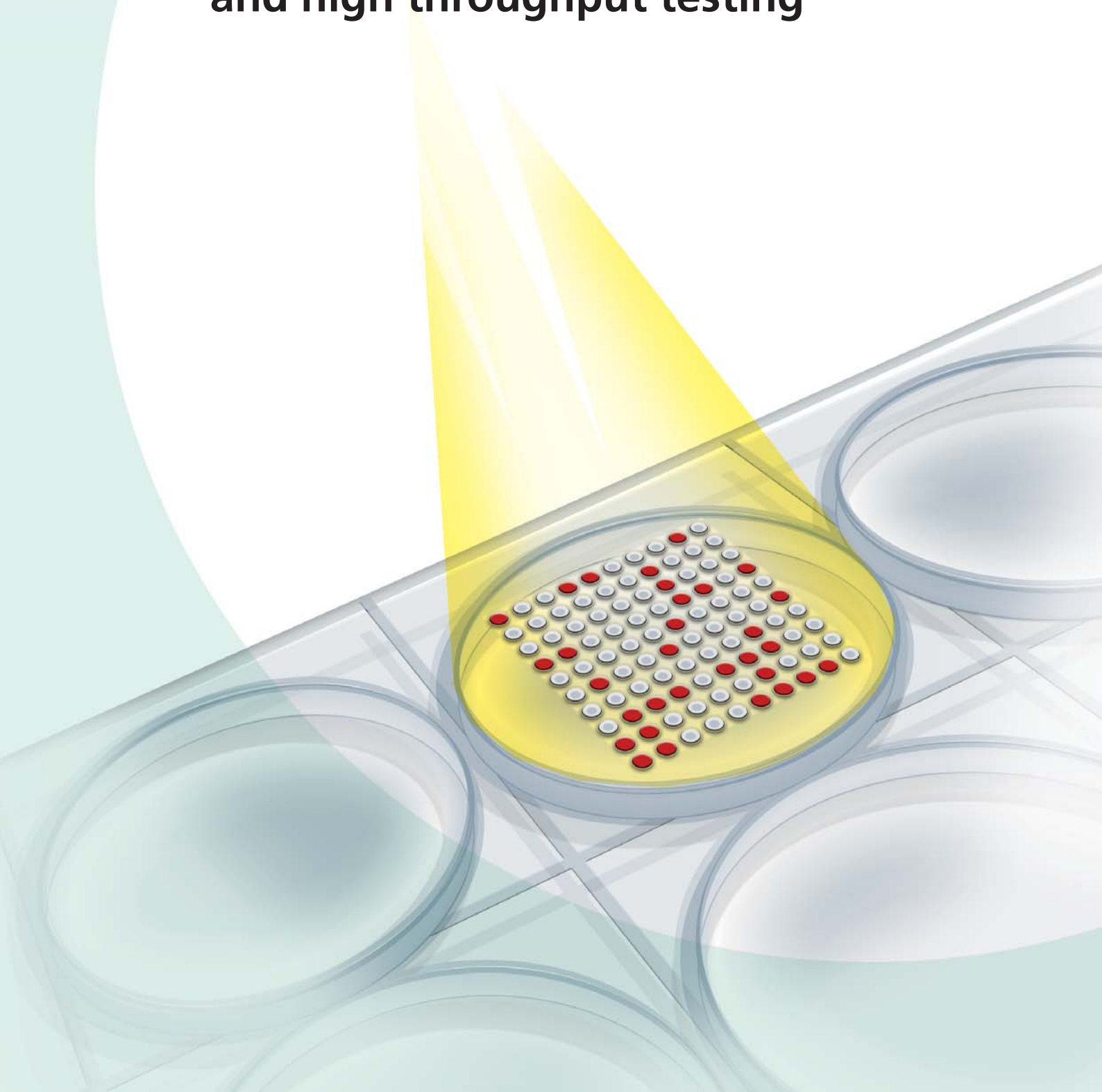


HISTO SPOT SSO

**Fully automated HLA-SSO assay –
ideal for on-call
and high throughput testing**



Test principle: Miniature reverse SSO "blot"

