March 14, 1972 H. SCHARF ET AL 3,649,465

SPINNER FLASK

Filed June 24, 1969 2 Sheets-Sheet 1

FIG. 1

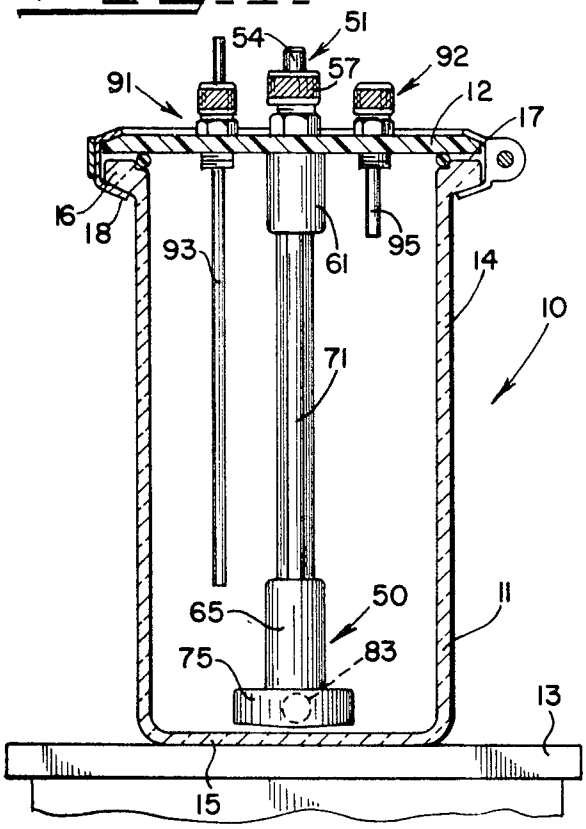


FIG. 5

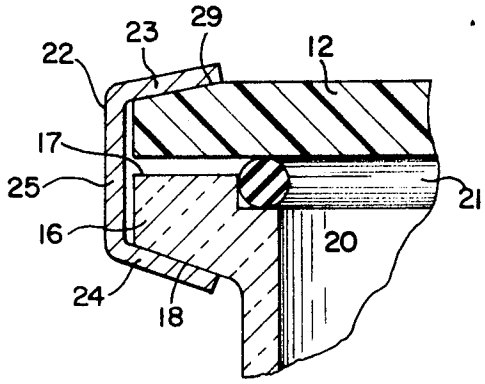
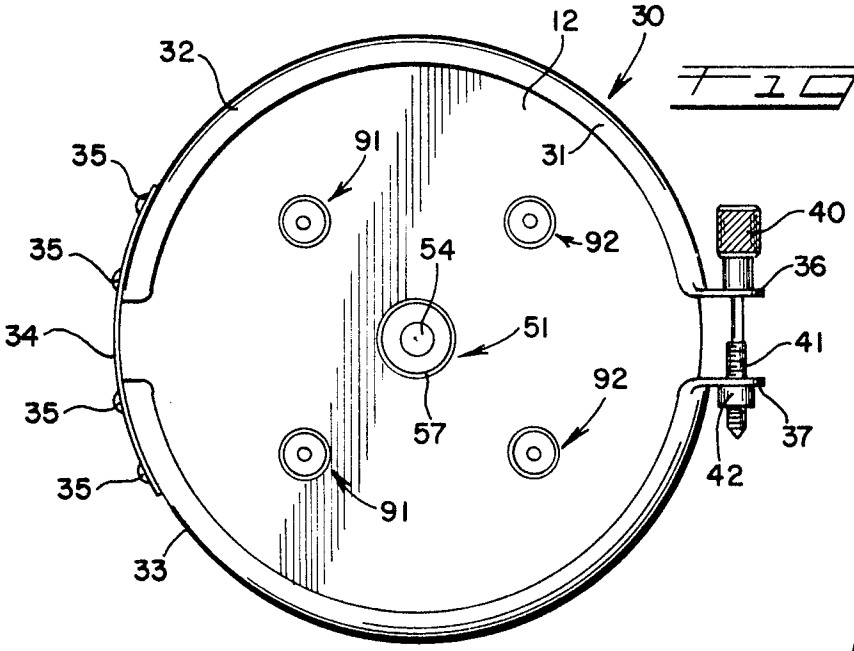


FIG. 3



INVENTORS
HAROLD SCHARF
CHARLES E. BENDER
DOUGLAS S. FRASER

BY Robert E. Wagner ATT'Y.

