MOLECULAR DIVERGENCE PATTERNS WITHIN THE GONDWANAN LIVERWORT GENUS *JENSENIA* (PALLAVICINIACEAE, HEPATICOPHYTINA, BRYOPHYTA). STUDIES IN AUSTRAL TEMPERATE RAIN FOREST BRYOPHYTES 25

FRIEDERIKE SCHAUMANN*, TANJA PFEIFFER AND WOLFGANG FREY

ABSTRACT. For six of the seven species of the genus *Jensenia* (*J. pisicolor, J. connivens, J. erythrostans, J. diff timis, J. spinosa* and *J. decipiens* [Pallaviciniaceae, Hepaticophyta]) geomolecular divergence patterns are evaluated by comparison of the cpDNA *trnT*-*trnL* 5' exon intergenic spacer and *trnL* intron and nrDNA ITS2 sequences. All studied *Jensenia* taxa are characterized by a low intra- and interspecific sequence variation in the investigated DNA regions. The species are differentiated by a sequence divergence of 0–2.0% (*trnT-L* spacer) and 0–1.3% (*trnL* intron and ITS2), respectively. A comparison with the molecular data of five *Pallavicinia* s.str. species supports the delimitation of *Jensenia* as a distinct taxon.


INTRODUCTION

*Jensenia* Lindb. (Pallaviciniaceae) consists of about 7–8 species (Grolle 1964, Forrest et al. 2004) with southern temperate and alpine tropical distribution (Fig. 1.1). The genus is lacking in Laurasia, the family Pallaviciniaceae in general is presumably of Gondwanan origin (cf. Schuster 1982, Frey 1990). Together with *Pallavicinia* Gray and *Hattorianthus* R. M. Schust. & Inoue, *Jensenia* belongs to the subfamily Pallavicinioideae.

Morphologically, *Jensenia* is differentiated from its close relative *Pallavicinia* s.str. by the dendroid habit with regular 2 or 3(–4)× bifurcate terminal branching of the frond (Fig. 1.2, 3), and the gynoecea, which are strictly confined to frond bifurcations and are always situated above a terminal branch in the lower part of the aerial frond (Grolle & Piippo 1986). In literature the systematic position of *Jensenia* is discussed controversially. Following Gottsche (1864), Lindberg (1868), Stephani (1900) and Verdoorn (1932), Schuster (1963) and Grolle (1964) recognize the dendroid species as a distinct genus. This concept is followed by various authors (e.g., Grone 1980, Scott 1985, Grolle & Piippo 1986, Engel 1990, Perold 1993, Gradstein et al. 2001) and was recently confirmed by morphological studies (Schuette & Crandall-Stotler 2002). In contrast, Schifner (1893), Hässel de Menéndez (1961), Schuster (1992) and authors dealing with the New Zealand flora (e.g., Allison & Child 1975, Campbell et al. 1975, Glenny 1998) do not accept the dendroid species as a separate genus, but classify them as a subgenus of *Pallavicinia* s.l. (*Mittenia*...
Fig. 1. World distribution of the genus *Jensenia* (1 *J. spinosa*, 2 *J. pischolor*, 3 *J. connivens*, 4 *J. florschuetzii*, 5 *J. erythropus*, 6 *J. diffomis*, 7 *J. decipiens*) and habit of South American *J. pischolor* (2; voucher W. Frey & F. Schaumann 01-257) and Australasian *J. connivens* (3; voucher W. Frey & T. Pfeiffer 98-T27).
In this work, *Pallavicinia* s.str. and *Jensenia* are treated as separate genera. *Pallavicinia* s.str. species are prostrate (to ascending), but never dendroid as the species of *Jensenia*. *Jensenia* species prefer terricoleous sites, moist earth and clayey soil, in permanently atmospherically humid, more or less shaded habitats in montane and alpine environments. In the Neotropics *Jensenia* seems to be primarily alpine; below the forest line the species occur mostly in artificial habitats such as on steep, cut earth banks and along trails (Gradstein et al. 2001). The latter habitats are also characteristic for the New Zealand *J. connivens* (Colenso) Grolle.

