

بِسْمِ اللَّهِ الرَّحْمَنِ الرَّحِيمِ

مجله تخصصی  
تاکسونومی و مورفولوژی

علمی-پژوهشی

سال چهارم - شماره نهم - زمستان ۱۳۹۱

مجلهٔ تاکسونومی و بیوسیستماتیک بر اساس ابلاغیه شماره ۳/۱۱/۹۵۵ مورخ ۱۳۸۸/۰۶/۳۱ کمیسیون بررسی نشریات علمی وزارت علوم تحقیقات و فناوری، دارای درجه علمی-پژوهشی و شماره استاندارد بین‌المللی (شاپا) ۸۹۰۶-۲۰۰۸ (نسخه چاپی) و شماره استاندارد بین‌المللی ۲۱۹۰-۲۳۲۲ (نسخه الکترونیک) از سازمان اسناد و کتابخانه ملی جمهوری اسلامی ایران می‌باشد.

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چاپ و لیتوگرافی: انتشارات دانشگاه اصفهان

ناشر: دانشگاه اصفهان

انتشار: بهار ۱۳۹۲

تاکسونومی و بیوسیستماتیک  
سال چهارم - شماره سیزدهم - زمستان ۱۳۹۱  
شماره استاندارد بین‌المللی: ۸۹۰۶-۲۰۰۸  
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علمی-پژوهشی

صاحب امتیاز: معاونت پژوهش و فناوری دانشگاه اصفهان

دانشگاه اصفهان

سر دبیر: دکتر محمدرضا رحیمی نژاد رنجبر

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دکتر علی اکبر احسانپور	استاد - دانشگاه اصفهان
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صفحه‌آرای تخصصی: فریبا هادیان

ناشر: انتشارات دانشگاه اصفهان

نشانی: اصفهان - خیابان هزار جریب - دانشگاه اصفهان - ساختمان کتابخانه مرکزی - معاونت پژوهش و فناوری

طبقه دوم - اداره چاپ، انتشارات و مجلات - کد پستی: ۸۱۷۴۶۷۳۴۴۱ - دفتر مجله تاکسونومی و بیوسیستماتیک

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### معرفی مجله تاکسونومی و بیوسیستماتیک

مجله تاکسونومی و بیوسیستماتیک به صورت فصلنامه و هر سه ماه یکبار توسط دانشگاه اصفهان منتشر می‌شود. هدف از انتشار این مجله معرفی یافته‌های علمی استادان و پژوهشگران در زمینه تاکسونومی و بیوسیستماتیک، به ویژه با تأکید بر خزانه وراثتی جانداران (یوکاریوت‌ها و پروکاریوت‌ها) در ایران می‌باشد. مجله علمی - پژوهشی تاکسونومی و بیوسیستماتیک در زمینه‌های معرفی تاکسون‌های جدید، مرور نامگذاری تاکسون‌ها، طبقه‌بندی تاکسون‌ها، معرفی روش‌های جدید ایجاد و تحلیل داده‌ها، ژن‌اکولوژی، ژنتیک جمعیت‌ها و تنوع وراثتی، تنوع زیستی و فیلوژنی تاکسون‌ها، مقاله‌های اصیل پژوهشی را به صورت مقاله کامل (Full Paper) و مقاله کوتاه (Short Paper) پس از داوری دقیق به چاپ می‌رساند.

**پیش از ارسال مقاله، روش تدوین و نگارش مقاله خود را به دقت با مطالب زیر مطابقت فرمایید.**

### نکات قابل توجه

- ۱- در مقاله، قواعد دستور زبان فارسی و رسا بودن جملات مورد توجه ویژه قرار گیرد.
- ۲- مقالاتی که برای چاپ در این مجله ارسال می‌گردد نباید قبلاً چاپ شده باشد (مگر در شکل خلاصه در گردهمایی‌ها) همچنین نباید به طور همزمان برای چاپ به مجلات دیگر ارایه شده باشد.
- ۳- مسؤلیت مطالب مندرج در مقاله بر عهده نویسنده یا نویسندگان مقاله است.
- ۴- مجله در قبول، رد و اصلاح مقاله‌ها آزاد است.
- ۵- استفاده از مندرجات مجله با ذکر مأخذ آزاد است.
- ۶- مقاله‌های دریافتی توسط هیأت تحریریه با همکاری متخصصان امر داوری می‌گردد و در صورت تصویب با رعایت نوبت به چاپ می‌رسد. تصمیم نهایی برای چاپ مقاله توسط هیأت تحریریه صورت می‌گیرد.

### نحوه تدوین مقاله

- ۱- مقاله بایستی به زبان فارسی تهیه شود (به استثناء مقاله‌های پژوهشگران خارجی که باید به زبان انگلیسی باشد) و هر مقاله باید یک چکیده به زبان انگلیسی داشته باشد؛ این شرط تا زمانی که زبان مجله تغییر نکرده است پا برجا خواهد بود.
- ۲- هر مقاله علمی - پژوهشی بایستی به ترتیب دارای قسمت‌های: عنوان، مشخصات مؤلف یا مؤلفان و نشانی دقیق همراه با شماره تلفن و نشانی پست الکترونیک فرستنده (مسئول مکاتبات)، چکیده فارسی، واژه‌های کلیدی، مقدمه، مواد و روش‌ها، نتایج، بحث، جمع‌بندی، قدردانی، منابع، Abstract و Key words باشد.
- ۳- تایپ مقاله با نرم‌افزار Microsoft Office Word 2003، به صورت یک رو، در کاغذ A4، با حاشیه‌های متن ۳ سانتی‌متر و به صورت یک ستونی و با فاصله خطوط ۱ سانتی‌متر (single) انجام شود.
- ۴- مقاله نباید از ۱۵ صفحه چاپ شده در مجله (حدود ۶ هزار کلمه) تجاوز کند.
- ۵- از درج پاورقی برای بیان توضیحات انگلیسی و فارسی و برعکس خودداری شود و در صورت نیاز در درون پرانتز و در متن مقاله آورده شود.
- ۶- شکل‌ها و جدول‌ها شماره‌گذاری شده و به همراه زیرنویس آنها در متن مقاله آورده شود؛ در نرم‌افزار Word، فرمت شکل‌ها در بخش Text Wrapping، به صورت In line with text انتخاب شود. از ارسال شکل‌های گروه‌بندی شده (Group) اکیداً خودداری شود؛ نمودارها به صورت دو بعدی و سیاه و سفید طراحی شوند و الزاماً از حالت سه بعدی خارج شوند.

عنوان: شامل کوتاه ترین عبارتی خواهد بود که به طور کلی گویای محتوای مقاله باشد، خط فارسی عنوان 16 B Lotus Bold و انگلیسی 14 Times New Roman Bold است.

نام و نشانی نگارندگان: مسؤولیت ترتیب نام نگارندگان بر عهده نویسنده مسؤول خواهد بود. درج شماره مربوط به نشانی هر نگارنده پس از نام نگارنده به صورت بالا نویس (Superscript) است؛ علاوه بر درج شماره مربوط، یک ستاره برای نام نویسنده مسؤول (Corresponding Author) درج شود. نشانی ها به ترتیب و با خط 12 B Lotus Bold و 11 Times New Roman Bold در زیر نام نویسندگان ذکر می گردد. نشانی پست الکترونیک مسؤول مکاتبات با خط 10 Times New Roman Bold نوشته شود.

نمونه فارسی

معرفی گونه ای جدید در جنس *Centaurea* از ایران  
علیرضا اسدی<sup>۱\*</sup>، محمد کیانی<sup>۲</sup> و شهریار نظری<sup>۲</sup>  
<sup>۱\*</sup> دانشگاه اصفهان گروه زیست شناسی، <sup>۲</sup> مرکز تحقیقات زیستی  
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چکیده: خط 11 B Lotus و 10 Times New Roman شامل ۱۰۰ تا ۲۵۰ کلمه و بدون هر گونه کلمه اختصاری

واژه های کلیدی: حداکثر حاوی شش کلمه مرتب شده بر اساس حروف الفبا

مقدمه، مشاهدات، مواد و روش ها، نتایج، بحث و نتیجه گیری، قدردانی و منابع: 11 Times New Roman و 13 B Lotus

Abstract و Key words: 12 Times New Roman

عنوان جدول در بالای جدول و عنوان شکل (شامل نمودار، تصویر، دیاگرام، گراف، ...) در زیر آنها نوشته شود.

11 B Lotus و 9 Times New Roman

نمونه: شکل ۱-، شکل ۲-، جدول ۱-، جدول ۲-

نحوه مرجع دهی:

الف) مرجع دهی در متن (References in text): در متن به صورت نام نویسنده و یا نویسندگان (بدون نام کوچک) و سال انتشار نوشته شود.

نمونه فارسی: یک نویسنده: (بهارلو، ۱۳۸۸)، دو نویسنده: (قاسم زاده و اشتری، ۱۳۶۵)، سه نویسنده و بیشتر: (شریعت مدار و همکاران، ۱۳۷۶)

نمونه انگلیسی: یک نویسنده: (Davis, 1985)، دو نویسنده: (Dagan and Zohary, 1970)، سه نویسنده و بیشتر: (Johnson et al., 2000)

کلمه et al., بایستی به صورت مورب باشد (این کلمه لاتین است).

ب) مرجع دهی در بخش منابع (References list): فهرست منابع بایستی به ترتیب حروف الفبا مرتب شده ابتدا منابع فارسی و سپس منابع خارجی آورده شود.

ب-۱) مرجع دهی به مقاله (Paper): به ترتیب شامل: نام نویسنده یا نویسندگان، سال، عنوان، نام کامل مجله، شماره مجلد، شماره صفحات.

ب-۱-۱) مقاله با یک نگارنده

نمونه فارسی: بحرانی، ص. (۱۳۷۵) بررسی گوناگونی ژنتیکی در گونه های وحشی (*T. urartu* and *T. boeoticum*) با استفاده از الکتروفورز پروتئین بذر. مجله بذر و نهال ۲: ۱-۹.

نمونه انگلیسی:

- Noda, K. (1981) C-banding technique for Wheat chromosomes. Wheat Information Service 52(8): 29-31.

ب-۱-۲) مقاله با دو نگارنده:

نمونه فارسی: ولی پور، ع. و حسینی، ا. (۱۳۷۶) بررسی پراکنش گیاهان مقاوم به شوری در ایران. مجله زیست‌شناسی ۳(۵): ۷۵ - ۹۱.

نمونه مثالی انگلیسی:

- Baum, B. R. and Appels, R. (1992) Evolutionary change at the 5s DNA loci of species in the Triticaceae. *Plant Systematics and Evolution* 183: 195-208.

ب-۱-۳) مقاله با سه نگارنده و بیشتر:

نمونه فارسی: ولی پور، ع.، حسینی، ا. و امینی، ا. ر. (۱۳۷۶) بررسی پراکنش گیاهان مقاوم به شوری در ایران. مجله زیست‌شناسی ۳: ۷۵ - ۹۱.

نمونه انگلیسی:

- Jain, S. K., Qualset, C. O., Bhatt, G. M. and Wu, K. K. (1975) Geographical patterns of phenotypic diversity in a world collection of durum wheats. *Crop Science* 15: 404-700.

ب-۲) مرجع دهی به کتاب (Book): به ترتیب شامل: نام نویسنده یا نویسندگان، سال، عنوان کتاب، شماره Edition

در صورت وجود، نام مؤسسه انتشاراتی، نام اولین شهری که انتشار در آن انجام گرفته است.

نمونه فارسی: مظفریان، و. (۱۳۷۳) کورموفیت‌های ایران. جلد ۴، مرکز نشر دانشگاهی، تهران.

نمونه انگلیسی:

- Stace, C. A. (1989) *Plant Taxonomy and Biosystematics*. Edward Arnold, London.
- Rice, E. L. (1984) *An Introduction to Microbiology*. 2<sup>nd</sup> ed., Academic Press, New York.

مرجع دهی به ترجمه فارسی کتاب:

استیس، سی. ای. (۱۳۷۵) تاکسونومی گیاهی و سیستماتیک زیستی. ترجمه خسروی، الف. انتشارات دانشگاه شیراز، شیراز.

ب-۳) مرجع دهی به بخشی از کتاب (Chapter in Book) که هر بخش دارای نویسنده جداگانه باشد:

نمونه انگلیسی:

- Morrison, L. A. (1993) *Triticum-Aegilops* systematics: taking an integrative approach. In: *Biodiversity and Wheat Improvement* (ed. Damania, A. B.) 59-66. John Wiley & Sons, New York.
- Sears, E. R. (1956) The systematic, cytology and genetics of wheat. In: *Handbuch der Pflanzenzucht*. (eds. Kapparet, H. and Rudorf, W.) 2: 164-187. Paul Parey, Berlin and Humburg.

ب-۴) مرجع دهی به پایان‌نامه کارشناسی ارشد یا دکترا: نام نویسنده، سال، عنوان پایان‌نامه، مقطع تحصیلی، نام دانشگاه، نام شهر، نام کشور.

نمونه فارسی: حسین پور، م. (۱۳۶۵) تاکسونومی و بیوسستماتیک جنس *Cardaria* L. در ایران. رساله دکتری، دانشگاه اصفهان، اصفهان، ایران.

نمونه مثالی انگلیسی:

- Hassanpour, S. M. (2006) Study of Biosystematic of the genus *Rhamnus*. Ms.c. Thesis, University of Isfahan, Isfahan, Iran.

ب-۵) مرجع دهی به Patent:

- Suzuki, T., Ohishi, N. and Yagi, K. (2000) Methods of obtaining a composition 9-cis  $\beta$ -Carotene in high purity. US Patent 6057484.

ب-۶) مرجع دهی به همایش‌ها (سمینارها، سمپوزیوم‌ها، کنگره‌ها، میتینگ‌ها و ...): به ترتیب شامل: نام نویسندگان، سال انتشار، عنوان مقاله، دوره و نام همایش، محل برگزاری، شهر، کشور.

نمونه فارسی: رنگی پور، ا.، افشارزاده، س.، بلالی‌دهکردی، غ. ر. و صاحبی، ج. (۱۳۸۷) مطالعه جنس لویی در رودخانه زاینده‌رود. اولین همایش ملی زیست‌شناسی گیاهی، دانشگاه پیام نور، تالش، ایران.

نمونه انگلیسی:

- Mason-Gamer, R. J. and Helfgott, D. M. (2002) Molecular phylogenetic investigation of allopolyploid *Elymus* in North America. 4<sup>th</sup> International Triticeae Symposium, Prague, Czech Republic.
- ب-7) مرجع دهی به مقاله های کامل همایش ها (سمینارها، سمپوزیومها، کنگره ها، میتینگ ها و ...) (Proceedings):  
به ترتیب شامل: نام نویسندگان، سال انتشار، عنوان مقاله، دوره و نام همایش، محل برگزاری، شهر، کشور.  
نمونه فارسی: صفوی، و. و شریعتی، م. (۱۳۸۶) تأثیر الیستور متیل جاسمونات بر ستر بتاکاروتن در جلبک سبز تک سلولی *Dunaliella salina*. مجموعه مقالات دومین همایش ملی زیست شناسی سلولی و ملکولی، کرمان، ایران.  
نمونه انگلیسی:
- Mohsenzadeh, S. (1996) Study of nitrogen fertilizer time and amount on seed production and other characterizations of Sorghum. In: Proceeding of the 4<sup>th</sup> Iranian Congress of Agriculture and Plant Breeding, Isfahan, Iran.
  - Shariati, M. and Lilley, R. McC. (1993) Triggering of glycerol synthesis in *Dunaliella tertiolecta* at constant osmotic pressure. 33<sup>rd</sup> Annual General Meeting of Australian Society of Plant Physiologist. Perth, Australia.
- ب-8) مرجع دهی به اینترنت: مرجع دهی به نشانی های اینترنتی تقریباً فاقد اعتبار بوده و پیشنهاد می شود استفاده نگردد.  
در مواقعی که ناگزیر از استفاده محدود از آن باشد نام نویسنده، زمان چاپ و در انتها نیز زمان استخراج از اینترنت درج گردد.  
نمونه:
- Rotblat, J. (2000) Fifty Pugwash conferences: a tribute to Eugene Rabinowitch. Retrieved from <http://www.pugwash.org/reports/pac/pac256/otblat.htm>. On: 22 June 2001.
- پ) شکل ها و جدول ها: شکل ها و جدول ها به ترتیب ذکر شده درون متن قرار بگیرند، توضیحات شکل ها در پایین و توضیحات جدول ها در بالای آنها نوشته شود.

درستی نام علمی گونه های گیاهی از لحاظ صفت گونه ای و نام آتور در پایگاه جهانی فهرست نام های گیاهی

[www.ipni.org](http://www.ipni.org) بررسی شود.

نحوه ارسال مقاله

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## مجله علمی - پژوهشی تاکسونومی و بیوسیستماتیک

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آمارجیت سینگ، سودان، آشوک کومار و سوشانت شارما



## تشریح مقایسه‌ای برخی از گونه‌های تیره خشخاش (Papaveraceae) در ایران

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#### چکیده

در مطالعه حاضر، بررسی مقایسه‌ای صفات تشریحی ساقه، رگبرگ اصلی برگ‌های قاعده‌ای و میوه در خانواده Papaveraceae (به جز جنس *Papaver*) در ایران ارائه شده است. در این پژوهش، ۴۵ صفت تشریحی مربوط به ساقه، رگبرگ اصلی و میوه در گونه‌های *G. elegans*, *G. haussknechtii*, *G. corniculatum*, *Glaucium grandiflorum*, *Chelidonium majus* مورد بررسی قرار گرفته است همچنین، وجود کرک غده‌ای در *G. pulchrum* و *G. oxylobum* برای نخستین بار گزارش شد. از بین صفات بررسی شده، چندین صفت از جمله: شکل جفت، زاویه برچه‌ای، شکل زاویه برچه‌ای، تعداد لایه‌های پارانشیم داخلی و خارجی دیواره تخمدان، تعداد دستجات آوندی جفت در میوه، وجود حفره مرکزی در میوه، وجود کرک‌های غده‌ای، تعداد دستجات آوندی، تعداد دستجات بافت آبکش، عدم وجود بافت استحکامی در اطراف دستجات آوندی در رگبرگ اصلی برخی گونه‌ها متفاوت است، که می‌تواند در جداسازی گونه‌های جنس *Glaucium* مفید باشد. صفات تشریحی رگبرگ اصلی پهنک در این جنس‌ها، به رغم برخی از تفاوت‌ها، اساساً ساختار مشابهی دارد. بنابراین، صفات تشریحی رگبرگ به تنهایی نمی‌تواند معیار مناسبی برای شناسایی و تشخیص گونه‌های جنس *Glaucium* و چهار جنس دیگر باشد. همچنین، صفات تشریحی ساقه، معیار مناسبی برای تشخیص و جداسازی گونه‌های جنس *Glaucium* و سایر جنس‌ها نیست. از سوی دیگر، صفات تشریحی میوه در شناسایی و جدا کردن گونه‌های جنس *Glaucium* و همچنین چهار جنس دیگر مفید است به ویژه، برش عرضی میوه *H. pendulum* اختلاف قابل توجهی را در مقایسه با دیگر جنس‌ها نشان می‌دهد. برخی از صفات تشریحی میوه در جنس *Glaucium* می‌تواند در گروه‌بندی تاکسونومیک این جنس مفید باشد. مهم‌ترین این صفات، شکل جفت تخمدان است.

**واژه‌های کلیدی:** آناتومی، *Papaveraceae*, *Roemeria*, *Heypocoum*, *Glaucium*, *Chelidonium*، ایران

## تنوع زیستی گیاهی پنج مانداب مهم شهرستان بابل، استان مازندران

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### چکیده

این پژوهش، شامل مطالعه فلور و بررسی تنوع زیستی پنج مانداب مهم مرزون آباد، لنگور، بصرآ، رمنت و آغوزین در شهرستان بابل در شمال ایران است. برای تحقق مطالعات فلورزیستی در این مانداب‌ها، همه گیاهان آوندی در طی دو فصل رویشی (۱۳۸۹-۱۳۹۰) جمع‌آوری گردید. تعداد کل ۱۹۶ گونه متعلق به ۱۳۸ جنس و ۵۸ خانواده گیاهی شناسایی شد. تیره‌های Poaceae (۲۴ گونه)، Cyperaceae (۱۹ گونه)، Asteraceae (۱۶ گونه)، Fabaceae (۱۳ گونه) و Polygonaceae (۹ گونه) دارای بیشترین غنای گونه در منطقه مورد مطالعه بودند. جنس‌هایی که بیشترین تعداد گونه را در بردارند عبارتند از: *Cyperus* (۸ گونه)، *Polygonum* (۷ گونه) و *Potamogeton* (۴ گونه). در بین گیاهان منطقه، تروفیت‌ها با ۳۷ درصد فراوان‌ترین شکل زیستی منطقه را تشکیل می‌دهند. از لحاظ پراکنش جغرافیایی، بیشترین گونه‌ها متعلق به عناصر چند ناحیه‌ای با ۵۴/۵ درصد هستند. زیستگاه‌های مختلف این مانداب‌ها ارزیابی شد. از میان پنج مانداب شهرستان بابل، مانداب مرزون آباد بیشترین تعداد گونه‌ها (۱۱۱) را به خود اختصاص داده است و مانداب لنگور با ۶۳ گونه در رتبه دوم قرار دارد. به علاوه، مقایسه‌ای بین اطلاعات به دست آمده از این مانداب‌ها با سایر مانداب‌های شمال ایران نیز انجام گردید که نشان‌دهنده برخی شباهت‌ها و اختلافات بین مناطق مختلف مورد بررسی است. طبق ضریب تشابه سورنسون مشخص شد که شباهت اندکی بین گونه‌های گیاهی این پنج مانداب با یکدیگر وجود دارد که دلیل آن مساحت متفاوت و فاصله نسبتاً زیاد میان آنهاست.

**واژه‌های کلیدی:** استان مازندران، پراکنش جغرافیایی، شکل زیستی، فلور، گیاهان آبی، مانداب‌های بابل

## بررسی بیوسیستماتیک گونه‌های *Phalaris L.* (خانواده غلات) در ایران

مریم کشاورزی\*، مهناز خاکسار و پریناز قدم

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### چکیده

الگوی باندینگ پروتئین ذخیره‌ای بذر بر اساس الکتروفورز SDS-PAGE با استخراج عصاره پروتئینی گونه‌های *Phalaris* در ایران بررسی شد. نتایج نشان داد که دو وارته *P. Paradoxa L.* یعنی *Praemorsa Paradoxa* و *Paradoxa* ارتباط بسیار نزدیکی دارند. ارتباط بسیار نزدیکی نیز بر مبنای ضریب شباهت بالای پروتئینی ( $I=0/385$ ) بین دو گونه *P. Arundinacea L.* و *P. brachystachys Link.* مشاهده شد. نتایج الکتروفورزی با نتایج بررسی‌های پیشین در مورد ساختمان تشریحی و تنوع ریختی مقایسه شده است.

واژه‌های کلیدی: *Phalaris*، روابط تاکسون‌ها، SDS-PAGE

## معرفی فلور، شکل زیستی و پراکنش جغرافیایی گیاهان جنگل‌های پست نور و سیسنگان

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### چکیده

مناطق پست هیرکانی (خزری) شامل لکه‌های به جامانده از جنگل‌های خزان‌کننده اروپا-سیبری است که در سه استان گیلان، مازندران و گلستان پراکنده است. نور و سیسنگان دو تکه بزرگ از این جنگل‌های پست هستند که با عنوان "پارک جنگلی" در مفهوم "منابع طبیعی ایران" طبقه‌بندی شده‌اند. با وجود برخی مطالعات محلی بر روی این جنگل‌ها، هنوز دانش کافی در مورد فلور و پوشش گیاهی این مناطق وجود ندارد. گونه‌های گیاهی جمع‌آوری شده از این مناطق نشان‌دهنده وجود ۲۲۵ گونه گیاهی متعلق به ۱۷۵ جنس و ۷۷ تیره گیاهی است. Poaceae با ۲۸، Asteraceae با ۱۸ و Rosaceae با ۹ گونه، به ترتیب بیشترین غنای گونه‌ای را نشان می‌دهند. جنس‌های دارای بیشترین تعداد گونه به ترتیب *Carex* (با ۶ گونه)، *Veronica* (با ۵ گونه) و *Euphorbia*، *Polygonum* و *Solanum* (هر کدام با ۴ گونه) هستند. به لحاظ طیف شکل زیستی، تروفیت‌ها با ۳۰/۲٪ اشکال زیستی غالب را تشکیل می‌دهند و به دنبال آن، ژئوفیت‌ها (۲۷/۱٪)، همی کریتوفیت‌ها (۲۰/۹٪) و فانروفیت‌ها (۱۸/۲٪) قرار دارند. فلور این مناطق، عمدتاً از عناصر چندناحیه‌ای با ۶۰ تاکسون (۲۷/۳٪) و سپس عناصر اروپا-سیبری/ایرانی-تورانی/مدیترانه‌ای با ۴۳ تاکسون (۱۹/۵٪) تشکیل شده است. درصد هر کدام از عناصر جغرافیایی و اشکال زیستی به طور اختصاصی برای هر جنگل ارائه می‌شود. بر اساس شاخص تشابه سورنسن، برخی شباهت‌های فلورستیکی بین دو جنگل وجود دارد. جنگل‌های پست نور و سیسنگان، به علت فشار فعالیت‌های انسانی و چرای دام، در معرض خطر حذف گونه‌های گیاهی و یا تغییر جوامع طبیعی هستند.

واژه‌های کلیدی: فلور، جنگل پست هیرکانی، شکل زیستی، نور و سیسنگان

## مطالعه سیتوتاکسونومی چهار جمعیت *Astragalus anserinifolius* Boiss. بخش *Malacothrix* Bunge از ایران

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### چکیده

رفتار و تعداد کروموزوم‌های میوزی چهار جمعیت ایرانی گونه *Astragalus anserinifolius* Boiss. از جنس گون، بخش *Malacothrix* مورد مطالعه قرار گرفت. تمامی این جمعیت‌های خودرو دیپلوئید بوده، عدد کروموزومی  $2n=2x=16$  را نشان دادند که مطابق با عدد پایه پیشنهادی  $x=8$  از ICPN است. گرچه در تمامی تاکسون‌ها جفت شدن کروموزوم‌ها و جدا شدن آنها در مرحله میوز منظم بود، لیکن بی‌نظمی‌های میوز شامل درجات متفاوتی از کروموزوم‌های جدا افتاده و چسبندگی کروموزوم‌ها در متافاز I، چند هسته‌ای و شمار متفاوتی از کروموزوم‌های تأخیری، پیشرو و پل در آنافاز I/تلوفاز I، ناهمزمانی هسته‌ها و مهاجرت زود هنگام کروموزوم‌ها در متافاز II و تأخیر و تشکیل پل‌ها و سیتومیکسی در آنافاز II/تلوفاز II مشاهده شد.

**واژه‌های کلیدی:** بخش *Malacothrix*، *Astragalus anserinifolius*، عدد کروموزومی، بی‌نظمی‌های میوز

## مطالعه روابط فنتیک میان جمعیت‌های طبیعی گیاه شیرین بیان (تیره باقلاییان) در ناحیه زاگرس مرکزی ایران بر اساس داده‌های ریخت‌شناسی کمی، ترکیبات فلاونوئیدی و محتوای گلیسیریزین

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### چکیده

روابط فنتیک میان ۳۵ نمونه جمعیتی جمع‌آوری شده از جمعیت‌های طبیعی دو وارسته از گونه *Glycyrrhiza glabra* در ناحیه زاگرس مرکزی ایران مورد مطالعه قرار گرفته است. ۲۱ صفت ریخت‌شناسی کمی روی ۲۷ نمونه جمعیتی اندازه‌گیری شد و آنالیزهای مختصات اصلی، خوشه‌بندی، K-means و MDS روی داده‌های ریخت‌شناسی صورت گرفت. محتوای فلاونوئیدی ۲۴ نمونه جمعیتی استخراج و با روش کروماتوگرافی لایه نازک خالص سازی و بررسی در سطح تعیین اسکلت و کلاس فلاونوئیدی صورت گرفت. محتوای گلیسیریزین ریزوم‌ها با استفاده از روش پردازش تصویر در ۲۴ نمونه به صورت کمی اندازه‌گیری شد. نتایج آنالیزهای چند متغیره روی داده‌های ریخت‌شناسی و داده‌های پروفایل لکه‌های فلاونوئیدی نشان داد که می‌توان نمونه‌های جمعیتی مطالعه شده را بر اساس موقعیت جغرافیایی محل جمع‌آوری نمونه‌ها به دو گروه عمده تقسیم نمود. اندازه سطح بذر، متغیرترین صفت ریخت‌شناسی کمی بر اساس مقادیر CV تشخیص داده شد و عرض نیام در عریض‌ترین بخش، کمترین تغییرات را نشان داد. نمونه‌های هر دو وارسته، فلاونوئیدهای متنوعی را از کلاس‌های فلاون و فلاونول تولید کرده بودند. در مجموع، تعداد ۷ ترکیب فلاونوئیدی مختلف با Rf‌های متمایز از دو وارسته بدست آمد. نتایج، سطوح متوسطی از تنوع میان نمونه‌های مورد مطالعه را نشان داد. نتایج این مطالعه، جداسازی دو وارسته در منابع موجود بر اساس یک تک صفت کیفی را تأیید می‌کند. ریزوم‌های هر دو وارسته، مقادیر مشابهی از گلیسیریزین و انواع مشابهی از فلاونوئیدها را در پروفایل کروماتوگرام‌های لایه نازک ارائه نمودند که اهمیت و کاربرد یکسان ریزوم‌های هر دو وارسته را از نظر کاربرد به عنوان دارویی گیاهی در طب سنتی نشان می‌دهد.

**واژه‌های کلیدی:** ایران، زاگرس، شیرین بیان، فلاونوئید، گلیسیریزین، ریخت‌شناسی



## گزارش به روز شده‌ای از گندمیان پنجاب (هندوستان)

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### چکیده

تیره Poaceae چهارمین تیره بزرگ گیاهان گل‌دار است. این تیره دربردارنده ۷۰۰-۸۰۰ جنس و ۱۱۰۰۰-۱۳۰۰۰ گونه با پراکنش جهانی است. این خانواده، از اهمیت بوم‌شناختی و اقتصادی بی‌نظیری برخوردار است. پیدایش این تیره به دوره کرتاسه نخستین و گونه‌گونی عمده آن به دوره سنوزوئیک میانی بازمی‌گردد. در حال حاضر، این تیره نزدیک به یک پنجم سطح خشکی‌ها را پوشش می‌دهد و تقریباً در تمامی زیستگاه‌ها در سطح جهان وجود دارد. با وجود شرایط بوم‌شناختی نیمه گرمسیری و موقعیت اقتصادی منطقه پنجاب، گندمیان بخش قابل ملاحظه‌ای را در منطقه تشکیل می‌دهند. با وجود اهمیت قریب به اتفاق، مطالعات تاکسونومیک در مورد گندمیان در این منطقه از توجه کافی برخوردار نبوده است. تنها در مطالعات Sharma و Khosla (۱۹۸۹) است که گونه‌های گندمیان به زیرتیره‌ها و طایفه‌ها طبقه‌بندی شده‌اند. با این حال، پس از تأسیس کارگروه تبارشناسی گندمیان (GPWG) جهان تحولی نوین در زمینه طبقه‌بندی گندمیان داشته است. اما کشور هند تا آنجا که به تنوع گندمیان مربوط می‌شود هنوز به طور کامل مورد توجه قرار نگرفته است. تحقیقات ما برای اکتشاف و رده‌بندی فلور تیره گندمیان منطقه، تلاشی برای تقویت و به روزرسانی اطلاعات درباره تنوع آن در منطقه مورد مطالعه است. مطالعات حاضر، جمع‌بندی ۱۹۲ مجموعه گونه از جمله ۷ گزارش جدید را ارائه کرده است. نمونه‌های گونه‌ای از زیرتیره‌ها عبارتند از: Aristidoideae (۵)، Arundinoideae (۵)، Bambusoideae (۴)، Chloridoideae (۵۵)، Centothecoideae (۱)، Erhartoideae (۳)، Panicoideae (۹۸) و Pooideae (۲۱).

**واژه‌های کلیدی:** هندوستان، تیره گندمیان، پنجاب، رده‌بندی، تاکسونومی، گرمسیری



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## Conclusion

The reclassification of grasses of the region proposed in the present paper shall provide new direction to future studies in the group. This is in consonance with a world wide renewal of interest in systematics and evolution of this group. Apart from their conventional uses, grasses have emerged as model plant species for events in plant development and as indicators of environmental changes. Intensive explorations need to be carried out to identify candidate grass species for these areas in the modern biology. The usefulness of intensive explorations is indicated by the fact that the seven new species have been identified in the present study.

