US00RE39566E

(19) **United States**

(12) **Reissued Patent**

Elsener et al.

(10) **Patent Number: US RE39,566 E**

(45) **Date of Reissued Patent: Apr. 17, 2007**

(54) **THERMOCYCLER AND LIFTING ELEMENT**

(75) Inventors: **Donat Elsener**, Thun (CH); **Daniel Ryser**, deceased, late of Stafa (CH); by **Werner Ryser**, legal representative, Huhen (CH)

(73) Assignee: **Applera Corporation**, Foster City, CA (US)

(21) Appl. No.: **11/116,441**

(22) Filed: **Apr. 28, 2005**

Related U.S. Patent Documents

Reissue of:

(64) Patent No.: **6,555,792**
Issued: **Apr. 29, 2003**
Appl. No.: **09/672,726**
Filed: **Sep. 28, 2000**

(30) **Foreign Application Priority Data**

Sep. 29, 1999 (CH) 1782/99

(51) **Int. Cl.**

B01L 3/00	(2006.01)
B01L 7/00	(2006.01)
G01N 35/00	(2006.01)
C12M 1/38	(2006.01)
H05B 3/22	(2006.01)

(52) **U.S. Cl.** **219/428**; 219/385; 219/433; 435/288.4; 435/809

(58) **Field of Classification Search** ..... None
See application file for complete search history.

(56) **References Cited**

U.S. PATENT DOCUMENTS

3,080,759	A		3/1963	McQuaid
3,634,651	A	*	1/1972	Siegel et al.
3,933,165	A		1/1976	Budzak et al.
4,094,641	A		6/1978	Friswell
4,096,965	A		6/1978	Lessnig et al.
4,909,992	A	*	3/1990	Björkman
4,948,564	A		8/1990	Root et al.
5,030,418	A	*	7/1991	Miyata
5,159,197	A		10/1992	Wannlund
5,210,015	A		5/1993	Gelfand et al.
5,282,543	A		2/1994	Picozza et al.
5,346,672	A	*	9/1994	Stapleton et al.
5,378,433	A		1/1995	Duckett et al.
5,459,300	A	*	10/1995	Kasman

(Continued)

FOREIGN PATENT DOCUMENTS

DE	195 01 298 C1		2/1996
DE	197 39 119 A1		3/1999
DE	19739119	*	3/1999
EP	0 379 437 B1		7/1990
EP	542422	*	5/1993

(Continued)

OTHER PUBLICATIONS

U. Landegren et al., "A Ligase–Mediated Gene Detection Technique," Science, 241:1077–80 (Aug. 1988).

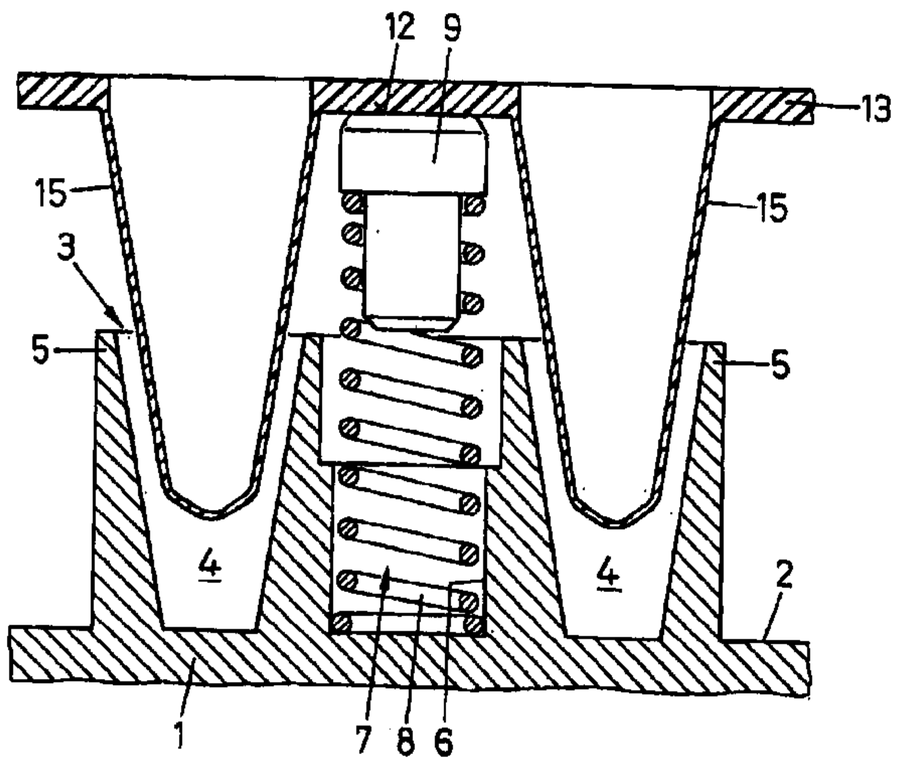
(Continued)

Primary Examiner—Joseph Pelham
(74) *Attorney, Agent, or Firm*—Finnegan Henderson Farabow Garrett & Dunner, L.L.P.