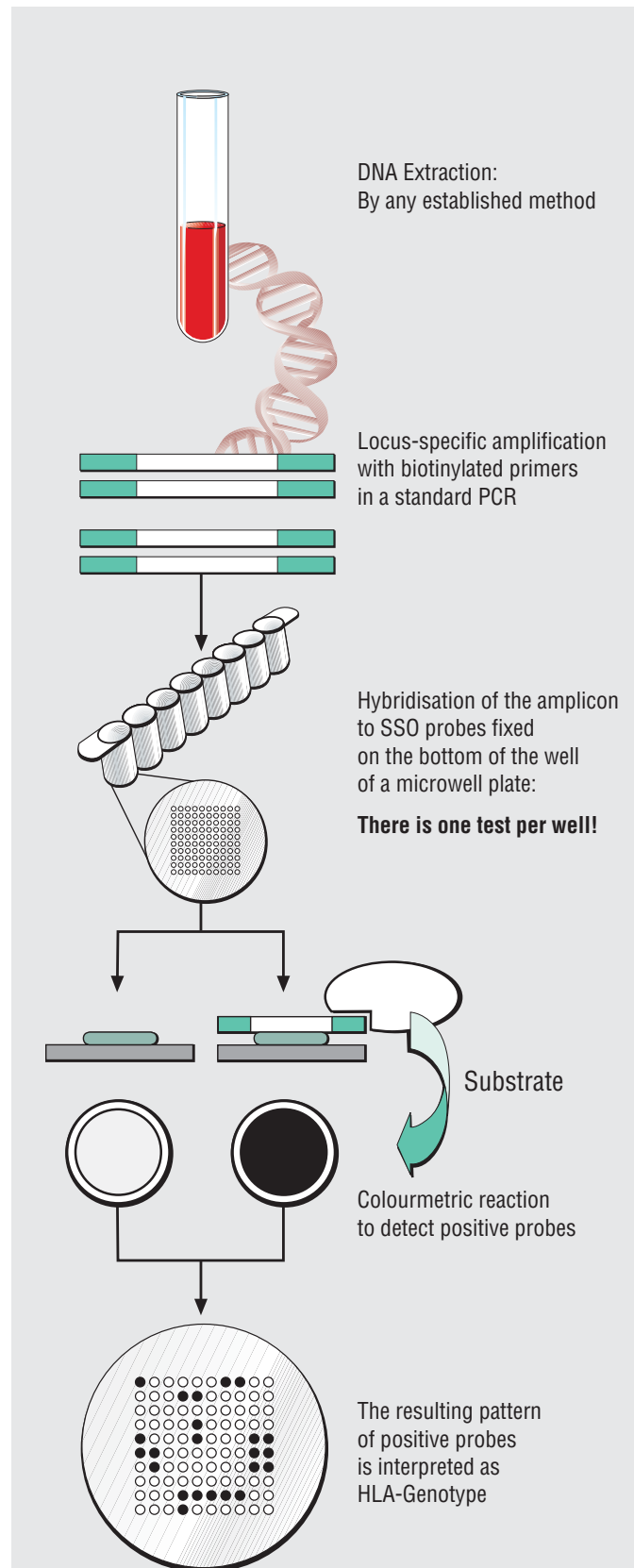
The **HISTO SPOT SSO** assay in combination with the **Mr.SPOT** processor represents the most automated typing system for low to medium resolution HLA typing on the market.

Advantages:

- Fully automated SSO assay
- Flexible: 1 to 96 tests per run
- Fast: 3 hours from sample to result for a single type
- Resolution better than low resolution SSP, and comparable with other SSO tests
- easy to handle
- reagents provided in working dilution
- no extra washing runs necessary
- convenient interpretation software included
- potential for high resolution typing in the future

With HISTO SPOT on-call testing can now be automated because of the speed of the process in combination with a very low hands-on time and simple test procedure.

The flexibility and convenience of the system makes HISTO SPOT the ideal solution for low as well as high throughput requirements.



The Mr.SPOT processor

The Mr.SPOT processor runs the whole assay from amplicon to result. After PCR amplification the amplicon tray is placed into Mr.SPOT. It adds hybridisation buffer to each well and transfers each amplicon together with the hybridisation buffer to the test well containing the immobilized SSO probes.

