TECHNICAL WHITE PAPER

3D Electron Diffraction and 4D-STEM on Your SEM

How hybrid pixel detector technology opens up advanced transmission electron methods for scanning electron microscope users

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ABSTRACT

The scanning electron microscope (SEM) is one of the most widely used analytical instruments in science and industry. Yet most SEM users tap only a fraction of its potential: surface imaging with secondary and backscattered electrons, possibly combined with energy-dispersive X-ray spectroscopy (EDS). Advanced transmission methods — three-dimensional electron diffraction (3D-ED) and four-dimensional scanning transmission electron microscopy (4D-STEM) — have transformed structural analysis in transmission electron microscopy (TEM), enabling atomic-resolution structure determination of crystals too small or beam-sensitive for X-ray diffraction, and mapping nanoscale strain, orientation, and electric fields across materials. Until recently, these methods were inaccessible on a standard SEM.

This white paper explains what 3D-ED and 4D-STEM are, why they matter, and how the ASI FeliS hybrid pixel detector brings these capabilities to a standard SEM platform. It is written for SEM users with a working knowledge of electron microscopy but no prior background in electron diffraction.

KEYWORDS: scanning electron microscopy, 3D electron diffraction, 4D-STEM, hybrid pixel detector, Timepix3, Medipix3, ptychography, crystal structure determination, orientation mapping, FeliS

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1. Introduction: What Can Your SEM Really Do?

If you use a scanning electron microscope, chances are you spend most of your time acquiring secondary-electron or backscattered-electron images, perhaps combined with energy-dispersive X-ray (EDS) analysis. These are powerful techniques — but they address only two questions: what does the surface look like, and what elements are present?

There is a third question — what is the atomic structure of the material? That is equally important for understanding material properties, reactivity, and failure mechanisms. Answering has traditionally required dedicated instruments: an X-ray diffractometer for bulk measurements or a transmission electron microscope (TEM) for nanoscale work. Both represent major investments in equipment, sample preparation, and operator expertise.

Over the past decade, electron diffraction methods developed for TEM have matured enormously. Three-dimensional electron diffraction (3D-ED) can now determine the crystal structure of particles just a few hundred nanometers across — smaller than can be handled by X-ray methods. Four-dimensional scanning transmission electron microscopy (4D-STEM) maps strain, orientation, and local electric fields with nanometer resolution across extended areas. These methods have transformed structural biology and materials science on TEM platforms.

The same methods are now becoming accessible on SEM, at lower cost, on instruments that are already installed in thousands of laboratories worldwide. The key enabling technology is the hybrid pixel detector: a new class of area detector that combines single-electron sensitivity, zero noise, and fast frame rates in a format that fits directly under the pole piece of a standard SEM.

This document explains what 3D-ED and 4D-STEM are, why they are scientifically valuable, how they work technically, and what the ASI FeliS detector does to make them practical on a SEM. No prior knowledge of electron diffraction is assumed.

2. SEM: A Powerful Platform with Untapped Potential

2.1 A Brief History

The scanning electron microscope was commercialized in the 1960s and has since become the workhorse imaging instrument of materials science, geology, biology, and semiconductor

manufacturing. Unlike the TEM, which passes an electron beam through a thin sample and forms an image from transmitted electrons, the SEM scans a focused beam across the surface and collects signals generated by the interaction of electrons with the near-surface region.

The three most common SEM signals are secondary electrons (SE, emitted from the top few nanometers of the surface and primarily carrying topographic information), backscattered electrons (BSE, reflecting compositional contrast via average atomic number), and characteristic X-rays (used in EDS elemental analysis). Together these provide rich information about morphology and composition — but nothing directly about crystal structure.

2.2 EBSD: The First Step Toward Structure

Electron backscatter diffraction (EBSD) was the first structural technique to be widely adopted on SEM. In EBSD, the sample is tilted to approximately 70° and backscattered electrons that exit the surface at shallow angles form a diffraction pattern (a Kikuchi pattern) on a phosphor screen. These patterns are indexed computationally to yield local crystal orientation, phase, and grain boundary information across polycrystalline materials.

EBSD is now a standard analytical module sold by all major SEM manufacturers and is routinely used in metallurgy, geology, and semiconductor failure analysis. However, EBSD has fundamental limitations. It is a surface technique, probing only the top few tens of nanometers. It requires a reflective geometry that is incompatible with standard imaging, and it struggles with nanocrystalline, amorphous, or beam-sensitive materials.

The next step — transmission geometry — removes many of these constraints and opens the full power of electron diffraction to SEM users.