(57) **ABSTRACT**

Some of the blind holes (**6**) between indentations (**4**) of a heating surface (**3**) contain lifting elements (**7**) which, after opening of a cover, release a microtitre plate (**13**) from the heating surface (**3**) and raise said microtitre plate about 2 to 3 mm, so that it can be removed without application of force. Each lifting element (**7**) consists of a coil spring (**8**) and a contact pin (**9**) made of, for example, PEEK which is inserted into said coil spring and presses with a round flat abutting surface (**12**) against the lower surface of the microtitre plate (**13**). The spring constant of the lifting element (**7**) is about 6 N/mm.

33 Claims, 3 Drawing Sheets



U.S. PATENT DOCUMENTS

5,464,541	A		11/1995	Aysta et al.
5,475,610	A		12/1995	Atwood et al.
5,538,848	A		7/1996	Livak et al.
5,582,665	A		12/1996	Eigen et al.
5,602,756	A		2/1997	Atwood et al.
5,604,130	A		2/1997	Warner et al.
5,616,301	A		4/1997	Moser et al.
5,681,492	A	*	10/1997	Van Praet
5,710,381	A		1/1998	Atwood et al.
5,721,136	A		2/1998	Finney et al.
5,741,463	A		4/1998	Sanadi
5,780,717	A		7/1998	Wise et al.
5,928,907	A		7/1999	Woudenberg et al.
6,015,674	A		1/2000	Woudenberg et al.
6,159,368	A		12/2000	Moring et al.
6,162,400	A		12/2000	Schembri
6,190,619	B1		2/2001	Kilcoin et al.
6,197,572	B1		3/2001	Schneebeli
6,251,662	B1		6/2001	Day
6,272,939	B1		8/2001	Frye et al.
6,315,957	B1		11/2001	Feygin et al.
6,406,670	B1	*	6/2002	Earley et al.
6,423,948	B1	*	7/2002	Kwasnlski et al.
6,514,750	B2		2/2003	Bordenkircher et al.
6,638,761	B2		10/2003	Shin et al.
6,719,949	B1		4/2004	Barzilai et al.
2002/0028507	A1		3/2002	Heimberg et al.

FOREIGN PATENT DOCUMENTS

EP	0 542 422	A1	5/1993
EP	0 606 534	B1	7/1994
EP	0 810 030	B1	12/1997
EP	0 810 030	A1	12/1997
EP	0810030	*	12/1997
EP	0 836 884	A2	4/1998
EP	0 895 240	A1	2/1999
EP	0 955 097	A1	11/1999
EP	1 088 590	A1	4/2001
GB	1 427 034		3/1976
JP	S63-008537		1/1988
JP	H05-501647		4/1993
JP	H06-233670		8/1994
JP	H07-5180		1/1995
JP	2645916		5/1997
JP	2727015		12/1997
JP	H09-325100		12/1997
JP	H10-267933		10/1998
JP	H11-326157		11/1999
JP	2001-149801		6/2001
WO	WO 90/08298	A1	7/1990
WO	WO 91/17239	A1	11/1991
WO	WO 97/36681	A1	10/1997
WO	WO 98/42442	A1	10/1998
WO	WO 98/43740	A2	10/1998
WO	WO 98/56506	A1	12/1998
WO	WO 99/17881	A1	4/1999
WO	WO 99/20395	A1	4/1999
WO	WO 01/28684	A2	4/2001

OTHER PUBLICATIONS

D. Nickerson et al., "Automated DNA diagnostics using an ELISA–based oligonucleotide ligation assay," Proc. Natl. Acad. Sci USA, 87:8923–27 (Nov. 1990).

D.C. Uber et al., "Application of Robotics and Image Processing to Automated Colony Picking and Arraying," Bio Techniques, vol. 11, No. 5, 642–44 (1991).

Peter Jones et al., "Integration of Image Analysis and Robotics into a Fully Automated Colony Picking and Plate Handling System," Nucleic Acids Research, vol. 20, No. 17, 4599–4606 (1992).

P. Grossman et al., "High–density multiplex detection of nucleic acid sequences: oligonucleotide ligation assay and sequence–coded separation," Nucl. Acids Res., 22:4527–34 (1994).

Co–pending U.S. Appl. No. 09/848,270, Inventors: Frye et al., Filed: May 4, 2001, Title: System and method for filling a substrate with a liquid sample.

Co–pending U.S. Appl. No. 09/977,225, Inventors: Freudenthal et al., Filed Oct. 16, 2001, Title: System for filling substrate chambers with liquid.

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,141, "Notice Declaring Interference," Paper No. 1, dated Aug. 13, 2003.

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,141, "Interference Initial Memorandum," attached to Aug. 13, 2003 Notice Declaring Interference (Paper No. 1).

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,141, "Shin List of Intended Preliminary Motions Under 37 C.F.R. § 1.633," dated Sep. 29, 2003.

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,141, "Elsener List of Intended Preliminary Motions," dated Sep. 29, 2003.

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,141, "Order Setting Times," Paper No. 18, dated Oct. 2, 2003.

