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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 5 :
G02B 21/34, G01N 1/30

A1

(11) International Publication Number: WO 94/23326

(43) International Publication Date: 13 October 1994 (13.10.94)

(21) International Application Number: PCT/GB94/00671

(22) International Filing Date: 30 March 1994 (30.03.94)

(30) Priority Data:
9306815.3 1 April 1993 (01.04.93) GB

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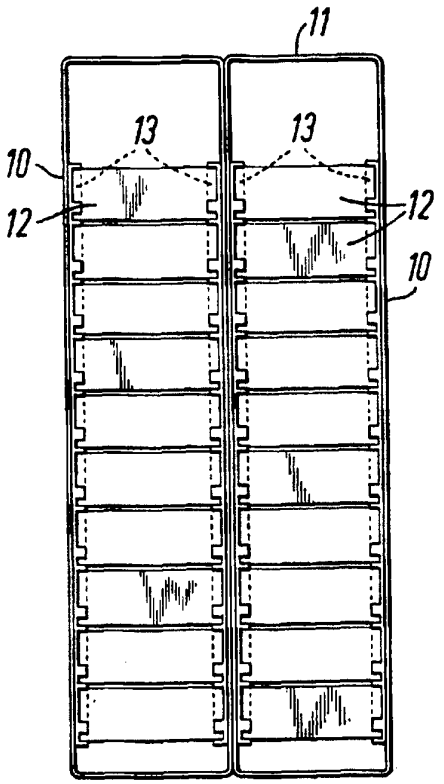
(81) Designated States: GB, JP, US, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE).

Published
With international search report.

(54) Title: MICROSCOPE SLIDE HOLDER

(57) Abstract

A microscope slide holder has a planar hollow frame (10) and series of clips (13) for retaining a number of microscope slides (12) in a generally flat plane. The holder is used for uniform processing, e.g. in-situ hybridisation, of the slides. A slim reaction chamber (35) can receive a slide holder for treatment using a minimum of reagent. A treatment apparatus receives a number of such chambers for heating and cooling.



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Microscope Slide Holder

This invention relates to a microscope slide holder, particularly a slide holder suitable for use in in-situ nucleic acid hybridisation.

Nucleic acid hybridisation is generally carried out with the target nucleic acid to be analysed immobilised in a solid support, particularly nylon membrane. In-situ hybridisation is a more sophisticated technique that enables the visualisation and the localisation of specific target DNA or RNA molecules in tissue sections, single cells or chromosome preparations. In this case, the support needs to be in the form of a microscope slide which can be examined in a conventional microscope. The success of any hybridisation process depends crucially upon control of temperature and time in the hybridisation cycle. Up to now, in-situ hybridisation has only been carried out using slides handled manually and individually between the various processes e.g. hybridisation, thermal cycling and slide washing. It has been very difficult to perform these processes accurately and reproducibly.

One key to achieving efficient in-situ hybridisation is the use of a slide holder which enables treatment of a plurality of slides at one time. Accordingly, the invention provides a microscope slide holder comprising a generally rectangular planar frame for receiving a plurality of microscope slides, having retaining means on the frame for individually retaining said slides in edge-to-edge relationship so that they cover space substantially in the plane of the frame.

Preferably, the retaining means comprise individual clips in which the slides are a sloppy fit and which permit movement of the slides perpendicular to the plane of the frame.

According to a further feature of the invention, there is provided a liquid reagent container for receiving said slide holder, which has a size closely matching that of a frame and into which said frame can be inserted through an open edge of the container.

Further, there is provided a processing unit having temperature control means and a plurality of slots for receiving a corresponding plurality of reagent containers as mentioned above.

Using such apparatus, 20 or more microscope slides can be put through identical treatment procedures, including temperature cycling, washing in water or other reagent when inserted in a reagent container, or heating in a suitable oven.