*Jensenia pasicolor* (Hook.f. & Taylor) Grolle has an amphi-southatlantic distribution (South Georgia, Tristan da Cunha, Prince Edward Is., Crozet Is., Amsterdam I., Kerguelen Is., Falkland Is., Tierra del Fuego, Patagonia, Valdivian Region north to 40°10’S; Frey & Schaumann 2002, Grolle 2002; see Fig. 1). *Jensenia connivens* occurs in New Zealand (Grolle 1964; Ratkowsky 1987 excluded it from Tasmania). Scott (1985) lists the latter species from SE Australia. *Jensenia connivens* is here not considered to occur in SE Australia, since the species’ description and photograph by Scott (1985) as well as herbarium specimen (voucher B.Fuhrer & G.Scott, Bryo 236017 in B) determined by Scott belong to *Pallavicinia rubristipa* Schiffn. *Jensenia pasicolor* and *J. connivens* are habitually very similar if not identical (Scott 1985) and may be regarded as Gondwanan, respectively palaeoaustral taxa, and the presumable progenitor of derived alpine tropical species. These latter alpine tropical species include the neotropical *J. difformis* (Nees) Grolle (incl. *J. wallisii* [J.B.Jack & Steph.] Grolle; which is probably conspecific with *J. difformis* [Grolle 1964], after Hässel de Menéndez [1961], however, conspecific with *J. erythropus*) from the Andes, SE-Brazil, Mt. Roraima, and Costa Rica (new record, Holz et al. 2001), *J. erythropus* (Gottsche) Grolle (with two varities, var. *erythropus* and var. *nobandae* Gronde [Gronde 1980]; widespread, páramos of Costa Rica, high Andes from Bolivia to Venezuela, Sierra de Itatiaia; Gronde 1980, Gradstein et al. 2001) and *J. florschuetzii* Gronde (páramos of Colombia and northern Ecuador) as well as the Indo-Malayan *J. decipiens* (Mitt.) Grolle (Sri Lanka, Sumatra, Java, Celebes, Sabah, Mindanao, Luzon, Papua New Guinea). The only African representative of the genus, *J. spinosa* (Lindenh. & Gottsche) Grolle occurring from Malawi, Tanzania, Rwanda, Zaire to South Africa, Mauritius, Réunion and St. Helena (Perold 1993), may also be a Gondwanan relict species, with an independent evolution since the disruption of Africa from the palaeoaustrial region ca. 105 mio. years ago (McLoughlin 2001).

DNA sequencing offers a suitable tool to clarify the systematic status of *Jensenia* and the molecular divergence patterns within the taxon. Sequences of the cpDNA (*trnT-trnL* spacer, *trnL 5’exon, trnL intron*) and nrDNA (ITS2) are compared. These are suitable markers for the distinction of Pallaviciniacean species (compare Schaumann et al. 2003 for *Symphyogyna*). Furthermore, geomolecular divergence patterns of the Gondwanan relict species (*J. pasicolor, J. connivens, J. spinosa*) and presumable modern taxa from secondary speciation centers in the Neotropics and Indo-Malaysia are assessed.
MATERIAL AND METHODS

Plant material. The collecting data as well as further relevant data (GenBank accession numbers etc.) of the investigated specimens are listed in Table I. The trnL_UAA intron sequence of Jensenia connivens (1) was analyzed by M. Stech. Fresh plant material of J. florschuetzii was not available.

