VEGETATIONAL PATTERN WITHIN AN ABANDONED HAY-FIELD ADJACENT TO OLD-GROWTH AT GRIFFITH WOODS, HARRISON COUNTY, KENTUCKY

[See Part I in separate document]

II: COMPOSITIONAL GRADIENTS IN RELATION TO DISTANCE FROM ROAD (WITH PROBABLE POLLUTION) AND APPARENT EFFECTS OF HERBIVORY

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Preface for current drafts (Jan 2016). I have updated Part I (from 2014-15) and now completed Part II in a separate file. My goal for these materials is to publish a succinct summary in a standard scientific journal, and to develop further plans for research at this site. Their current format resembles a thesis or dissertation, with too much detail for regular publication in a standard journal. It is most important for interested people to communicate more and develop cooperative goals, ideally across the central Bluegrass region.

PART II: ABSTRACT [see Part I for title page and overall summary]

Given apparent effects of deer browsing (and perhaps other herbivory) on spatial pattern of woody plants in a central Kentucky hayfield abandoned for seven years, herbaceous composition was examined in more detail using Detrended Correspondence Analysis. With data from 199 20×5 m plots, Axis One score declines linearly away from the busy two-lane highway (US 62) that runs along the northwest long edge of this rectangular 250×175 m field. Relationship of Axis One to elevation, which declines irregularly away from the road, is weaker and it is reversed along rows at greater distance from the road, from positive to negative correlation. An initial set of soil samples, plus patterns in species' scores on the ordination and their known ecological characteristics, indicate that Axis One is primarily controlled by a general decline in soil fertility away from the road, but reversed locally in gullies. Such decline can be largely attributed to eutrophication from the road (calcium, nitrogen, phosphorous, etc.) rather than any natural pattern in the soil, which appears to be relatively uniform across the field. However, a detailed soil survey is still needed in this field, pending further cooperative work with managers. Axis Two of the ordination reveals a partially independent gradient that appears to be controlled by herbivory. This interpretation follows from indications of deer-browsing in 2007 (concentrated along a diagonal across the field), provisional mapping of deer trails in 2016, and patterns of individual woody species. White ash (dominant), maples and hackberry concentrate in less browsed zones; scattered red cedar, thorny Rosales, oaks and hickories, in more browsed zones. As well as patterns in the vegetation, signs of browsing on transects of planted blue ash provide support, tending to increase along Axes One (away from road) and Two. Based on these initial results, it is proposed to continue the research, with more concentration on patterns in soil and signs of herbivory, including smaller mammals. Such work is important for understanding how restoration should proceed from old fields to woods.

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INTRODUCTION

During 2003-2007, an old hayfield in Harrison County, Kentucky, became used for an initial planting of selected native plant species, as part of general restoration at what became the Griffith Woods Wildlife Management Area. The 745-acre “Silver Lake Farm”, with old growth known as Griffith Woods, provides the best opportunity for restoration of something like the original woodland on uplands of the central Bluegrass region (Campbell 2013, 2015). This region had unusual vegetation before Virginian settlement, due to highly fertile soils and influence from larger herbivores, but almost all of the uplands are now converted to farmland.

This field—dubbed the “Collection Field”—is located between US 62 and one of the best patches of old-growth woods at the farm. It became a convenient place for early trials in planting some species, and for general demonstration of vegetation at the farm. The plantings were laid out in a systematic grid, in order to allow future experimental use, and in 2007 a complete survey of the vegetation was made (Campbell 2015). Although the geology, soils and topography in this field are relatively uniform, a surprising degree of spatial pattern emerged in data from the vegetation survey, and from growth and mortality of the more abundantly planted species. The spatial patterns, especially in woody species, could be largely interpreted in terms of patterns in effects of browsing by deer, and perhaps other mammals (Campbell 2015).

The second report on this field, presented here, investigates the patterns with more rigor, focussing on multivariate analysis of variation in the herbaceous vegetation. Indicators of browsing in 2007 are combined with a more detailed survey of deer trails in January of 2016. Also, more information is assembled to investigate potential relationships with variation in soil across the field.

METHODS

In 2003, the Collection Field was organized into ten unmowed rows, each about 7.5 m wide and parallel to the road. Strips of about 10 m wide were mowed between these rows about twice per year from 2004 to 2007. The vegetation survey of 2007 has been detailed in the first report (Campbell 2015). The survey was made during September within 199 largely unmowed plots, each 20 × 5 m [100 m^2] and contiguous along the 20 row sections. Within these plots, all vascular plant species were recorded. Each herbaceous or subshrubby species was assigned a cover-class, using the following quasi-logarithmic eight-point scale: 0 = absent; 1 = present: 0–0.1%; 2 = 0.1–0.3%; 3 = 0.3–1%; 4 = 1–3%; 5 = 3–10%; 6 = 10–30%; 7 = 30–100%. These were rough visual estimates, and they often included vegetation along the edges of plots, where there was more influence from adjacent mowing and associated edge-effects. With exclusion of shrubs and trees, but inclusion of ground-covering woody vines, the number of species was 98 (Table 2). Nomenclature generally follows Weakley (2015). Sedges (*Carex*) were not identified to species with confidence, but later observations in the field did improve the suggested identifications. The list also excludes nine early flowering annuals that were recorded later, in 2008-2014: *Cardamine hirsuta, Cerastium glomeratum, Cruciata pedemontana, Erigeron philadelphicus, Geum vernum, Lamium purpureum, Myosorus macrosperma, Thlaspi alliaceum* and *Veronica arvensis*. The matrix of 199 plots and 98 species was ordinated using Detrended Correspondence Analysis (DCA: Hill & Gauch 1980) in PC-ORD software (McCune & Mefford 1999). There was no need for transformation of data, using the above 8-point scale.

Due to various practical limitations, a complete soil survey has not yet been conducted in this field, but two row sections were sampled in order to explore the range of variation. Samples were taken at intervals of 5 m in complete transects along the front and back row sections in the northern 2/3 of the field: rows N1 and N10 in Figure 1. These samples were dug from the top 10 cm, excluding leaves, twigs and other debris. They were pooled for each of the 20 × 5 m plots, with five samples thus contributing equally to each plot; samples from edges were split. The pooled samples were dried, ground and analyzed using using standard methods, in the Soil Testing Laboratory at the University of Kentucky. Soil pH was determined in 1 molar KCl and in Sikora II buffer (Sikora 2015). Nutrients were extracted with Mehlich III. Organic matter and nitrogen contents were determined as percentages. In addition, average height of goldenrod (*Solidago altissima*) was estimated visually around each sampled point, and the mean of these values was computed for each plot. These heights were then used as an overall "phytometer" of annual growth by the herbaceous vegetation within the field.

During the initial vegetation survey in 2007, scattered obvious indications of browsing by deer or other herbivores were noted in each plot; see Campbell (2015) for details. Also, a systematic assessment of apparent damage to the planted blue ashes was made. However, since major effects of herbivory were not anticipated, there was no consistent independent survey of indications throughout the field. Therefore, in January 2016 a provisional map of deer trails was made. Transects were walked between all rows, and pronounced ("major") deer trails that crossed into adjacent plats were noted. These trails were mostly about 20-30 cm wide, with ground vegetation only 0-10 cm tall and some soil exposed. In addition, less obvious trails were noted but with less precision; in some cases these "minor" trails appeared to involve rabbits or other mammals, rather than deer. In some areas, trails ran parallel to the rows and these were generally counted twice for each adjacent plot, especially if they appeared to meander between plots. In the analysis below, numbers of trails on the inside (away from US 62) of the front rows (S1 and N1) were doubled, because the outer side of these rows runs along the mowed right-of-way of US 62, where an assessment of trails was difficult.

Figure 1. The Collection Field at Griffith Woods, 12 Nov 2004, from Google Earth, centered on 38.34°N, 84.35°W. Road along upper edge is US 62, running to right at 43.5° NE. Rows are about 17.5 m apart and parallel to the road, except for slight divergence towards the lower left. Rows are numbered 1 to 10 on "south" side (S to left) and "north" side (N to right). Front rows (N1 and S1) are 15-20 m from the edge of asphalt. The series of plots within rows are indicated by numbers at upper and lower sides: 12 in all N rows; 9, 8 or 7 in S rows.