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Taxon	1	2	3	4	5	6
	Stewart (1869)	Sabnis (1940)	Nair (1978)	Sharma and Bir (1978)	Sharma and Khosla (1989)	Present Work
<i>C. citratus</i> (DC.) Stapf. (1906) 357	-	-	-	-	-	+*
<i>C. jwarancusa</i> (Jones) Schult. (1824) 458	+	+	+	-	+	+
[Syn. <i>Andropogon schoenanthus</i> (L.) Spreng. (1896) 204. (2)]						
<i>C. martinii</i> (Roxb.) Watson (1882) 392	-	-	+*	-	+	+
<i>C. nardus</i> (L.) Rendle (1899) 155	-	-	-	-	+*	+
<i>Dichanthium annulatum</i> (Forssk.) Stapf (1917) 178	+	+	+	+	+	+
[Syn. <i>Andropogon annulatus</i> Forssk. (1775) 173 (1)]						
<i>D. caricosum</i> L. A. Camus (1921) 459	-	-	-	-	+*	+
<i>D. foveolatum</i> (Delile) Roberty (1960) 170	-	+*	-	-	+	+
[Syn. <i>Andropogon foveolatus</i> Delile (1812) 16 t. 82. (2)]						
<i>Eulaliopsis binata</i> (Retz.) C. E. Hubb. (1935) 3262	-	+*	-	-	+	+
[Syn. <i>Ischaemum angustifolium</i> Hook. (1889) 241 (2)]						
<i>Hackelochloa granularis</i> (L.) O. Kuntze. (1891) 776	-	-	-	-	+*	+
<i>Hemarthria compressa</i> (L. f.) R. Br. (1810) 207	-	+*	+	+	+	+
[Syn. <i>Rottboellia compressa</i> L. (1781) 114 (2)]						
<i>Heteropogon contortus</i> (L.) P. Beauv. ex Roem. & Schult. (1817) 836	+	+	+	+	+	+
[Syn. <i>Andropogon contortus</i> L. (1753) 1045 (2)]						
<i>Imperata cylindrica</i> (L.) P. Beauv. (1812) 165	-	+*	+	+	+	+
[Syn. <i>I. arundinacea</i> Cyprill. (1792) 27, t. 11 (2)]						
<i>Ischaemum rugosum</i> Salisb. (1791) 1	-	-	-	-	+*	+
<i>Iseilema prostratum</i> (L.) Andersson (1856) 251	-	+*	+	-	+	+
[Syn. <i>I. wightii</i> Andersson (1856) 251 (2)]						
[ <i>Andropogon prostratus</i> L. (1856) 251 (3)]						
<i>Lasiurus indicus</i> (Boiss.) Henr. (1941) 414	-	+*	+	-	+	+
[Syn. <i>Elionurus hirsutus</i> auct. (1881) 68 (2)]						
<i>Pogonatherum crinitum</i> (Thunb.) Kunth. (1906) 178	-	-	-	-	-	+*
<i>P. Paniceum</i> (Lam.) Hack. (1906) 178	-	+*	+	-	+	+
[Syn. <i>P. saccharoideum</i> P. Beauv. (1812) 176, 177 (2)]						
<i>Rottboellia cochinchinensis</i> (Lour.) Clayton (1981) 817	-	-	-	-	+*	+
[ <i>R. exaltata</i> L. f. (1779) 40 (5)]						
<i>Mnesithea laevis</i> (Retz.) Kunth (1829) 154	-	+*	-	-	-	[-]
[Syn. <i>Rottboellia perforata</i> Roxb. (2)]						
<i>Saccharum bengalense</i> Retz. (1789) 16	+	+	+	+	+	+
[Syn. <i>S. sara</i> Roxb. (1820) 249 (1)]						
[ <i>S. arundinaceum</i> Hook. f. (1786-1787) 14 (2)]						
<i>S. officinarum</i> L. (1753) 54	+	-	+	+	+	+
<i>S. ravennae</i> L. (1774) 88	-	+*	+	+	+	+
[Syn. <i>Erianthus ravennae</i> Beauv. (1812) 162, 177 (2)]						
<i>S. spontaneum</i> L. (1771) 183	+	+	+	+	+	+
<i>Sorghum bicolor</i> (L.) Moench (1794) 207	+	-	+	+	+	+
[Syn. <i>S. vulgare</i> Pers. (1805) 101 (1)]						
[ <i>S. cernuum</i> (Ard.) Host (1809) 2 (3)]						
<i>S. halepense</i> (L.) Pers. (1805) 101	+	+	+	+	+	+
[ <i>Andropogon halpensis</i> Brot. (1804) 89 (2)]						
<i>Themeda anathera</i> (Nees ex Steud.) Hack. (1889) 669	-	-	-	-	-	+*
<i>T. quadrivalvis</i> (L.) O. Kuntze. (1891) 794	-	-	-	+*	+	+
[Syn. <i>Anthistiria ciliata</i> L. (1781-1782) 113 (1)]						
<i>Vetiveria zizanioides</i> (L.) Nash (1903) 67	+	+	+	+	+	+
[Syn. <i>Anatherum muricatum</i> Beauv. (1812) 150 (1)]						
[ <i>Andropogon squarrosus</i> of Hook. f. (1896) 186 (2)]						
<i>Zea mays</i> L. (1753) 971	+	-	+	+	+	+
<b>Species Number (Cumulative)</b>	<b>40</b>	<b>74</b> <b>(93)</b>	<b>105</b> <b>(133)</b>	<b>73 (147)</b>	<b>146 (185)</b>	<b>187</b> <b>(192)</b>

Taxon	1	2	3	4	5	6
	Stewart (1869)	Sabnis (1940)	Nair (1978)	Sharma and Bir (1978)	Sharma and Khosla (1989)	Present Work
<i>P. maximum</i> Jacq. (1781) 2, 13	-	-	+	+	+	+
<i>P. miliaceum</i> L. (1753) 58	+	-	+	-	+	+
<i>P. paludosum</i> Roxb. (1820) 310	-	-	-	-	+	+
<i>P. virgatum</i> Roxb. ex Steud. (1841) 262	-	-	+	-	+	+
[Syn. <i>P. psilopodium</i> Trin. (1826) 217 (5)]						
<i>P. repens</i> L. (1762) 86	-	-	-	-	+	+
<i>P. tenellum</i> Roxb. (1854) 21	-	-	+	-	+	+
[Syn. <i>P. trypheron</i> Schult. (1824) 244 (3)]						
<i>Paspalidium flavidum</i> (Retz.) A. Camus (1922) 419	-	-	+	+	+	+
<i>Paspalum dilatatum</i> Poir. (1804) 35	-	-	-	-	+	+
<i>P. longifolium</i> Roxb. (1820) 283	-	+	+	-	-	+
[Syn. <i>P. longiflorum</i> Retz. (1786) 15 (2)]						
<i>P. notatum</i> Fluegge. (1810) 106	-	-	-	-	+	+
<i>P. scrobiculatum</i> L. (1767) 29	+	-	+	-	+	+
<i>P. vaginatum</i> Sw. (1788) 21	-	+	+	+	+	+
[Syn. <i>P. sanguinale</i> L. (1896) 15 (2)]						
[ <i>P. paspaloides</i> (Michx.) Scribn. (1894) 29 (4)]						
<i>Pennisetum glaucum</i> (L.) R. Br. (1810) 195	+	+	+	+	+	+
[Syn. <i>Penicillaria spicata</i> Willd. (1809) 1037 (1)]						
[ <i>P. typhoides</i> (Burm.) Stapf et C.E. Hubb. (1933) 271 (2)]						
<i>P. orientale</i> (L.) C. Rich. (1805) 72	-	+	+	-	+	+
<i>P. polystachyon</i> (L.) Schult. (1824) 146	-	-	-	-	+	+
<i>P. purpureum</i> Schumach. (1827) 44	-	-	-	+	+	+
<i>Setaria intermedia</i> Roem. & Schultz. (1817) 489	-	-	-	+	+	+
[Syn. <i>S. tomentosa</i> (Roxb.) Kunth (1829) 47 (4)]						
<i>S. italica</i> (L.) P. Beauv. (1812) 170, 178	-	-	-	-	+	+
[Syn. <i>Panicum italicum</i> R.Br. (1753) 56 (2)]						
<i>S. pumila</i> (Poir.) Roem. & Schult. (1817) 891	-	+	+	-	+	+
[Syn. <i>S. glauca</i> (L.) P. Beauv. (1812) 168, 169. (3)]						
<i>S. sphacelata</i> (Schumach.) Stapf et C.E. Hubb. (1929) 195	-	-	-	-	+	+
<i>S. verticillata</i> (L.) P. Beauv. (1812) 178	-	-	+	+	+	+
[Syn. <i>Panicum verticillatum</i> L. (1762) (3)]						
<i>Urochloa panicoides</i> P. Beauv. (1812) 53	-	-	+	+	+	+
<b>Tribe: Arundinelleae</b>						
<i>Arundinella nepalensis</i> Trin. (1826) 62	-	-	-	-	+	+
<b>Tribe: Andropogoneae</b>						
<i>Andropogon glomeratus</i> (Walter) Britton (1888) 67	+	-	-	-	-	+
[Syn. <i>Chrysopogon glaucoptis</i> Stend (1934) 139 (1)]						
<i>A. pumilus</i> Roxb. (1894) 496	-	-	+	-	-	+
<i>Apluda mutica</i> L. (1753) 82	-	-	+	-	+	+
[Syn. <i>A. varia</i> Hack. Subspecies <i>aristata</i> Hack. (1889) (199) (2)]						
[ <i>A. aristata</i> L. (1756) 303 (5)]						
<i>Arthraxon lancifolius</i> (Trin.) Hochst. (1856) 188	-	+	-	-	+	+
[Syn. <i>Andropogon monticola</i> Schult. (1827) 665 (2)]						
<i>A. prionooides</i> Steud. (1956) 399	-	-	-	-	+	+
<i>Bothriochloa insculpta</i> (Hochst.) A. Camus (1931) 165	-	-	-	-	+	+
<i>B. bladhii</i> (Retz.) S.T. Blake. (1969) 62	-	-	-	-	+	+
[ <i>B. odorata</i> (Lisboa) A. Camus (1931) 165 (5)]						
<i>B. ischaemum</i> L. (1936) 201	-	-	+	-	-	+
<i>B. pertusa</i> (L.) A. Camus (1931) 164	-	-	+	+	+	+
<i>Capillipedium huegelii</i> (Hack.) A. Camus (1921) 308	-	-	-	-	+	+
<i>C. parviflorum</i> (R.Br.) Stapf (1917) 169	-	-	-	-	+	+
<i>Chrysopogon serrulatus</i> Trin. (1832) 318	-	+	+	-	+	+
[Syn. <i>Andropogon monticola</i> Schult. (1896) 193 (2)]						
[ <i>C. fulvus</i> (Spreng.) Chiov. (1929) 327 (3)]						
<i>Coix lacryma-jobi</i> L. (1753) 972	-	-	-	-	+	+
<i>Cymbopogon commutatus</i> (Steud.) Stapf (1907) 211	-	-	+	-	+	+
[Syn. <i>C. parkeri</i> Stapf. (1929) 10 (3)]						

Taxon	1	2	3	4	5	6
	Stewart (1869)	Sabnis (1940)	Nair (1978)	Sharma and Bir (1978)	Sharma and Khosla (1989)	Present Work
<i>Sporobolus coromendelianus</i> (Retz.) Kunth ((1829) 68	-	+*	-	+	+	+
<i>S. diandrus</i> (Retz.) P. Beauv. (1812) 26, 147, 178	-	+*	+	+	+	+
<i>S. fertilis</i> Steud. (1965) 291	-	+*	-	-	+	+
[Syn. <i>S. indicus</i> (L.) R. Br. (1810) 170 (2)]	-	+*	-	-	+	+
<i>S. helvolus</i> (Trin.) T. Durand. & Schinz (1895) 820	-	+*	+	-	+	+
[Syn. <i>S. glaucifolius</i> (Hochst. ex Steud.) Hochst. ex T. Durand & Schinz (1854) 154 (2)]	-	+*	+	-	+	+
<i>S. ioclados</i> (Nees ex Trin.) Nees (1841) 161	-	-	+*	+	+	+
[Syn. <i>S. marginatus</i> Hochst. ex A. Rich. (1850) 397 (3)]	-	-	+*	+	+	+
<i>S. tenuissimum</i> (Mart. ex Schrank) Kuntze (1898) 369	-	+	+	-	-	+
[Syn. <i>S. minutiflorus</i> (Trin.) Link (1827) 88 (2)]	-	+	+	-	-	+
<b>Subfamily : Panicoideae</b>						
<b>Tribe: Isachneae</b>						
<i>Isachne albens</i> Trin. (1828) 8, 85	-	-	+*	-	-	[-]
<i>I. himalaica</i> Hook. f. 1897(1896) 23	-	+*	-	-	-	+
[Syn. <i>I. australis</i> R. Br. (1810) 196 (2)]	-	+*	-	-	-	+
<b>Tribe: Paniceae</b>						
<i>Alloteropsis cimicina</i> (L.) Stapf (1919) 487	-	-	-	-	+*	+
<i>Brachiaria brizantha</i> (Hochst. ex A. Rich.) Stapf (1919) 531	-	-	-	-	+*	+
<i>B. distachya</i> (L.) Stapf (1919) 565	-	-	-	-	+*	+
<i>B. mutica</i> (Forssk.) Stapf (1919) 526	-	-	-	-	+*	+
<i>B. ramosa</i> (L.) Stapf (1919) 542-544	-	+*	+	+	+*	+
[Syn. <i>Panicum ramosum</i> L. (1767) 29-30 (2)]	-	+*	+	+	+*	+
<i>B. reptans</i> (L.) C. Gardner & C.E. Hubb. (1938) pl 3363, f. 3	-	+*	+	-	+	+
[Syn. <i>Panicum prostratum</i> Lam. (1791) 171 (2)]	-	+*	+	-	+	+
<i>Cenchrus biflorus</i> Roxb. (1820) 238	-	+*	+	+	+	+
[Syn. <i>C. catharticus</i> Delile. (1839) 4 (2)]	-	+*	+	+	+	+
<i>C. ciliaris</i> L. (1771) 302	+	+	+	+	+	+
[Syn. <i>Pennisetum cenchroides</i> Rich. ex pers. (1805) 72 (1)]	+	+	+	+	+	+
<i>C. echinatus</i> L. (1753) 1050	+	-	-	-	-	+
<i>C. pennisetiformis</i> (Hoscht. & Steud.) (1854) 109	-	-	-	+*	+	+
<i>C. prieurii</i> (Kunth) Maire (1931) 523	-	-	+*	-	-	+
<i>C. setigerus</i> Vahl (1805) 395	-	-	+*	+	+	+
<i>Digitaria abludens</i> (Roem. & Schult.) Veldkamp (1973) 53-55	-	-	-	-	+*	+
<i>D. bicornis</i> (Lam.) Roem. & Schult. (1817) 470	-	-	-	-	+*	+
<i>D. ciliaris</i> (Retz.) Koeler (1802) 27	-	-	-	-	-	-
[Syn. <i>D. biflorus</i> Willd. (1809) 92 (3)]	-	-	+*	+	+	+
[Syn. <i>D. adscendens</i> (Kunth) Henrard (1934) 92 (4)]	-	-	+*	+	+	+
<i>D. longiflora</i> (Retz.) Pers. (1805) 85	-	-	-	-	+*	+
<i>D. nodosa</i> Parl. (1842) 39	-	-	+*	-	-	+
<i>D. radicata</i> (J. Presl.) Miq. (1857) 437	-	-	-	-	+	+
<i>D. sanguinalis</i> Pers. (1771) 52	+	-	-	-	-	+
<i>D. setigera</i> Roth (1817) 474	-	-	-	-	+*	+
<i>D. stricta</i> Roth ex Roem. & Schult. (1817) 474	-	-	+*	+	+	+
<i>Echinochloa colonum</i> (L.) Link. ((1833) 209	-	+*	+	+	+	+
[Syn. <i>Panicum colonum</i> L. (1759) 870 (2)]	-	+*	+	+	+	+
<i>E. crusgalli</i> (L.) P. Beauv. (1812) 161	-	+*	+	-	+	+
[Syn. <i>Panicum crusgalli</i> L. (1753) 56 (2)]	-	+*	+	-	+	+
<i>E. stagnina</i> (Retz.) P. Beauv. (1812) 171	-	-	-	-	+*	+
<i>Eriochloa fatmensis</i> (Hoscht.) W. D. Clayton (1975) 108	-	-	-	+*	+	+
[Syn. <i>E. nubica</i> (Steud.) Hack. & Stapf ex Thell. (1919) 697 (4)]	-	-	-	+*	+	+
<i>E. procera</i> (Retz.) C.E. Hubb. (1930) 256	-	-	+*	-	-	+
<i>Oplismenus burmanii</i> (Retz.) P. Beauv. (1812) 168,169	-	-	-	+*	+	+
<i>O. compositus</i> (L.) P. Beauv. (1812) 168,169	-	-	+*	+	+	+
<i>Panicum antidotale</i> Retz. (1786) 17	+	+	+	+	+	+
[Syn. <i>P. miliare</i> Tam. (1791) 173 (1)]	+	+	+	+	+	+
<i>P. atrosanguineum</i> Hochst. ex A. Rich. (1851) 375	+	+	+	-	-	+
[Syn. <i>P. hydaspicum</i> Edgew. (1862) 207 (1)]	+	+	+	-	-	+



Taxon	1	2	3	4	5	6
	Stewart (1869)	Sabnis (1940)	Nair (1978)	Sharma and Bir (1978)	Sharma and Khosla (1989)	Present Work
<i>Cynodon barberi</i> Rang. & Tadol. (1916) 846	-	-	-	-	+	+
<i>C. dactylon</i> (L.) Pers. (1805) 85	+	+	+	+	+	+
<i>Melanocenchris abyssinica</i> (R. Br. ex Fresen.) Hochst. (1855) 274	-	-	+	-	+	+
<i>M. jacquemontii</i> Jaub. et Spach. (1851) 36, t.325 [Syn. <i>Gracilea royleana</i> Hook. f. (1896,1897) 284 (2)]	-	+	+	-	-	+
<i>Ochthochloa compressa</i> (Forssk.) Hilu (1981) 560 [ <i>Eleusine flagellifera</i> Nees (1842) 220 (2)]	-	+	+	-	+	+
[Syn. <i>E. compressa</i> (Forssk.) Asch. & Schweinf. ex C. Chr. (1922) 12 (3)]	-	+	+	-	+	+
<i>Oropetium biflorus</i> Stapf (1820) 98	-	+	-	-	-	[-]
<i>O. thomaeum</i> (L. f.) Trin. (1820) 98 pl. 3	-	+	+	-	+	+
<i>Perotis hordeiformis</i> Nees (1838, 1841) 247- 248	-	-	-	+	+	+
<i>P. indica</i> (L.) Kuntze (1891) 787	-	-	+	-	-	+
<i>Tetrapogon tenellus</i> (J. König ex Roxb.) Chiov. (1908) 352 [Syn. <i>Chloris tenella</i> J. König ex Roxb. (1820) 330 (2)]	-	+	+	-	-	+
<i>T. villosus</i> Desf. (1799) 389, pl. 255 [Syn. <i>Chloris villosa</i> (Desf.) Pers. (1805) 87 (2)]	-	+	-	-	-	+
<i>Tragus racemosus</i> (L.) All. (1785) 241 [Syn. <i>T. roxburghii</i> Panigrahi (1974) 496 (3)] [ <i>T. biflorus</i> Schult. (1824) 205 (4)]	-	+	+	+	+	+
<i>Tripogon jacquemontii</i> Nees ex Steud. (1892) 85	-	-	-	-	-	+
<i>Zoysia matrella</i> (L.) Merr. (1912) 20, 230	-	-	-	-	-	+
<b>Tribe: Eragrostideae</b>						
<i>Acrachne racemosa</i> (Heyne. ex Roem. & Schult.) Ohwi (1947) 1	-	+	-	-	+	+
[Syn. <i>Eleusine verticillata</i> (Roxb.) (1820) 346 (2)]	-	-	-	-	+	+
<i>Cleistogenes gatacrei</i> (Stapf) Bor (1960) 487	-	-	-	-	+	+
<i>Dactyloctenium aegyptium</i> (L.) Willd. (1809) 1029 [Syn. <i>Eleusine aegyptia</i> (L.) Desf. (1798) 85 (2)]	+	+	+	+	+	+
<i>D. aristatum</i> Link (1827) 59	-	-	-	-	+	+
<i>D. indicum</i> Boiss. (1859) 131	-	-	+	-	+	+
<i>Desmostachya bipinnata</i> (L.) Stapf (1900) 632 [Syn. <i>Eragrostis cynosuroides</i> (Retz.) P. Beauv. (1812) 71, 162, 174 (1)]	+	-	+	+	+	+
<i>Dinebra retroflexa</i> (Vahl) Panz. (1813) 59-60	-	-	-	+	+	+
<i>Eleusine coracana</i> (L.) Gaertn. (1788) 8	+	-	-	+	+	+
<i>E. indica</i> (L.) Gaertn. (1788) 8	-	-	-	-	+	+
<i>Eragrostiella nardoides</i> (Trin.) Bor (1940) 270	-	-	-	-	+	+
<i>Eragrostis atrovirens</i> (Desf.) Trin. ex Steud. (1840) 562	-	-	-	+	+	[-]
<i>E. cilianensis</i> (Bellardi) Vignolo ex Janch. (1907) 110 [Syn. <i>E. major</i> L. (1809) 14,24 (2)]	-	-	+	-	+	[-]
<i>E. ciliaris</i> (L.) R.Br. (1818) 478	-	-	+	+	+	+
<i>E. coarctata</i> Stapf 313 (1897).	-	-	+	-	-	+
<i>E. diarrhena</i> (Schult. & Schult f.) Steud. (1854) 266	-	-	+	+	+	+
<i>E. diplachnoides</i> Steud. (1854) 268	-	-	-	-	+	+
<i>E. gangetica</i> (Roxb.) Steud. (1854) 266	-	-	+	-	+	+
<i>E. japonica</i> (Thunb.) Trin. (1830) 405 [Syn. <i>E. interrupta</i> (Thunb.) Trin. (1812) 71, 162,175 (2)]	-	+	+	+	+	+
<i>E. minor</i> Host (1809) 15 [Syn. <i>E. poaeoides</i> P. Beauv. (1812) 162 (3)]	-	+	+	+	+	+
<i>E. nutans</i> (Retz.) Nees ex Steud. (1840) 563	-	-	+	-	-	+
<i>E. pilosa</i> (L.) P. Beauv. (1812) 71, 162, 175	-	+	+	+	+	+
<i>E. tenella</i> (L.) P. Beauv. Roem. & Schult. (1817) 576	-	-	+	+	+	+
<i>E. tremula</i> Hochst. ex Steud. (1854) 269	-	+	+	+	+	+
<i>Leptochloa chinensis</i> (L.) Nees (1824) 4	-	-	-	-	+	+
<i>L. paniceae</i> (Retz.) Ohwi (1941) 311 [Syn. <i>L. filiformis</i> (Pers.) P Beauv. (1812) 163,166 (2)]	-	+	+	+	+	+
<i>Neyraudia arundinacea</i> (L.) Henrard (1929) 8	-	-	-	-	+	+

Taxon	1	2	3	4	5	6
	Stewart (1869)	Sabnis (1940)	Nair (1978)	Sharma and Bir (1978)	Sharma and Khosla (1989)	Present Work
<i>Briza minor</i> L. (1753) 70	-	-	-	-	-	+
<i>Lolium temulentum</i> L. (1753) 83	-	+	+	+	+	+
<i>Phalaris minor</i> Retz. (1783) 8	-	+	+	-	-	+
<i>Poa annua</i> L. (1753) 68	-	+	-	+	+	+
<i>Polypogon fugax</i> Nees ex Steud. (1854) 184	-	-	-	-	+	+
<i>P. monspeliensis</i> (L.) Desf. (1798) 67	-	+	+	+	+	+
<i>Rostraria cristata</i> (L.) Tzvelev (1970, 1971) 47	-	-	+	+	+	+
[Syn. <i>Lophochloa phleoides</i> (Vill.) Rchb. (1830) 42 (3)]	-	-	+	+	+	+
<i>R. pumila</i> (Desf.) Tzvelev (1970) 48	-	-	+	-	+	+
[Syn. <i>Lophochloa pumila</i> (Desf.) Bor (1960) 445 (3)]	-	-	+	-	+	+
<b>Tribe: Meliceae</b>						
<i>Melica</i> sp.	+	-	-	-	-	[-]
<b>Tribe: Stipeae</b>						
<i>Stipa orientalis</i> Trin. (1829) 83	-	+	-	-	-	[-]
<b>Tribe : Nardeae</b>						
<i>Nardus stricta</i> L. (1753) 53	+	-	-	-	-	[-]
<b>Subfamily: Arundinoideae</b>						
<b>Tribe: Arundineae</b>						
<i>Arundo donax</i> L. (1753) 81	+	+	-	+	+	+
<i>Elytrophorus spicatus</i> Willd A. Camus (1923) 547	-	-	-	-	-	+
<i>Phragmites australis</i> (Cav.) Trin. ex Steud. (1840) 143						
[Syn. <i>Arundo phragmites</i> L. (1753) 81 (1)]	+	+	-	-	+	+
[ <i>P. communis</i> Trin. (1820,1822) 134 (2)]						
<i>P. karka</i> (Retz.) Trin. ex Steud. (1841) 324	-	+	+	+	+	+
<b>Sub family: Danthoideae</b>						
<b>Tribe: Danthoneae</b>						
<i>Schismus arabicus</i> Nees (1841) 422	-	-	+	-	-	[-]
<b>Subfamily : Aristidoideae</b>						
<b>Tribe : Aristideae</b>						
<i>Aristida adscensionis</i> L. (1753) 82	+	+	+	+	+	+
[Syn. <i>A. depressa</i> Retz. (1786) 22 (1)]						
<i>A. funiculata</i> Trin. et Rupr. (1842) 159	-	+	+	-	+	+
<i>A. histricula</i> Edgew. (1862) 208	-	+	+	-	-	+
<i>A. hystrix</i> L. f. (1781,1782) 113	-	+	+	-	-	+
<i>A. mutabilis</i> Trin. & Rupr. (1842)150-151	-	-	+	-	-	+
<i>Stipagrostis hirtigluma</i> (Steud. ex Rupr. & Trin.) De Winter (1963) 134, 136	-	+	-	-	-	+
[Syn. <i>Aristida hirtigluma</i> Steud. ex Trin. & Rupr. (1842)171-172 (2)]						
<b>Subfamily: Chloridoideae</b>						
<b>Tribe: Pappophoreae</b>						
<i>Enneapogon cenchroides</i> (Licht. ex Roem. et Schult.) C.E. Hubb (1934) 119	-	-	+	-	-	+
<i>E. desvauxii</i> P. Beauv. (1812) 82, t. 16, f. 11						
[Syn. <i>Pappophorum brachystachyum</i> Jaub. & Spach. (1850) 365 (3)]	-	-	+	-	-	+
<i>E. persicus</i> Boiss. (1844) 71						
[Syn. <i>Pappophorum aucheri</i> Jaub. & Spach. (1851)32,323 (2)]	-	+	+	-	-	+
<b>Tribe: Cynodonteae</b>						
<i>Aeluropus lagopoides</i> (L.) Trin. ex Thwaites (1864) 374						
[Syn. <i>A. repens</i> Trin. (1848) 462 (1)]	+	+	-	-	-	+
[ <i>A. villosus</i> Trin. ex C.A. (1896) 334 (2)]						
<i>Chloris barbata</i> Sw. (1797) 200	-	-	-	-	+	+
<i>C. dolichostachya</i> Lag. (1816)	-	-	+	-	+	+
<i>C. gayana</i> Kunth (1830) 293, 58	-	-	-	-	+	+
<i>C. montana</i> Roxb. (1820) 331	-	-	+	-	+	+
<i>C. virgata</i> Sw. (1797) 203	-	-	+	+	+	+
<i>Crypsis schoenoides</i> L. (1791) 166, 42, f. 1	-	-	+	-	-	-

Even though research in grasses around the world and within the country has extended to several aspects, basic exploration and inventorization is far from complete.

Our studies brings the cumulative number of species to 192 with 7 new species reports for the region *Briza minor*, *Cymbopogon citratus*, *Elytrophorus spicatus*, *Pogonatherum crinitum*, *Themeda anathera*, *Tripogon jacquemontii*, *Zoysia matrella* (Table 1).

Among the subfamilies, Panicoideae is the best represented in the region. Within this subfamily 4 tribes with 98 species is the most well represented. It is expectedly so since Panicoid grasses are known to dominate the warmer regions like the area of present investigations. The species representation of subfamilies is: Bambusoideae (4), Ehrhartoideae (3), Centothecoideae (1), Pooideae (21), Aristidoideae (5), Arundinoideae (5) and Chloridoideae (55).

Table 1. A comparative list of grass species of Punjab

# Numbers (1, 2, 3 etc.) refer to authors named in the head row who reported under these synonyms, (+) = Reported; (-) = Not reported; (\*) = First reports; [-] = Species not recovered.

Taxon	1	2	3	4	5	6
	Stewart (1869)	Sabnis (1940)	Nair (1978)	Sharma and Bir (1978)	Sharma and Khosla (1989)	Present Work
<b>Subfamily: Bambusoideae</b>						
<b>Tribe: Bambuseae</b>						
<i>Bambusa glaucescens</i> (Willd.) Siebold ex Munro (1868) 89	-	+*	-	+	-	+
[Syn. <i>B. nana</i> Roxb. (1832) 199 (2)]#						
<i>B. nutans</i> G.C.Wall. ex Munro (1868) 92	-	-	-	+*	+	+
<i>B. vulgaris</i> Schrad. ex J.C. Wendl. (1808) 26	+	-	-	+	+	+
[Syn. <i>B. arundinacea</i> (Retz.) Willd. (1799) 245 (1)]						
<i>Dendrocalamus strictus</i> (Roxb.) Nees (1834) 476-477	+	-	-	-	+	+
<i>Drepanostachyum falcatum</i> (Nees) Keng f. (1983) 16	+	-	-	-	-	+
[Syn. <i>Arundinaria falcata</i> Nees (1834) 478 (1)]						
<b>Subfamily: Ehrhartoideae</b>						
<b>Tribe: Oryzeae</b>						
<i>Leersia hexandra</i> Sw. (1788) 21	-	-	-	-	+*	[-]
<i>Oryza rufipogon</i> Griff. (1851) 5, t. 144	-	-	-	+*	+	+
<i>O. sativa</i> L. (1753) 353	+	+	+	+	+	+
<b>Subfamily: Centothecoideae</b>						
<b>Tribe: Thysanolaeneae</b>						
<i>Thysanolaena maxima</i> (Roxb.) Kuntze (1891) 794	-	-	-	+*	+	+
<b>Subfamily: Pooideae</b>						
<b>Tribe: Triticeae</b>						
<i>Secale cereale</i> L. (1753) 84	+	-	-	-	-	+
<i>Hordeum aegiceras</i> Nees ex Royle (1839-1840) 418	+	-	-	-	-	+
<i>H. vulgare</i> L. (1753) 84-85						
[Syn. <i>H. coeleste</i> (L.) P. Beauv. (1812) 114 (1)]	+	-	+	+	+	+
[ <i>H. hexastichum</i> L. (1753) 85 (1)]						
<i>Triticum aestivum</i> L. (1753) 85	+	+	+	+	+	+
[Syn. <i>T. vulgare</i> Vill. (1787) 153 (2)]						
<i>T. compactum</i> Host (1809) 4,7	-	-	-	+*	-	+
<i>T. durum</i> Desf. (1798) 114	+	-	-	+	-	+
<i>T. sphaerococcum</i> Percival (1921) 157, 321 f. 202	-	-	-	+*	-	[-]
<b>Tribe: Bromeae</b>						
<i>Bromus tectorum</i> L. (1753) 77	-	+*	-	-	-	+
<b>Tribe: Poeae</b>						
<i>Alopecurus nepalensis</i> Trin. ex Steud. (1854) 148	-	+*	+	-	+	+
<i>Avena fatua</i> L. (1753) 80	+	+	+	+	+	+
<i>A. sativa</i> L. (1753) 79	+	-	+	-	-	+
<i>A. sterilis</i> L. (1762) 118	-	-	+*	-	+	+
<i>Agrostis gigantea</i> Roth (1788) 31	-	+*	-	-	-	+
[ <i>A. alba</i> L. (1753) 63 (2)]						

to prepare the final prints.

### Identification

Identification of species was done with the help of the floras and compilations mentioned above as well as those of Clayton and Renvoize (1986) and Cope (1982). Specimens were deposited in the Departmental Herbarium.

### Results and Discussion

Table 1 presents a comparative statement of grass species reported in some of the important floristic compilations of the region. Irrespective of the schemes of classification followed by these authors, we have annotated the species into the latest system of the GPWG (2001). Compared to just 5-6 subfamilies in the earlier systems of classification (Prat, 1960; Caro, 1982; Clayton and Renvoize, 1986; Watson and Dallwitz, 1992) the GPWG system recognizes 12 subfamilies with the thirteenth group 'Incertae Sedis' of uncertain affinities. Furthermore, the subfamilies have been classified into 42 tribes. Therefore, reclassification into the GPWG system has involved a revision in subfamilial and tribal affiliation of several species listed in Table 1.

A close scrutiny of various columns of Table 1 clearly shows that grasses received scant attention in the earliest floristic compilations and that work on their exploration and inventorization has made a staggered progress.

Stewart (1869) listed only 40 grass species, whereas Kashyap (1936) did not include grasses in his work. Sabnis (1940) prepared an alphabetic list of 74 species taking the cumulative total to 93 which further increased to 133 with Nair (1978) and 147 with Sharma and Bir (1978). However, the compendium by Sharma and Khosla (1989) registered addition of thirty eight new species taking the cumulative total to 185.

However, since the last revision of the grass flora of the region, grass systematics has picked momentum across the globe mainly after the establishment of the Grass Phylogeny Working Group and their system of classification (GPWG, 2001). This has aroused interest and provided invaluable guidance in systematics and evolution of grasses of the world. Several researchers have studied this interesting and promising group of plants. These studies relate to the fundamental questions relating to grass organography and development (Kellogg, 2000), morphological nature of the spikelets and its parts (Ambrose *et al.*, 2000) and origin of C<sub>4</sub> anatomy (Gaut and Doebley, 1997; Kellogg, 2000). Systematic analysis of grasses has also picked momentum (Linder and Rudall, 2005; Soreng *et al.*, 2007). Within subfamilies, large and complex genera have been revised (Salamin *et al.*, 2002; Saarela *et al.*, 2003; Spangler, 2003; Molina and De Agradar, 2004; Finot *et al.*, 2005; Zuloaga and Morrone, 2005). Owing to their cosmopolitan distribution and occurrence in varied habitats and sharply defined phases in vegetative and reproductive phenology, grasses are being considered as ideal indicator of climate change (Yuan *et al.*, 2007).

Work on Indian grasses has resulted in new species reports (Gopalan and Chandrasekaran, 2001; Veldkamp and Salunkhe, 2000; Kiranraj, 2008; Takhar and Katewa, 2008; Sur, 2001; Ravi *et al.*, 2001; Kumar *et al.*, 2008) documentation of intraspecific diversity through molecular and other methods (Chandra *et al.*, 2004; Saxena and Chandra, 2006) and ethanobotanical uses (Sahu *et al.*, 2010). Ecological studies relate to species association (Soodan *et al.*, 2009) habitat preference and dispersion (Bazzaz, 1991; Sharma *et al.*, 2010).

and scents (Kaul and Vats, 1998; Khanuja *et al.*, 2005; Kim *et al.*, 2005; Bhuiyan *et al.*, 2008; Sujatha, 2010). Grasses also comprise the main source of green cover of our lawns and landscape for tourism and sports. Also, their use in handicraft and cottage industry is well known.

Grasses assume even a greater importance in areas like Punjab which has a tropical/subtropical ecology and an agrarian economy. But grasses have not been given sufficient attention in the floristic compilations of the region. Some of the earliest works have made no mention of the group (Bamber, 1916; Kashyap, 1936). Others have given only an alphabetic list of species with sketchy descriptions (Stewart, 1869; Sabnis, 1940; Nair, 1978; Sharma and Bir, 1978; Sharma, 1990). It is only in the work of Sharma and Khosla (1989) that grass species have been classified into sub families and tribes. With the establishment of Grass Phylogeny Working Group and their publication of a revised and phylogenetic classification (GPWG, 2001), we have witnessed a great upsurge in research on various aspects of grasses across the globe. Amidst a scenario of renewed interest in various aspect of grasses, India has been identified as a 'seriously under-collected country' as far as inventorization of grass diversity is considered (Kellogg, 2006).

We have initiated work on a systematic inventorization, description and classification of the grass species of Punjab and adjoining hills according to the latest format and classification system proposed by GPWG (2001). In the paper, we present a comparative and updated account of the reports of grasses from the region.

## **Materials and Methods**

### **Area of study**

The entire state of Punjab is located between 29° 30' to 30°0' N and 73° 55' to 76° 50' E along with the adjoining hills were surveyed and explored for species diversity of grasses in the entire range of habitats occupied by them. According to the classification of forest types proposed by Champion and Seth (1968), the vegetation type of Punjab falls under the subgroup 5B Northern Tropical Dry Deciduous Forests. The average elevation in the plain region is 200-300 AMSL and the annual rainfall ranges from 250mm to 1000 mm at different locations in the regions. In the hilly areas, there are small stretches of Subtropical Pine Forest and Himalayan Moist Forest in which the ground is covered by the stoloniferous grasses. During the extensive exploration and collection surveys, notes were taken on the economic and ethnobotanical significance, distribution range and the flowering and fruiting phenology of the species.

### **Taxonomic Description**

Taxonomic description of the species was done in sufficient details with special emphasis on diagnostic characters for identification and classification. Grasses have a special morphology. They have a fibrous root system and the stem has a horizontal portion below (rhizome) or above (stolon) the soil surface. The erect portion, the culm bears characteristic leaves with a sheath and a blade. The inflorescence is a panicle, a raceme or a spike modified in various ways. The basic unit of the inflorescence is the spikelet which and typically, consists of two glumes enclosing one or more florets. Each bisexual floret consists of a lemma, a palea, 2-3 lodicules, 3 stamens and a pistil with a bifid plumose stigma. Besides, the spikelet may bear male and reduced florets. The description was based on characters of morphology and micromorphology of the vegetative (culm, leaf, blade, ligule, collar etc.) and reproductive (inflorescence, spikelet, floret, caryopsis etc.) parts. Stereoscopic study was followed by drawings which were inked to prepare the plates which were computer scanned

## **An updated conspectus of grasses of Punjab (India)**

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### **Abstract**

Poaceae is the fourth largest family of the flowering plants. It includes about 700-800 genera and 11000-13000 species distributed worldwide. The family has unmatched ecological and economic importance. With its origin in the early cretaceous and major diversification in the mid Cenozoic, the family at present covers nearly a fifth of land surface and occurs in nearly all the habitats of the world. With a sub tropical ecology and an agrarian economy of Punjab, grasses comprise the most significant group in the region. Despite an overwhelming significance, taxonomic studies in grasses have not received sufficient attention in the region. It is only in the work of Sharma and Khosla (1989) that grass species have been classified into subfamilies and tribes. However, after the establishment of the Grass Phylogeny Working Group (GPWG) the world has witnessed a renaissance in grass systematics. But, India remains an 'undercollected' country as far as grass diversity is concerned. Our work on the exploration and systematics of the grass flora of the region is an effort to consolidate and update the information on the diversity of grasses of the studied area. The present studies have brought the cumulative species number to 192 including seven new reports. The species representation of subfamilies is: Aristidoideae (5) Arundinoideae (5) Bambusoideae (4), Centothecoideae (1), Chloridoideae (55) Ehrhartoideae (3), Panicoideae (98) and Pooideae (21).

**Key words:** Cumulative, India, Poaceae, Punjab, Systematics, Taxonomy, Tropical

### **Introduction**

The grass family Poaceae (R. Br.) Barnh. is the fourth largest family of the flowering plants. It includes about 700-800 genera and 11000-13000 species distributed worldwide (Clayton and Renvoize, 1986; Watson and Dallwitz, 1999). In the classification of grasses proposed by Grass Phylogeny Working Group (GPWG) the genera have been put into forty two tribes and twelve subfamilies besides a group of uncertain affinities, Incertae Sedis. Apart from a high degree of taxonomic diversity, the family has unmatched ecological and economic importance. With its origin in the early cretaceous and major diversification in the mid cenozoic, the family at present covers nearly a fifth of land surface (Arabaci and Yildiz, 2004; Ture and Bell, 2004) and occurs in nearly all the habitats of the world (Clayton and Renvoize, 1986; Ture and Bocuk, 2007). All the cereals and millets are cultivated grasses. Sugarcane, the main source of sugar around the world is also a cultivated grass species. Besides, grasses constitute the main source of forage and fodder for livestock. Apart from food and fodder several grasses are used to extract of aromatic oils

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data neither separated the two varieties nor accessions separated based on their geographical location of populations, although some meaningful groups were obtained. K-means clustering and NMDS analyses confirmed the groupings. Populations from which the accessions were obtained, could be grouped into two Northern and Southern groups in central Zagros region, with a weak support from morphological data.

Hayashi *et al.* (2005a) divided strains of *G. uralensis* into two types based on their *cp-rbcL* sequences (the GA type and AT type). He reported that there were no correlations between the chemotype and the *rbcL* genotype. Accessions of the two varieties of *G. glabra* in our study were not separate in cluster analysis as both produced similar flavonoid spot profiles.

Separation of major polar flavonoids of bulked extractions from each variety resulted in identification of seven flavonoids which had not been reported before. The major non-polar flavonoid of this species without considering the variety of samples was reported as glabridin (Hayashi *et al.*, 2003b).

Our solvent systems efficiently extracted and purified seven flavonoid skeletons which were not reported before for roots and rhizomes of *G. glabra* varieties. These flavonoids shared the prenyl group on aromatic ring-A (position at carbon 6) and differed in *R<sub>f</sub>* values and other substitution properties.

Glabridin which was a major non-polar constituent of underground parts of licorice was not a major flavonoid constituent in our results.

Both varieties consisted of flavones and flavonols. It is expected that *G. glabra* var. *glandulifera* exhibit more pharmacological properties due to the presence of an extra hydroxyl group on carbon number 7 of skeleton number 3 (Figure 10), although this claim must be examined at the variety level.

Closest relatives of *G. glabra* which was studied by Hayashi *et al.* (2005b) based on *rbcL* sequences, were *G. inflata* and *G. uralensis*. These taxa represented a clade that produced glycyrrhizin (an oleanane-type triterpene saponin) as the major constituents in rhizomes. Their sister group to *G. glabra* clade consisted of *G. echinata*, *G. macedonica* and *G. pallidiflora*; which did not produced glycyrrhizin as the major constituent (they produced macedonoside C as the major constituent in rhizomes). Our results provided more detailed information about *G. glabra* populations in Fars province at the variety level and refined the results of a previous study by Haji-Mehdipour *et al.* which reported Fars populations among top three populations regarding glycyrrhizin contents of rhizomes (Haji-Mehdipour *et al.*, 2008).

Finally, while the phylogenetic relationship between *glycyrrhiza* spp. at species level is clear, the infra-specific relationships at population level are still not known. Phenetic relationships between varieties of *G. glabra* which was studied here claimed that their classification under the putative species was accurate, as they shared similarities in amounts of glycyrrhizin produced in rhizomes, morphological characters and flavonoid constituents. However, the varietal rank of these taxa may be changed to forma according to suggestions made by Brummitt (1990). Results of this study showed intermix of both populations and their characteristics regarding morphology, flavonoids and glycyrrhizin contents, support this suggestion.

