to perform 2-dimensional analysis means that it
can also be applied to X-ray images of physically sliced coral skeletal cores. X-rays are added to
CoralCT as image files and the observer interface (Figs. 2-5) is the same, except without the option
to change slice thickness, location, or rotation.

2.2.6. CoralCT organization
CoralCT is integrated with a file transfer protocol (FTP) server. The application runs locally on an observer’s computer, but all files are stored on the server. This setup enables any observer to work on any coral skeletal core that has been added to the CoralCT server. When a core is chosen, the CT scan or image data are downloaded from the FTP server and then automatically opened in the CoralCT app. Likewise, all observer-generated files are sent back to the server. These include the locations of observer-defined density bands, and any output growth-rate data. All growth rate data are publicly accessible via download through the CoralCT app, unless the owner of a particular core has chosen to restrict access.

Use of CoralCT, whether to analyze cores or download datasets, requires creating a username and password linked to a verifiable email account. This requirement enables several features. First, core owners can limit access to growth-rate datasets of their cores to a specific username. Thus, there should be no concern to adding cores to the CoralCT server before publication, and then the data can be released for public access upon publication. Additionally, keeping track of which datasets were generated by which observer enables multi-observer analysis, and potentially observers could be weighted in such an analysis by a skill metric; this is not done in CoralCT, but could be done offline because of the CoralCT user-based system. Finally, when observers process a core, they are prompted to rank the clarity of the density banding pattern on a qualitative scale of 0-10, which could allow subsequent weighting of cores based on a metric of relative quality of bands (i.e., difficult-to-interpret cores should not necessarily be excluded, but arguably should contribute less to a site- or region-average since the data are less certain).

CoralCT enables observers to save their work on a core and return at a later time to continue their progress. Whenever an observer either submits a core for growth-rate analysis or clicks the “Save and exit” button, their work (i.e., their banding maps) are saved on the server by username.
If that same observer selects that core at a later time, their progress will automatically be downloaded from the server and the core will open as they last left it. Additionally, because downloading CT scans can take tens of minutes, there are two ways to avoid repeatedly downloading the same core to work on it across multiple sessions. Observers can click the “Open last scan” button on the main menu to load the core they most recently worked on (Fig. 10). This circumvents downloading the core data from the server again, and rather opens the most recent CT scan in the local directory. By default, CT scans or X-ray images are saved in a temporary folder that is erased each time an observer chooses a new core to download from the server. The reason for this is that the CT scan data in particular are large enough that an observer may fill their local hard drive after working on multiple cores. However, an observer may use the “Save CT data on your drive” checkbox to have each downloaded core saved in a separate local folder by core name (Fig. 10). This will allow the observer to later use the “Open saved scan” button to choose any of the core folders they have previously worked on to open it again for band identification without the need for repeated downloading from the server (Fig. 10).

When an observer submits a core for growth-rate processing, the observer must define the calendar year represented by the space between their first two defined density bands. CoralCT will display the core’s collection month and year when the observer is making this decision. Density-banding, while annual, does not necessarily coincide with calendar years. For example, the skeleton formed from one low-density band to the next may represent January-to-December, but it could also represent June-to-May or any other 12-month period of the year. The observer must interpret the core and their identified banding to estimate which calendar year overlaps most with their upper-most year. For example, if an observer identified a first band very nearly at the top of the core, and the core was collected in January 2018, the first year should be considered 2017.
Similarly, if there is a partial year of skeleton accreted above the top-most identified band and the core was collected in June 2013, the first year should be considered 2012. In other cases, the decision can be more difficult. For example, in Figures 3-4, the first observer-defined band is very nearly at the top of the core, with perhaps one month of growth above it (i.e., approximately 10% of the typical band distance is visible between the top band and the top of the core), but this core was collected in August 2019. Thus, the most appropriate choice is to consider the first band to be 2019 because it appears to include the majority of 2019, but this is a narrow majority (an estimated 7 months).

There are several additional organizational features of CoralCT. Observers can add notes in two different ways. While identifying density bands, observers can add notes that will be visible to other observers of the same core. For example, an observer may make note of their interpretation of intra-annual banding (e.g. Fig. 3) or comment on which settings produced the clearest views of banding patterns. Separately, an observer may add notes to a separate dialog box when submitting a core for growth-rate processing. These processing notes will be added to the growth-rate output files and may contain information such as whether the bands became difficult to interpret beyond a certain point, whether additional work is required on the core, or how the observer interpreted the calendar year of the first band. Finally, while identifying bands, an observer can save screenshots of the core, or view screenshots saved by other observers. As with the first type of notes, the screenshots provide a mechanism for sharing advice or providing explanation of interpretation among observers. The screenshots can be annotated with text boxes to facilitate this record keeping. All of these note-sharing features are optional.
Figure 10. Main navigation screen of CoralCT. This image shows the general application appearance immediately after logging in, and in this case the dropdown menus have been used to select core “T900” from Dongsha Atoll in the South China Sea. Clicking “Open CT scan” would load the core into the screen shown in Figures 2-5.

