Annals of Botany **82**: 843–847, 1998
Article No. bo980774

Variation and Allometry of Seed Weight in *Aeschynomene americana*

JIANHUA ZHANG

Department of Agronomy and Plant Genetics, University of Minnesota, St. Paul, MN 55108, USA

Received: 2 February 1998 Returned for revision: 12 July 1998 Accepted: 21 August 1998

One hundred and fifty seeds from each of 72 *Aeschynomene americana* populations with world-wide origin were weighed individually and the mean and distribution of seed weight were calculated and compared between populations. Mean seed weight varied less than five-fold between populations with about 80% having a mean seed weight between 2–3 mg. However, differences between the largest and smallest seeds (2–8 mg) ranged two-17-fold within populations. Populations that had larger mean seed weights also had larger maximum and minimum seed weights but not larger standard deviations than those with smaller mean seed weights. Seed distributions for populations with a mean seed weight ⩽ 2 mg were positively skewed while those for all populations with a mean seed weight ⩾ 4 mg were negatively skewed. Populations that flowered later produced smaller seeds than those that flowered earlier.

Key words: Seed weight, weight distribution, inter-population variation, correlation among traits, *Aeschynomene americana*.

INTRODUCTION

Variations in seed weight between or within plant species are due to the evolutionary responses of plants to (*a*) maximize the potential fitness by producing a large number of seeds and (*b*) increase the chances of establishment of the resulting seedlings through a great allocation of maternal resources to individual seeds (Willson, 1983; Westoby, Rice and Howell, 1990; Westoby, Jurado and Leishman, 1992). Because massive seed production and great allocation to individual seeds are mutually exclusive due to the limitation of maternal resources, compromise between the two reproductive strategies produces a wide range of seed weights depending on various biological and environmental factors (Mazer, 1989; Leishman, Westoby and Jurado, 1995). Results have shown that plant species occupying closed or dry habitats usually produce larger seeds than those found in open or moisture-rich habitats (Salisbury, 1942, 1974; Baker, 1972; Mazer, 1989). Seed weight also varies with the height, growth form and dispersal mode of plants within different communities (Mazer, 1989; Leishman *et al*., 1995; Lord, Westoby and Leishman, 1995). Significant dependence of seed weight on environmental or other plant variables is considered by some researchers to be evidence of ecological relations between variables (Leishman *et al*., 1995; Westoby, Leishman and Lord, 1995). However, others suggest that correlations between seed weight and environmental or other plant variables across different species may not reflect true ecological relationships between the two sets of variables. The observed trend using plants across phylogenies may have been confounded by phylogenetic constraints (Harvey, Read and Nee, 1995*a*, *b*). In fact, several studies did show the existence of phylogenetic patterns underlying the relationships between seed weight and other plant attributes (Mazer, 1989; Lord *et al*., 1995). It is thus important to minimize the confounding effects through phylogenetic correction of the data (Harvey and Pagel, 1991; Frumhoff and Reeve, 1994) or, alternatively, by focusing on phylogenetic lineages such as plants from different populations of a given species or different species from a given genus to reduce the risk of biased interpretations of the results.

Reports on inter-population variation in seed weight and their correlation with environmental or other plant variables are surprisingly limited in the literature. The scarcity of studies on this particular issue does not necessarily mean that inter-population variation in seed weight is minimal or of little ecological significance. Rather, researchers generally agree that variation in seed weight does have significant impact on plant growth and establishment (Fenner, 1985; Roach and Wulff, 1987). The magnitude and mechanisms underlying inter-population variation in seed weight represent an understudied area in plant ecology where some fundamental questions remain to be answered. For example, studies have documented variation in seed weight both between and within plant populations (Hawke and Maun, 1989; Michaels *et al*., 1988; Aronson, 1992; Zhang and Hamill, 1996). Great variation within populations, which can be many-fold in magnitude (Hawke and Maun, 1989; Zhang and Maun, 1990; Banovetz and Scheiner, 1994), may result in overlapping seed weight distributions and decrease the likelihood of differentiation between populations. Little effort has been made to evaluate the extent of variation in seed weight within and between populations and their relative contribution to the total variability within a species. Furthermore, previous studies focused mainly on mean seed weight for comparisons between species or populations. Populations with different mean seed weights are expected to have evolved under different selection pressures (Willson, 1983; Westoby *et al*., 1990; 1992). However, populations

0305-7364/98/120843+05 $30.00/0

with similar mean seed weight may differ in seed weight distribution and will probably have experienced different selection pressures as well. Focusing on mean seed weight alone could result in failure to detect ecologically meaningful variation in seed weight between populations. Finally, plant traits are commonly intercorrelated (Schlichting, 1986, 1989) and seed weight has been shown to correlate with other plant traits such as plant height and growth form (Mazer, 1989; Leishman *et al*., 1995). Thus, it is also worthwhile examining the pattern and degree of integration between seed weight and other traits across populations.