In order that the invention shall be clearly understood, one or more exemplary embodiments will now be described with reference to the accompanying drawings, in which:

Fig. 1 shows a microscope slide holder in plan view;

Fig. 2 shows a perspective view of the holder;

Fig. 3 shows three different forms of slide clip;

Fig. 4 shows a perspective view of a different slide holder;

Fig. 5 shows two views of a further form of clip;

Figs. 6 and 7 show different forms of clip employing bent wire;

Fig. 8 shows a processing unit; and

Fig. 9 shows liquid reagent containers for the slide holders.

In Fig. 1, two identical rectangular planar frames 10 are joined side-by-side. Each frame is dimensioned so as to receive ten standard microscope slides disposed in edge-to-edge relationship so that they cover substantially the whole space within the frame. A part of the frame is left empty to provide a form of handle 11 at the top. Each microscope slide 12 is supported at its ends by retaining means in the form of clips 13. The clips are designed so that the frame 10 can be laid horizontally or positioned vertically without the slides 12 falling out.

The same slide frame is shown in Fig. 2 in perspective view.

The precise nature of the clips 13 is illustrated in Fig. 3 which shows three different versions. The clip on the left has two projections 14 which are a press fit in holes 15 provided in the long sides 16 of the frames. In the middle and right-hand versions the clips engage the frames by virtue of channels 17 along their sides which are a snap fit onto the sides of the frames. In all three cases, the microscope slides 12 are shown to be a loose fit in the clips, such that if the frame is tilted slightly out of the vertical, the slides can tilt forwards (as illustrated) so that they are no longer coplanar but rather positioned in a series of parallel spaced-apart planes. In this position, it can be seen that each individual slide can be removed between two fingers of one hand. Conversely, the slides are easily placed in the clips by presenting them in a slightly tilted position and dropping them past the centre support 18 in each case. It can also be seen that if the frame is tilted backwards in the picture all the slides come to rest against the backs 20 of the clips and are in no danger of falling out if the frame is

positioned horizontally.

Fig. 4 shows a modified form of frame in which the individual clips 13 are formed as projections on a continuous moulded strip 21. Similar geometrical considerations apply as in Fig. 3.

Fig. 5 shows in greater detail a possible form of clip seen in perspective from both sides. For positioning the clips on the frame, two projections 24 are provided on one face, with a centre projection 25 having an enlarged sprung head, all of which can engage releasably in holes in the side of the frame 16. At the side of the clip which receives the microscope slide, a back surface 26 extends between L-shaped end retainers 27. A centre support 28 allows a slide inserted in the clip to tilt forwards without falling out.

Figs. 6 and 7 illustrate clips which are formed as a series of bent sections of a wire 30 extending the length of the frame. In Fig. 7, the clips only hold the microscope slides at one end and one of the slides 31 is shown in an angled position as it is inserted or extracted. In order to achieve this, the clip portion is swung in torsion around the vertical axis of the continuous wire 30.

Fig. 8 shows a processing unit having five slots 33 in its top surface. Each slot is designed to receive a complete microscope slide holder for treatment. Each holder is preferably first inserted in an envelope-like container 35, as illustrated in Fig. 9. Certain of the slots 33, for example numbers 1 and 2, may be provided with temperature control means such as a water bath or sources of radiant heat; others of the slots may accept slide holders and/or reagent containers for maintenance

at ambient temperature.

Fig. 9 shows a number of liquid reagent containers 35, one containing a microscope slide holder as previously described. The handle portion 11 of the frame projects at the top of the container. The container 35 is a relatively tight fit around the slide holder and the holder is inserted through one open edge 36 of the container. It can be seen that this arrangement minimises the quantity of liquid reagent which has to be placed in the container in order to treat the microscope slides. Moreover, since the liquid quantity is small, it can rapidly and accurately be heated or cooled. Each container has a widened top 37 to simplify filling and emptying.