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Hayashi *et al* (2003b) reported Glabridin as the major flavonoid in underground parts of *G. glabra* collected from Kazakhstan. They also reported Rutin (RT), Isoquercitrin (IQ), Pinocembrin (PN), Licoflavanone (LF) as the four major flavonoids identified from the leaves of same specimens.

In this study, seven more flavonoids with different *R<sub>f</sub>* values were separated in this species; three flavonoids in rhizomes of the var. *glandulifera* and four flavonoids in rhizomes of the var. *glabra*.

### Glycyrrhizin contents of rhizomes

Glycyrrhizin contents of rhizomes in 24 accessions belonging to the two studied varieties were measured using image processing technique. Results showed that accessions were highly variable (from 0.03 to 0.23 percent of dry weight), so that the most glycyrrhizin rich accession (S15, Kuhmareh, 0.23% DW) had more than seven folds glycyrrhizin than the glycyrrhizin-poor accession (S20, Khafr, 0.03% DW). Both the richest and poorest accessions belonged to variety *glandulifera* and both were collected from locations of similar soil texture (clay). Quantities of glycyrrhizin in rhizomes of accessions collected from sandy soils (S4, S10, S12, S17, S18, S21 and S28) were also diverse, suggesting that neither soil texture nor variety (taxonomic rank) were main factors affecting the glycyrrhizin amount in rhizomes. Hayashi and co-workers claimed that glycyrrhizin contents of rhizomes of *Glycyrrhiza glabra* were 10.5% of dry weight (Hayashi *et al.*, 2000). In another report by the same author, glycyrrhizin contents in the underground parts of 3-years-old cultivated *G. uralensis* (China type) was 2.08 to 5.12% of dry weight; relatively higher than those of the Kazakhstan type (0.75 - 2.55% of dry weight) and both were much higher than those of Iranian natural populations studied here (Hayashi *et al.*, 2005a).

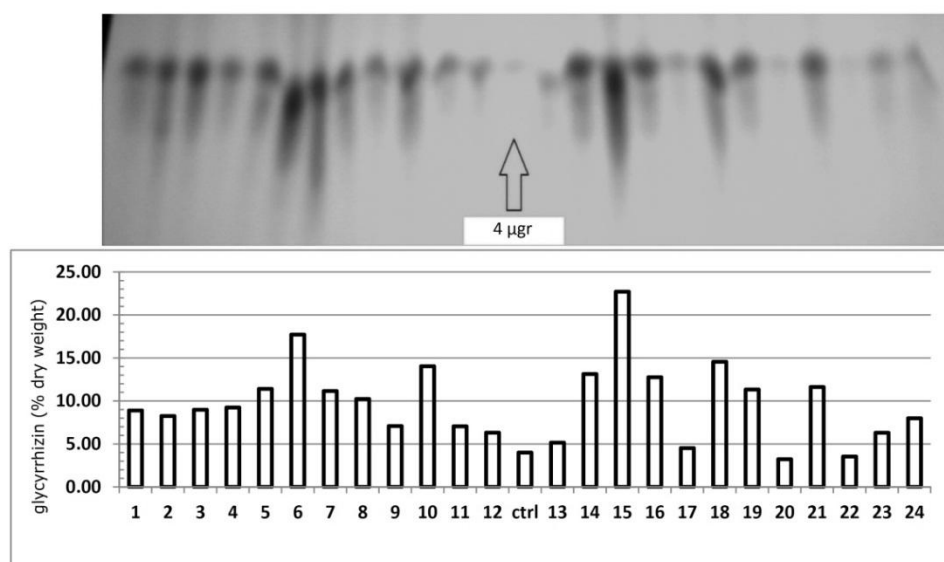


Figure 10. Quantification of glycyrrhizin contents of rhizomes using image analysis. Control lane contained 4µg of glycyrrhizin standard (pure).

### Conclusions

The two varieties of *G. glabra* studied here, grew together forming mixed populations. Intermixture of closely related taxa in licorice plants was already reported for *G. glabra* and *G. uralensis* in a previous study by Hayashi *et al.* (2003b). Cluster analysis of morphological

applying shift reagents (Markham, 1982) showed that three flavonoid skeletons were involved in the separated flavonoids. Variety *glandulifera* consisted of 3 flavonoids, while variety *glabra* consisted of four flavonoids. Properties of each skeleton are presented in Table 4 and skeletons are themselves shown in Figure 10.

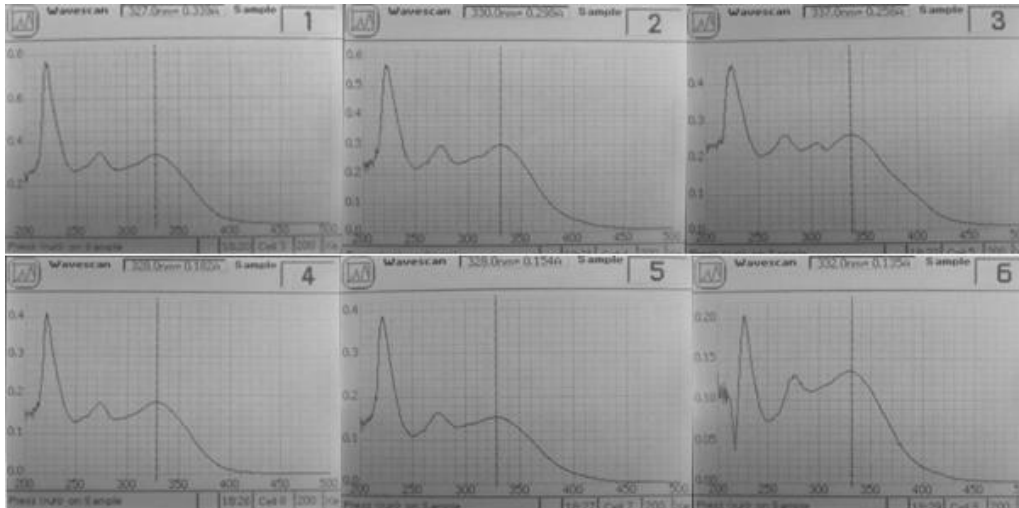


Figure 8. Exemplar UV spectra of methanolic and shift reagents for band#4 in *G. glabra* var. *glabra*. 1: MeOH spectrum, 2: AlCl<sub>3</sub> spectrum, 3: MeOH spectrum, 4: MeOH spectrum, 5: NaOAc spectrum, 6: H<sub>3</sub>BO<sub>3</sub> spectrum.

Both varieties consisted of Flavones and Flavonols. Spots number 1, 5, 6 in variety *glandulifera* and spots number 1, 6 in variety *glabra* were not flavonoids, but phenolic compounds. Those spots were excluded from further identification. The flavonoid skeletons of spots number 2 and 4 in variety *glandulifera* were flavones. Spots number 2, 4 and 5 in variety *glabra* were also flavones. Spot number 3 in variety *glandulifera* and the same number in variety *glabra* were flavonols, although they were not the same. The flavonol in variety *glandulifera* was characterized by an additional hydroxyl group on the carbon number 7 of aromatic ring A.

Table 4. Major flavonoid skeletons identified from licorice rhizomes.

Variety	Spot	Skeleton	R <sub>f</sub>	detail	detail	Fig#
<i>G. glabra</i> var <i>glandulifera</i>	2	Flavone	0.735	5-OH, 6-prenyl		1
	3	Flavonol	0.604	5-OH, 6-prenyl	7-OH	3
	4	Flavone	0.542	5-OH, 6-prenyl		1
<i>G. glabra</i> var <i>glabra</i>	2	Flavone	0.695	5-OH, 6-prenyl		1
	3	Flavonol	0.561	5-OH, 6-prenyl		2
	4	Flavone	0.500	5-OH, 6-prenyl		1
	5	Flavone	0.454	5-OH, 6-prenyl		1

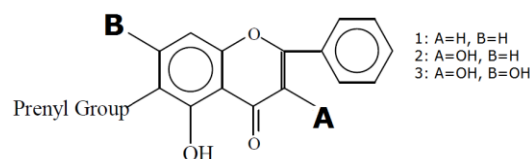


Figure 9. Major flavonoid skeletons identified from licorice. Three flavonoids with skeleton 1, 3 were identified from var. *glandulifera* and four flavonoids with skeleton 1, 2 were identified from var. *glabra*.

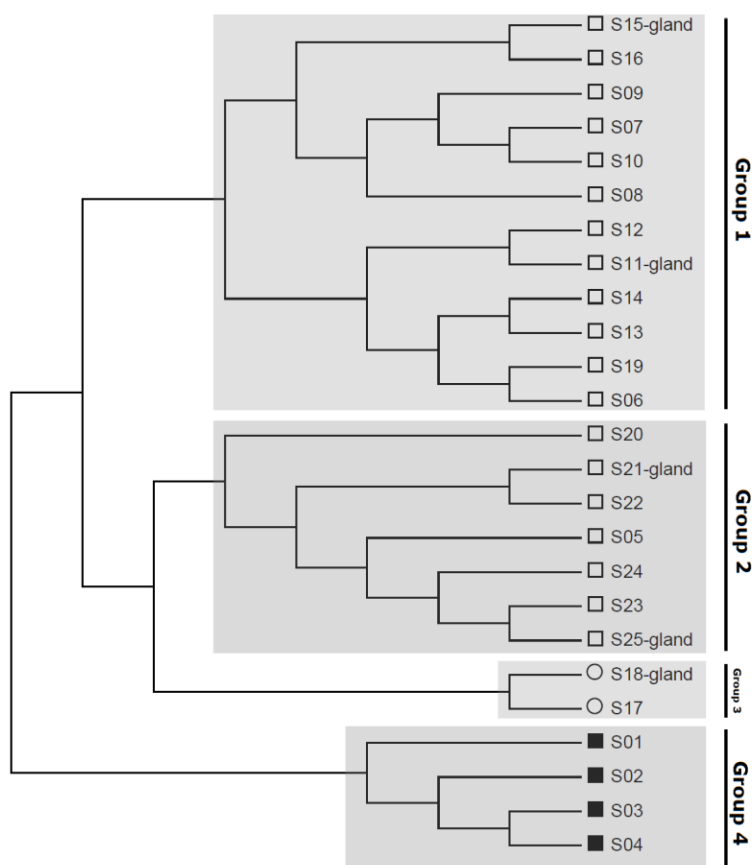


Figure 6. Phenetic relationships between licorice accessions based on flavonoid spots profile. Dendrogram based on data from TLC-35 visualized by UV 254nm and UV 366nm.

### Flavonoid Identification

Flavonoid constituents of rhizomes were separated by using TLC chromatography. The solvent system used for separation of flavonoid spots (see material and method) was also used for purification of each flavonoid. Inspection of chromatograms under UV254nm showed that six bands could be extracted and purified for each variety. Chromatograms of flavonoid constituents of each variety (A: variety *glandulifera*, B: variety *glabra*) are presented in Figure 7 and  $R_f$  values are reported in Table 4.  $R_f$  values ranged from 0.454 to 0.735 for mentioned solvent system and TLC type (refer to Material and Method) and 25°C Temperature.

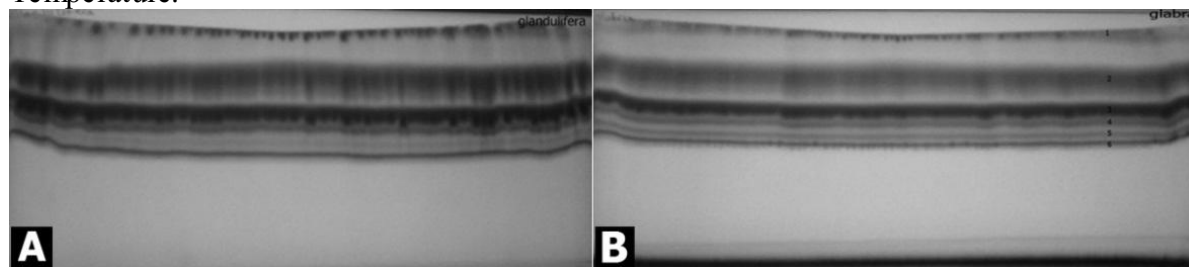


Figure 7. Chromatograms for separation and identification of major flavonoids in A: var. *glandulifera* and B: var. *glabra*.  $R_f$  values are reported in Table 4. Twelve bands were recovered from two TLC plates from which seven different flavonoids were identified (Table 7 and Figure 9).

UV spectrophotometry of each constituent in the range of 200-500 nm before and after

The solvent system used for TLC-35 was a polar system, lacking non-polar components used in TLC-34. It also had increased proportions for polar solvents compared to the solvent system used for TLC-33. Therefore, it was expected that TLC-35 effectively separate the polar flavonoid constituents.

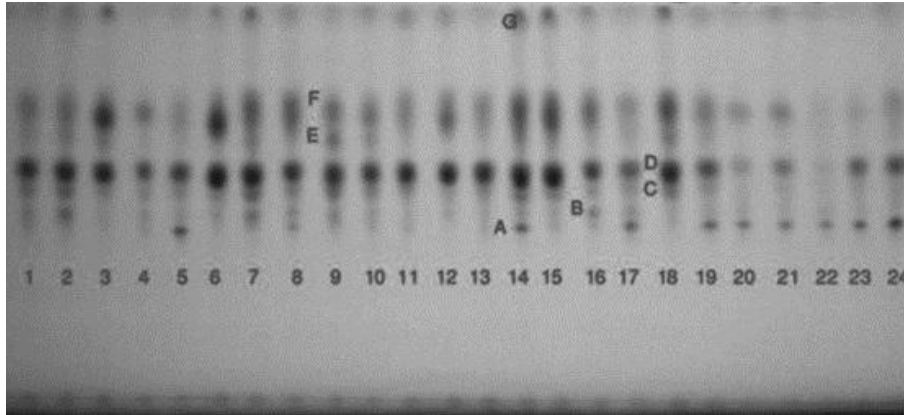


Figure 5. Flavonoid spots on chromatogram of TLC-35. Numbers beneath chromatogram correspond to accessions. For accession names and the solvent system used, refer to text.

Figure 6 shows phenetic relationships between accessions based on flavonoids spots. Four main clusters (groups 1-4) of accessions were identified in the resultant dendrogram. Group 1 and group 2 consisted of a mix of 19 accessions from both varieties. Two subgroups were identified in group 1 while members of group 2 were chained. Members of the second subgroup in group 1 were arranged based on their population distance. Populations of accessions S11 and S12 (both from Darab), S13 and S14 (from Firouzabad and Kuhmareh) were close together with similar soil textures. Accessions S17 and S18 made in small cluster that was also observed in the dendrogram of morphological data. They were geographically located in close distances and were related to different varieties. The fourth group consisted of accessions S01 to 04; all located in Eghlid (NE of Shiraz). Members of this group were also closely related in the dendrogram obtained from morphological data.

Clustering of accessions in groups 1 and 2 were not exactly based on the geographical locations of their populations, however, analyses were robust when different qualitative similarity coefficients were utilized (only results of DICE similarity coefficient are presented).

Accessions studied here could be assigned to two groups; NE populations in Eghlid, Marvdasht and Sepidan and the rest of populations in NW and South of the studied area.

Hayashi *et al.* (2000) in their phylogenetic study on licorice species using *rbcL* sequence claimed that It was difficult to distinguish the variation in GL-producing species by *rbcL* sequence, since they were very similar in all of the *Glycyrrhiza* species. Our results from morphological and flavonoids spots profiles showed that although a clear-cut grouping was not achieved, variations at the infra-specific level could be elucidated. The variation of flavonoids in leaves of *Glycyrrhiza* species was reported to be higher than that of rhizomes (Hayashi *et al.*, 2003a; 2003b), which makes them proper markers for further investigation of phenetic relationships in central Zagros region populations. However, flavonoid spot profiles of rhizomes of the studied accessions in this study were also good enough to reveal phenetic relationships among them.

$K < 6$ . Each obtained cluster for  $K=6$  consisted of from 2 to 8 accessions. Results were compatible to those of cluster analysis using SM coefficient and ME sorting method (Figure 3). Clusters obtained in K-means-MDS analysis ( $K=6$ ) were identical to corresponding clusters in the unrooted tree (Figure 3), confirming the robustness of analyses and further confirmed the partitioning of accessions into two major groups consisting of a total of six subgroups.

The first cluster consisted of accessions S02, S04-S06, S17-S19, S21, S25-S30 and S32-S35. The rest of accessions fell into the second cluster. Membership of S29, S30 and S32 in the first cluster and the membership of S08 and S09 in the second cluster were inconsistent with partitioning of accessions along NW-SE. However, the grouping of the rest of accessions was consistent to geographical partitioning.

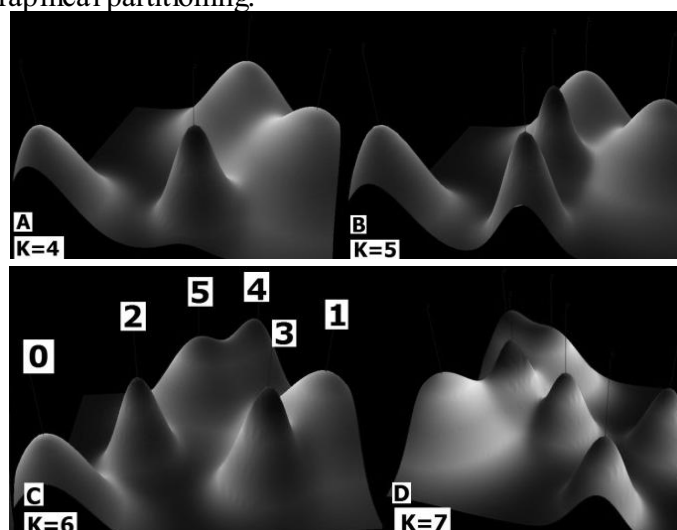


Figure 4. K-means-MDS clustering. Six clusters were defined based on maximized internal similarities in groups. For membership of each accession in each cluster refer to text and table 3.

Table 3. Membership of accessions in each cluster ( $K=4$  to  $K=7$ ).  $K=6$  was the best solution based on maximized internal similarities in defined groups.

Cluster	0	1	2	3	4	5	6
$K=4$	29, 30, 32, 33	27, 28, 34, 35, 07, 08, 09, 11, 14, 12, 13	15, 16	05, 06, 04, 02, 17, 18, 19, 21, 25, 26			
$K=5$	29, 30, 32, 33	27, 28, 34, 35, 07, 08, 09, 11, 14, 12, 13	15, 16	25, 26	05, 06, 04, 02, 17, 18, 19, 21		
$K=6$	29, 30, 32, 33	05, 06, 04, 02, 17, 18, 19, 21	15, 16	25, 26	27, 28, 34, 35	07, 08, 09, 11, 14, 12, 13	
$K=7$	07, 08, 09, 11, 14, 12, 13	05, 06, 04, 02, 17, 18, 19, 21	15, 16	25, 26	27, 28, 34, 35	29, 30	32, 33

### Flavonoids spot profiles

A total of 95 spots (bands) were scored for 25 accessions (TLC-33 using solvent system [chloroform (25%), acetic acid (25%), butanol (25%), methanol (25%)]). Number of scored bands in TLC-34 (for the 24 accessions analyzed using solvent system [water (20%) ethanol (20%) butanol (10%) chloroform (5%) acetic acid (10%) acetonitril (10%) metanol (20%) acetone (5%)]) was 89 bands, while the number of scored bands in TLC-35 (for 24 accessions analyzed using solvent system [water (50%) ethanol (15%) butanol (20%) acetic acid (10%) chloroform (5%)]) was 135 bands. TLCs were visualized under UV254nm and UV366nm. Data obtained from TLC-35 were adopted and profiles for 2-dimensional TLC for selected accessions was checked to make sure that all the possible bands were separated in TLC-35 (Figure 5).

Principal coordinate analysis of qualitative (0/1) morphological data was implemented using Jaccard coefficient (Jaccard, 1908), scattered accessions on the PCO 3-D plot. Relationships between accessions were evaluated by superimposition of a minimum-length spanning tree (Rohlf, 1975) and rotating the plot (Figure 3A). Cluster analysis of qualitative (0/1) morphological data using ME sorting method (Tamura *et al.*, 2007) of Simple Matching coefficient (SM) similarities, grouped 27 accessions based on geographical location of populations from which the accession was collected (Figure 3B). Resultant groupings were not clear cut; however, meaningful clusters were obtained from both analyses. Two main clusters were observed on the unrooted tree (dendrogram). One consisted of accessions S15 (Kuhmareh), S16 (Pasargad), S07, S08 (Marvdasht), S09 (Sepidan), S11 (Darab), S12 (Darab) and S13 (Firuzabad) and S14 (Kuhmareh). The other cluster consisted of the rest of accessions. Members of this cluster belonged to the variety *glabra*, except S11 and S15. Intermix of accessions belonging to the two varieties studied were also observed in other clusters on the phenogram.

Close phenetic relationship between accessions S05, S06 (collected from Beyza), S04, S02 (collected from Eghlid) and a sub cluster consisted of accessions S27 (Nourabad), S28 (Yasouj), S34 (Eghlid) and S35 (Shahrekord), all belonging to the variety *glandulifera* is shown on the phenogram. Cluster analyses of both quantitative and qualitative (0/1) morphological data failed to separate accessions based on taxonomic rank, nor “clearly” grouped accessions based on their population geographic location, although, interpretable clusters were obtained. A North-South partitioning of populations may be deduced. Populations in the first cluster were located in southern part of study area (except for S08 and S09) and populations in the second cluster were located in northern part (except for S29, S30 and S32).

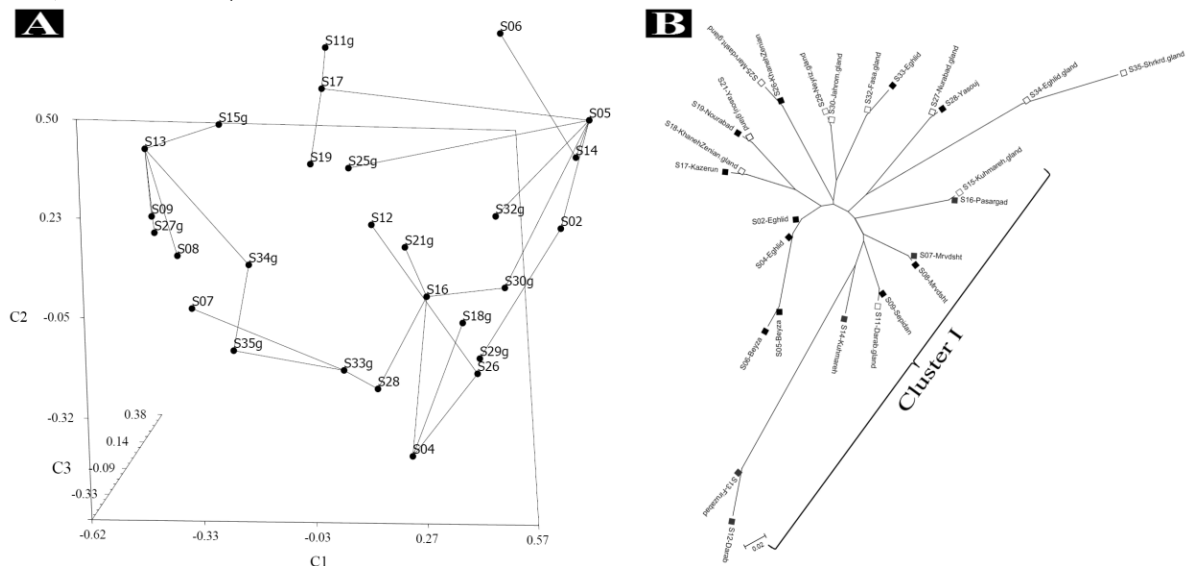


Figure 3. Multivariate analysis of qualitative (0/1) morphological data showing phenetic relationships between 27 accessions. A: Principal Coordinate Analysis using Jaccard coefficient (Jaccard, 1908). A minimum-length spanning tree (Rohlf, 1975) is superimposed on PCO plot, showing the relationships between objects (accessions). B: Unrooted tree obtained from cluster analysis based on ME sorting method (Tamura *et al.*, 2007) of Simple Matching coefficient similarities calculated by using NTSYS-pc software package (Rohlf, 2000). Open boxes are var. *glandulifera* and solid boxes are var. *glabra* (branch lengths proportional to distances).

Results of K-means-Clustering and MDS (Multi-Dimensional Scaling) showed that six clusters of accessions could be defined. Internal similarity tended to be reduced for  $K > 6$  or

and dendrogram was constructed using Tamura's ME sorting method (Tamura *et al.*, 2007).

#### Purification and identification of flavonoids

Close spots of total flavonoid were put on a horizontal line on a thin layer to separate constituent flavonoids. Skeleton of purified flavonoids were identified using UV-spectrophotometry (200-500 nm). Major substitutions on flavonoid skeleton were identified using NaOAc, H<sub>3</sub>BO<sub>3</sub>, HCl, AlCl<sub>3</sub> shift-reagents according to Markham (1982).

#### Extraction and quantitation of glycyrrhizin

Glycyrrhizin was extracted and purified from 2 gr of dried and grinded rhizomes of each accession using the method as described by Shabani *et al.* (Shabani *et al.*, 2009). Thickened roots (rhizomes) of each accession were dried and grinded to fine powder and 2 gr of each material was used for extraction. Ten µl of each extract was placed on UV254F silica-gel and run using a solvent system consisting of [chloroform: 64, methanol: 50, water: 10]. Chromatogram was photographed digitally (Figure 11) and the image was processed using ImageJ software package (Rasband, 2011) to quantify the amount of glycyrrhizin. Pure glycyrrhizin standard (1 mg/ml) was used for calibration. Measures were reported as percent of dry weight.

### Results and discussion

#### Morphology

Twenty-one quantitative morphological characters were measured across twenty-seven accessions of two varieties (*G. glabra* var. *glabra* and *G. glabra* var. *glandulifera*). Each character was measured up to 6 times to calculate the averages and coefficients of variation (CV). Most variable character was seed area (S-S) and least variable characters were i) Widest portion of legume (Lg-W) and ii) Length of sub-terminal leaflet at left (Le-L3) based on CV- and P-values, respectively.

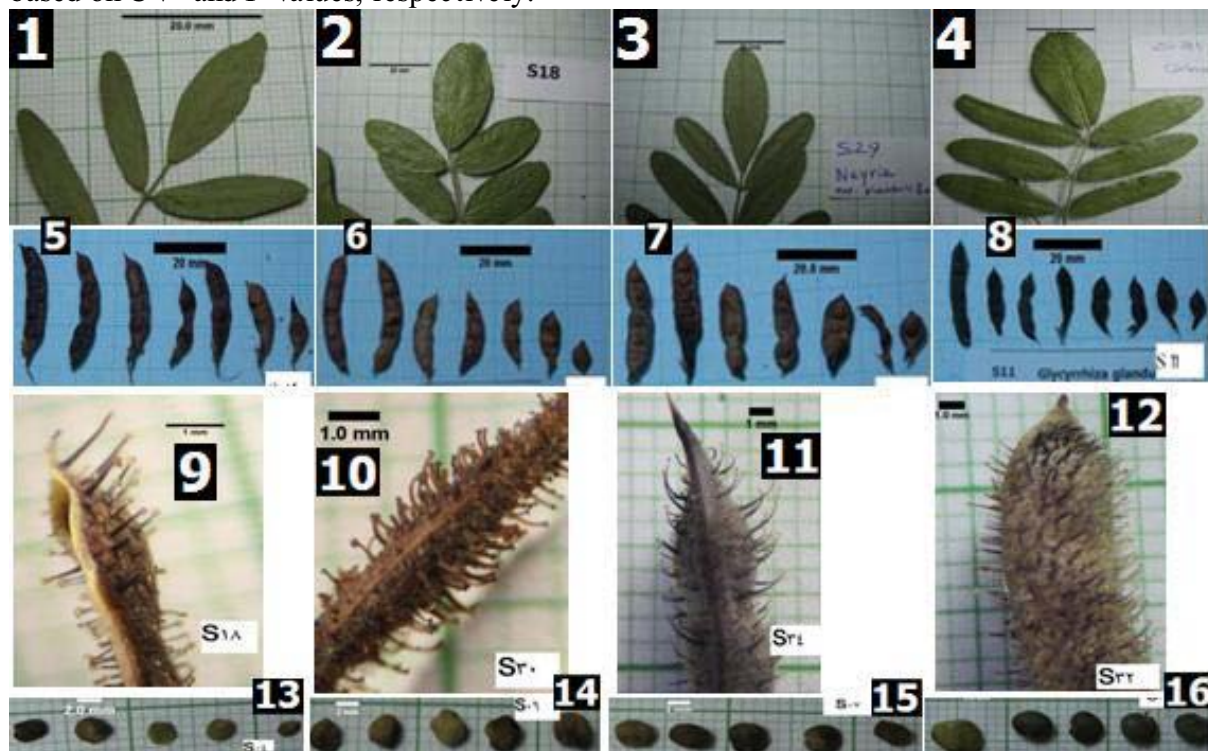


Figure 2. Exemplar photos in analysis of morphological variation in leaflets, legumes and seeds quantitative characters. Leaflets (1-4), Legumes (5-8), Fuzz on legumes (9-12) and Seeds (13-16).



### Morphological analysis

Twenty-one quantitative morphological characters (Table 2) were measured on 27 population accessions. Each character measured up to 6 times on collected materials. Data, including length, perimeter and seed area, legumes, leaflets, etc. were obtained from calibrated digital images and entered into a raw data matrix. Averages measurements of each character were calculated and P-value was defined as (Max-Min)/Average to determine variability of each character across all samples. P- and CV (coefficient of variation) values for each character are presented in Table 2. Then, data were encoded as qualitative data and new data matrix were formed on the basis of midpoints of distribution histograms. Qualitative data matrix was used for multivariate analyses (PCA, NMDS and Cluster analysis). Multivariate analyses were performed using NTSYS-pc software package (Rohlf, 2000). Cluster analysis based on SM similarity coefficient for qualitative data and dendrogram were constructed using ME sorting method of Tamura *et al.* (2007).

Table 2. Quantitative morphological characters

	Character Name	Character Code	P-value	CV
1	Number of ovule per legume	Lg-No	2.238	53.73
2	Number of seed per legume	Lg-Ns	2.477	60.77
3	Total length of legume	Lg-L1	1.716	35.15
4	Length of style	Lg-L2	2.928	55.73
5	Widest portion of legume	Lg-W	0.933	18.49*
6	Legume area	Lg-S	2.056	49.69
7	Legume perimeter	Lg-P	1.581	33.72
8	Length of terminal leaflet	Le-L1	0.771	20.5
9	Widest portion of lamina	Le-L2	0.793	22.8
10	Length of terminal leaflet petiolule	Le-C1	1.642	48.33
11	Length of sub-terminal leaflet at left	Le-L3	0.650*	20.82
12	Length of sub-terminal leaflet petiolule at left	Le-C2	1.410	34.09
13	Length of sub-terminal leaflet at right	Le-L4	0.679	21.81
14	Length of sub-terminal leaflet petiolule at right	Le-C3	1.046	29.1
15	Terminal leaflet area	Le-S1	0.706	19.71
16	Terminal leaflet perimeter	Le-P1	0.709	19.85
17	Seed max diameter	S-a	2.317	39.07
18	Seed min diameter	S-b	2.613	44.82
19	Seed area	S-S	6.146**	122.18**
20	Seed perimeter	S-P	2.611	39.27
21	Fuzz length	F	1.638	41.01

\*minimum and \*\*maximum values of P (see text) and cv.

### Profile of flavonoid spots

Flavonoids were extracted from 5 gr of dried and grinded rhizomes of each sample using methanol (80%) according to Markham (1982). Flavonoids were separated from water-insolubles by vacuum drying then dissolved in water and then further extraction by dissolving in n-butanol. Extractions were vacuum-dried and dissolved in 5 ml pure methanol. Flavonoids were separated on UV254F silica-gel thin-layers using an optimized solvent system [water: 50, ethanol: 15, butanol: 20, acetic acid: 10, chloroform: 5], then visualized under UV254nm. Chromatograms were photographed digitally to inspect the spots and score. Presence or absence of each flavonoid spot was scored as 1/0; data were entered into a raw data matrix. Multivariate analyses (PCA and Cluster analysis) were performed in NTSYS-pc software package. Cluster analysis was performed using DICE similarity coefficient as the coefficient of choice for qualitative data (Duarte *et al.*, 1999)

Table 1. Sample code, scientific names, locality, soil texture and GPS coordinates of specimens were studied.

Sample Code	Scientific Name	Locality	Alt. (m)	Soil Texture	GPS Coordinates
S01	<i>Glycyrrhiza glabra</i> var	Fars prov.: Eqlid-Dasht-e Bakian	2230	Clay	30°54'38.46"N 52°41'26.95"E
S02	<i>Glycyrrhiza glabra</i> var	Fars prov.: Eqlid-Dasht-e Bakian	2230	Sand	30°54'38.46"N 52°41'26.95"E
S03	<i>Glycyrrhiza glabra</i> var	Fars prov.: Eqlid- Dasht-e Namdan	2230	Clay	30°54'38.46"N 52°41'26.95"E
S04	<i>Glycyrrhiza glabra</i> var	Fars prov.: Eqlid- Dasht-e Namdan	2230	Sand	30°54'38.46"N 52°41'26.95"E
S05	<i>Glycyrrhiza glabra</i> var	Fars prov.: Beyza-Ali Abad	1520	Clay	30°05'4.32"N 52°21'20.65"E
S06	<i>Glycyrrhiza glabra</i> var	Fars prov.: Beyza-Doshman Ziyari	1520	Clay	30°42'3.09"N 52°21'54.62"E
S07	<i>Glycyrrhiza glabra</i> var	Fars prov.: Marvdasht- Dasht-e Miagh	1580	Clay	29°51'42.44"N 52°45'15.69"E
S08	<i>Glycyrrhiza glabra</i> var	Fars prov.: Marvdasht- Ramjerd	1620	Clay	30°51'6.66"N 52°34'38.78"E
S09	<i>Glycyrrhiza glabra</i> var	Sepidan- Kamhor	2240	Clay	30°26'18.26"N 51°52'44.81"E
S10	<i>Glycyrrhiza glabra</i> var	Sepidan- Kamhor	2240	Sand	30°26'18.26"N 51°52'44.81"E
S11	<i>Glycyrrhiza glandulifera</i> var	Fars prov.: Darab	1180	Clay	28°45'23.61"N 54°30'57.82"E
S12	<i>Glycyrrhiza glabra</i> var	Fars prov.: Darab	1180	Sand	28°45'23.61"N 54°30'57.82"E
S13	<i>Glycyrrhiza glabra</i> var	Fars prov.: Firooz Abad- Joukan	1600	Clay	28°52'43.07"N 52°33'13.94"E
S14	<i>Glycyrrhiza glabra</i> var	Fars prov.: Koochmarreh Sorkhi	1032	Clay	29°16'27.83"N 52° 9'4.98"E
S15	<i>Glycyrrhiza glandulifera</i> var	Fars prov.: Koochmarreh Sorkhi	1032	Clay	29°16'27.83"N 52° 9'4.98"E
S16	<i>Glycyrrhiza glabra</i> var	Fars prov.: Pasargad	1700	Clay	30°12'41.04"N 53°12'9.70"E
S17	<i>Glycyrrhiza glabra</i> var	Fars prov.: Kazeroon	860	Clay	29°38'11.38"N 51°40'33.23"E
S18	<i>Glycyrrhiza glandulifera</i> var	Fars prov.: Khaneh Zenian	1560	Sandy-Clay	29°40'37.41"N 52° 9'34.02"E
S19	<i>Glycyrrhiza glabra</i> var	Fars prov.: Nour Abad	920	Clay	30°7'25.99"N 51°33'45.86"E
S20	<i>Glycyrrhiza glabra</i> var	Fars prov.: Khafr	1410	Clay	28°59'17.50"N 53°12'16.87"E
S21	<i>Glycyrrhiza glabra glandulifera</i> var	Kohgilouyeh & Boyerahmad prov.: Yasouj	2535	Clay	30°39'28.59"N 51°33'53.52"E
S22	<i>Glycyrrhiza</i> Spp.	Hamedan prov.: Hamedan	1813	n/a	34°49'32"N 48°33'26.87"E
S23	<i>Glycyrrhiza glabra</i> var	Esfahan- Mobarakeh	1570	Clay	32°20'4.57"N 51°30'33.42"E
S24	<i>Glycyrrhiza</i> Spp.	Azerbaijan prov.: Orumiyeh	n/a	n/a	37°33'23.86"N 45° 7'3.76"E
S25	<i>Glycyrrhiza glabra glandulifera</i> var	Fars prov.: Marvdasht- Ramjerd	1620	Clay	30°51'6.66"N 52°34'38.78"E
S26	<i>Glycyrrhiza glabra</i> var	Fars prov.: Khaneh Zenian	1560	Clay	29°40'37.41"N 52° 9'34.02"E
S27	<i>Glycyrrhiza glandulifera</i> var	Fars prov.: Noor Abad	920	Clay	30°7'25.99"N 51°33'45.86"E
S28	<i>Glycyrrhiza glabra</i> var	Kohgilouyeh & Boyerahmad prov.: Yasouj	2535	Sand	30°39'28.59"N 51°33'53.52"E
S29	<i>Glycyrrhiza glandulifera</i> var	Fars prov.: Neiriz	1795	Clay	29°11'37.51"N 54°20'57.71"E
S30	<i>Glycyrrhiza glandulifera</i> var	Fars prov.: Jahrom	1050	Clay	28°28'54.43"N 53°33'13.68"E
S31	<i>Glycyrrhiza glabra</i> var	Fars prov.: Ghader Abad	1900	Clay	30°16'59.57"N 53°16'0.10"E
S32	<i>Glycyrrhiza glandulifera</i> var	Fars prov.: Fasa	1370	Clay	28°56'11.38"N 53°43'4.37"E
S33	<i>Glycyrrhiza glabra</i> var	Fars prov.: Eqlid	2150	Clay	30°54'38.46"N 52°41'26.95"E
S34	<i>Glycyrrhiza glandulifera</i> var	Fars prov.: Eqlid	2354	Clay	30°54'38.46"N 52°41'26.95"E
S35	<i>Glycyrrhiza glandulifera</i> var	Chamahal & Bakhtiari prov.: Shahrekord	2200	Clay	32°18'9.40"N 50°52'52.89"E

*G. glabra* var. *glandulifera* (WALDST. & KIT.) Boiss. (Rechinger, 1984).

