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# United States Patent [19] Chang

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- [54] **METHOD OF AND APPARATUS FOR CELL PORTION AND CELL FUSION USING RADIOFREQUENCY ELECTRICAL PULSE**
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- [21] Appl. No.: **555,873**
- [22] Filed: **Jul. 19, 1990**

### Related U.S. Application Data

- [63] Continuation-in-part of Ser. No. 238,670, Aug. 30, 1988, Pat. No. 4,970,154, which is a continuation-in-part of Ser. No. 106,282, Oct. 9, 1987, Pat. No. 4,822,470.
- [51] Int. Cl.<sup>5</sup> ..... **C12M 1/42; C12N 13/00; C12N 15/00; C12N 15/87**
- [52] U.S. Cl. .... **435/287; 435/173.4; 435/173.5; 435/173.6; 435/555; 935/52; 935/85; 935/89; 935/93**
- [58] Field of Search ..... **422/22; 435/173, 287; 935/85, 89, 93, 52**

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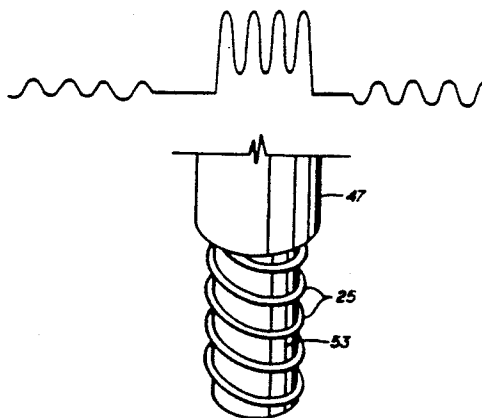
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### [57] ABSTRACT

Disclosed are an apparatus and a method for the poration and fusion of cells using high-power radiofrequency electrical pulses. The electrodes of the apparatus can be hand held or part of integrated equipment with special containers for cells. The electrodes, which are positioned equidistant from each other, are attached to a high power function generator. The power function generator can apply a continuous AC electrical field and/or a high-power pulsed radiofrequency electrical field across the electrodes. The alternating electrical field induces cell congregation by the process of dielectrophoresis. The high-power pulsed radiofrequency electrical field porates or fuses the cells. The method has the ability to porate or fuse biological cells with a very high efficiency. The method can be used to fuse or porate a variety of cells including animal cells, human cells, plant cells, protoplasts, erythrocyte ghosts, liposomes, vesicles, bacteria and yeasts. The method has the unique ability to porate or fuse cells in very small or very large numbers. During the poration or fusions, a variety of chemical agents including DNA, RNA, antibodies, proteins, drugs, molecular probes, hormones, growth factors, enzymes, organic chemicals and inorganic chemicals can be introduced into these cells. The method can also be used to produce new biological species, to make hybridoma cells which produce animal or human monoclonal antibodies and to insert therapeutic genes into human cells which can be transplanted back into the human body to cure genetic diseases.

12 Claims, 12 Drawing Sheets



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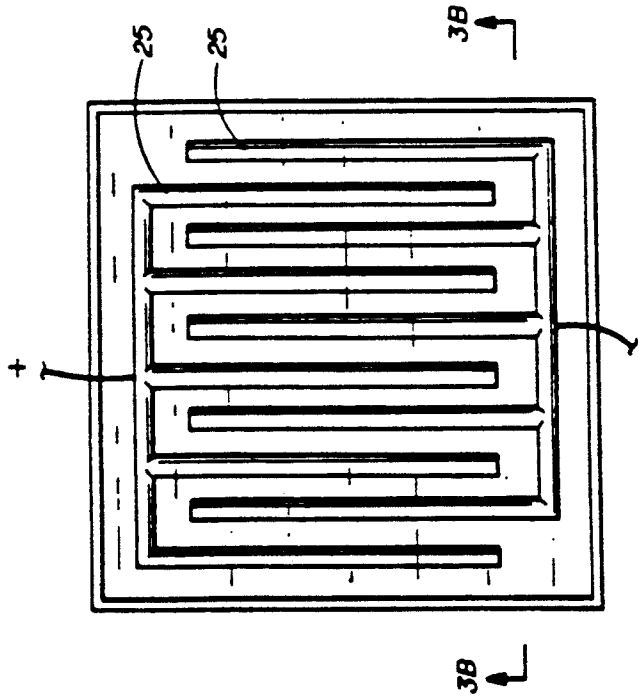