The microscope slide holder described is advantageous in that a plurality of slides can be uniformly treated; each slide can easily be inserted and removed from the holder; and the holder can be positioned vertically to be placed in a reagent container or positioned horizontally for placing on a heater plate for temperature treatment. In particular, when placed horizontally, the slides still have a freedom within the clips to move perpendicular to their own planes. This enables a holder full of slides to be placed on a horizontal heated surface which is slightly smaller than the internal dimensions of the frame and the clips. When this is done, the slides necessarily come to lie flat in contact with the heated plate, while the frame can drop slightly and indeed comes to exert a slight downward pressure on all the slides. By contrast, if all the slides are rigidly held in the frame, any slide not exactly coplanar with the majority would not lie in face-to-face contact with the heater plate.

The apparatus described above is particularly adapted to the process of in-situ hybridisation and provides a means of carrying out all the processes necessary in uniform fashion for a relatively large number of samples.

CLAIMS

1. A microscope slide holder comprising a generally rectangular planar frame (10) for receiving a plurality of microscope slides (12), having retaining means (13) on the frame for individually retaining said slides (12) in edge-to-edge relationship so that they cover space substantially in the plane of the frame.

2. A microscope slide holder as claimed in claim 1 wherein said retaining means (13) are arranged along at least one side (16) of the frame.

3. A microscope slide holder as claimed in claim 1 or 2 wherein the retaining means (13) are formed as a series of inward projections (18, 20, 26, 27, 28) on the frame (10).

4. A microscope slide holder as claimed in claim 3 wherein the retaining means for one or more slide(s) is a discrete unit (13, 21) having said projections, which unit is clipped (14, 15, 17) onto said frame.

5. A microscope slide holder as claimed in claim 1 or 2 wherein the retaining means are formed as clips bent at intervals in a continuous metal member (30) which extends parallel along a side of the frame.

6. A microscope slide holder as claimed in any preceding claim which includes a handling portion (11) at one end of the frame which is not provided with retaining means for slides.

7. A microscope slide holder as claimed in any preceding claim wherein the retaining means (13) retains each slide without gripping it.

8. A microscope slide holder as claimed in any preceding claim wherein the holder retains the sides (12) when the frame is disposed in both a generally vertical and a generally horizontal plane.

9. A microscope slide holder as claimed in claim 7 wherein slides can be tilted about an edge to insert or remove them in or from the retaining means.

10. A reaction apparatus comprising a microscope slide holder as claimed in any preceding claim, and a chamber (35) for receiving liquid reagent which chamber has internal dimensions very slightly larger than the external dimensions of the slide holder.

11. A reaction apparatus as claimed in claim 10 wherein the slide holder is fully accommodated within the chamber with only a handling portion (11) accessible outside it.

12. A reaction apparatus as claimed in claim 10 or 11 wherein the chamber is open along one edge (36).

13. A reaction apparatus as claimed in claim 12 wherein the said open edge (36) is splayed to form a pouring lip (37).

14. A treatment apparatus comprising a housing and heating and cooling means with control means on or in the housing, wherein the housing has one or more top apertures (33) which receive one or more reaction apparatuses as claimed in any of claims 10 to 13.