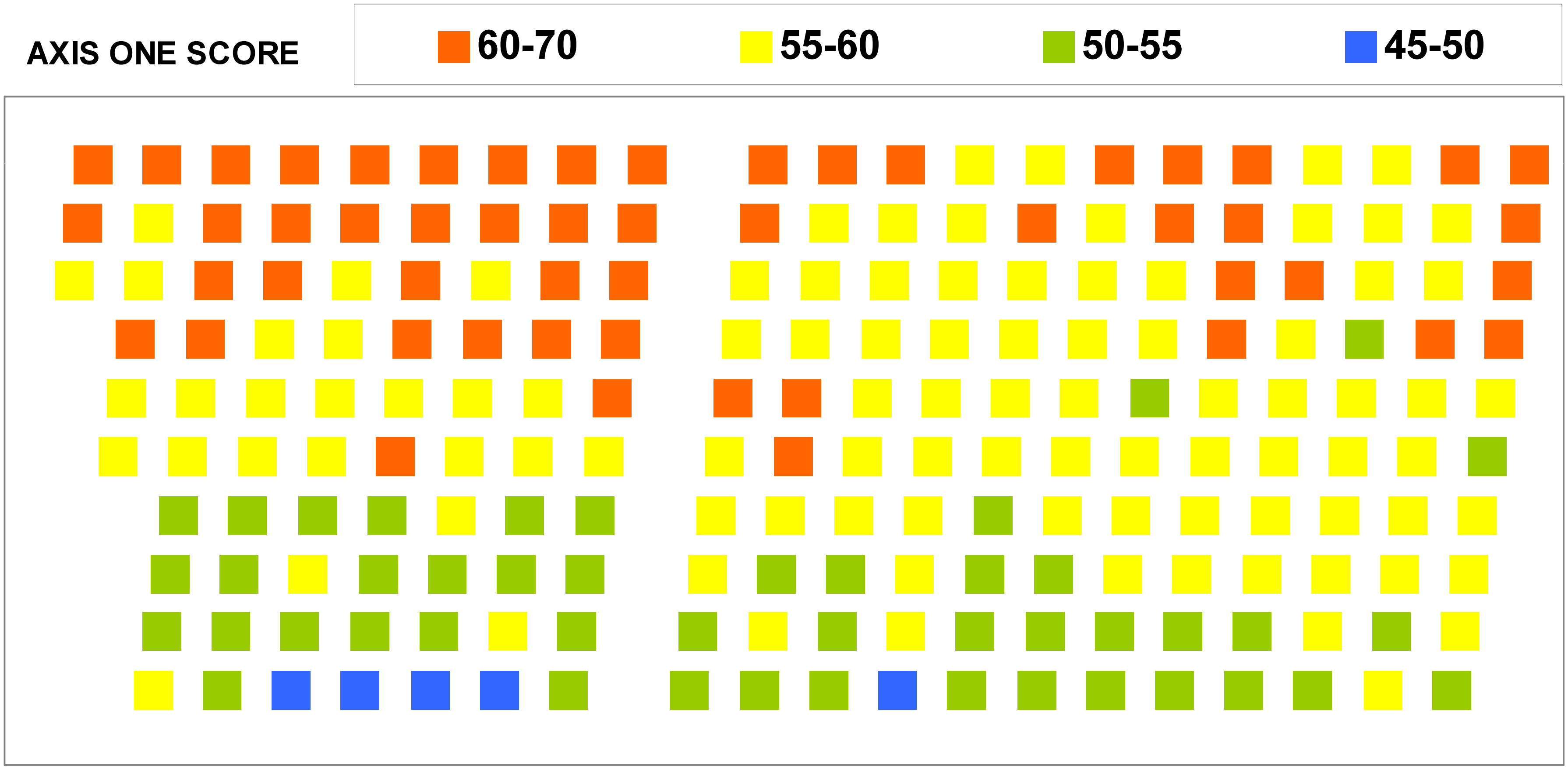

Figure 2a. Trend in ordination axis scores within plots: Axis One.
See Part I (Figure 1) to compare with trends in topography and vegetation; the road (US 62) runs along the upper edge of this map.

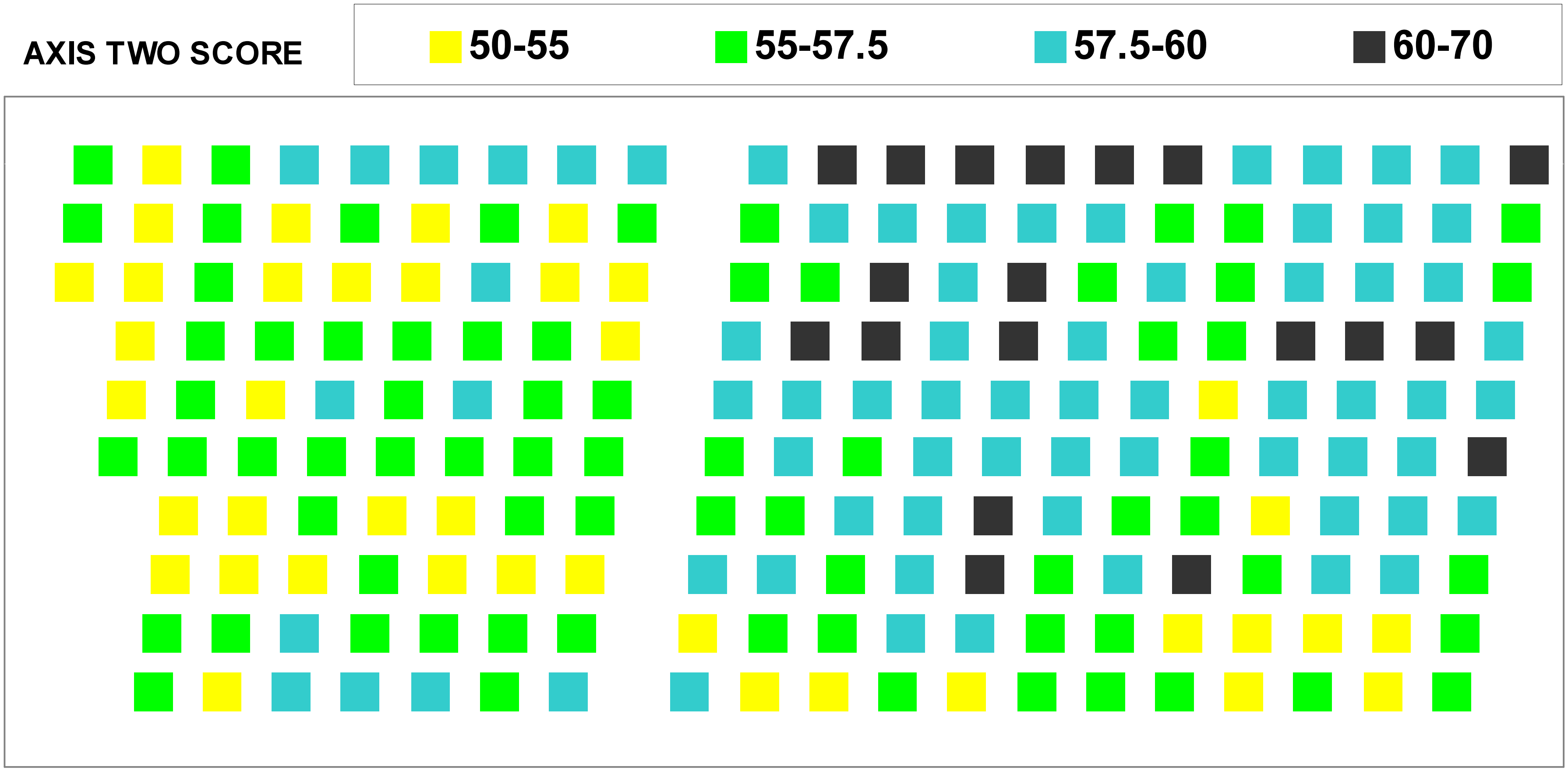

Figure 2b. Trend in ordination axis scores within plots: Axis Two.

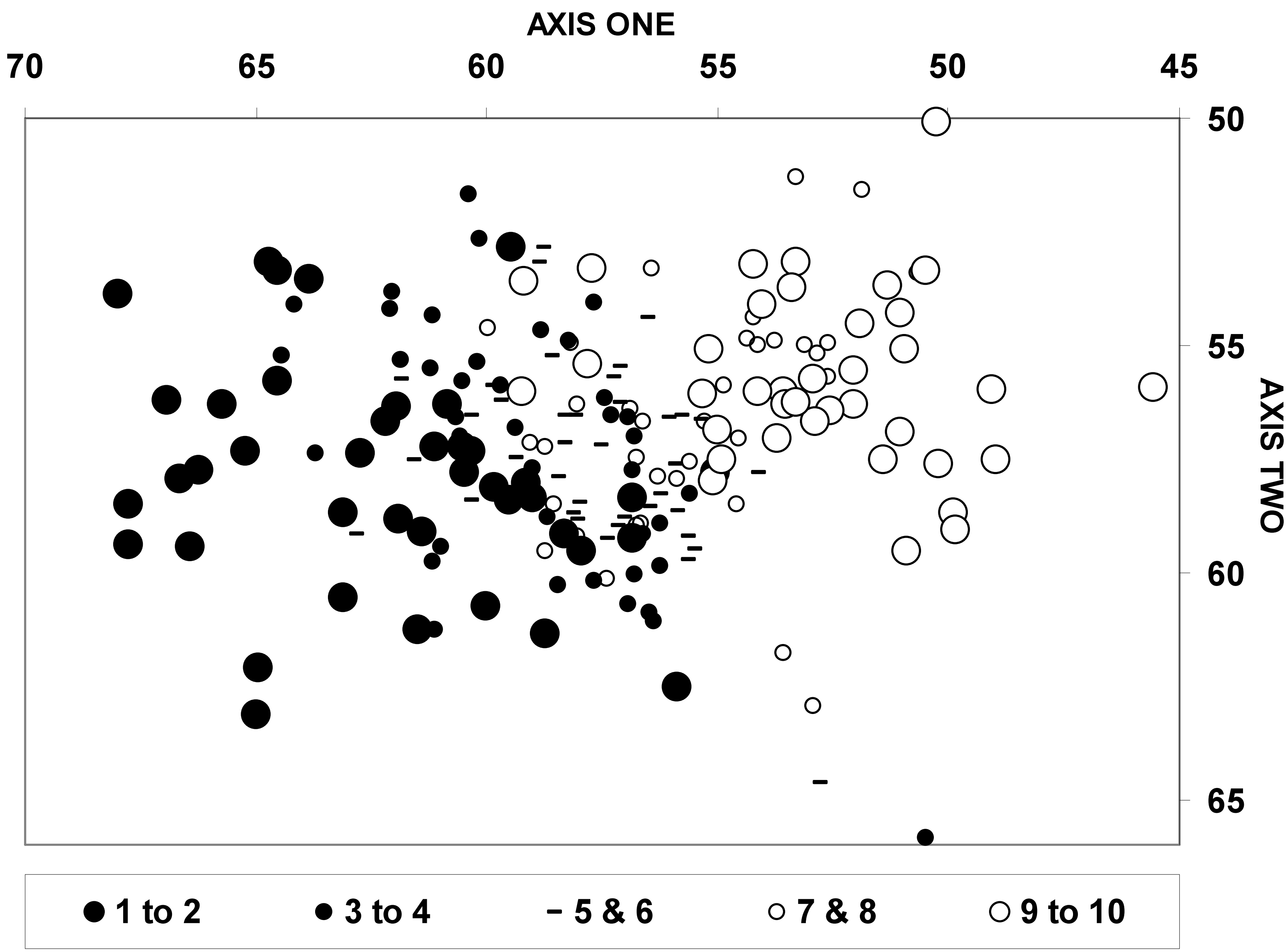

Figure 3a. Plot location in field overlaid on ordination: distance from road (as row number). Numbers are in units of 17.5 m (i.e. 1 to 2 = 17.5-35 m, etc.).

