

**Improving procedures
for obtaining Sanger sequences from old herbarium specimens**
Mika Bendiksby, Lisbeth Thorbek, Charlotte Bjorå, Rune Halvorsen

ESM Table 1, 1/2

| Coll. | Extr. | Taxon | Voucher information | L | LF | 16 | K | SG | AH | A2 | SS |
|-------|-------|---|--|-------|-------|-------|-------|-----|-----|----|----|
| 1826 | 2009 | <i>Lagopsis marrubiastrum</i> (Stephan) Ikonn.-Gal. | Bunge s.n., 1826 (C) | 0,n,r | 0,n,r | 0,n | — | — | — | — | — |
| 1842 | 2010 | <i>Stachys acerosa</i> Boiss. | Th. Kotschy s.n., 24.05.1842 (O) | 1 | 1 | 1 | 1,f | — | — | — | — |
| 1848 | 2009 | <i>Eriophyton rhomboideum</i> (Benth.) Ryding | T. Thomson (C) | 1,n | 1,n | 1 | 1,n | 1 | 1 | — | 1 |
| 1848 | 2009 | <i>Lagopsis marrubiastrum</i> (Stephan) Ikonn.-Gal. | T. Thomson s.n., no date (C) | 1,n | 1,r | 1,f | 1,f,r | — | — | — | — |
| 1849 | 2004 | <i>Lamium incisum</i> Willd. | Engler s.n., Sept. 1849 (WU) | 0,n | 0,n,r | 0,n | 0,r | — | 0 | — | — |
| 1850 | 2009 | <i>Anisomeles heyneana</i> Benth. | Stocks 3649 (O) | 1,r | 1,r | 1 | 1,f,r | — | — | — | — |
| 1853 | 2009 | <i>Lamium multifidum</i> L. | HDP s.n., May 1853 (O) | 1,n | 1,n | 1 | — | 1 | 1 | 1 | — |
| 1853 | 2010 | <i>Stachys neuracalycina</i> Boiss. | I. Branche 88 (C) | 0 | 0 | 0 | — | — | — | — | — |
| 1862 | 2004 | <i>Lamium galactophyllum</i> Boiss. and Reut. | E. Bourgeau 223 (WU) | 1 | 1 | 1 | — | 1 | 1 | 1 | — |
| 1875 | 2004 | <i>Lamium hybridum</i> Vill. | Kerner 397 (WU) | 1 | 1 | 1 | — | 1 | 1 | — | — |
| 1879 | 2009 | <i>Eriophyton rhomboideum</i> (Benth.) Ryding | Dr. J.E.T. Aitchison 831 (C) | 1 | 1 | 1,n | 1,f,r | — | — | — | — |
| 1879 | 2009 | <i>Eriophyton rhomboideum</i> (Benth.) Ryding | A.W. Heyde 106 (UPS) | 0,n | 0,n | — | — | — | — | — | — |
| 1879 | 2009 | <i>Stachyopsis oblongata</i> (Schrenk) Popov and Vved. | A. Regel s.n., 08.06.1879 (C) | 0,n | 0,n | 0,n | — | — | — | — | — |
| 1882 | 2009 | <i>Lamium amplexicaule</i> L. var. <i>aleppicum</i> (Boiss. and Hausskn.) Bornm. | Th. Pichler s.n., anno 1882 (WU) | 1 | 1 | 1 | — | 1 | 1 | — | 1 |
| 1882 | 2004 | <i>Galeopsis pyrenaica</i> Bartl. | G. Gautier s.n., 12.07.1882 (WU) | 0 | 0 | — | — | — | — | — | — |
| 1883 | 2009 | <i>Hypogomphia turkestanica</i> Bunge | A. Regel s.n., 18.04.1883 (C) | 0,n | 0,n | 0,n | 0,f,r | — | — | — | — |
| 1884 | 2009 | <i>Lamium garganicum</i> subsp. <i>corsicum</i> (Gren. and Godr.) Mennema | D.C. Forsyth Major s.n., 15.5.1884 (UPS) | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 1884 | 2010 | <i>Matsimurella chinensis</i> (Benth.) Bendiksby | Dr. A. Henry 5220 (H) | 0,n | 0,n | 0,n | — | — | — | — | — |