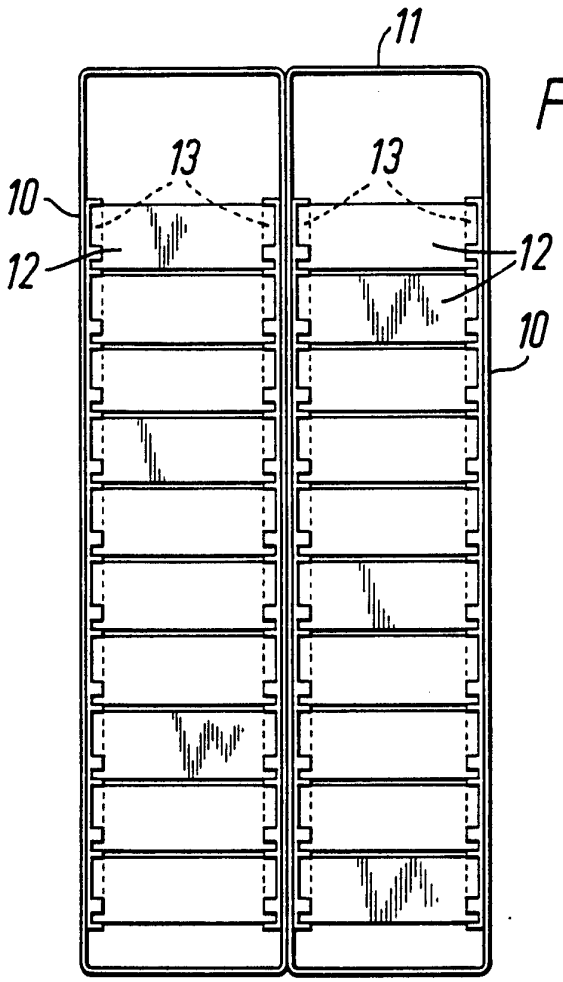


Fig. 1

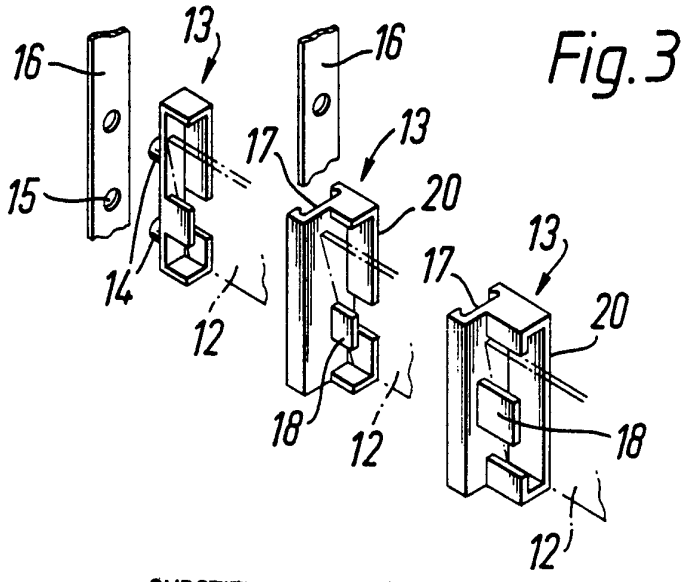


Fig. 3

Fig. 2

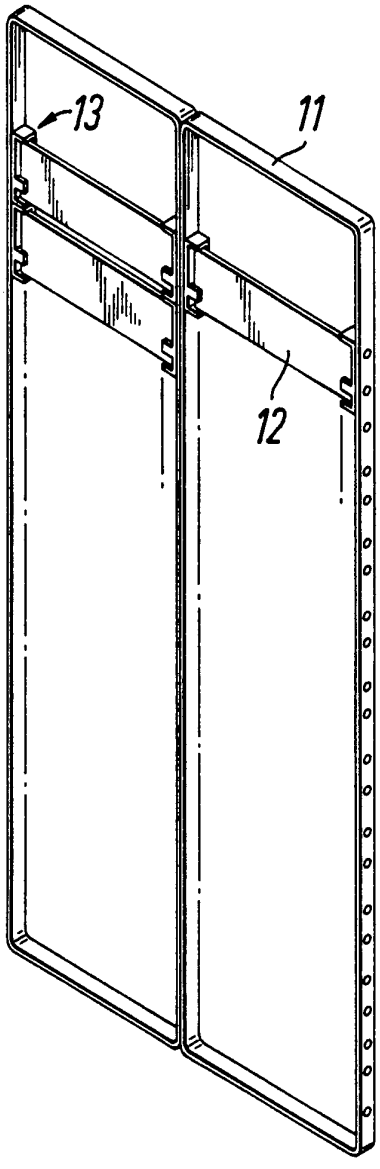