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,141, "Order," Paper No. 19, dated Oct. 7, 2003.

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,411, "Summary of Telephone Conference," Paper No. 22, dated Oct. 28, 2003.

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,141, "Shin Stipulation to Level of Ordinary Skill in the Art," datead Nov. 14, 2003.

Elsener et al. v. *Shin et al.,* Patent Interference No. 105,141, "Summary of Telephone Conference," Paper No. 24, dated Nov. 18, 2003.

Elsener et al. v. *Shin et al.*, Patent Interference No. 105,141, "Shin Revised Stipulation to Level of Ordinary Skill in the Art," dated Nov. 25, 2003.

Elsener et al. v. *Shin et al.*, Patent Interference No. 105,141, "Senior Party Notification of Change of Ownership of the Elsener Patent and Request for Termination of the Interference Proceedings Under 37 C.F.R. § 1.602," dated Jun. 10, 2004.

Elsener et al. v. *Shin et al.*, Patent Interference No. 105,141, "Judgment—Rule 602," Paper No. 40, dated Jul. 20, 2004.

Opposition to EP 1 088 590 B1 (European Patent Application No. 00 810 855.7), filed Jan. 16, 2003 at European Patent Office, on behalf of AB Applied Biosystems.

Opposition to EP 1 088 590 B1 (European Patent Application No. 00 810 855.7), Decision on the Termination of the Opposition Proceedings, issued Jun. 30, 2004, with English translation.