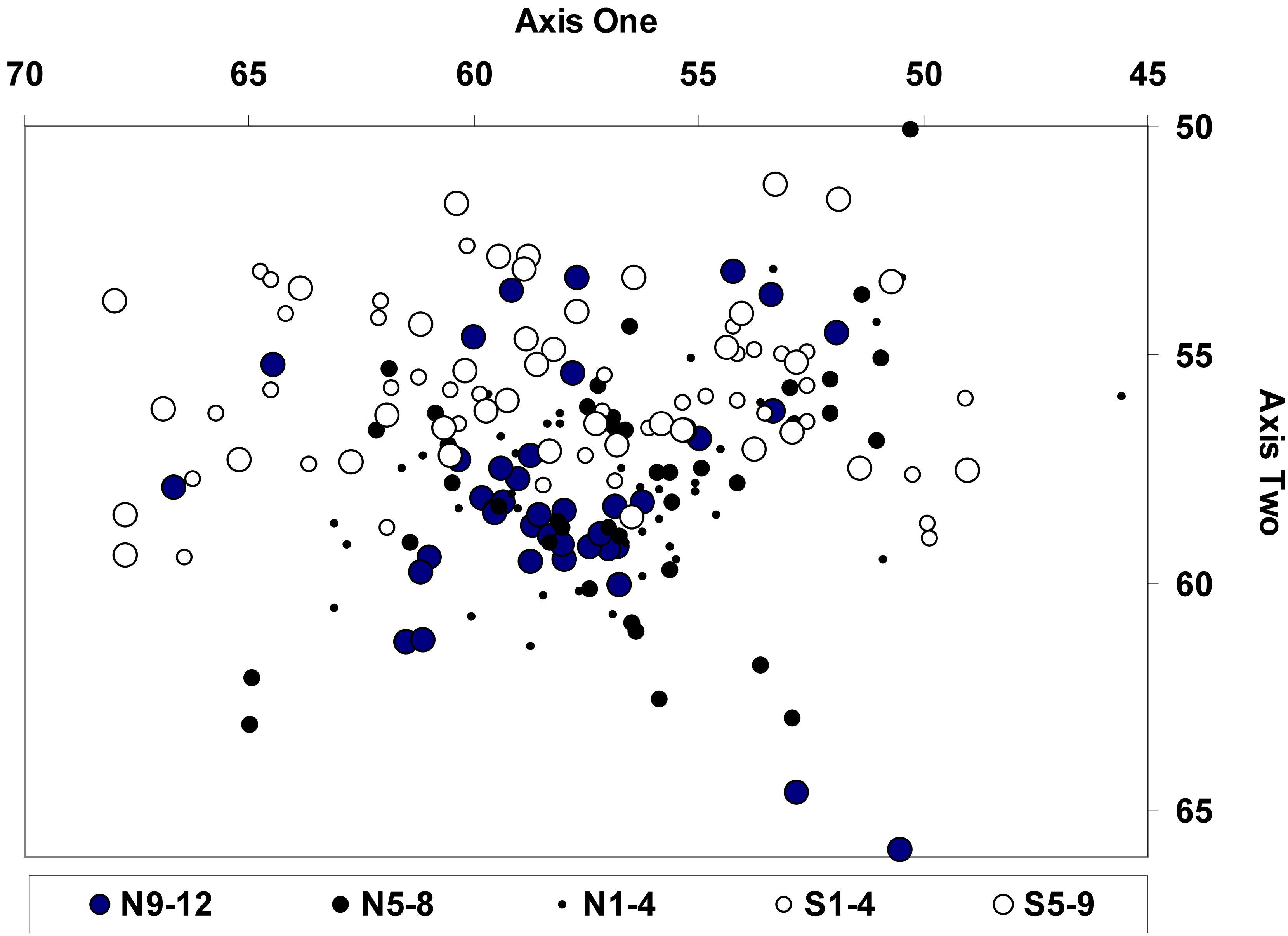

Figure 3b. Plot location in field overlaid on ordination: northern versus southern location in rows (centers 20 m apart). Correlation with Axis Two: r = 0.41, P < 0.0001.

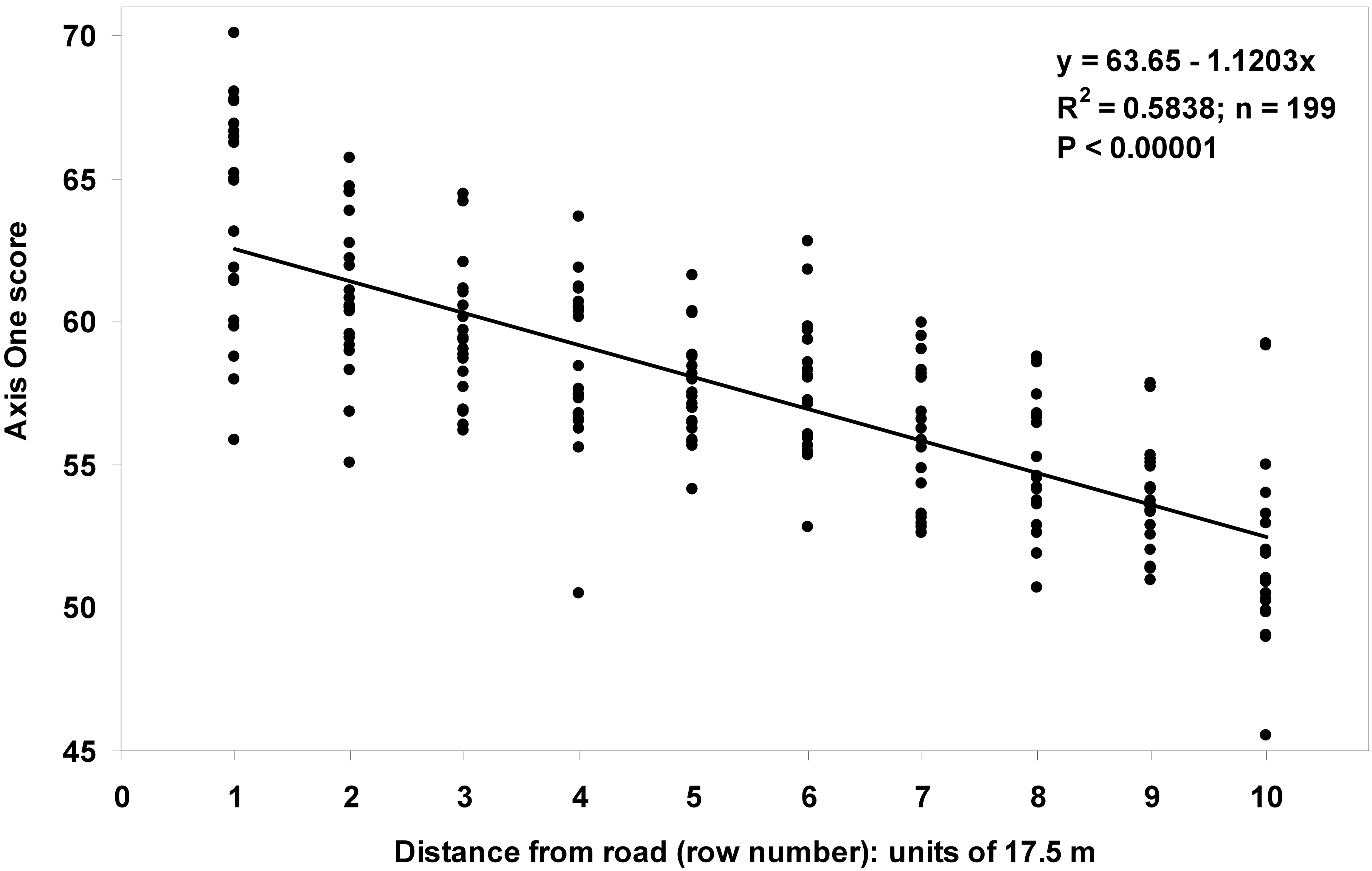

Figure 4a. Plot scores in relation to distance from road (row number): Axis One.