Fig. 4

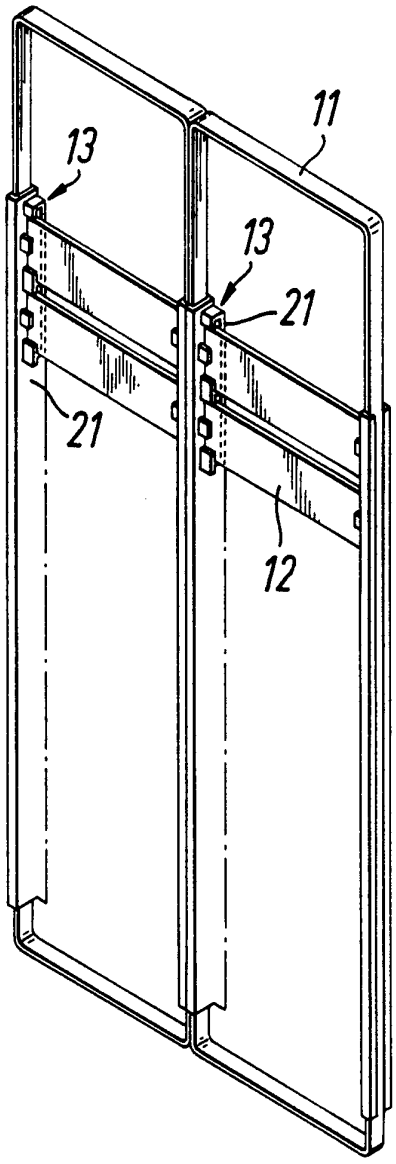


Fig.6

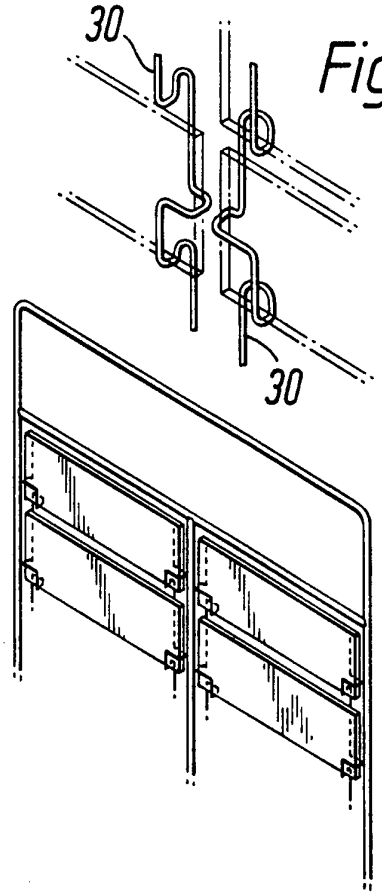


Fig.7

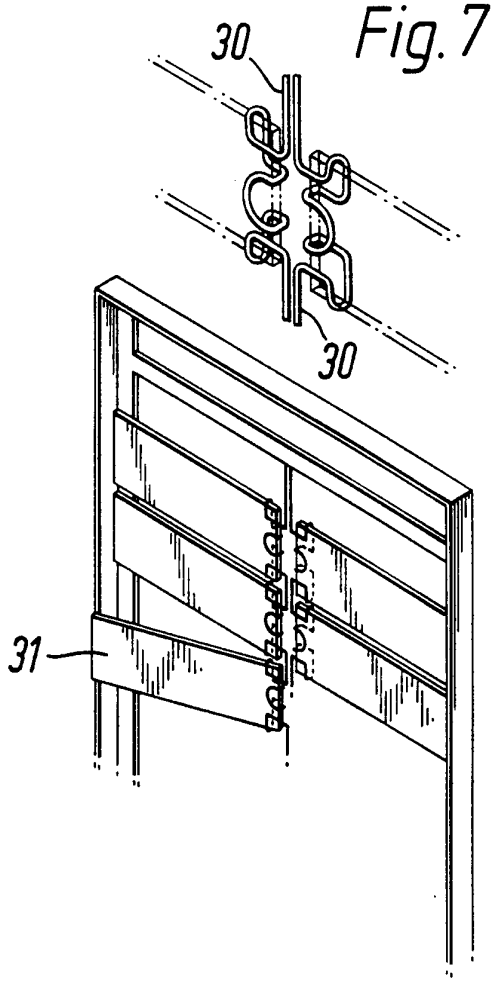


