# Conditional Imaging with NIS-Elements

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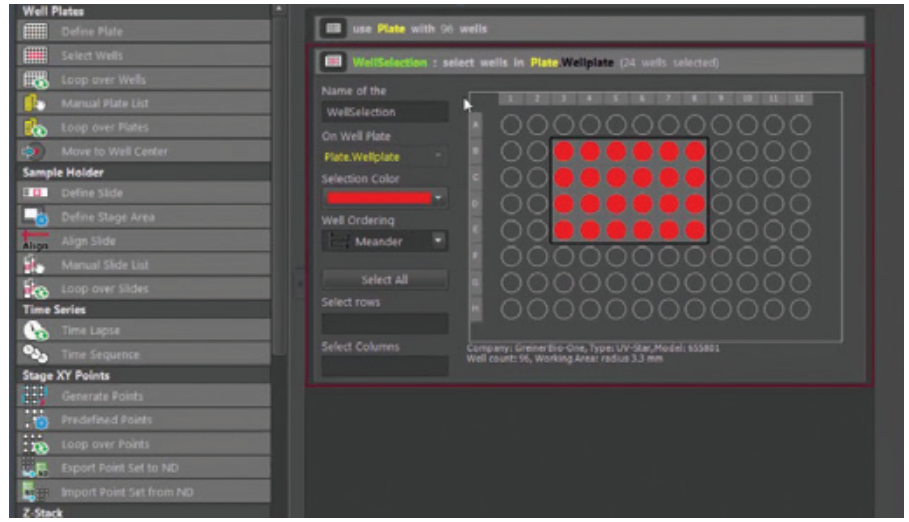
As technologies in light microscopy advance, a common bottleneck in the acquisition of experimental data lies with the researcher themselves. The requirement to be present alongside a system, repeatedly capturing images and making decisions based on collected recordings takes time and costs money in research projects. It can also lead to conscious or unconscious bias in how fields of view are selected.

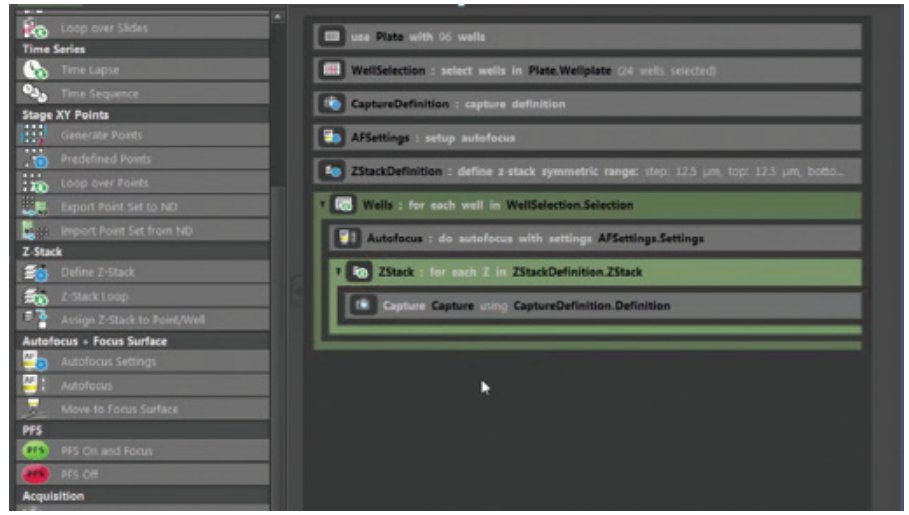
For those capable of building and running a customised system with their own software controlling acquisition, writing comprehensive scripts and macros has always been an effective means of reducing repetitive actions on a microscope. However, this has historically required a high degree of expertise and it can be time-consuming to adapt to new assays or samples. Some degree of automation has been integrated into manufacturers' software; however, this tends to be either relatively basic in application or somewhat cumbersome, which limits the flexibility and capability of the system. Custom scripts have been possible with the support of manufacturer software development teams but this can be slow and costly for the researcher to implement.