March 14, 1972 H. SCHARF ET AL 3,649,465

SPINNER FLASK

Filed June 24, 1969 2 Sheets-Sheet 2

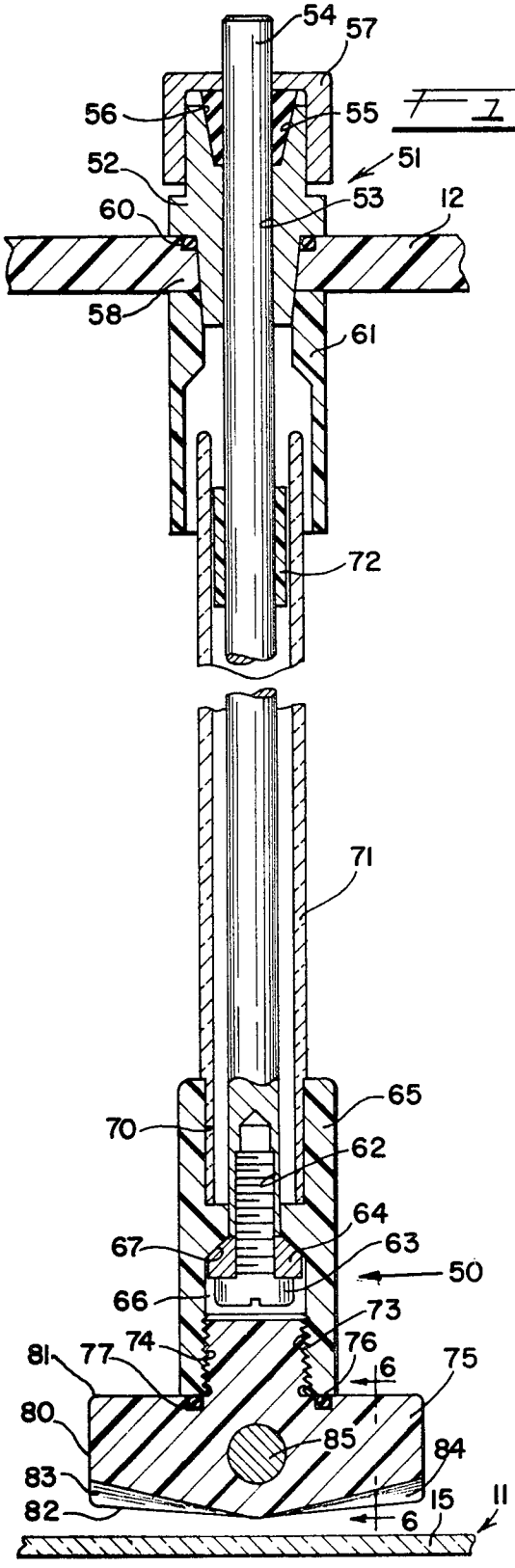


FIG-2-

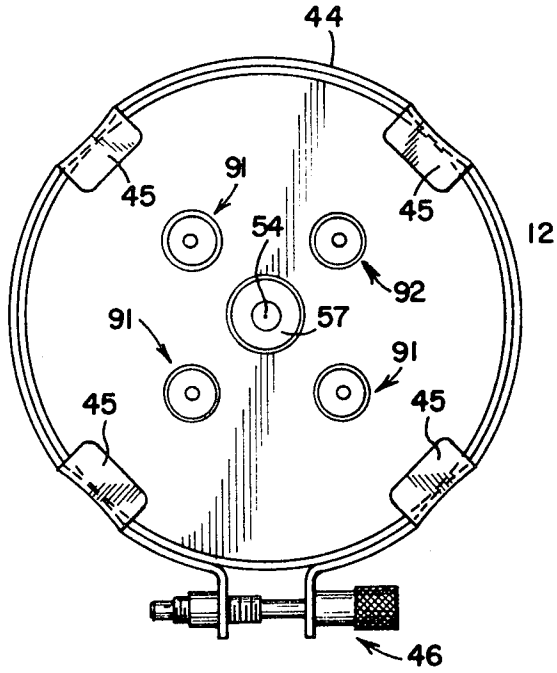


FIG-4-

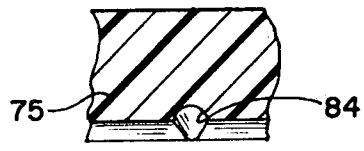


FIG-6-

INVENTORS
HAROLD SCHARF
CHARLES E. BENDER
DOUGLAS S. FRASER

BY Robert E. Wagner ATT'Y.

