# BF Protocol FROG SPAWN

Aim To record the phenology of the spawning of the common frog in selected ponds and ditches by assessing the number of egg masses, as an indicator of the 'health' of frog populations. To identify any significant changes in those populations and to measure some physical characteristics of the ponds which may affect breeding success

Rationale The common frog (Rana temporaria) has been selected for inclusion in the ECN programme as an example of a ubiquitous predatory amphibian. Adult frogs feed mainly on insects, slugs and snails but also eat woodlice and the larvae of moths and butterflies. During the breeding season, frogs live in shallow ponds and ditches, but spend much time on land during the rest of the year. The frog population is therefore affected by conditions on land as well as in the shallow water bodies in which they breed. The changing status of frogs and other amphibians in Britain has been studied and reported (Cooke & Arnold 1982); populations have decreased, particularly during the 1960s (Cooke 1972), as a result of agricultural drainage, modification of breeding sites, and pollution by fertilizers and from other sources. In the last two decades there has been an increased awareness of several effects of the acidification of fresh waters by airborne pollutants and this has generated a parallel interest in the effects of increased acidity and concentrations of aluminium on larval amphibians (Cummins 1986; Beattie, Tyler-Jones & Baxter 1992). Numerous laboratory studies have demonstrated sublethal and lethal effects of acid conditions on embryonic and larval amphibians, and evidence of acid-related stress and mortality has also been found under field conditions (Cummins 1990).

It is difficult to monitor populations of adult frogs, but a rough approximation of frog colony size in a pond can be obtained by counting the number of egg clumps and multiplying by two; this assumes that each female produces one clump and that there is an even sex ratio (Cooke 1975). Frogs usually spawn in shallow water, up to about 15 cm depth, and the spawn masses and the water in which they rest are therefore usually reasonably accessible for monitoring purposes.

# Method Equipment

A pH meter suitable for use in the field, and a maximum/minimum thermometer are available from laboratory suppliers. Other equipment, such as a water depth measurement pole, is easily made.

#### Location

One or more shallow ponds or wet ditches will be selected, preferably those in which frogs are known to have bred in recent years and which are situated conveniently for frequent visits to record stages in development.

## Sampling

#### Biological

The time at which frog breeding starts in the UK varies greatly; in some years it may begin during December in Cornwall whilst not starting until April at high altitudes in the Pennines and in Scotland. In a particular pond, however, annual variation in the date of spawning tends to be rather small. Recorders will check the pond(s) weekly from about 1 January, or from an earlier or later date where local knowledge is available, to ascertain and record the date on which male frogs congregate in the spawning areas and begin calling. Thereafter the pond(s) will, if possible, be visited daily until the first eggs have hatched. At

sites where daily visits are impossible, because of time and distance constraints, visits should be made as often as possible, with a minimum of weekly visits, until the first eggs have hatched.

A record is made of the date on which spawn is first seen, and on subsequent daily or weekly visits the number of new spawn masses which have appeared since the last visit are recorded. Each new spawn mass is marked by carefully attaching a coloured thread to its edge, using a large bodkin needle. This will ensure that a mass is not counted twice. Newly deposited egg masses can be recognised because the eggs are packed tightly and the jelly capsules surrounding the embryos will not have expanded. Threads can be removed from egg masses which have been in the water for several days. The total surface area of water occupied by the spawn masses will be estimated in square metres. A percentage estimate of dead or obviously diseased eggs will be made. Where visits are made less frequently than daily, and in any case when the total number of spawn masses exceeds 100, it will be impossible to count new masses and only the total surface area of water occupied by the spawn masses will be estimated in square metres. The date on which embryos are first seen to have hatched from the eggs is recorded. Once embryos have started to hatch, weekly visits and recording (see next paragraph) are sufficient and will continue until newly metamorphosed frogs are seen leaving the pond, or for a period of 16 weeks from the time when spawn was first observed, whichever is the shorter.

#### **Physico-chemical**

At the time when spawn is first seen on the pond a 250 ml water sample is taken from the spawning area, using a bottle which has been rinsed in pond water before filling. Analysis of this sample provides a means of characterising ponds across the network and an annual baseline against which any subsequent changes in chemical composition of individual ponds can be assessed. Conductivity and pH are measured on unfiltered water according to methods in the ECN Initial Water Handling (WH) Protocol, which also describes the method for filtering the sample. After filtering, the water is analysed for dissolved Na<sup>+</sup>, K<sup>+</sup>, Ca<sup>2+</sup>, Mg<sup>2+</sup>, Fe<sup>2+</sup>, Al<sup>3+</sup>, NH<sub>4</sub><sup>+</sup>-N, Cl<sup>-</sup>, NO<sub>3</sub><sup>-</sup>-N, SO<sub>4</sub><sup>2-</sup>-S, PO<sub>4</sub><sup>3-</sup>-P, alkalinity and dissolved organic carbon. If there is subsequently a high and unexplained mortality of spawn or tadpoles, a further water sample is to be taken for the chemical analyses specified above, as an aid to explaining the cause of such mortality.

pH, temperature and water depth are important factors affecting the breeding success of frogs and these will be measured and recorded at weekly intervals between the date of first spawning and the date when newly metamorphosed frogs are first seen leaving the pond.

pH will be measured weekly at each of three re-locatable, random positions immediately outside the spawning area and close to the edge of the pond at a water depth of 50 mm. Ideally, a field pH meter should be used, its electrode being placed in the water with its tip at a depth of 50 mm.