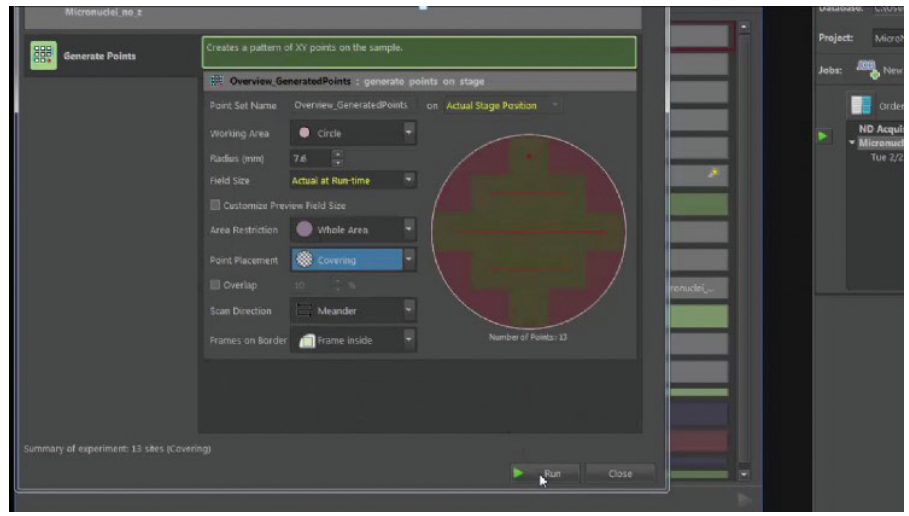
The JOBS module for Nikon's NIS Elements software changes this by offering researchers without programming expertise the possibility of unlocking the full potential of their system in terms of both flexibility and massively increasing their productivity by having the system run in an unattended and unbiased, reproducible way. By fully integrating with microscope configuration, acquisition and analysis, its applications are practically unlimited. With a simple or advanced interface, even the most inexperienced user can create entire workflows to automate experiments efficiently.

Essentially, JOBS is a user-friendly interface that allows microscopists to easily create workflows capable of acquiring or analysing images based on variable conditions. Requiring no prior programming experience, a 'job' can be created within minutes, capable of carrying out an experiment using all manner of microscopy equipment. For a simple example, a researcher can instruct JOBS to count how many detected cells the system has acquired and then decide whether to image more to complete the dataset, or end the experiment. For a more advanced development, the job could measure the intensities of each object of interest to determine the levels of fluorescence, discard objects that are too dim to analyse, seek and find new ones and then, once the desired dataset is complete, email the user an initial analysis of the data and shutdown the system.

For those with experience of programming languages, recognisable 'if' and 'while' functions make up the backbone of such conditional imaging decisions. A large and ever-growing function library allows for maximum control with minimal user input to create powerful jobs. For the more advanced user, not only can entire imaging experiments be created and managed remotely over long time periods, but simplified workflows can be developed for other users. One function of JOBS allows for a custom message to appear, capable of prompting the user to make a decision. Uses could include providing updates on an experiment, providing troubleshooting tips if any problems are detected (for example, a low intensity image may prompt the user to increase the gain of the detector) or




*Figures 1 to 3. Highlighting the use of the NIS-Elements JOBS wizard to set automated experiments.*

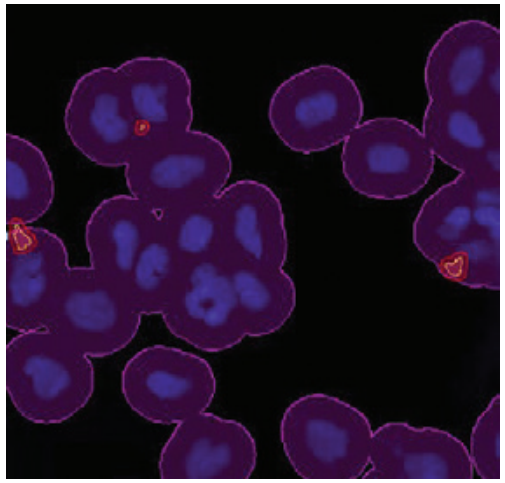


*Figure 4. Segmenting DAPI micronuclei from nuclei in NIS-Elements JOBS software.*

developing a friendly wizard for the user to run an entire experiment based solely on answering questions about what they wish to achieve.