In this study, the extent of between- and within-population variation in seed weight of 72 *Aeschynomene americana* populations were investigated and the dependence of mean seed weight on five measurements taken from the maternal plants was examined.

MATERIALS AND METHODS

Aeschynomene americana L. is an annual or perennial herbaceous shrub known as a green manure or pasture legume in tropical areas around the world (Singh, 1968; Hodges *et al*., 1982; Bishop, Ludke and Rutherford, 1985).

The wide spread of *A. americana* results from natural distribution and importation for agricultural use. Populations from different locations vary greatly in morphology, phenology and several reproductive traits, including time of flowering and flower size (Bishop, Pengelly and Ludke, 1988). These variations are probably due to natural selection because artificial selection for particular traits is not common practice for this species.

Seeds of 72 *A. americana* populations were obtained from a larger pool of collections of the CSIRO Division of Tropical Crops and Pastures Research Station at Samford, Queensland, Australia. The 72 selected populations are all annuals originating from more than 17 different countries in America (58 populations), Asia (3), Africa (2), and some unknown locations (8) ranging from 4°07′ N to 22°24′ N. The seed for each population was obtained by growing ten plants per population for one generation under similar conditions in an experimental garden located at the Samford Pasture Research Station. During the growing season, plant height, crown diameter, leaf/shoot ratio, days to flowering, and duration of seed development were measured. Results are reported elsewhere (Bishop *et al*., 1988). Mature seeds were collected from the ten plants of each population, cleaned, and then stored at 4 °C before use (Dr. B. Pengelly, pers. comm.). The seed samples used in this study were taken randomly from well mixed seed collections and then screened visually to exclude insect damaged and defective seeds.

In 1995, a subsample of 150 seeds was randomly selected from each of the 72 populations by dividing and subdividing the seed sample. A sample size of 150 seeds was chosen in this study because it was the maximum number of seeds that could be processed given time and labour constraints. Preliminary analyses of the data showed that the mean seed weight of a given population varied only within a range of 2% when the sample size exceeded 130 seeds, suggesting that data generated using the 150 seeds were reliable. The selected seeds for each population were air dried for 2 weeks and weighed (to the nearest 0·01 mg) individually using an electronic balance.

TABLE 1. *Description of plant and seed variables used in this study*

Variable	Mean ± s.e.
Crown height (cm)	144 ± 38
Crown diameter (cm)	166 ± 29
Dimension (crown height × diameter) (cm^2)	23665 ± 913
Leaf/shoot ratio (%)	33·15 ± 3·95
Days to flowering (d)	111 ± 23
Duration of seed development (d)	41 ± 14
Mean seed weight (mg)	3·27 ± 0·93

Mean ± s.e. are calculated across all 73 populations.

A box plot (Kuo, McDonald and Fox, 1992) was used to show the percentiles of seed weight distribution for each of the 72 populations. Inter-population differences in mean seed weight were examined using ANOVA (Proc GLM; SAS, 1990) with population as a fixed factor. Population was considered a fixed factor because the 72 populations were all annuals subjectively selected from a larger pool of *A. americana* populations that had sufficient seed. The proportion of the total variance in seed weight due to between- and within-population variation was calculated by dividing the sum of squares of each factor by the total sum of squares derived by the maximum likelihood method (Proc Varcomp; SAS, 1990). The skewness of seed weight distribution within each population was compared with a normal distribution following Zar (1984). To determine whether mean seed weight depended on any of the measured traits of the maternal plants, the calculated mean seed weight of the 72 populations was regressed against each of the measurements of the maternal plants (Table 1).