2.3 Why SEM Rather Than TEM?

SEM offers several practical advantages over TEM for transmission experiments. The sample chamber is larger, accommodating bulkier sample holders, heating stages, cooling stages, and in-situ deformation rigs. The accelerating voltage in a SEM (typically 10–30 kV) is much lower than in TEM (80–300 kV), which means less beam damage to sensitive biological and organic samples. Access to SEM time is generally far easier and less expensive than TEM time. And critically: close to a million SEMs are already installed worldwide. Adding transmission diffraction capability to an existing instrument, rather than purchasing a dedicated TEM, dramatically lowers the barrier to adoption.

TEM-quality diffraction data from your SEM: more samples per day, lower beam damage, no separate booking queue. The trade-off is resolution and scattering strength. For many applications, SEM geometry is entirely adequate.

3. Electron Diffraction in Transmission: The TEM Revolution

3.1 Bragg Diffraction and the Unit Cell

When a beam of electrons (or X-rays) passes through a crystalline material, it is scattered by the periodic arrangement of atoms. Constructive interference occurs at specific angles defined by Bragg's law: $n\lambda = 2d \cdot \sin\theta$, where d is the spacing between crystal planes, λ is the electron wavelength, and θ is the diffraction angle. The resulting diffraction pattern is a fingerprint of the crystal structure: the positions of diffraction spots reveal the unit cell dimensions, and their intensities carry information about the arrangement of atoms within the unit cell.

In a conventional single-crystal X-ray diffraction experiment, a macroscopic crystal (typically 0.1–1 mm) is rotated while diffraction data are collected from many orientations. Computational methods then reconstruct the three-dimensional electron density of the unit cell, and from it the positions of all atoms. This approach is the gold standard of structural characterization and has underpinned the determination of hundreds of thousands of crystal structures deposited in the Cambridge Structural Database and the Protein Data Bank.

3.2 The Nanocrystal Problem

The critical limitation of X-ray diffraction is crystal size. Crystals must be large enough to diffract X-rays measurably, which means at minimum a few micrometers for synchrotron sources and typically 50–100 µm for laboratory diffractometers. Many scientifically and industrially important materials do not form large crystals. Zeolites, metal-organic frameworks, pigments, pharmaceuticals, and minerals often exist only as nanoparticles or microcrystalline powders. Powder X-ray diffraction can identify phases and measure unit cell parameters but generally cannot solve unknown structures without additional prior knowledge.

Electrons interact with matter far more strongly than X-rays. A crystal just 100–500 nm across — one that would produce no measurable X-ray diffraction signal — scatters electrons strongly enough to yield a high-quality single-crystal diffraction pattern. This is the fundamental motivation for electron diffraction: it extends crystallographic structure determination to the nanoscale.

3.3 From Single Patterns to Three-Dimensional Data

A single electron diffraction pattern records only a two-dimensional slice through the three-dimensional reciprocal lattice of the crystal. To reconstruct the full three-dimensional structure, data must be collected from many orientations. In TEM, this is achieved by tilting the sample systematically while recording a diffraction pattern at each tilt angle.

Early electron crystallography collected a limited number of zone-axis patterns and used them in combination to determine structure. The decisive advance came with automated diffraction tomography (ADT) and precession electron diffraction tomography (PEDT), developed in the mid-2000s, which automated the collection of complete tilt series and greatly improved data quality by reducing the contribution of multiple scattering (dynamical diffraction). The method was subsequently renamed three-dimensional electron diffraction (3D-ED) and microcrystal electron diffraction (MicroED). It is now a standard analytical capability at electron microscopy centers worldwide.

4. Three-Dimensional Electron Diffraction (3D-ED)

4.1 How a 3D-ED Experiment Works

In a 3D-ED experiment, the microscope operates in diffraction mode with the beam focused on or near the sample. A tilt series is recorded: the sample is rotated through a range of angles (typically ±60° or more) while diffraction patterns are acquired continuously or at discrete steps. The resulting dataset is a three-dimensional map of diffraction intensities in reciprocal space.

The data are then processed in software to index the diffraction patterns, merge them into a coherent dataset, and extract structure factor amplitudes. Finally, structure solution — determining where the atoms are — is carried out using the same algorithms used in X-ray crystallography: direct methods, charge flipping, or Patterson methods. Refinement against the experimental data yields the final structural model.

The entire pipeline from data collection to refined structure can now be completed in under an hour for straightforward cases, using freely available open-source software such as PETS2, Instamatic, and SHELX, or commercial packages.

4.2 Technical Requirements

A 3D-ED experiment requires three hardware components beyond a standard TEM or SEM:

- A tilt stage capable of high-angle, continuous rotation. Most SEM and TEM stages can be retrofitted or replaced.
- An area detector able to record the full diffraction pattern simultaneously. The detector must be sensitive enough to record weak reflections without saturating on strong ones, and fast enough to acquire patterns at the tilt rate of the stage.
- Software to control the data collection, process the raw images into a reciprocal-space dataset, and solve and refine the structure.