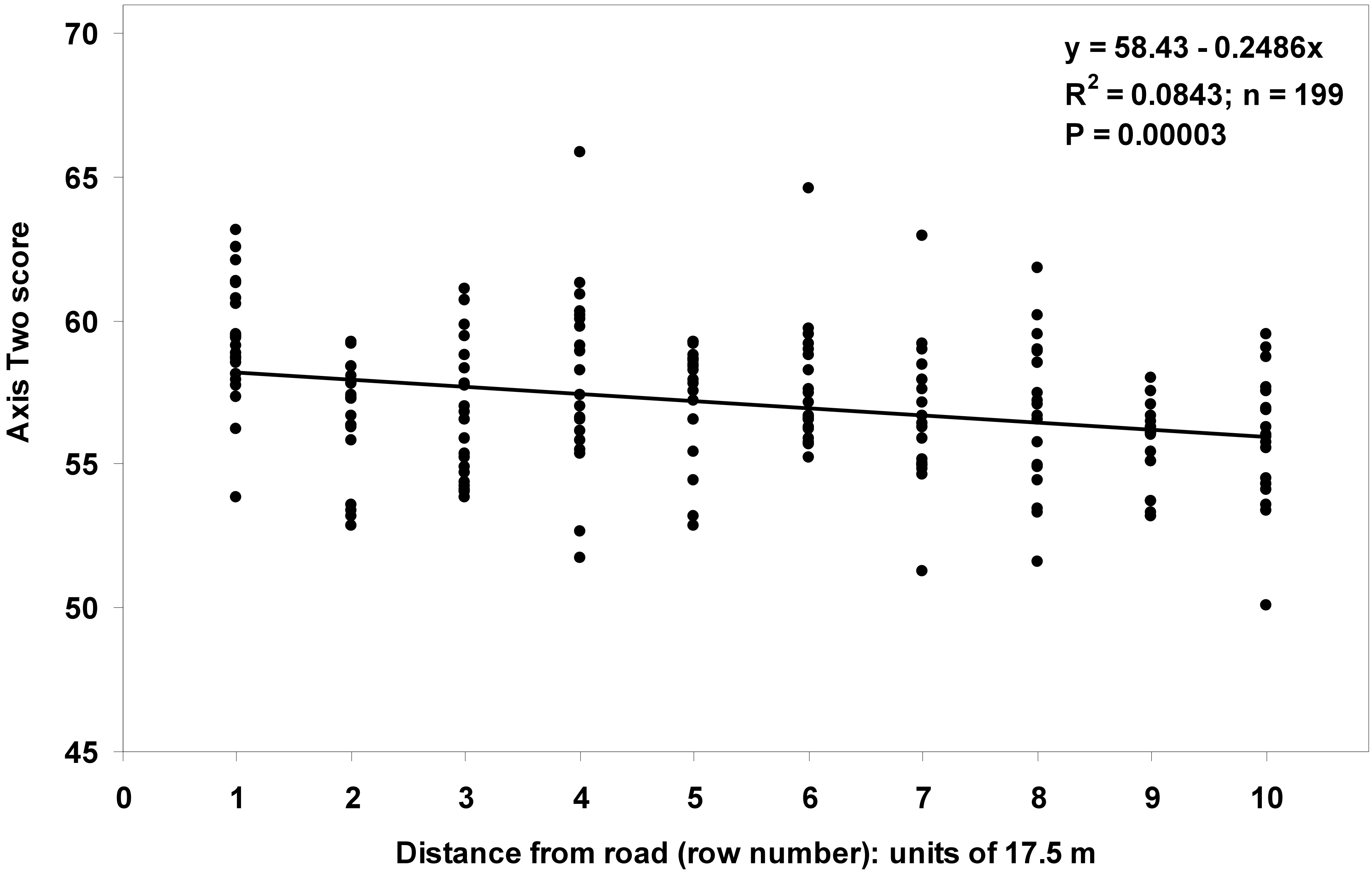

Figure 4b. Plot scores in relation to distance from road (row number): Axis Two

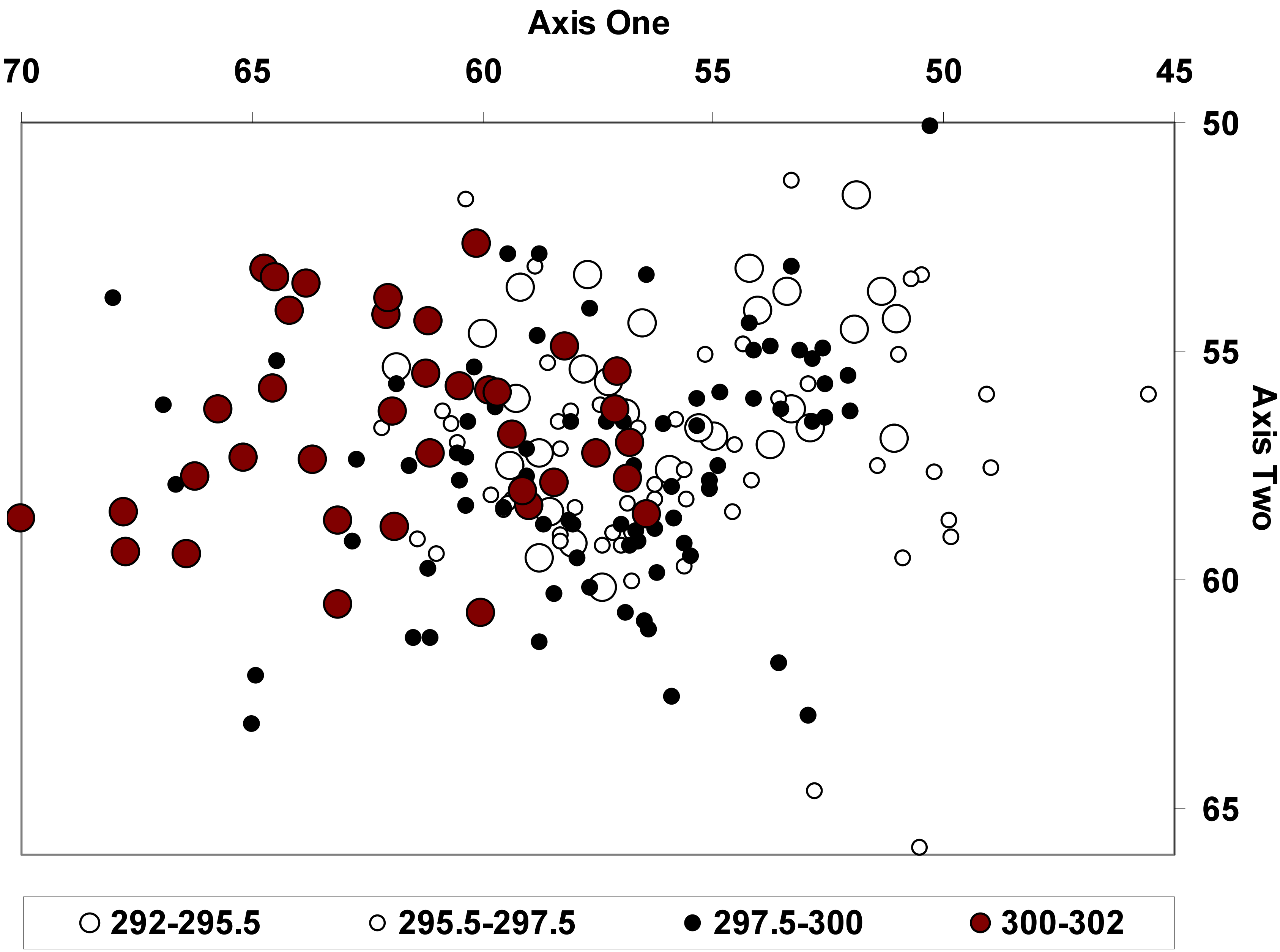

Figure 5. Elevation classes in m a.s.l., overlaid on ordination Axes One and Two.