| 1888 | 2004 | <i>Lamium amplexicaule</i> L. var. <i>aleppicum</i> (Boiss. and Hausskn.) Bornm. | O. Stapf. 209 (WU) | 1 | 1 | 1 | — | 1 | 1 | — | 1 |
| 1890 | 2003 | <i>Lamium x holsaticum</i> E.Krause | Wettstein s.n., anno 1890 (WU) | 1 | 1 | 1 | — | 1 | 1 | 1 | — |
| 1893 | 2004 | <i>Galeopsis x pernhofferi</i> Wettst. | D. Pernhoffer 1326 (WU) | 1 | 1 | — | — | 1 | 1 | 1 | 1 |
| 1894 | 2004 | <i>Lamium galactophyllum</i> Boiss. and Reut. | P. Sintensis 5538 (WU) | 0,n | 0,n | 0,n | 0,f,r | 0,r | — | — | — |
| 1894 | 2009 | <i>Lamium macrodon</i> Boiss. et Huet. | P. Sintenis 15477 (WU) | 1 | 1 | 1 | — | 1 | 1 | 1 | 1 |
| 1895 | 2004 | <i>Lamium eriocephalum</i> Benth. | Gerolle 340 (WU) | 1 | 1 | 1 | — | 1 | 1 | 1 | — |
| 1896 | 2003 | <i>Ballota spinosa</i> Lk. | C. Baenitz/Bertrand 1132506 (US) | 0 | — | — | — | — | — | — | — |
| 1898 | 2009 | <i>Hypogomphia turkestanica</i> Bunge | O. Paulsen 275 (C) | 1 | 1 | 1 | 1,n | — | — | — | — |
| 1902 | 2004 | <i>Lamium album</i> L. subsp. <i>crinitum</i> (Montbret & Aucher ex Benth.) Mennema | J. Bornmüller 7947 (WU) | 1 | 1 | 1 | 1 | 1 | 1 | 1 | — |
| 1902 | 2004 | <i>Lamium macrodon</i> Boiss. et Huet. | E. Zederbauer s.n., May 1902 (WU) | 1 | 1 | 1 | — | 1 | 1,r | 1 | — |
| 1902 | 2009 | <i>Lamium macrodon</i> Boiss. et Huet. | E. Zederbauer s.n., June 1902 (WU) | 1,n | 1,n | 1,n | — | 0 | 1,r | — | — |
| 1902 | 2010 | <i>Lamium tomentosum</i> Willd. | J. et A. Bornmüller s.n., 17.7.1902 (S) | 1 | 1 | 1 | — | 1 | 1 | 1 | 1 |
| 1903 | 2009 | <i>Lamium paczoskianum</i> Vorosch. ined. | C. Roth s.n., April 1903 (S) | 1 | 1 | 1 | — | 1 | 1 | — | — |
| 1904 | 2009 | <i>Lamium galactophyllum</i> Boiss. and Reut. | E. Koenig s.n., 7.6.1904 (WU) | 1 | 1 | 1 | — | 1 | 1 | 1 | 1 |
| 1905 | 2010 | <i>Lamium hybridum</i> Vill. | J.E. Palmér s.n., May 1905 (UPS) | 1 | 1 | 0 | — | 1 | 1 | — | 0 |
| 1906 | 2003 | <i>Ajugoïdes humilis</i> (Miq.) Makino | T. Makino 41905 (US) | 0,n | 0,n | 0,n | — | — | — | — | — |
| 1908 | 2004 | <i>Lamium hybridum</i> Vill. | M. and M. Malzéville 2436 (WU) | 1 | 1 | 1 | — | 1 | 1 | — | — |
| 1909 | 2009 | <i>Holmskioldia sanguinea</i> Retz. | Brixen s.n., 06.09.1909 (O) | 1 | 1 | 1 | 0 | — | — | — | — |
| 1909 | 2003 | <i>Lamium bifidum</i> Cirillo | A Latzel s.n., 29.3.1909 (UPS) | 1 | 1 | 1 | — | 1 | 1 | 1 | 1 |
| 1910 | 2009 | <i>Petitia domingensis</i> Jacq. | M. Fuertes 195 (O) | 1,r | 1,r | 1,n | 1,f,r | — | — | — | — |
