# VISIOPHARM®

AUGMENTED PATHOLOGY

Precision Pathology

High-throughput pathology

# Stereotopix™ Research

**– powered by newCAST™**

RUO

Researchers use quantitative microscopy in many ways with the goal of producing high-quality, quantitative results to support the development, submission, and award of grants and publications. Principal Investigators often need to obtain unbiased structural information (volume, area, length and numbers) from many types of tissue and from images created on a variety of different modalities.

Stereotopix™ will provide laboratories with an ultra-flexible stereology software solution to support the requirements of research.

The Visiopharm stereology software STEREOTOPIX™ is driven by newCAST™, which was developed in close collaboration with world-leaders in the field and is the most comprehensive, robust, user friendly and versatile stereological system available today.

## Advantages of Stereotopix™:

- Unbiased quantification of tissue changes
- Highly configurable stereology toolbox
- Automated physical fractionator with image analysis driven sampling
- Multiple image modalities: whole slide, brightfield, fluorescence and more
- Developed and tested by leading experts

## A design based stereology solution

Design based stereology software, ensures unbiased estimates of structural information (volume, area, length, and number) from 2-D tissue sections. Stereology (newCAST) from Visiopharm is a complete, well documented solution that has been cited in over 800 publications just since 2010.

## Combine probes for more quantitative power

The Stereology module has a comprehensive selection of stereological probes that can be combined to allow for simultaneous analysis of several end-points. Modern stereological methods introduce a need for efficient handling of local thickness measurement and guard zones at artificial edges for calculation of exact sampling probabilities. Local thickness measurement and guard zone handling is seamlessly integrated into Stereology.

## An organized, intuitive workflow

Sample, count, document, calculate, publish – it's that simple. Stereology will support users through each step of the process yielding powerful results of unbiased estimates that will meet or exceed the high standards of leading scientific journals for publication.

## Increase sampling efficiency

The Stereology module can be expanded with The Proportionator™ which greatly increases sampling efficiency since practically all fields of view will contain the feature of interest.

## Automated physical fractionator

Visiopharm's patented Autodisector™ makes it possible to work fast and efficiently with the Physical Disector principle, and thereby reduce or eliminate frequently occurring practical problems due to section shrinkage, staining penetration, opaque tissues, and large objects.



**Figure 1**
Start with the basic software for Stereology (dark green). For enhanced capabilities, add the relevant modules from the outer ring (light green).

## Developed and tested by leading experts

Visiopharm's newCAST™ is the backbone of Stereotopix™ and originates from the stereology epicenter of Aarhus University, Denmark, developed and tested with Profs. H.J. Gundersen and J.R. Nyengaard. New methods and tools for Stereotopix™ are routinely developed, implemented and tested in close collaboration with the Stereology Research Laboratory.
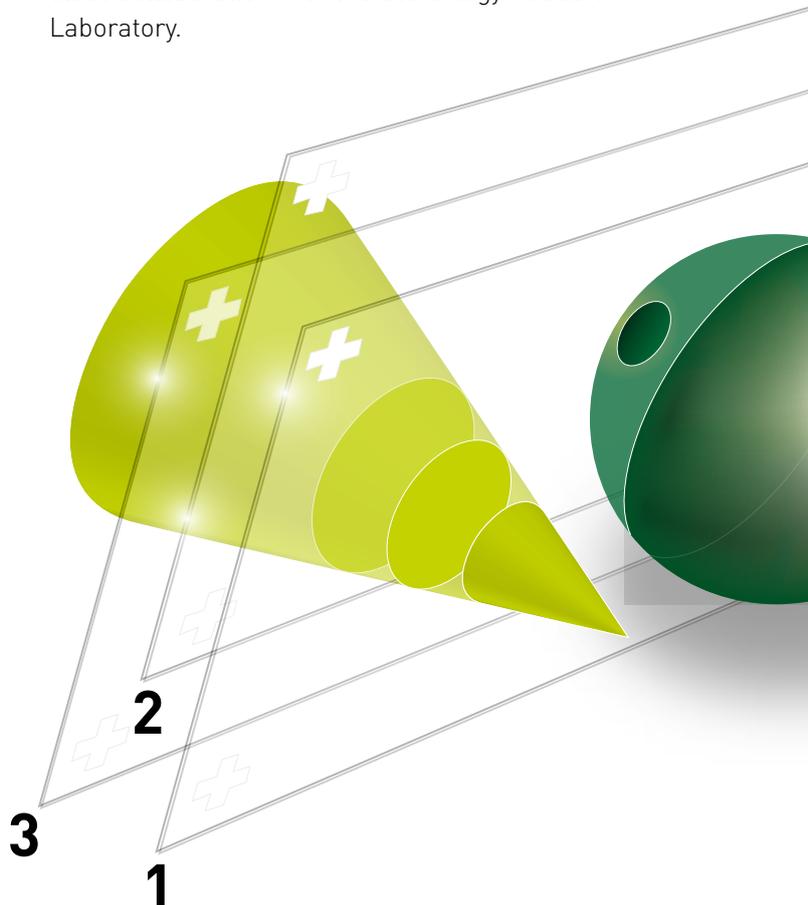
# Virtual™