Licorice species have been considered as the most important taxa in the genus; they have long been used as medicinal plants; their most important constituent (glycyrrhizin,  $C_{42}H_{72}O_{16}$ ) is widely used as a natural sweetener and a pharmaceutical agent due to its anti-inflammatory and hepatoprotective properties (Hayashi and Sudo, 2009). Hayashi *et al.* (2005a) studied the relationships between 10 strains of so called economically important species of the genus *Glycyrrhiza* and divided them into two types (consisting GA and AT genotypes). The study failed to correlate chemotypes to genotypes, however, in other phylogenetic studies by Hayashi *et al.* (2000; 2005b), *G. glabra* was considered as a close relative to *G. inflata* and *G. uralensis*; a clade that produced glycyrrhizin as the major constituent. Other recent studies considered intra-specific variation of licorice *s. l.* species, quality control and authentication methods (Yao *et al.*, 2008; Khan *et al.*, 2009; Daei *et al.*, 2010; Zhang *et al.*, 2011). Variations in glycyrrhizin contents of licorice roots were evaluated by Haji-Mehdipour *et al.* (2008) in a number of wild populations of the species in Iran. Fars province population was reported among the three higher glycyrrhizin producing sites.

Flavonoids (also referred to as Vitamin P) are a class of plant secondary metabolites. The term is the general name of the compounds based upon a fifteen-carbon skeleton. At the simplest level, the skeleton consists of two phenyl rings (A- and B-rings) connected by a three-carbon bridge (C-ring). In general, plants alone possess the biosynthetic ability of the flavonoids (Mobh, 1939). This study was aimed to assess phenetic relationships between wild populations of the species *G. glabra* (licorice) in the central Zagros region of Iran (including Fars province), using quantitative morphological data, flavonoid spots and glycyrrhizin content profiles.

## Material and Method

Plant material was collected from wild populations of *G. glabra* throughout Zagros Mountain chain of Iran with the emphasis on central Zagros region. Locations of the studied specimens are shown on the map (Figure 1) and the corresponding list is presented in Table 1. Rhizome samples for flavonoid extraction and glycyrrhizin content analyses were selected from those rhizomes with 1-2 mm diameter collected in August-September, from the 0-300 mm soil layer (Douglas *et al.*, 2004; Bolouri-Moghaddam *et al.*, 2009).

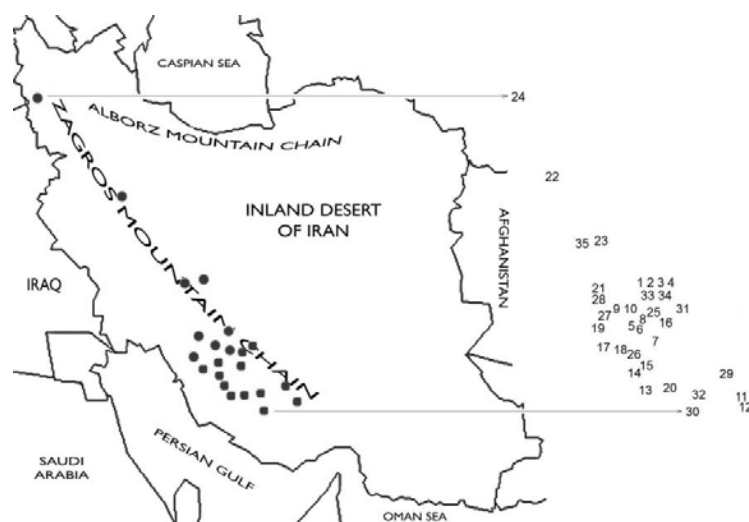


Figure 1. Locations of populations where licorice specimens were collected

## Phenetic relationships among natural population accessions of *Glycyrrhiza glabra* L. (Fabaceae) in central Zagros region of Iran, based on quantitative morphology, flavonoids and glycyrrhizin contents data

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### Abstract

Phenetic relationships among thirty five accessions from natural populations of two varieties of *Glycyrrhiza glabra* in central Zagros region of Iran were studied. Twenty one quantitative morphological characters were measured for twenty seven accessions. PCO, clustering, K-means and MDS analyses were performed on morphological dataset. Polar flavonoid constituents of twenty four accessions were extracted, purified using TLC and characterized at the skeleton class level. Glycyrrhizin contents of rhizomes in twenty four accessions were quantified using image processing methods. Results of multivariate analysis of both morphological and flavonoid spot profile data showed that accessions could be partitioned into two main groups based on geographical locality of the populations. The most variable morphological trait based on CV values, was seed area and the least variable one was Legume width in the widest portion. Accessions of both varieties produced various flavonoids of class flavones and flavonols. Seven flavonoid constituents from the two varieties were separated based on different *R<sub>f</sub>* values. The results revealed that there were moderate (not prominent) levels of variation between the studied accessions. Separation of the varieties based on the single qualitative character in the available literature, was confirmed. Rhizomes of both varieties showed similar amounts of glycyrrhizin and almost similar types of flavonoids in their TLC profiles, suggesting that both were equivalent as herbal drugs in folk medicine.

**Key words:** Flavonoid, *Glycyrrhiza glabra*, Glycyrrhizin, Iran, Morphology, Zagros

### Introduction

Licorice refers to roots and rhizomes of *Glycyrrhiza glabra*; one of the about 18 accepted congeneric species in Leguminosae family (IPNI, 2008; WCSP, 2012), originated in Mediterranean region. They are distributed and grow wild throughout the northern hemisphere. Six species of the genus were reported for the Flora Iranica by Rechinger (1984): *G. bucharica* Regel, *G. aspera* Pall., *G. uralensis* Fisch. ex DC., *G. glabra* L., *G. echinata* L. and *G. macedonica* Boiss. & Orph. *Glycyrrhiza glabra* L. (Syn.: *G. violacea* Boiss.) is represented in Iran by two resembling varieties: *G. glabra* var. *glabra* (the autonym) and

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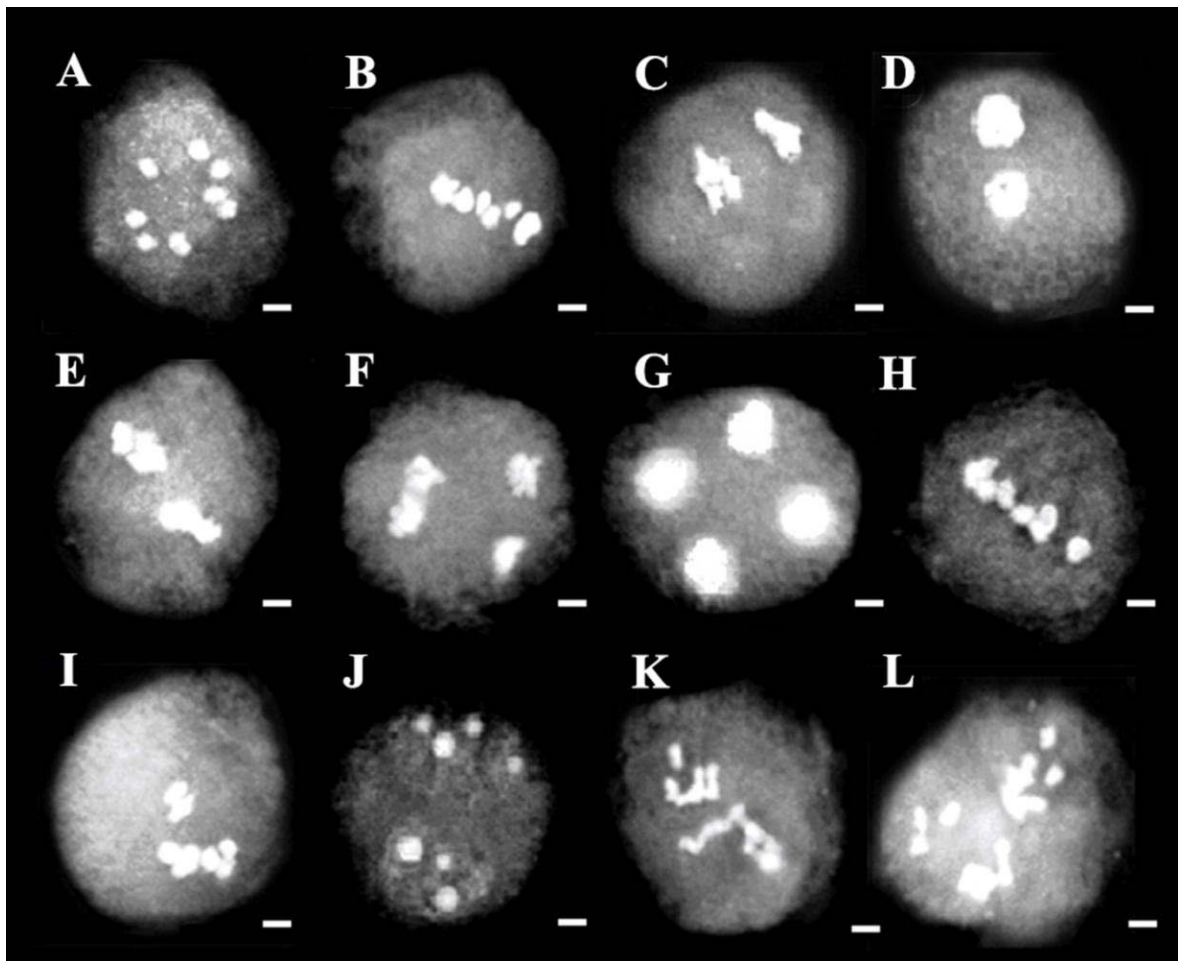


Figure 7. A-L. Representative meiotic cells in the population ANS28. A) Diakinesis; B) Metaphase I; C) Anaphase I; D) Telophase I; E) Metaphase II; F) Anaphase II; G) Telophase II; H) Fragmented chromosome in metaphase I; I) Precocious migration of chromosomes to the pole in metaphase I; J) Trinucleate in telophase I and one micronucleus; K) Asynchronous nucleus; L) Precocious migration of chromosomes in metaphase II. Scale bar: 3  $\mu$ m.

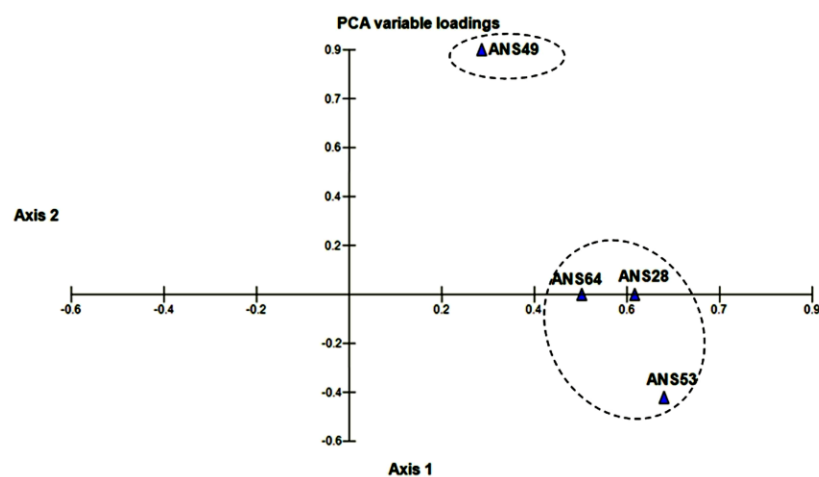


Figure 8. PCA analysis of different populations of *A. anserinifolius* based on cytogenetic data (abbreviations are as listed in Table 1).



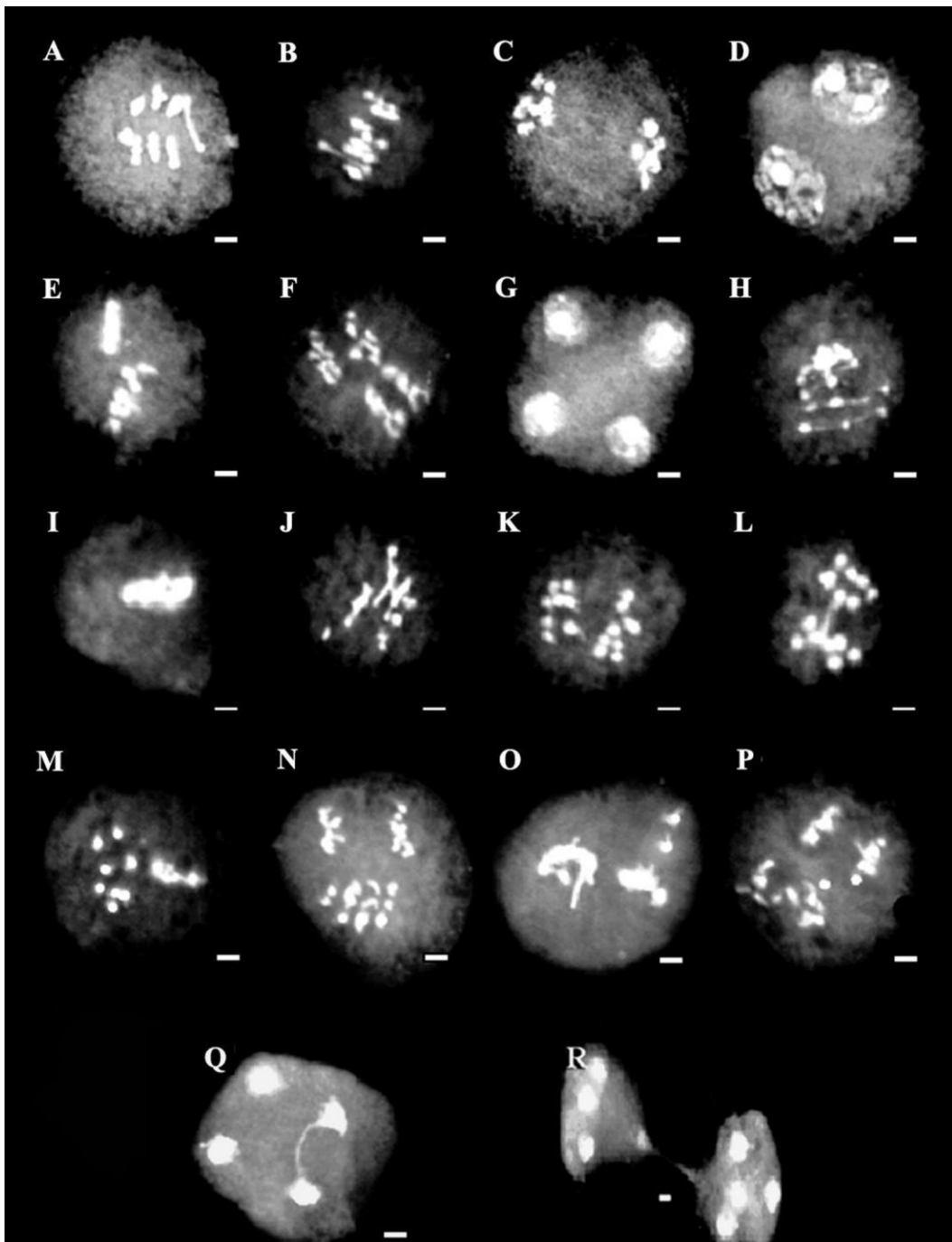


Figure 6. A-L) Representative meiotic cells in the population ANS64. A) Diakinesis; B) Metaphase I; C) Anaphase I; D) Telophase I; E) Metaphase II; F) Anaphase II; G) Telophase II; H) Fragmented chromosomes in metaphase I; I) Sticky chromosomes in metaphase I; J) Precocious chromosome migration to the poles in metaphase I; K) Laggard in anaphase I; L) Bridge in anaphase I. M-R) Representative meiotic cells in the population ANS64. M-N) Asynchronous nucleus; O) Precocious migration of chromosomes to the poles in metaphase II; P) Laggards in anaphase II; Q) Bridge in anaphase II; R) Cytomixis in telophase II. Scale bar: 3  $\mu$ m.

characters responsible for such variations affected meiotic behavior of the taxa, and thus, results from meiotic analysis support morphological groups well.

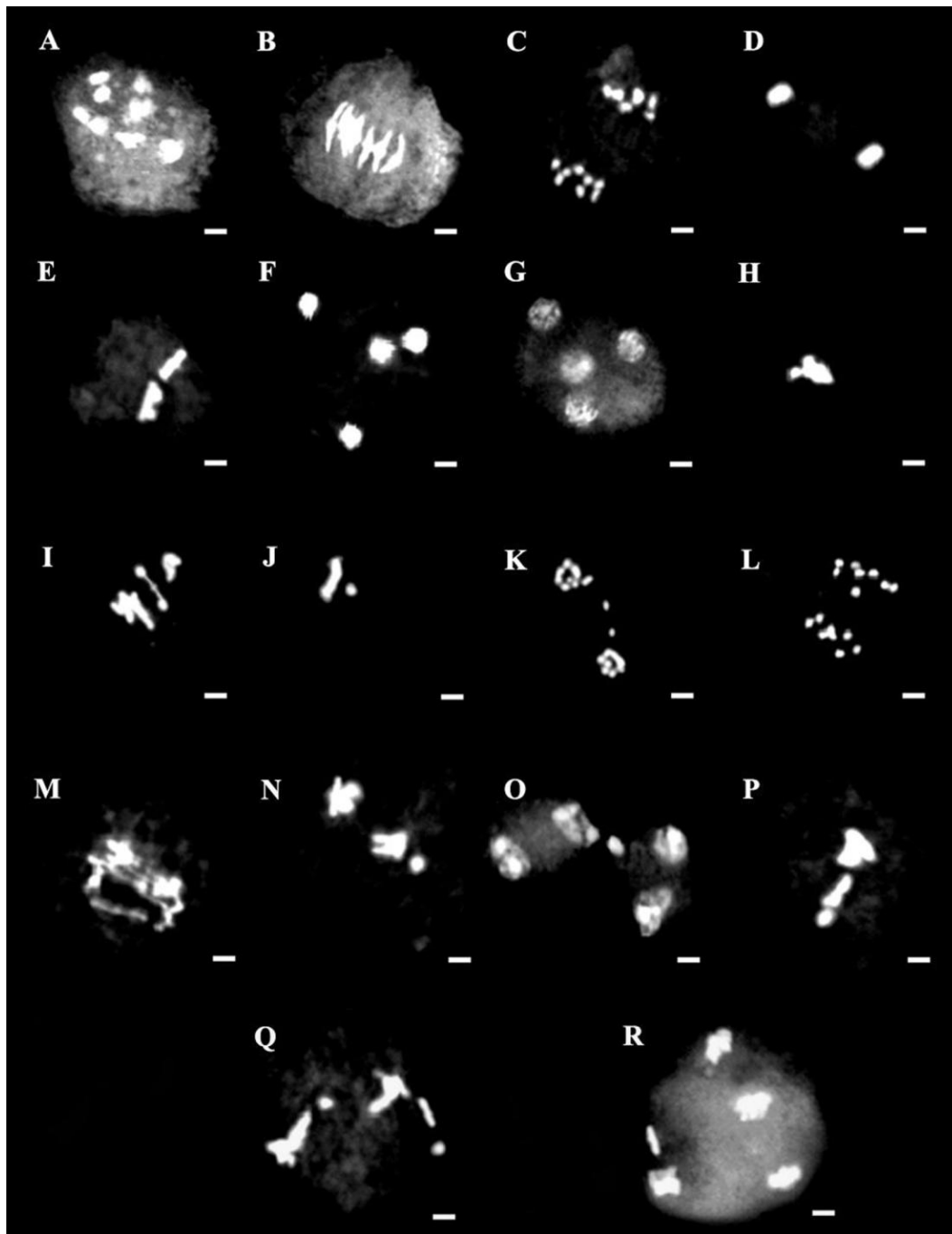


Figure 5. A-L) Representative meiotic cells in the population ANS49. A) Diakinesis; B) Metaphase I; C) Anaphase I; D) Telophase I; E) Metaphase II; F) Anaphase II; G) Telophase II; H) Sticky chromosomes in diakinesis; I) Fragmented and sticky chromosomes in metaphase I; J) Precocious chromosome migration in metaphase I; K-L) Laggard in anaphase I. M-R) Representative meiotic cells in the population ANS49. M) Bridges in Anaphase I; N) Forward chromosome in anaphase I; O) Cytomixis in telophase I; P) Asynchronous nucleus; Q) Precocious chromosome migration to the pole in metaphase II; R) Laggard in anaphase II. Scale bar: 3  $\mu$ m.

### **Cytomixis**

The phenomenon of cytomixis consists in the migration of chromosome between meiocytes through cytoplasmic connection. cytomixis, which is principally a type of meiotic abnormality resulting in changes in gametic chromosome number through migration of chromosomes between adjacent PMCs, could be considered as a process of evolutionary significance in plant populations (Ghanima and Tallat, 2003; Ghaffari, 2006). The factors responsible for cytomixis are rather ambiguous. Some possible causes attributed to cytomixis are the effect of fixation (Gottschalk, 1970), mechanical injury (Sarvella, 1958), pathological conditions (Boback and Herich, 1978), temperature anomalies (Basavaiah and Murthy, 1987), polyploid level (Verma *et al.*, 1984), hybrid condition (Yen *et al.*, 1993), cell response as a consequence of pesticides and antibiotic dosages (Kumar and Sinha, 1991), abnormal genetic behavior due to treatment with a chemical mutagen (Kumar and Srivastava, 2001; Kumar and Sharma, 2002), crop culture condition (Pierozzi and Benatti, 1998), failure of cell wall formation during premeiotic mitosis (Kamra, 1960), and genetically controlled behavior (De Mantu and Sharma, 1983). This phenomenon was observed in the population ANS49 at telophase I (Figure 5-O) and in the population ANS64 at telophase II (Figure 6-R, Table 3).

### **Asynchronous nucleus**

Asynchrony in nucleus was observed in all populations at metaphase II (Figures 4-P, Q; 5-P; 6-M, N; 7-K), among which the population ANS53 showed the highest frequency. Asynchronism was also seen in the population ANS53 at anaphase I (Table 3).

### **Precocious migration**

Precocious migration of chromosomes to the poles was observed in all populations at metaphase I and II stages (Figures 4-K, L, T; 5-J, Q; 6-J, O; 7-L). The highest frequencies of such chromosome migrations at metaphase I and II were observed in the populations ANS53 and ANS64, respectively (Table 3).

### **Micronucleus**

As a consequence of precocious migration of univalent, non-oriented bivalents and laggard chromosomes, some micronuclei were observed in telophase I only in the population ANS 28 (Figure 7-J).

Results obtained from PCA analysis based on cytogenetic data showed an inter population variation as well as morphological characters and resulted in dividing taxa into two groups but with different members in comparison to morphology. So that the populations ANS28, ANS64, ANS53 were divided in group 1 and the population ANS49 in group 2 (Figure 8). The population ANS49 is separated from other populations by high score in the formation of fragmented and sticky chromosomes at metaphase I (25.6% and 39.58%, respectively).

Results from PCA analysis of morphological characters represented variation only between different populations of *A. anserinifolius*. On the other hand, the responsible reasons were not strong enough to justify variation at interspecific level leading to a new species or even at intraspecific level for separating a new subspecies or a new variety. According to some previous works on the genera *Astragalus* and *Onobrychis* of the family Fabaceae (Ranjbar *et al.*, 2009, 2010b, 2011), the agreement between groupings resulted from morphological and meiotic analyses, occurred when the taxa demonstrate well inter/intraspecific variations in phenetic analysis. Quantitative/qualitative morphological

The population ANS53 also showed chromosome bridges at metaphase II. The thickness of bridges observed and the number of the chromosomes involved in their formation varied among different meiocytes in different species.

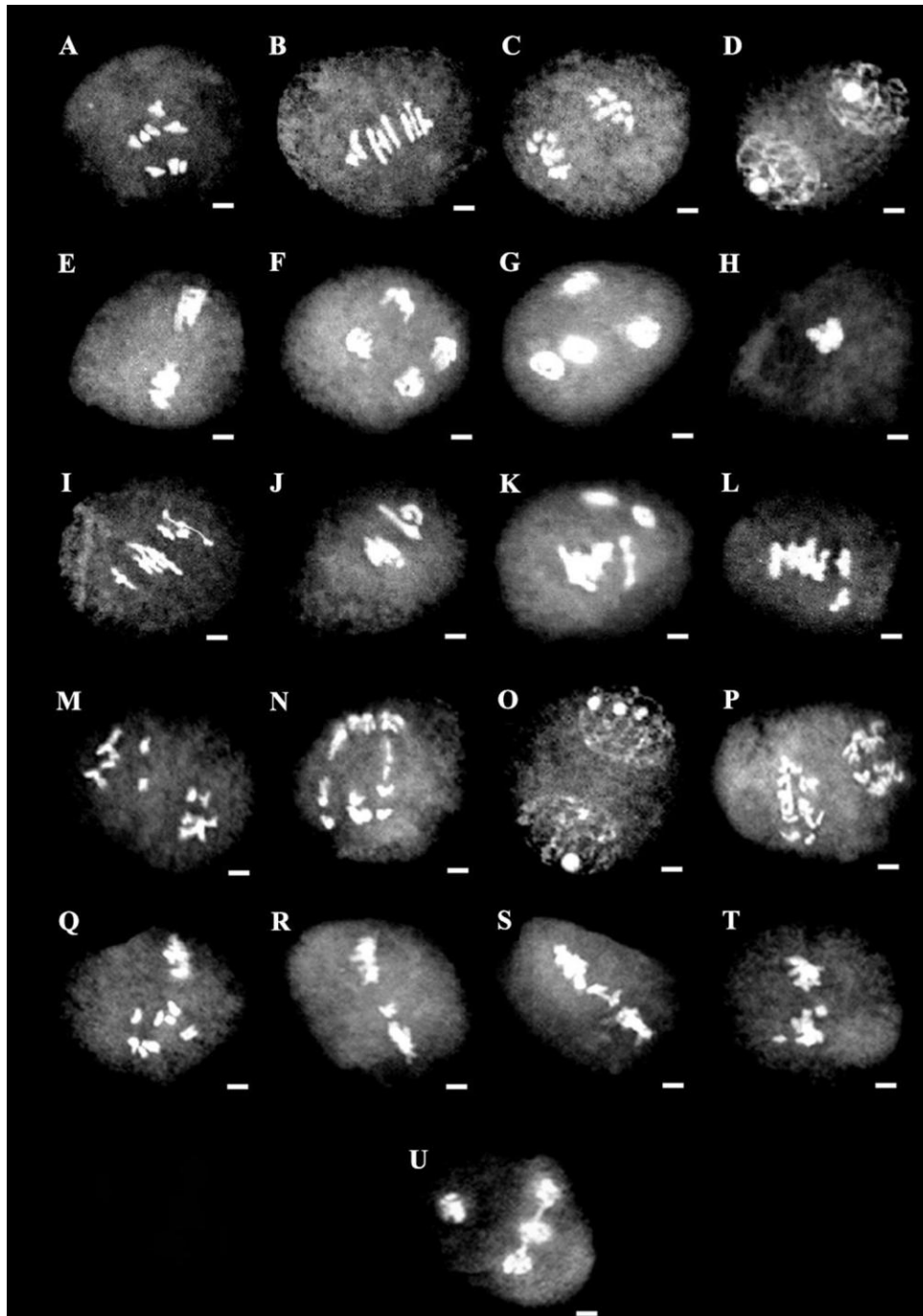


Figure 4. A-L) Representative meiotic cells in the population ANS53. A) Diakinesis; B) Metaphase I; C) Anaphase I; D) Telophase I; E) Metaphase II; F) Anaphase II; G) Telophase II; H) Sticky chromosomes in diakinesis; I) Fragmented chromosomes in metaphase I; J) Sticky and fragmented chromosomes in metaphase I; K-L) Precocious migration of chromosomes to the poles in metaphase I; M-U) Representative meiotic cells in the population ANS53. M) Laggards in anaphase I; N) Bridges in anaphase I; O) Trinucleate in telophase I; P-Q) Asynchronism in meiosis; R) Fragmented chromosome in metaphase II; S) Bridge in metaphase II plates; T) Precocious chromosome migrating to the poles; U) Bridge in late anaphase II. Scale bar: 3  $\mu$ m.

Table 3. Number of pollen mother cells (PMCs) analyzed and percentage of PMCs meiotic behavior in different populations of *A. anserinifolius*

Meiotic characters	ANS53	ANS49	ANS64	ANS28
Number of counted cells	1573	660	1161	1424
% D/MI	17.16	21.81	15.15	18.53
% Fragmented chromosome	19.25	25.6	7.59	1.13
% Bridge	0.3	0	0	0
% Chromosome stickiness	18.14	39.58	17.61	0
% Precocious migration	10.74	7.63	4.54	0.37
% AI/TI	29.43	17.42	13.52	13.55
% Laggard chromosome	2.59	2.6	0.63	0
% Bridge	1.29	2.6	1.91	0
% Forward chromosome	0.64	1.73	0.63	0
% Micronucleus	0	0	0	0.37
% Polynucleate	0.43	0	3.18	5.69
% Asynchronous nucleus	0.64	0	0	0
% Cytomixis	0	0.86	0	0
% MII	8.77	13.78	5.25	8.14
% Asynchronous nucleus	48.55	15.38	21.31	43.1
% Fragmented chromosome	1.44	2.19	0	3.44
% Bridge	0.72	0	0	0
% Precocious migration	7.24	10.98	14.75	4.31
% AII/TII	44.62	46.96	66.06	100
% Laggard chromosome	0	0.32	1.17	0
% Forward chromosome	0	0	0	0
% Bridge	0.56	0	0.13	0
% Precocious migration	0	0	0	0
% Multipolar cell	0.14	0	0	0
% Cytomixis	0	0	0.13	0

### Laggard, forward, sticky and fragmented chromosomes

Fragmented chromosomes, being unable to orient at the metaphase plate, were observed during metaphase I or metaphase II (Figures 4-I, J, R, 5-I, 6-H, 7-H). The highest frequencies of fragmented chromosomes of metaphase I and metaphase II cells were observed in populations ANS49 and ANS28, respectively. Laggard chromosomes were observed during anaphase I in populations ANS53, ANS49 and ANS64 (Figures 4-M; 5-K, L; 6-K); and during anaphase II in populations ANS49 and ANS64 (Figures 5-R; 6-P). According to Niclas and Ward (1994), non-oriented bivalents may be related to impaired attachment of kinetochores to the spindle fibers. Pagliarini (1990) reported that laggards may result from late chiasma terminalization (Souza *et al.*, 2006). These laggards might have degenerated or may have resulted in the formation of polyads particularly at the resting phase (Basi *et al.*, 2006). Forward chromosomes were seen during anaphase I in the populations ANS53, ANS49 and ANS64, among which ANS49 showed the highest frequency (Table 3). Sticky chromosomes were observed at diakinesis and metaphase I in the populations ANS53, ANS49 and ANS64 (Figures 4-J; 5-H, I; 6-I), among which ANS49 had the highest frequency. Chromosome stickiness might have been caused by genetic and environmental factors. However, several agents have been reported to cause chromosome stickiness (Pagliarini, 2000).

### Chromosome bridges

Chromosome bridges resulting from stickiness were observed at anaphase I and anaphase II stages in the populations ANS53, ANS49 and ANS64 (Figures 4-N, S, U; 5-M; 6-L, Q).

**Yazd:** Tang-e Chenar toward Mehriz, 15 km to Mehriz, 2018 m, 2010.4.10, Ranjbar & Mahmoudian 22653 (BASU); Bafgh, Bahabad, 1994.7.3, Jafarynejad & Javadian 404 (Animal & Natural Resources Research Center of Yazd).



Figure 3. Isotype of *A. anserinifolius* Boiss. (Aucher-Eloy 4410 P)

### Cytogenetics

All the wild populations of *A. anserinifolius* studied here possessed  $2n=2x=16$  chromosome number and showed regular bivalent pairing and chromosome segregation at meiosis. They were similar in life history, breeding system, ecology, and geographical distribution in Iran (Figure 1). However, some meiotic abnormalities were observed in different populations included the occurrence of varied degrees of sticky, fragmented and forward chromosomes in anaphase I, laggards and bridges in anaphase I to telophase II, asynchronism, precocious chromosome migration and cytomixis (Table 3; Figures 4-7).

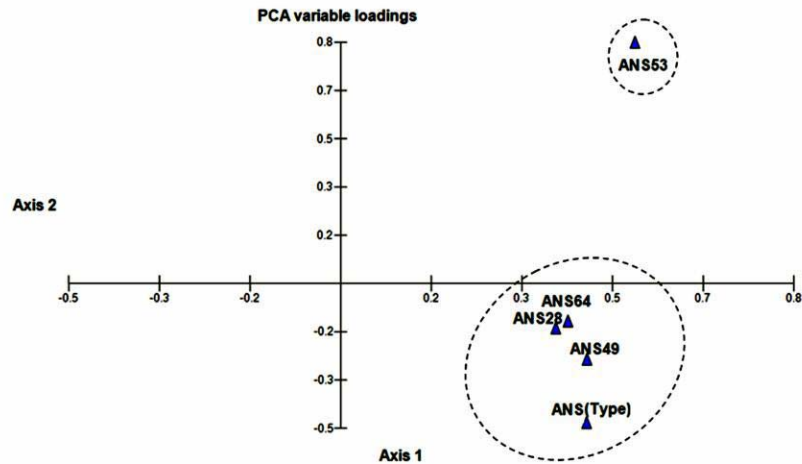


Figure 2. PCA analysis of different populations of *A. anserinifolius* based on morphological characters (abbreviations as listed in Table 1).

*A. anserinifolius* Boiss. 1843, *Diagn. pl. orient.*, ser. 1, 2: 76 - *Malacothrix* - **Holotype**: [Iran] ad sinum Persicum, *P.M.R. Aucher-Eloy* 4410 (G-BOIS!; iso: BM, FI: foto MSB, G!, K!, LE, OXF, P!, W!) [1 sheet in W!: foto MSB *Aucher* 4410, 'erroneously ad Ispahan!'] (Figure 3).

Plants 11-24 cm tall, with developed stems. Stipules ca. 2-3 mm long, pilose, free from one another and the petiole. Leaves ca. 3 cm long, petiole 0.5-1 cm, both petiole and rachis covered with appressed or subpatent hairs. Leaflets 9-13 pairs, contiguous, complicate, obovate, ca. 6 mm long and 4 mm broad, both sides covered with appressed hairs. Inflorescence axillary, spherical or elliptic in flowering, elongated in fruit, flowers numerous, peduncle ca. 5 cm long, covered with short appressed hairs. Bracts linear, ca. 4 mm long, pilose. Pedicels ca. 1 mm long, pilose. Calyx campanulate or shortly tubular, at the base gibbous, ca. 10 mm long, covered with white hairs, rarely with some scattered black hairs, the teeth subulate, ca. 7 mm long. Corolla yellow or pale violet. Standard ca. 15 mm long, the limb elliptic, emarginated at apex. Wing-petals ca. 15 mm long, the limb narrowly elliptic, dilated toward the apex, round-tipped, at the base auriculate, equaling the claw. Keel ca. 10 mm long, the limb oblique-elliptic, equaling the claw. Ovary pilose, sessile. Pods ca. 9 mm long and 4 mm broad, covered with spreading long soft white hairs, bilocular.

### Specimens seen

**Kerman:** Sarcheshmeh toward Pariz, 5 km before Pariz, 2295 m, 2010.4.11, Ranjbar & Mahmoudian 22264 (BASU); Sarcheshmeh toward Sirjan, After Pariz, Pasujan village, 2206 m, 2010.4.12, Ranjbar & Mahmoudian 22609 (BASU); Baft, Azad University of Baft, 2174 m, 2010.4.13, Ranjbar & Mahmoudian 22528 (BASU); Anar toward Shahr-e Babak, 30 km after Anar, 60 km before Shahr-e Babak, 1982 m, 2010.4.11, Ranjbar & Mahmoudian 22649 (BASU); 66 km to Sirjan, 35 km after Shahr-e Babak, 1816 m, Ranjbar & Mahmoudian 22601 (BASU).

**Isfahan:** 10 km SW Mourchekhort to Natanz, 1600 m, 2002.5.11, Rahiminejad, Sahebi & Ghaemmaghani 13224 (Isfahan University Herbarium); Isfahan University Campus, 4574 (Isfahan University Herbarium); Isfahan University Campus, Ahmad Zarrehabadi 6792 (Isfahan University Herbarium).

## Cytogenetics

Chromosome number and meiotic behavior were studied in 4 populations of *A. anserinifolius*. For each population, 15 flower buds from at least 2 plants at an appropriate stage of development were fixed in 96% ethanol, chloroform and propionic acid (6 : 3 : 2) for 24 h at room temperature and then stored in 70% alcohol at 4°C until used. Anthers were squashed and stained with 2% acetocarmine. All slides were made permanent by the Venetian turpentine. Photographs of chromosomes were taken on an Olympus BX-41 photomicroscope at an initial magnification of 1000 X. Chromosome counts was made from well-spread metaphases in intact cells, by direct observation and from photomicrographs. Voucher specimens were kept at BASU, Hamedan, Iran (Table 1).

Table 1. Localities of the species used in this study

Species	Voucher specimens	Locality	Altitude (m)	Collector name	Date	Abbreviation
<i>A. anserinifolius</i>	BASU 22653	Yazd: Tang-e Chenar toward Mehriz, 15 km to Mehriz	2018	Ranjbar & Mahmoudian	10.4.2010	ANS53
<i>A. anserinifolius</i>	BASU 22649	Kerman: Anar toward Shahr-e Babak, 30 km after Anar, 60 km before Shahr-e Babak	1982	Ranjbar & Mahmoudian	11.4.2010	ANS49
<i>A. anserinifolius</i>	BASU 22264	Kerman: Sarcheshmeh toward Pariz, 5 km before Pariz	2295	Ranjbar & Mahmoudian	11.4.2010	ANS64
<i>A. anserinifolius</i>	BASU 22528	Kerman: Baft, Azad University of Baft	2174	Ranjbar & Mahmoudian	13.4.2010	ANS28

Table 2. Morphological characters of four populations of *A. anserinifolius* compared with its type specimen

Morphological characters	ANS64	ANS53	ANS49	ANS28	ANS (Type)
Plant height (cm)	19	35	19	18	17.5
Leaf length (cm)	2.5	2.5	2.5	2.25	3
Petiole length (cm)	0.75	1	0.85	0.75	0.75
Leaflet number	9.5	6.5	10	11	11
Leaflet length (mm)	5.5	6.5	6	4.5	6
Leaflet width (mm)	2	2.5	2.5	1	4
Stipule length (mm)	4	4	4	4	2.5
Peduncle length (cm)	4	5.5	3.75	4.5	5
Bract length (mm)	3.5	5	3.5	2.5	4
Calyx length (mm)	13	14.5	16.5	12.5	13
Calyx teeth length (mm)	8	8	9.5	6.5	7
Standard length (mm)	12	12.5	13	11.5	15
Standard width (mm)	5	5	5.5	5	5
Keel length (mm)	9	9	10	8.5	13
Wing length (mm)	11.5	11	12.5	10	15

## Results and discussion

### Morphology

Results from morphological study showed an inter population variation within *A. anserinifolius*, so that its different populations were divided into two groups. The populations ANS-type, ANS28, ANS64 and ANS49 were placed in group 1 and the population ANS53 in group 2 (Figure 2). Group 2 with a single population (ANS53) was separated from the other populations by its higher plant height and lower number of leaflets (Table 2).