| 1910 | 2010 | <i>Stachys heraclea</i> All. | Elias s.n., 01.07.1910 (C) | 1 | 1 | 1 | — | — | — | — | — |
| 1911 | 2004 | <i>Stachys aspera</i> Michx. | F.F. Forbes s.n., 27.06.1911 (UPS) | 1 | 1 | 1 | — | — | — | — | — |
| 1912 | 2003 | <i>Lamium moschatum</i> Mill. | S. Linder s.n., 4.11.1912 (UPS) | 1 | 1 | 1 | 1 | 1 | 1 | 1 | 1 |
| 1913 | 2010 | <i>Moluccella bucharica</i> (B. Fedtsch.) Ryding | B. Fedtschenko 7441 (C) | 1 | 1 | 1 | — | — | — | — | — |
| 1914 | 2002 | <i>Galeopsis segetum</i> Neck. | Landmark, A. 12 (O) | 1 | 1 | 1 | — | — | — | — | — |
| 1914 | 2004 | <i>Galeopsis reuteri</i> Rchb. | C. Bicknell and L. Pollini 2344 (WU) | 0,n | 0,n | — | — | — | — | — | — |
| 1914 | 2010 | <i>Microtoena patchoulii</i> (C.B.Clarke ex Hook.f.) C.Y.Wu and S.J.Hsuan | Levro 21009 (L) | 0,n | 0,n | 0,f,n | — | — | — | — | — |
| 1916 | 2004 | <i>Lagopsis supina</i> (Steph. ex Willd.) Ikonn.-Gal. | Siméon s.n., (28.03.1916) 17 (C) | 1,n | 1,r | 1,f | 1,f,r | — | — | — | — |
| 1918 | 2009 | <i>Loxocalyx quinquenervis</i> Hand-Mazz. | Dr. Heinr. Frh. V. Handel-Mazzetti 12507 (C) | 1,n,r | 1,n,r | 1,f | — | — | — | — | — |
| 1919 | 2010 | <i>Micotoena delavayi</i> Prain. | T. Siméon 1291 (C) | 1 | 1 | 1 | — | — | — | — | — |
| 1920 | 2005 | <i>Gomphostemma javanicum</i> (Blume) Benth. | M. Ramos and C. Edano s.n., June 1920 (US) | 0 | 0 | 0 | — | — | — | — | — |
| 1920 | 2009 | <i>Loxocalyx ambiguus</i> (Makino) Makino | Tomitaro Makino 41885 (S) | 0,n,r | 0,n,r | 0,n | 0,f,r | 0,r | — | — | 0 |
| 1929 | 2003 | <i>Paraphlomis javanica</i> (Blume) Prain | J. F. Rock 1097 (US) | 1 | 1 | 1 | 1 | — | — | — | — |
| 1921 | 2009 | <i>Matsimurella chinensis</i> (Benth.) Bendiksby | Wang Te Hui 272 (C) | 0 | 1 | 0 | 0 | — | — | — | — |
| 1921 | 2009 | <i>Vitex trifolia</i> L. <i>trifolia</i> | D. W. Garber 622 (O) | On | On | On | — | — | — | — | — |
| 1921 | 2010 | <i>Stachys angustifolia</i> M.Bieb. | Soska s.n., 28.07.1921 (O) | 1 | 1 | 1 | 1 | — | — | — | — |

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ESM Table 1, 2/2

| Coll. | Extr. | Taxon | Voucher information | L | LF | 16 | K | SG | AH | A2 | 5S |
|-------|-------|---|--|-----|-----|-----|-------|-----|----|----|----|
| 1922 | 2005 | <i>Achyrosperrum parviflorum</i> S.Moore | R.A. Dummer 5603 (US) | 1 | 1 | 1 | 1 | — | — | — | — |
| 1922 | 2004 | <i>Leucas volkensii</i> var. <i>parviflora</i> Sebald | Rob, E. and Th. C. E. Fries s.n., 16.01.1922 (UPS) | 1 | 1 | 1 | — | — | — | — | — |
| 1923 | 2009 | <i>Hemigenia pedunculata</i> Diels | M. Koch s.n., 02.11.1923 (O) | 1 | 1 | 1 | 1 | — | — | — | — |