DNA preparation, PCR and sequencing. For DNA extraction from herbarium tissues and fresh plant material the method of Doyle & Doyle (1990) was followed with the exception of using only 76% (v/v) ethanol to wash the pellets after precipitation with cold isopropanol. PCR reactions with primers C_M/D_M for the cpDNA trnL_UAA intron, A/D_s for the cpDNA trnT_UGU-L_UAA intergenic spacer (A, C_M, D_M slightly modified after Taberlet et al. 1991, the internal intron primer D_s [5'-GTG TCC TTC GAG TCT CTG CAC-3'] was developed for liverworts [M. Stech, pers. comm.] and 5.8F/25R for nrDNA internal transcribed spacer (ITS2; after Baldwin 1992; primer sequences are available on request), purification of PCR products and sequencing follow the method described in Quandt et al. (2001).

Alignment and tree construction. The alignment of the sequences was created manually with the alignment editor Align32 (Hepperle 1997). Species of the sister genus Symphyogyna (S. hymenophyllum and S. podophylla) were used as outgroup (subfam. Symphyogynoideae [Trev.] Schust., Schuster 1992). PAUP4.0b10 (Swofford 2002) was used for the calculation of molecular trees. Maximum parsimony and likelihood analyses were performed with three different data sets: cpDNA (trnT-trnL spacer, trnL 5'exon, trnL intron), nrDNA (ITS2) and combined investigated cpDNA/nrDNA regions. The first eight positions of the trnT-trnL spacer were excluded due to incomplete sequences. The lengths of the trnT-trnL spacer given in the Results are based on this data set, and values are hence smaller than the ‘real’ lengths. Analyses were conducted including and excluding ambiguous sequence parts (trnT-trnL spacer: positions 255–274, trnL intron: positions 111–137, ITS2: positions 31–59). Heuristic searches were performed with the following options: all characters unweighted and unordered, multistate characters interpreted as uncertainties, gaps coded as missing data, performing TBR branch swapping, collapse zero length branches, MulTrees option in effect. Heuristic bootstrap searches were performed with 1000 replicates, 1000 random addition replicates per bootstrap replicate, and the same options in effect.

Maximum likelihood analyses were executed using the models and parameters that best fit the data as evaluated by Modeltest v.3.06 (Posada & Crandall 1998) employing the winModeltest front-end v4b (Patti 2002). For the nrDNA regions the TrN+G and GTR+G models, for the cpDNA regions the HKY+G and TrN+I models and for the combined data sets the TrN+G and TrN+I models were performed. Likelihood bootstrap analyses were performed with 500 replicates, 10 random addition replicates per bootstrap replicate, and the same options in effect.

RESULTS

In the six investigated Jensenia species the trnL intron length ranges from 314–315 bp; the trnT-L spacer has a length of 251–254 bp. In the studied species of
Table 1. List of investigated specimens with abbreviations (abbr.), origin, sampling data, herbarium (herb.) in which a voucher is held, and GenBank accession numbers (acc. no.). W. Frey = private herbarium W. Frey, H. Kürschner = private herbarium H. Kürschner, both Berlin.

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**Jensenia connivens** (Colenso) Grolle
- 1. New Zealand, South Island, Westland National Park: W. Frey & T. Pfeiffer, 98Mo11
  - Origin: W. Frey, CHR
  - Sampling data: 8100
  - Herb.: CHR 527432
  - Acc. no.: AY547526, AY547511
  - Origin: W. Frey, CHR
  - Sampling data: 8100
  - Herb.: CHR 527432
  - Acc. no.: AY547526, AY547511

**Jensenia decipiens** (Mitt.) Grolle
- 1. Papua New Guinea: T. Koponen, 31048
  - Origin: B
  - Sampling data: B 270408
  - Herb.: Bryo 219635
  - Acc. no.: AY547532, AY547516
- 2. Philippines, Luzon: M. Jacobs, B 448
  - Origin: B
  - Sampling data: B 234544
  - Herb.: Bryo 219635
  - Acc. no.: AY547532, AY547516
- 3. West-Irian: P. Hiepko & W. Schultze-Motel, 2051
  - Origin: B
  - Sampling data: B 270408
  - Herb.: Bryo 219635
  - Acc. no.: AY547532, AY547516