## Whole Slide Stereology

Visiopharm's Virtual™ makes it possible to work directly on scanned virtual (whole slide) images from all major slide scanner manufacturers.

### Hardware independent stereology

Upgrading your current Stereology system to use whole slide imaging with Visiopharm's Virtual™ will allow you to instantly benefit from the technical advances in virtual microscopy. Visiopharm software will read the whole slide and you perform the stereological analysis completely separate from hardware. You no longer have to perform analysis for hours in a small dark room with no windows!

### Much faster

The analysis is up to 10 times more efficient using whole slides for your stereological analysis due to several time-saving steps:

- The overview image is instantly created during sampling, the field of view is shown promptly with no need to focus or move the stage.

- It is easy to analyze the next slide as all images are immediately pre-imported into the database. No need to mount a new slide on the microscope – they are already in the database.

Efficiency is further enhanced using the automated add-on modules from Visiopharm: Autodisector™ and Proportionator™.

### Time to change?

Is your microscope hardware getting old and giving you frequent issues? Is your computer slow, needs replaced but you are uncertain if your old hardware is still compatible with a new computer? Maybe it is time to start using whole slide technology?
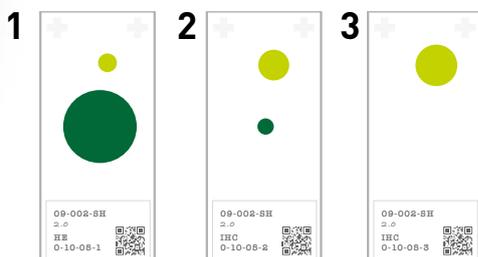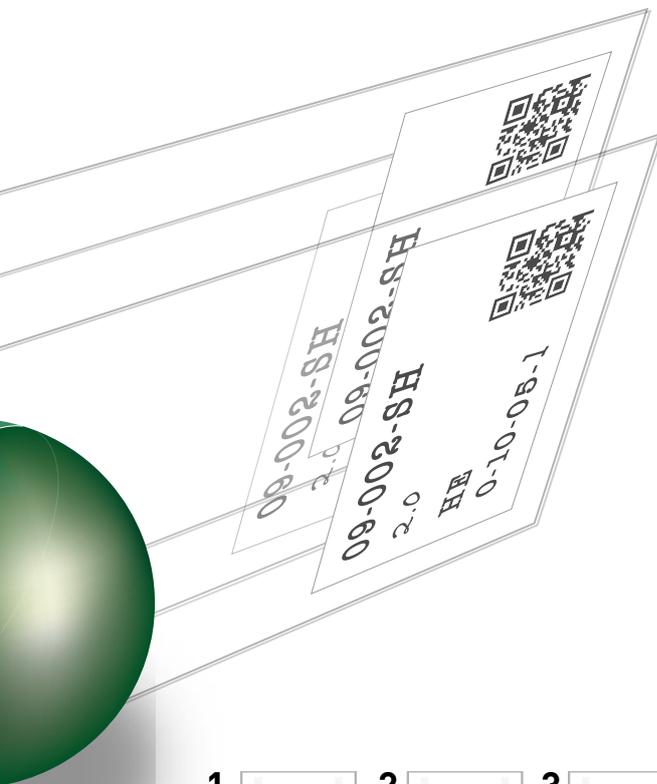
### Advantages of Virtual™:

- Analyze anywhere due to separated image acquisition and analysis.

- Possibility for image/slide sharing with other labs

- Navigate the virtual slide and change magnifications as if on a live microscope

- Save your counts directly on the image. The counts can be reviewed later for documentation.

- No need to calibrate or solve hardware issues on the microscope!

