Midi Parasep

Faecal Parasite Concentrator

For *in vitro* use only

INTENDED USE

Midi Parasep is a closed system for a clean and efficient concentration of intestinal parasites from human faecal probes. The simple 4 step kit provides a fast and simple method to concentrate helminth ova as well as protozoan cysts/oocysts.

SUMMARY AND EXPLANATION

The microscopic examination of stool specimen enables a diagnosis of intestinal parasitic infection. Faecal concentration has become a routine procedure as it allows the detection of small numbers of organisms that may be missed using other methods. Sedimentation is designed to separate protozoan organisms and helminth eggs and larvae from faecal debris by centrifugation.

PRINCIPLE OF THE TEST

The faecal sample is taken with the spoon on the filter and mixed into the tube with the solutions. After a short mixing and centrifugation step, Midi Parasep is reopened and the sediment is ready for microscopy.
It is a single use, disposable device offering significant time saving as well as prevention against cross- contamination. The unique tangential hexagonal filter provides a fast and reliable filtration of the sample, leading to a clean background.

REAGENTS

Midi Parasep is available in 2 different kits providing all material and solutions needed for 40 tests: Midi Parasep Formalin and Midi Parasep SAF
All materials are supplied ready to use.

Composition:

Midi Parasep consists of 2 parts: the solution tube (mixing chamber) and the filter with attached conical tube. In each kit, 2 reagents are provided:

Already filled in the 40 mixing chambers (6.0 mL). In Midi Parasep Formalin, it is a 10% formalin solution with Triton X.

One vial containing 80 mL of Ethyl Acetate for emulsification of the sample.

STABILITY AND STORAGE

The kits are stable until the expiry date stated on the packing.

The liquids must be discarded according to the correct protocol.

Parasep Products:

Midi Parasep, kit with empty components, 50 tests.....909000
Midi Parasep Formalin (40 tests, kit with all necessary solutions, with spatula).....905000
Ethyl Acetate, 250 ml (Reorder only).....1473

PRECAUTIONS

For professional use only. For *in vitro* diagnostic use only

1. **Formalin**:
   H302 Harmful if swallowed
   H317 May cause an allergic skin reaction
   H332 Harmful if inhaled
   H341 Suspected of causing genetic defects
   H350 May cause cancer
   P280 Wear protective gloves/protective clothing/eye protection/face protection.
   P301 + P312 + P330 IF SWALLOWED: Call a POISON CENTER or doctor/physician if you feel unwell. Rinse mouth.
   P308 + P313 IF exposed or concerned: Get medical advice/attention.
   P333 + P313 If skin irritation or rash occurs: Get medical advice/attention.
2. Ethyl Acetate:
   H225 Highly flammable liquid and vapour.
   H319 Causes serious eye irritation.
   H336 May cause drowsiness or dizziness.
   P210 Keep away from heat/sparks/open flames/hot surfaces. No smoking.
   P261 Avoid breathing vapour/spray.
   P303+361+353 IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water/shower.
   P304+340 IF INHALED: Remove person to fresh air and keep comfortable for breathing.
   P305+351+338 IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
3. All patient samples should be treated as potentially infectious. User must wear protective gloves, eye protection and laboratory coats when performing the test.

SPECIMEN

Use fresh or preserved stool.

Stool specimens could be preserved in formalin for up to one year.

MATERIALS REQUIRED BUT NOT PROVIDED

Centrifuge with 50 mL bucket holder
Pipette for 2.0 mL
Microscope
Slides/coverslips

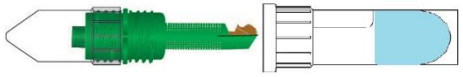
TEST PROTOCOL

Please adhere to the following guidelines when handling Midi Parasep. To avoid cross contamination, the Midi Parasep device should remain closed at all times except when introducing the sample or when retrieving the final concentrated sample for examination.

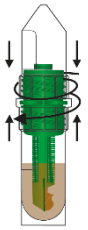
1. Sample Preparation

Unscrew lid. **Add 2.0 mL of Ethyl Acetate to the mixing chamber.** When using unfilled Midi Parasep, **prepare fixing solution and fill in 6.0 mL before adding Ethyl Acetate**. Alternatively, *Ethyl Acetate can be added after the sample through the filter thimble* (see 2).