Hence, investigations in different aspects can be useful to resolve taxonomic problems of this problematic group. This work follows previous studies conducted on leguminous fodder species in Iran (Ranjbar and Karamian, 2004; Ranjbar, 2007a, 2007b; Ranjbar *et al.*, 2009) and aims to: increase the knowledge about patterns of morphological variation, chromosome numbers and meiotic behaviour in the four populations of *A. anserinifolius* belonging to *A. sect. Malacothrix*. Such findings can be helpful in understanding the relationships between the chromosomal criteria and taxonomic delimitations.

## Materials and methods

### Morphology

This study is mainly based on field observations during several excursions in Iran and on herbarium materials. The field studies were carried out in different parts of Iran, primarily in central and southeastern Iran (Figure 1). All vouchers have been preserved in BASU, Hamedan, Iran. Also, several sheets were examined for each taxon from the following herbaria: W, P, TARI, BASU, Herbarium of Isfahan University and Herbarium of Research Centers of Natural Resources and Animal Affairs of Yazd and Hormozgan. The specimens studied morphologically are listed in Table 1 and used as operational taxonomic units (OTUs). A total of 14 quantitative characters related to vegetative and reproductive organs were studied in the 4 populations of *A. anserinifolius* (Table 2). Data were entered onto a computerized spreadsheet program, Microsoft Excel version 7. The spreadsheet was later transformed into a file format suitable for phenetic analysis. Principal component analysis (PCA) was performed using MVSP version 3.2 (Kovach, 1985-2004) and used to determine inherent or natural groupings.

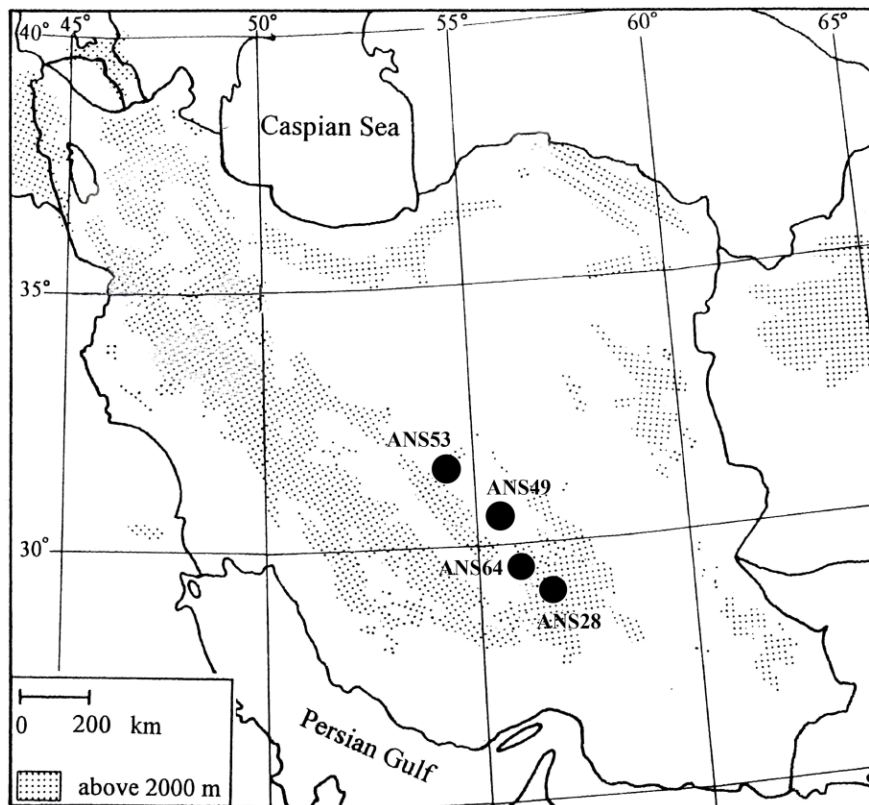


Figure 1. Distribution map of *A. anserinifolius* in Iran

## Cytotaxonomy study of four populations of *Astragalus anserinifolius* Boiss. of section *Malacothrix* Bunge from Iran

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### Abstract

In this research, meiotic chromosome number and the behavior of four populations of *Astragalus anserinifolius* Boiss. of *Astragalus* sect. *Malacothrix* were studied. All wild populations were diploid and showed  $2n=2x=16$  chromosome number, consistent with the proposed base number of  $x=8$  from IPCN. Although all taxa displayed regular bivalent pairing and chromosome segregation at meiosis, some meiotic abnormalities included varied degrees of fragmented and sticky chromosomes in metaphase I, polynucleate and a variable number of laggards, forwarded chromosomes and bridges in anaphase I/telophase I, asynchronous nucleus and precocious chromosome migration in metaphase II and laggards, bridges and cytomixis in anaphase II/telophase II were observed.

**Key words:** *Astragalus* sect. *Malacothrix*, *A. anserinifolius*, chromosome number, meiotic abnormalities

### Introduction

*Astragalus* L. (Fabaceae) is the most diverse genus in the southwest Asia (ca. 1000 spp). With more than 840 species, it is the largest genus in the flora of Iran and the most problematic group in the legume systematic (Lock and Simpson 1991; Yakovlev *et al.*, 1996; Ranjbar and Maassoumi, 1998; Ranjbar and Karamian, 2002, 2003; Ranjbar *et al.*, 2005, 2010a, 2010b, 2010c, 2010d, 2011). *Astragalus* sect. *Malacothrix*, with about 90 species in Iran, is one of the largest sections within the genus (Podlech *et al.*, 2010). Bunge (1868) placed the section together with seven other sections into subgenus *Hypoglottis*. All members of the subgenus share similar morphological characters such as dense capitates or spike like inflorescences. In recent Podlech's typification system (1982), this subgenus is no longer upheld, because of nearly continuous transitions to other groups of the genus. Most of the cytological studies in the genus have concentrated on the chromosome count (Aryavand, 1983; Maassoumi, 1987; Bader and Sherif, 2007; Sheidai *et al.*, 1996). The basic chromosome number ( $x=8$ ) and the two ploidy levels ( $2n=2x=16$ ,  $2n=4x=32$ ) are present in the genus of the old world.

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## Discussion

The knowledge of the floristic composition of an area is a prerequisite for any ecological and phytogeographical study and conservation management (Siadati *et al.*, 2010). Noor and Sisangan forests are considered as remnants of Caspian lowland forests. For the first time, Barzehkar (1994) conducted a preliminary study on vegetation of Noor forest, but detail floristic accounts of this area is still lacking. On the other hand, Sisangan forest is characterized as a unique lowland forest due to occurrence of Hyrcanian endemic plant, *Buxus hyrcana*. The latter species constitutes some rare pure stands across Hyrcanian lowland and submountain forests (Zohary, 1973; Rastin, 1983; Hamzeh'ee *et al.*, 2008; Asadi *et al.*, 2011). Likewise, Noor forest is characterized by the occurrence of *Populus caspica*, a rare and endangered tree species in Iran. Sisangan forest has lower plant diversity compared to Noor forest due to high density of box trees and shrubs in the former. According to a subjective observation, soil texture of the two forests is relatively different from each other and it might be considered as the main cause of differences in the floristic composition and vegetation structure between the two areas.

Based on Sørensen's formula, the obtained similarity between the two forests was about 60 % which indicates rather high similarity of floristic compositions between the forests due to their placement within the lowlands and lacking altitudinal gradients. However, the occurrence of *Buxus* as mono-dominant woody species in some parts of Sisangan forest makes its floristic composition slightly different from Noor forest Park.

Since the life form classification is based essentially on plant reaction to climate, the individual spectrum should tell us much about macroclimatic patterns at field sites (Pears, 1985). Although, therophytes occur abundantly in desert areas (Archibold, 1995), more or less high occurrence of this life form indicates some anthropogenic and over-grazing effects in the study areas (Grime, 2001; Naqinezhad *et al.*, 2006). Similar proportion of therophytes has been previously observed in some other studied ecosystems (Ghahreman *et al.*, 2006, Naqinezhad *et al.*, 2006, Ghahremaninejad *et al.*, 2011). The high percentage of therophytes in the life form spectrum were also encountered elsewhere (Ozen and Kilinch, 2002; Severoglu *et al.*, 2011). The occurrence of therophytes in the Sisangan forest is more prominent than in the Noor forest due to more anthropogenic effects in the former. Following therophytes, geophytes are next dominant life forms. The high proportion of geophytes is consistent with the results of some floristic studies in some other forest areas in the Hyrcanian district (e.g. Ghahreman *et al.*, 2006; Akbarinia *et al.*, 2004; Razavi, 2008; Siadati *et al.*, 2010).

Similar to previous investigations (Ghahreman *et al.*, 2006; Naqinezhad *et al.*, 2010; Ghahremaninejad *et al.*, 2011), pluriregional species constitute a remarkable proportion of the studied flora. These elements can be observed in the lower altitudes of some mountainous systems (Hegazy *et al.*, 1998). Euro-Siberian elements constitute the large proportions of both total flora and flora of each studied forest separately. The occurrence of these elements reflects the phytogeographical link of the studied area with the Euro-Siberian region (e.g. Zohary, 1973; Takhtajan, 1986; Akhiani *et al.*, 2010).

Noor and Sisangan forests are the last remnants of the lowland Hyrcanian forests. These highly threatened ecosystems possess two rare and endemic/subendemic species (*Populus caspica* and *Buxus hyrcana*) which have been drastically exterminated from other areas of the Hyrcanian forests. Conservation policies upon the areas should be applied seriously in order to decrease further damaging effects.

within the Noor forest were geophytes (29.7%), therophytes (29.2%) and hemicryptophytes (19.5%) followed by phanerophytes (17.3%), hydrophytes (2.1%), chamaephytes and helophytes (1.1%). Nevertheless, the dominant life forms in the Sisangan forest were therophytes (29.9%), geophytes (29.6%) followed by hemicryptophytes, phanerophytes (21.9%) and chamaephytes (0.7%).

The total flora was composed mostly of pluriregional elements with 60 taxa (27.3%), followed by Euro-Siberian/Irano-Turanian/Mediterranean elements with 43 taxa (19.5%) (Figure 6). The ratio of endemism was 6.4% and included 14 taxa in the two studied forests. The flora of both forests was mostly composed of pluriregional elements with 52 taxa (28.1%) in Noor and 33 taxa (24.6%) in Sisangan forest, followed by Euro-Siberian (16.8%), Euro-Siberian/Irano-Turanian/Mediterranean (14%), Euro-Siberian/Mediterranean (9.2%), Euro-Siberian/Irano-Turanian (8.2%), endemics (7%), sub-cosmopolitan (5.4%), cosmopolitan (4.9%), Irano-Turanian/Mediterranean (4.3%), Irano-Turanian (1.6%) and Mediterranean (0.5%) elements in Noor forest and Euro-Siberian/Irano-Turanian/Mediterranean (18.7%), Euro-Siberian (18%), Euro-Siberian/Irano-Turanian (9.7%), ES/M (8.2%), endemics and sub-cosmopolitan (5.2%), Irano-Turanian/Mediterranean (4.5%), cosmopolitan (3.7%), Irano-Turanian (1.5%) and Mediterranean (0.7%) in Sisangan forest.

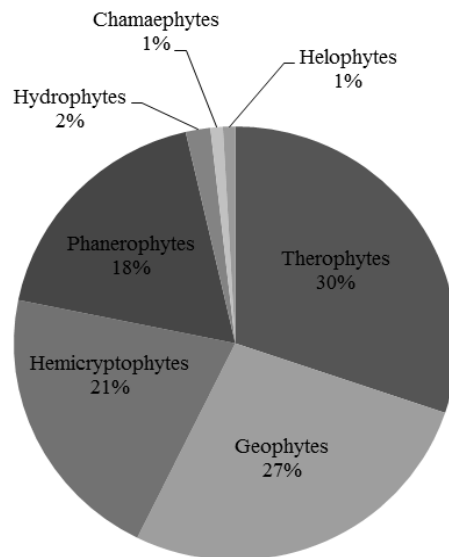


Figure 5. Life form spectrum of studied flora of Noor and Sisangan forests

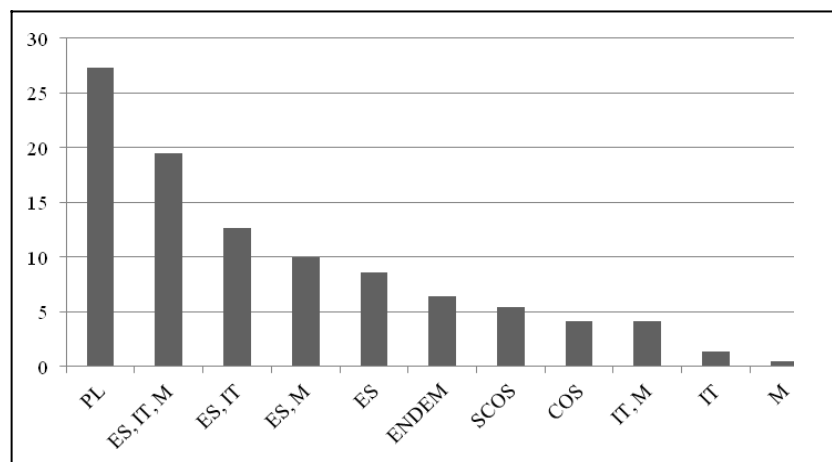


Figure 6. Percentage of main chorotypes of plants studied in Noor and Sisangan forests

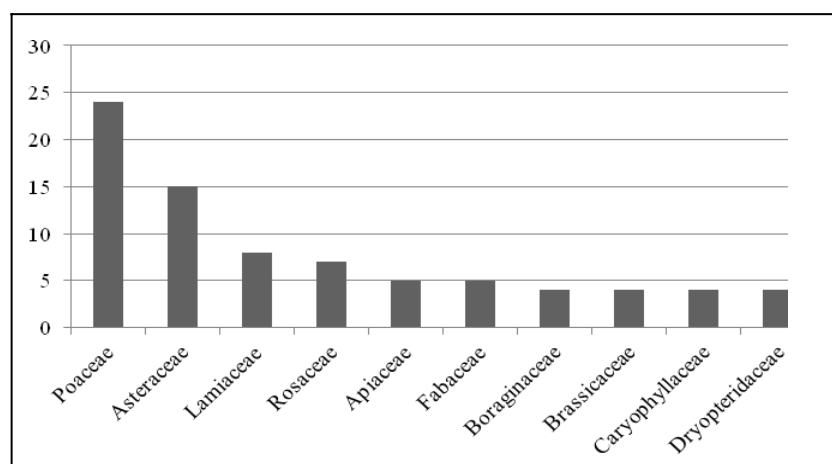


Figure 2. The richest families in terms of number of genera

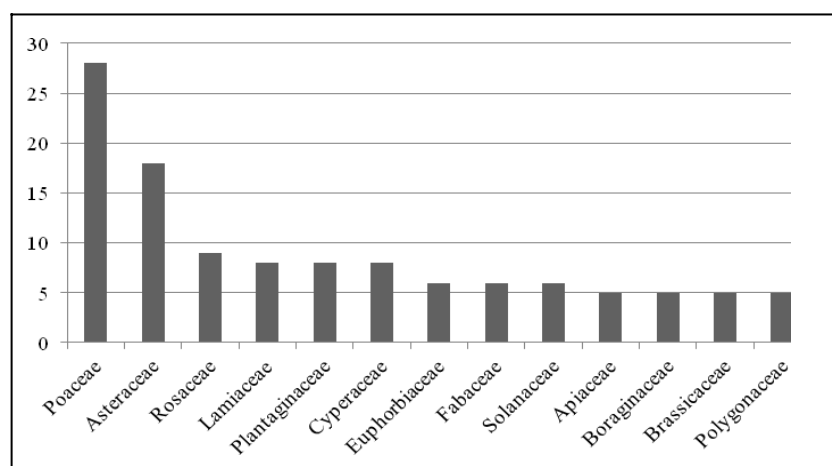


Figure 3. The richest families in terms of number of taxa

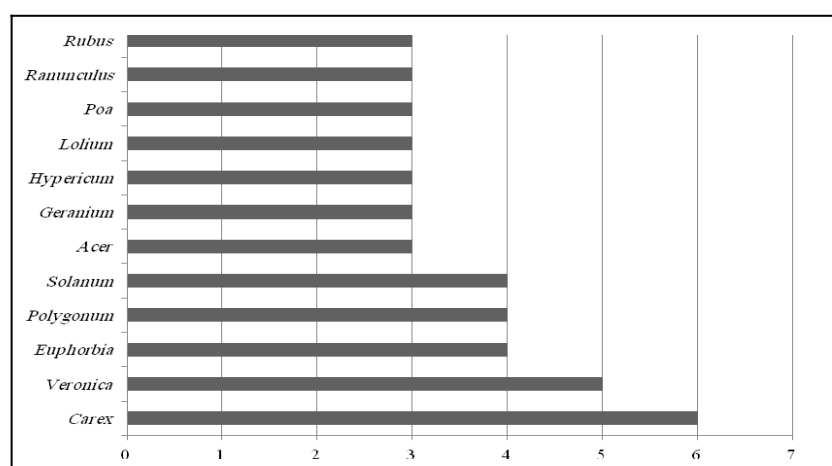


Figure 4. The genera with the largest number of species

### Life form and chorotype spectrum

In the assessment of life form spectrum in the two forests, the dominant life forms were therophytes, which constituted 30.2% of the studied flora, followed by the geophytes (27.1%), hemicryptophytes (20.9%) and phanerophytes (18.2%) (Figure 5). Detailed surveys of life form spectrum in the two studied forests showed that the dominant life forms



Taxon	Life form	Chorotype	Habitat
<i>Cyperus difformis</i> L.	Th	PL	NM
<i>Cyperus rotundus</i> L.	GR	COS	NM
<b>Dioscoreaceae</b>			
<i>Tamus communis</i> L.	GC	ES, IT2, M, N Africa	SM
<b>Iridaceae</b>			
<i>Iris pseudacorus</i> L.	GR	ES, M, N Africa	NI-NM
<b>Juncaceae</b>			
<i>Juncus inflexus</i> L.	Hel	PL	NM
<b>Liliaceae</b>			
<i>Ornithogalum kochii</i> Parl.	GB	ES, [IT2-Iran]	NI
<i>Scilla gorganica</i> Speta	GB	Endem (Iran-Hyr)	NI
<b>Orchidaceae</b>			
<i>Limodorum abortivum</i> (L.) Sw.	GR	ES, M [IT2-Iran]	SI
<i>Listera ovata</i> (L.) R.Br.	GR	PL	NI
<i>Ophrys sphegodes</i> Mill. subsp. <i>sphgodes</i>	GT	ES, M	NI-SI
<b>Poaceae</b>			
<i>Aegilops tauschii</i> Coss.	Th	IT2, Cau	SM
<i>Alopecurus myosuroides</i> Huds. var. <i>myosuroides</i>	Th	ES, IT, M	NM
<i>Arthraxon hispidus</i> (Thunb.) Makino	Th	PL	NM
<i>Brachypodium sylvaticum</i> (L.) P.Beauv.	Hem	ES, IT2	NI-SM
<i>Briza minor</i> L.	Th	ES, M, IT1,2, N Africa	SM
<i>Bromus japonicus</i> var. <i>japonicus</i> Thunb.	Th	PL	NM
<i>Catapodium rigidum</i> (L.) C.E.Hubb. ex Dony	Th	ES, M, IT2	SM
<i>Cynodon dactylon</i> (L.) Pers.	Hem	COS	NM-SM
<i>Digitaria sanguinalis</i> (L.) Scop.	Th	PL	NM
<i>Echinochloa crus-galli</i> (L.) P.Beauv. var. <i>crus-galli</i>	Th	SCOS	NM
<i>Eleusine indica</i> (L.) Gaertn.	Th	SCOS	NM-SM
<i>Hordeum glaucum</i> Steud.	Th	IT, M, N Africa [SS]	NM-SM
<i>Lolium multiflorum</i> Lam.	Th	ES, IT2, M, N Africa	SM
<i>Lolium perenne</i> L.	Hem	PL	SM
<i>Lolium rigidum</i> Gaudin	Th	ES, M, IT2	NM
<i>Lophochloa phleoides</i> (Vill.) Rchb.	Th	PL	NM-SM
<i>Microstegium vimineum</i> (Trin.) A.Camus	Th	PL (Neophyte)	NI-SM
<i>Milium vemale</i> M.Bieb.	Th	ES, IT	NI
<i>Oplismenus undulatifolius</i> (Ard.) P.Beauv.	Hem	ES, M	NI-SI-NM-SM
<i>Paspalum dilatatum</i> Poir.	GR	PL (Neophyte)	NM-SM
<i>Phragmites australis</i> (Cav.) Steud.	Hel	PL	NM
<i>Poa annua</i> L.	Th	SCOS	NI-SI-SM
<i>Poa nemoralis</i> L.	GS	PL	NI-SI
<i>Poa trivialis</i> L.	GS	ES, IT, M	NI-SI
<i>Polypogon monspeliensis</i> (L.) Desf.	Th	PL	NM
<i>Setaria glauca</i> (L.) P.Beauv.	Th	SCOS	NM-SM
<i>Sorghum halepense</i> (L.) Pers.	GR	PL	NM
<i>Vulpia myuros</i> (L.) C.C.Gmel.	Th	M, IT2,4	SM
<b>Ruscaceae</b>			
<i>Ruscus hyrcanus</i> Woronow	Ch	IT2, Cau, Hyr	NI-SI
<b>Smilacaceae</b>			
<i>Smilax excelsa</i> L.	Ph	M, Cau, Euxino-Hyr	NI-SI
<b>Typhaceae</b>			
<i>Sparganium erectum</i> L.	Hyd	ES, M, N Africa [IT2]	NM

Table 2. Number of families, genera and species in the main taxonomic groups

Plant Groups	Families	Genera	Species
Eudicots	56	123	161
Monocots	12	40	50
Pteridophytes	9	12	14

The largest families in terms of number of genera were Poaceae (24 genera), Asteraceae (15 genera) and Lamiaceae (8 genera) (Figure 2). Poaceae (28 spp.), Asteraceae (18 spp.) and Rosaceae (9 spp.) showed the highest species richness respectively (Figure 3). The genera with the largest number of species were *Carex* (6 spp.), *Veronica* (5 spp.) and *Euphorbia*, *Polygonum* and *Solanum* (each with 4 spp.) respectively (Figure 4).

Taxon	Life form	Chorotype	Habitat
<i>Cyclamen coum</i> Mill. subsp. <i>caucasicum</i> (K.Koch.) Meikle	GT	Cau, Euxino-Hyr	NI-SI
<i>Primula heterochroma</i> Stapf	Hem	Endem (Hyr) [Semnan]	NI
<b>Ranunculaceae</b>			
<i>Batrachium trichophyllum</i> (Chaix) Bosch	Hyd	SCOS	NM
<i>Ranunculus dolosus</i> Fisch. & C.A.Mey.	Th	Endem (Hyr)	NI
<i>Ranunculus muricatus</i> L.	Th	IT, M, Cau, N Africa	NI-SM
<i>Ranunculus ophioglossifolius</i> Vill.	Th	ES, M, Euxino-Hyr, N Africa [IT2]	NM
<b>Rhamnaceae</b>			
<i>Paliurus spina-christi</i> Mill. var. <i>spina-christi</i>	Ph	M, IT2,3	NM-SM
<b>Rosaceae</b>			
<i>Crataegus microphylla</i> K.Koch	Ph	Cau, Euxino-Hyr [Krym, E Bulgaria]	NI-SI
<i>Geum urbanum</i> L.	GR	ES, IT2,3, N Africa	NI-NM-SM
<i>Mespilus germanica</i> L.	Ph	M (E), IT2, ES (Cau+Euxino-Hyr+S Europe)	NI-SM-NM
<i>Potentilla reptans</i> L.	Hem	ES, IT, M, N Africa [SS]	NI
<i>Prunus divaricata</i> Ledeb. subsp. <i>caspiaca</i> (Kov. & Ekim.) Browicz	Ph	Cau, Hyr [IT]	NI-SI
<i>Rubus caesius</i> L.	Ph	ES, IT	NI-SM
<i>Rubus persicus</i> Boiss.	Ph	Endem (Hyr)	NI-SM
<i>Rubus sanctus</i> Kuntze	Ph	IT, M, Cau	NM-SM
<i>Sanguisorba minor</i> Scop.	Hem	ES, M, IT2,3, N Africa	NI-NM
<b>Rubiaceae</b>			
<i>Galium ghilanicum</i> Stapf	Th	IT, Cau	NI-SM
<b>Salicaceae</b>			
<i>Populus caspica</i> (Bornm.) Bomm.	Ph	IT2,3, Cau	NI
<i>Salix alba</i> L.	Ph	ES, IT, M, N Africa	NM
<i>Salix</i> sp.	Ph		SI-SM
<b>Sapindaceae</b>			
<i>Acer cappadocicum</i> Gled.	Ph	Euxino-Hyr, Cau [Pak]	SI
<i>Acer velutinum</i> Boiss. var. <i>glabrescens</i>	Ph	Endem (Hyr)	NI-SI
<i>Acer velutinum</i> Boiss. var. <i>velutinum</i>	Ph	Endem (Hyr)	NI
<b>Scrophulariaceae</b>			
<i>Scrophularia vernalis</i> L. subsp. <i>clausii</i> (Boiss. & Buhse) Grau.	Hem	Hyr [IT-Azer+Semnan]	NI
<i>Verbascum</i> sp.	Hem		SM
<b>Solanaceae</b>			
<i>Atropa belladonna</i> L.	GR	ES, M	SM
<i>Physalis alkekengi</i> L.	GR	ES, IT2,3,4	SM
<i>Solanum dulcamara</i> L.	Ph	IT2, ES (Cau, S Russia)	NI-NM
<i>Solanum kieseritzkii</i> C.A.Mey.	GR	Endem (Hyr)	NM
<i>Solanum nigrum</i> L.	Th	COS	NI-SI-SM
<i>Solanum sisymbriifolium</i> Lam.	Ph	PL	SM
<b>Tamaricaceae</b>			
<i>Tamarix ramosissima</i> Ledeb.	Ph	ES, IT	NM
<b>Ulmaceae</b>			
<i>Ulmus glabra</i> Huds.	Ph	ES, [IT, M]	SI
<i>Ulmus minor</i> Mill.	Ph	ES, M, N Africa	NI-SI-NM
<i>Zelkova carpinifolia</i> Dippel	Ph	Cau, Euxino-Hyr [IT2-Iran]	NI-SI-NM
<b>Urticaceae</b>			
<i>Parietaria officinalis</i> L.	GR	ES [IT, M]	NI
<i>Urtica dioica</i> L.	GR	PL	NI-SI
<b>Verbenaceae</b>			
<i>Verbena officinalis</i> L.	Hem	SCOS	NM-SM
<b>Violaceae</b>			
<i>Viola alba</i> Besser	GR	ES, M, N Africa	NI-SI
<i>Viola odorata</i> L.	GR	ES, IT, M, N Africa	NI-SI
<b>Zygophyllaceae</b>			
<i>Tribulus terrestris</i> L. var. <i>orientalis</i>	Th	PL	NM
<b>Monocotyledones</b>			
<b>Alismataceae</b>			
<i>Alisma plantago-aquatica</i> L.	Hyd	PL	NM
<b>Araceae</b>			
<i>Arum maculatum</i> L.	GR	ES	NI
<i>Lemma minor</i> L.	Hyd	COS	NM
<b>Cyperaceae</b>			
<i>Carex divulsa</i> Gooden. subsp. <i>divulsa</i>	GR	ES, IT, M, N Africa	NI-SI
<i>Carex grioletii</i> Roem. ex Schkuhr	GR	M, Cau, Euxino-Hyr	NI
<i>Carex remota</i> L.	GR	ES, M, N Africa	NI-SI
<i>Carex songorica</i> Kar. & Kir.	GR	IT2,3,4, ES (Cau+Russia) [East Asia]	NM
<i>Carex strigosa</i> Huds.	GS	ES	NI-SI
<i>Carex sylvatica</i> Huds.	GR	ES, N Africa [Altai Mts]	NI-SI

Taxon	Life form	Chorotype	Habitat
<i>Centaurium pulchellum</i> (Sw.) Druce	Th	ES, IT, N Africa	SM
<b>Geraniaceae</b>			
<i>Geranium columbinum</i> L.	Hem	ES, M, N Africa [Azer]	NM-SM
<i>Geranium molle</i> L.	Th	ES, IT, M, N Africa	NI-SM
<i>Geranium robertianum</i> L.	Hem	PL	SI-SM
<b>Hamamelidaceae</b>			
<i>Parrotia persica</i> C.A.Mey	Ph	Hyr [Azer]	NI-SI
<b>Hypericaceae</b>			
<i>Hypericum androsaemum</i> L.	Ch	ES, M, N Africa [IT-Azer+Turk]	NI
<i>Hypericum hirsutum</i> L.	Hem	ES (Cau+E+Euxino-Hyr), NW Africa	NI
<i>Hypericum perforatum</i> L.	Hem	PL	NM-SM
<b>Juglandaceae</b>			
<i>Pterocarya fraxinifolia</i> (Poir.) Spach	Ph	Cau, Euxino-Hyr	NI-SI
<b>Lamiaceae</b>			
<i>Ajuga reptans</i> L.	GS	ES [M+N Africa]	NI
<i>Clinopodium umbrosum</i> (M. Bieb.) K. Koch	GR	Cau, Euxino-Hyr [Afgh+Him+N India]	NI-SM
<i>Lamium album</i> L. subsp. <i>album</i>	GR	ES, IT	NI-SI
<i>Lycopus europaeus</i> L.	GS	PL	NI-NM
<i>Mentha aquatica</i> L.	GS	PL	NI-SM
<i>Prunella vulgaris</i> L.	GR	PL	NI-SM
<i>Scutellaria tournefortii</i> Benth.	GR	Hyr [Azer+Afgh]	NI-SM
<i>Teucrium hyrcanicum</i> L.	GR	Cau (Transcau), Euxino-Hyr	NM-SM
<b>Linaceae</b>			
<i>Linum bienne</i> Mill.	Hem	ES, IT 2, M, N Africa	SM
<b>Lythraceae</b>			
<i>Lythrum salicaria</i> L.	Hem	SCOS	NM
<i>Punica granatum</i> L.	Ph	PL	NI-NM
<b>Malvaceae</b>			
<i>Malva</i> sp.	Th		NM
<i>Sida rhombifolia</i> L.	Hem	PL (Neophyte)	NM-SM
<i>Tilia dasystyla</i> Steven	Ph	ES	SI-SM
<i>Tilia</i> sp.	Ph		SM
<b>Moraceae</b>			
<i>Ficus carica</i> L.	Ph	M, IT 2, Cau	NI-SI
<i>Morus alba</i> L.	Ph	IT	NI-SI
<b>Oleaceae</b>			
<i>Fraxinus excelsior</i> L. subsp. <i>coriariifolia</i> (Scheele) E. Murray.	Ph	Cau Euxino-Hyr [IT 2]	NI
<i>Jasminum fruticans</i> L.	Ph	M, Cau, N Africa [IT 2+S Europe]	NM
<b>Onagraceae</b>			
<i>Circaea lutetiana</i> L.	GR	ES, IT, M, N Africa	NI-SI
<i>Epilobium hirsutum</i> L.	GR	PL	NI-NM
<b>Orobanchaceae</b>			
<i>Orobanche cernua</i> Loefl.	Hem	PL	NI
<b>Oxalidaceae</b>			
<i>Oxalis comiculata</i> L.	Th	COS	NI-SM
<b>Papaveraceae</b>			
<i>Chelidonium majus</i> L.	Hem	ES, M, IT 3, N Africa	SM
<b>Phytolaccaceae</b>			
<i>Phytolacca americana</i> L.	Hem	PL (Neophyte)	SM
<b>Plantaginaceae</b>			
<i>Kickxia elatine</i> (L.) Dumort. subsp. <i>crinite</i> (Mabille.) Greuter.	Th	IT, M	NM
<i>Plantago lanceolata</i> L.	Hem	ES, IT, M, SS, N Africa	NM-SM
<i>Plantago major</i> L.	Hem	SCOS	NM-SM
<i>Veronica anagallis-aquatica</i> L. subsp. <i>michauxii</i> (Lam.) A. Jelen.	Hem	IT	NM
<i>Veronica arvensis</i> L.	Th	ES, IT, M	NI
<i>Veronica crista-galli</i> Steven	Th	Cau, Hyr	NI-SI
<i>Veronica francispetae</i> M.A.Fisch.	Th	Endem (Iran-Hyr)	NI-SI
<i>Veronica persica</i> Poir.	Th	SCOS	NI
<b>Polygonaceae</b>			
<i>Polygonum hydropiper</i> L.	Th	ES, IT, M	NM
<i>Polygonum hyrcanicum</i> Rech.f.	Hem	Endem (Iran-Hyr) [Alborz]	NM-SM
<i>Polygonum lapathifolium</i> L.	Th	ES, IT, M	NI-NM-SM
<i>Polygonum patulum</i> M.Bieb.	Hem	M, IT 2,3, Cau	NM
<i>Rumex sanguineus</i> L.	Hem	ES [M]	NI-SM
<b>Portulacaceae</b>			
<i>Portulaca oleracea</i> L.	Th	COS	NM
<b>Primulaceae</b>			
<i>Anagallis arvensis</i> L.	Th	ES, IT	SM

Taxon	Life form	Chorotype	Habitat
<i>Conyzanthus squamatus</i> (Spreng.) Tamamsch.	Hem	PL (Neophyte)	NM-SM
<i>Eclipta prostrata</i> (L.) L.	Th	PL	NM-SM
<i>Senecio vernalis</i> Waldst. & Kit.	Hem	ES, IT2	SM
<i>Siegesbeckia orientalis</i> L.	Th	PL	NI-SM
<i>Sonchus asper</i> (L.) Hill	Th	IT, M	NM
<i>Sonchus oleraceus</i> L.	Th	PL	NI-SM
<i>Tagetes minuta</i> L.	Th	PL (Neophyte)	SM
<i>Willemetia tuberosa</i> Fisch. & C.A.Mey. ex DC.	GT	Cau, Hyr, IT2	NI
<i>Xanthium brasiliacum</i> Vell.	Th	M, IT2	NM
<b>Betulaceae</b>			
<i>Alnus glutinosa</i> (L.) Gaertn.	Ph	Cau, Euxino-Hyr	NI-SI
<i>Alnus subcordata</i> C.A.Mey.	Ph	Endem (Hyr)	NI
<i>Carpinus betulus</i> L.	Ph	ES [Alborz]	NI-SI
<b>Boraginaceae</b>			
<i>Cynoglossum officinale</i> L.	Hem	ES, M, Euxino-Hyr [IT2,3,4]	NI
<i>Lindelofia kandavanensis</i> Bomm. & Gauba	Hem	Endem (Iran-Hyr)	NI-NM-SM
<i>Buglossoides purpureocaerulea</i> (L.) I.M. Johnst.	Hem	ES [M]	SM
<i>Lithospermum officinale</i> L.	GR	ES, M, IT2,3,4	SM
<i>Nonea lutea</i> (Desr.) A.DC.	Th	ES (Cau) [IT2]	NI-NM
<b>Brassicaceae</b>			
<i>Brassica toumefortii</i> Gouan	Th	M, SS, IT2,3, N Africa	NM
<i>Cardamine hirsuta</i> L.	Th	SCOS	NI
<i>Cardamine tenera</i> Boiss.	GR	Cau (Hyr)	NI
<i>Lepidium ruderale</i> L.	Hem	IT2	NM-SM
<i>Nasturtium officinale</i> R.Br.	Hem	ES, IT, M	NM
<b>Buxaceae</b>			
<i>Buxus hyrcana</i> P.ojark.	Ph	Endem (Hyr)	SI-SM
<b>Campanulaceae</b>			
<i>Campanula rapunculoides</i> L.	Hem	ES [IT, M]	SM
<b>Cannabaceae</b>			
<i>Celtis australis</i> L.	Ph	M, N Africa	SI-SM
<b>Caryophyllaceae</b>			
<i>Cerastium glomeratum</i> Thuill.	Th	PL	NI-NM-SM
<i>Minuartia hybrida</i> (Vill.) Schischk.	Th	M, ES (European Russia), IT1,2	SM
<i>Polycarpon tetraphyllum</i> (L.) L.	Th	M, IT2 [SS]	SM
<i>Stellaria media</i> Cirillo	Th	COS	NI-SI-NM-SM
<b>Amaranthaceae</b>			
<i>Chenopodium album</i> subsp. <i>album</i> L.	Th	SCOS	SM
<b>Convolvulaceae</b>			
<i>Calystegia sepium</i> (L.) R. Br.	GR	PL	NI-NM-SM
<i>Calystegia silvatica</i> (Kit.) Griseb.	GR	ES [IT-Azer]	SM
<b>Cornaceae</b>			
<i>Cornus australis</i> C.A.Mey.	Ph	ES, IT1,2	NI-SM
<b>Crassulaceae</b>			
<i>Sedum stoloniferum</i> S.G.Gmel.	Hem	Cau, Euxino-Hyr	NM
<b>Dipsacaceae</b>			
<i>Dipsacus pilosus</i> L.	Hem	ES	NM
<b>Ebenaceae</b>			
<i>Diospyros lotus</i> L.	Ph	Cau (Transcau), Euxino-Hyr [Himalaya]	NI-SI
<b>Euphorbiaceae</b>			
<i>Acalypha australis</i> L.	Th	PL (Neophyte)	NM
<i>Euphorbia amygdaloides</i> L.	GR	ES, M, N Africa	NI-SM
<i>Euphorbia peplus</i> L.	Th	PL	NI-SM
<i>Euphorbia</i> sp.	Th		NM
<i>Euphorbia turcomanica</i> Boiss.	Th	IT2,3,4, Cau	NM-SM
<i>Mercurialis perennis</i> L.	GR	ES [N Africa]	NI
<b>Fabaceae</b>			
<i>Albizia julibrissin</i> Durazz.	Ph	Euxino-Hyr [China+Japon]	SI
<i>Coronilla varia</i> L. subsp. <i>varia</i>	Hem	ES, M, IT2	NM-SM
<i>Gleditsia caspica</i> Desf.	Ph	Endem (Hyr) [Turk]	NI
<i>Lotus corniculatus</i> L.	Hem	PL	SM
<i>Medicago lupulina</i> L.	Hem	PL	NM
<i>Trifolium campestre</i> Schreb.	Th	ES, M, IT1,2, N Africa [SS]	NM-SM
<i>Trifolium resupinatum</i> L. var. <i>resupinatum</i>	Th	ES, M, IT2,3,4, N Africa	NM-SM
<i>Vicia tetrasperma</i> (L.) Schreb.	Hem	PL	NM
<b>Fagaceae</b>			
<i>Quercus castaneifolia</i> C.A.Mey.	Ph	Endem (Hyr) [Khor]	NI-SI
<b>Gentianaceae</b>			

Table 1. Checklist of identified plant species in Noor and Sisangan lowland forests

**Symbols and abbreviation used in Table 1:**

Life forms: Chamaephyte (Ch), Bulbose geophyte (GB), Geophyte with corm (GC), Rhizomatose geophyte (GR), Stoloniferous geophyte (GS), Tuber geophyte (GT), Hemicryptophyte (Hem), Helophyte (Hel), Hydrophyte (Hyd), Phanerophyte (Ph), Therophyte (Th).