# Autodisector™

## The Automated Physical Fractionator

Visiopharm's patented Autodisector™ makes it possible to work fast and efficiently with the stereology physical fractionator principle by automating multiple steps leading to perfectly aligned adjacent sections. The Autodisector™ technology is tissue independent and provides perfectly matched disector pairs on any kind of tissue at any magnification.

## An automated physical fractionator

Visiopharm's patented Autodisector™ makes it possible to reduce or eliminate frequently occurring practical problems in thick sections such as section shrinkage, staining penetration, opaque tissues, and large objects.

## No more problems with stain penetration

With the Autodisector™ there is no need for thick sections or the struggle to determine if you have good staining penetration. Thin sections can now be used ensuring good staining penetration of antibodies-including standard kits, and auto-staining methods.

## Not sensitive to section shrinkage

Often thick sections collapse especially during dehydration, or in other steps of the manual staining process. With thin sections, this no longer becomes a concern.

## No limitation on the size of the objects counted

There are no limitations regarding the size of objects that may be counted, for example counting glomeruli in the kidney cortex is not a problem. Also, the time spent counting with the automated physical disector is comparable to the time spend on the optical disector, plus you avoid all changes associated with thick sections.

## Guided sampling

Guided sampling can be done with the Proportionator™ sampling principle- a new general sampling principle in stereology that uses image analysis to perform a non-uniform sampling based on the extent of the feature of interest. This is in principle a biased method – but the sampling probability of each field is known, allowing for calculation of unbiased estimates. The Proportionator™ sampling design is based on the sampling with Probability Proportional to Size (PPS).

## Increased efficiency

When available, whole slide images will help the sampling and counting go even faster with the Autodisector™. The Autodisector™ provides a lot of automation in the sampling process including automatic tissue detection, tissue alignment of sections, automated sampling and storage of paired fields of view as TIFFs. When it's time to count, and analyze, field of view images are ready and can be loaded as an offline sampling also from remote workstations. Because the sampling images are stored (including counts and measurements) the entire process is documented and provides a method to analyze the same data by different researchers to check for inter observer differences.

## Advantages of Autodisector™:

- An organized intuitive workflow

- Automated tissue detection saves time delineating tissue.

- Excellent automated section alignment using both boundary and tissue features

- Automated field of view sampling for offline analysis

- Works on wholes slides as well as glass slides on microscopes

# Proportionator™

## Image analysis driven sampling

The Proportionator™ is a very efficient sampling principle in stereology that uses image analysis to perform a non-uniform sampling based on the extent of the feature of interest in each field of view. With the image analysis driven Proportionator™ sampling, the feature of interest will be present in almost all sampling positions even if the feature is inhomogeneously distributed.

### Powered by image analysis

The Proportionator™ provides the tools necessary to design an image analysis protocol to guide the sampling process. The image analysis protocol will detect the relevant feature (e.g. area of stained cells) and calculate the weight of the feature in all the fields of view. This gives a field with a high weight value a higher probability of being sampled than a field of view with a low weight. For each sampled field the weight value and the sampling probability are logged.

### Greatly increase sampling efficiency

The Proportionator™ greatly increases sampling efficiency since practically all fields of view will contain the feature of interest. Recently a comparative study* showed that the Proportionator™ was twice as fast as normal meander sampling and in combination with Visiopharm's Autodisector™ it became three times faster (Keller et al., 2013, Improving efficiency in stereology: a study applying the proportionator and the autodisector on virtual slides. Journal of Microscopy, 251: 68–76).

### Save time, reduce expenses

Large scale stereology research projects now become possible with the Proportionator™. The largest increase in efficiency is on inhomogeneous tissue, like ß-islets in the pancreas or purkinje cells in the cerebellum. However, there is always a net gain in efficiency when using Proportionator sampling.
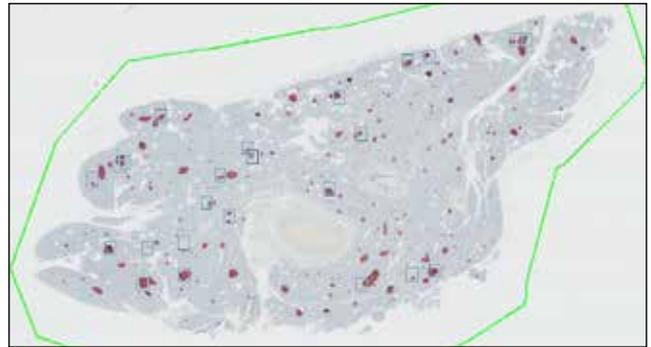


Proportionator sampling in a monkey pancreas. The positions are sampled using an image analysis protocol tuned to find the beta islets (red). Note that almost all sampling positions overlay beta islets.