Fig.5

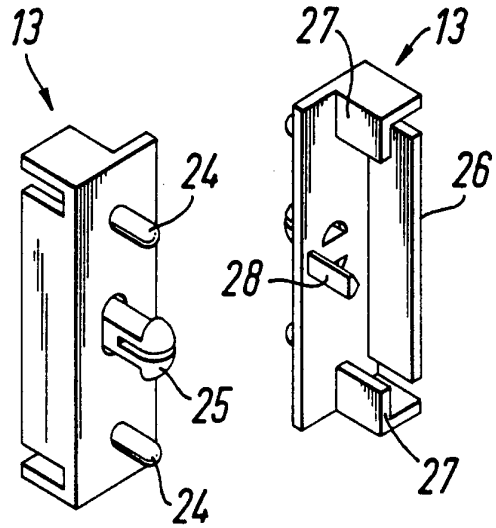


Fig.8

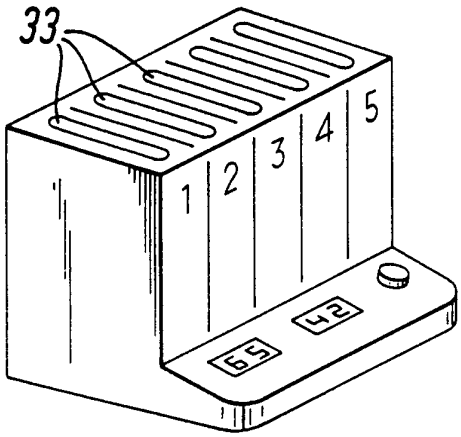
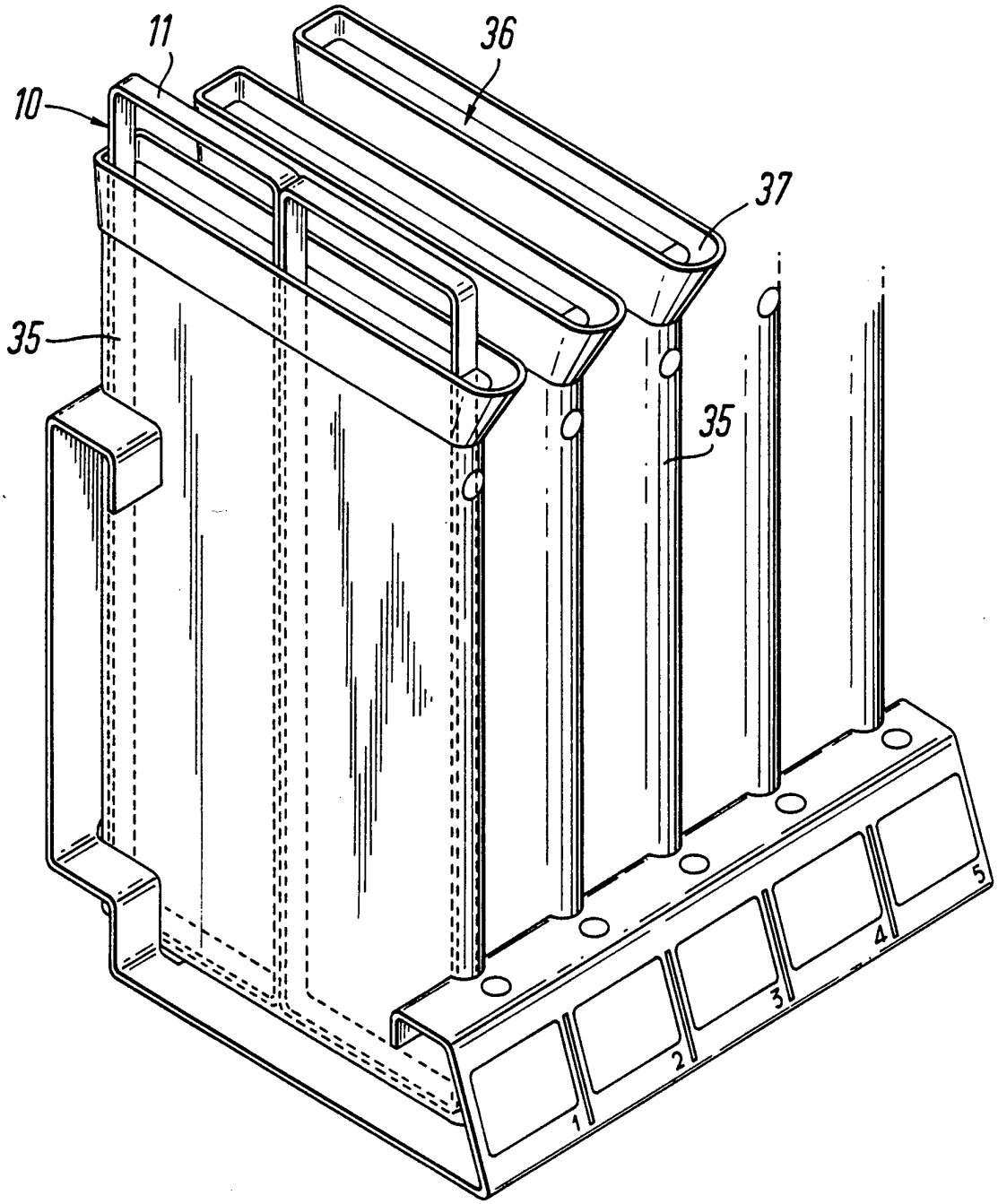