**Jensenia diffinis** (Nees) Grolle
- Ecuador, Prov. Zamora-Chinchipe: H. Kürschner, G. Parolly & D. Wagner, 02-619
  - Origin: B
  - Sampling data: B 270408
  - Herb.: Bryo 219635
  - Acc. no.: AY547532, AY547516

**Jensenia erythropus** (Gottsche) Grolle var. nobandae Gronde
  - Origin: B
  - Sampling data: B 270408
  - Herb.: Bryo 219635
  - Acc. no.: AY547532, AY547516
- 2. Colombia, Boyaca: A.M. Cleef, 9780
  - Origin: B
  - Sampling data: B 270408
  - Herb.: Bryo 219635
  - Acc. no.: AY547532, AY547516

**Jensenia picrocolor** (Hook.f. & Taylor) Grolle
- Kerguelen Archipel: E. Aubert de la Rue
  - Origin: B
  - Sampling data: B 270408
  - Herb.: Bryo 037979
  - Acc. no.: AY547532, AY547516
- 2. Chile, X. region, Cordillera Pelada: W. Frey & F. Schaumann, Mo 01-257
  - Origin: B
  - Sampling data: B 270408
  - Herb.: Bryo 037979
  - Acc. no.: AY547532, AY547516
- 3. Chile, X. region, Reserva Nacional de Llanquihue: W. Frey & F. Schaumann, 01-382a
  - Origin: B
  - Sampling data: B 270408
  - Herb.: Bryo 037979
  - Acc. no.: AY547532, AY547516

**Jensenia spinosa** (Lindenb. & Gottsch.) Grolle
- Tanzania, Morogoro District: T. Pócs, R. Ochrya & H. Bednarek-Ochrya, Ser. VIII, No. 197
  - Origin: B
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  - Herb.: Bryo 037979
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*Pallavicinia longispina* Steph.
Taiwan

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*Pallavicinia hyellii* (Hook.) Carruth.
Rwanda

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*Pallavicinia rubristipa* Schiffn.
Australia, Vic.

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*Pallavicinia subelliiata* (Austin) Steph.
1 Japan, Fukayabakei

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2 Japan, between Sennin-bushi and Akanuma

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*Pallavicinia xiphoideas* (Hook.f. & Taylor) Trevis.
1 Chile, X. region, near Valdivia

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2 New Zealand, South Island, Fiordland National Park

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*Symphyogyna hymenophyllum* (Hook.) Mont. & Nees
New Zealand, North Island, Urewera National Park

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*Symphyogyna podophylla* (Thunb.) Mont. & Nees
Chile, X. region, Parque Nacional Puyehue

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Pallavicinia s.str., the trnL intron varies from 320 bp to 328 bp and the trnT-L spacer from 242–252 bp. In taxa of the Symphyogyna outgroup the trnL intron sequences are 326–331 bp and the trnT-L spacer 249–250 bp long. The trnL 5’ exon comprises 35 bp in all studied taxa. In the cpDNA alignment 189 of the 663 positions are variable (29%). Of the variable positions, 148 (78%, 22% of total positions) are parsimony-informative.

ITS2 has a length of 233–234 bp in Jensenia and of 224–236 bp in Pallavicinia s.str. In the outgroup, ITS2 sequence length varies from 216 to 218 bp. Of the 281 positions 97 (35%) are variable. Of the variable positions, 61 (63%, 22% of total included positions) are parsimony-informative. In the combined cpDNA and nrDNA data set 944 positions were used for tree construction. 284 (30%) of the positions are variable and 209 of the variable positions (74%, 22% of total included positions) are parsimony-informative.