FIG. 3A

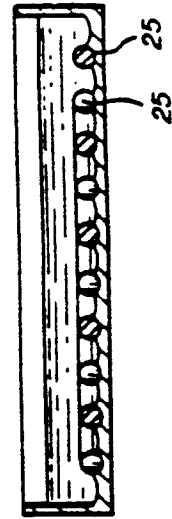


FIG. 3B

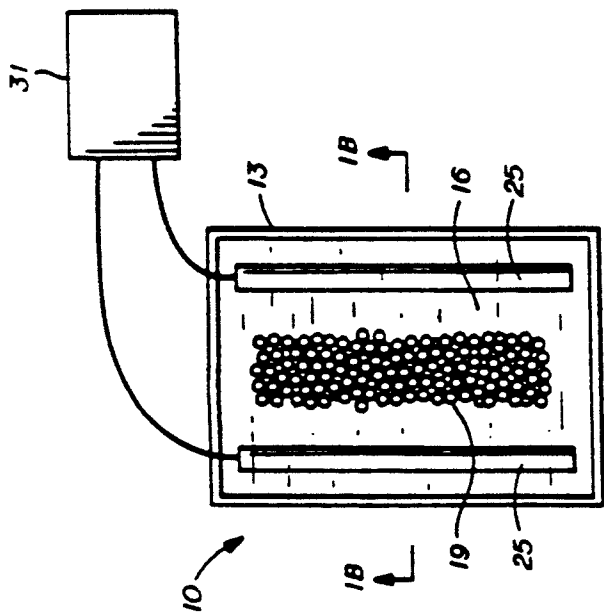


FIG. 1A

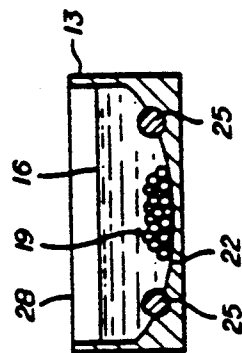
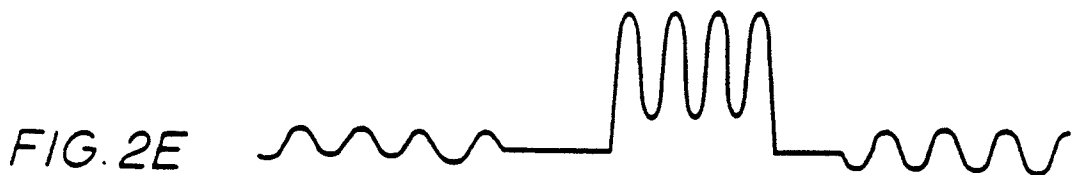
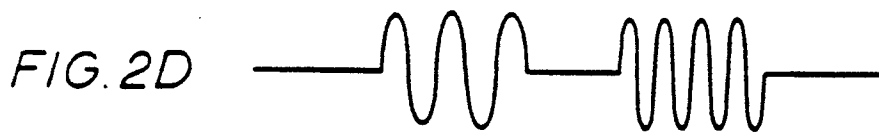
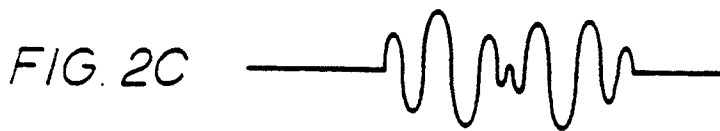
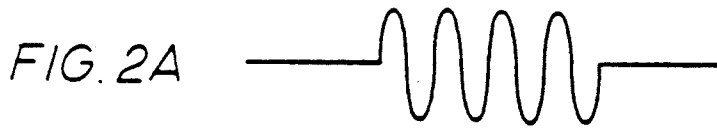