Chorotypes: Caucasian (Cau), Cosmopolitan (COS), Endemic (En), Euro-Sibirian (ES), Euxino-Hyrcanian (Euxino-Hyr), Hyrcanian (Hyr), Irano-Turanian (IT), Mediterranean (M), Pluriregional (PL), Subcosmopolitan (SCOS), Saharo-Sindian (SS), North (N), South (S), East (E), West (W), Afghanistan (Afgh), Azerbaijan (Azer), Himalaya (Him), Khorasan (Khor), Mountains (Mts), Pakistan (Pak), Temperate (Temp), Transcaucasus (Transcau), Turkmenistan (Turk).

Habitats: Inside of Noor forest (NI), Margin of Noor forest (NM), Inside of Sisangan forest (SI), Margin of Sisangan forest (SM).

Taxon	Life form	Chorotype	Habitat
<b>Pteridophytes</b>			
<b>Aspleniaceae</b>			
<i>Asplenium adiantum-nigrum</i> (L.)	GR	PL	NI-SI
<i>Phyllitis scolopendrium</i> (L.) Newman	GR	PL (N Temp.)	NI-SI
<b>Dennstaedtiaceae</b>			
<i>Pteridium aquilinum</i> (L.) Kuhn	GR	COS	NI
<b>Dryopteridaceae</b>			
<i>Dryopteris pallida</i> (Bory) Fomin	GR	M [Hyr]	NI
<i>Polystichum aculeatum</i> (L.) Roth	GR	PL	NI
<i>Polystichum woronowii</i> Fomin	GR	Euxino-Hyr	NI
<b>Equisetaceae</b>			
<i>Equisetum telmateia</i> Ehrh.	GR	PL (N Temp.)	NI-NM
<b>Onocleaceae</b>			
<i>Matteuccia struthiopteris</i> (L.) Tod.	GR	PL	NI
<b>Ophioglossaceae</b>			
<i>Ophioglossum vulgatum</i> L.	GR	PL (N Temp.)	NI-SI
<b>Polypodiaceae</b>			
<i>Polypodium vulgare</i> L.	GR	PL	NI-SI
<b>Pteridaceae</b>			
<i>Adiantum capillus-veneris</i> L.	GR	PL	NI
<i>Pteris cretica</i> L.	GR	PL	NI-SI
<i>Pteris dentata</i> Forssk.	GR	PL	NI
<b>Woodsiaceae</b>			
<i>Athyrium filix-femina</i> (L.) Roth.	GR	PL (N Temp.)	NI
<b>Angiosperms</b>			
<b>Dicotyledones</b>			
<b>Adoxaceae</b>			
<i>Sambucus ebulus</i> L.	GR	ES, IT 2, M, N Africa	NI-NM-SM
<b>Amarantaceae</b>			
<i>Alternanthera sessilis</i> (L.) DC.	Th	PL (Neophyte)	NM
<i>Amaranthus hybridus</i> L.	Th	PL (Neophyte)	SM
<i>Amaranthus blitum</i> L.	Th	PL	SM
<b>Apiaceae</b>			
<i>Bupleurum marschallianum</i> C.A.Mey.	GR	Cau, Hyr [Azer]	NM-SM
<i>Eryngium caucasicum</i> Trautv.	Hem	IT 2, 3, 4, Cau	NM-SM
<i>Pimpinella affinis</i> Ledeb.	Hem	IT 2, Cau, Euxino-Hyr	NI-SM
<i>Sanicula europaea</i> L.	Hem	ES [M]	NI
<i>Torilis arvensis</i> Link	Th	PL	NI-SI
<b>Apocynaceae</b>			
<i>Periploca graeca</i> L.	Ph	M(E), ES(Euxino-Hyr+S)	NI-NM
<b>Araliaceae</b>			
<i>Hedera pastuchovii</i> Woronow	Ph	Cau (Transcau), Hyr	NI-SI
<b>Asteraceae</b>			
<i>Artemisia annua</i> L.	Th	M, IT 2, 3, Cau	NM-SM
<i>Bidens tripartita</i> Bigelow	Th	PL	NM
<i>Carduus arabicus</i> Jacq.	Th	M, IT 1, 2	NM-SM
<i>Carpesium abrotanoides</i> L.	Hem	PL	SM
<i>Carpesium cernuum</i> L.	Hem	PL	NI-NM-SM
<i>Cichorium intybus</i> L.	Hem	PL	NM-SM
<i>Cirsium arvense</i> (L.) Scop.	Hem	PL	NI
<i>Conyza bonariensis</i> (L.) Cronquist	Th	COS	NI-NM-SM
<i>Conyza canadensis</i> (L.) Cronquist	Th	SCOS (Neophyte)	SM

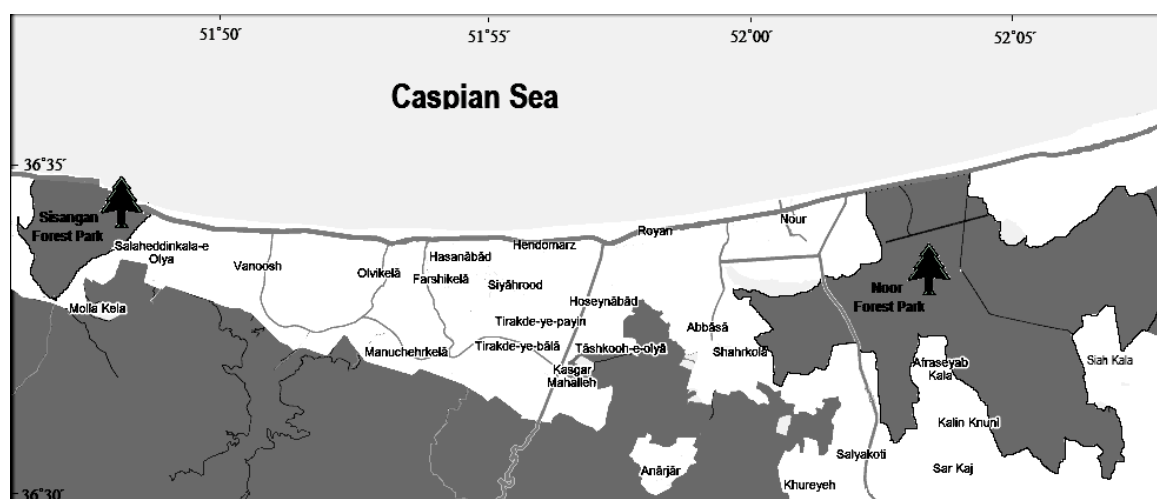


Figure 1. Locations of the Noor and Sisangan forests in the Hyrcanian lowland area

### Data collection

Data was collected during March to November of 2011. Floristic data were collected by using 62 relevés with the area of 400 m<sup>2</sup> surface in Noor and 20 relevés with the area of 100 m<sup>2</sup> in Sisangan forests. Different sizes of relevés were allocated to different vegetation physiognomy and plant density and richness. Sisangan forest was covered mainly by mono-dominant *Buxus hyrcana* trees or shrubs with low herbaceous ground vegetation. Other species found beyond the relevés were also considered for our floristic survey. Plant determination was carried out using Rechinger (1963-2010), Assadi *et al.*, (1988-2011), Davis (1965-1988), Komarov (1968-1980), Townsend *et al.*, (1966-1985), Ghahreman (1979-2003). Moreover, the ferns were identified using Khoshravesh *et al.*, (2009). All plant names and their authors were checked by IPNI website ([www.ipni.org](http://www.ipni.org)). The classification system of The angiosperm phylogeny group (2009) was used for family names. The life form of each species followed Raunkiaer's classification system (Raunkiaer, 1934). The terminology and delimitation of the main phytocoria was based on the concepts applied by Zohary (1973), Léonard (1988) and Takhtajan (1986). In this article, PL (Pluriregional elements) are plants ranging in distribution over three phytogeographical regions, SCOS (sub-cosmopolitan elements) are plants ranging in distribution over most continents but not all of them and COS (cosmopolitan elements) referring to plants that have a broad worldwide distribution. Floristic similarity between two forests was evaluated using Sørensen's (1948) similarity index.

### Results

Floristic study in Noor forest showed the occurrence of 185 plant species belonging to 149 genera and 68 families while the number of determined plants in the Sisangan forest was 137 species belonging to 111 genera and 57 families. Totally, 225 plant species from 176 genera and 77 families were collected from these two forests. Among the 77 plant families, nine families were Pteridophytes and 68 were Angiosperms (Table 1). Eudicots with 56 families, 123 genera and 160 species were the richest group, while monocots had 12 families, 40 genera and 50 species in the studied flora (Table 2).

vegetation and floristic investigations have been conducted especially in the mountain and sub-mountain forests of Hyrcanian area (e.g. Djazirei, 1965; Mobayen and Tregubov, 1970; Dorostkar and Noirfalise, 1976; Assadollahi, 1980; Mossadegh, 1981; Hamzeh'ee, 1994; Akhiani, 1998; Klein, 2001; Akbarinia *et al.*, 2004; Esmailzadeh *et al.*, 2007; Atashgahi *et al.*, 2009; Naqinezhad *et al.*, 2010). However, necessity of an extensive floristic and vegetation studies in the remained lowland patches is felt more than ever. There are some specific investigations particularly on the lowland areas (Rastin, 1983; Tabari *et al.*, 2002; Ghahreman *et al.*, 2006; Hamzeh'ee *et al.*, 2008; Naqinezhad *et al.*, 2008; Ghahremaninejad *et al.*, 2011; Asadi *et al.*, 2011). For instance, Tabari *et al.* (2002) provided some information on the distribution and vegetation structure of *Fraxinus excelsior* in the lowland Hyrcanian forests. Likewise, a detailed vegetation and floristic survey was done particularly on the *Alnus glutinosa* subsp. *barbata* patches in northern Iran (Ghahreman, *et al.*, 2006; Hamzeh'ee *et al.*, 2008).

Noor and Sisangan are two large patches of such lowland forests classified as “natural forest parks” in the context of “Iranian Natural Resources” (Mazandaran Natural Resources Office, 2012). It may be proclaimed that these forests are the only remnants of Caspian lowland forests, due to the destruction and severe damage by animals and humans in recent years (Barzehkar, 1994; Hamzeh'ee *et al.*, 2008). Sisangan is known by the occurrence of pure stands of *Buxus hyrcana*, an endemic Hyrcanian woody species which has been largely destroyed from the other parts of the Hyrcanian forest. Likewise, one of the main habitats of *Populus caspica*, a rare tree in the area, is located in Noor forest. Detailed floristic and vegetation studies should be conducted to provide a basic framework for further ecological and conservational studies on these highly threatened ecosystems. By now, no comprehensive study has been accomplished in these two areas particularly on all parts of the forests and forest margins. The aims of the current investigation are (1) representing a complete and updated checklist of all plant species of these two areas; (2) assessing some species-related characters such as life form and chorology in the areas; and (3) comparing the flora of the two forest areas with each other and with other forest areas studied in the Hyrcanian area.

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Noor forest is located between 52° 00' to 52° 06' E and 36° 32' to 36° 34' N with a 3600 hectare surface area. The forest is surrounded by main transitional Noor-Nowshahr road in the north, Noor-Chamestan road in the west, Afraseyabkola village in the south and Hashemrud river and Izdeh forests in the east. The area is generally flat. Sisangan forest is located between 51° 47' to 51° 49' E and 36° 33' to 36° 34' N, in 27 km east of Nowshahr, Mazandaran. The total area of the forest is approximately 602 hectare and is generally flat. The forest faces Caspian Sea and main transitional road in the north, Tooskatook village in the west, Salahedinkala in the east and south (Figure 1).

## A contribution to flora, life form and chorology of plants in Noor and Sisangan lowland forests

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### Abstract

Lowland Hyrcanian (Caspian) areas possess a number of important remnant patches of deciduous Euro-Siberian forests distributed sparsely in the three Iranian provinces, Guilan, Mazandaran and Golestan. Noor and Sisangan are two large patches of such lowland forests classified as “natural forest parks” in the context of “Iranian Natural Resources”. In spite of a few local studies, broad knowledge upon the flora and vegetation of these areas are lacking. A total of 225 species belonging to 175 genera and 77 plant families were collected from the studied areas. The largest families in terms of species richness, were Poaceae (28 spp.), Asteraceae (18 spp.) and Rosaceae (9 spp.), respectively. The genera with the largest number of species were *Carex* (6 spp.), *Veronica* (5 spp.) and *Euphorbia*, *Polygonum*, *Solanum* (each with 4 spp.), respectively. In the assessment of life form spectrum, the dominant life forms were therophytes (30.2%), followed by the geophytes (27.1%), hemicryptophytes (20.9%) and phanerophytes (18.2%). The flora was mostly composed of pluriregional elements with 60 taxa (27.3%), followed by Euro-Siberian/Irano-Turanian/Mediterranean elements with 43 taxa (19.5%). Life form spectra and chorotype percentages were discussed for each study area separately. According to Sørensen's (1948) similarity index, there was a remarkable similarity between two forest areas. Noor and Sisangan forests were highly threatened ecosystems in case of species loss and changing natural communities due to occurrence of anthropogenic and over-grazing effects.

**Key words:** Flora, Lowland Hyrcanian forest, Life form, Noor and Sisangan

### Introduction

Hyrcanian (Caspian) forests extend from Talish area of Azerbaijan Republic in the west to Golestan National Park of Golestan in the east and constitute a green cover across the northern slopes of the Alborz Mountains. These forests are related to an unique climate with high annual precipitation (ranging from 600 to 2000 mm) and considered to be largely compatible with the Euro-Siberian forest structure (Frey and Probst, 1986). The forests of the south Caspian area have been severely degraded and deforested; particularly, in the alluvial and lowlands where only small remnants of the forests exist now. Due to this land conversion, many plant species were restricted to isolated remnants of a formerly more widespread lowland habitats (Ghahreman *et al.*, 2006). A considerable number of

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(Keshavarzi *et al.*, 2007). As this species is tetraploid with  $2n=28$  (Baldini, 1995) it is not surprising. Sometimes this variability is related to soil conditions. Rich or poor soils cause difference of morphological features of *P. minor* individuals. Despite high variation of this species *P. brachystachys* ( $2n=12$ ) and *P. paradoxa* ( $2n=14$ ) are diploids with more similar populations (Baldini, 1995). As *P. arundinacea* is out of selection it was supposed to show more variability. We thought that low variability could be due to insufficient sampling from localities. A vast field study and new collections from different species of *Phalaris* is recommended. As enzyme electrophoresis are capable presenting inter- and intra- specific variations and also some morpho-geographic subspecies could be separated.

## R

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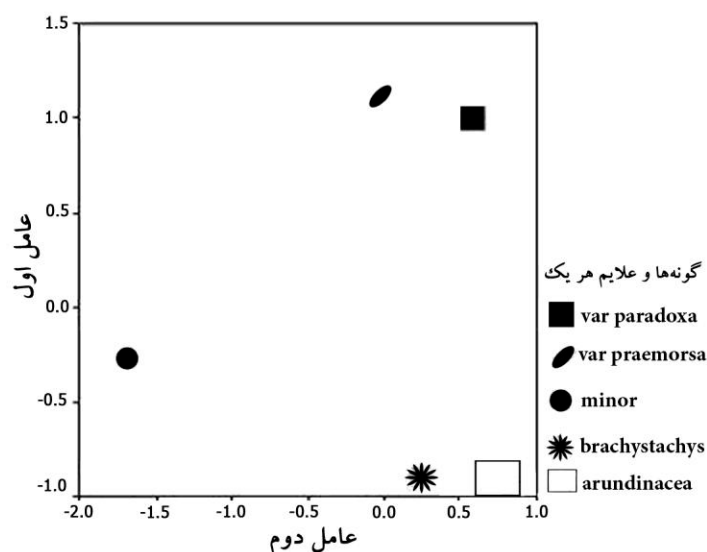


Figure 3. PCA ordination of the *Phalaris* species based on SDS-PAGE characters

Authors are convinced of existence of two centers of variation for this taxon in the whole world: Mediterranean region with 11 species and South West of USA with 4 species. There are four species of this genus in Iran, all belonging to the Old World species group (Baldini, 1993). *P. minor* has a vast distribution region in Irano-Touranian and Saharo-Sindian phytogeographic region (Keshavarzi *et al.*, 2007). *P. paradoxa* has a high morphological variability. Morphological studies in Iran revealed that two variations of *P. paradoxa* (*P. paradoxa* var. *praemorsa* and *P. paradoxa* var. *paradoxa*) make a closely related group also with *P. minor*. *P. brachystachys* is related to these taxa at the level 10. *P. arundinacea* which is the only perennial species of these taxa in Iran which makes a separate cluster and it is far from the other species.

*Phalaris* species of Iran showed differences in leaf anatomical structure and leaf dorsal epidermis (Keshavarzi *et al.*, 2009). Main differences were observed in hair type and frequency, stomata number in dorsal leaf area, stomata size and general outline of leaf cross sections. These were of diagnostic value and an identification key was made based on these features. Anatomical studies revealed that two varieties of *P. paradoxa* were distinguished from each other while morphological studies (Keshavarzi *et al.*, 2011) indicated a close relationship between these two. Main morphological diagnostic features for these varieties, which are used in identification keys, are the shape of rudimentary spikelets and ligule surface. In *P. paradoxa* var. *praemorsa* rudimentary spikelets were club like and ligule surface lacked hair while *P. paradoxa* var. *paradoxa* showed no club like rudimentaries and had hairy ligule.

In the meantime, anatomical and morphological studies both confirmed the separation of the studied taxa and the present result of SDS-PAGE was in accordance with previous results although there were some differences in clustering patterns.

Electrophoretic data also confirmed a close relationship and identity between the two variations of *P. paradoxa*. *P. arundinacea* was very different from the other species of this genus morphologically, but it was located near them according SDS-PAGE data. Hucle and Matus (1999) stated that *P. minor*, being an auto-tetraploid taxon, is a highly variable species within this genus according results of studying its enzyme electrophoretic patterns. We found out high morphological variability in different accessions of this species as well

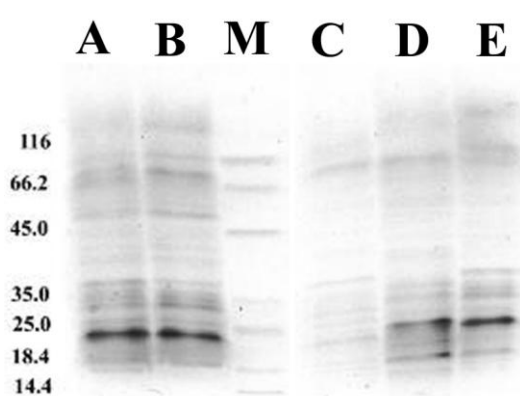


Figure 1: Seed protein banding profile of wild *Phalaris* species of Iran. A) *P. paradoxa* var. *paradoxa*; B) *P. paradoxa* var. *praemorsa*; C) *P. brachystachys*; D) *P. arundinacea*; E) *P. minor*; M) marker.

Table 4. Factor analysis results based on SDS-PAGE electrophoretic characters for *Phalaris* species of Iran

Band no.	1 <sup>st</sup> factor	2 <sup>nd</sup> factor	3 <sup>rd</sup> factor
1	--	--	0.79
2	--	--	0.70
3	--	--	0.70
7	0.84	--	--
8	--	5.10	--
10	0.84	--	--
11	0.96	--	--
13	0.96	--	--
14	0.96	--	--
15	0.96	--	--
16	0.84	--	--
17	0.96	--	--
18	0.84	--	--
21	0.96	--	--
24	0.96	--	--

Cluster analysis result is shown in WARD dendrogram (Figure 2). UPGMA dendrogram was similar to WARD one. The taxa are clearly separated based on electrophoretic data of seed storage proteins. Results revealed that two varieties of *P. paradoxa* as var. *praemorsa* and var. *paradoxa* were closely related. High similarity index is a reflex of genomic identity ( $J=0.857$ ). Dendrogram showed close relationship and high protein similarity ( $J=0.583$ ) between *P. arundinacea* and *P. brachystachys*. On the other hand, *P. minor* comprised a separate cluster itself. Ordination of studied taxa based on PCA (Figure 3), showed that there was a concordance with cluster analysis.

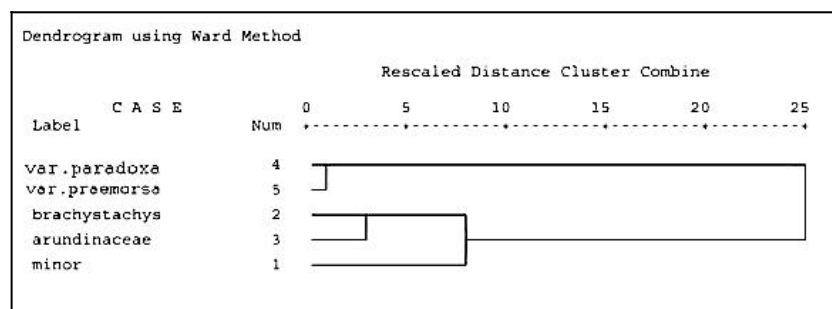


Figure 2. Dendrogram depicting clustering by WARD method of Taxa of *Phalaris*, by cluster analysis of seed storage protein

Table 2. Seed storage protein banding profiles of seed samples of *Phalaris* species native to Iran (1- band is present in the seed sample, 0- band is absent in the seed sample).

Taxon	<i>P. paradoxa</i> var. <i>praemorsa</i>	<i>P. paradoxa</i> var. <i>paradoxa</i>	<i>P. minor</i>	<i>P. brachystachys</i>	<i>P. arundinacea</i>
1- 0.26	1	1	1	0	1
2 -0.27	0	1	0	0	1
3- 0.29	0	1	0	0	1
4 -0.31	1	1	1	1	1
5- 0.33	1	1	1	1	1
6-0.35	1	1	1	1	1
7- 0.36	1	1	1	0	0
8- 0.37	0	0	0	1	1
9-0.38	1	0	0	0	0
10-0.39	1	1	1	0	0
11-0.40	1	1	0	0	0
12-0.41	0	0	0	0	1
13-0.42	1	1	0	0	0
14-0.43	1	1	0	0	0
15-0.45	1	1	0	0	0
16-0.47	1	1	1	0	0
17-0.48	1	1	0	0	0
18-0.51	1	1	1	0	0
19-0.53	0	0	1	0	0
20-0.55	1	1	1	1	1
21-0.57	1	1	0	0	0
22-0.58	0	0	1	1	1
23-0.60	1	1	0	1	0
24-0.62	1	1	0	0	0
25-0.65	1	1	1	1	1

Table 3. Jaccard similarity index based on electrophoretic data of seed storage protein in *Phalaris* taxa native to Iran

Case	Jaccard Measure				
	1:M	2:B	3:A	4:P.P	5:P.R
1:M <i>P. minor</i>	-	0.429	0.438	0.455	0.476
2:B <i>P. brachystachys</i>	0.429	-	0.583	0.273	0.286
3:A <i>P. arundinacea</i>	0.438	0.583	-	0.348	0.250
4:P.P <i>P. paradoxa</i> var. <i>paradoxa</i>	0.455	0.273	0.348	-	0.857
5:P.R <i>P. paradoxa</i> var. <i>praemorsa</i>	0.476	0.286	0.250	0.857	-

morphology in Iran. The objective of this study was to assess the level of seed electrophoretic patterns of *Phalaris* taxa in Iran. We tried to reveal the degree of coincidence between morphological variations and SDS-PAGE profiles for *Phalaris* native to Iran for the first time.

### Materials and Methods

In this study, five populations were chosen to study electrophoretic pattern of seed storage proteins. Seed samples were obtained from the sources indicated in Table 1. 1 gr of seed was used from each accession. Voucher specimens for this study were collected from the wild and all have been deposited at the Herbarium of Alzahra University, Tehran.

Table 1. Origin of seed samples of *Phalaris* species native to Iran

Taxon	Voucher details
<i>P. minor</i>	Tehran, Vanak 1700 m, Keshavarzi, 83m19.
<i>P. brachystachys</i>	Tehran, Karaj, Mardabad, 1267 m, Keshavarzi, 1383b8.
<i>P. paradoxa</i> var. <i>paradoxa</i>	Khuzestan, 15 km to Izae, 827 m, Nanaii, 85pp3.
<i>P. paradoxa</i> var. <i>praemorsa</i>	Mazandaran, Sari, Sameskandeh, 100 m, Khaksar, 85pp7.
<i>P. arundinacea</i>	Tehran to Chalous, Dizin, 2700 m, Khaksar, 85a 1.

The final extract was loaded on SDS PAGE and stained by coomassie brilliant blue (Lammeli, 1970). We used Jaccard similarity coefficient. In statistical analysis, presence or absence of each band was considered as a qualitative feature. Then, the dendrogram was constructed using WARD hierarchical and UPGMA clustering by SPSS software ver. 11. In order to find the most variable protein band in studied taxa, principal component analysis was done. Standard proteins ( $\beta$ -galactosidase, Ovalbumin, Lactate dehydrogenase, lactoglobulin- $\beta$ , Lysozyme and Bovine serum albumin) were used to evaluate the molecular weight of the unknown proteins. The protein density was determined by Bradford Protocol. Banding patterns were studied and R.F. values were measured. We used Jaccard similarity coefficient.

### Results and Discussion

SDS-Page electrophoretic data were analyzed (Table 2). Jaccard similarity index was evaluated (Table 3). Totally 25 bands were observed for these taxa. The 4<sup>th</sup>, 5<sup>th</sup>, 6<sup>th</sup>, 20<sup>th</sup> and 25<sup>th</sup> bands were common in studied taxa. While band numbers 11, 13, 14, 15, 17, 21 and 24 were only observed in *P. paradoxa* var. *paradoxa* and *P. paradoxa*. Band number 9 was exclusively observed in *P. paradoxa* var. *praemorsa*. Merely in *P. arundinacea* the band number 12 was shown. Band number 19 was found only in *P. minor*. All of the studied taxa had band number one but not *P. brachystachys*. The highest numbers of bands were observed in *P. paradoxa* var. *paradoxa* and the least one in *P. brachystachys* (Figure 1).

In order to find most variable protein band in the studied taxa, principal component analysis was implemented. Primitive analysis showed that three first factors were responsible for the 95% of total studied variation in taxa. In the first factor with almost 61% of the total variation, bands number 1, 7, 10, 11, 14, 15, 16, 17, 18, 21 and 24 had the highest positive correlations. Bands number 8, 12 and 22 had the highest negative correlation. In the second factor with near 20% of observed variation, band number 8, had the highest positive correlation and band number 19 had the highest negative one. In the third factor with 14.16% of total variation, bands number one to three had the highest positive correlations (Table 4).

## Biosystematic study of *Phalaris* L. species (Poaceae) in Iran

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### Abstract

This study dealt with banding patterns of seed storage proteins using sodium dodecylsulphate-polyacrylamide gel electrophoresis (SDS-PAGE) with extract of bulked seeds of *Phalaris* L. species of Iran. The results showed that two varieties of *P. paradoxa* L. as var. *praemorsa* and var. *paradoxa* L. were closely related. A close relationship and high protein similarity ( $J=0.583$ ) were found between *P. arundinacea* L. and *P. brachystachys* Link.. Electrophoretic results were compared with previous anatomical and morphological studies.

**Key words:** *Phalaris*, Taxon relationships, SDS-PAGE

### Introduction

The genus *Phalaris* L. has had a complicated taxonomic and nomenclatural history (Baldini, 1993, 1995). It comprises 22 species of annual and perennial grasses in temperate regions throughout the world. These are commonly adventives species of open habitats. There are 4 species and 5 taxa of *Phalaris* in Iran (Bor, 1970): *P. minor* Retz., *P. brachystachys* Link., *P. paradoxa* L. (with 2 varieties) and *P. arundinacea* L.. These species are distributed in various regions of Iran. They are among important forage plants. Members of the genus *Phalaris* display many variations on the standard structure of the inflorescence (Bor, 1968). There has been no report of systematic study on *Phalaris* species of Iran.

Many authors demonstrated that sodium dodecylsulphate-polyacrylamide gel electrophoresis (SDS-PAGE) of proteins extracted from seeds produces a composite pattern for the phenotypes in the analyzed population (Yusaf *et al.*, 2006; Sheidai *et al.*, 2008). SDS-PAGE can be used to characterize the seed protein banding profiles of species and cultivars in several grass genera, compare the cultivars of different geographical origin and provide taxonomically useful descriptors that are substantially free from environmental influence. This procedure has provided useful data for many grasses as *Lolium* L. and *Festuca* L. complex (Aiken *et al.*, 1992), *Dactylis* L. and *Leucopoa* Griseb. (Aiken *et al.*, 1998). There was no report of SDS-PAGE in *Phalaris* species but limited variation was found out by means of Isozymes in annual *Phalaris* species (Hucle and Matus, 1999).

Seed protein banding profiles have proved to provide informative supplementary data for morphological features in resolving problems in the grass taxonomy. In this research, we tried to resolve *Phalaris* species relationship by using SDS-PAGE as supplementary data for

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Taxa	Habitat	Life form	Chorotype	Locality	Hb. No. (FAR)
<b>Poaceae</b>					
<i>Aegilops tauschii</i> Coss.	Ma	Thr	ES, IT, M	AW	13535
<i>Agrostis stolonifera</i> L.	Ma	Geo	ES, IT, M	BW	13605
<i>Alopecurus arundinaceus</i> Poir. var. <i>arundinaceus</i>	Ma	Geo	PL	AW	13534
<i>Arundo donax</i> L.	Ma	Geo	ES-IT-M	MW, BW	13794
<i>Bromus japonicus</i> Thunb. var. <i>japonicus</i>	Ma	Thr	PL	BW	13596
<i>Briza minor</i> L.	Ma	Thr	ES, M	BW	13723
<i>Calamagrostis epigejos</i> (L.) Roth	Ma	Geo	PL	MW	13726
<i>Catabrosa aquatica</i> P. Beauv.	Em	Hyd	PL	MW, LW	13500
<i>Cynodon dactylon</i> (L.) Pers.	Ma	Geo	PL	MW	13795
<i>Dactylis glomerata</i> L.	Ma	Hem	PL	RW	13722
<i>Digitaria sanguinalis</i> (L.) Scop.	Ma	Thr	PL	MW, AW	13667
<i>Echinochloa crus-galli</i> (L.) P. Beauv.	Em	Thr	PL	RW	13585
<i>Eleusine indica</i> (L.) Gaertn.	Ma	Thr	PL	MW	13635
<i>Lolium perenne</i> L.	Ma	Hem	COSM	RW	13567
<i>Lophochloa phleoides</i> (Vill.) Richb.	Ma	Thr	PL	RW	13588
<i>Paspalum dilatatum</i> Poir.	Ma	Geo	PL	BW, MW	13604
<i>Paspalum distichum</i> L.	Em	Geo	COSM	RW, MW, LW	13587
<i>Phalaris arundinacea</i> L.	Ma	Geo	PL	MW	13724
<i>Phleum paniculatum</i> Huds. var. <i>ciliatum</i> (Boiss.) Bor	Ma	Thr	ES	MW	13725
<i>Phragmites australis</i> (Cav.) Steud.	Em	Hyd	COSM	MW, RW, BW	13732
<i>Polypogon monspeliensis</i> (L.) Desf.	Ma	Thr	PL	MW, LW	13502
<i>Polypogon semiverticillatus</i> (Forssk.) H. Hyl.	Ma	Thr	PL	MW	13664
<i>Setaria glauca</i> (L.) P. Beauv.	Ma	Thr	PL	LW, BW	13621
<i>Sorghum halepense</i> (L.) Pers.	Ma	Geo	PL	MW	13659
<b>Potamogetonaceae</b>					
<i>Potamogeton crispus</i> L.	Su	Hyd	PL	MW, LW	13560
<i>Potamogeton lucens</i> L.	Su	Hyd	PL	RW, LW, MW	13575
<i>Potamogeton nodosus</i> Poir.	Su	Hyd	PL	MW	13647
<i>Potamogeton pectinatus</i> L.	Su	Hyd	COSM	LW, MW	13558
<b>Sparganiaceae</b>					
<i>Sparganium erectum</i> L. subsp. <i>neglectum</i> (Beeby) K. Richter	Em	Geo	ES	AW, LW, MW	13536
<b>Typhaceae</b>					
<i>Typha angustifolia</i> L.	Em	Hyd	PL	RW	13713
<i>Typha caspica</i> Pobed.	Em	Hyd	ES (Eux-Hyr)	AW	13738
<i>Typha domingensis</i> Pers.	Em	Hyd	PL	LW	13608
<i>Typha latifolia</i> L.	Em	Hyd	COSM	RW, MW	13797

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Taxa	Habitat	Life form	Chorotype	Locality	Hb. No. (FAR)
<i>Veronica anagallis-aquatica</i> L.	Em	Hem	PL	AW, LW	13551
<i>Veronica persica</i> Poir.	Ma	Thr	PL	BW	13598
<i>Veronica polita</i> Fr.	Ma	Thr	PL	BW	13599
<b>Solanaceae</b>					
<i>Datura innoxia</i> Mill.	Ma	Thr	PL	MW	13771
<i>Datura stramonium</i> L.	Ma	Thr	PL	MW	13773
<i>Solanum persicum</i> Willd. ex Roem. & Schult. subsp. <i>persicum</i>	Ma	Pha	ES, IT	AW, MW	13743
<i>Solanum nigrum</i> L.	Ma	Thr	PL	BW, MW	13703
<b>Tamaricaceae</b>					
<i>Tamarix ramosissima</i> Ledeb.	Ma	Pha	PL	LW	13614
<b>Urticaceae</b>					
<i>Parietaria officinalis</i> L.	Ma	Hem	ES	MW	13511
<i>Urtica dioica</i> L.	Ma	Hem	PL	MW	13798
<i>Urtica urens</i> L.	Ma	Thr	PL	MW	13799
<b>Verbenaceae</b>					
<i>Phyla nodiflora</i> (L.) Greene	Ma	Hem	PL	BW, MW	13760
<i>Verbena officinalis</i> L.	Ma	Hem	PL	RW, MW	13577
<b>Monocotyledones</b>					
<b>Alismataceae</b>					
<i>Alisma plantago-aquatica</i> L.	Em	Hyd	COSM	LW, BW	13549
<i>Sagittaria sagittifolia</i> L.	Em	Hyd	ES-IT-M	LW	13686
<b>Butomaceae</b>					
<i>Butomus umbellatus</i> L.	Em	Hyd	ES-IT-M	RW, LW, BW, MW	13610
<b>Cyperaceae</b>					
<i>Bolboschoenus affinis</i> Drobow	Em	Hyd	PL	RW, MW, LW	13581
<i>Carex divulsa</i> Stokes subsp. <i>divulsa</i>	Ma	Geo	ES-IT-M	RW	13584
<i>Carex riparia</i> (R. Br.) Poir.	Em	Hyd	ES	MW, RW	13525
<i>Carex sylvatica</i> Huds.	Em	Geo	ES-M	AW	13721
<i>Carex songorica</i> Kar. & Kir.	Em	Geo	IT, ES	MW, RW, AW	13517
<i>Cyperus alternifolius</i> L.	Ma	Geo	PL	MW	13656
<i>Cyperus difformis</i> L.	Ma	Thr	PL	MW	13630
<i>Cyperus fuscus</i> L.	Em	Thr	PL	LW, MW	13687
<i>Cyperus longus</i> L.	Ma	Geo	ES-IT-M	MW	13772
<i>Cyperus odoratus</i> L. subsp. <i>Transcaucasicus</i> (Kuk.) Kukkonen	Ma	Geo	ES, IT	BW, MW	13643
<i>Cyperus pygmaeus</i> Rottb.	Ma	Thr	PL	MW	13632
<i>Cyperus rotundus</i> L.	Ma	Hem	COSM	RW, BW, MW	13564
<i>Cyperus serotinus</i> Rottb.	Em	Hyd	PL	AW	13736
<i>Fimbristylis bisumbellata</i> Bubani	Ma	Thr	PL	BW, MW	13709
<i>Pycreus flavesence</i> (L.) Reichenb.	Em	Geo	PL	MW	13680
<i>Pycreus flavidus</i> (Retz.) T.Koyama	Em	Thr	PL	LW	13720
<i>Schoenoplectus lacustris</i> (L.) Palla	Em	Hyd	ES, IT	MW, LW, RW	13545
<i>Schoenoplectus litoralis</i> (Schrad.) Palla	Em	Hyd	ES, IT, M	AW	13526
<i>Schoenoplectus mucronatus</i> (L.) Palla	Em	Hyd	PL	BW, AW	13696
<b>Iridaceae</b>					
<i>Iris pseudacorus</i> L.	Em	Hyd	ES	MW, LW, AW	13515
<b>Juncaceae</b>					
<i>Juncus articulatus</i> L.	Ma	Geo	PL	BW, AW	13700
<i>Juncus inflexus</i> L.	Ma	Hel	PL	AW	13740
<i>Juncus littoralis</i> C.A.Mey.	Ma	Geo	IT-M	LW, MW	13616
<i>Juncus acutus</i> L.	Ma	Geo	PL	LW	13625
<b>Lemnaceae</b>					
<i>Lemna minor</i> L.	Fl	Hyd	PL	MW, LW, AW	13658
<b>Hydrocharitaceae</b>					
<i>Najas graminea</i> Delile	Su	Thr	PL	MW	13731