2.2.7. Metadata standards

All core submissions must have required metadata to be used in CoralCT. Required metadata includes genus, latitude, longitude, and collection year as absolute minimums. Ideally, metadata also includes water depth and collection month. Optional metadata may include core-specific density calibration parameters (with slope and intercept in the form of HU = density x
slope + intercept), whether the scan is a piece of a core that scanned in multiple sessions, and whether the scan needs to be flipped upside down to be oriented correctly.

CT scans typically contain certain metadata required as part of the file format. CoralCT requires “SliceLocation” (defines the vertical position in the image stack), “PixelSpacing” (defines the image resolution), and both “RescaleSlope” and “RescaleIntercept” (defines how the image data are converted to the HU scale).

X-ray images must be in .tiff format, and required metadata include whether the image is an X-ray positive or negative, and the resolution of the image (in dpi, or digital pixels per inch). While CT scans can all be placed on the standard HU scale for conversion to absolute density units (g cm$^{-3}$), X-rays will depend on the nature of how the X-ray was taken and the image was saved. For absolute density data to be extracted from X-rays, each image must have a calibration between density and grayscale of the image. Previous works have often placed sets of standards of known density in X-ray images to accomplish this. If no density calibration exists, X-ray images can be used only for linear extension measurements, but not density or calcification rate.

2.2.8. Submitting CT scans or X-ray images to the server

CoralCT can only be used on cores that have been added to its server. A key motivation of CoralCT is to enable broad collaborations and multi-observer analyses of coral skeletal cores (see Section 5), and a pre-requisite to achieving this goal is for all available skeletal core CT scans or X-rays to be shared. Users can click the “Submit data” button on the main menu (Fig. 10), and from there choose whether they are submitting a folder of DICOM files, a zipped file containing DICOM files, or an X-ray image. Core data submissions should also include required metadata.
(Section 2.2.7), submitted as a spreadsheet, preferably in the template provided for CoralCT (Supplement 1).

2.2.9. Accessing output growth parameter datasets

Output growth parameter data are saved in a standardized format as a text file (see Supplement 2). Users may access datasets by clicking the “Access data” button on the main menu (Fig. 10), which then shows dropdown menus that can be used to select the desired dataset. These output data can be downloaded per individual observer of a specific core, all observers of a core, or all observers of all cores in a region. In any case, a separate text file is downloaded onto the user’s local computer for each observer-core combination.

2.2.10. Additional instructions, user guide, and application updates

A more detailed description of how to use every feature of CoralCT is provided in a separate user guide (Supplement 3). The most up-to-date version of these instructional materials, as well as the CoralCT application itself, can be found at www.coralct.com.

3. Assessment of the method

We compare multiple approaches to quantifying annual extension and density in a ~70 year-long coral skeletal core (“T900”) collected from Dongsha Atoll in the South China Sea. These approaches include (i) the new CoralCT framework described here, (ii) an earlier version of the program (“coralCT”; DeCarlo and Cohen, 2016) that worked by tracing corallites throughout skeletal cores instead of the orthogonal simulated corallites we introduce here, (iii) two simulated X-rays images, which are each analyses conducted on one slice at a single rotation (the two
analyses were done at 0° and 90° rotations), (iv) using medical imaging software as has been used previous in coral growth studies (Cantin et al., 2010; Carilli et al., 2012), and (v) using CoralCT but by two additional observers from all the other analyses in (i)-(iv). The medical imaging software Horos was used for (iv), which works the same as the software Osirix used in other coral growth studies (e.g., Carilli et al., 2012). In Horos, we used the measure tool to draw a single line connecting each successive pair of density bands, and HU were extracted for density along this 1-pixel wide line. We do not separately present calcification rate results here because calcification is simply the product of annual extension and density.

