

Faecal Parasitology: Concentration methodology really does matter!

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Abstract

Since the establishment of UKNEQAS Faecal Parasitology in 1986, failure to report the parasite species present in EQA specimens and recovering lower numbers of parasite stages than those seen pre-distribution by UKNEQAS Parasitology have always been performance issues. Therefore a questionnaire was sent to the 618 participants in the Scheme to establish the routine method they used to examine for parasite stages in faecal samples. Of the 200 laboratories who responded, all used a concentration method based on the Modified Ridley-Allen technique (1) but with variations in the centrifugal force, centrifugal time, pore size of the sieve and use of a solvent. Therefore faecal concentrations were done in UK NEQAS to assess the effect of these parameters. The number of parasite stages recovered increased with centrifugal force, centrifugal time and use of a solvent.

Introduction

The use of a faecal concentration method for the examination of faeces for parasitic diseases increases the likelihood of finding ova, cysts and larvae in faecal specimens particularly in those specimens, where they are present in too few numbers to be seen by direct microscopy.

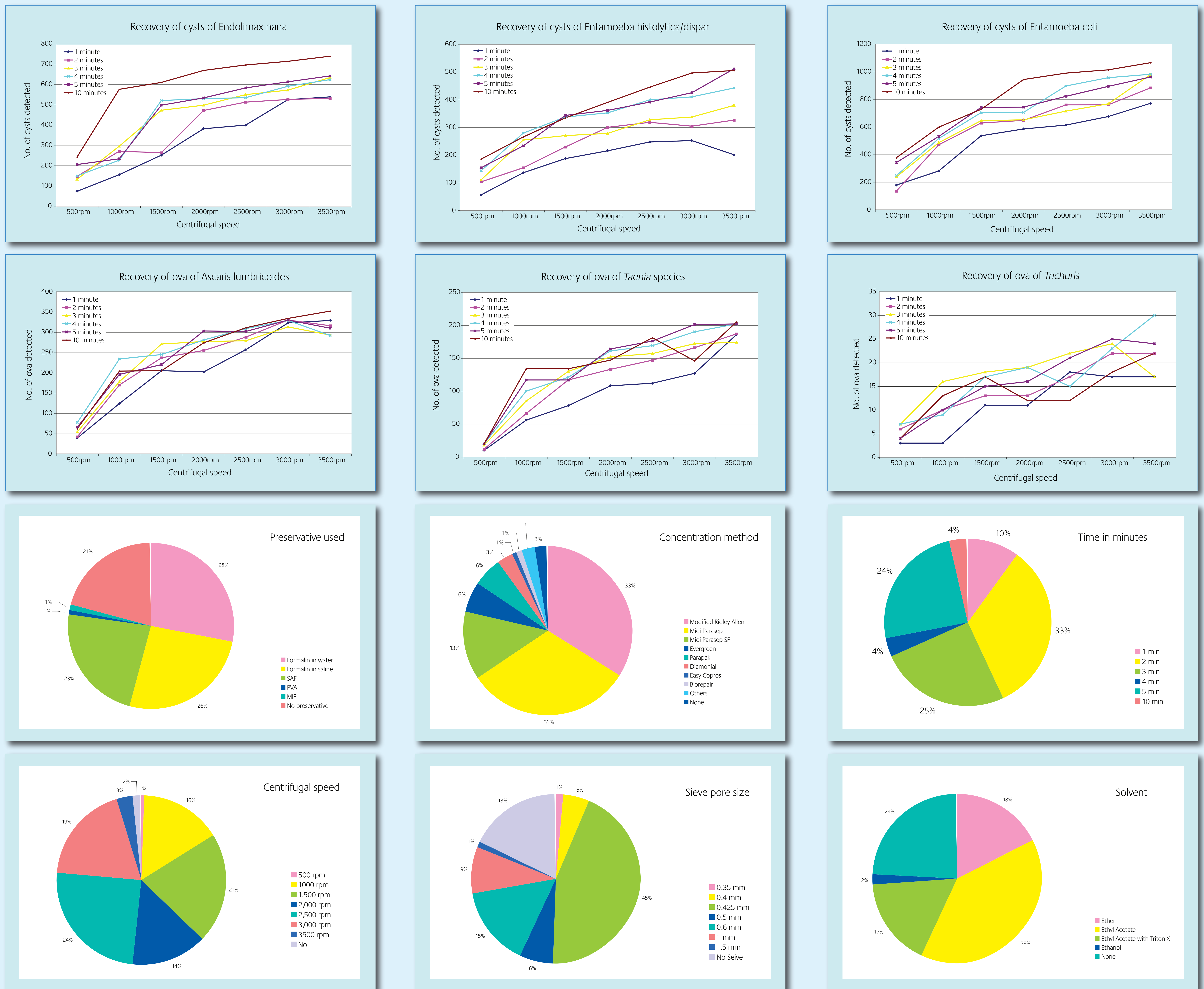
In response to concerns by UK NEQAS Parasitology regarding participants failing to report parasites in UK NEQAS faecal parasitology specimens and also noting that they were recovering lower numbers of ova and cysts than those seen by UK NEQAS in the pre-distribution examination of the specimens, we enquired regarding the routine method they used to examine for parasite stages in faecal samples.