The shift from film and CCD cameras to fast, sensitive direct electron detectors was the key enabling step that made 3D-ED practical. Modern hybrid pixel detectors can record thousands of diffraction patterns per minute, enabling automated data collection on multiple particles in a single session.

4.3 What Can 3D-ED Solve?

3D-ED has been applied successfully to a wide range of material classes where other structural methods struggle or fail:

- Pharmaceutical compounds and drug metabolites that crystallize only as fine powders
- Metal-organic frameworks (MOFs) and porous materials with large, complex unit cells
- Natural minerals and geological samples, including newly discovered phases
- Industrial catalysts and zeolites, where understanding pore geometry and active site geometry is commercially important
- Biological macromolecules such as small membrane proteins and peptides (under the name MicroED)
- Battery electrode materials and other functional materials where phase identification and structure determination inform electrochemical performance

A landmark example is the structure determination of the zeolite catalyst SSZ-52, published in 2015, whose structure had resisted X-ray analysis for years and was solved by 3D-ED in a single session.

The Cambridge Structural Database now contains thousands of structures solved by 3D-ED, and the method is increasingly used as a routine complement to powder X-ray diffraction in pharmaceutical development.

Penghan Lu and colleagues at Forschungszentrum Jülich have demonstrated 3D-ED on a SEM using ASI FeliS, solving crystal structures from nanoparticles that were impossible to characterize by any other method at that laboratory. Early results by Daen Jannis at EMAT (University of Antwerp) similarly show the power of transmission hybrid pixel detectors for advanced electron diffraction experiments [5].

4.4 3D-ED on SEM: Differences and Advantages

Moving 3D-ED from TEM to SEM introduces both advantages and constraints. The lower accelerating voltage (30 kV vs 80–300 kV) means stronger electron-matter interaction: diffraction signals are stronger for a given particle size, and radiation damage to sensitive samples is reduced. The larger sample chamber allows more flexible stage geometries and easier integration of sample heating, cooling, or gas injection. And because SEM instruments are far more widely available than TEMs, SEM-based 3D-ED is accessible to a much larger research community.

The main constraint is the reduced penetrating power of lower-energy electrons: samples must be thinned to below approximately 200–300 nm for good diffraction data quality. Sample preparation methods are discussed in Section 8.

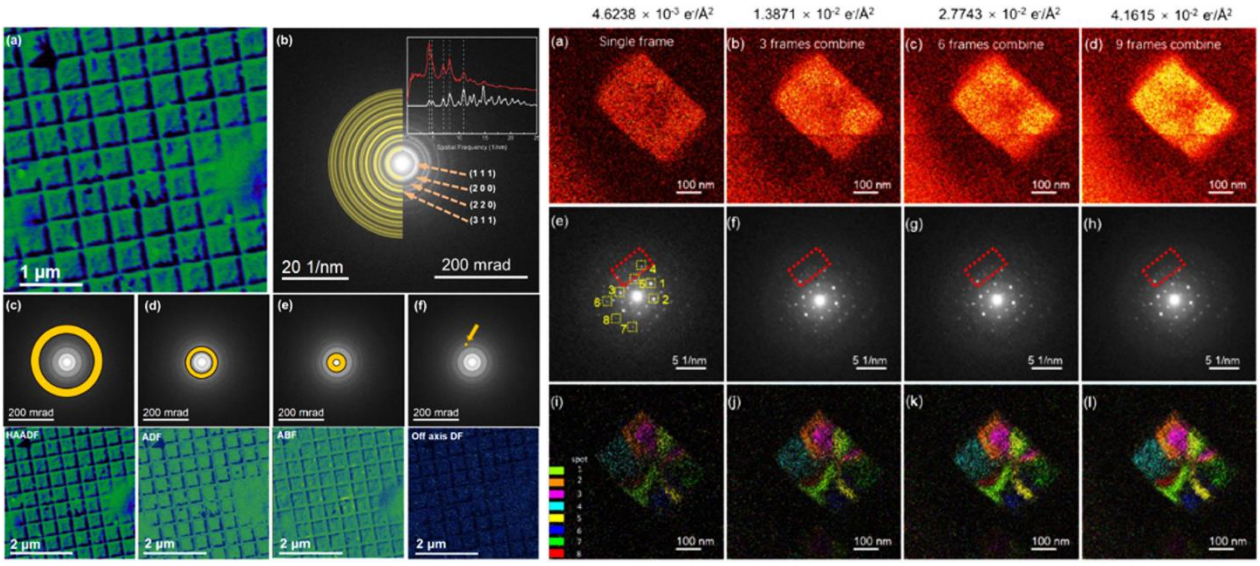

Figure 1. Orientation mapping and diffraction indexing performed with FeliS, highlighting its capability for high-speed, data-rich diffraction analysis. Adapted from [9].