The studied cpDNA and nrDNA regions show a low sequence divergence in the studied species of Jensenia. The trnL 5’ exon is identical in all Jensenia taxa. Interspecific differentiation ranges from 0–5 substitutions in the trnT-L spacer, 0–4 substitutions in the trnL intron and 0–3 substitutions in the ITS2 region (interspecific substitution percentage: trnL intron as well as ITS2 0–1.3%, trnT-L spacer 0–2.0%). Intraspecific differentiation is also very low (intraspecific substitution percentage: trnT-L spacer 0–0.8%, trnL intron 0–0.3%, ITS2 0–0.4%). In comparison to other genera of the Pallaviciniaceae (Pallavicinia and Symphyogyna, compare also Schaumann et al. 2003), the inter- and intraspecific substitution percentage in the trnL intron, trnT-L spacer and ITS2 region of Jensenia is very low (Tables 2–4).

A geographical differentiation between the species of Jensenia is discernible when regarding the percentage of substitutions in the combined data set of the trnL intron/trnT-L spacer and ITS2 sequences (Table 4). According to the molecular data, Jensenia piscicolor is more similar to the northern South American species J. difformis and J. erythropus as well as the African J. spinosa than to the Australasian J. connivens and Indo-Malayan J. decipiens. Jensenia decipiens is most distant from all other Jensenia species. This geomolecular differentiation is only partially displayed in the calculated molecular trees (see Figs. 2, 3).

The maximum parsimony analysis of the combined cpDNA and nrDNA data set resulted in 63 most parsimonious trees; the strict consensus of these trees is shown in Fig. 2 (length 408 steps, CI=0.7990, RI=0.8746). The likelihood analysis of the combined cpDNA and nrDNA calculated with the GTR+G model resulted in the phylogram shown in Fig. 3 (ln L=−3208.74312). Both trees are very similar, support Jensenia as a monophylum and demonstrate three Pallavicinia s.str. clades. A well-supported clade is formed by P. lyelli, P. longispina and P. subciliata, another solely by P. xiphoides samples and a third by P. rubristipa. Both dendrograms differ only in the relationship within Pallavicinia s.str.

All dendrograms, calculated with the three different data sets and model types as well as with the variable positions excluded (data only partially shown), show Jensenia as a well-supported monophylum supported by high bootstrap values (e.g., Figs. 2, 3). Within Jensenia the trees only show slight differences. Except for dendrograms based solely on cpDNA, Jensenia decipiens and J. connivens form one clade (bootstrap values 54–62%;
Fig. 2. Strict consensus of 63 most parsimonious trees (length 408 steps, CI=0.7990, RI=0.8746) inferred from combined cpDNA and nrDNA sequences (trnT-L spacer, trnL 5’ exon, trnL intron and ITS2) of 21 specimens of Pallaviciniaceae (compare Table 1) by a heuristic search with PAUP 4.0b10. Symphyogyna hymenophyllum and S. podophylla were used as outgroup. Numbers above branches indicate bootstrap values >50% from 1000 replicates with 1000 random addition replicates per bootstrap replicate.
Fig. 3. Maximum likelihood phylogram (lnL = −3208.74312) based on combined cpDNA and nrDNA sequences (trnT-L spacer, trnL 5′exon, trnL intron and ITS2) of 21 specimens of Pallaviciniaceae (compare Table 1) with PAUP 4.0b10. *Symphyogyna hymenophyllum* and *S. podophylla* were used as outgroup. Numbers above branches indicate bootstrap values from 500 replicates with 10 random addition replicates per bootstrap replicate.
Table 2. Percentage of substitutions [%] in the trnT-L spacer and trnL intron within the Pallaviciniaceae. (For values on interspecific substitution percentages in the trnL intron within Symphyogyna compare Schaumann et al. 2003)

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Table 3. Percentage of substitutions [%] in the ITS2 region within the Pallaviciniaceae. (For values on interspecific substitution percentages within Symphyogyna compare Schaumann et al. 2003)