* cited by examiner

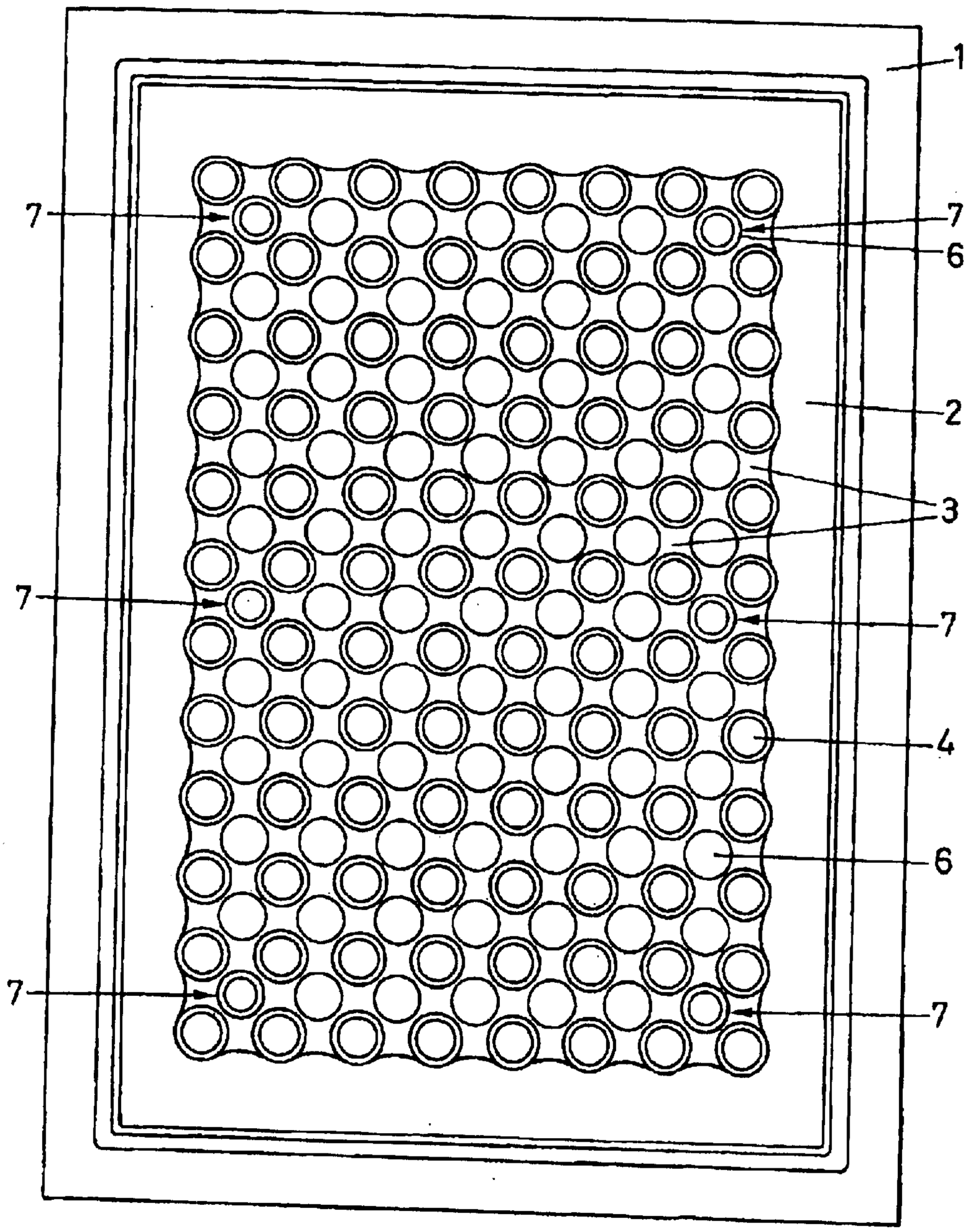


FIG. 1

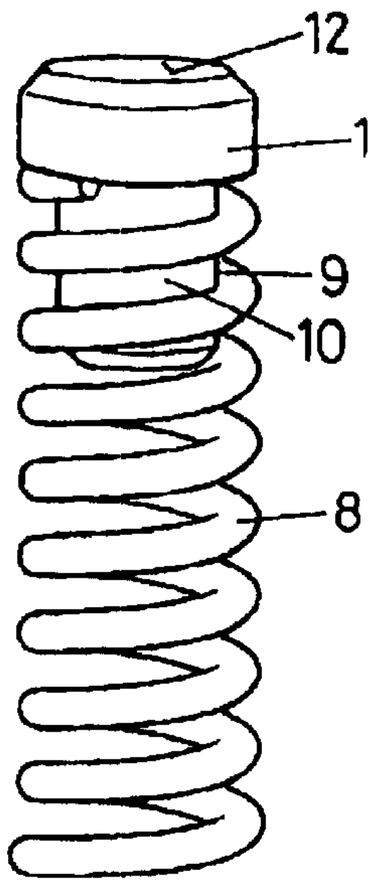


FIG. 3

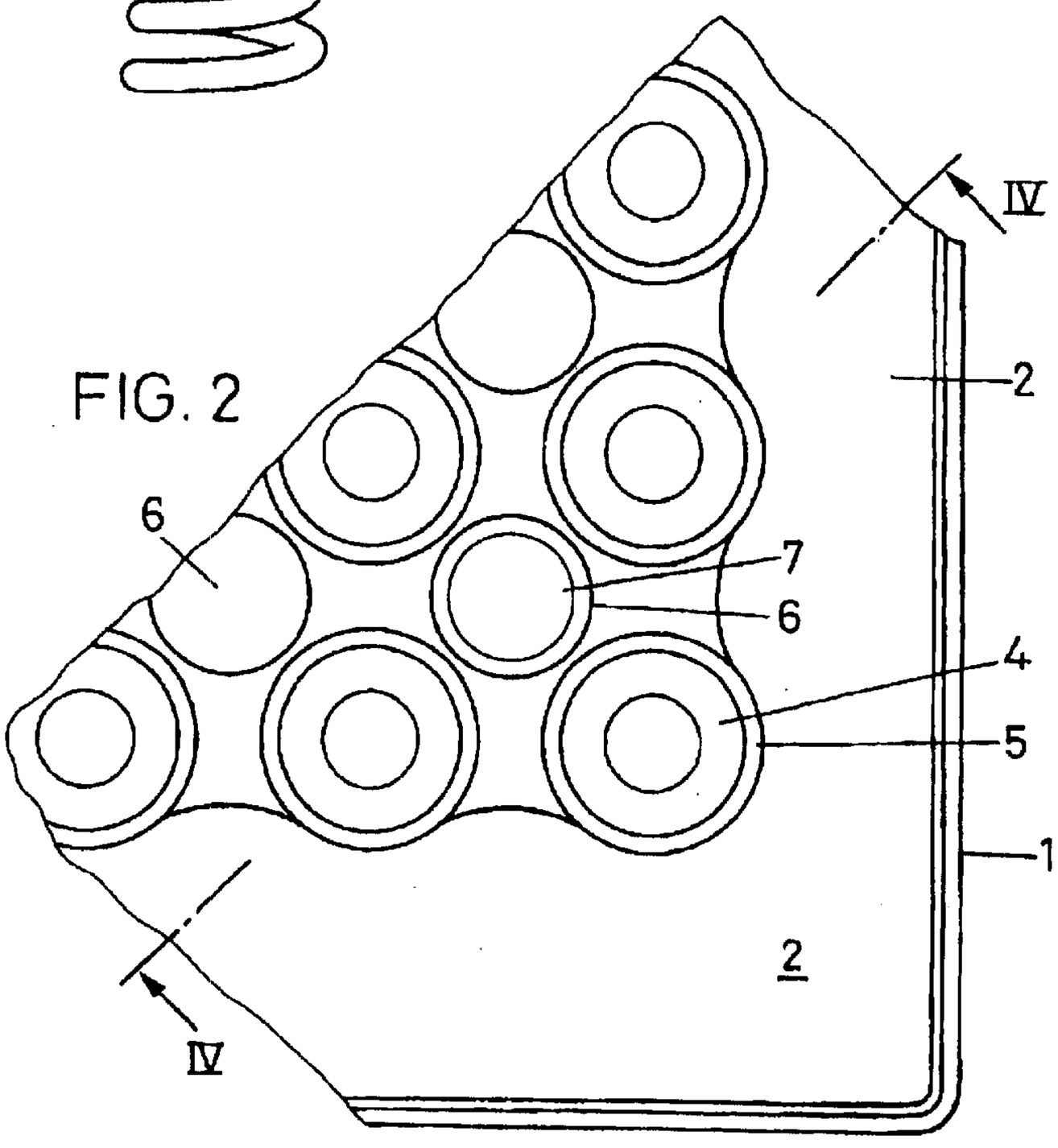


FIG. 2

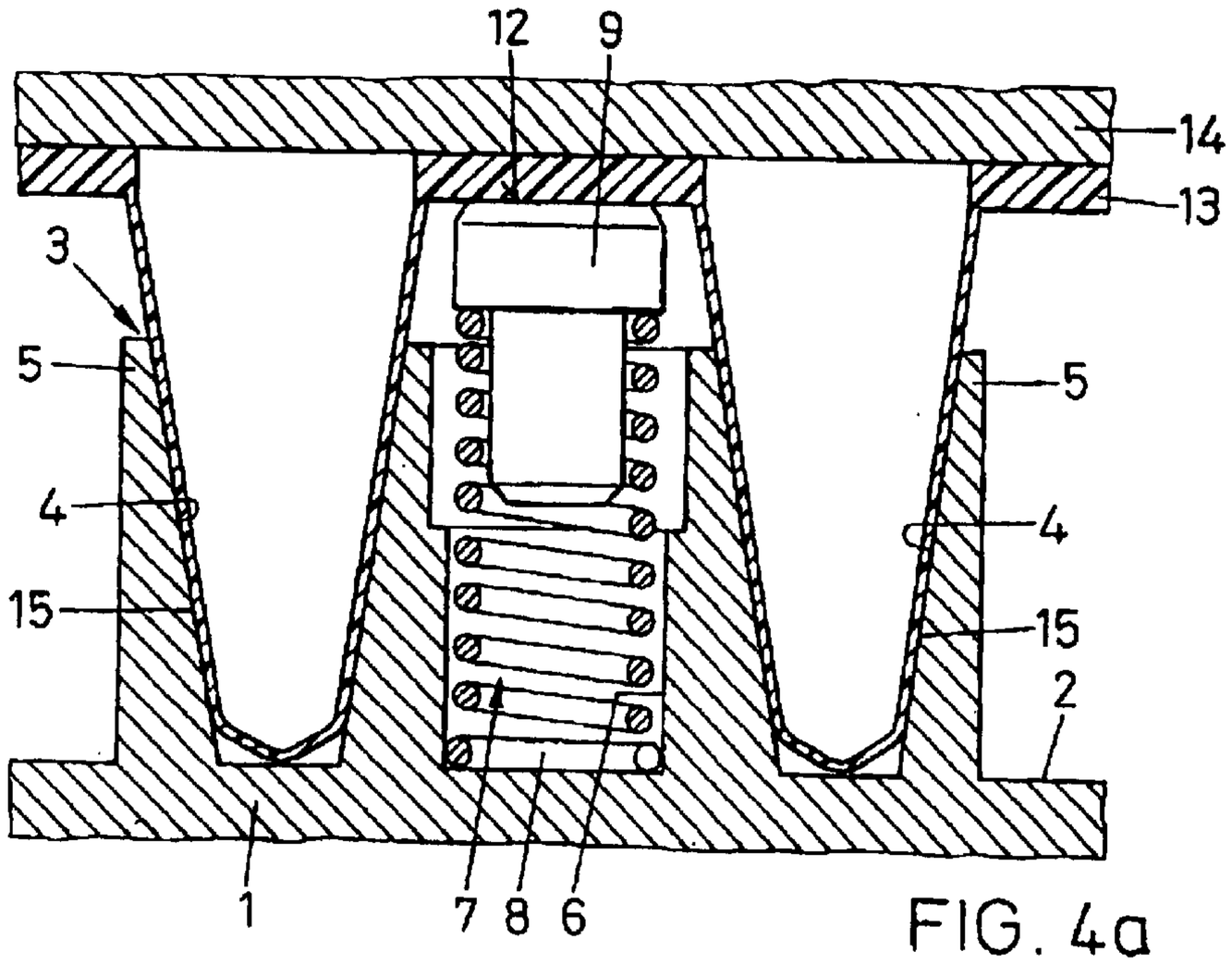


FIG. 4a

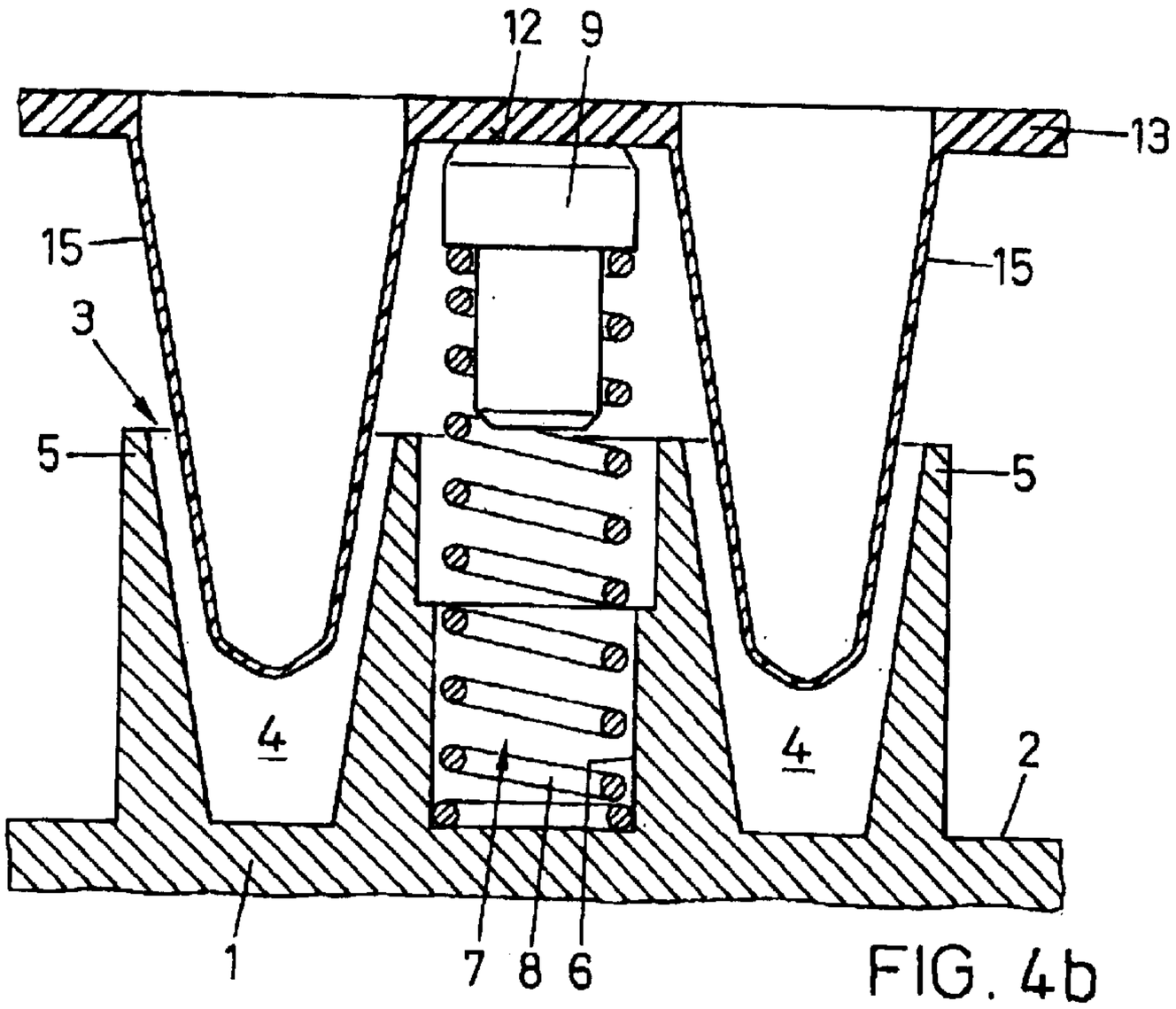


FIG. 4b

THERMOCYCLER AND LIFTING ELEMENT

Matter enclosed in heavy brackets [ ] appears in the original patent but forms no part of this reissue specification; matter printed in italics indicates the additions made by reissue.

FIELD OF THE INVENTION

The invention relates to a thermocycler. Such devices are used for subjecting the content of the wells of microtitre plates to temperature cycles which initiate specific chemical reactions. It also relates to lifting elements for use in thermocyclers.

PRIOR ART

In known thermocyclers of the generic type, there is the problem that the microtitre plate which, in the interests of good heat transfer, rests closely against the heating surface frequently becomes baked onto it and can then be detached from it only with very great difficulty. This either necessitates complicated manipulations or requires suitable and correspondingly heavy and expensive handling devices for applying relatively large forces of 150 N or more. A possible aid is the use of Teflon spray, which can prevent the microtitre plate from baking on. However, this must be repeated for every plate and complicates the procedures.

SUMMARY OF THE INVENTION

It is the object of the invention to improve a known thermocycler of the generic type so that the microtitre plates can be raised and removed after each treatment without particular application of force. This object is achieved by the features in the characterizing clause of claim **1**.

It has been found that, as a result of the measures according to the invention, the microtitre plate is raised after removal of the cover, which permits convenient gripping and lifting thereof without application of force. This may substantially facilitate the manual removal of the microtitre plate, but in particular the removal can also be effected without any manual intervention, by means of handling devices of the type otherwise usual in the laboratory.

Furthermore, the invention provides particularly suitable lifting elements for thermocyclers according to the invention.

BRIEF DESCRIPTION OF THE DRAWINGS

The invention is explained in more detail below with reference to Figures which show only an embodiment.