5. 4D-STEM: Scanning at the Nanoscale

5.1 Concept

Four-dimensional scanning transmission electron microscopy (4D-STEM) extends the scanning transmission approach to full diffraction pattern recording. In conventional STEM, a focused electron probe is scanned across the sample and one or more integrated signals (bright-field, annular dark-field) are recorded at each probe position to form an image. In 4D-STEM, instead of integrating the transmitted signal, a full two-dimensional diffraction pattern is recorded at each probe position.

The result is a four-dimensional dataset: two spatial dimensions (the scan position) and two reciprocal-space dimensions (the diffraction pattern). This dataset is extraordinarily rich: it contains, in principle, all of the information that the transmitted electron beam carries about local structure, orientation, strain, electric fields, and composition.

5.2 Orientation and Phase Mapping

One of the most practically useful applications of 4D-STEM is virtual imaging and orientation mapping. By computationally selecting different regions of the diffraction patterns in post-processing, one can reconstruct images with arbitrary contrast: bright-field, dark-field, or annular dark-field, without the need for physical apertures. Orientations of crystalline grains can be mapped by correlating each diffraction pattern against a library of simulated patterns, yielding maps with a spatial resolution limited only by the probe size.

This is in some ways the transmission equivalent of EBSD, but without the severe geometric constraints of the tilted-sample backscatter geometry. 4D-STEM orientation maps can be acquired on flat samples at normal incidence, are sensitive to all crystal orientations, and can detect phase boundaries and grain orientations at nanometer resolution in a single pass.

5.3 Strain Mapping

When a crystal is strained, for example, near a grain boundary, a dislocation, or the interface between two lattice-mismatched layers, the positions of diffraction discs in the convergent-beam pattern shift slightly from their unstrained values. These shifts, measured in post-processing across the 4D dataset, yield quantitative maps of local lattice strain with sub-nanometer spatial resolution and sensitivity to strains of order 0.1%.

Strain mapping has applications in semiconductor device characterization (where built-in strain affects carrier mobility), in the study of fatigue and deformation in structural alloys, and in the characterization of epitaxial thin films and multilayers.

5.4 Ptychography: Breaking the Resolution Limit

Ptychography is a phase-retrieval method that uses the full 4D dataset to reconstruct the complex exit wavefunction of the electron probe after passing through the sample. Because the wavefunction rather than just its intensity is recovered, ptychography provides much higher information content than conventional imaging.

In TEM-based 4D-STEM, ptychographic reconstruction has achieved atomic resolution images from samples that conventional STEM imaging cannot resolve. On a SEM, the lower accelerating voltage limits the achievable resolution, but ptychography can still substantially improve contrast compared to conventional transmission imaging, particularly for light-element materials or thin biological samples where phase contrast is important, but amplitude contrast is weak.

The link to SEM's original mission: high-resolution imaging – is direct: ptychography via 4D-STEM on SEM breaks through the resolution limits that bright-field transmission imaging imposes and does so without the investment of a high-end aberration-corrected TEM.

5.5 Electric and Magnetic Field Mapping

The center-of-mass of the diffraction disk in a 4D-STEM dataset shifts in response to local electric fields in the material (differential phase contrast, DPC imaging). This enables direct imaging of built-in electric fields across p-n junctions, ferroelectric domain walls, and charged defects at nanometer spatial resolution. On SEM, the technique is applicable to samples where field strengths are large enough to produce measurable disk deflections at the lower electron energies used.

6. The Role of the Detector: Why Hardware Defines the Method

6.1 From Film to CCD to Direct Electron Detectors

Electron diffraction experiments were recorded on photographic film for most of the twentieth century. Film has excellent spatial resolution and a large dynamic range, but digitization is slow and labor-intensive, and quantitative intensity extraction is imprecise. CCD cameras coupled to

scintillators and fiber-optic tapers were introduced in the 1990s and enabled digital data collection, but introduced noise and limited the achievable dynamic range.

The decisive technological shift came with the development of direct electron detectors: devices in which the electron beam strikes the active sensor layer directly, without an intervening scintillator or fiber optic. Two main detector architectures have emerged: monolithic active pixel sensors (MAPS) and hybrid pixel detectors. Both outperform CCD-based cameras on essentially all metrics relevant to electron diffraction.