A maximum/minimum thermometer will be set up in an area of open water immediately outside the spawning area by attaching the thermometer to a float so that it is held horizontally in the water at a depth of 50 mm. A suitable float can be made from a sturdy plastic bottle, its buoyancy being adjusted by adding water to ensure that the thermometer is held at the correct depth. Alternatively the thermometer can be attached to the lower surface of an appropriately sized block of wood which floats with the thermometer in the shade. The float is attached to the bank with one or more lines so that is held in position. The water temperature at the time of first spawning is recorded and the thermometer is subsequently read and re-set each week.

#### **BF** Protocol

Water depth is measured weekly by placing a pole, graduated in centimetres, in the water as near as practicable to the centre of the area occupied by spawn.

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**References** Beattie, R.C., Tyler-Jones, R. & Baxter, M.J. 1992. The effects of pH, aluminium concentration and temperature on the embryonic development of the European common frog, *Rana temporaria. Journal of Zoology*, **228**, 556-570.

**Cooke, A.S.** 1972. Indicators of recent changes in status in the British Isles of the frog (*Rana temporaria*) and the toad (*Bufo bufo*). *Journal of Zoology*, **167**, 161-178.

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**Cooke, A.S. & Arnold, H.R.** 1982. National changes in status of the commoner British amphibians and reptiles before 1974. *British Journal of Herpetology*, **6**, 206-207.

**Cummins, C.P.** 1986. Effects of aluminium and low pH on growth and development in *Rana temporaria* tadpoles. *Oecologia*, **69**, 248-252.

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# Specification of results and recording conventions

The measurement variables listed below are those required for each BF sampling location at an ECN Site. Sites submitting data to the ECNCCU should refer to the accompanying Data Transfer documentation for the specification of ECN dataset formats, available on the restricted access Site Managers' extranet. Contact <u>ecnccu@ceh.ac.uk</u> if you need access to this documentation.

The first 4 key parameters uniquely identify a sample or recording occasion in space and time, and must be included within all datasets:

- <u>Site Identification Code</u> (e.g. T05)
- Core Measurement Code (e.g. PC)
- Location Code (e.g. 01)
- Sampling Date (/time)

Unique code for each ECN 'core measurement' Each ECN Site allocates its own code to replicate sampling locations for each core measurement (e.g. for different surface water collection points) Date on which sample was collected or data recorded. This will include a time element

where sampling is more frequent than daily

Unique code for each ECN Site

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### Core measurement: vertebrates – frog spawn (BF Protocol)

Frog spawn monitoring is conducted annually at selected ponds and ditches, taking daily measurements of spawn and weekly measurements of pond environment. A single sample of pond water, taken at the time of first spawning, is analysed annually for a set of chemical determinands (see below). The pond is checked weekly from the 1 January for signs of frogs congregating and calling. Daily records are made of spawn development from the time the frogs are first seen congregating until the first eggs have hatched. Weekly records of pond pH and temperature are made between the date of first spawning and the date on which the newly metamorphosed frogs are first seen leaving the pond.

		Precision of
Variable	Units	recording
Pond ID	as location code (01,0	2, etc)
Date frogs first seen congregating		
Date of first spawning		
Date of first hatching		
Date newly metamorphosed frogs first	seen leaving	
Pond sample		
Site Identification Code		
Core Measurement Code		
Location Code		
Sampling date		<b>•</b> <i>i</i>
pH	pH scale	0.1
Conductivity	µS cm	0.1
Alkalinity	mg l	3 significant figures
Na	mg l	3 significant figures
K 2+	mg l	3 significant figures
	mg I	3 significant figures
Mg =- <sup>2+</sup>	mg I	3 significant figures
Fe	mg I	3 significant figures
	mg I	3 significant figures
	mg I	3 significant figures
	mg i	3 significant figures
	mg I	3 significant figures
NO <sub>3</sub> - N	mgi	3 significant figures

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SO4 <sup>2-</sup> -S	mg l <sup>-1</sup>	3 significant figures
Dissolved organic carbon	mg l <sup>-</sup> '	3 significant figures
Pond monitoring (weekly) Site Identification Code Core Measurement Code Location Code Recording (Sampling) date Date Min/May thermometers set		
Depth at centre of spawning area	cm	0.1
Minimum temperature	°C	1
Maximum temperature	°C	1
pH (at Position 1)	pH scale	0.1
pH (at Position 2)	pH scale	0.1
pH (at Position 3)	pH scale	0.1
Spawn monitoring (daily) Site Identification Code Core Measurement Code Location Code Recording (Sampling) date		
Number of new snawn masses	count	1
Total surface area of pond covered by	m <sup>2</sup>	0.1
spawn		
Percentage dead or diseased eggs	%	1

# **Recording forms**

A standard field recording form is available from the CCU. An example is provided in Appendix II.