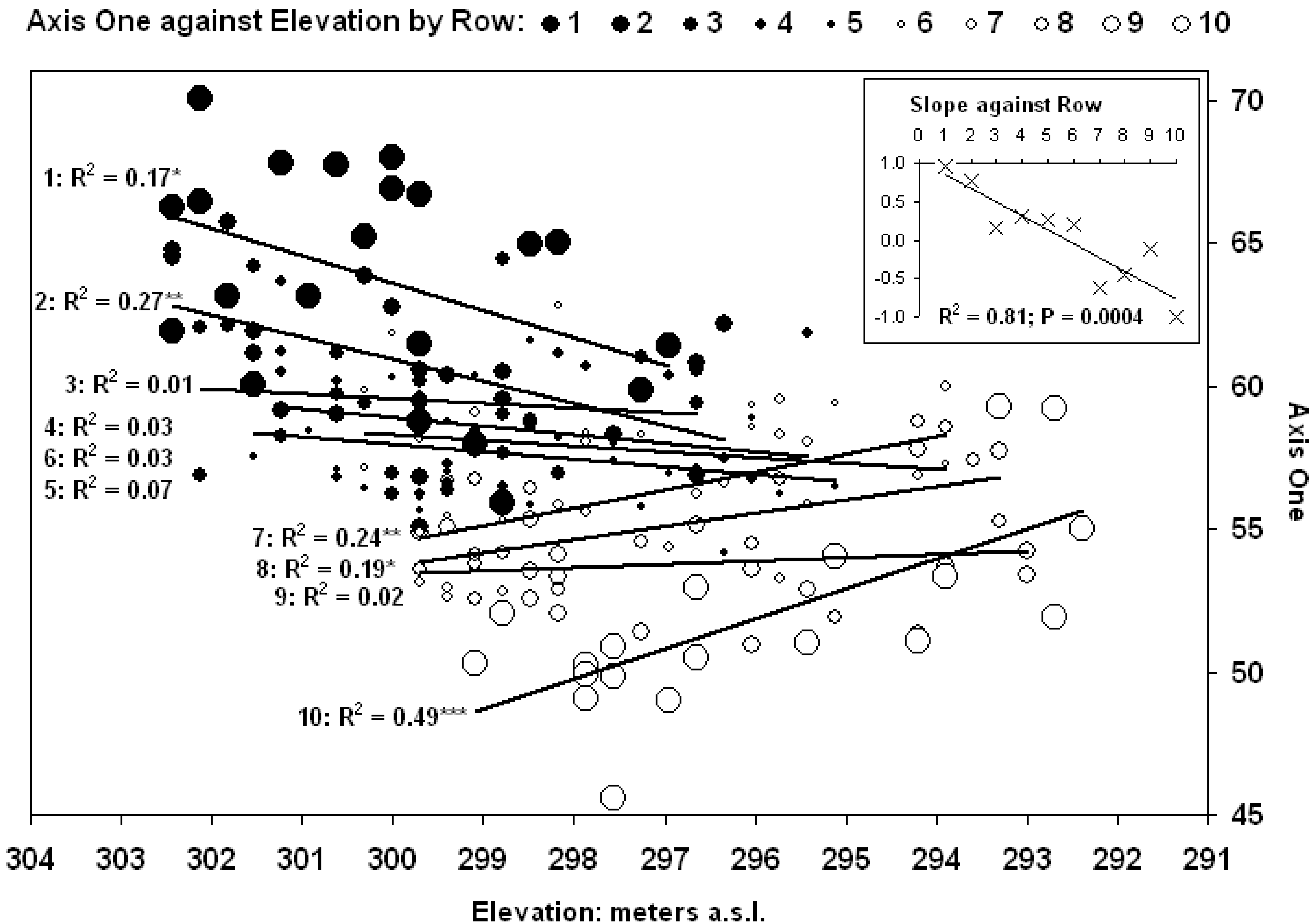

Figure 6. Elevation of plots in relation to Axis One score and row number. In overall correlation of Axis One score with elevation, R^2 = 0.19 (P = 0.007).

Figure 6: caption continued.

Linear trends are fitted to data from each of the ten rows, with significance indicated by asterisks after the proportional sums of squares (R^2), as follows:

* 0.1 > P > 0.05; ** 0.05 > P > 0.01; *** 0.01 > P.

The inset at upper right shows the slope for each of these linear trends in relation to row number (= distance from road in units of 17.5 m).

The two following trends are not shown.
(1) Best polynomial fit of Axis One versus elevation is cubic: $R^2 = 0.294$; $n = 199$; $P < 0.00001$ (compare with Figure 4a above); there is no significant trend with data below 297 m a.s.l.
(2) Linear regression of elevation against distance from road: $R^2 = 0.349$; $P < 0.00000001$.

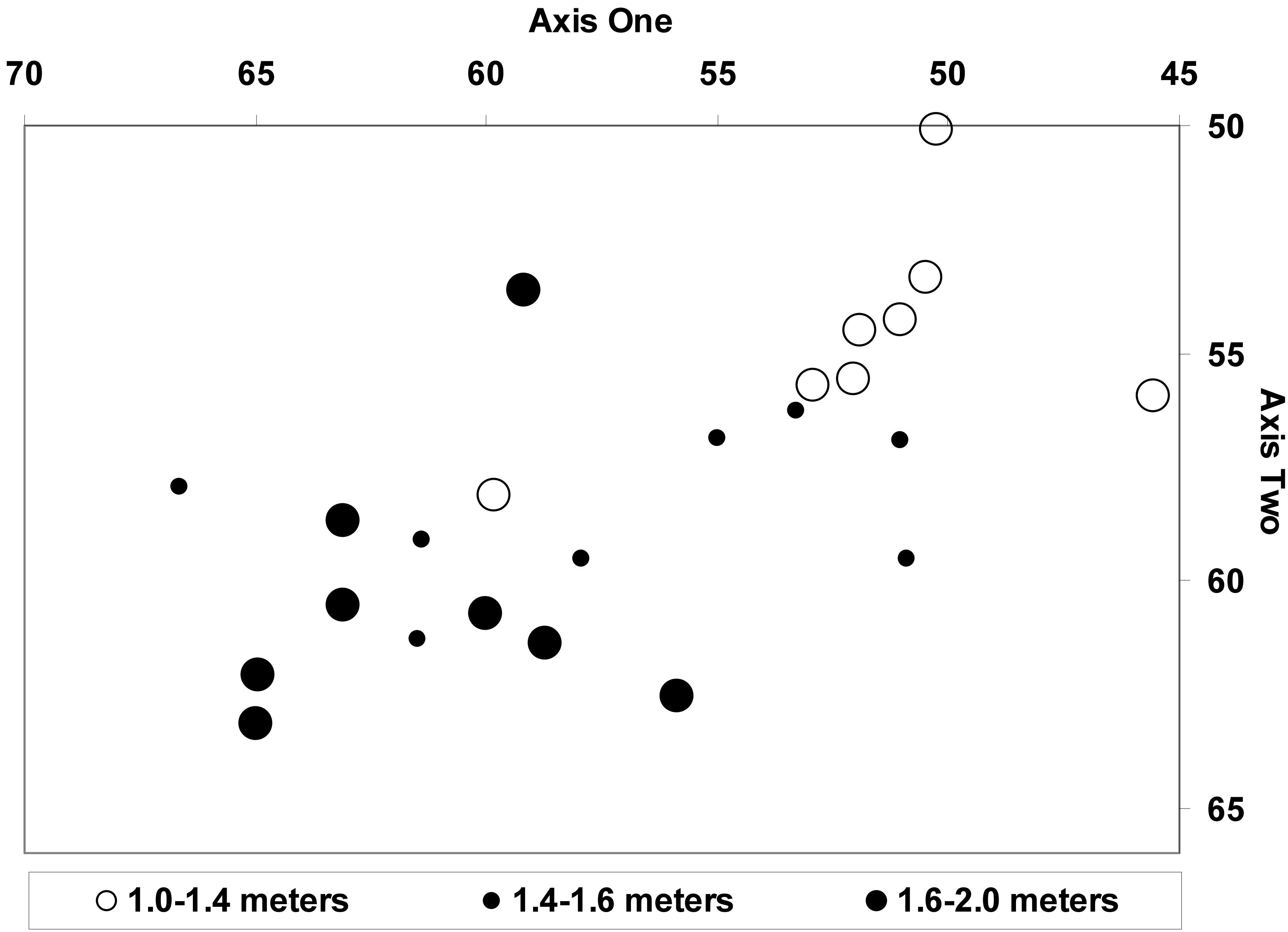

Figure 7a. Mean goldenrod height, overlaid on ordination; initial sample of 24 plots. See Table 1 for correlations.

