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#### **Technical Note**

# A simple program to measure and analyse tree rings using Excel, R and SigmaScan

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#### ABSTRACT

I present a new software that links a program for image analysis (SigmaScan), one for spreadsheets (Excel) and one for statistical analysis (R) for applications of tree-ring analysis. The first macro measures ring width marked by the user on scanned images, stores raw and detrended data in Excel and calculates the distance to the pith and inter-series correlations. A second macro measures darkness along a defined path to identify latewood–earlywood transition in conifers, and a third shows the potential for automatic detection of boundaries. Written in Visual Basic for Applications, the code makes use of the advantages of existing programs and is consequently very economic and relatively simple to adjust to the requirements of specific projects or to expand making use of already available code.

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#### Introduction

The analysis of tree rings typically involves measuring the distance between ring boundaries, comparing measurements between cores and checking for errors. While the fastest way to measure tree rings is to measure prepared cores or stem discs directly (e.g., with the LinTab<sup>TM</sup> System [Frank Rinn, Heidelberg, Germany]), this method is very time-consuming if rings are sometimes indistinct, in which case it becomes necessary to compare and re-measure the original cores (Levanič, 2007). Here, measuring digitized images that can be re-analysed easily is an advantage.

Several programs are available to measure tree rings on digital images, differing in cost and capacity. A good overview of tree ring software can be found at http://web.utk.edu/~grissino/software.htm. While the more upmarket programs provide many extras, they may not be optimal for all purposes. For instance, in tropical tree rings, which are difficult to identify and cross-date, it is useful to compare cores on several images that can be scrolled up and down individually to search for matching features, but WinDENDRO<sup>TM</sup> and Lignovision<sup>TM</sup> can open only one image at the time (Table 1).

Writing completely new software for the analysis of tree-ring images for some specific needs tends to be too complex or time-consuming for most researchers. However, different programs are available that are able to perform part of the task necessary, such as image analysis, detrending or graphical presentations. If these

I here present simple procedures to measure and evaluate digitized cores, combining existing programs via VBA macros. While the software was designed to help with the analysis of tropical tree rings that are difficult to date, the first macro, with only about 160 lines of effective code, is simple to change and features such as graphic presentation and simple statistical evaluation can be modified in Excel without any code-writing. The macros combine SigmaScan Pro, a program for image analysis, Excel, a spreadsheet program with simple graphic and statistic features, and R, a powerful open-source statistical package. It was developed and tested with SigmaScan Pro 5.0 (Systat Software Inc., Chicago, USA), Excel 2003 (Microsoft Inc., Redmont, USA) and R 2.9.0 (R Development Core Team, 2009, http://www.R-project.org) on a PC with Windows XP as well as on a Mac with Windows XP.

#### Handling

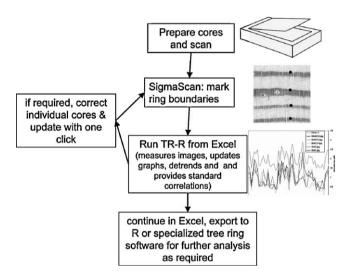
The following is a typical workflow (see Fig. 1) for tree-ring analysis using the described program. More details for the practical use are provided in the accompanying manual. Some technical details are given in the following chapters, the full source code with comments can be seen by opening the macro in Excel. Prepare samples (cores or discs) and obtain a high-resolution image. Many researchers produce images from sanded increment cores with a

programs can run macros in a common environment such as Visual Basic for Applications (VBA), they can be linked to exchange data or commands even if their programming languages differ. Compared to writing new software it then becomes relatively easy to produce versatile and flexible software for specific applications such as treering analysis.

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Table 1
Comparison of TR R with commercial software for image analysis of tree rings. Costs are approximate and for single-user licences, except Lignovision, which is for up to five

	Cost	Parallel images open	Skip gaps	Early/latewood	Follow-on analysis	Data formats
TR R	Free (700 € SigmaScan; R, RExcel free; Excel generally available)	Limited by computer memory	Y	Not implemented, but rel. easy if EW/LW identifiable by darkness	Detrending, cross-correlation, plotting of multiple series	Currently only xls and as exported from Excel
Windendro	900 €Mini 2700 € Basic 5300 € Regular	1 (but multiple paths per image: 4 in basic, 128 in regular)	Y	N (basic) Y (regular)	Detrending, visual and numeric crossdating, master series	Proprietory, Excel, Tuscon
Lignovision	1000 €	1	N	Y	Plots single series	Heidelberg, Excel



