## Claims

1. A method for a cell-based drug screening assay comprising the steps of:

- culturing a cell population in a culturing environment,
- defining at least two monitoring points in time in dependency of at least one of the cell types, physiological characteristics of the cells, physiological characteristics of formed-tissue, the mode of action of a drug substance and the culturing environment,
- applying drug substances to the cultured cells at least at one treatment point in time,
- monitoring of the effect of said drug substance on cells or formed tissues at least at said two monitoring points in time.

2. The method of claim 1, wherein said monitoring points in time are selected from the group consisting of

- a time point relevant for an early-stage of a specific cell, preferably between four and seven days after culturing the cells in the environment, and
- a time point for a late-stage, preferably fourteen to seventeen days after culturing the cells in the environment, and
  at a point in time defined by the absence or presence of a specific physiological characteristic e.g. hypoxia or change in cell growth kinetics,
  and
- a time point relevant for measuring drug efficacy after a drug-free recovery phase.

3. The method of claim 1 and 2, wherein monitoring of cell proliferation continues one to seven or more days after termination of the drug application.

4. The method of claims 1 to 3, wherein cells are cultured in an environment, which comprises a structural compound and a linker compound, preferentially comprising a multi-branched polyethylene glycol with vinyl sulfone end groups and a peptide comprising at least two cysteines or, generally thiol groups enabling polymerizing to a three-dimensional matrix.

5. The method of claim 4, wherein said three-dimensional environment promotes three-dimensional growth of the encapsulated cells, preferentially spheroid growth.

6. The method of claim 4 and 5, wherein said three-dimensional environment is cast in a recipient such as a gel casting device or a standard cell-culture device, preferably a 384-well plate.

7. The method of claim 1 to 6, wherein said cells is selected of the group of any type of cell, cell line, differentiated cell, and cancer cell such as colon, pancreatic and ovarian cancer cell.

8. The method of claim 1 to 6, wherein said cells are cells of which the growth is promoted by the drug treatment.

9. The method of claim 1 to 8, wherein said drug substances are selected of the group of

(i) any chemical, natural or synthetic substance and any substance of known drug screens; or

(ii) any combination of substances according to i);

(iii) any substance(s) according to i) or ii), either in solution or combined with any carrier, nanoparticle or delivery device.

10. The method of claim 1 to 9, wherein monitoring of cell characteristics is assayed by
metabolic activity
and/or flow cytometrie,
and/or DNA content,
and/or protein quantification,
and/or cell counting,
and/or image-based analysis,
and/or levels of hypoxic and angiogenic markers,
and/or Ribonucleic acid (RNA) levels,
and/or protein expression levels,
and/or mass spectrometry,
and/or immunohistochemistry.

11. The method of claim 1 to 10, wherein the maximally tolerated dose of a drug substance is determined for a given type of cell.

12. The method of claim 1 to 11, wherein the lowest effective dose of a drug substance is determined for a given type of cell.

13. The use of the method of claim 1 to 12 for evaluating
(i) reprogramming of differentiated cells with respect to an applied substance and
(ii) differentiation of stem cells with respect to an applied substance.

## Claims

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- applying drug substances to the cultured cells at least at one treatment point in time,
- monitoring of the effect of said drug substance on cells or formed tissues at least at said two monitoring points in time.

2. The method of claim 1, wherein said monitoring points in time are selected from the group consisting of

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- a time point relevant for measuring drug efficacy after a drug-free recovery phase.

3. The method of claim 1 and 2, wherein monitoring of cell proliferation continues one to seven or more days after termination of the drug application.

4. The method of claims 1 to 3, wherein cells are cultured in an environment, which comprises a structural compound and a linker compound, preferentially comprising a multi-branched polyethylene glycol with vinyl sulfone end groups and a peptide comprising at least two cysteines or, generally thiol groups enabling polymerizing to a three-dimensional matrix.

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6. The method of claim 4 and 5, wherein said three-dimensional environment is cast in a recipient such as a gel casting device or a standard cell-culture device, preferably a 384-well plate.

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(i) reprogramming of differentiated cells with respect to an applied substance and

(ii) differentiation of stem cells with respect to an applied substance.