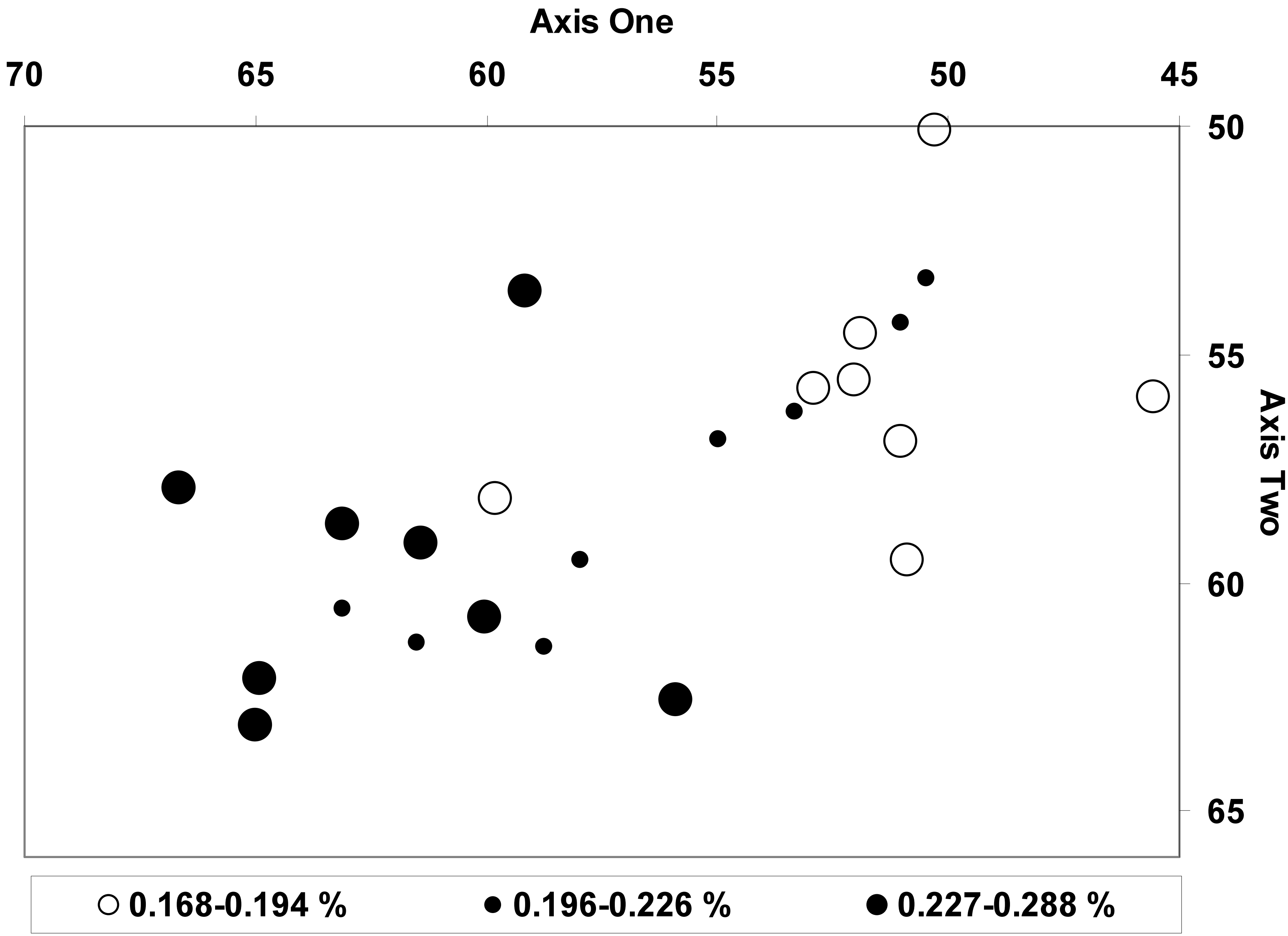

Figure 7b. Total nitrogen in soil, overlayed on ordination: initial sample of 24 plots. See Table 1 for correlations.

Table 1. Soil parameters: correlations with each other, ordination axes, and elevation.

Attributes	pHK	pHS	Ca	Zn	OM	P	TN	GH	Mg	K
pHK		0.98	0.98	0.96	0.95	0.91	0.90	0.71	0.43	0.31
pHS	0.98		0.95	0.93	0.92	0.87	0.85	0.68	0.39	0.29
Ca	0.98	0.95		0.96	0.94	0.91	0.85	0.61	0.40	0.41
Zn	0.96	0.93	0.96		0.98	0.92	0.88	0.64	0.39	0.40
OM	0.95	0.92	0.94	0.98		0.91	0.92	0.66	0.45	0.40
P	0.91	0.91	0.91	0.92	0.91		0.92	0.69	0.40	0.35
TN	0.90	0.85	0.85	0.88	0.92	0.92		0.82	0.56	0.19
GH	0.71	0.61	0.61	0.64	0.66	0.69	0.82		0.51	-0.15
Mg	0.43	0.40	0.40	0.39	0.45	0.40	0.56	0.51		-0.10
K	0.31	0.29	0.41	0.40	0.40	0.35	0.19	-0.15	-0.10	
Axis 1	0.68	0.66	0.58	0.63	0.64	0.63	0.79	0.77	0.70	-0.26
Axis 2	0.60	0.56	0.55	0.55	0.57	0.44	0.62	0.72	0.56	-0.15
R1 elev.	-0.47	-0.46	-0.51	-0.45	-0.45	-0.36	-0.17	0.36	0.48	-0.62*
R10 elev.	-0.50	-0.39	-0.48	-0.74**	-0.28	-0.69*	-0.58*	-0.79**	0.12	0.28
Means for Rows One and Ten plus their ratios; with T-tests * $P < 0.05$, ** $P < 0.01$										
R1 mean	5.48	6.84	2874	3.05	4.47	40.7	0.231	1.62	172	119
R10 mean	4.68	6.58	1660	1.35	3.60	23.6	0.195	1.38	133	130
R1 / R10	1.17**	1.04**	1.73**	2.3**	1.24**	1.73*	1.19**	1.17**	1.29**	0.91

Table 1. Explanation: attributes are as follows; close ranking reflect close correlation. Standard correlation coefficients are presented among the following attributes; significance at $P < 0.0001$ is indicated by deep shading; significance at $P < 0.05$ is indicated by light shading.

pHK: soil pH in 1 Molar KCl.
Ca: soil calcium in mg per kg
OM: soil organic matter percentage
TN: total soil nitrogen percentage
Mg: soil magnesium in mg per kg
pHS: soil pH in Sikora II buffer
Zn: soil zinc in mg per kg
P: soil phosphorous in mg per kg
GH: mean goldenrod height in meters
K: soil potassium in mg per kg

Axis 1 and Axis 2: correlations are shown with each of above attributes; n = 24 in each case.

R1 elevation: correlations are shown with elevation in the front row (row one); n = 12.
R10 elevation: correlations are shown with elevation in the back row (row ten); n = 12.
Significance of T-tests is indicated by * for $P < 0.05$ and ** for $P < 0.01$.

R1 mean: mean value is shown for each attribute in the front row (row one); n = 12.
R10 mean: mean value is shown for each attribute in the back row (row ten); n = 12.
R1 / R10: ratio of mean in front to mean in back row is shown; significance of T-tests is indicated by * for $P < 0.05$ and ** for $P < 0.01$.

Note that uncontrolled variables here are in the adjacent tree-lines, which contain nitrogen-fixing black locust in places. However, TN was not higher in plots adjacent to black locust thickets, which tended to occur instead adjacent to plots with relatively low TN: 0.218 in R1, n = 5; 0.173 in R10, n = 2. This trend is not significant but will deserve deeper exploration.

RESULTS

Ordination of Plots: Apparent Relationship to Soil

The first three axes of the ordination (detrended correspondence analysis) have eigenvalues (before detrending) of 0.18, 0.12 and 0.11. The lengths of these gradients, in terms of weighted mean standard deviations of species' scores within samples (Hill & Gauch 1980), are 2.1, 1.7 and 1.8. Only the first two axes are interpreted here. The third axis has little or no segregation of common species. It is correlated with northern versus southern position in the field (N12 to S9 in Figure 1; $r = 0.36$, $n = 199$, $P < 0.0001$), but less strongly than the second axis ($r = 0.41$).

Unexpectedly, the first axis ("Axis One") is related to distance from the road (Figures 2a and 3a). Plot scores decline linearly away from the road for the whole 160 m distance from row one to row ten (Figure 4a). Elevations also generally decline away from the road, but covariance of the first axis with elevation is much weaker than its covariance with distance from the road: $R^2 = 0.19$ (Figure 6) versus 0.58 (Figure 4a). Moreover, the relationship to elevation within rows changes gradually from negative closer to the road (rows 1-6) to positive farther from the road (rows 6-10); see Figure 6 for details.

These trends along Axis One suggests a significant effect of pollution from the road, with local reversal at lower elevation away from road, perhaps due to drainage of nutrients into gullies. The initial surveys of soils in rows one and ten support this interpretation. Most of the soil attributes are correlated with each other (Table 1), especially pH, calcium (Ca), zinc (Zn), organic matter, phosphorous (P) and nitrogen (N). All of the attributes are positively correlated with Axis One score, except potassium (K). However, the strongest correlations with Axis One occur in total nitrogen ($r = 0.79$) and goldenrod height ($r = 0.77$). Goldenrod height appears to be a good indicator of overall fertility; its strongest correlation is with total nitrogen (0.82). All attributes except potassium are significantly higher in row one than in row ten, with the largest ratios in zinc, calcium and phosphorous. As with Axis One and elevation, the soil attributes tend to have more negative relationships to elevation in row ten than in row one (especially goldenrod height). But only goldenrod height and magnesium have positive (albeit non-significant) relationships to elevation within row one.

Interpretation of these edaphic trends in terms of pollution from the road is supported by the finding of gravel, asphalt and glass fragments in soil samples from row one, but none in samples from row ten. These obvious fragments, about 0.5-3 cm in size, constituted only about 0.5 to 5% of each initial soil sample. This debris was removed before analysis, but it is likely that additional smaller or more degraded fragments were overlooked. The debris was found only in plots 6 to 9 along row one (Figure 1), which matched the lowest section along this row at 297-298 m a..s.l. versus 299-302 m. It would appear that this debris had been washed down into the field from the roadside.

Ordination of Plots: Apparent Relationship to Herbivory

The second axis ("Axis Two") is also correlated with most of the soil attributes, but more weakly than is the first axis (Table 1). The edaphic relationship generally extends from the upper right sector to the lower left sector of the ordination, as presented in Figures 3, 5 and 7. Axis scores are reversed within these diagrams to ease comparison with results of Part I in this study (Campbell 2015), and with conceptual gradients in the overall "Herbivore Hypothesis" (Campbell 2012). Indications of herbivory do appear related to Axis Two, but any simple "herbivory gradient" is initially obscured by correlation with the predominant edaphic gradient.

Analysis of spatial pattern in this field during 2007 indicated a concentration of browsing effects by deer along a diagonal pathway, with three estimated zones: "central pathway"; "transition"; and "little influence" (Campbell 2015). There is slightly more overall segregation of these zones along Axis Two than along Axis One (Figure 8). The "central" and "transitional" zones are strongly concentrated in the upper half of the ordination, with lower Axis Two scores. However, the zone of "little influence" is widespread over most of the ordination. Recorded signs of browsing by deer in plots during 2007 (excepting data from the planted blue ash) are virtually absent from the lower left sector of the ordination (Figure 9a). There is no monotonic trend along either axis (Figure 9a), but these signs are strongly concentrated in a central zone of the ordination, with a distinct peak in frequency along Axis One (Figure 9b).