Methods

618 participants subscribe to the UKNEQAS Parasitology Scheme. A questionnaire was sent to all participants to establish their method for concentrating faecal samples for the examination of parasites. 200 laboratories responded and their results were analysed. Following the analysis, faecal concentrations using the Parasep® faecal concentration method were performed in UK NEQAS on specimens containing a range of ova and cysts incorporating the variation in centrifugal force and centrifugal time reported in the participants questionnaires.

Results

Results of UKNEQAS Parasitology comparing different Centrifugal speed and Centrifugal time



Discussion

Although 96% of participants use a concentration method to examine faecal samples for parasites and all use a method based on the Modified Ridley Allen technique (1), there were variations in the centrifugal speed, centrifugal time, pore size of the sieve and solvent used. Previous studies have already shown that

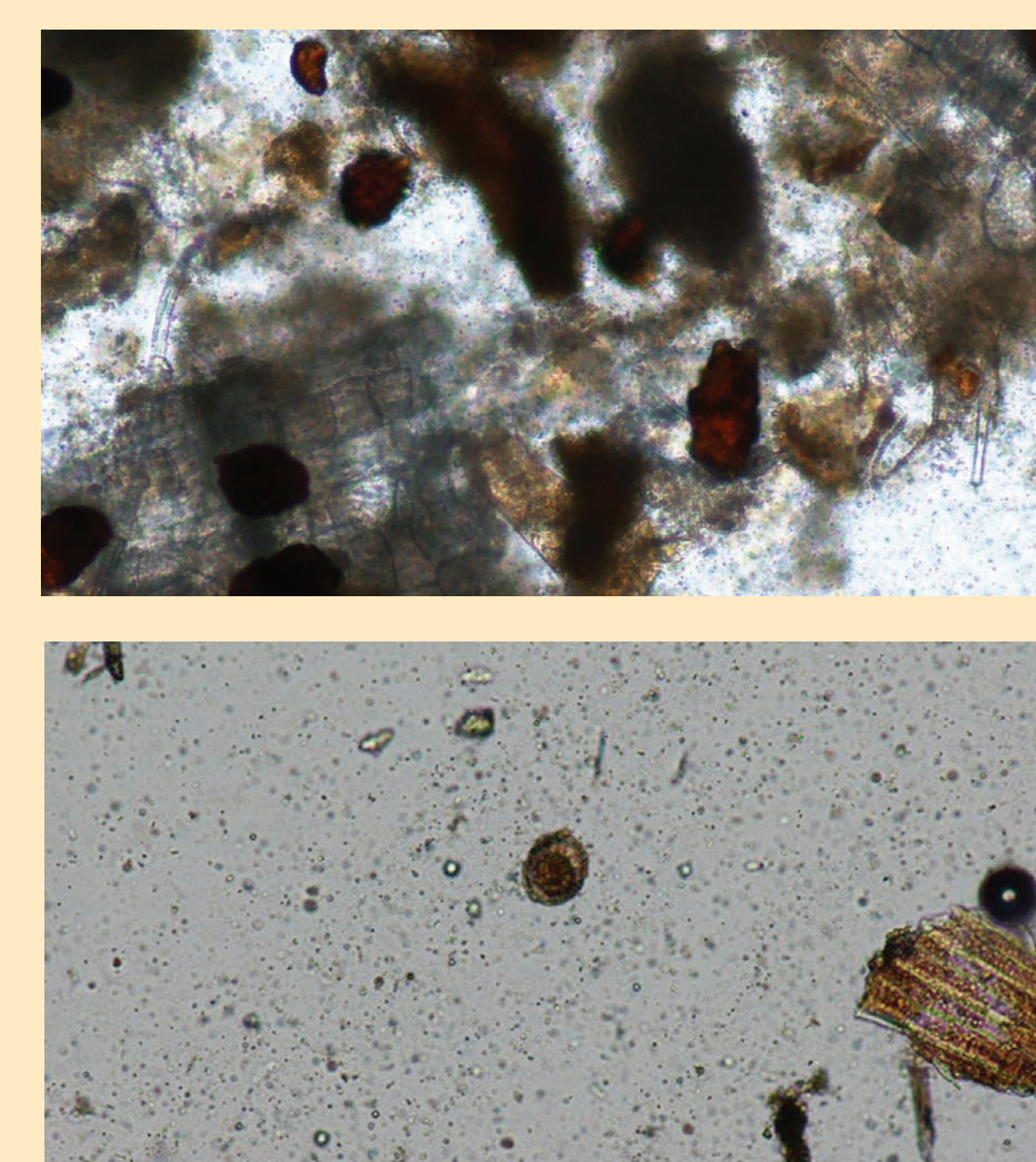
- 10% formalin in water is more effective than formalin in saline since the saline solution can affect the specific gravity such that cysts in particular may become trapped in the fatty layer (1).
- A sieve with a larger pore size results in an increase in the amount of debris making the deposit more difficult to examine.
- The recovery of parasite stages is significantly less if a solvent and more debris is present not used when concentrating a stool specimen (2).

An ovum of *Ascaris lumbricoides* in a specimen concentrated with ethyl acetate. The same specimen concentrated without solvent