United States Patent Office 3,649,465
Patented Mar. 14, 1972

3,649,465

SPINNER FLASK

Harold Scharf, Charles E. Bender, and Douglas S. Fraser, New Palz, N.Y., assignors to The Virtis Company, Inc., Gardiner, N.Y.

Filed June 24, 1969, Ser. No. 836,091
Int. Cl. C12b 1/00
U.S. Cl. 195—143 6 Claims

ABSTRACT OF THE DISCLOSURE

A spinner flask arrangement for tissue and virus culture having a vessel closed by a cover with sealing means interposed. A novel clamp holds the cover assembled to the vessel and biases the sealing means for fluid-tight integrity. The cover supports a magnetically driven cylindrical agitator having a novel mounting to prevent cell grinding and bearing residue from contaminating the culture. The agitator shape prevents cell damage from impacts and assures good mixing.

This invention relates to improvements in spinner flask arrangements and, more specifically, is directed to a new and improved agitator and vessel which may be used for tissue and virus culture work, biological and chemical manipulations and like techniques wherein a liquid or semi-liquid substance is required to be agitated under controlled circumstances.

Tissue culture work has increased in recent years because of the increased need for inquiry into the behavior of cells, isolated bits of tissue, cellular phenomena in living organisms and the like. The increased activity no doubt is brought about by the biologists' and medical researchers' continued interest in cancer research, viruses and the like. In such studies and related tissue culture techniques, it is desirable to provide an environment for the culture study which includes controlled agitation under controlled or isolated circumstances for extended durations. During such studies, it is absolutely essential that the flask or container in which the tissue culture medium is located be maintained completely sterile and free from any contact with foreign or unsterile equipment. In some studies, some engineering variables such as temperature and pressure need to be carefully controlled. Also, because of the rather fragile nature of some cells, it is desirable that agitation be accomplished in such a fashion to eliminate cell damage from grinding or impacting through action of the agitating means.

Due to the need for complete isolation during culture studies, magnetic drives for agitators have been used in the past with a degree of success. Problems have been encountered, however, in supporting the magnetically driven agitators to eliminate the unwanted cell grinding and cell impacting while providing a suitable drive arrangement which will allow the most positive type of magnetic coupling in a fashion which would assure the contents of the vessel or flask to be free of possible contamination. A further problem has existed in maintaining a tight seal at the juncture of the cover and flask without using sealing substances such as silicon grease or the like which could have a deleterious effect on the culture medium undergoing study and, in some instances, contaminated it to a point that would render it useless, oftentimes, occurring after weeks of preparation and agitation.

The present invention provides a simplified solution to the problems outlined above in the form of an improved spinner flask arrangement for tissue and virus culture study. The vessel in the arrangement is of conventional shape, being of generally cylindrical design with a flat bottom wall and a flanged upper margin and being fairly easy to mold and, therefore, less costly than special types of flasks for culture work presently known in the prior art. A cover of generally disc-like shape is provided and has a chamfer or frusto-conical surface at its peripheral margin. The flask in the region of the flanged upper margin is formed with an axially facing groove which receives an O-ring and cooperates with the cover to form a vacuum-tight seal, eliminating the need for silicon greases and the like.

A clamp arrangement is uniquely designed to exert clamping pressure along the frusto-conical surfaces to drive the cover into fluid-tight engagement with the sealing means and flask and, therefore, provide the necessary fluid-tight integrity. Any number of ports may be formed in the cover and closed with non-toxic stoppers or compression-type fittings which provide a measure of flexibility in the studies and permit the controlled addition of nutrients, removal of samples, monitoring of the temperature and pressure through probes or the like under the controlled conditions required.

In the center of the cover is provided a novel holder which supports the spindle supporting the agitating means. This type of holder permits free vertical adjustment of the spindle to provide the most positive magnetic coupling under the existing conditions. A novel bearing support arrangement is also provided having a protective shroud which assures that any bearing residue developed during agitation will be positively prevented from entering the media undergoing culture study. The agitator is located centrally of the flask and is of generally cylindrical shape having an end portion formed to a conical shape of reduced height. Two grooves are formed along the surface of the cone which are shallow and generally arcuate shaped when viewed in cross section. This provides proper agitation and prevents the congregation of cells in any one location, resulting in a smooth mixing pattern of generally spiral configuration over a wide range of speeds. The agitator is made in two parts including an agitator head and a hub portion, permitting separation of the two and assuring ease of assembly to a support spindle. The head is of cylindrical shape yet contains an elongated magnet which is cast in the head with the axis thereof positioned along the diameter of the cylinder close to the frusto-conical portion of the head. The agitator head may be formed of an inert substance such as a polymerized tetrafluoroethylene such as the plastic sold under the trademark "Teflon" by the E. I. du Pont Company, Wilmington, Del. Plastics of this type have a built-in natural lubricity which assures free running of any engaging parts. Further advantages of the spinner flask arrangement of the present invention will become apparent upon consideration of the objects achieved and a detailed description of a preferred embodiment.