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<td>Pallavicinia</td>
<td>6.9–20.6</td>
<td>3.6–18.4</td>
<td></td>
</tr>
<tr>
<td>Symphyogyna</td>
<td>15.3–17.8</td>
<td>14.7–24.7</td>
<td>6.3–15.6</td>
</tr>
</tbody>
</table>

Table 4. Percentage of substitutions [%] in the combined trnT-L spacer, trnL intron and ITS2 sequence within the genus Jensenia. (*: value refers only to cpDNA)

<table>
<thead>
<tr>
<th>Distribution</th>
<th>Taxa</th>
<th>J. pisicola</th>
<th>J. erythropus</th>
<th>J. diiformis</th>
<th>J. spinosa</th>
<th>J. connivens</th>
<th>J. decipiens</th>
</tr>
</thead>
<tbody>
<tr>
<td>Southern South America</td>
<td>J. pisicola</td>
<td>0–0.2*</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>subtantarctic islands</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Northern South America</td>
<td>J. erythropus</td>
<td>0.2</td>
<td>0.2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Northern South America</td>
<td>J. diiformis</td>
<td>0.2</td>
<td>0.2</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Africa</td>
<td>J. spinosa</td>
<td>0.2</td>
<td>0.2</td>
<td>0.2</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>New Zealand</td>
<td>J. connivens</td>
<td>0.4–0.7</td>
<td>0.4–0.7</td>
<td>0.4–0.7</td>
<td>0.4–0.7</td>
<td>0.4</td>
<td></td>
</tr>
<tr>
<td>Indo-Malaysia</td>
<td>J. decipiens</td>
<td>0.8</td>
<td>0.8</td>
<td>0.7</td>
<td>0.7–1.1</td>
<td>0.2*–0.5*</td>
<td></td>
</tr>
</tbody>
</table>

see Figs. 2, 3). In maximum likelihood phylograms Jensenia pisicola from Kerguelen Island is not part of the J. pisicola clade, but is displayed as most distant from all other Jensenia species (see Fig. 3; supported, if at all, by a low bootstrap value 52%). Also J. erythropus from Colombia is occasionally depicted on an own branch (see Fig. 3).

These dendrograms as well as the equally parsimonious trees mostly differ in the topology of Pallavicinia s.str. Especially the position of Pallavicinia rubristipa varies. Maximum parsimony and likelihood analyses conducted with the ITS2 sequence and partially the combined cpDNA and nrDNA data set support an affinity of P. rubristipa to
_Jensenia_ (see also Figs. 2, 3). In the ITS2 maximum parsimony phylogram this is supported by a high bootstrap value (data not shown). In some combined cpDNA and nrDNA as well as the nrDNA maximum likelihood dendrograms, _Pallavicinia_ s.str. forms a monophylum (no bootstrap support).

**DISCUSSION**

The species of _Jensenia_ are morphologically, ecologically and molecularly clearly differentiated from _Pallavicinia_ s.str. The taxon forms a well-supported monophylum on the molecular level (Figs. 2, 3). All investigated _Jensenia_ species are characterized by a low sequence divergence in the investigated cpDNA (trnT-L spacer/trnL intron) and nrDNA (ITS2) regions. The recognition of a separate taxon is therefore justified. The substitution percentages between _Jensenia_ and _Pallavicinia_ are as high as between other genera of the Pallaviciniaceae (s. Table 2, 3) suggesting a delimitation of _Jensenia_ on generic rank. However, when _Jensenia_ is accepted as a genus, _Pallavicinia_ s.str. is paraphyletic in several phylograms (cf. Figs. 2, 3). This is mainly due to _P. rubristipa_, which shows similarities to _Jensenia_ taxa in the ITS2 region. Further molecular studies with a broader sampling of Pallaviciniacean species even show polyphyly of _Pallavicinia_ (own unpublished data). Morphologically _Jensenia_ is clearly delimited as a genus (e.g., Grolle 1964, Grolle & Piippo 1986, Schuette & Crandall-Stotler 2002) and therefore here retained at this rank.