Taxa	Habitat	Life form	Chorotype	Locality	Hb. No. (FAR)
<i>Lythrum salicaria</i> L.	Em	Hel	PL	MW, BW, RW	13756
<b>Malvaceae</b>					
<i>Abutilon theophrasti</i> Medik.	Ma	Thr	PL	MW, RW	13670
<b>Moraceae</b>					
<i>Ficus carica</i> L. subsp. <i>carica</i>	Ma	Pha	IT-M	AW	13786
<i>Morus alba</i> L.	Ma	Pha	IT	AW	13785
<b>Nelumbonaceae</b>					
<i>Nelumbium nuciferum</i> Gaertn.	Fl	Hyd	PL	MW	13788
<b>Nymphaceae</b>					
<i>Nympha alba</i> L.	Fl	Hyd	ES-M	AW, MW, BW, LW, RW	13787
<b>Onagraceae</b>					
<i>Epilobium hirsutum</i> L.	Ma	Geo	PL	LW, AW	13734
<b>Oxalidaceae</b>					
<i>Oxalis corniculata</i> L.	Ma	Thr	PL	RW, BW	13571
<b>Phytolaccaceae</b>					
<i>Phytolacca americana</i> L.	Ma	Hem	PL	MW	13789
<b>Plantaginaceae</b>					
<i>Plantago major</i> L.	Ma	Hem	PL	LW, BW	13540
<b>Polygonaceae</b>					
<i>Polygonum aviculare</i> L.	Ma	Thr	PL	BW	13757
<i>Polygonum barbatum</i> L.	Ma	Geo	PL	BW	13708
<i>Polygonum hydroppiper</i> L.	Ma	Thr	ES, IT	MW	13776
<i>Polygonum hyrcanicum</i> Rech. f.	Ma	Hem	ES	BW, LW	13759
<i>Polygonum lapathifolium</i> L. subsp. <i>lapathifolium</i>	Ma	Thr	ES, IT	MW, BW	13745
<i>Polygonum patulum</i> M. Bieb.	Ma	Thr	ES, IT	LW	13607
<i>Polygonum persicaria</i> L.	Ma	Thr	PL	RW, AW	13579
<i>Rumex pulcher</i> L.	Ma	Hem	ES, IT, M	RW	13569
<i>Rumex sanguineus</i> L.	Ma	Hem	ES	RW	13790
<b>Portulacaceae</b>					
<i>Portulaca oleracea</i> L.	Ma	Thr	ES, IT, M	MW	13791
<b>Primulaceae</b>					
<i>Anagalis arvensis</i> L.	Ma	Thr	PL	MW, LW	13550
<i>Samolus valerandi</i> L.	Em	Hem	PL	LW, MW	13554
<b>Punicaceae</b>					
<i>Punica granatum</i> L.	Ma	Pha	ES, IT	LW	13556
<b>Ranunculaceae</b>					
<i>Batrachium trichophyllum</i> (Chaix) Bosch	Su	Hyd	PL	AW, LW, MW	13537
<i>Ranunculus ophioglossifolius</i> Vill.	Em	Thr	ES, IT, M	RW	13589
<i>Ranunculus marginatus</i> d'Urv.	Ma	Thr	PL	MW	13651
<i>Ranunculus scleratus</i> L.	Em	Thr	PL	AW	13532
<b>Rosaceae</b>					
<i>Potentilla reptans</i> L.	Ma	Hem	ES, IT	LW, RW	13792
<i>Rubus caesius</i> L.	Ma	Pha	ES, IT	LW, RW	13618
<i>Rubus hyrcanus</i> Juz.	Ma	Pha	ES	LW	13716
<i>Rubus sanctus</i> Schreb.	Ma	Pha	ES, IT	LW, RW	13617
<b>Rubiaceae</b>					
<i>Galium elongatum</i> C. Presl	Em	Hyd	ES	BW	13593
<i>Galium ghilanicum</i> Stapf	Ma	Thr	ES, IT, M	RW, LW	13582
<b>Salicaceae</b>					
<i>Populus nigra</i> L.	Ma	Pha	ES, IT, M	RW, MW	13793
<i>Salix alba</i> L.	Ma	Pha	ES, IT	AW, LW	13796
<b>Scrophulariaceae</b>					
<i>Kickxia elatine</i> (L.) Dumort.	Ma	Thr	M	MW, LW	13662
<i>Verbascum punalense</i> Boiss. & Buhse	Ma	Hem	ES, IT	LW	13684

Taxa	Habitat	Life form	Chorotype	Locality	Hb. No. (FAR)
<i>Rorripa islandica</i> (Oeder) Borbas	Em	Geo	PL	AW, LW	13528
<i>Sisymbrium irio</i> L.	Ma	Thr	PL	MW, BW	13641
<b>Callitrichaceae</b>					
<i>Callitriche brutia</i> Petagna	Su	Hem	ES-M	AW, MW	13538
<b>Campanulaceae</b>					
<i>Campanula rapunculus</i> L.	Ma	Hem	ES (Hyr)	MW	13510
<b>Caprifoliaceae</b>					
<i>Sambucus ebulus</i> L.	Ma	Geo	PL	LW, MW, RW	13611
<b>Caryophyllaceae</b>					
<i>Silene latifolia</i> Poir.	Ma	Hem	ES, IT, M	MW	13514
<b>Ceratophyllaceae</b>					
<i>Ceratophyllum demersum</i> L.	Su	Hyd	PL	MW, LW, RW, BW	13730
<b>Chenopodiaceae</b>					
<i>Chenopodium ambrosioides</i> L.	Ma	Hem	PL	MW	13781
<i>Chenopodium album</i> L.	Ma	Thr	COSM	BW, MW	13766
<i>Chenopodium rubrum</i> L.	Ma	Thr	PL	BW	13761
<b>Convolvulaceae</b>					
<i>Calystegia sepium</i> (L.) R. Br.	Ma	Geo	PL	LW	13552
<i>Convolvulus arvensis</i> L.	Ma	Hem	COSM	LW	13685
<b>Cuscutaceae</b>					
<i>Cuscuta campestris</i> Yunck.	Ma	Thr	COSM	MW	13672
<b>Euphorbiaceae</b>					
<i>Acalypha australis</i> L.	Ma	Thr	PL	MW, AW	13782
<i>Chrozophora oblique</i> (Vahl) Juss. ex Spreng	Ma	Thr	IT	MW, BW	13666
<i>Euphorbia helioscopia</i> L.	Ma	Thr	ES, IT, M	MW	13520
<i>Euphorbia peplus</i> L.	Ma	Thr	ES, IT, M	MW, BW	13750
<i>Euphorbia virgata</i> Waldst. & Kit.	Ma	Hem	ES-IT-M	MW, BW	13654
<i>Ricinus communis</i> L.	Ma	Hem	PL	MW	13783
<b>Fabaceae</b>					
<i>Sequigera varia</i> Lassen	Ma	Hem	IT	BW, LW	13597
<i>Glycyrrhiza echinata</i> L.	Ma	Geo	ES, IT, M	BW	13705
<i>Lathyrus aphaca</i> L.	Ma	Thr	ES, IT, M	MW	13503
<i>Lathyrus hirsutus</i> L.	Ma	Hem	ES-IT-M	RW	13572
<i>Lotus corniculatus</i> L.	Ma	Hem	PL	LW, MW	13689
<i>Medicago lupulina</i> L.	Ma	Hem	PL	BW	13602
<i>Medicago polymorpha</i> L.	Ma	Thr	IT, M	LW	13563
<i>Melilotus indicus</i> (L.) All.	Ma	Thr	PL	MW	13660
<i>Trifolium campestre</i> Schreb.	Ma	Thr	ES-IT-M	BW	13595
<i>Trifolium lappaceum</i> L.	Ma	Thr	ES-IT-M	MW	13764
<i>Trifolium repens</i> L.	Ma	Geo	ES, IT, M	MW	13770
<i>Trifolium resupinatum</i> L.	Ma	Thr	ES, IT, M	MW, LW	13509
<i>Vicia sativa</i> L.	Ma	Thr	ES, IT, M	MW	13505
<b>Haloragaceae</b>					
<i>Myriophyllum verticillatum</i> L.	Su	Hyd	COSM	LW, MW	13626
<b>Hypericaceae</b>					
<i>Hypericum perforatum</i> L.	Ma	Hem	PL	LW, BW	13628
<b>Lamiaceae</b>					
<i>Lycopus europaeus</i> L.	Ma	Geo	PL	MW	13748
<i>Marrubium vulgare</i> L.	Ma	Geo	PL	LW	13546
<i>Mentha aquatica</i> L.	Em	Geo	ES	MW	13774
<i>Mentha langifolia</i> (L) Hudson	Ma	Hem	PL	LW	13775
<i>Teucrium hyrcanicum</i> Steud.	Ma	Geo	ES (Hyr)	LW, MW	13619
<b>Lentibulariaceae</b>					
<i>Utricularia australis</i> R. Br.	Su	Hyd	PL	MW	13637
<b>Lythraceae</b>					
<i>Ammania baccifera</i> L.	Ma	Thr	PL	BW	13706

Taxa	Habitat	Life form	Chorotype	Locality	Hb. No. (FAR)
<b>Dennstaedtiaceae</b>					
<i>Pteridium aquilinum</i> (L.) Kuhn	Ma	Geo	COSM	MW	13778
<b>Equisetaceae</b>					
<i>Equisetum arvense</i> L.	Em	Geo	PL	MW	13506
<i>Equisetum ramosissimum</i> Desf.	Ma	Geo	PL	MW	13678
<i>Equisetum telmatia</i> Ehrh.	Ma	Geo	PL	BW	13600
<b>Salviniaceae</b>					
<i>Salvinia natans</i> (L.) All.	Su	Hyd	PL	RW, BW	13573
<b>Woodsiaceae</b>					
<i>Athyrium filix-femina</i> (L.) Roth	Ma	Geo	COSM	MW	13524
<b>Spermatophyta</b>					
<b>Angiospermae</b>					
<b>Dicotyledones</b>					
<b>Amaranthaceae</b>					
<i>Alternanthera sessilis</i> (L.) R. Br.	Ma	Thr	PL	BW, MW	13698
<i>Amaranthus blitoides</i> S.Watson var. <i>blitoides</i>	Ma	Thr	PL	MW, BW	13669
<i>Amaranthus retroflexus</i> L.	Ma	Thr	PL	BW	13800
<i>Amaranthus viridis</i> L.	Ma	Thr	PL	MW	13668
<b>Apiaceae</b>					
<i>Berula angustifolia</i> F. K. Mertens et W. D. J. Koch	Ma	Hyd	PL	BW	13769
<i>Bupleurum marschallianum</i> C.A. Mey.	Ma	Thr	ES-IT	MW	13749
<i>Daucus carota</i> L. var <i>sativus</i>	Ma	Hem	IT-M	LW, RW	13570
<i>Eryngium caucasicum</i> Trautv.	Ma	Hem	ES, IT, M	MW, LW	13673
<i>Hydrocotyle vulgaris</i> L.	Em	Geo	ES	AW	13529
<i>Oenanthe aquatica</i> (L.) Poir.	Em	Hem	ES-IT	MW, RW	13629
<i>Pimpinella affinis</i> Ledeb.	Ma	Hem	PL	MW	13648
<i>Turgenia latifolia</i> (L.) Hoffm.	Ma	Thr	ES- M-IT	RW	13718
<b>Asteraceae</b>					
<i>Artemisia annua</i> L.	Ma	Thr	ES, IT, M	MW	13779
<i>Bidens tripartita</i> L.	Ma	Thr	PL	MW, BW	13701
<i>Carduus arabicus</i> Jacq.	Ma	Thr	ES, IT, M	RW	13714
<i>Centaurea iberica</i> Trevir. ex Spreng.	Ma	Thr	PL	LW, RW	13692
<i>Cirsium vulgare</i> (Savi) Ten.	Ma	Hem	PL	RW	13715
<i>Conyza bonariensis</i> (L.) Cronquist.	Ma	Thr	COSM	MW, BW	13753
<i>Conyzanthus squamatus</i> (Spreng.) Tamamsch.	Ma	Hem	PL	MW	13747
<i>Crepis pulchra</i> L.	Ma	Thr	ES, IT, M	LW	13562
<i>Cichorium intybus</i> L.	Ma	Hem	PL	MW	13649
<i>Eclipta prostrata</i> (L.) L.	Em	Thr	PL	RW, MW, BW	13565
<i>Lactuca serriola</i> L.	Ma	Hem	PL	LW	13691
<i>Senecio vernalis</i> Waldst. & Kit.	Ma	Thr	ES, IT, M	RW	13719
<i>Sonchus asper</i> (L.) Hill. subsp. <i>glaucescens</i> (Jordan) Ball.	Ma	Hem	PL	LW, RW	13541
<i>Sonchus oleraceus</i> L.	Ma	Thr	PL	BW	13735
<i>Xanthium spinosum</i> L.	Ma	Thr	PL	MW	13674
<i>Xanthium strumarium</i> L.	Ma	Thr	PL	BW, LW	13762
<b>Boraginaceae</b>					
<i>Heliotropium europaeum</i> L.	Ma	Thr	ES, IT	MW	13661
<i>Myosotis palustris</i> Lam.	Em	Geo	COSM	AW	13530
<b>Brassicaceae</b>					
<i>Capsella bursa-pastoris</i> (L.) Medicus	Ma	Hem	PL	MW	13645
<i>Cardamine hirsuta</i> L.	Em	Thr	COSM	MW	13780
<i>Nasturtium microphyllum</i> Boenn. ex Rchb.	Em	Hyd	PL	AW	13531
<i>Nasturtium officinale</i> W.T.Aiton	Em	Hyd	PL	AW, MW, LW	13504
<i>Raphanus raphanistrum</i> L. subsp. <i>raphanistrum</i>	Ma	Thr	PL	LW	13624

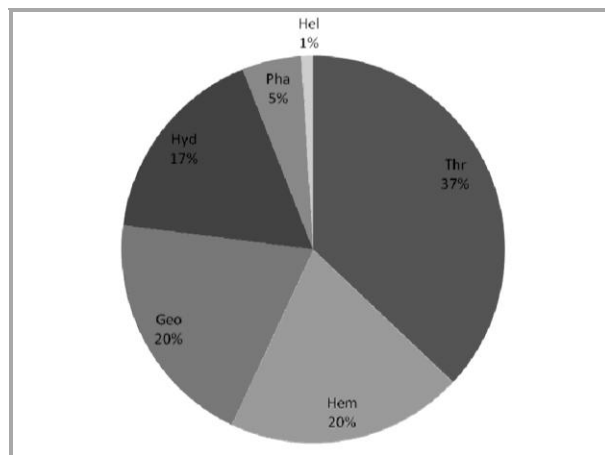


Figure 4. Proportion of different life forms (%) identified in the five important wetlands of Babol. Abbreviations: Thr=Therophyte, Hem=Hemicryptophytes, Pha=Phanerophytes, Hel=helophytes, Geo=geophyte.

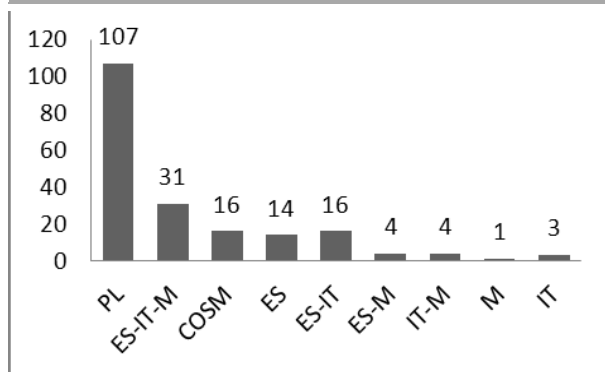


Figure 5. Proportion of various chorotypes (%) in the studied wetland sites. Abbreviations: IT=Irano-Turanian, M=Mediterranean, ES=Euro-Siberian, PL=Pluriregional, COSM=Cosmopolitan.

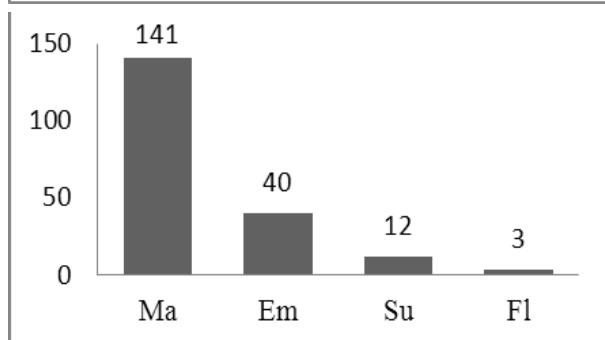


Figure 6. Proportion of species richness in different habitats of Babol wetlands. Em (emergent plants), Fl (floating plants), Ma (marginal plants), Su (submerged plants).

Table 4. Floristic list of the five important wetlands of Babol.

Symbols and abbreviations used in the table:

**Life form:** Geo (Geophyte), Hel (Helophyte), Hem (Hemicryptophyte), Hyd (Hydrophyte), Pha (Phanerophyte), Thr (Therophyte)

**Chorotype:** COSM (Cosmopolitan), ES [Euro-Siberian (Eux-Hyr=Euxino-Hyrcanian, Hyr=Hyrcanian)], IT (Irano-Turanian), M (Mediterranean), PL (Pluriregional)

**Habitat and Ecology:** Aq (Aquatic habitats), Em (Emergent plant), Fl (Floating plant), Hyg (Hygrophyte), Ma (Marginal plant), Su (Submerged plant)

**Location:** AW (Aghoozbon wetland), BW (Bosra wetland), LW (Langoor wetland), MW (Marzoonabad wetland), RW (Ramenet wetland).

Taxa	Habitat	Life form	Chorotype	Locality	Hb. No. (FAR)
<b>Pteridophyta</b>					
<b>Adiantaceae</b>					
<i>Adiantum capillus veneris</i> L.	Ma	Geo	PL	MW	13523
<b>Azollaceae</b>					
<i>Azolla filiculoides</i> Lam.	Su	Hyd	PL	LW, AW	13690

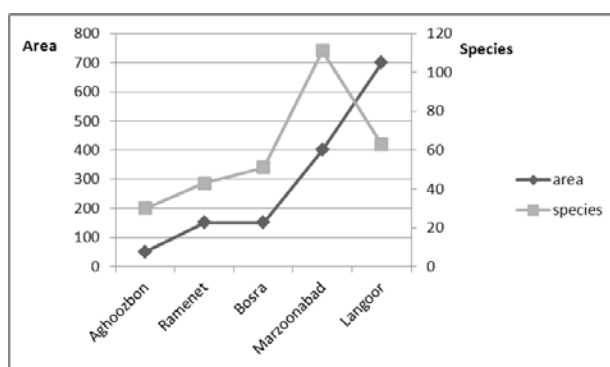


Figure 3. The number of species according to their surface area in the five wetlands of study area

### Life forms

In the total assessment of life from spectrum, therophytes made up 37% of the vegetation and were the dominant biological type in the studied area, followed by hemicryptophytes with 20%, as the second dominant life form (Figure 4). Although, therophytes occurred abundantly in desert areas (Archibold, 1995), its high presence was attributed to human activities and extensive grazing. This effect was previously observed in other studied ecosystems as well (Ghahreman *et al.*, 2006; Ejtehadi *et al.*, 2003; Khodadadi *et al.*, 2009).

### Chorological spectrum

The chorotype distributions of species in these five wetlands are as given (Figure 5). As it is shown, the flora of the study areas is much affected by pluriregional elements due to two reasons. First, the humid and wet habitats dominating the area, that harbor the bulk of the pluriregional plants adapted to wet places. Second, human activities that are responsible for the establishment of widespread weeds (Archibald, 1995; Naqinezhad *et al.* 2006).

### Habitat

The results of this study showed the existence of three different habitats in the studied area as follows (Figure 7):

1-Habitat for marginal plants: These habitats were usually situated on wet places near to wetlands, plains, rivers, etc. i.e. *Euphorbia helioscopia*, *Marrubium vulgare*, *Fimbristylis bisumbellata*, *Silene latifolia*, *Juncus acutus*, *Lathyrus hirsutus*, *Bupleurum marschallianum*, *Polygonum lapathifolium*, *Ranunculus marginatus*, *Verbascum punalense*.

2-Habitat for the emergent plants, these habitats contained marshlands and places out of open water area. Plants of this habitat had the high ability to absorb large amount of water. These habitats placed at second status after marginal habitats. Some species of this habitat were: *Hydrocotyle vulgaris*, *Oenanthe aquatica*, *Eclipta prostrata*, *Nasturtium microphyllum*, *Lythrum salicaria*, *Samolus valerandi*, *Ranunculus ophioglossifolius*, *Carex songorica*, *Sparganium erectum*, *Sagittaria sagittifolia*, *Phragmites australis*.

3-Habitat for open water plants: These parts were characterized with some floating and submerged plants. There were fewer species existed in this habitat. Species adapted to these habitats were: *Ceratophyllum demersum*, *Salvinia natans*, *Callitriche brutia*, *Batrachium trichophyllum*, *Lemna minor*, *Nympha alba*.

A column in Table 4 is relevant to habitat diversity of plant species. The number of plant species (in number) which can be found in each habitat is summarized in Figure 6.

Table 2. Comparative floristic richness and taxonomic diversity. Myankaleh (Ejtehadi *et al.*, 2003); Anzali (Ghahreman and Attar, 2003); Amirkelayeh (Ghahreman *et al.*, 2004); Boujagh (Naqinezhad *et al.*, 2006); Fereydoonkenar (Hoseinzadeh, 2007).

	Present study	Myankaleh	Anzali	Amirkelayeh	Boujagh	Fereydoonkenar
Total number of taxa (T)	196	242	291	320	248	247
Total number of genera (G)	138	169	194	213	164	176
Total number of families (F)	58	48	68	76	62	73
T/G	1.4	1.4	1.8	1.5	1.5	1.4
G/F	2.4	3.5	2.9	2.8	2.6	2.4

### Vegetation of the wetlands

The main structure of vegetation of whole areas of the studied wetlands were relatively similar to the vegetation of other wetlands of the northern Iran (e.g. Naqinezhad *et al.*, 2006; Asri and Eftekhari, 2002). The vegetation of these five wetlands were also relatively different from each other. The Langoor wetland which had the surface area more than 700 ha was the largest wetland in Babol. *Nymphaea alba* was the dominant aquatic plant in this wetland. This plant was considered as a monodominant vegetation in this wetland in the area. The Marzoonabad wetland which had the surface area as much as 400 ha was located in second status. There were many plant communities such as *Nymphaea alba-Nelumbium nuciferum* and *Phragmites australis-Sparganium erectum* in the wetland. The Bosra wetland which was more than 150 ha had the marshy mood in some parts. The special characteristic of this wetland was the occurrence of high density of *Phragmites australis* that served as a very valuable nesting places for lots of waterfowls. There was an obvious community of *Typha domingensis-Sparganium erectum* in the Ramenet wetland. Finally, one of the major characteristics of the Aghoozbon wetland was the occurrence of *Paspalum distichum* vegetation in its Islands.

A floristic resemblance study was done to show the level of similarity between the five wetlands, using Sørensen's (1948) similarity index (Table 3). Based on the obtained results the similarity level varied between 4.1% (Ramenet-Aghoozbon) to 29.8% (Marzoon abad-Langoor). This revealed that the floristic similarity was related to each wetland surface area and immigration of the variety of waterfowl that was carried plants seeds from other wetlands (Naqinezhad *et al.*, 2006). It can be seen frequently between Marzoonabad and Langoor wetlands. Furthermore, less similarity between Ramenet and Aghoozbon wetlands was derived mostly from human activities and their relatively far distance. Moreover, frequency of species in each wetland indicated that with the increase in wetland surface, the number of species increases except for the Langoor wetland. This could be due to water deficiency, human interference and lack of migratory waterfowls in the Langoor wetland (Figure 3).

Table 3. Comparison of the flora of the five wetlands with each other using Sørensen's coefficient (references are as Table 1) (in percentage).

	Langoor	Bosra	Ramenet	Aghoozbon
Marzoonabad	29.8	27.1	20.7	15.7
Langoor		17.5	24.5	21.7
Bosra			17	7.5
Ramenet				4.1

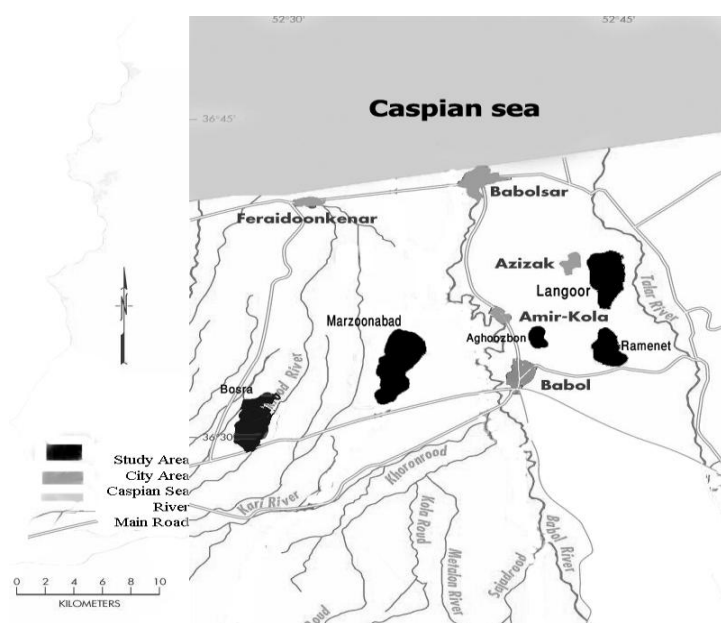


Figure 1. Location map of the five important wetlands of Babol

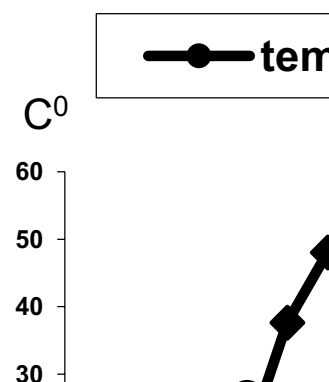


Figure 2. The climatic diagram based on the data from Gharakheil meteorological station covering the years 1979-2004

## Results and Discussion

In this study, a total of 196 species of vascular plants were identified from Babol wetlands, which belonged to 58 families and 138 genera. There were different number of families, genera and species among various taxonomic groups (Table 1). The richest families in terms of the number of taxa were Poaceae (24), Cyperaceae (19), Asteraceae (16) and Fabaceae (13) respectively. Nine families possessed two taxa and the remaining 28 families had just one taxon. As for the species richness, genera with four and exceeding species were: *Cyperus* (eight species), *Polygonum* (seven species), *Potamogeton* (four species), *Juncus* (four species), *Carex* (four species), *Trifolium* (four species) and *Typha* (four species). The most species-rich wetland was Marzoonabad with 111 plant taxa and the lowest species-rich wetland was Aghoozbon with 30 plant taxa. This could be due to the occurrence of more divers' habitats and different surface area in the Marzoonabad wetland. In Langoor wetland, there were 63 taxa from 56 genera and 31 families. There were 111 plant taxa from 86 genera and 45 families in the Marzoonabad wetland. In Bosra wetland, there were 51 plant taxa from 42 genera and 26 families. In Ramenet wetland, 43 plant taxa from 38 genera and 21 families were collected and determined. In a study on Aghoozbon wetland, 30 plant taxa within 27 genera and 18 families were determined. The ratios of species/genera and genera/families for the Babol wetlands indicated a higher taxonomic diversity as compared to other wetland areas, but these wetlands had fewer species than others, because of lower surface area and higher habitat homogeneity (Table 2).

Table 1. The number of families and genera in the taxonomic groups

	Pteridophyta	Angiospermae	
		Monocotyledones	Dicotyledones
Family	6	11	41
Genus	6	38	94
Species	8	62	126



Langoor, Bosra, Ramenet and Aghoozbon are important ecosystem in the north of Iran. Studying these wetlands is very important because they serve as a very valuable resting, nesting and wintering places for a wide variety of waterfowls. These wetlands also play the critical role in restoring water, which is required for cultivating activities in summer. Because of variety in the climatic conditions, there are rather remarkable and unique wetlands with exclusive characteristic in different parts of Iran. Some floristic and ecological studies have been conducted on these valuable ecosystems e.g. Hashilan wetland (Karami *et al.*, 2001) in the west and Parishan wetland (Dolatkahi *et al.*, 2010) in the south of Iran. Nevertheless, many floristic and vegetation studies of the wetland habitats in Iran were concentrated along the southern Caspian shore, i.e. Amirkelayeh lagoon and coasts of Lahijan-Langerud (Asri and Moradi, 2004; Ghahreman *et al.*, 2004), Anzali lagoon (Ghahreman and Attar, 2003; Asri and Eftekhari, 2002), Miankaleh wildlife refuge (Sharifnia *et al.*, 2007; Ejtehad *et al.*, 2003; Asri *et al.*, 2007), Boujagh National Park (Naqinezhad *et al.*, 2006), Estil wetland (Khodadadi *et al.*, 2009), Solukli wetland in Golestan national Park (Akhani, 1998), Gomishan lagoon (Karimi, 2010). The aims of the study were to present: (1) a checklist of all vascular plants found in these five wetlands, (2) spectrum of life form and phytogeographical data across the whole wetlands together with detailed information about the habitats, life form and chorology for each species, (3) a comparison between the results of Babol wetlands and other wetlands and (4) a solution for protecting these wetlands from serious destruction.

## Materials and Methods

### Study area

The wetlands of Babol are located in Mazandaran province, northern Iran, between 52° 35' - 52° 45' E and 36° 31' - 36° 37' N. The studied area covers five wetlands namely Marzoonabad, Langoor, Bosra, Ramenet and Agoozbon. Marzoonabad and Basra wetlands are located beside Babol-Amol road. Langoor and Agoozbon wetlands are near Babol-Bahnamir road and finally Ramenet wetland is placed beside Babol-Kiakola road. All these wetlands are located in the plain part of Babol (Figure 1). There are many cultivated places and also some canals around of the studied wetlands. These canals carry water from wetland to cultivated farms. The total surface and the mean altitude of the Babol wetlands are 1470 ha and 14.7 m respectively. The rainiest month is October. The mean annual precipitation is 738.7 mm and the mean annual temperature is 16.3 °C. The maximum and minimum mean temperatures are 29.3 °C, and 4.5 °C, respectively. The ombrothermic diagram of the studied area was prepared according to climate data obtained from the Gharakheil meteorological station (Figure 2). Three rivers namely Talar, Babol and Haraz provide water for these five wetlands and then empty their water into the Caspian sea.

### Data collection and Analyses

In order to survey the flora of Babol wetlands, topographic maps were provided at first. Then, the specimens were collected in different seasons. The collected samples were then identified and named based on the classification and terminology applied to various Flora, such as: Flora Iranica (Rechinger, 1963-1998), Flora of Iran (Assadi, 1988-2007) and Flora of Turkey (Davis, 1965-1988). All plant specimens were deposited in the Farabi Herbarium (FAR), Tehran. Life forms were named following the Raunkiaer's classification (Raunkiaer, 1934) and chorology of species is based on Zohary (1973) and Takhtajan (1986) viewpoints. The habitat and color flower of each species were carefully noted while collecting the samples.

## Plant diversity of five important wetlands of Babol Mazandaran province, Iran

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### Abstract

This study deals with the flora of five important wetlands namely Marzoonabad, Langoor, Bosra, Ramenet and Aghoozbon and their surroundings in Babol, Mazandaran province. In order to carry out a floristic survey in these wetlands, all vascular plants were collected during two growing seasons (2010-2011). We encountered 196 species belonging to 138 genera and 58 families. The largest families in the studied area were Poaceae with 24 species, Cyperaceae with 19 species, Asteraceae with 16 species, Fabaceae with 13 species and Polygonaceae with 9 species, respectively. Genera represented by the greatest number of species were *Cyperus* (8), *Polygonum* (7) and *Potamogeton* (4). Classification based on life form indicated that the therophytes (37%) comprised the largest proportion of the plants in the studied area. From chorological point of view, the largest proportion of the flora belonged to the pluriregional elements (54.5%). Various habitats of the wetland are discussed. Among the five wetlands of Babol, Marzoonabad had the highest number of species (111) and Langoor with 63 species placed on second. Moreover, a comparison between the data collected here and other northern Iranian wetlands has been provided which indicated some similarities and dissimilarities between different studied wetlands. According to Sørensen's (1948) similarity index, there are less similarities between the species of the five wetlands of Babol because they have different surface area and there is no relation between them.

**Key words:** Aquatic plants, Babol wetlands, Chorology, Flora, Life form, Mazandaran province

### Introduction

Wetlands are valuable ecosystems which provide abundant services and materials with economic value, not only to the adjacent local populations but also to regional communities, providing valuable services such as water quality improvement, flood mitigation, erosion control and recreational enrichment (Mitsch and Gosselink, 2000). Destroying wetlands by means of drainage and pollution, which have derived from wastewater of agriculture and industries are substantial problems for the world wetlands. The occurrence of exotic species such as *Azolla filliculoides* on the water surface of wetlands is the major concern in these aquatic ecosystems in the north part of Iran. The wetlands of Babol namely Marzoonabad,

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differences. According to anatomical study of *Roemeria* and the transverse incision of blade of two species there were some significant differences as following:

- Lack of external collenchyma (such as *G. contortuplicatum*) and external parenchyma in the main vein in *Roemeria hybrida*.
- Lack of strengthening tissue surrounding the vascular bundles in *Roemeria refracta* (like *H. pendulum*)
- Presence of 1 and 3 vascular bundle in blade cross section provided from *R. refracta* and presence *R. hybrida* respectively.

Anatomical characteristics of the fruit in *Roemeria* which differed between these two species as follows: lack of hair in cross cutting the fruit of *Roemeria refracta* and its presence in *R. hybrida* and the difference in the overall shape of fruit in the transect so the shape of fruit in *R. refracta* was almost quadrangular (four corners) and the shape in *R. hybrida* was triangular. Anatomical study of the stems of species in this genus did not show very important difference. Blades transect study in two other genera such as *Hypocoum* and *Chelidonium* showed structurally no significant difference with respect to *Glaucium* and *Roemeria*. Cross section of blade in *C. majus* showed more similarity with *G. oxylobum*. Transect of fruit in *H. pendulum* was different from other genera. The general form of the transect was rectangular and lacking any placental bulge so identification of the placental place was difficult. External sclerenchyma could be seen only on the vascular bundles. As like as *Glaucium*, fruit in *C. majus* posed prominent placenta and the placenta shape in latter species was dentiformis. Identification of some species based only on morphological characteristics was difficult and even sometimes impossible. Previous studies and this study on anatomical characters of *Glaucium* indicated that these traits could identify and separate these species more accurately in some cases.

Anatomical study of blade in these genera indicated, in spite of some differences, the fundamental structure was similar, so cross section of blade solely could not be a good scale for separating species of genus *Glaucium* and identification of four genera. Fruit anatomical characters not only were useful in separating species of genus *Glaucium*, but also, for identification and determination of the four other genera. Stem anatomical study indicated that these four genera were very similar in this character, so stem anatomy characters was neither a suitable scale for separating species of *Glaucium*, nor for other genera. However, anatomical characteristics of the fruit, stem and blade in the four genera could confirm previous studies and had a good agreement with previous studies. (Solereeder, 1908; Metcalfe and Chalk, 1950).

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Table 4. Anatomical characters of the stem in studied species

Abbreviations: A: cuticle's thickness, B: epidermis thickness, C: parenchyma cortex thickness, D: scleranchyma cortex thickness, E: external phloem width, F: internal phloem width, G: xylem width. The unit of all quantity is micrometer.

Species	A	B	C	D	E	F	G
<i>G. haussknechtii</i>	10	30	60	200	40	30	300
<i>G. grandiflorum</i>	10	20	60	330	90	50	260
<i>G. fimbriigerum</i> Boiss. subsp. <i>Annuum</i>	5	10	35	80	15	70	150
<i>G. fimbriigerum</i> subsp. <i>ophycarpum</i>	10	20	72	180	30	130	60
<i>G. pulchrum</i>	10	35	150	50	40	20	70
<i>G. oxylobum</i>	10	20	220	130	80	10	110
<i>R. refracta</i>	15	20	30	80	50	40	80
<i>R. hybrida</i>	10	10	30	150	70	60	130
<i>Hypecoum pendulum</i>	10	10	30	70	50	25	120
<i>Chelidonium majus</i>	10	10	20	130	130	40	100

Results of present study showed, anatomical characters of midrib in *Glaucium* species led to finding some traits which could be used for identification for different species. In vascular structure of midrib in most *Glaucium* species, bicollateral phloem and scleranchymatous sheath of vascular bundles could be seen (Figure 1: n, v). Among species of this genus, *G. contortuplicatum* possessed collateral phloem and lacked external collenchyma which was distinguished from other species. This species was easily recognized from other species by its helical fruit. A central cavity can be observed midrib in *G. oxylobum* which in turn, could be used as a differentiating feature for this species. Multicellular single row hairs in *G. grandiflorum* and *G. corniculatum* could be seen (Figure 1: c, f). Although, Metcalfe and Chalk (1950) studied anatomical traits of Papaveraceae but they didn't report on glandular hairs. In this study, glandular hairs were observed in *G. oxylobum* and *G. pulchrum*.