It is an object of this invention to provide a new and improved spinner flask arrangement.

It is a further object of this invention to provide a new and improved spinner flask arrangement having a novel mounting for the agitating means.

It is a further object of this invention to provide a spinner flask arrangement for use in tissue culture studies which will provide positive protection against the entry of contaminants into the culture media undergoing study.

It is a further object of this invention to provide a spinnner flask arrangement having a uniquely shaped and mounted agitating means to provide a good agitation pattern and prevent bearing residue contamination of the culture study.

It is a still further object of this invention to provide a new and improved spinner flask arrangement for use in tissue culture studies and the like wherein the cover is fluid-tightly mounted on the vessel without the need

for vacuum greases and the like and having a unique clamping arrangement for ease of assembly of the cover to the vessel.

Objects in addition to those set forth above will become apparent upon reference to the accompanying drawings wherein:

FIG. 1 is a cross-sectional view of a spinner flask arrangement in which the vessel and cover are illustrated in cross section and the agitating means is a full elevation with a magnetic stirrer base shown fragmentarily;

FIG. 2 is an enlarged cross-sectional view of the agitating means and supporting arrangement with the cover shown fragmentarily in cross section;

FIG. 3 is an enlarged top plan view of the flask shown in FIG. 1 illustrating one form of clamping arrangement;

FIG. 4 is a view similar to FIG. 3, however, on a reduced scale, illustrating a modified form of clamping arrangement;

FIG. 5 is an enlarged cross-sectional view of the cover, flask, clamp and seal; and

FIG. 6 is a fragmentary cross-sectional view taken along the line 6—6 of FIG. 2.

Referring now to FIG. 1, reference numeral **10** indicates a spinner flask arrangement including a vessel or flask **11** having a disc-like cover **12**. The flask **11** is positioned on the bed of a magnetic stirrer indicated generally at **13**, this being of known type and having a conventional variable speed control motor driving a conventional variable speed control motor driving a rotating magnet for magnetically coupling and driving magnetic stirrers.

The vessel **11** includes cylindrical side walls **14** integral with a generally flat bottom wall **15**. An enlarged flange **16** having a generally flat top **17** and frusto-conical underside **18** is formed at the top of the side fall **14**, as is best seen in FIG. 5. The upper margin or flat top **17** is formed with a shoulder or rabbit groove **20** around its inner periphery to receive and support a nannular O-ring **21** of resilient material. A circumferential clamp **22** has upper and lower flange portions **23** and **24** which project from the main body **25** of the clamp over the cover **12** and under the frusto-conical wall **18** of the flange **16**, functioning to hold the cover **12** on the vessel **11** in a manner to exert axial force to compress the annular seal **21**. In FIG. 5, the clamp is in a semi-relaxed condition.

Two forms of clamping arrangements are shown in FIGS. 3 and 4. In FIG. 4, the clamp **30** is provided with a continuous flange **31** along the top of the cover **12**. This corresponds to the flange **23** shown in the cross-sectional view of FIG. 5. A continuous flange of similar shape is formed on the underside of the clamp. The clamp **30** is formed in two separate sections **32** and **33** which are joined together through a spring steel section **34** which is fastened to each section by rivets **35** or the equivalent. The opposite or free ends of sections **32** and **33** are flat and are bent to form flanges **36** and **37** which are generally parallel to each other when the clamp is in the clamped position. A knurled knob **40** on the end of a threaded shaft **41** cooperates with a threaded boss **42** joined to the flange **37** to constrict the clamp circumferentially. As seen in FIG. 5, the cover **12** is provided with a chamfer or frusto-conical surface at its outer periphery to develop an axial component of force during clamp constriction to draw the cover tightly onto the top **17** of the flange **16** of the vessel **11**. Reversal of rotation releases the clamp sections for lateral expansion and removal.

