Liver Fluke – Monitoring/Diagnostic Tools.

Diagnosis of liver fluke infection in the live animal is not straightforward but there are useful diagnostic tests available, each with their respective pros and cons, depending on the stage of fluke present in animals, time of year etc. Selected tests can be used to (i) monitor fluke infection and/or (ii) detect treatment outcomes and test for resistance (Table 22).

Table 22. Diagnostic tests for liver fluke:

	Age of fluke (weeks)													Comments	Best time to use
Diagnostic Test	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**12+**		
Liver/bile duct enzymes														Not specific for liver fluke, not recommended as a stand-alone test, requires supporting evidence	Any time of year, but requires careful interpretation
Serum/blood ELISA														Earliest reliable indicator (2-4 weeks post-infection) specific for liver fluke. Can stay +ve, even after successful treatment	From mid-summer onwards, best in young, first season grazing animals e.g. sentinel lambs
Faecal or coproantigen (cELISA)														Second earliest indicator (6-7 weeks post-infection), good indicator of treatment outcome	From late summer onwards, depending on the weather, can be used in young & older animals
Faecal egg count (FEC)														Latest indicator (10-12 weeks post-infection), works well as composite test, good monitoring tool	From autumn onwards, depending on the weather, can be used on young & older animals

Liver & bile duct enzymes (GLDH and/or GGT)

This is a blood test which detects biochemical changes associated with liver and/or bile duct damage from as early as 1-2 weeks post infection., However, it is not specific for liver fluke and will reflect other liver/bile duct problems, levels also fluctuate and are unstable. Therefore, it is not recommended as a stand-alone test because it requires supporting evidence.

Serum/antibody ELISA

Also, a blood test which detects antibodies circulating in response to fluke infection. It is very specific for liver fluke and provides the earliest indicator of fluke infection, with animals becoming positive (seroconverting) within 2-4 weeks of infection. Antibodies can persist in the blood for several months, so the test can remain positive, even after successful treatment. This means the ELISA test is most useful as an early indicator of infection in a season using first season animals e.g. spring-born lambs as sentinels. It is less useful in older animal (Gordon *et al.*, 2012).

Faecal or coproantigen ELISA (cELISA)

This test is a specific and sensitive non-invasive faecal test, which can detect infection a few weeks before eggs appear (i.e. 6-7 weeks post-infection, compared to 10-12 for FEC). Samples are relatively easy to collect and store, but the test does require a specialist laboratory to run it. cELISA is specific for liver fluke and does not cross-react with rumen fluke, which is increasingly found as a co-infection. However, the test does not work well as a composite (pooled) test and **should only be used on individual animal samples**. This is because it relies on a threshold value to indicate a positive result and mob samples can dilute the antigen levels if some animals are negative. However, this test is a good indicator of treatment outcome and has become the default choice for flukicide resistance testing via the coproantigen reduction test (or CRT), even without an accompanying FEC.

Faecal egg count /detection (FEC)

Another non-invasive faecal test but can only detect adult (patent / egg-laying) fluke infection, which typically means it is valid 10 -12 weeks after the animals were infected. Samples are easy to collect and the FEC method itself is relatively easy to perform and can detect liver fluke and rumen fluke eggs simultaneously. However, fluke eggs are typically shed sporadically meaning a FEC can generate false negative results, so care is needed in interpretation. A composite (pooled) FEC is a useful monitoring tool and FEC can also be adapted for flukicide efficacy testing e.g. composite TCBZ-R FECRT. (Daniel, *et al.*, 2012).