Fig. 9



INTERNATIONAL SEARCH REPORT

Intern ial Application No
PCT/GB 94/00671

A. CLASSIFICATION OF SUBJECT MATTER
IPC 5 G02B21/34 G01N1/30

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 5 G02B G01N

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C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
Y	FR,A,2 508 184 (SMITHKLINE BECKMAN) 24 December 1982 see page 8, line 22 - page 10, line 30; figures 4,5,12	1-3,6-8, 10-14
Y	US,A,3 667 088 (F.D.KURTZ) 6 June 1972 see the whole document	1,2,8
Y	EP,A,0 415 400 (G.MENZEL GLASBEARBEITUNGSWERK) 6 March 1991 see column 3 - column 6; figures 1-18	1-3,7,8
Y	US,A,2 864 105 (J.ROSE) 16 December 1958 see the whole document	6
Y	GB,A,219 119 (F.W.JACKSON) 9 November 1983 see page 4; figures 5-16	10-13

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Y	US,A,3 132 744 (B.C.SCHARF) 12 May 1964 see column 3; figures 1-4	10-13
Y	WO,A,92 21953 (MEDITE GESELLSCHAFT FÜR MEDIZINTECHNIK MBH) 10 December 1992 see abstract; claim 42; figures 1-12	14

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Patent document cited in search report	Publication date	Patent family member(s)		Publication date
FR-A-2508184	24-12-82	US-A-	4453807	12-06-84
		CA-A-	1179534	18-12-84
		DE-A,C	3222594	05-01-83
		GB-A,B	2107664	05-05-83
		JP-A-	58002817	08-01-83
		US-A-	4501495	26-02-85
US-A-3667088	06-06-72	NONE		
EP-A-0415400	06-03-91	US-A-	5044500	03-09-91
		CA-A-	2023604	02-03-91
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WO-A-9221953	10-12-92	DE-A-	4117833	03-12-92
		EP-A-	0586494	16-03-94