RESULTS

Mean seed weights of the 72 populations varied less than five-fold from 1·4–6·4 (0·42–1·31s.d.) mg with about 80% of the populations having a mean seed weight ranging from 2–3 mg. Within a given population, a difference of 2–8 mg (2–17-fold) was observed between the largest and smallest seeds. Over 90% of the populations had a 2–4 mg difference in weight between seeds of the two extreme weights. Populations having large mean seed weights also had large maximum ($y = 3{\cdot}66 - 0{\cdot}08x + 0{\cdot}14x^2$, where y is maximum and x is mean seed weight, $R^2 = 0{\cdot}741$, $P < 0{\cdot}001$) and minimum ($y = -0{\cdot}293 + 0{\cdot}468x$, where y is minimum and x is mean seed weight, $R^2 = 0{\cdot}417$, $P < 0{\cdot}001$) seed weights, but not large standard deviations ($R^2 = 0{\cdot}184$, $P > 0{\cdot}05$).

Differences in mean seed weight between populations were statistically significant (Table 2) according to analysis of variance. Subsequent multiple range tests between means of 2556 possible pairs of populations revealed significant differences in mean seed weight in 1858 (72·69%) of the comparisons. The population ranked 71 in mean seed weight had absolutely larger seeds than those ranked 1, 2, 4, 5, 7, 16 and 17 as the range of seed weight distribution of the

TABLE 2. *ANOVA table and the relative contribution of between and within population variance to the total variation in seed weight of* A. americana

Source	d.f.	MS	F	P	Variance explained (%)
Between population	71	113·78	220·65	< 0·0001	52·94
Within population	10585	0·5156			47·06

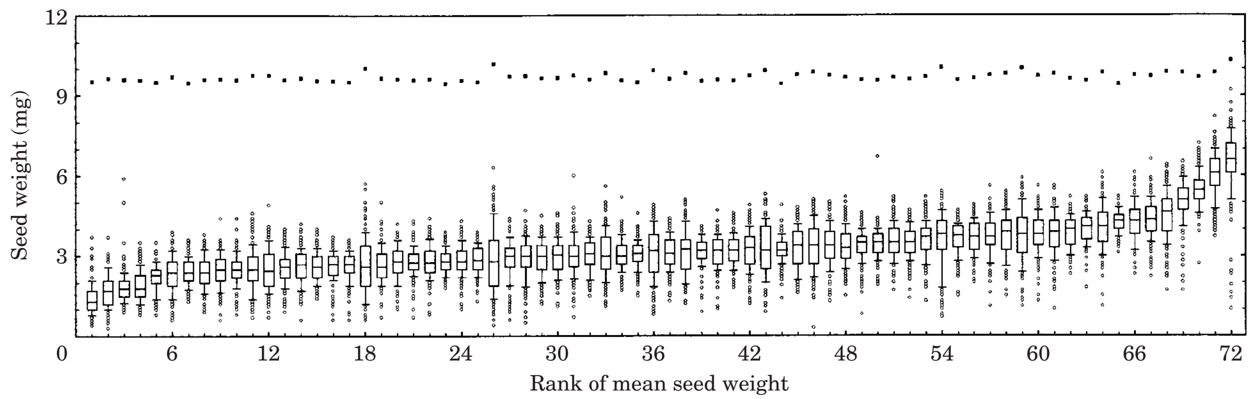

FIG. 1. Box plot showing the 50 (solid line), 25–75 (box) and 10–90 (capped bars) percentile of seed weight distribution within each population. Data points outside the 10–90 percentile are denoted by open circles. Populations of *A. americana* were ranked along the horizontal axis based on mean seed weight. Solid squares indicate the standard deviation (increased by 9 for presentation purpose) of mean seed weight for each population.

former did not overlap at all with that of the latter populations (Fig. 1). When focusing on the distribution of 80 % of the seeds that are closer to the population mean (the range denoted by capped bars), all the populations ranked below 5 did not overlap at all with those ranked 63 or above. The standard deviation of mean seed weight varied between 0·42 and 1·32 (mean = 0·69) independent of mean seed weight. The standard deviation of mean seed weight across population means was 0·86.