**Fig. 1.** Typical workflow for tree ring measurement including the described macro. Ring boundaries are marked in SigmaScan only. The macros for measurement and synchronisation are run from within Excel, SigmaScan needs to be running but at this stage the user operates only within Excel. If individual samples are later corrected, the user switches to SigmaScan, running the macro again in Excel will update all measurements and statistics

high-end desktop scanner, but obtaining images with a microscope and stitching adjacent parts of a core together is also an option (Levanič, 2007). The image resolution needs to balance the amount of detail required and the size of the image files, a resolution of 1200–2400 dpi is almost always sufficient and substantially less (300–600 dpi) is sufficient for samples with clear ring boundaries. SigmaScan supports a large range of image formats, including BMP, JPEG, PCX, TGA, TIF and PSD, b/w as well as colour images. The size of the image as well as the number of images open at the same time are basically limited by computer memory, but should not be unnecessarily large to avoid slow performance.

Open the image with SigmaScan and use the overlay draw mode of SigmaScan (Mode > Overlay draw mode) to mark ring boundaries with a dot. An overlay in SigmaScan is a layer placed onto the original image but stored in a separate file, so that the original image is not modified. Since only one overlay (the standard is green) is needed to mark ring, four other colours can be used to mark additional features or write comments to the image. The size of the dot can be set in Tools > Cursor/Pen size, 5–9 pixel is mostly useful, depending on the image size and how easily the marks should be visible. Dots on successive ring boundaries should be approximately along a straight line, which is best achieved by following the rays from bark to pith. If this is not possible because the core does not exactly follow the rays from bark to pith, start a new line along a different ray by placing a second dot on the same ring boundary

(see arrows in Fig. 2 and details in the next chapter). When all ring boundaries have been marked, save and close the file, the overlay with the ring boundaries will be saved in a separate file with the extension "ov2".

Open TR R in Excel (click "Activate macros" when asked) and check that in the sheet "setup" the directory of the image files, the outer year and the calibration (in pixels per mm, e.g., 94.5 for a resolution of 2400 dpi) are correct. Make sure SigmaScan and RExcel are open. Put the name of the image files (including the extension but excluding the directory) in the third row of the sheet "Detr" and start the macro "MeasureRing" (via Extra > Macro > Macros or Alt-F8). The detrended series are written below the cell with the filename, the raw data to the sheet "Raw". Pearson correlation coefficients and Gleichläufigkeit to the sheet "Cor" and some extra information to "Eval". Names and general layout of Excel sheets should not be changed unless users also modify the code in the macro that addresses these sheets. The name of the Excel file (TR R) can be changed as required. The raw-data measurement does not require R, so the program will run with Excel and SigmaScan alone. However, for detrending and the calculation of the correlation coefficient and Gleichläufigkeit (Eckstein and Bauch, 1969) R routines are called, and it is recommended to install R (http://www.R-project.org) and RExcel (http://rcom.univie.ac.at/) as well. For users familiar with R, this provides a seamless interface to a powerful and increasingly popular statistics and graphics software. Alternatively, Excel can be used for graphic display and simple statistics, or data can be exported and used in other tree ring software. Special export formats such as Heidelberg or Tuscon that some tree ring programs require are currently not provided. After running the macro, ring boundaries are tagged with the year (see Fig. 1 in the manual). This annotation is saved in a separate file and will be shown when the image file is opened in SigmaScan the next

Depending on the project and how complex cross-correlation turns out to be, users will validate the first dating by comparing the graphs of detrended series or by using correlation coefficients and Gleichläufigkeit. The sheet "Detr" shows the Pearson correlation and Gleichläufigkeit with the mean series, the sheet "Cor" has cross-correlations between each two series. Of course data can also be validated using other software such as COFECHA. When crosscorrelation indicates error in dating or problematic rings, images are compared in SigmaScan. A major advantage compared to other available software is that various images of different cores can be opened at the same time and compared by scrolling individual images up and down so that matching tree rings can be aligned. This is a substantial advantage when identifying ring boundaries is difficult and detailed anatomical comparisons between cores are necessary. When dots marking ring-boundaries are changed and the Excel macro is run, graphs and correlations in Excel are updated automatically. The number of files open at the same time is limited

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