6.2 Detector Architectures Compared

Detector type	MAPS (rolling shutter)	MAPS (global shutter)	Hybrid pixel	Fibre-coupled CCD
Single-electron sensitivity	Yes	Yes	Yes (zero noise)	No
Dynamic range	Moderate	Moderate	Very high (counting + ToT)	Low
Frame rate (fps)	100–400	100–400	Up to 10 000+	1–10
Pixel size (µm)	~5	~5	55	~15–25
Counting mode	No	No	Yes	No
Time resolution	No	No	1.56 ns (ToA)	No
Radiation hardness	Moderate	Moderate	Very high	Low
SEM-compatible	No (large TEM only)	Some	Yes (FeliS)	Rarely

Table 1. Comparison of detector technologies for transmission electron diffraction. ToT = Time over Threshold (proxy for energy deposition); ToA = Time of Arrival.

6.3 The Hybrid Pixel Advantage

In a hybrid pixel detector, the sensor layer (which absorbs the incident electron) is bonded to a separate readout chip via microscopic solder bumps (“bumps”) on a per-pixel basis. Each pixel operates independently and can process signals simultaneously, enabling true single-electron counting. The readout chip contains threshold discrimination circuitry in every pixel: only signals above the threshold (corresponding to a single electron interaction) are counted, and electronic noise below the threshold is entirely rejected.

The consequence is a detector with effectively zero readout noise in counting mode. This is qualitatively different from any CCD or MAPS detector, where noise accumulated over an exposure limits sensitivity. For electron diffraction, it means that weak reflections near the background limit can be detected reliably, improving the completeness and accuracy of the diffraction dataset.

Hybrid pixel detectors also offer a very high dynamic range: while operating in counting mode for weak signals, many designs (including those based on the Timepix3 and Medipix3 ASICs) can simultaneously measure the energy deposited by each electron via the Time over Threshold (ToT) signal. This allows strong reflections to be measured without saturating, even when weak reflections are still counted individually.

6.4 The Medipix/Timepix Family

The dominant hybrid pixel chip platform for scientific instrumentation outside high-energy physics is the Medipix/Timepix family, developed by CERN's Medipix collaboration. The collaboration includes research institutions across Europe and has produced several chip generations, each licensed to commercial manufacturers for instrument development.

- Medipix3: optimized for high dynamic range imaging in counting mode, with charge-summing across pixels to improve spatial resolution at low energies.
- Timepix3: adds per-pixel time-of-arrival (ToA) measurement with approximately 1.56 ns resolution, enabling time-resolved experiments and simultaneous spatial and temporal event reconstruction.

ASI's detectors are based on the Timepix3 ASIC, manufactured by CERN and licensed by ASI. The sensor layer is a 300 µm thick silicon sensor, with 256×256 pixels at 55 µm pitch, for a total active area of 14×14 mm². The data-driven readout architecture means that only pixels that register an electron hit send data, enabling sustained hit rates far exceeding those of frame-based cameras.

A separate commercial family, based on technology developed at Paul Scherrer Institut (PSI) and commercialized by Dectris, uses the EIGER and JUNGFRAU ASICs. These are widely used at synchrotron X-ray sources and are also available for electron microscopy. The architectural choices differ in detail (pixel pitch, dynamic range mode, readout scheme) but the fundamental principle – hybrid pixel single electron counting – is the same.

7. The ASI FeliS: Hybrid Pixel Detection for SEM

7.1 Design Philosophy

The FeliS is ASI's hybrid pixel detector purpose-built for the transmission geometry on scanning electron microscopes. The core challenge is integration: a SEM pole piece has limited working distance and chamber space, the sample is positioned above the detector, and no modifications should interfere with standard SEM imaging, EDS, or EBSD. The FeliS addresses this through a retractable lid mechanism: the detector is stored under a protection lid during conventional SEM use. The lid slides open below the sample when transmission experiments are required.

The FeliS uses the Timepix3 chip and can therefore operate in both frame-based and data-driven modes. In 4D-STEM applications, frame-based acquisition synchronized to the SEM scan delivers a full diffraction pattern at every scan point. In 3D-ED applications, the continuous data-driven mode records every electron hit with its precise position and time stamp, maximizing the sensitivity for weak diffraction signals during a tilt series.

