or protozoa. Thus, the teaching of WO 2015/044191 A1 does not allow to distinguish between cells of the same host material, in particular between eukaryotic cellular bodies of the same host material.

Besides that, it is known from WO 2007/056027 A1 to use electrical fields instead of applying heat to liquids for the reduction of cells. The electrodes used in WO 2007/056027 A1 are made of a rather thick dielectric material, which is covered with electrically conductive material for connection to a power supply.

A disadvantage of the assembly in WO 2007/056027 A1 is the requirement of relatively high potential differences between the electrodes due to the large distance between the electrodes. The large distance between the electrodes also causes inhomogeneities in the applied electric fields, which leads to irregular lysis along the electrodes. Additionally, the dielectric material makes up the main part of the electrode, as the dielectric material has to be rather thick in order to be coated with electrically conductive material. Thus, microfluidic applications, such as a tip for a pipette, cannot be realized with the teaching of WO 2007/056027 A1.

Another disadvantage of the known methods is that larger cells are more easily lysed than smaller cells due to their size. Therefore, if one desires to lyse small cells in a biological sample made up of small and large cells, the larger cells are inevitably lysed as well.

Accordingly, it is an objective of the present disclosure to eliminate or at least alleviate the disadvantages of the known methods and devices of the prior art.

In particular, it is an objective of the present disclosure to provide a method for specific electroporation and/or lysis of eukaryotic cellular bodies in a biological sample with at least two subgroups of eukaryotic cellular bodies for microfluidic applications, which method allows the specific electroporation and/or lysis of a subgroup, preferably of only one subgroup, of eukaryotic cellular bodies in a sample with at least two subgroups of eukaryotic cellular bodies.

Further, it is an objective of the present disclosure to provide a device for specific electroporation and/or lysis of eukaryotic cellular bodies in a biological sample for microfluidic

applications. In another example it is an objective of the present disclosure to provide a method for manufacturing a device suitable for specific electroporation and/or lysis cellular bodies in a biological sample. In another example it is an objective of the present disclosure to provide a method for specific electroporation and/or lysis of eukaryotic cellular bodies in a biological sample using the device.

Summary

In an aspect, the disclosure provides a method of targeted electroporation and/or lysis of eukaryotic cellular bodies in a biological sample with at least two subgroups of eukaryotic cellular bodies, wherein each subgroup has a different susceptibility to electroporation and/or lysis in electric fields, comprising the following steps:

- transferring the biological sample in a chamber,
- exposing the biological sample to an electric field in the chamber, wherein the electric field is generated by at least two electrodes which are coated with a dielectric material with a relative permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more and
- choosing the electric parameters of the electric field such as the field strength, the frequency or the wave form so that the subgroups are differently affected by said electric field for electroporation and/or lysis.

In another aspect, there is provided a device, preferably handheld device, suitable for targeted electroporation and/or lysis of eukaryotic cellular bodies in a biological sample, comprising at least one chamber for receiving the biological sample and at least two electrodes for generating an electric field in the chamber, which electrodes are coated with a dielectric material with a permittivity greater than 1, preferably greater than 3.9, more preferably 60 or more, wherein the distance between the electrodes is below 1 mm, preferably below 550 µm, more preferably below 100 µm, but greater than 5 µm, wherein the device further comprises an adjustment unit that enables the adjustment of electric parameters of the electric field such as the field strength, the frequency or the wave form.

In another aspect, there is provided a method of targeted electroporation and/or lysis of cellular bodies in a biological

sample, comprising the following steps:
- actuating a piston of an air displacement pipette such that the biological sample is transferred into a chamber within a tip of the pipette
- exposing the biological sample to an electric field in the chamber, wherein the electric field is generated by at least two electrodes which are coated with a dielectric material with a relative permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more, wherein the distance between the electrodes is below 1 mm, preferably below 550 µm, more preferably below 100 µm, but greater than 5 µm and
- choosing the electric parameters of the electric field such as the field strength, the frequency or the wave form so that the cellular bodies are lysed and/or electroporated.

In another aspect, there is provided a method for manufacturing a device, preferably handheld device, particularly an air displacement pipette, for targeted electroporation and/or lysis of eukaryotic cellular bodies, wherein at least two electrodes are coated with a layer of dielectric material with a permittivity greater than 1, preferably greater than 3.9, more preferably 60 or more, wherein the electrodes are arranged such that the distance between the electrodes is below 1 mm, preferably below 550 µm, more preferably below 100 µm, but greater than 5 µm. Also provided is a method of targeted electroporation and/or lysis of eukaryotic cellular bodies in a biological sample, comprising the following steps:
- transferring the biological sample in a chamber,
- exposing the biological sample to an electric field in the chamber, wherein the electric field is generated by at least two electrodes which are coated with a dielectric material with a relative permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more, and
- choosing the electric parameters of the electric field such as the field strength, the frequency or the wave form so that eukaryotic cellular bodies in a biological sample are affected by said electric field for electroporation and/or lysis.