FIG. **1** shows a plan view of the heating plate of a thermocycler according to the invention,

FIG. **2** shows, on a larger scale, a cut-out from a plan view according to FIG. **1**,

FIG. **3** shows a perspective view of a lifting element according to the invention,

FIG. **4**a shows a section along IV—IV in FIG. **2**, in addition with microtitre plate and cover, and

FIG. **4**b shows a section corresponding to FIG. **4**a with the cover removed.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

The thermocycler, which may be suitable, for example, for holding an 8×12 microtitre plate having the dimensions

85 mm×130 mm, has a heating plate **1** which forms a heating surface **3** which is surrounded by an edge strip **2** and is somewhat higher than said edge strip and in which round indentations **4** are arranged in a regular square grid, each of which indentations is surrounded by an all-round wall **5** (FIG. **2**) projecting beyond the base level of the heating surface **3**. In each case, a blind hole **6** is provided between four indentations **4**.

Six lifting elements **7** are arranged in six of the blind holes **6** altogether, distributed approximately uniformly over the heating surface **3**. Each of the lifting elements **7** consists (FIG. **3**) of a cylindrical coil spring **8** of stainless steel, the lowermost winding of which is somewhat wider than the other windings, and a contact pin **9** whose approximately cylindrical shaft **10** is inserted into the upper end of the coil spring **8** and is held therein by friction.

The shaft **10** carries an approximately disc-like head **11** which projects laterally from it and against whose lower surface the upper end of the coil spring **8** abuts, while its upper surface forms a round flat abutting surface **12**. The contact pin **9** is rotationally symmetrical and is produced as a single piece from a heat-resistant plastic, such as PEEK, PTFE, FP, PPS or PI, for example by the injection moulding process. It may also consist of, for example, ceramic, but the production is then as a rule more complicated and more expensive. The contact pin **9** is between 3 mm and 8 mm, preferably about 6 mm, long. The diameter of the abutting surface **12** is between 3 mm and 7 mm, preferably about 5 mm.

The lifting element **7** has a length of between 15 mm and 20 mm, preferably of about 16 mm. Its spring constant in the relaxed position is between 5 N/mm and 7.5 N/mm, in particular 6 N/mm. It is of course also possible to choose other dimensions and properties in adaptation to different designs of the heating plate and depending on the density with which the lifting elements **7** are arranged on the heating surface and which is 1 per 18.4 cm^2 in the case described above and, as a rule, is at least 1 per 30 cm^2.

The coil spring **8** is dimensioned in each case so that the somewhat wider lowermost winding is slightly radially compressed in the blind hole **6** so that there is a frictional lock between said winding and the wall of the blind hole **6**. The lifting element **7** is thus adequately fixed but can nevertheless easily be removed. The other windings are free from the wall of the blind hole **6** so that the compression of the coil spring **8** is not hindered.

When the thermocycler is used, the microtitre plate **13**, which usually consists of plastic, e.g. polypropylene, is placed on the heating surface **3** (FIGS. **4**a, **4**b) manually or preferably by means of a suitable handling device, e.g. a robot arm, and a hinged cover **14** of the thermocycler is lowered onto said microtitre plate so that each of its wells is pressed into a corresponding indentation **4** and rests against its wall (FIG. **4**a). This ensures good heat transfer between the heating plate **1** and the samples in the wells **15**. The coil springs **8** of the lifting elements **7**, which, in the relaxed state, project about 6 mm above the edges of the walls **5**, are compressed by the pressure exerted by the microtitre plate **13** on the abutting surfaces **12** of its contact pins **9** and are shortened by 2 to 3 mm.

After the thermal treatment of the samples in the microtiter plate, which, for example to initiate a PCR reaction, may undergo a relatively large number of temperature cycles, each of which may consist of, for example, heating from 4° C. to 96° C. with subsequent cooling to 4° C., the cover **14** is swivelled up again. Each of the compressed

lifting elements **7** exerts an upward force of about 15 N on the microtitre plate **13**. This is sufficient to detach the microtitre plate **13** from the heating surface **3** even if it is baked onto the latter and to raise it, possibly with a delay of a few seconds (FIG. **4**b). The microtitre plate **13** raised in this manner and no longer connected to the heating surface **3** can now be removed easily and without application of great force, which again can be effected by a robot arm.

It has been found that it is generally sufficient if the lifting elements together exert a force of about 0.8 N/cm^2, preferably 1 N/cm^2, on the microtitre plate. Contact pins made of PEEK have proved suitable in that they are thermally stable and do not bake onto microplates of the conventional materials, such as polypropylene, so that the slight frictional lock is sufficient to hold the lifting elements **7** in the blind holes **6**.

Apart from the lifting elements **7**, the thermocycler can correspond to a known type, e.g. PTC 225 Tetrad from MJ Research, Inc. It is also possible to retrofit known thermocyclers with lifting elements.

What is claimed is:

1. A thermocycler having a heating plate which forms a heating surface for holding a microtitre plate whose wells are held in indentations provided in the heating surface, and have a cover which can be lowered and raised relative to the heating surface, said cover serving for pressing the microtitre plate against the heating surface, wherein a plurality of elastically compressible lifting elements for rising and detaching of the microtitre plate from the heating surface are distributed over the heating surface, said lifting elements projecting beyond the edges of the indentations at least when the cover is raised.