FIG. 1B



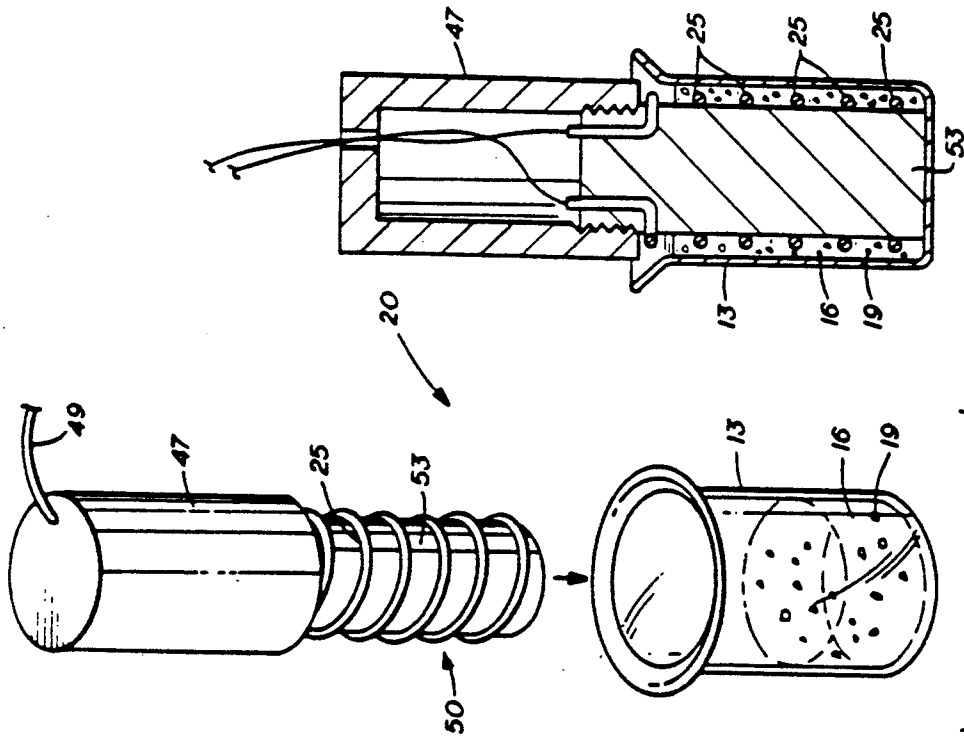


FIG. 5B

FIG. 5A

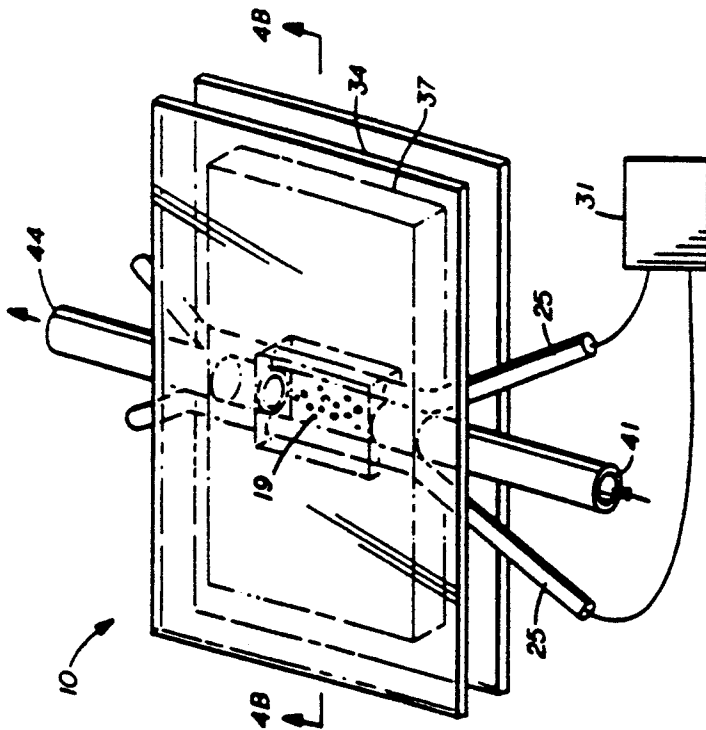


FIG. 4A

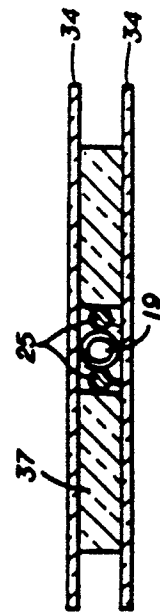


FIG. 4B