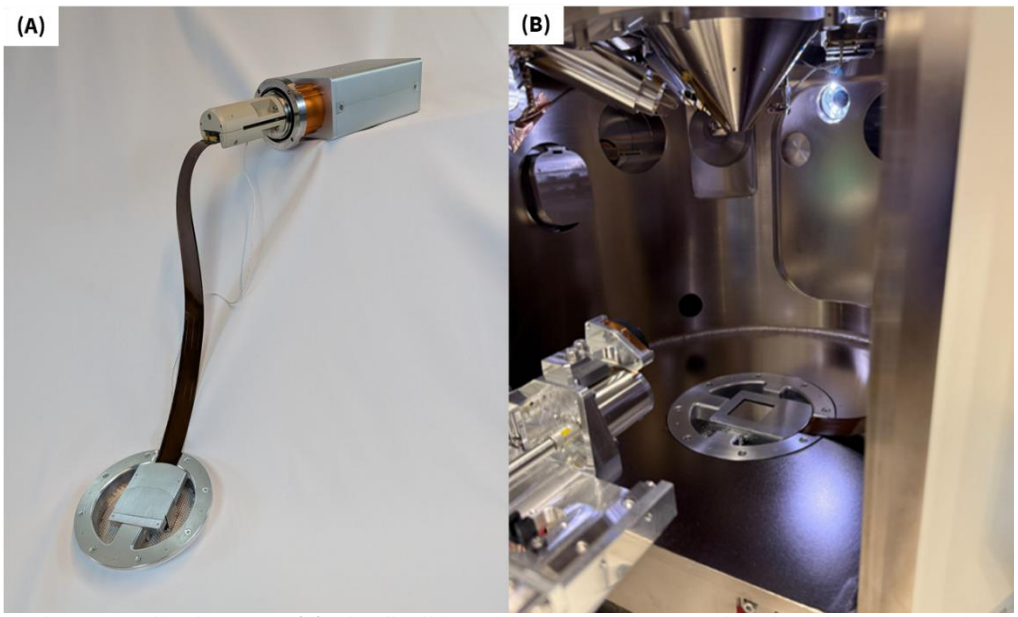

Figure 2. The ASI FeliS detector. (A) The flexible cable-mounted sensor head enables integration into a wide range of SEM chambers while allowing optimization of the projection geometry. (B) FeliS installed in a Zeiss Gemini SEM chamber at FAU-Erlangen-Nürnberg. A motorized protective lid shields the sensor from contamination and supports lift-off characterization, plasma cleaning, and other workflows without requiring chamber venting.

7.2 Key Specifications

Parameter	Value
ASIC	Timepix3 (CERN Medipix collaboration)
Pixel matrix	256 × 256 pixels
Pixel pitch	55 µm
Active area	14.1 × 14.1 mm²
Sensor	300 µm silicon (standard)
Energy range	10 – 40 kV (SEM operating range)
Readout mode	Data-driven (event-based) and frame-based
Max frame rate	Up to 10 000 frames/s (frame-based mode)
Counting threshold	Single-electron threshold (~800 eV equivalent)
Time resolution	~1.56 ns per-pixel ToA (Timepix3)
Retraction mechanism	Motorized retractable lid for in-vacuum switching
Vacuum compatibility	SEM chamber compatible (UHV-safe materials)

Table 2. FeliS key specifications. ToA = Time of Arrival.

7.3 Integration and Workflow

The FeliS integrates with the SEM via 10 Gb Ethernet to the acquisition computer. The detector occupies a standard port of the SEM chamber (e.g. CF65) and is mounted on a dedicated adapter flange below the sample stage, depending on the SEM model. The retractable lid is actuated by a small, motorized drive inside the chamber; switching between transmission and conventional mode takes under 30 seconds without breaking vacuum.

For 3D-ED, the SEM's built-in stage controls the tilt series, and the FeliS acquisition software (based on the ASI SERVAL platform) records the diffraction patterns synchronized with stage movements. Data are exported in standard formats compatible with PETS2 and Instamatic for structure solution. For 4D-STEM, the scan signal from the SEM column triggers event-mode acquisition on the FeliS, and the resulting 4D dataset is exported for processing in tools such as py4DSTEM or LiberTEM.

