

FECAL EGG COUNTING PROCEDURE – Janice Liotta

1. Using a tongue depressor, weigh out 2 to 4 gm of feces into beaker.
2. Break up the fecal pellets and add the correct amount of flotation solution to the feces to make a slurry. You'll need a total of 56 ml flotation solution for 4 g of feces, 42 mls for 3 g or 28 ml for 2 gm. **It is easiest to add just a little of your floatation solution to first break up the fecal pellets and then add the remainder of the solution.** For example, for 4 g of feces, you can add about 20 ml flotation solution to help break up the feces using the tongue depressor to break lumps. Then bring the slurry up to the 60 ml mark on your beaker using the remainder of your flotation solution. **. If you do not have a scale, keep in mind that a gram of feces is approximately 1 ml in volume. Thus, you can put 28 mls of solution into a beaker and then add enough feces to bring the liquid level to 30 mls, etc.**
3. Add a stir bar, and stir on a magnetic stirrer at medium speed for 5 min. **OR** put in a leakproof jar and shake vigorously for 5 minutes.
4. At the end of 5 minutes, while mixture is still stirring, draw about 1 ml fecal suspension from the upper layers of the slurry into your syringe.
5. Load one side of counting chamber carefully to avoid producing bubbles – each chamber holds about .15 ml of slurry and repeat sampling and loading procedure for second side of chamber.
6. Let preparation stand a minimum of 5 min (examine it at least by 20 min.)
7. Place chamber on microscope and examine with 10 X objective (Adjust the focus until you can see grid lines clearly and then refine your focus to the air bubble layer).
8. Count eggs in both sides of chamber- each chamber or grid has six sections. Do not count eggs outside the grid. Calculate the number of eggs per gram of feces: (side 1 + side 2) X 50

Flotation solutions:

1. Epsom salts (Magnesium sulfate) – dissolve 400 g (~ 1 2/3 cup) in 1 liter of water **OR**
2. Granulated sugar – dissolve 1 lb (454 g) in 355 ml of hot water (~1 ¼ cups of sugar per cup of water)
3. Saturated table salt – add salt to water until no longer going into solution, about 305 g of salt per liter of water. 350 g + 303 ml or 1.28 cup.

McMaster slide:

The egg counting chambers or “McMaster slide” can be ordered from Chalex Corporation, 5004-228th Ave SE, Issaquah, WA 98029, (1-425-391-1169, Fax 425-391-6669, email: chalexcorp@att.net, <http://WWW.VETSLIDES.COM>). Another option is <http://fecsource.com/order.html>. Counting chambers come with either etched or green grids. The etched grids are harder to see than the higher contrast green grids.

Microscope:

Ideally, you need a compound microscope with a 10X eyepiece and 10X lenses (it will also have 4X and 40X lenses) with an internal light source. Reconditioned microscopes work fine and can be ordered on the web from places like Associated Microscope (1-800-476-3893, <http://www.associatedmicroscope.com/>) or obtained from high schools or colleges that have not turned in older microscopes yet for reconditioning.

Magnetic stirrer:

Refurbished magnetic stirrers are also available on the web. One source is Medical Resources USA (1- 800 330-3591, <http://www.medicalresources.com/>)

Notes: Fecal egg counts are a useful measure of potential pasture contamination. They are not necessarily correlated to actual worm numbers or to the severity of parasitic disease. Some types of worms lay more eggs than other types and some worms are more dangerous than others. Paired samples from the same animals before and after (7-10 days) deworming can help determine the effectiveness of an anthelmintic treatment. <90% reduction and <60% reduction in fecal egg counts suggest mild and severe resistance, respectively.

McMASTER QUANTITATIVE FECAL EGG COUNTING METHOD

EQUIPMENT, SUPPLIES AND REAGENTS:

Tongue depressors/applicator sticks
Balance
50 –100 ml beaker
Flotation solution – made with table salt, granulated sugar or Epson salts
1 cc syringes or transfer pipets
Advanced equine counting chamber – McMaster Slide

Saturated Salt Flotation Solution

Add salt to water with constant stirring until no longer going into solution

Approximately 350 g / 1 liter (0.8 lbs/ 1 quart) . 350 g =303 ml or 1.28 cups of table salt

Epson Salt Flotation Solution - dissolve 400 g (~ 1 ⅔ cup) in 1 liter of water

Sugar Flotation Solution – dissolve 1 lb (454 g) in 355 ml of hot water (~1 ¼ cups of sugar per cup of water)

PROCEDURE:

1. Place beaker on balance and tare it.
2. Using tongue depressor, weigh out 3 gm of feces into beaker.
3. Add approximately 15 ml flotation solution.
4. Mix well with tongue depressor to break lumps.
5. Bring up to 45 ml with flotation solution.
6. Continue mixing with applicator sticks for several minutes
7. While mixture is still stirring, draw about 1 ml fecal suspension into syringe or transfer pipet
8. Load one side of counting chamber carefully to avoid producing bubbles.
9. Repeat sampling and loading procedure for second side of chamber.
10. Let preparation stand 5 min (examine it at least by 20 min).
11. Place chamber on microscope and examine with 10X objective.
12. Count eggs in both sides of chamber.
13. Calculate eggs per gram:

45 ml final volume, $\text{epg} = (\text{side 1} + \text{side 2}) \times 50$

Note: The minimum detection limit is 100 eggs per gram.

COMMENTS:

The McMaster counting chamber is available from Chalex Corporation (formerly Advanced Equine Products), 5004-228th Ave SE, Issaquah, WA 98029 USA, Tel: 425-391-1169, Fax: 425-391-6669, Email: chalexcorp@att.net , Website: www.vetslides.com . Cost is approximately \$20 per chamber. The volume under each grid for this chamber is 0.15 ml. Counting chambers with etched grids are less expensive and last longer than the easier- to-see green grids.

Also if you have a smaller amount of feces, this table may help:

Amount of feces (g)	Amount of flotation solution needed to break up fecal pellets (ml)	Total volume of flotation solution to add (ml)	Total volume of solution plus feces (ml)
2 grams	10 ml	28 ml	30 ml
3 grams	15 ml	42 ml	45 ml
4 grams	20 ml	56 ml	60 ml