Signs of recent browsing on the planted blue ash during 2007 (with random-systematic planting along 12 of the 20 row sections) increased from lower left to upper right sectors of the ordination, generally away from the road (Figure 10a, b). In contrast, resprouting of leading stems, suggesting earlier damage during 2004-2006, was most frequent in the lower left sector. In Part I of this study (Campbell 2015), zones of high versus low mortality in the planted blue ash were estimated. Plots in the high mortality zone are concentrated in a central zone along Axis One, and the proportion of these plots increases into upper sectors of the ordination, with lower Axis Two scores (Figure 11a, b).

The more recent survey of deer trails in January 2016 reveals intense pattern over the field (Figure 12a). Major trails are more frequent in upper sectors of the ordination, with lower Axis Two scores, and there is no significant trend along Axis One (Figure 13a). With the addition of minor trails, perhaps made by rabbits or other other animals rather than deer, trends are similar but slightly less significant (Figures 12b and 13b).

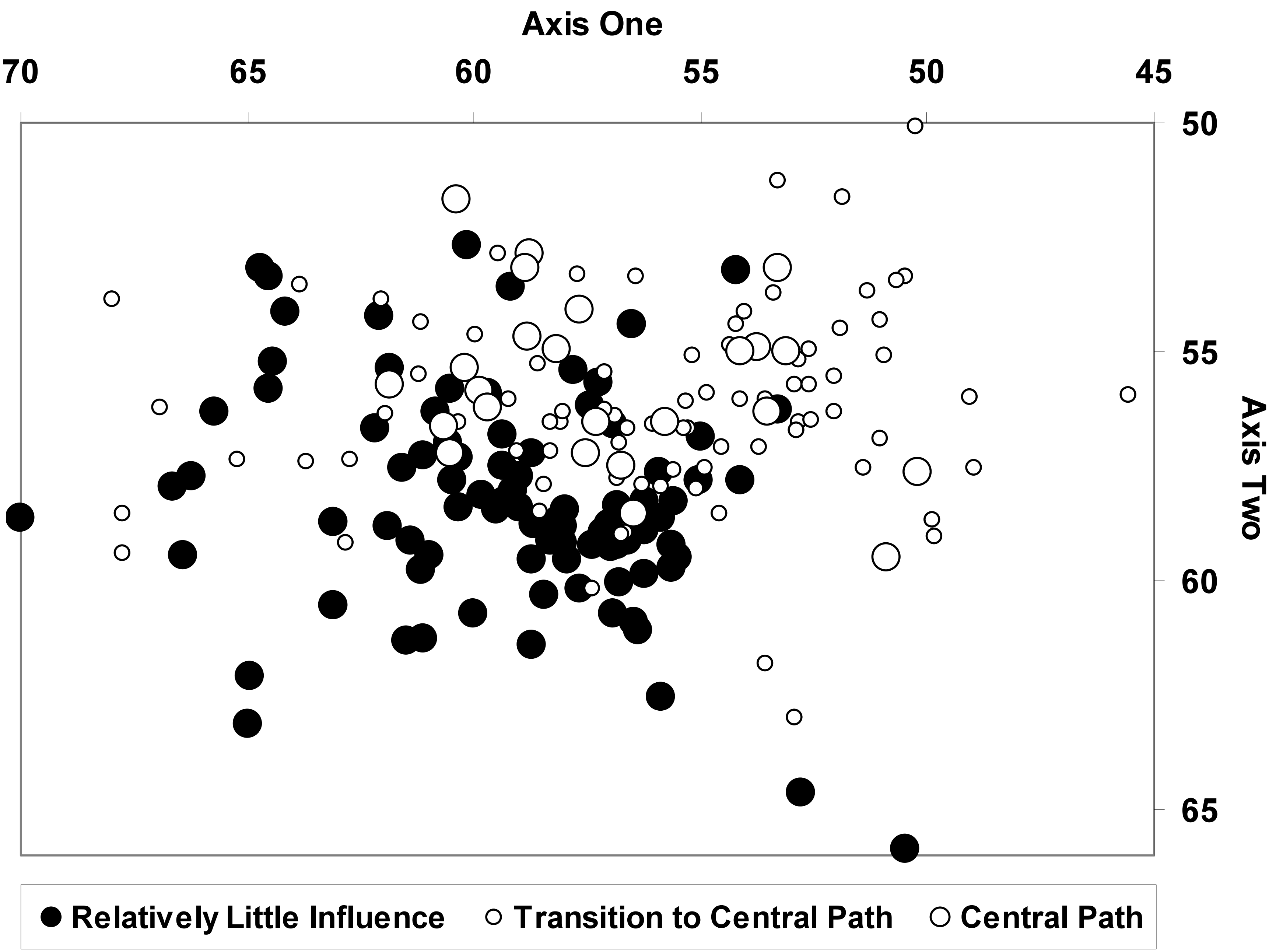

Figure 8. Estimated zones of deer influence overlaid on ordination Axes One and Two.

Figure 8: caption continued.
See Part I for details of how these zones were estimated, especially trends in woody species. Using ANOVA and Tukey's range test, these three zones have some significant differences in mean scores along Axes One and Two ($\pm$ standard errors as follows). There is a general trend from lower left to upper right, but "Central Path" is concentrated at central Axis One scores.
(1) For Axis One: $F = 14.07$; $P < 0.0001$. Mean for "Central Path" = 57.00 $\pm$0.66 (n = 24); for "Transition" = 56.06 $\pm$0.50 (n = 84); for "Zone of Little Influence" = 59.21 $\pm$0.36 (n = 91). Central versus Transition is non-significant; Central versus Little Influence has $P < 0.05$; Transition versus Little Influence has $P < 0.01$.
(2) For Axis Two: $F = 23.9$; $P < 0.0001$. Mean for Central = 55.69 $\pm$0.38; for Transition = 56.22 $\pm$0.23; for Little Influence = 58.27 $\pm$0.25. Central versus Transition is non-significant; Central versus Little Influence has $P < 0.01$; Transition versus Little Influence has $P < 0.01$.