### A complete workflow for stereology

Visiopharm's powerful Stereotopix™ solution, with the help of the Proportionator™, provides a complete workflow for high volume, efficient use of stereology. First an image analysis protocol is created, guided sampling is performed, counting is done with one of the several geometric probes offered, and the calculation of the weight-corrected counting results are done automatically and presented in an excel data sheet. The unbiased estimate can then be calculated using the weight-corrected results, and the CE of the estimate can be directly calculated from the sampling data in the Calculator.

### Advantages of Proportionator™:

- Increase speed with guided sampling powered by image analysis

- Greatly increase sampling efficiency especially in combination with Autodisector™ and Virtual™

- Save time, reduce expenses

- Detect small objects effectively

- A complete workflow for stereology

## Training and support

Visiopharm offers on-site, as well as on-line, software training conducted by highly skilled Visiopharm employees. The training can lead to new Standard Operating Procedures and experimental protocols. We also offer the Visiopharm Academy in which small groups of users join for 2-3 days of theory and practical exercises - guiding users every step of the way.

Visiopharm also offers on-line installation and support, allowing our technical experts to quickly assist in real time solving issues with software, servers and hardware among other things - supporting users every step of the way.
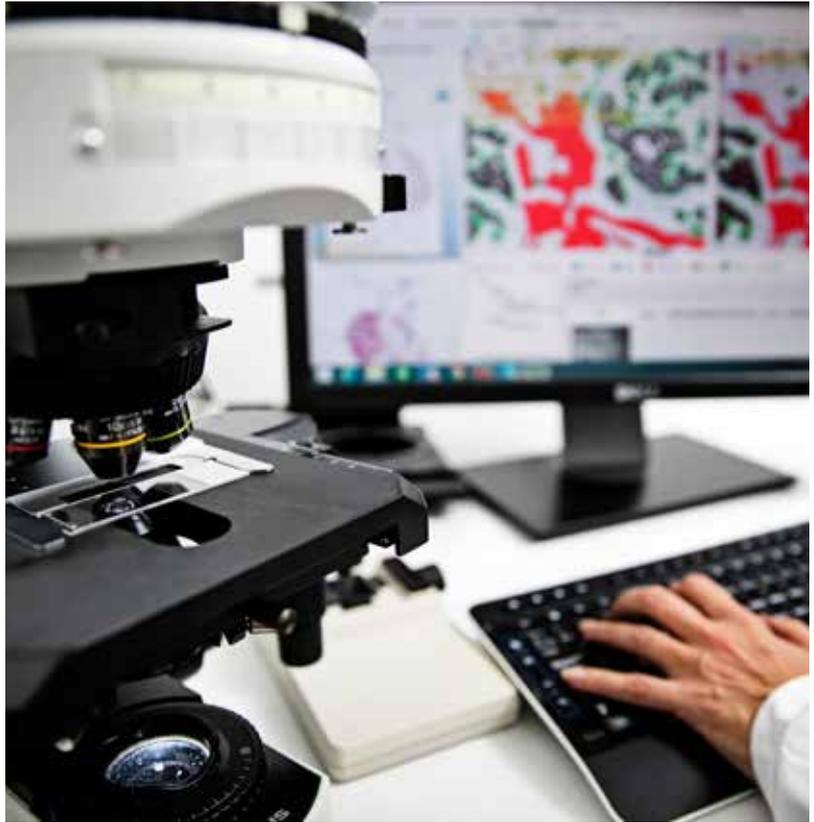
## About Visiopharm

Visiopharm is a vendor-neutral provider of Augmented Pathology comprising high-throughput and precision pathology for digital and automated test applications. Through its many partnerships and expertise in intelligent image analysis Visiopharm solutions span across the entire digital pathology workflow modernising and improving anatomic pathology laboratory routines worldwide. In EU Visiopharm offers several IVD algorithms as an aid to the pathologist doing breast cancer diagnostics. Recently Visiopharm launched a new Viewer completely innovating smooth and seamless navigation for routine analysis.

Visiopharm's software is featured in over 1200 scientific publications, and has over 800 licenses placed, with countless users, in more than 30 countries. Headquartered in the Medicon Valley of Denmark, with a branch office in Sweden, the Netherlands, and North America in Broomfield, Colorado.

# VISIOPHARM®

AUGMENTED PATHOLOGY