Martin Godfrey reports his findings from a recent study of *Cephaloziella* chromosomes.

Some chromosome counts for the genus *Cephaloziella*

arly in 2011, Des Callaghan asked me if I would do chromosome counts on a number of *Cephaloziella* species which he was studying. There are only a few published chromosome counts for this genus (Smith, 1996; Paton, 1999), so this was an opportunity to break new ground as well as confirm the published counts for *C. massalongi* and *C. nicholsonii* (Newton in Paton, 1984).

Methods

Martha Newton has published an excellent guide to bryophyte chromosomes (Newton, 1989) which formed the basis for this project. All of the counts were done using mitotic preparations of growing shoot tips but, as on previous occasions, I found that I needed to experiment a bit to achieve satisfactory results.

The living specimens were placed in small jars, liberally sprayed with water, sealed and allowed to grow for at least 2 weeks to ensure that there were plenty of growing shoot tips. The tips were harvested by cutting 5–10 mm lengths of shoots which were fixed overnight in 3:1 absolute alcohol/glacial acetic acid. My main departure from Newton was to stain the fixed shoots for 3–4 hours prior to the excision and squashing

△ C. stellulifera. D. Callaghan

of the growing tips as I have found that this gives more consistent results. I also found that with *Cephaloziella* I got better staining using aceto-orcein rather than lacto-propionic orcein. After staining, around 0.5 mm of the growing tip was excised in a drop of 45% acetic acid and squashed in the usual way. As the chromosomes were rather small, counts were made using a $\times 20$ eyepiece with an oil immersion lens – in some cases a green filter was used to improve contrast.

Getting decent chromosome figures in *Cephaloziella* proved surprisingly difficult, especially in the case of *C. divaricata* which took many attempts. To check my technique I grew some *Cephalozia bicuspidata* alongside the *C. divaricata*, fixed and treated it in the same way and got excellent chromosome preparations, so it appears there is something about *Cephaloziella* which makes this genus rather intractable to standard techniques.

Results

Good counts were obtained for all of the species provided, and the results are summarized in Table 1. Not surprisingly, the new counts reflected what seems to be the underlying chromosome number for the genus, n=18, and it was pleasing

to confirm the known counts. In particular, finding that n=36 for the Welsh *C. nicholsonii* gives added support to the view (unpublished) that this species may well have arisen as a diploid of *C. massalongi*.

Duplicates of the specimens have been sent to RBGE and NHM for DNA sequencing studies. The vouchers have been retained in the personal herbarium of Des Callaghan.

Concluding remarks

I would strongly recommend that if you have not tried this very useful technique, you should do so. No special equipment beyond that typically owned by an active bryologist is needed and, whilst a bit of experimentation, practice and patience is required to get decent results, that can be said of most bryological techniques. Obtaining halfway decent chromosome figures is probably simpler than cutting reasonable leaf sections for a beginner.

Martin F. Godfrey

e martinandrosie@aol.com

References

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Table 1. Chromosome count results

*All specimens were collected and determined by Des Callaghan.

Specimen no.	Taxon	Habitat	Site	Date collected	Vc.	Altitude (asl)	Chromosome count
1295	C. divaricata	On old mine spoil beside stream in open	Hermon Copper Bog SSSI	23.02.11	48	150	18 (new count)
1294	C. hampeana	Thin humic soil over old mine spoil in open	Hermon Copper Bog SSSI	23.02.11	48	150	18 (new count)
1259	C. massalongi	Partially worked metal ore seam at mouth of old copper mine	Figra Mine	15.01.11	48	230	18
1281	C. massalongi	On base of old mine building wall beside small stream in open mixed woodland	Pant-y-wrach	04.02.11	48	40	18
1302	C. massalongi	On soil deep under overhang of stream bank	Hermon Copper Bog SSSI	23.02.11	48	120	18
1282	C. massalongi	On old mine spoil of leat bank in mixed woodland	Coed Aberglaslyn	05.02.11	49	60	18
1279	C. nicholsonii	On old copper mine spoil on north- facing moorland slope	Drws-y-coed	06.02.11	49	160	36
1284	C. rubella	On stream bank beside old copper mine in mixed woodland	Pant-y-wrach	04.02.11	48	40	18 (new count)
1260	C. stellulifera	Open area of flat spoil in old copper mine	Figra Mine	15.01.11	48	230	18 (new count)
1280	C. stellulifera	On old copper mine spoil on north- facing moorland slope	Drws-y-coed	06.02.11	49	160	18 (new count)