According to present study, some anatomical features of fruit in *Glaucium* can be used in grouping of the genus. The most important traits were the apparent shape of ovarian placenta (Figure 2: b, d, f, h, j, l, n and p). Placenta types were observed as following:

- 1- Triangular: *G. haussknechtii*, *G. corniculatum* (Figure 2: a and e)
- 2- Dentiformis: *G. elegans* (Figure 2: k)
- 3- Heart-shaped: *G. oxylobum*, *G. pulchrum* (Figure 2: o, m)
- 4- Bean-shaped: *G. grandiflorum*, *G. fimbriigerum* Boiss. subsp. *annuum*, *G. fimbriigerum* subsp. *ophycarpum* (Figure 2: c, g and i)

Another distinguishing character was carpellary angle. Carpellary angle varied from 95° in *G. pulchrum* to 170° in *G. oxylobum* (Figure 2: m and o) which was classified as following:

Group I: Lunar-shaped: *G. haussknechtii*, *G. corniculatum*, *G. elegans* and *G. pulchrum* (Figure 2: a, e and k).

Group II: Linear-formed: *G. oxylobum* and *G. fimbriigerum* subsp. *ophycarpum* (Figure 2: g and o).

Group III: V-shaped: *G. fimbriigerum* Boiss. subsp. *annuum* and *G. grandiflorum* (Figure 2: i).

In addition to the above mentioned traits, the differences in the number of external and internal layers of parenchyma in the ovary wall, the number of vascular bundles of placenta and type of hairs might be suitable in identification of unknown species. Considering the anatomical traits of fruit in *Glaucium*, identification of two species, *G. haussknechtii* and *G. grandiflorum*, from each other and other species of *Glaucium* was easily possible. Anatomical characteristics of the stem in species of this genus did not show very important

Table 2. ...

Species	K	L	M	N	O	P	Q	R	S
<i>G. corniculatum</i>	150	20	70	130	80	60	5	2180	210
<i>G. elegans</i>	170	20	40	210	-	100	5	2100	260
<i>G. grandiflorum</i>	180	20	70	150	80	110	4	1500	200
<i>G. contortuplicatum</i>	150	-	60	170	50	150	3	1500	300
<i>G. pulchrum</i>	100	30	-	110	-	140	3	900	200
<i>G. hussknechtii</i>	150	10	20	100	70	85	4	1300	150
<i>G. oxylum</i>	220	10	40	230	70	100	5	2260	200
<i>Roemeria hybrida</i>	110	35	50	65	-	-	1	1200	-
<i>R. refracta</i>	110	-	20	120	30	100	3	1500	230
<i>Hypecoum pendulum</i>	250	35	70	150	-	-	5	1500	250
<i>Chelidonium majus</i>	50	15	60	90	10	90	5	1200	120

Table 3. Anatomical characters of the capsule in studied species

Abbreviations: A: Papile presence, B: Papile layers' number, C: Papile's shape, D: cuticle's thickness, E: epidermis thickness, F: external parenchyma layer number, G: upper parenchyma layer thickness, H: lower parenchyma layer number, I: lower parenchyma layer thickness, J: laticiferous tubes location, K: bundle vessel width, L: upper scleranchyma length, M: placenta's shape, N: Bundle vessle number of placenta, O: shape's Leaflet Angle between, P: Leaflet Angle between, Q: Wall thickness, R: placenta's thickness, S: Attache wall, an: angular, em: embowed, fi: Five form, g: globular, li: linear, l: long, Mo: Monoseriate, Mu: Multiseriate, o: oblong, re: renal, p: Paranchym, pd: Paranchym down, pu: paranchya up, s: short, sc: semicircle, su: sunken, sp: spear, to: tooth, tr: triangle. The unit of all quantity is micrometer. The unit of all quantity is micrometer.

Species	A	B	C	D	E	F	G	H	I	J
<i>G. corniculatum</i>	+	Mu	l, g	10	40	10	20	3	110	Pd, sc/v, sc/p
<i>G. elegans</i>	+	Mo, Mu	s, se, l, o	10	30	3	50	5	100	sc/v, sc/p
<i>G. fimbrilligerum</i> Boiss. subsp. <i>Annuum</i>	+	Mu	l, o	5	40	3	40	6	130	sc/v, sc/p
<i>G. fimmbrilligerum</i> subsp. <i>ophycarpum</i>	+	Mu	L, g	10	50	5	110	4	80	
<i>G. grandiflorum</i>	+	Mu	S, se	10	30	3	50	7	80	sc/v, sc/p
<i>G. hussknechtii</i>	+	Mu	l, o	10	30	5	80	7	220	sc/v, sc/p
<i>G. pulchrum</i>	-	-	-	20	30	6	100	4	60	sc/v, sc/p
<i>G. oxylum</i>	+	Mu	L, g	10	15	3	30	4	70	sc/v, sc/p
<i>R. refracta</i>	-	-	-	10	40	4	170	1-3	80	P
<i>R. hybrida</i>	+	Mu	sp	10	20	6	160	4	100	P
<i>Hypecoum pendulum</i>	-	-	-	23	30	2	150	-	-	sc/p, p
<i>Chelidonium majus</i>	-	-	-	10	6	1	20	1	15	sc/p

Species	K	L	M	N	O	P	Q	R	S
<i>G. corniculatum</i>	90	250	tr	4	em	130	450	600	130
<i>G. elegans</i>	50	100	to	6	em	125	390	530	700
<i>G. fimbrilligerum</i> Boiss. subsp. <i>Annuum</i>	20	110	re	7	an	110	370	1000	1500
<i>G. fimmbrilligerum</i> subsp. <i>ophycarpum</i>	40	130	re	5	li	160	300	1100	450
<i>G. grandiflorum</i>	50	100	re	8	an	115	300	650	700
<i>G. hussknechtii</i>	40	150	tr	4	em	130	400	580	100
<i>G. pulchrum</i>	50	70	fi	6	sc	95	300	750	250
<i>G. oxylum</i>	70	120	fi	2	li	170	300	800	430
<i>R. refracta</i>	15	-	su	1	sc	93	350	345	-
<i>R. hybrida</i>	45	-	su	1	sc	100	320	320	-
<i>Hypecoum pendulum</i>	50	100	su	1	sc	130	600	550	
<i>Chelidonium majus</i>	10	-	to	1	Li	170	100	250	200

Thicknesses of the upper parenchyma in *C. majus* and *R. refracta* in comparison with *R. hybrida*, *H. pendulum* and species of *Glaucium* had considerable differences (Figure 1: u-z and aa, bb, cc, dd). Thicknesses of the lower parenchyma in *C. majus* in comparison with other genera had considerable differences. Against *R. hybrida*, *C. majus* and most species of *Glaucium*, upper collenchyma did not exist in *Roemeria refracta* and *H. pendulum* and *R. hybrida* phloem was collateral (Figure 1: u-z and aa, bb, cc, dd). Vascular bundles of midrib in *R. refracta* were one, in *R. hybrida* three and five in *C. majus* and *H. pendulum*. The minimum number of midrib vascular bundles among all four genera belonged to *R. refracta* (Figure 1: u-z and aa, bb, cc, dd). There were some laticifer tubes around vascular bundles, parenchyma, especially sclerenchymatous sheath, cone-shaped, multicellular multi-row hairs were observed in *R. refracta*, while long multicellular and single row hairs were recognized in *R. hybrida* and *C. majus*. No hair was seen in *H. pendulum* (Figure 1: u-z and aa, bb, cc, dd).

**Stem:** Cuticle and epidermal thickness in *R. hybrida*, *H. pendulum*, *C. majus* were similar, but more thickness was measured in *R. refracta*; maximum thickness of cuticle among four genera belonged to *R. refracta* (Figure 3: m-t). Parenchyma thickness did not show significant differences in the four mentioned species (Figure 3: m-t). Sclerenchyma cortex thickness, width of external and internal phloem in *R. refracta* and *H. pendulum* in comparison with *R. hybrida*, *C. majus* had considerable differences. Laticifer tubes in *Roemeria* species could be seen around vascular bundle and in *C. majus* and *H. pendulum* clearly in the cortex and sclerenchymatous sheath (Figure 3: m-t).

**Fruit:** Cuticular thickness in *H. pendulum* was more than three other species (Figure 2: q-x). Inner parenchyma layers were superseded by sclerenchymatous in *H. pendulum* (Figure 2: v). Placenta was planar and had a vascular bundle in *Roemeria* species, while in *C. majus* was dentiform with a vascular bundle (Figure 2: q-x). Semi-circular carpellary angle varies from 90° to 130° in *R. refracta*, *R. hybrida* and *H. pendulum*, respectively (Figure 2: q, s, u and w). Linear carpellary angle (170°) was seen in *Chelidonium majus*. Laticifer tubes could be found in placental sclerenchyma and in wall parenchyma in *R. hybrid*. Multicellular spear-like hairs were observed in *Roemeria*, but the others lacked such a character (Figure 2: q and r).

Table 2. Anatomical characters of basal leaf in studied species.

Abbreviations: A: Papile presence, B: Papile layers' number, C: Papile's shape, D: upper cuticle's thickness, E: lower cuticle's thickness, F: upper epidermis thickness, G: lower epidermis thickness, H: upper collenchyma thickness, I: lower collenchyma thicknes, J: upper parenchyma layer thickness, K: lower parenchyma layer thickness, L: upper phloem width, M: lower phloem width, N: xylem width, O: upper tissue strength thickness, P: lower tissue strength thickness, Q: Bundle vessle number, R: Length leaflet, S: Bundle vessel distance between. l: long, Mu: multiseriate, Mc: multicellular, o: oblong, On: Onion shape, g: glandular, u: uniseriate. The unit of all quantity is micrometer.

Species	A	B	C	D	E	F	G	H	I	J
<i>G. corniculatum</i>	+	Un	l, Mc	15	13	15	15	25	120	250
<i>G. elegans</i>	-	-	-	10	10	30	10	30	146	300
<i>G. grandiflorum</i>	+	Un	l, Mc	5	5	28	20	50	40	330
<i>G. contortuplicatum</i>	-	-	-	20	20	40	35	-	40	200
<i>G. pulchrum</i>	+	Mu	l, Mc, g	20	20	20	30	30	140	350
<i>G. husskenechtii</i>	-	-	-	20	10	15	15	50	70	100
<i>G. oxylobum</i>	+	-	l, g	10	10	40	40	55	280	430
<i>Roemeria hybrida</i>	+	Mu	On, l	10	15	40	20	20	30	55
<i>R. refracta</i>	+	Un	l, Mc	10	10	20	30	-	50	320
<i>Hypecoum pendulum</i>	+	Un	l, Mc	10	10	30	35	-	-	190
<i>Chelidonium majus</i>	+	Un	l, Mc	10	10	10	15	30	80	50

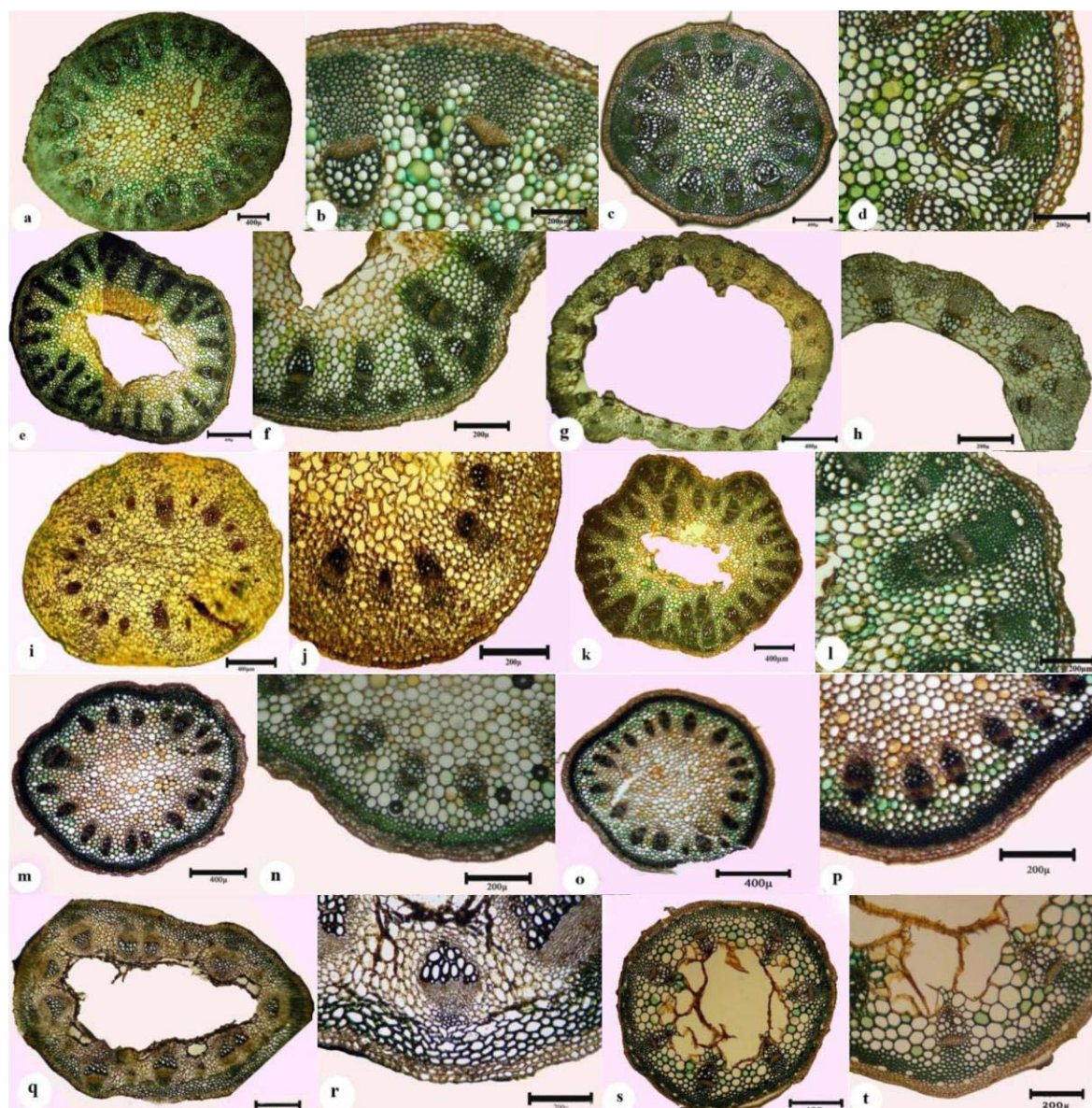


Figure 3. Cross section of stem in species: *G. grandiflorum* (a, b), *G. haussknechtii* (c, d), *fimbrilligerum* subsp. *annuum* (e, f), *G. fimbrilligerum* subsp. *ophycarpum* (g, h), *G. pulchrum* (i, j), *G. oxylum* (k, l), *Roemeria hybrida* (m, n), *Roemeria refracta* (o, p), *Hypocoum pendulum* (q, r), *Chelidonium majus* (s, t). Scale bars in the photos are as: a=400µm, b=200µm, c=400µm, d=200µm, e=400µm, f=200µm, g=400µm, h=200µm, i=400µm, j=200µm, k=400µm, l=200µm, m=400µm, n=200µm, o=400µm, p=200µm, q=400µm, r=200µm, s=400µm, t=200µm.

### ***Roemeria*, *Hypocoum* and *Chelidonium***

**Midrib:** Upper cuticle thickness did not show considerable differences in *R. refracta*, *R. heybrida*, *C. majus* and *H. pendulum* but less thickness was measured in most species of *Glaucium* (Figure 1: u-z and aa, bb, cc, dd). Lower cuticle thickness in *R. refracta* was more than *R. heybrida*, *C. majus* and *H. pendulum* but in comparison with most species of *Glaucium* did not show considerable differences. Upper and lower epidermises in *C. majus* in comparison with *H. pendulum*, *R. refracta* and *R. heybrida* had less thickness (Figure 1: u-z and aa, bb, cc, dd).





Figure 2. Cross section of fruit in species: *G. corniculatum* (a, b), *G. grandiflorum* (c, d), *G. haussknechtii* (e, f), *G. fimbrilligerum* subsp. *ophycarpum* (g, h), *G. fimbrilligerum* subsp. *annuum* (i, j), *G. elegans* (k, n), *G. pulchrum* (m, n), *G. oxylum* (o, p), *Roemeria hybrida* (q, r), *Roemeria refracta* (s, t), *Hypecoum pendulum* (u, v), *Chelidonium majus* (w, x).

Scale bars in the photos are as: a=400 $\mu$ m, b=200 $\mu$ m, c=400 $\mu$ m, d=200 $\mu$ m, e=400 $\mu$ m, f=200 $\mu$ m, g=400 $\mu$ m, h=200 $\mu$ m, i=400 $\mu$ m, j=200 $\mu$ m, k=400 $\mu$ m, l=200 $\mu$ m, m=400 $\mu$ m, n=200 $\mu$ m, o=400 $\mu$ m, p=200 $\mu$ m, q=400 $\mu$ m, r=30 $\mu$ m, s=400 $\mu$ m, t=200 $\mu$ m, u=400 $\mu$ m, v=200 $\mu$ m, w=400 $\mu$ m, x=30 $\mu$ m.

p). Several layers of parenchyma cells were recognized below the epidermis (Figure 2: a-p). The external area included 3-4 layers of chlorenchyma cells while 10 to 12 layers of the sclerenchymatous cells were observed in the internal region (Figure 2: a-p). At the center, vascular bundles were arranged in one row (Figure 2: a-p). The sclerenchyma tissue of pericarp was considerably occupied by laticifer tubes (Figure 2: a-p). Cuticle thickness varied from 5  $\mu\text{m}$  in *G. fimbriigerum* Boiss. subsp. *annuum* to 20  $\mu\text{m}$  in *G. pulchrum* (Figure 2: i and m). Epidermal cell thickness varied from 15 to 50  $\mu\text{m}$  in *G. oxylobum* and *G. fimbriigerum* subsp. *ophycarpum* (Figure 2: g and o). Inner parenchyma layer thickness varied from 20  $\mu\text{m}$  in *G. corniculatum* to 90  $\mu\text{m}$  in *G. fimbriigerum* subsp. *ophycarpum*. Number of internal layers of parenchyma ranged from three in the *G. corniculatum* to seven in *G. grandiflorum* and *G. haussknechtii* (Figure 2: a, c and e). Thickness of internal parenchyma varied from 60  $\mu\text{m}$  in *G. pulchrum* to 220  $\mu\text{m}$  in *G. haussknechtii*. Thickness of sclerenchyma tissue of fruit wall varied from 70  $\mu\text{m}$  in *G. pulchrum* to 250  $\mu\text{m}$  in *G. corniculatum* (Figure 2: a and m). The thickness of the tissue showed evident differences in *G. corniculatum* with respect to other species (Figure 2: a-p). Laticifer tubes could be seen in the sclerenchymatous sheath of the vascular bundles, the fruit wall, placenta and internal parenchyma layers (Figure 2: a-p). Length of vascular bundles in fruit wall varied from 20  $\mu\text{m}$  in *G. fimbriigerum* Boiss. subsp. *annuum* to 90  $\mu\text{m}$  in *G. corniculatum* (Figure 2: g and b). Carpellary angle varied from 95° in *G. pulchrum* to 170° in *G. oxylobum* (Figure 2: m and o). Carpellary angle form was crescent in *G. corniculatum*, *G. haussknechtii*, *G. elegans* (Figure 2: a, e and k), linear in *G. oxylobum* and *G. fimbriigerum* subsp. *ophycarpum* (Figure 2: g and o), V-shaped in *G. grandiflorum*, *G. fimbriigerum* Boiss. subsp. *annuum* (Figure 2: i and k) and semi-circular in *G. pulchrum* (Figure 2: m). Thickness of ovary wall varied from 300  $\mu\text{m}$  in *G. oxylobum*, *G. grandiflorum* and *G. fimbriigerum* subsp. *ophycarpum* to 450  $\mu\text{m}$  in *G. corniculatum*. The connected wall of placenta to the ovary varied from 100  $\mu\text{m}$  in *G. haussknechtii* to 1500  $\mu\text{m}$  in *G. fimbriigerum* Boiss. subsp. *annuum* (Figure 2: f-i). Thickness of placenta varied from 530  $\mu\text{m}$  in *G. elegans* to 1100  $\mu\text{m}$  in *G. fimbriigerum* subsp. *ophycarpum* (Figure 2: g-k). Among studied species triangular-shaped placenta could be seen in *G. haussknechtii* and *G. corniculatum* (Figure 2: b and f); dentiform in *G. elegans* (Figure 2: l), heart-shaped in *G. oxylobum* and *G. pulchrum* (Figure 2: n and p) and the bean-shaped could be found in *G. grandiflorum*, *G. fimbriigerum* Boiss. subsp. *annuum* and *G. fimbriigerum* subsp. *ophycarpum* (Figure 2: d, h and j). Placental thickness varied from 530  $\mu\text{m}$  in *G. elegans* to 1100  $\mu\text{m}$  in *G. fimbriigerum* subsp. *ophycarpum* (Figure 2: a, s, e, g, i, k, m and o). Total numbers of placental vascular bundles were two in *G. oxylobum*, four in *G. corniculatum*, *G. haussknechtii*, five in *G. fimbriigerum* subsp. *ophycarpum*, six in *G. elegans* and *G. pulchrum*, seven in *G. fimbriigerum* Boiss. subsp. *annuum* and eight in *G. grandiflorum* (Figure 2: a, s, e, g, i, k, m and o). Multicellular appressed trichomes in some species such as *G. corniculatum*, *G. grandiflorum* and *G. haussknechtii* were recognized, while glandular hairs could be found in *G. oxylobum* and *G. fimbriigerum* subsp. *ophycarpum* (Figure 2: a, s, e, g, i, k, m and o).

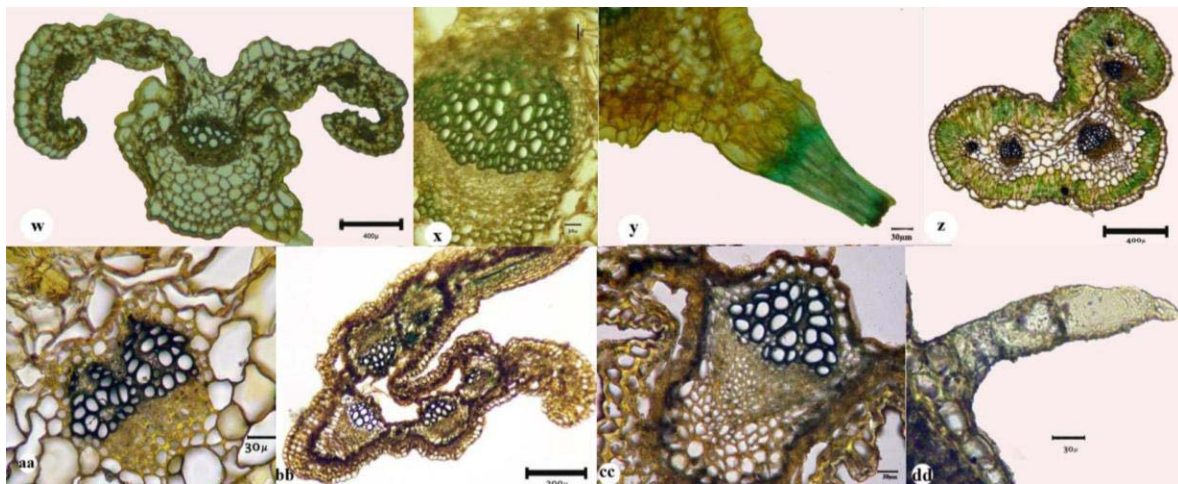


Figure 1. Cross section of midrib in species: *G. corniculatum* (a, b, c), *G. grandiflorum* (d, e, f), *G. haussknechtii* (g, h, i), *G. pulchrum* (j, k, l), *G. contortuplicatum* (m, n), *G. elegans* (o, p, q), *G. oxylobum* (r, s, t), *Roemeria hybrida* (u, v), *R. refracta* (w, x, y), *Hypocoum pendulum* (z, aa), *Chelidonium majus* (bb, cc, dd).

Scale bars in the photos are as: a=200 $\mu$ m, b=30 $\mu$ m, c=30 $\mu$ m, d=200 $\mu$ m, e=30 $\mu$ m, f=30 $\mu$ m, g=200 $\mu$ m, h=30 $\mu$ m, i=30 $\mu$ m, m=200 $\mu$ m, n=30 $\mu$ m, o=200 $\mu$ m, p=30 $\mu$ m, q=30 $\mu$ m, r=200 $\mu$ m, s=30 $\mu$ m, t=30 $\mu$ m, u=400 $\mu$ m, v=30 $\mu$ m, w=400 $\mu$ m, x=30 $\mu$ m, y=30 $\mu$ m, z=400 $\mu$ m, aa=30 $\mu$ m, bb=200 $\mu$ m, cc=30 $\mu$ m, dd=30 $\mu$ m.

Minimum thickness (5  $\mu$ m) of cuticle belonged to *G. grandiflorum* (Figure 1: d) and maximum (20  $\mu$ m) was observed in *G. contortuplicatum*, *G. pulchrum* and *G. haussknechtii* (Figure 1: g, j and m). Upper epidermis cells were usually larger than the lowers (Figure 1: a, d, g, j, m, o and r). Diameter of epidermal cells varied from 15  $\mu$ m in *G. corniculatum* and *G. haussknechtii* to 40  $\mu$ m in *G. oxylobum* and *G. contortuplicatum* (Figure 1: o and r). Thickness of upper collenchyma varied from 25  $\mu$ m in *G. corniculatum* to 55  $\mu$ m in *G. oxylobum* (Figure 1: a, d, g, j, m, o and r). There was no upper collenchyma in *G. contortuplicatum*. Thickness of lower collenchyma ranged from 40  $\mu$ m in *G. grandiflorum* and *G. corniculatum* to 280  $\mu$ m in *G. oxylobum*; lower parenchyma thickness varied from 40  $\mu$ m in *G. grandiflorum* and *G. contortuplicatum* to 280  $\mu$ m in *G. oxylobum*. Upper parenchyma thickness varied from 100  $\mu$ m in *G. haussknechtii* to 430  $\mu$ m in *G. oxylobum*. The range of vascular bundles varied from 110  $\mu$ m in *G. pulchrum* to 230  $\mu$ m in *G. oxylobum* (Figure 1: k and s). The range of fiber thickness at the upper surface of vascular bundles varied from 25  $\mu$ m in *G. corniculatum* to 50  $\mu$ m in *G. oxylobum*. Also, in most studied species, phloem tissue was bicolateral except for *G. contortuplicatum* (Figure 1: n). The range of xylem thickness varied from 80  $\mu$ m in *G. elegans* to 50  $\mu$ m in *G. grandiflorum*, *G. corniculatum*. Midrib cross section in *G. oxylobum* cleared a central cavity (Figure 1; r). In some species such as *G. contortuplicatum*, *G. haussknechtii* and *G. elegans* no hairs were observed on the surface of blade (Figure 1: i and q). Transverse section of the stem included cuticle layer, epidermal cells, parenchyma and vascular bundles. Cortex composed of parenchyma cells and one layer of sclerenchymatous cells. At the central part of stem, the medulla could be seen which was composed of many separated spherical cells, the center was occupied by a large cavity (Figure 3: a-l). Cuticular thickness in *G. fimbriigerum* Boiss. subsp. *annuum* was 5  $\mu$ m and in other species was 10  $\mu$ m (Figure 3: g). Measured characteristics were shown in the Tables 2, 3 and 4 (Figure 3: a-l).

**Fruit:** A cross section of fruit showed the placenta in the internal surface and two large cavities. There was a layer of small regular epidermis cells (Figure 2: b, d, f, h, j, l, n and

## Results and Discussion

### *Glaucium*

**Midrib and Stem:** In transverse section of the midrib, upper surface was flat and the lower surface was raised (Figure 1, a-t). A thick cuticle was observed on the outer surface of the epidermis (Figure 1: a, d, g, j, m, o and r). Several laminar layers of collenchyma were recognized under the epidermis (Figure 1: a, d, g, j, m, o and r). Under the collenchyma, parenchyma and in the central part, vascular bundles could be seen (Figure 1: a-t). Which were surrounded by sclerenchymatous sheaths (Figure 1: a-t). The continuity of the epidermal cells was interrupted by the presence of hairs. Some of the hairs were short, others were long (Figure 1: c, f, i, l, q and t). There were multilayer hairs (covering hairs) in some species such as *G. pulchrum* (Figure 1: l). These hairs were shorter (3 to 4 cells) and thicker than single row ones and had more cells (Figure 1: a, c, f, i, l, q and t).

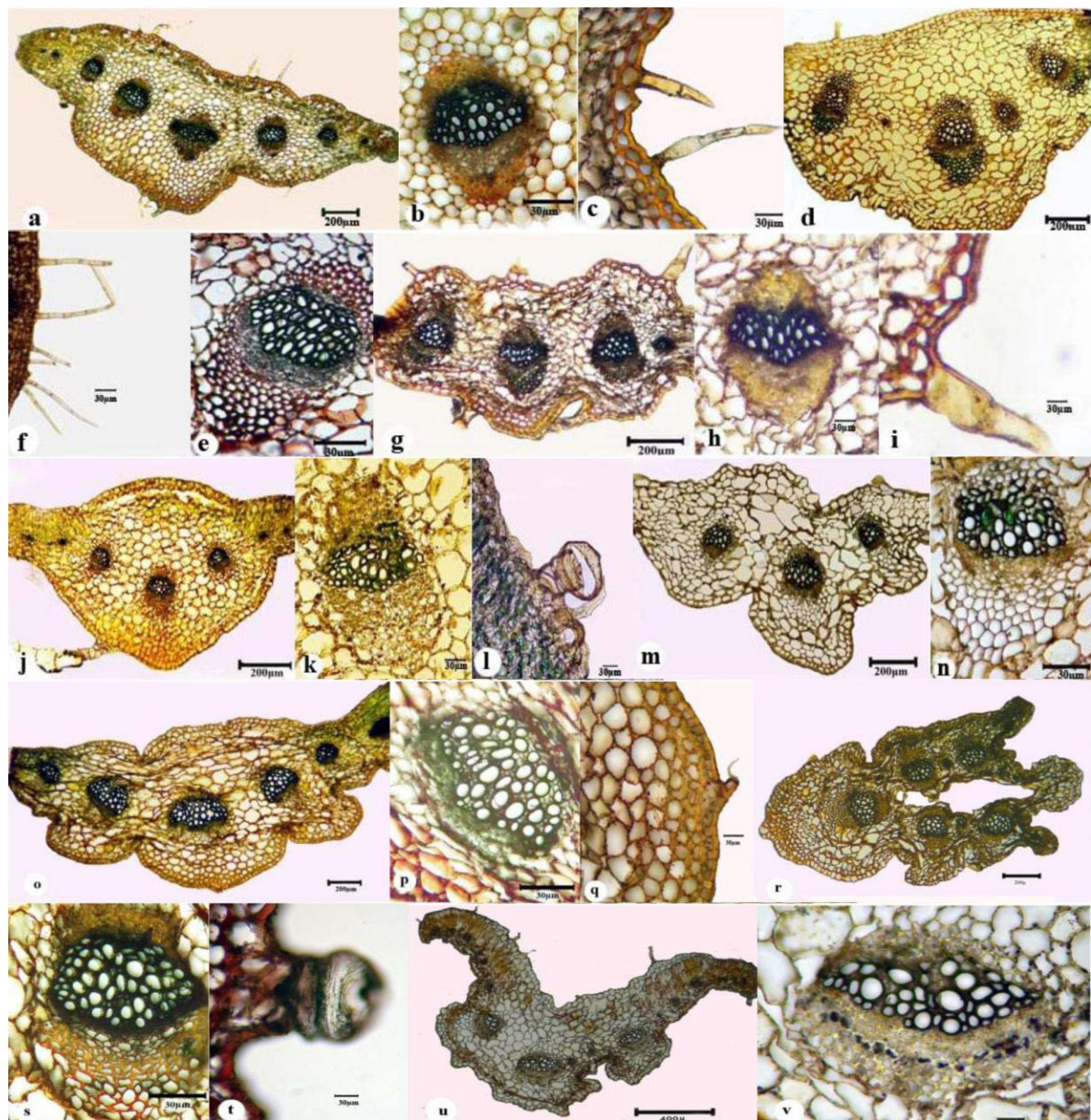


Figure 1. ...

Cullen, 1966). Moreover, Mobayen (1985) introduced two subspecies *G. fimbrilligerum* Boiss. subsp. *annuum* and *G. fimbrilligerum* subsp. *Ophyocarpum*.

Anatomical structure of leaves (Dickson, 1935; Esau, 1977; Fahn, 1990; Batanouny, 1992) and laticifers (Solereeder, 1908) have been presented for some species. Solereeder (1908) claimed that the type of laticifers, composition and location of them in different organs are among traits that are taxonomically valuable. Metcalfe and Chalk (1950) reported several anatomical traits of poppy family that exclusively can be taxonomically useful in the identification and delimitation of the species.

Furthermore, anatomical characteristics of *Glaucium flavum* Cr. have been investigated (Dickson, 1935; Esau, 1977; Fahn, 1990; Batanouny, 1992). The most important studies, in respect of anatomy, are restricted to few economical species such as *Papaver somniferum* (Dickinson and Fairbairn 1975) and *Glaucium flavum* (Nessler, 1992; Bercu *et al.*, 2006) which mainly concerned to ultra-structure of alkaloid sac and laticifers, respectively. Azizian and Alishahi Norani (1997) studied anatomical characteristics of fruit and blade with emphasis on latex tubes in species of *Glaucium*. Furthermore, Carlquist and Hoekman (1985) studied anatomical structure of wood in *Romneya* and *Dendromecon*. Carlquist and Zona (1988) continued his studies in cooperation with Zona on structure of wood in *Papaveraceae*. Some anatomical features of midrib and fruit of *Glaucium* are of diagnostic value (Solereeder, 1908; Metcalfe and Chalk, 1950). Anatomical characteristics of the fruit, stem and petiole in four studied genera confirm the results of the previous studies. Because of the high variation of morphological characters of *Glaucium*, this research was aimed to 1) provide some anatomical characters of above- named genera, specifically *Glaucium*; 2) assess these characters' value in sorting out of the species.

### Materials and methods

In this survey, all voucher specimens are deposited at TUH (acronyms according to Holmgren *et al.*, 1990) listed in Table 1. Because of the high variation of morphological diagnostic features of these species and difficulty of their identification, only representative specimens of any species were used in the study. For anatomical studies, dried basal leaves, fruits and stems were fixed in FAA 70 (Formalin, Glacial acetic acid and 70% Ethanol, 5: 5: 90, respectively), cross sections were made at the middle of blade, fruit, stem and were stained white methyl green and bismarck brown colors and then photographed by Leitz light microscope model Wetzlar, Nikon camera (Coolpix S10). For measuring required characters, Mesurepro software model HASP 2.17 was used.

Table 1. *Glaucium* species, their localities and voucher specimens

Species	Locality
<i>Glaucium elegans</i>	Tehran: Jajrud, Azad, Ganjalizadeh TUH-8864
<i>G. contortuplicatum</i>	Mazandaran: 40 km to Amol, near Andovar village, Attar, Okhovat & Mehdigholi, TUH-26352
<i>G. fimbrilligerum</i>	Tehran: ozaneh near Firuzkuh, Ghahreman, Aghostin, Shikholeslam, TUH-941
<i>G. pulchrum</i>	Zanjan: Abhar, Yazdan dust, TUH-8884
<i>G. corniculatum</i>	Azərbayjan: 10 km after Oshnavieh to Urumieh, American _ Iranian Expedition, TUH-34697
<i>G. grandiflorum</i>	Kermanshah: Bakhtaran. Ghahreman, TUH-8964
<i>G. haussknechtii</i>	Zanjan: Arijan vallage, Zarre, TUH-12633
<i>G. oxylobum</i>	Esfahan: 60 km to Delijan from Esfahan, American _ Iranian Expedition, TUH-33936
<i>Roemeria refracta</i>	Lorestan: Khorramabad, Veissina, Dogar. Veiskarami, TUH-23625
<i>R. hybrida</i>	Khorasan: Gonabad. Mobayen, TUH-24313
<i>Hypocoum pendulum</i>	Lorestan: Khorramabad, Chegeni Khatereh, Veiskarami, TUH-23626
<i>Chelidonium majus</i>	Gilan: Lahijan, Mobayen, TUH-8860

## Comparative anatomy of some selected species of the poppy family (Papaveraceae) in Iran

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### Abstract

In the present study, a comparative anatomical study of stem, midrib of basal leave and fruit is provided. In this paper, 45 anatomical characters of stem, midrib and fruit of some species such as *Glaucium grandiflorum*, *G. fimbriigerum*, *G. corniculatum*, *G. contortuplicatum*, *G. haussknechtii*, *G. elegans* and other species such as *Roemeria hybrida*, *R. refracta*, *H. pendulum* and *C. majus* are presented. In this study, we report the presence of glandular hair in *G. oxylobum* and *G. pulchrum* for the first time. Among the examined traits, some traits such as shape of placenta, the presence of hairs, carpellary angle, the shape of carpellary angle, the number of external and internal layers of parenchyma of outer wall of ovary, the number of vascular bundles of placenta in the fruit and the presence of the central cavity, existence of hairy glands, the number of vascular bundles of phloem, lack of strengthening tissue surrounding the vascular bundles in the midrib of some species are different and the differences are significant enough to be useful in the delimitation of some species of *Glaucium*. Anatomical features of midrib in these genera indicate, in spite of some differences, the fundamental structure is similar, so cross section of blade by itself cannot be a good scale for identification of the species of the genus *Glaucium* and other genera. Anatomical features of stem are neither a suitable scale for separating species of *Glaucium*, nor for other genera. Anatomical features of fruit are useful in identification and determination species of genus *Glaucium*, but also, for four other genera, especially cross section of the fruit of *H. pendulum* showed significant differences in comparison with other genera. Some anatomical features of fruit in *Glucium* can be used in taxonomically grouping of the genus. The most important traits are the apparent shape of ovarian placenta.

**Key words:** Anatomy, *Chelidonium*, *Glaucium*, *Hypocoum*, *Roemeria*, Papaveraceae, Iran

### Introduction

Poppy family (Papaveraceae) comprises of approximately 26 to 42 genera and 690 to 800 species in the world (Judd *et al.*, 1999). The members of Papaveraceae are shrub, herbaceous perennials and annuals distributed in the temperate and the subtropical regions of the world. Among five genera of family Papaveraceae in Iran, *Glaucium*, *Hypocoum*, *Chelidonium* and *Roemeria* consist of 10, 1, 1 and 2 species, respectively (Rechinger and

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