In the modified form of clamp shown in FIG. 4, the clamp body **44** is continuous, having spaced flanges with frusto-conical surfaces above and below as indicated at **45**. These flanges cooperate with the cover **12** in the fashion mentioned above with an adjusting knob arrangement **46** functioning in the same manner as that described with respect to FIG. 3. In each of the clamping arrangements of FIGS. 3 and 4, the clamp is laterally expanded to permit removal of the cover **12**. Expansion is possible after the adjusting knob is rotated to the released position.

As seen in FIG. 2, an agitating means, indicated generally at **50**, is supported from the cover **12** by a spindle support or holder arrangement indicated generally at **51**. The spindle holder consists of a threaded body **52** having a central passage **53** adapted to receive an agitator supporting spindle **54**. The spindle **54** may be formed of stainless steel, glass, plastic or the like.

A frusto-conical bore **55** is formed at the upper end of the holder body **52** and receives a resilient washer or ferrule **56** of similar shape. A gland or jam nut **57** forces the ferrule or washer **56** into the bore, constricting it tightly about the spindle **54** to mechanically lock it against axial movement and simultaneously form a fluid-tight seal. The lower end of the holder body **52** is externally threaded as at **58** and sealed to the cover **12** through an O-ring **60**. The threads **58** project well beneath the cover and receive a complementarily threaded shroud **61** which projects downwardly into the vessel **11** for reasons to become apparent.

The spindle **54** is internally threaded as at **62** at its lower end to receive a cap screw **63** which functions to join a bearing **64** formed of glass or equivalent material at the lower end of the spindle. An agitator hub **65** has a central bore **66** which includes a frusto-conical shoulder **67** in opposed engagement with a similarly shaped surface on the bearing **64**. The upper end of the bore is counterbored as at **70** and receives the lower end of a shroud **71** with a press fit. The upper end of the shroud **71** projects upwardly in circumferentially spaced relation to the spindle **54**. The upper end of the shroud **71** is loosely received within the shroud **61** on the cover to form a foam baffle. A plastic bearing **72** is carried on the spindle **54** at its upper end and has its outer circumferential surface in bearing engagement with the inner surface of the shroud **71**. In this manner, stability of the agitating means **50** is assured.

At the lower end of the agitator hub **65**, the bore **66** is enlarged and internally threaded as at **73** to cooperate with external threads **74** formed on the agitator head **75**. The agitator head **75** may be provided with an annular groove **76** which receives a seal such as an O-ring **77** or the like to seal the hub to the head **75** isolating the bearing from the culture media. The agitator head **75** is of generally cylindrical shape, having a smooth circumference **80**, radial end wall **81** at the top and conical end wall **82** in closely spaced relation with the bottom wall **15** of the vessel **11**. An Alnico or other type of magnet **85** having good magnetic strength is imbedded within the agitator head **75** and functions to magnetically couple the agitator head to the drive arrangement shown at **13** in FIG. 1. The conical end **82** of the agitator head **75** is formed with grooves **83** and **84** of semi-circular cross section and diminishing cross section as shown in FIG. 6. These shallow grooves are sufficient to cause good agitation of the culture media in a generally upward pattern and in a gentle manner to eliminate the creation of a vortex or cell damage by impacting. The culture is agitated with substantial uniformity and congregation of the cells in one area is eliminated. Cell grinding is likewise eleiminated because of the novel support of the agitator head **75**.

Referring once again to FIGS. 1, 3 and 4, the cover **12** is provided with additional openings which receive compression-type fittings **91** and **92** which are generally similar to that described through the compression fitting **51**. Four such fittings are shown in FIGS. 3 and 4, however, any number within the practical limitations of the vessel size may be included, being easily made since the cover is formed of polycarbonate. Such fittings facilitate the introduction of a probe such as that shown at **93** in FIG. 1 to permit temperature, pressure or other engineering variables to be measured without interruption of the sealed conditions. Sampling and insertion of nutrients

may be likewise accomplished through the other ports via a tube 95 extending through the flttings shown in FIG. 1.

While the present invention has been described in connection with spinner flask arrangements, it is obvious that the unique principles may be applied to vessels of various shapes as well as in conjunction with culture studies employing filters and the like.