Another strong potential for the JOBS software is the removal of user-derived bias in scientific methodology. Choosing regions of interest to image, selecting the brightest objects and acquiring the healthiest cells can all be automated and determined by the system based on thresholds or pre-determined configurations, allowing for a truly randomised series of image locations, or an accurate sample-set representative of a population.

Combined with a high throughput system, such as those with plate loaders and microfluidics, JOBS has the potential to drive experiments with vast numbers of samples, boosting *n* values of experimental data, being left unattended from hours to days.

Dr Michael Delves of Imperial College London has been using the JOBS package to screen drugs for malaria prevention. Using 384-well plates containing 'miniature cultures' of *Plasmodium falciparum* gametocytes (the sex cells of the malaria parasite), a series of candidate drugs can be administered for transmission-blocking effects. Due to the need to adjust temperatures during the experiment to simulate parasites being taken up into the mosquito, maintaining focus during time-lapse imaging becomes an obstacle due to the expansion and contraction of the plate.

To allow for reliable and unsupervised experiments to take place, Dr Delves's group have created a JOB that first focusses on the well of interest, and then images every well in a loop at 10 frames per second

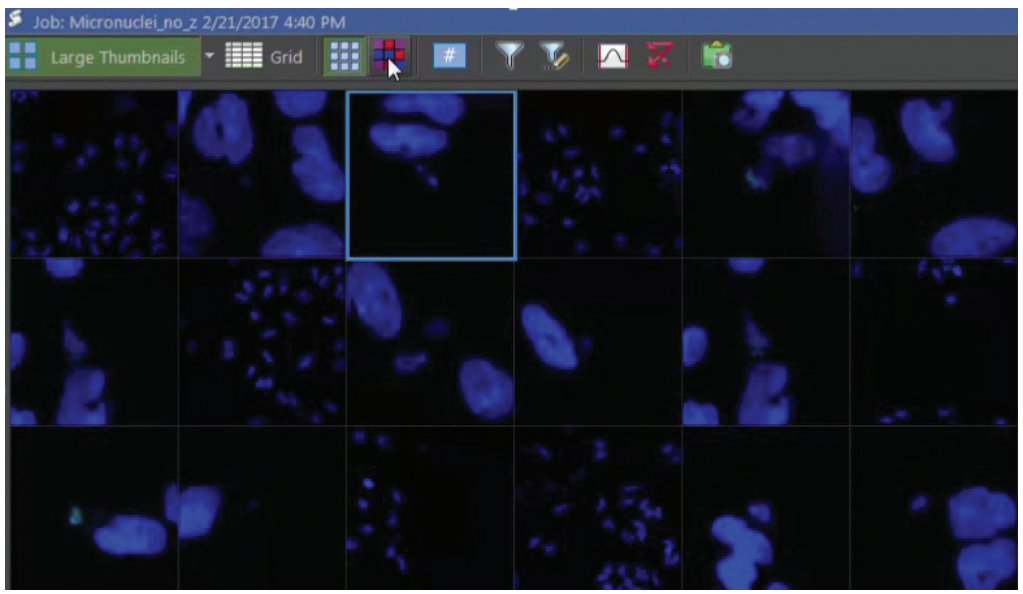



*Figure 5. Thumbnails view helps the user to easily identify micronuclei.*

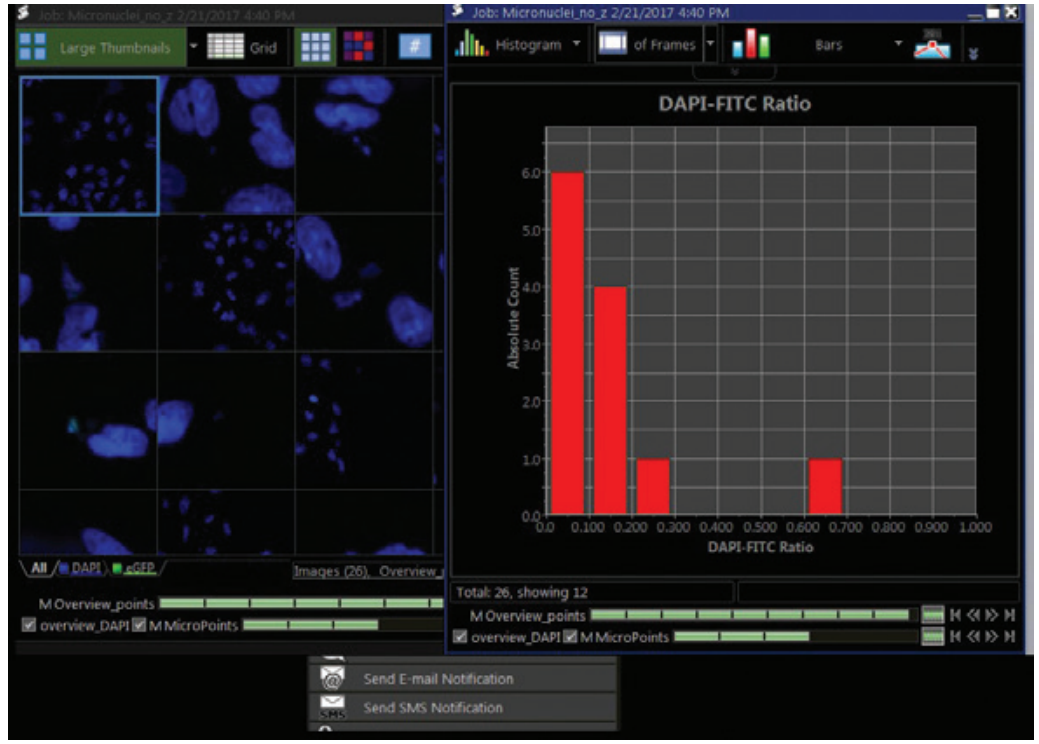



*Figure 6. Graphs showing the count of micronuclei in each image are created effortlessly.*

every 1-2 seconds. This can then be fed into on-the-go image analysis all within the JOBS module.

After having incorporated JOBS functionality into imaging experiments, Dr Delves achieves a screening rate of 12,000 drugs per week; a vast improvement over past manual imaging of just eight drugs per day. The JOB is now shared and updated with collaborators and pharmaceutical companies across the globe, and is easily emailed to any group with a Nikon system to conduct identical experimental protocols.