| 1923 | 2004 | <i>Lamium flexuosum</i> Ten. | O. Gavioli 2942 (WU) | 0,n | 0,n | 0,n | 0,f,r | 0,r | — | — | — |
| 1923 | 2009 | <i>Microcorys loganiacea</i> F. Muell. | M. Koch s.n., 19.11.1923 (O) | 1 | 1 | 1 | 1 | — | — | — | — |
| 1924 | 2003 | <i>Lamium album</i> L. ssp. <i>barbatum</i> (Siebold and Zucc.) Mennema | H. Smith 6513 (S) | 1 | 1 | 1 | — | 1 | 1 | 1 | 1 |
| 1924 | 2009 | <i>Stachys affinis</i> Bunge | Harry Smith 5839 (UPS) | 1 | 1 | 1 | — | — | — | — | — |
| 1925 | 2009 | <i>Hypogomphia turkestanica</i> Bunge | A. Vvedensky 211 (C) | 0,n | 0,n | 0,n | 0,f,r | — | — | — | — |
| 1925 | 2009 | <i>Lamium amplexicaule</i> L. var. <i>orientale</i> (Pacz.) Mennema | G. Kleopow 5000 (S) | 1 | 1 | 1 | — | 1 | 1 | 1 | — |
| 1925 | 2010 | <i>Acrotome pallescens</i> Benth. | Arthur Örtendahl s.n., 1925 (UPS) | 1 | 1 | 1 | — | — | — | — | — |
| 1926 | 2009 | <i>Colebrookea oppositifolia</i> Sm. | Navkumar Datt Gairola 03 (S) | 1 | 1 | 1 | 1 | — | — | — | — |
| 1926 | 2010 | <i>Eremostachys sogdiana</i> Pazij and Vved. | M. Popov 6294 (C) | 1 | 1 | 1 | — | — | — | — | — |
| 1926 | 2009 | <i>Lamium flexuosum</i> Ten. | H. Lindberg 3722 (S) | 1 | 1 | 1 | — | 1 | 1 | 1 | — |
| 1926 | 2010 | <i>Stachyopsis oblongata</i> (Schrenk) Popov and Vved. | M. Popov and A. Vvedensky s.n., 07.1926 (H) | 1 | 1 | — | — | — | — | — | — |
| 1926 | 2010 | <i>Stachys arenaria</i> Vahl | J. Lid s.n., 21.04.1926 (O) | 0 | 0 | — | — | — | — | — | — |
| 1926 | 2010 | <i>Stachys saxicola</i> Coss. and Balansa | J. Lid s.n., 21.04.1926 (O) | 1 | 1 | 1 | — | — | — | — | — |

ESM Table 1: List of herbarium specimens tested that were collected prior to 1927, sorted by year of collection (Coll.). Additional information includes year of DNA extraction (Extr.), voucher information and genetic markers for which attempts at amplification were made for subsets of taxa. Successful/unsuccessful amplification using the ‘regular procedure’ is marked with 1/o. Amplification not attempted is marked with “—”. Sequence obtained via nested PCR is marked with n. Sequence obtained via the ‘replicate’ PCR procedure is marked with r. Sequence obtained by amplification of two shorter fragments is marked with f (possible for the *trnL-trnF* region, *rps16* and *matK*; see Fig. 1). DNA templates that did not generate any sequence are shaded in gray. Abbreviations: The plastid regions, L= *trnL* intron, LF= *trnL-trnF* spacer, 16= *rps16* intron, K= *matK*, SG= *trnS-trnG*, and AH= *psbA-trnH* spacer, and the nuclear regions, A2=RNA polymerase A2, and 5S=nuclear ribosomal 5S-NTS.