2. Emulsification and Filtration



Introduce a pea sized (0.5 g) faecal sample into the mixing chamber using the spoon in the end of the Midi Parasep filter thimble. More is not necessary, but would lead to darker background.

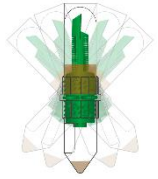


Seal Midi Parasep by screwing in the filter thimble into the sedimentation cone unit. Vortex to emulsify the sample with the sedimentation cone pointing upwards.

As alternative to 1), Ethyl Acetate can be added directly before centrifugation:

remove the upward pointing conus, add Ethyl Acetate through the filter thimble without opening the mixing chamber again. Reclose and mix.

3. Centrifugation

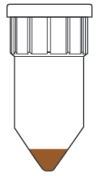


Invert Midi Parasep and centrifuge at 1,000 g for one minute or 500 g for five minutes. Midi Parasep fits all 50 mL centrifuge buckets. To calculate the required rpm for any centrifuge:

$$RPM = \sqrt{\frac{g}{1.12r}} \times 1.000$$

rpm: rotor speed in revs/min.
g: centrifugal force
(for Midi Parasep: 1.000 x g or 500 x g)
r: radius, horizontal distance between sedimentation cone tip and spindle centre, measured in mm.

4. Sediment recovery and examination



Open very slowly to avoid aerosol release. **Unscrew the conus, discard the rest (the filter thimble remains in the mixing chamber).** Loosen the fatty plug and **pour off the supernatant carefully. Transfer 1-2 drops of the sediment to a microscopic slide for examination.**

One drop of Lugol can be added to identify protozoans when necessary.

The sediment should be used within 10 minutes to avoid the resubstitution into the mixture. The lid of the mixing chamber fits the conus too, samples can therefore be stored if desirable.

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USEFUL HINTS

1. Sometimes, the addition of a few drops of NaCl solution to the sediment could be useful, especially when it is very dark. That would simplify microscopy.
2. To identify protozoan cysts and to improve the contrast of helminth eggs, it is useful to compare unstained and Lugol stained samples.
3. If desirable, Ethyl Acetate can be added after the introduction of the faecal sample. For this purpose, reopen the filled Midi Parasep (remove conus). Then, add the amount of Ethyl Acetate through the filter and closethe system again.
4. Helminths have variations in their egg laying cycles, means that although infected, there may be no parasite eggs found in the faeces. Therefore, it is useful, to examine 3 different stool samples from one patient on 3 consecutive days.

PERFORMANCE DATA

A comparative study was performed between the Paraprep system and the modified Ridley-Allen concentration method which is an open technique. One hundred faecal samples, both fresh and preserved, were examined in duplicate by both techniques. Theywere containing a wide range of ova, larvae, cysts and oocysts as follows:

- 26 faecal samples were contaminated with ova; 21 of which contained only 1 species of helminth and 5 contained 2 or 3 species of helminths.
- 24 faecal samples were protozoan cysts or oocysts positive; 15 of which contained only one species of protozoa and 9 contained 2 or more protozoa.
- 50 faecal samples were negative from ova, cysts or larvae. A comparable recovery of parasites was noted in both methods.

REFERENCES

1. Garcia, LS., Bruckner, DA., Diagnostic Medical Parasitology, Elsevier, N.Y. 1988
2. Perry, JL, *et al.*, *Parasite detection efficiencies of five stool concentration systems*, J Clin. Micro., 28:1094, 1990
3. Allen, AVH., Ridley, DS., J Clin Pathol.,1970 Sep; 23(6): 545-546.

CALCULATION OF NECESSARY RPM

The RPM of the used centrifuge can be obtained from the following table, simply by measuring the distance between spindle centre of the rotor and the sedimentation conus end of the Paraprep device. All data are for 1,000 x g, the numbers in the right column represent the rpm for the centrifuge used:

DISTANCE IN MM	RPM
140	2.500
150	2.400
160	2.400
170	2.300
180	2.200
190	2.200
200	2.100
210	2.100
220	2.000
230	2.000
240	1.900
250	1.900