The core exhibited multi-decadal variability in extension rate, and these large-scale variations were broadly captured by all the techniques (Fig. 11) but with different levels of variance and noise. The orthogonal simulated corallites (i.e., CoralCT) produced similar results to the previously-described corallite tracing method (DeCarlo and Cohen, 2016), differing on average by only 1.8% among the 76 years measured (i.e., the mean of absolute values of percent differences between the two 76-year time series). CoralCT also produced consistent results whether it was used in 3-dimensional mode or on single 2-dimensional slices (i.e., simulated X-rays), with average differences of only 1.6%. Conversely, there were larger differences (6.3% on average) between CoralCT and medical imaging software. Between Observer 1 and Observers 2 and 3, there were average differences of 11% and 3%, respectively, with maximum annual differences of 69% and 36%, respectively.

Annual density differences between CoralCT and other techniques followed similar patterns as extension rate (Fig. 12). Density derived from corallite tracing and from simulated X-rays differed from CoralCT by 1.6% and 1.3%, respectively, on average among years. The difference between CoralCT and the medical software Horos was larger, at 4% on average.
Between Observer 1 and Observers 2 and 3, there were average differences of 2.8% and 0.9%, respectively. These differences among observers are a result of differences in the locations within the core that each observer defined the annual density bands. For instance, Observer 2 defined a large-growth year at 1952, whereas the other observers interpreted this as two smaller-growth years, 1951 and 1952. There was anomalously high density in the portion of core that Observers 1 and 3 defined as 1951, but Observer 2’s banding definition included this density peak within 1952. As a result, the portions of core prior to ~1952 began to be assigned to different years among observers, causing the density time series to be effectively lagged relative to each other, and this created relatively large differences in annual density up to 13%. 
Figure 11. Assessment of annual extension rate measured with various techniques in core T900.

(a) Time series of raw annual extension rate, (b) time series of percent differences between the CoralCT method presented here and other techniques, (c-d) histograms of percent differences between the CoralCT method presented here and other techniques. Solid and dashed lines in (f) approximate the separate histograms for Observer 2 and 3, respectively. X-rays refer to simulated X-rays (i.e., single 2-dimensional slices).
Figure 12. Assessment of annual density measured with various techniques in core T900. (a) Time series of raw annual density, (b) time series of percent differences between the CoralCT method presented here and other techniques, (c-d) histograms of percent differences between the CoralCT method presented here and other techniques. Solid and dashed lines in (f) approximate the separate histograms for Observer 2 and 3, respectively. X-rays refer to simulated X-rays (i.e., single 2-dimensional slices).

4. Discussion
CoralCT fills an existing gap in infrastructure for standardized, collaborative, and archived analysis of growth rates in coral skeletal cores. The flexibility in the user interface and automated processing allows CoralCT to be used on cores from any coral genera, scanned with any CT instrument, and interpreted for banding patterns by anyone with a computer connected to the internet. Perhaps most crucially, CoralCT archives all raw CT scan and X-ray image data, as well as observer density-banding maps, and makes publicly accessible the calculated growth rate data for all cores (unless the core owner restricts access prior to publication).

Among the methods performed by a single observer, the annual extension and density results were broadly similar (Figs. 11-12). The earlier version of coralCT (DeCarlo and Cohen, 2016), which worked by tracing corallite paths between annual density bands, was an effective technique for ensuring the 3-dimensional orientation in which extension rate was measured represented the growth direction of the coral. However, the downside to the corallite-tracing approach is that corallites are not always traceable in relatively low resolution CT scans (i.e., pixel spacing of 0.2 mm or greater) or in coral genera with complex skeletal architecture such as *Orbicella*, *Diploria*, or *Diploastrea*. The simulated corallites presented here in the new version of CoralCT produced extension rates within 2% of the traced corallites, suggesting that the simulated-corallite method is representative of the orientation of growth and effectively captures annual linear extension. A key advantage of the simulated corallites is that they are simply extended orthogonal to the observer-defined bands, thereby offering a more flexible approach that works equally well across different CT scans and coral genera.

We compared 3-dimensional and 2-dimensional analysis by measuring growth on a 3-dimensional CT scan and on simulated X-rays (i.e., single 2-dimensional slices) of the same core. The 2-dimensional analysis produced extension rates that were on average within 2% of the 3-dimensional analysis.
dimensional analysis, although some years had differences as great as 5%. Since 2-dimensional and 3-dimensional analyses captured the main multi-decadal variations in extension and density (Figs. 11-12), both approaches appear effective means of analyzing coral growth parameters. However, it is possible that for other cores not analyzed here, the differences could be larger. Additionally, an advantage of 3-dimensional analysis is that in some cores there may not be a single slice in which all of the density bands are clearly visible, which then requires band identification across multiple slices in different rotations or locations in the core. Thus, 3-dimensional analysis offers an overall better representation of the growth orientation in skeletal cores and in some cases will be needed to effectively identify all density bands as the growth direction changes downcore. However, cores with clear banding visible in one slice may be effectively analyzed in 2-dimensions by defining bands on that single slice, an approach which can substantially reduce the time required to process growth parameters.