In other words, the disclosure provides a method of targeted electroporation and/or lysis of eukaryotic cellular bodies in a

biological sample with at least a first and a second group or subgroup of eukaryotic cellular bodies, each group or subgroup having an electroporation and/or a lysis rate, wherein the biological sample is transferred into a chamber and exposed to an electric field in the chamber which is generated by at least two electrodes which are coated with a dielectric material with a relative permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more, wherein for isolating and/or delivering a compound into the cells of the first group or subgroup the electric parameters such as the field strength, the frequency or the wave form of the electric field are chosen to conduct electroporation and/or lysis the first group or subgroup with a different electroporation and/or a lysis rate than the second group or subgroup.

In another example, the disclosure provides a device, preferably handheld device, suitable for targeted electroporation and/or lysis of eukaryotic cellular bodies in a biological sample with at least subgroups of eukaryotic cellular bodies, wherein cellular bodies of each subgroup have a different susceptibility to electroporation and/or lysis in electrical fields, comprising at least one chamber for receiving the biological sample and at least two electrodes for generating an electric field in the chamber, which electrodes are coated with a dielectric material with a permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more, wherein the distance between the electrodes is below 1 mm, preferably below 550 µm, more preferably below 100 µm, wherein the device further comprises an adjustment unit that enables the adjustment of electric parameters of the electric field such as the field strength, the frequency or the wave form so that the subgroups are differently affected with electroporation and/or lysis.

In other words, the disclosure provides a device, preferably handheld device, for targeted electroporation and/or lysis of cellular bodies that comprises a tip for a pipette with at least one chamber for receiving a biological sample and at least two electrodes for generating an electric field in the chamber, which electrodes form an inner surface of the chamber and which electrodes are coated with a dielectric material with a relative permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more, wherein the distance between the elec-

trodes is below 1 mm, preferably below 550 µm, more preferably below 100 µm, wherein the device further comprises an adjustment unit that enables the adjustment of electric parameters of the electric field such as the field strength, the frequency or the wave form.

In another example, the disclosure provides a device that is an air displacement pipette with a suction and pressure means for creating a reduced pressure in a receiving chamber, wherein receiving chamber is suitable for receiving a biological sample, wherein the chamber has at least two electrodes for generating an electric field in the chamber, which electrodes are each on an inner surface of the chamber and which electrodes are coated with a dielectric material with a relative permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more, wherein the distance between the electrodes is below 1 mm, preferably below 550 µm, more preferably below 100 µm. Preferably the device further is connected to or comprises an adjustment unit that enables the adjustment of electric parameters of the electric field such as electric field strength, electric pulse frequency or electric pulse wave form of the electric field.

In another example the disclosure provides a method of targeted electroporation and/or lysis of cellular bodies in a biological sample, comprising the following steps:

- actuating the suction and pressure means of an air displacement pipette such that the biological sample is transferred into a chamber within a tip of the pipette
- exposing the biological sample to an electric field in the chamber, wherein the electric field is generated by at least two electrodes which are coated with a dielectric material with a relative permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more, wherein the distance between the electrodes is below 1 mm, preferably below 550 µm, more preferably below 100 µm, and
- choosing the electric parameters of the electric field such as the field strength, the frequency or the wave form so that the cellular bodies are lysed and/or electroporated.

In another example the disclosure provides a method for manufacturing a device, preferably handheld device, particularly a tip for a pipette, for targeted electroporation and/or lysis of eukaryotic cellular bodies, wherein at least two electrodes are

coated with a layer of dielectric material with a permittivity greater than 3.9, preferably greater than 9, more preferably 60 or more, wherein the electrodes are arranged such that the distance between the electrodes is below 1 mm, preferably below 550 µm, more preferably below 100 µm.

All examples relate to each other and any example described for one aspect commensurately also relates to all other aspects and examples. E.g. the biological sample described for the method or the means for the method can be included or can be part of the device. The device or any part thereof can be used in the inventive methods.

Detailed description of the invention

The present invention provides a method that allows the targeted electroporation and/or lysis of a subgroup of eukaryotic cellular bodies in a biological sample with at least two subgroups of eukaryotic cellular bodies. In this disclosure, eukaryotic cellular bodies comprise nucleated eukaryotic cells, such as lymphocytes, as well as non-nucleated bodies stemming from eukaryotic cells, such as erythrocytes. Alternative terms for eukaryotic cellular bodies are eukaryotic cellular vessels or eukaryotic cellular corpuscles. The eukaryotic cellular bodies may stem from the same organism, e.g. mammals, humans or animals. The various types of eukaryotic cellular bodies have in common that they comprise an eukaryotic cell membrane that encloses an intracellular cavity, usually filled with an aqueous medium. The cell membrane is usually from an eukaryotic cell. The term eukaryotic cellular bodies is mostly associated with eukaryotic cells herein, but also includes bodies without a nucleus, such as erythrocytes. Without being limited to a particular theory, it appears that the eukaryotic cell wall is specifically susceptible for manipulation by the inventive methods for electroporation or lysis, i.e. inducing leakiness or rupture of the cell membrane, respectively. Surprisingly, it was found out by the inventors that eukaryotic cells and their non-nucleated cellular bodies each have particular discernible susceptibilities to the electric parameters in a chamber with coated electrodes according to the invention so that different eukaryotic cellular bodies can be individually targeted without affecting other eukaryotic cellular bodies. This allows specific treat-