With the present invention, cell grinding and impacting is completely eliminated. The cover and vessel are sealed by a simply designed, easily operable stainless steel clamp and sealing arrangement without the necessity of using sealing grease or other sealants which might cause contamination during the cell studies. The multi-ported top eliminates the need for multi-ported vessels while providing a measure of versatility in the application. With the foam baffle formed at the top of the shroud and the complete protection of the bearing surfaces, bearing residue is trapped in the agitator head, further preventing contamination. In addition, through the unique mounting arrangement, the drive magnet may be positioned relatively close to the driving magnet in the magnetic stirrer for a good positive coupling to permit driving at a wide range of speeds. The novelly shaped head also assures excellent agitation patterns. When made in the preferred form, the entire spinner flask arrangement is completely autoclaveable for ease of sterilization between experiments.

Upon a consideration of the foregoing, it will become obvious to those skilled in the art that various modifications may be made without departing from the invention embodied herein.

We claim:

1. In a spinner flask arrangement wherein a vessel is provided with a bottom wall and upstanding side walls, said vessel having an open end which is enclosed by a cover having outwardly facing frusto-conical shoulder surfaces to form an isolated volume, the improvement comprising, spindle support means having agitating means disposed for rotation at one end thereof and having its opposite end extending through said cover, means attaching said support means to said cover, clamping means exerting pressure on said outwardly facing frusto-conical shoulder surfaces of said cover and said side walls of said vessel, said spindle support means being surrounded along its lower portion thereof by support shroud means, said support shroud means having said agitating means attached thereto, said support shroud means being rotatable about said spindle support means, said spindle support means being surrounded along its upper portion thereof by a foam shroud means, said foam shroud means being maintained in fixed relation to said spindle support means and extending over and surrounding the upper end of said shroud means during agitation, said agitating means being suspended above said bottom wall of said vessel a sufficient distance to preclude cell grinding therebetween during agitation.

2. The improvement in spinner flask arragements as defined in claim 1 wherein said agitating means includes an agitator head of generally smooth cylindrical configuration having a generally elongated magnet imbedded therewithin, and having a downwardly conical shaped end, said conical shaped end having at least one groove formed therein extending from adjacent the apex of said cone to the base of said cone, said groove being semi circular in cross section and being effective to agitate the culture media contained within said spinner flask in a gentle, generally upward pattern, said smooth cylindrical configuration of said agitator head preventing cell impacting during rotation of said agitating means.

3. The improvement in spinner flask arrangements as defined in claim 1 wherein said spindle support means includes a spindle fixedly mounted in said cover and extending downwardly therefrom and substantially perpendicular thereto, said spindle having said agitating means disposed at its lower end and maintaining said agitating means above said bottom wall of said vessel, said support shroud means substantially enshrouding and surrounding said spindle along its length to a point adjacent said cover to prevent bearing residue produced during rotation of said agitating means from escaping into the contents of said vessel, said support shroud means being attached to said agitating means at its lower end and being rotatable with said agitating means about said spindle.

4. The improvement in spinner flask arrangements as defined in claim 1 wherein said agitating means includes a hub portion and said agitator head, said hub portion having a central bore therethrough for reception of said spindle, said central bore having a frusto-conical shoulder formed therein, a bearing supported on said spindle, said bearing having a frusto-conical bearing portion in supporting engagement with said frusto-conical shoulder of said hub, and means for fluid tightly adjoining said agitator head and hub for unitary rotation about the central axis of said spindle.

5. The improvement in spinner flask arrangements defined in claim 1 wherein said spinner flask includes axially spaced grooves disposed about the upper margin of the upstanding side walls of said vessel and resilient sealing means positioned in said grooves, said cover having an outwardly axially facing frusto-conical shoulder engaging said sealing means, and, clamping means having holding means disposed about the periphery of said cover and said upper margin of said upstanding walls of said vessel, said holding means engaging said cover and said vessel at the junction thereof at at least one point on the periphery of said cover, said holding means terminating in adjusting means movable to increase or decrease the diameter of said holding means engaging said frusto-conical portion of said cover and said upper margin of said upstanding walls of said vessel, a decrease in the diameter of said holding means exerting pressure on said cover to maintain said cover and said vessel in sealed engagement.

6. The improvement in spinner flask arrangements as defined in claim 5 wherein hinge means is formed on said clamping means to allow expansion of said holding means and lateral removal of said holding means and adjusting means from engagement with said cover and said vessel.

References Cited

UNITED STATES PATENTS

2,793,166	5/1957	Hatch	195—143
2,958,517	11/1960	Harker et al.	195—143
3,172,235	3/1965	Bjorklund	195—143
3,251,635	5/1966	Phillips	259—122
3,445,342	5/1969	Freedman	195—143

ALVIN E. TANENHOLTZ, Primary Examiner

U.S. Cl. X.R.

259—122