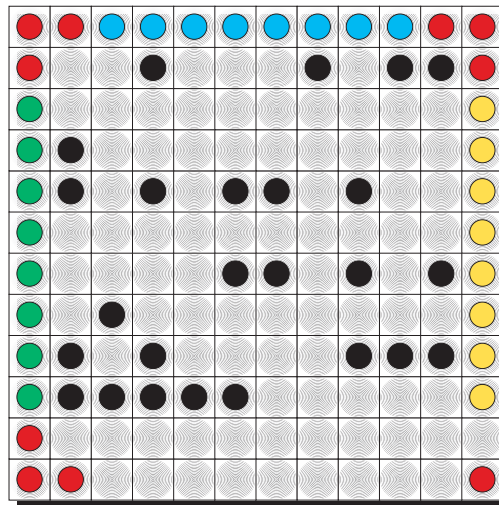
Then the processor performs all necessary washing steps and all pipetting steps for the colourmetric reaction.

Afterwards the resulting probe pattern (coloured dots in the bottom of each well) is photographed by the integrated camera and the image is transferred to the interpretation software.



Comparison of workflow for different SSO typing systems

Luminex	Strip Test	HISTO SPOT
<p>PCR set up: 30 min</p> <p>PCR run: 1,5 h</p> <p>Denaturation: 10 min pipetting 10 min incubation</p> <p>Neutralization: 5 min pipetting</p> <p>Hybridization: 5 min pipetting 15 min incubation</p> <p>Washing (3x): 5 min pipetting 5 min centrifugation</p> <p>Labeling: 5 min pipetting 5 min incubation 5 min pipetting 5 min incubation 10 min pipetting</p> <p>Load samples into Luminex Analyzer</p> <p>→ ~ 2 h hands on time</p> <p>Data acquisition Transfer data from Luminex</p>	<p>PCR set up: 30 min</p> <p>PCR run: 1,5 h</p> <p>Denaturation: 5 min pipetting 10 min incubation</p> <p>Prepare Reagents: 10 min pipetting</p> <p>Load amplicon into washer</p> <p>→ ~ 1 h hands on time</p> <p>Strip scanning and data acquisition</p> <p>Transfer data from scanner</p>	<p>PCR set up: 30 min</p> <p>PCR run: 1,25 h</p> <p>Load amplicon into Mr.SPOT</p> <p>→ ~ 0,5 h hands on time</p> <p>Data acquisition Transfer data from Mr.SPOT</p>
<p>Result after 5 h</p>	<p>Result after 5 h</p>	<p>Result after 3 h</p>



The interpretation software

The software analyses the image by comparing spot intensity to the background.

There is a positive control probe and a number of probes coding for the HLA locus typed and for the lot number of the test strip.

A pattern of positive probes is generated based on the strength of each probe versus a control probe. This pattern is matched to an HLA type.

Typing reports are given according to the latest IMGT/HLA nomenclature reports. Alternatively, it is possible to choose a result format based on CWD alleles (ASHI) or NMDP codes.

The red spots represent the control probes that have two functions: They are positive controls that indicate mastermix was added to the PCR run. They are also positional probes that enable the software to locate the picture.

The black spots represent positive specific probes. The last three black spots are control probes, positive for all DR alleles, which form the basis for defining the reactivity of the other probes in the well and indicate that the PCR was successful.

The green, blue and yellow spots identify the kit, lot and batch of the test respectively.

Traceability of samples and reagents

A barcode reader identifies the lots of all reagents and of the test-strips. This information will be included in the test reports. Additionally, a pattern of control spots in the test wells codes for the HLA locus and lot of the test.

After creating the sample sheet it is possible to print a layout for the setup of the PCR and for the correct positioning of the amplicon tray and the test strips in the processor.

The user is guided through the setup of the test by step-by-step instructions on the touchscreen of the Mr. SPOT processor. There are exact instructions on the minimum volumes of each reagent that are required for a specific run. The user has to confirm the addition of each reagent before the run starts.