Schuster (1992) places the species of _Jensenia_ in _Pallavicinia_ subgenus _Mittenia_, since some _Pallavicinia_ species are transitional between _Jensenia_ and _Pallavicinia_ s.str. The androecia in _Pallavicinia leviier Schiffn._ are situated in ill-demarcated rows and do not stand in two rows as typical for _Pallavicinia_ s.str. Additionally, some species intergrade between dendroid _Jensenia_ and the prostrate _Pallavicinia_ s.str. species. _Jensenia_ specimens always differentiate into creeping rhizomatous axes and an erect flabellate frond (2 or 3–4)× bifurcate terminal branching. Ascending axes, only 1–2 (very rarely 3)× bifurcate terminal branching, are known from _Pallavicinia_ s.str. (_P. longispina_ Steph., _P. rubristipa_, _P. ambigua_ [Mitten] Steph.). On the molecular level _Pallavicinia longispina_ and _P. rubristipa_ clearly belong to _Pallavicinia_ s.str. The ascending and dendroid taxa evolved independently within the Pallaviciniaceae, e.g., in _Jensenia, Pallavicinia_ s.str. (e.g., _P. longispina_) and _Symphyogyna_ spp. (Schaumann et al. 2003).

All species of the genus _Jensenia_ are characterized by a very low percentage of substitutions and no indels in the studied cpDNA and nrDNA regions (see Table 2, 3). As in other bryophytes divergence within the trnT-L spacer is higher than in the trnL intron (Meißner et al. 1998, Quandt et al. 2001, Pfeiffer et al. 2004). In liverworts trnL intron divergence values between 0 and 2.4% are common on infraspecific level and between 2.4–6.1% on interspecific level (Meißner et al. 1998, Pfeiffer et al. 2004). Infraspecific variation ranges in the trnT-L spacer from 0–3.4% and interspecific variation from 5.4–9.6% (Meißner et al. 1998, Pfeiffer et al. 2004). The substitution percentages of these DNA regions are not constant as often supposed but vary enormously in the studied genera of the Pallaviciniaceae (_Jensenia, Pallavicinia, Symphyogyna_; for data on _Symphyogyna_ cf. Schaumann et al. 2003).

_Jensenia_ belongs to the ancient Pallaviciniaceae (Schuster 1992) and probably has a
Gondwanan origin. The long geographical isolation led to the development of morphological clearly distinct species on former Gondwanan fragments. The observed high sequence homology of *Jensenia* species in the cpDNA (trnT-L spacer/trnL intron) and nrDNA (ITS2) regions can either be explained by a stenoevolutive behaviour of the genus or by an evolutionarily young age (which would contradict all previous morpho-ecological assessments).

**Interspecific relationships in *Jensenia***

A geographical divergence between the treated *Jensenia* species is observable, although the infrageneric molecular divergence is very low (Table 4). Taxa evolved in secondary distribution areas as the northern Andes and Indo-Malaysia are on the molecular level similar to their relatives in southern South America or New Zealand, respectively. *Jensenia pisicolor* of southern South America and from the subantarctic islands is closely related to *J. erythropus* and *J. difformis* from northern South America. Furthermore, *J. pisicolor* stands closer to the African *J. spinosa* than to *J. connivens* from New Zealand. The nearest relative of the Indo-Malayan *J. decipiens* is the latter species. In the phylograms both species form a clade (see Figs. 2, 3).

The habitual similarity of *J. connivens* and *J. pisicolor* supposed by Scott (1985), leading to his conclusion of their possible identity, is probably due to his misidentification of *J. connivens*. Fig. 1 clearly shows that both species differ in their habit. While *J. pisicolor* thalli possess small teeth, barely visible with a hand-lens, the teeth of *J. connivens* are conspicuous. Also the molecular data shows that *J. pisicolor* and *J. connivens* are distinct species.

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**REFERENCES**


