

CLAIMS

1. A method for subjecting a plurality of microwells containing cells to a *high-content* assay, said method comprising:
 - a) Acquiring at least one image of said plurality of microwells;
 - b) In said image, detecting a plurality of areas of interest, each area of interest corresponding to a single cell;
 - c) Measuring at least one derived property, and, optionally, at least one direct property of said areas of interest, where said one or more properties is a selection property;
 - d) Selecting a subset of said plurality of microwells, where said microwells belonging to the subset contain areas of interest selected based on said at least one selection property;
 - e) Extrapolating an output parameter from a property measured in the set of areas of interest selected, where said property is defined as output property, said output property being distinct from said selection properties where said output parameter is the processing of an output property measured in said set of areas of interest.
2. A method according to claim 1, wherein said direct property is a property associated with a single area of interest, i.e., a property measured by assessing the single area of interest and said derived property is a property associated with a multiplicity of areas of interest, i.e., a property which requires the assessment of two or more areas of interest to be measured.
3. A method according to one of claims 1 or 2, wherein one of said derived properties is a relationship property, or is a coexistence property, between one or more areas of interest included in the same microwell.
4. A method according to claim 1, wherein said selection is made

by imposing inclusion criteria, wherein said inclusion criteria comprise:

- identifying, from said derived and, optionally, direct measured properties, one or more selection properties;
- imposing, for each of said selection properties, the threshold value, or the range of values, within which said selection property must fall.

5. A method according to one of claims 1 to 4, wherein at least one of said selection properties is a cumulative property.
6. A method according to one of claims 1 to 5, which comprises the selection of a first set of areas of interest based on a first selection property and, within said first set of areas of interest, a selection of a subset of areas of interest based on a second selection property, preferably, said first selection property is a cumulative property and said first set of areas of interest corresponds to a subset of microwells and said second selection property is a direct or relative property, and said subset of areas of interest corresponds to a subset of cells embedded in said subset of microwells.
7. A method according to one of claims 1 to 6, wherein said output parameter is the result of any statistical processing of the output property measured in each area of interest which belongs to the set of areas of interest selected.
8. A method according to one of claims 1 to 7, wherein said at least one image is acquired with an image acquisition device configured to acquire at least one image of said plurality of microwells.
9. A method according to one of claims 1 to 8, wherein said image

is analyzed and processed to return a measurement of said properties, said analysis and processing process comprising the following steps:

- Identifying, in an image containing a plurality of microwells, the zones which correspond to the microwells;
- Detecting, within said zones corresponding to the microwells, a plurality of areas of interest, each area of interest corresponding to one of said cells contained in said plurality of microwells;
- Measuring at least one property of each of said areas of interest;
- Selecting a set of areas of interest based on one or more of said measured properties, said one or more properties defined as selection properties;
- Extrapolating an output parameter from measured properties in said set of areas of interest.

10. A method according to one of claims 1 to 9, wherein said plurality of microwells is embedded in a microfluidic device comprising at least 15,000, or at least 18,000, preferably 19,200 microwells.

11. A method according to one of claims 1 to 10, wherein said microwells are inverted open microwells, i.e., are microwells which have an upper end and a lower end both open.

12. A method according to one of claims 1 to 11, which subjects said plurality of microwells to a dynamic test, where several images of the same field are acquired at successive times (*time-lapse imaging*) and the measurement of said at least one property, at time t_0 and, subsequently, at time $t_1, t_2, \dots, t_n$, returns an analysis which reflects the variations of said property over time.

13. A method according to one of claims 1 to 12, wherein said cells,

while kept in said plurality of microwells, are exposed to one or more agents which impact said output parameter.

14.A system (1) for subjecting a plurality of microwells containing cells to a *high-content* assay, said system comprising:

- an image acquisition device (2) configured to acquire at least one image of said plurality of microwells (3); and
- a data processing unit (4) configured to:
 - In said image, detecting a plurality of areas of interest, each area of interest corresponding to a single cell;
 - measure at least one derived property, and, optionally, at least one direct property of said areas of interest, where said one or more properties is a selection property;
 - select a subset of said plurality of microwells, where said microwells belonging to the subset contain areas of interest selected based on said at least one selection property;
 - extrapolate an output parameter from a property measured in the set of areas of interest selected, where said property is defined as output property, said output property being distinct from said selection properties where said output parameter is the processing of an output property measured in said set of areas of interest.

15.A computer program for subjecting a plurality of microwells containing cells to a *high-content* assay, said computer program comprising instructions which, when the program is executed by a data processing unit, cause the processing unit to perform the following steps:

- Measuring at least one derived property, and, optionally, at least one direct property of said areas of interest, where said one or more properties is a selection property;

- Selecting a subset of said plurality of microwells, where said microwells belonging to the subset contain areas of interest selected based on said at least one selection property;
- Extrapolating an output parameter from a property measured in the set of areas of interest selected, where said property is defined as output property, said output property being distinct from said selection properties where said output parameter is the processing of an output property measured in said set of areas of interest.