- An in-house unpublished study showed that ethyl acetate used along with Triton X resulted in a greater recover of parasites than ether.

This study by UKNEQAS Parasitology showed that:

- The number of parasite stages increased with an increase in speed.
- Many participants centrifuge on a low speed for a longer time. However the graphs depicting the recovery of ova and cysts demonstrate that an adequate centrifugal force must be used since sufficient gravitational force is needed to sediment the ova and cysts.
- Centrifugal time at any given speed is also critical, since ova and cysts may remain in suspension if the sample is not centrifuged for the minimum required time
- Although the recovery of parasites was best if the samples were centrifuged for 3,500rpm for 10 minutes an increased amount of debris was observed making the deposit more difficult to examine.



Conclusions

- A faecal concentration method is essential to increase the sensitivity of finding parasite stages.
- Poor concentration technique leads to poor recovery of parasites.
- All laboratories should use the recommended centrifugal speed (3000rpm) and time (3 minutes) when using this technique to optimise diagnosis of faecal parasites by microscopy.

References

- (1) Allen A. V. H. and Ridley D. S. (1970): Further observations on the formalin-ether concentration technique for faecal parasites. J. Clin. Pathol. 23, 545 - 546.
- (2) Comparison between the Midi Parasep and Midi Parasep Solvent Free (SF) faecal parasite concentrators: A C Saez M M Manser, P L Chiodini; J Clin Pathol online June 16, 2011