The seed weight distribution was normal in nine, positively skewed (significantly different from a normal distribution at $P < 0{\cdot}05$) in 14, and negatively skewed (significantly different from a normal distribution at $P < 0{\cdot}05$) in 49 of the 72 populations. As a group, populations whose seed weight distributions were positively skewed had significantly ($P < 0{\cdot}005$) smaller mean seed weight ($2{\cdot}55 \pm 0{\cdot}16$ mg) than populations with a negatively skewed distribution ($3{\cdot}46 \pm 0{\cdot}013$ mg) according to Tukey's test. All the populations with a mean seed weight $\leqslant 2$ mg had a positively skewed seed weight distribution, while populations with a mean seed weight $\geqslant 4$ mg had a negatively skewed seed weight distribution (Fig. 2). Within the 2–4 mg weight range, where the mean seed weight of most populations resided, some populations had a positively skewed, some had a negatively skewed, and some had a normal seed weight distribution.

The mean seed weight of populations did not correlate with height, crown diameter, dimension, leaf/shoot ratio, or the duration of seed development of maternal plants (data not shown). However, the mean seed weight depended on the number of days of post emergence growth required for

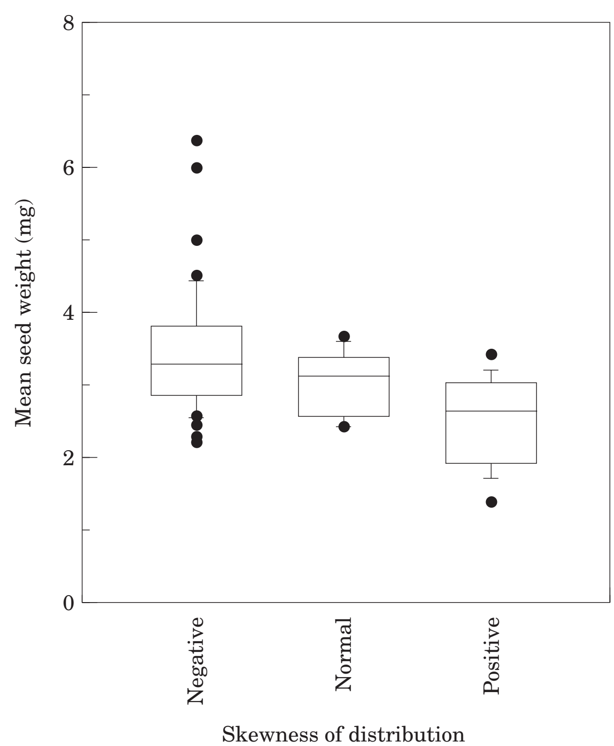

FIG. 2. Box plot showing the range of mean seed weight of the populations with negatively skewed, normal and positively skewed seed weight distribution. Horizontal lines indicate the 50 percentile, boxes the 25–75 percentile, capped bars the 10–90 percentile, and closed circles the data points outside the 10–90 percentile.

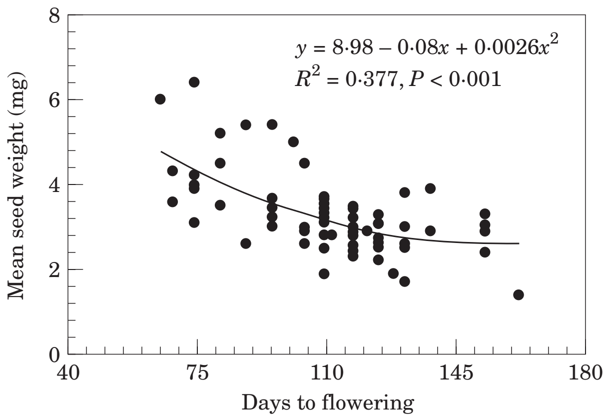

FIG. 3. Regression of mean seed weight against the number of days to flowering across the 72 *A. americana* populations.

the maternal plants to flower across populations. Early flowering populations produced larger seeds than those that flowered later in the growing season (Fig. 3).