With almost endless possibility and flexibility, the Nikon JOBS module has the potential to change how experiments are planned from the outset, providing a logical and adept assistant and imaging controller that can make the necessary pre-programmed decisions to run lengthy time courses over days and nights with just an email sent to the user once the analysis is complete.

As well as offering endless flexibility, JOBS also has the option of running wizards to easily guide users through launching more basic experiments. The software comes with a library of wizards for the most common experiments, making the system simple to use for less experienced users.

As computing technologies continue to advance, with 'big data' algorithms already providing inhuman levels of knowledge and prediction, and artificial intelligence on the horizon, the quintessential imaging routine of sitting by a microscope, lit by a monitor in the dark, is likely to become a distant bottleneck in the current research environment.

The Nikon JOBS module is compatible with any modern Nikon system, including widefield, confocal and super-resolution systems. It is also compatible with many 3rd party devices that are able to communicate with NIS Elements including LIPSI, the latest high content package. It is particularly complementary to Nikon's latest developments

and systems that offer large fields of view including the Ti2 inverted research microscope, Yokogawa CSU-W1 spinning disk and the latest in point scanning confocal technology, the all new A1R HD25 offering a 25 mm field of view, approximately double the area of conventional point-scanning confocals.

**Edited by Pierre-Emmanuel Monet, European Product Manager**

Pierre-Emmanuel has worked for Nikon Instruments for 13 years, and is currently European Product Manager. This involves product strategy, product launches, support and training for live cell microscopes, cameras & software. Pierre-Emmanuel specializes in NIS-Elements software and also works with the planning and development team in Japan to assist with developing some of the next generation of Nikon products.

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