Although analysis using medical imaging software captured the large-scale patterns in extension rate and density, there were important deviations from the CoralCT-based methods (Figs. 11-12). Extension rates differed by more than 6% on average, with differences as large as 30%. Similarly, annual density varied by 4% on average between the two methods, but with deviations reaching 20%. The main advantage of the medical software is that growth rates can be measured more rapidly than in CoralCT, but this approach also carries several disadvantages. Since measurements are made along a single line, noise is introduced into the results: for extension rate the observer’s choice of where laterally along a band to measure growth can influence the distance between bands, as can the observer’s ability to properly make measurements orthogonal to the density bands. For density, the single-line measurement will pass through low-density corallite centers and high-density thecal walls, adding variance to the bulk annual density measurements.
Drawing multiple measurement lines may reduce some of this noise, but that also reduces the measurement-speed advantage of this technique. Additionally, the largest drawback of using medical software is that current techniques do not allow traceable analyses that can be edited or inspected by other observers.

Multiple observers have greater potential to produce different results compared to the different measurement techniques. In particular, if observers identify different numbers of bands, then growth parameter time series from one observer will become lagged relative to another observer for all years prior to the year that was counted differently. In the analyses presented here, Observers 1 and 3 generally agreed well, with annual extension and density within 3% and 1%, respectively. However, Observers 1 and 2 had much larger mismatches, with extension varying on average by 11% and density prior to 1952 clearly being lagged between the two time series and with differences as great as 13%. Comboul et al. (2014) demonstrate how age-counting errors in coral cores hinder the ability to reconstruct interannual to decadal scale variability. Additionally, Comboul et al. (2014) present a method for optimally correcting counting errors, but doing so depends on making an assumption about the probability of miscounting each band, a quantity which is unknown and likely varies greatly between cores. Alternatively, analyzing cores with three or more observers enables cross-dating techniques (see Section 5 below) or more simply the identification of outlier observers or mismatched years (e.g., by identifying density lags, as described above in Section 3).

5. Recommendations

Coral growth parameters are known to be sensitive to climate variability. Across recent decades, when satellite-based observation systems provide comprehensive datasets of climate
variability, reconstructing growth in skeletal cores can improve understanding of coral sensitivity
to climate change (e.g., Cantin et al., 2010; Cooper et al., 2012; Tanzil et al., 2013; Yan et al.,
2019). Furthermore, when skeletal cores extend prior to the instrumental record, they can provide
reconstructions of climate that advances knowledge of multidecadal to centennial variability in the
ocean (e.g., Saenger et al., 2009). These approaches are analogous to dendrochronology, the study
of variations in annual ring widths of trees. However, variations in coral growth are typically
smaller than those of trees, which often have interannual ring-width variability of several-fold
magnitude (Speer, 2010). The maximum variations from the mean extension and density in the
Dongsha Atoll core presented here were approximately 40% and 10%, respectively, and these
values are characteristic of variations from other studies (e.g., Cantin et al., 2010; Cooper et al.,
Tanzil et al., 2013; Yan et al., 2019). This implies that coral cores need to be analyzed with
more precision than tree rings; a 10% random measurement error may have relatively little effect
on a tree-ring reconstruction but would blur patterns in coral extension rate and potentially override
any meaningful signal in a coral density reconstruction.

CoralCT offers a framework to accurately measure coral growth, and to do so in a
standardized and reproducible manner across multiple observers. Since CoralCT can only be used
on skeletal cores added to its companion server, anyone with a computer and internet connection
can install the CoralCT app and contribute to coral core analysis. This enables multiple laboratories
to conduct measurements in standardized ways, or even for the public to contribute (see Section
2.2.6 regarding observer tracking). Given the requirement for precise measurements of growth
parameters, and the potential for substantial differences in data generated by multiple observers,
there is a need in the field for skeletal cores to be analyzed by multiple observers with consistent
methods. With data from multiple observers for each core, cross-dating statistics (Speer, 2010) can
be applied to identify outlier observers and to create “master chronology” compilations of growth parameters from independent observers. Doing so should, in theory, reduce noise in the generated time series and improve confidence where there is good agreement between observers. While CoralCT offers advances in terms of simulated corallites (Section 2.2.4) and automated band detection (Section 2.2.3), the aspect of CoralCT most likely to move the needle on reproducibility in the field is the platform for data sharing and collaboration.

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References