DISCUSSION

Seed weight, which reflects potential food reserves available to subsequent seedlings, is commonly considered an important trait that determines the success of individual plants (Mazer, 1989; Westoby *et al*., 1990, 1992). Extensive studies have been conducted on the variability, ecological importance and evolution of seed weight in plants within a population, or species within or between communities (Staton, 1984; Mazer, 1989; Zhang and Maun, 1990; Leishman *et al*., 1995). Studies focusing on populations within a species are strikingly rare. This study shows a two–17-fold variation in seed weight within populations, but a less than five-fold variation in mean seed weight between populations of *A. americana*. Overlapping in seed weight distribution occurs, with a great contribution of the within-population components to the total variation in seed weight. This more or less fuzzy differentiation in seed weight between populations due to great variation within each population may be responsible for the lack of interest in seed weight at the inter-population level. However, populations of *A. americana* do differ significantly in seed weight in spite of great within-population variation. Differences in a few populations are based not only on population means but also on the weight distributions of individual seeds within populations. This magnitude of intraspecific difference in seed weight can be considered substantial because overlaps in seed weight distribution are even commonly observed between species from different families or higher orders (Westoby *et al*., 1995; Lord *et al*., 1995). The underlying mechanisms that have shaped the more or less discrete seed weight distribution between those *A. americana* populations are unclear, but the results reveal that there is indeed potential for substantial genetically based seed weight differentiation to occur between populations of a species.

Previous studies concerning inter-specific or inter-population variation in seed weight focused mainly on mean seed weight, with little attention being paid to the distribution of individual seeds within each species or population. According to this study, the 72 *A. americana* populations differ in mean seed weight as well as the pattern of weight distribution of individual seeds. Some populations have negatively, while others have positively skewed seed weight distribution (Fig. 2), although their means may or may not be similar. The outcome would be a positively or negatively skewed weight distribution of the resulting plants given that seedling size depends on seed weight (Black, 1958; Fenner, 1985; Roach and Wulff, 1987). Populations with a group of relatively large individuals and a few small ones (negatively skewed) are expected to have different patterns of intraspecific competition and population dynamics compared to populations with a group of relatively small individuals and a few large ones (positively skewed). This argument is supported by a recent study asserting that the success of a plant depends not only on its own size but also on the size of neighbouring plants, which varies depending on the size distribution of the seedling population (Zhang and Hamill, 1997). Furthermore, the amount and weight distribution of seeds in the soil seed bank may also vary between populations with different seed weight distribution because seed longevity depends greatly on seed weight (Fenner, 1985; Banovetz and Scheiner, 1994). In all these cases, differences in the pattern of seed weight distribution may affect the amount and pattern of seedling recruitment and dynamics.

Taller plants or plants experiencing strong competition normally produce larger seeds than shorter ones across different phylogenies in different communities and environments (Salisbury, 1974; Mazer, 1989; Leishman *et al*., 1995). This generalization may not hold for *A. americana*, in which mean seed weight is negatively correlated with the number of days required for the parent plants to flower. For annuals such as *A. americana*, the length of pre-flowering growth determines the competitive ability of plants and is closely related to final fitness of plants. Habitats where competition between plants is strong would probably favour prolonged vegetative growth and thus lower, rather than higher mean seed weights. However, similar studies on other plant species are needed before any conclusive remarks can be made.

ACKNOWLEDGEMENTS

Seeds of *A. americana* used in this study were kindly provided by Dr. B. Pengelly at the CSIRO Division of Tropical Crops and Pastures Research Station at Samford, Queensland, Australia. I thank an anonymous reviewer for constructive comments on an earlier version of the manuscript.

LITERATURE CITED

Aronson J. 1992. Evolutionary biology of *Acacia caven* (Leguminosae, Mimosoideae): intraspecific variation in fruit and seed characters. *Annals of Missouri Botanical Garden* **79**: 958–968.

Baker HG. 1972. Seed mass in relation to environmental conditions in California. *Ecology* **53**: 997–1010.

Banovetz SJ, Scheiner SM. 1994. The effects of seed mass on the seed ecology of *Coreopsis lanceolata*. *American Midland Naturalist* **131**: 65–74.

Bishop HG, Ludke DH, Rutherford MT. 1985. Glenn jointvetch: a new pasture legume for Queensland coastal areas. *Queensland Agricultural Journal* **111**: 241–245.

Bishop HG, Pengelly BC, Ludke DH. 1988. Classification and description of a collection of the legume genus *Aeschynomene*. *Tropical Grasslands* **22**: 160–175.

Black JN. 1958. Competition between plants of different initial seed sizes in swards of subterranean clover (*Trifolium subterraneum*) with particular reference to leaf area and the light microclimate. *Australian Journal of Agricultural Research* **9**: 299–318.

Fenner M. 1985. *Seed ecology*. New York: Chapman and Hall.