2. The thermocycler according to claim **1**, wherein the projection of the lifting elements is at least 2 mm, preferably at least 5 mm.

3. The thermocycler according to claim **1**, wherein the density of the lifting elements is at least 1 per 30 cm^2.

4. The thermocycler according to claim **1**, wherein each lifting element is removably fixed to the heating surface.

5. The thermocycler according to claim **1**, wherein each lifting element is inserted into a blind hole in the heating surface.

6. The thermocycler according to claim **4**, wherein the fixing of the lifting element is effected by frictional locking with the walls of the blind hole.

7. The thermocycler according to claim **1**, wherein the lifting element comprises an elongated spring element which is compressible in the longitudinal direction and carries a contact part which forms an abutting surface, oriented transversely to the longitudinal direction, at the upper end of the lifting element.

8. The thermocycler according to claim **7**, wherein the contact part consists of plastic, preferably PEEK, PTFE, FP, PPS or PI.

9. The thermocycler according to claim **7**, wherein the spring element is in the form of a coil spring and the contact part is in the form of a contact pin which comprises a shaft surrounded by the upper part of the coil spring and a laterally projecting head which rests on the upper end of the coil spring and whose upper surface forms the abutting surface.

10. The thermocycler according to claim **9**, wherein the lowermost winding of the coil spring is somewhat wider.

11. The thermocycler according to claim **9**, wherein the contact pin is rotationally symmetrical.

12. The thermocycler according to claim **11**, wherein both the shaft and the head of the contact pin are essentially cylindrical.

13. The thermocycler according to claim **7**, wherein the length of the lifting element is between 15 mm and 20 mm and the diameter of the abutting surface is at least 3 mm.

14. The thermocycler according to claim **7**, wherein the spring constant of the lifting element is at least 5 N/mm.

15. A thermocycler comprising:

a heating plate which forms a heating surface for holding a microtitre plate whose wells are held in indentations provided in the heating surface;

a cover which can be lowered and raised relative to the heating surface, said cover serving for pressing the microtitre plate against the heating surface; and

a plurality of elastically compressible lifting elements for rising and detaching of the microtitre plate from the heating surface positioned over the heating surface.

16. The thermocycler according to claim 15, wherein at least a portion of each lifting element is positioned above the edges of the indentations at least when the cover is raised.

17. The thermocycler according to claim 15, wherein said lifting elements are positioned between the heating surface and the microtiter plate at least when the cover is raised.

18. The thermocycler according to claim 15, wherein each lifting element is removably fixed to the heating surface.

19. The thermocycler according to claim 15, wherein each lifting element is inserted into a blind hole in the heating surface.

20. The thermocycler according to claim 18, wherein the fixing of the lifting element is effected by frictional locking with the walls of a blind hole in the heating surface.

21. The thermocycler according to claim 15, wherein at least one of the lifting elements comprises an elongated spring element which is compressible in the longitudinal direction.

22. The thermocycler according to claim 21, wherein the elongated spring element carries a contact part which forms an abutting surface, oriented transversely to the longitudinal direction, at the upper end of the lifting element.

23. The thermocycler according to claim 15, wherein at least one of the lifting elements comprises an elongated spring element in the form of a coil spring which is compressible in the longitudinal direction.

24. The thermocycler according to claim 23, wherein the lowermost winding of the coil spring is wider than the other windings of the coil spring.

25. A thermocycler comprising:

a heating plate which forms a heating surface for holding a microtitre plate whose wells are held in indentations provided in the heating surface;

a cover which can be lowered and raised relative to the heating surface, said cover serving for pressing the microtitre plate against the heating surface; and

a plurality of elastically compressible lifting elements for rising and detaching of the microtitre plate from the heating surface positioned over the heating plate.

26. The thermocycler of claim 25, wherein the lifting elements are positioned over the heating surface of the heating plate.

27. The thermocycler of claim 25, wherein the lifting elements project beyond the edges of the indentations at least when the cover is raised.

28. A thermocycler comprising:

a heating plate which forms a heating surface for holding a microtitre plate whose wells are held in indentations provided in the heating surface;

a cover which can be lowered and raised relative to the heating surface, said cover serving for pressing the microtitre plate against the heating surface; and

a plurality of lifting elements for rising and detaching of the microtitre plate from the heating surface positioned over the heating surface.

29. The thermocycler of claim 28, wherein the lifting elements are elastically compressible.

30. The thermocycler of claim 28, wherein the lifting elements comprise spring elements.

31. The thermocycler of claim 30, wherein the spring elements comprise coil springs.

32. The thermocycler of claim 28, wherein the lifting elements project beyond the edges of the indentations at least when the cover is raised.

33. A thermocycler comprising:

a heating plate which forms a heating surface for holding a microtitre plate whose wells are held in indentations provided in the heating surface;

a cover which can be lowered and raised relative to the heating surface, said cover serving for pressing the microtitre plate against the heating surface; and

at least one elastically compressible lifting element for rising and detaching of the microtitre plate from the heating surface, said lifting element positioned between the heating surface and the microtiter plate at least when the cover is raised.

* * * * *