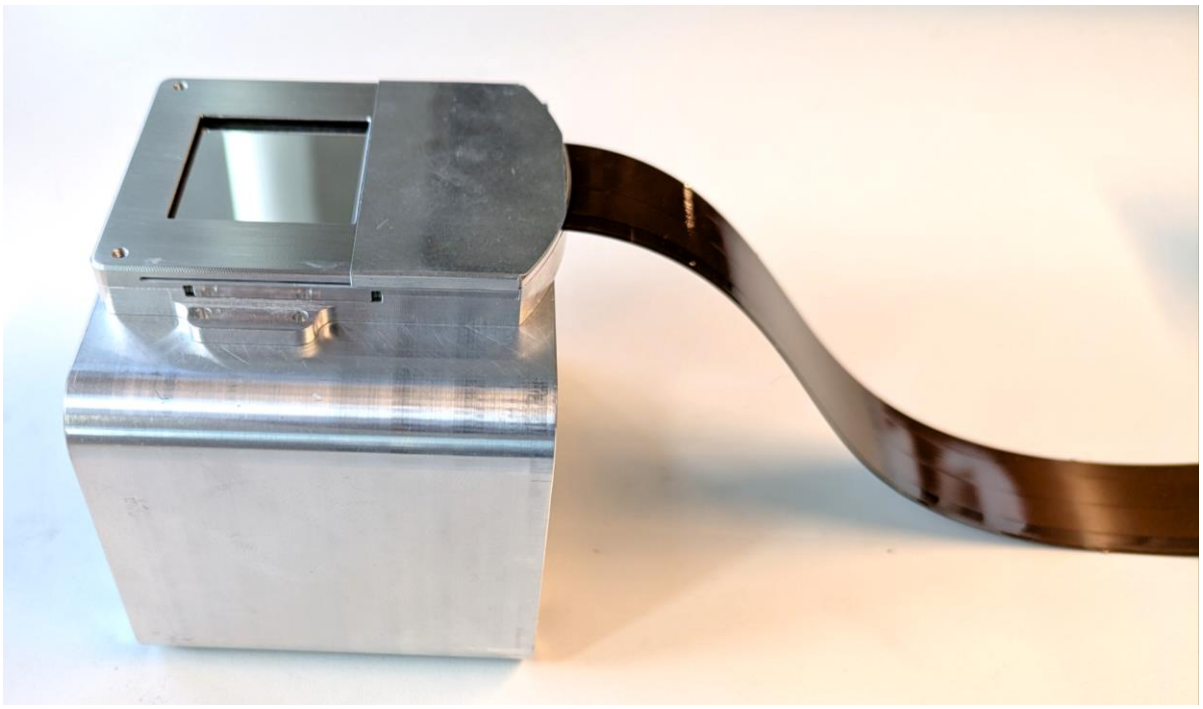

Figure 3. The FeliS sensor module. Four individual 256×256-pixel Timepix3 ASICs are assembled in a monolithic 2×2 configuration, forming a 512×512-pixel detector with 55×55 µm² pixel size. The compact sensor module is mounted on a dedicated PCB for direct placement below the SEM sample.

7.4 Why ASI?

Why choose the ASI FeliS?

- Timepix3 chip: zero-noise single-electron counting combined with per-pixel timing
- Purpose-built for SEM: the retractable lid design is the only solution that allows seamless switching between transmission and conventional SEM modes without venting the chamber.
- Open data ecosystem: SERVAL software outputs to open formats; direct compatibility with PETS2, Instamatic, LiberTEM, and py4DSTEM.
- Proven in peer-reviewed research: early-adopter publications from EMAT (Antwerp), FZ Jülich, and other leading centers demonstrate working 3D-ED and 4D-STEM on SEM.
- Expert support: ASI provides on-site installation, sample measurement support, and training by detector physicists and electron microscopists.

8. What Can and Cannot Be Done on a SEM

8.1 Physical Constraints

The lower accelerating voltage of a SEM (10–30 kV, typically 30 kV for diffraction experiments) compared to TEM (80–300 kV) has direct consequences for the physics of the experiment. The electron wavelength at 30 kV ($\lambda \approx 7$ pm) is larger than at 200 kV ($\lambda \approx 2.5$ pm), which means diffraction angles are larger for equivalent d-spacings. The interaction cross-section is larger, which means stronger signals from thinner samples, but also stronger multiple scattering for thicker samples.

The practical consequence is a sample thickness limit of approximately 100–300 nm for 3D-ED data of acceptable quality at 30 kV. Thicker samples produce excessive multiple scattering (dynamical diffraction), which distorts reflection intensities and degrades structure determination accuracy. For 4D-STEM applications, especially orientation mapping, somewhat thicker samples (up to ~500 nm) are acceptable because only diffraction pattern geometry rather than precise intensities is required.

The absence of a post-sample lens system in a SEM (unlike TEM) means that the diffraction camera length is fixed by the physical distance between sample and detector. For the FeliS, this is determined by the working distance and pole-piece geometry of the particular SEM model. Camera length calibration is performed once per instrument configuration.

Depending on the application requirements, the implementation of a projection lens is feasible on a project base.