Frumhoff PC, Reeve HK. 1994. Using phylogenies to test hypotheses of adaption: a critique of some current proposals. *Evolution* **48**: 172–180.

Harvey PH, Pagel MD. 1991. *The comparative method in evolutionary biology*. Oxford: Oxford University Press.

Harvey PH, Read AF, Nee S. 1995*a*. Why ecologists need to be phylogenetically challenged. *Journal of Ecology* **83**: 535–536.

Harvey PH, Read AF, Nee S. 1995*b*. Further remarks on the role of phylogeny in comparative ecology. *Journal of Ecology* **83**: 733–734.

Hawke MA, Maun MA. 1989. Intrapopulation variation in reproduction and seed mass of a beach annual: *Cakile edentula* var *lacustris*. *Journal of Coastal Research* **5**: 103–112.

Hodges EM, Kretschmer AE Jr, Mislevy P, Roush RD, Ruelke OC, Synder GH. 1982. Production and utilization of the tropical legume *Aeschynomene*. Circular S-290. Florida Agricultural Experimental Station, Institute of Foods and Agricultural Service, University of Florida, Gainsville.

Kuo J, McDonald S, Fox E. 1992. *SigmaPlot: user's mannual*. San Rafael, CA; Jandel Scientific.

Leishman MR, Westoby M, Jurado E. 1995. Correlates of seed size variation: a comparison among five temperate floras. *Journal of Ecology* **83**: 517–530.

Lord J, Westoby M, Leishman M. 1995. Seed size and phylogeny in six temperate floras: constraints, niche conservatism, and adaptation. *American Naturalist* **146**: 349–364.

Mazer SJ. 1989. Ecological, taxonomic, and life history correlates of seed mass among Indiana dune angiosperms. *Ecological Monograph* **59**: 153–175.

Michaels HJ, Benner B, Hartgerink AP, Lee TD, Rice S. 1988. Seed size variation: magnitude, distribution, and ecological correlates. *Evolutionary Ecology* **2**: 157–166.

Roach DA, Wulff RD. 1987. Maternal effects in plants. *Annual Review of Ecology and Systematics* **18**: 209–235.

Salisbury EJ. 1942. *The reproductive capacity of plants*. London: G. Bell and Sons.

Salisbury EJ. 1974. Seed size and mass in relation to environment. *Proceedings of Royal Society of London, Biological Sciences* **186**: 83–88.

SAS. 1990. *SAS/STAT user's guide. Version 6, fourth edn*. Cary, NC: SAS Institute Inc.

Schlichting CD. 1986. The evolution of phenotypic plasticity in plants. *Annual Review of Ecology and Systematics* **17**: 667–693.

Schlichting CD. 1989. Phenotypic plasticity in phlox. II. Plasticity of character correlations. *Oecologia* **78**: 496–501.

Singh RG. 1968. Changes in the physiochemical properties of soil on the decomposition of *Aeschynomene americana*. *Tropical Agriculture* **124**: 95–102.

Staton ML. 1984. Several variation in wild radish: effect of seed size on components of seedling and adult fitness. *Ecology* **65**: 1105–1112.

Westoby M, Jurado E, Leishman M. 1992. Comparative evolutionary ecology of seed size. *Trends in Ecology and Evolution* **7**: 368–372.

Westoby M, Leishman M, Lord JM. 1995. On misinterpreting the 'phylogenetic correction'. *Journal of Ecology* **83**: 531–534.

Westoby M, Rice B, Howell J. 1990. Seed size and plant growth form as factors in dispersal spectra. *Ecology* **71**: 1307–1315.

Willson MF. 1983. *Plant reproductive ecology*. New York: Wiley.

Zar JH. 1984. *Biostatistical analysis. 2nd edn*. Englewood Cliffs, N.J: Prentice-Hall, Inc.

Zhang J, Hamill AS. 1996. Responses of *Abutilon theophrasti* to agricultural management systems. *Weed Search* **36**: 471–481.

Zhang J, Hamill AS. 1997. Seed weight, intraspecific competition, and plant performance in *Abutilon theophrasti*. *Canadian Journal of Botany* **75**: 1614–1620.

Zhang J, Maun MA. 1990. Seed size variation and its effects on seedling growth in *Agropyron psammophilum*. *Botanical Gazette* **151**: 106–113.