8.2 Suitable Sample Types

Samples well-matched to 3D-ED on SEM include materials that form nanocrystalline or microcrystalline powders, are too beam-sensitive to tolerate the higher electron doses of TEM, or are available only in small quantities. Specific categories include:

- Pharmaceutical compounds and their polymorphs, including active pharmaceutical ingredients in formulated products
- MOFs and porous inorganic frameworks synthesized as nanoparticulate powders
- Environmental minerals and secondary phases in geological samples
- Battery electrode materials: lithiation/delithiation products, solid electrolyte interphase components

- Organic pigments, dyes, and coatings
- Nanoparticles synthesized by wet chemistry or vapor deposition

Samples that are poor candidates for SEM-based 3D-ED include those that cannot be thinned to below ~300 nm without destroying the structure of interest, very strongly scattering materials (heavy elements, thick crystals) where dynamical effects are severe even at 30 kV, and materials requiring the high spatial resolution of an aberration-corrected TEM for interpretation.

8.3 Sample Preparation

The sample preparation requirement for SEM-based 3D-ED is less demanding than for TEM, precisely because the stronger interaction at lower voltages. Three approaches are commonly used:

- Powder dispersal: nanoparticulate or microcrystalline powders are dispersed in a solvent (ethanol, isopropanol) by ultrasonication and drop-cast onto standard TEM support grids (holey carbon or silicon nitride membranes). This is entirely equivalent to TEM sample preparation and requires no specialized equipment.
- Crushing and dispersal: bulk polycrystalline samples are crushed in a mortar, dispersed in solvent, and drop-cast. Particle sizes of 100–1000 nm are typical after gentle crushing.
- FIB lamella: where a specific region of interest must be targeted (e.g., a grain boundary, an interface, or a precipitate), focused ion beam (FIB) milling can prepare a site-specific lamella thinned to below 300 nm. FIB preparation is standard on FIB-SEM instruments and is increasingly available on dedicated FIB workstations.

A particular advantage of SEM-based 3D-ED: the FIB-SEM combination instrument is extremely well suited to this workflow. Prepare a FIB lamella on the FIB column, transfer to the SEM stage, install the FeliS, and acquire a 3D-ED dataset from the same instrument in a single session.

8.4 Advantages of 30 kV Compared to TEM

- Lower beam damage: the inelastic scattering cross-section is higher at lower energies, but the damage per useful diffraction event is lower for many beam-sensitive organic and biological materials at 30 kV versus 200–300 kV.

- Larger sample chamber: SEM chambers can accommodate heating, cooling, gas injection, and mechanical testing stages not typically available on TEM. In-situ 3D-ED during phase transformations is feasible on a SEM.
- Accessibility: SEM time is cheaper and more available. Routine screening of many crystalline phases, which would require significant beam time allocation on a TEM, can be done cost-effectively on a SEM.

9. Summary and Outlook

9.1 Summary

This white paper has described two advanced transmission electron techniques: three-dimensional electron diffraction (3D-ED) and four-dimensional scanning transmission electron microscopy (4D-STEM). It explained how the ASI FeliS hybrid pixel detector makes them accessible on a standard SEM.

The key points are:

- 3D-ED enables single-crystal structure determination from particles as small as 100 nm, extending crystallography to sample sizes inaccessible by X-ray diffraction.
- 4D-STEM provides nanometer-resolution maps of crystal orientation, strain, phase distribution, and electric fields from a single scan dataset.
- Both methods are established on TEM and are now transitioning to SEM, driven by the development of fast, sensitive, noise-free hybrid pixel detectors.
- The SEM geometry offers lower beam damage, larger chamber flexibility, and dramatically greater instrument accessibility compared to TEM.
- The ASI FeliS, based on the Timepix3 chip, is the only purpose-built hybrid pixel detector for SEM transmission experiments, with a retractable mechanism that preserves full conventional SEM functionality.

9.2 Outlook

The adoption curve for new electron microscopy techniques follows a consistent pattern: a breakthrough in detector or source technology enables a new measurement, early adopters demonstrate compelling science, and the technique becomes routine once user-friendly software and installation protocols are established. EBSD followed this path over twenty years; 4D-STEM on TEM is following it now, with commercial software packages from Gatan, Thermo Fisher Scientific, and others appearing in the last two years.

SEM-based 3D-ED and 4D-STEM are at the early-adopter stage. The hardware is demonstrated and working. The software pipeline for structure determination is mature (PETS2, Instamatic, SHELX). The community of potential users is enormous: every materials characterization laboratory with a SEM and an interest in structure — which includes most pharmaceutical, geological, battery, and semiconductor research groups — is a potential adopter.

The question for most laboratories is not whether these methods will become routine, but whether to be among the early adopters who define the workflows and establish the techniques in their field, or to wait for full commoditisation.

Contact Amsterdam Scientific Instruments to discuss whether the FeliS is compatible with your SEM model, to request a demonstration measurement on your samples, or to receive a quotation. → www.amscins.com | info@amscins.com

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