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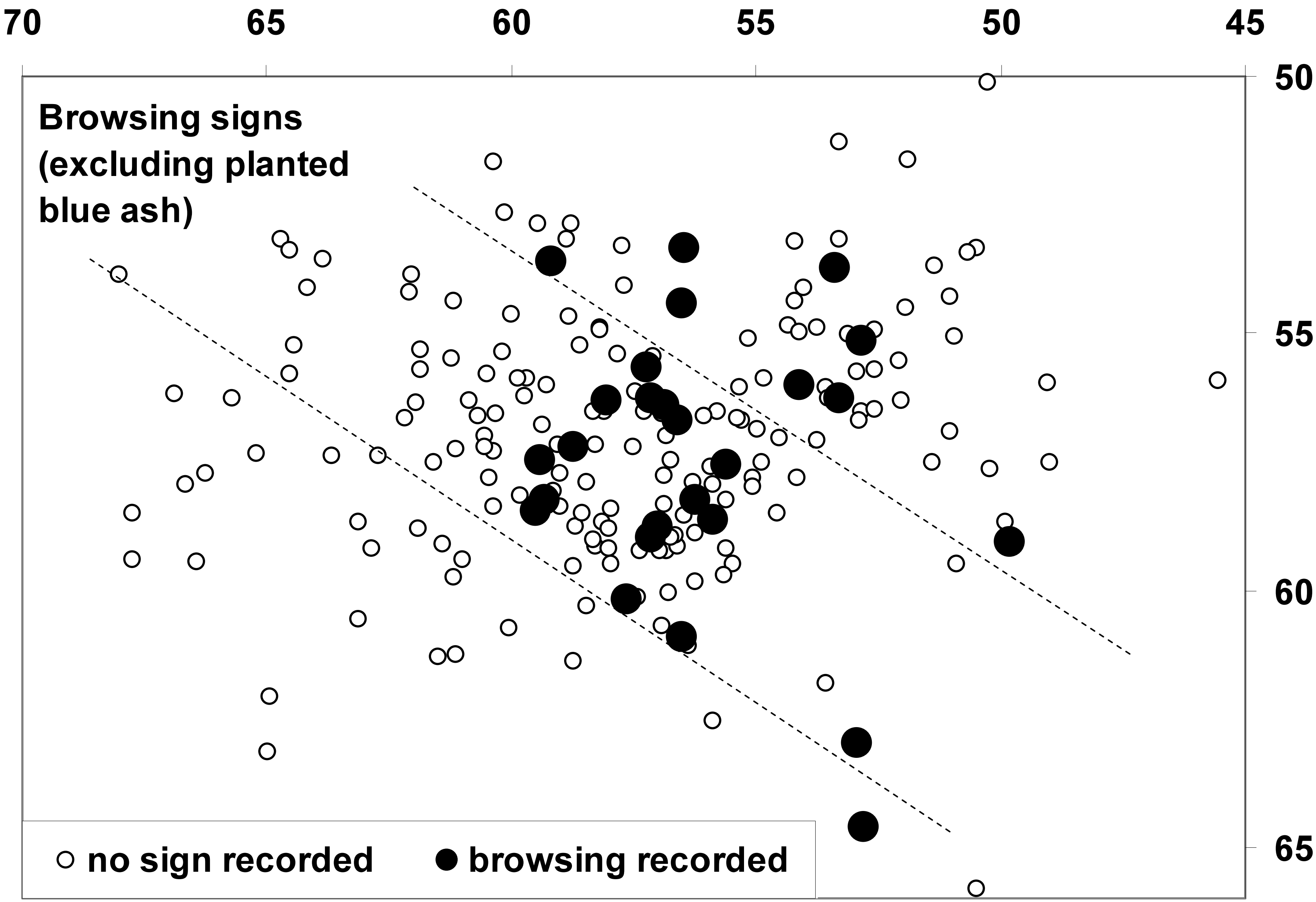
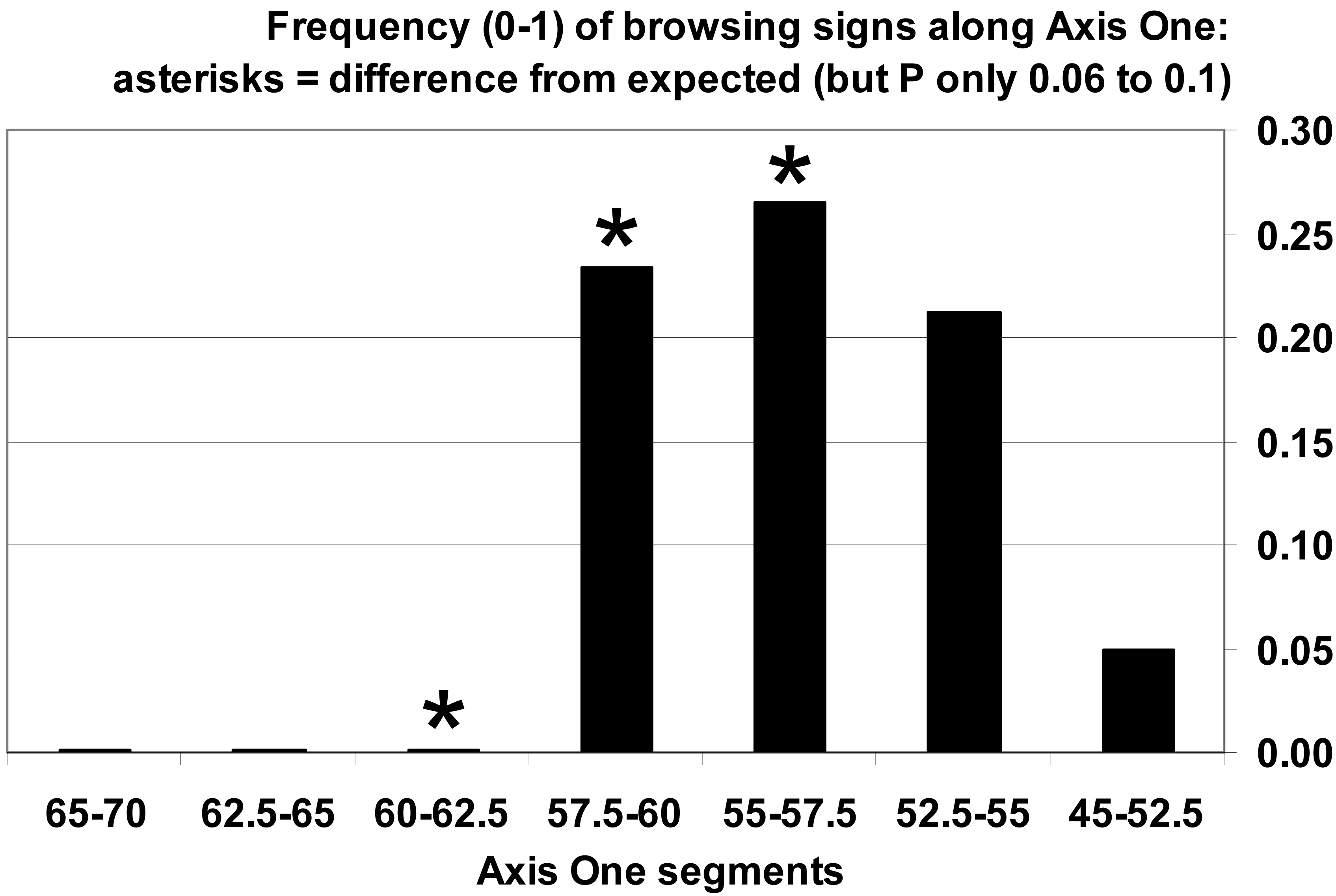
Figure 9 [next two pages].
A. Recorded signs of browsing by deer in plots, overlaid on ordination.
Signs of damage to the planted blue ash were excluded here, but see Figure 13.*
Dashed lines are fitted visually to indicate the consistent tendency for signs of herbivory to be concentrated in the upper right sector; see also Figures 13 and 14. Frequencies in these three zones here are 1/28 (4%), 17/97 (18%) and 8/51 (16%).
B. Trend in frequency of these browsing signs along Axis One.
The concentration at intermediate positions along the axis has only marginal significance.

* See Part I (p. 65): "About 59 direct signs of recent damage by herbivores were noted in Sep 2007, including 26 on the planted blue ash. Numbers were not sufficient for statistical comparison of plant species, but the most frequently noted species apart from the blue ash were white ash (9 cases) and bush honeysuckle (6 cases); other woody species included box-elder, sugar maple, roughleaf dogwood, red mulberry, Siberian crabapple and elderberry—none in the 'browsing-associated' group except the latter two (Table 4). Curiously, the few forbs with noted damage were among the 'browsing-associated' group: *Desmodium perplexum* and *Physalis longifolia*."


70
65
60
55
50
45
50
55
60
65
Browsing signs (excluding planted blue ash)
no sign recorded
browsing recorded

A

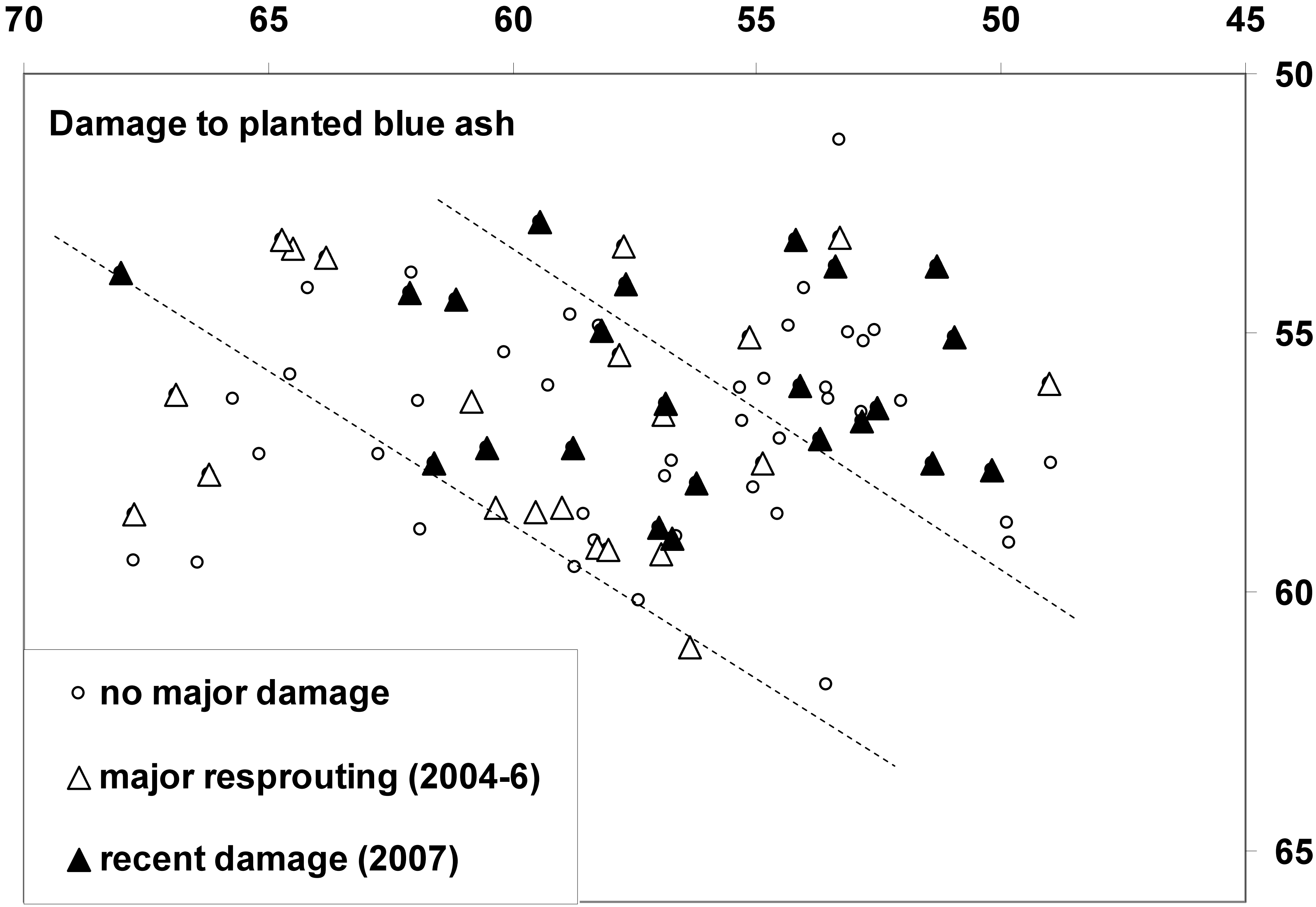

Frequency (0-1) of browsing signs along Axis One:
asterisks = difference from expected (but P only 0.06 to 0.1)
0.30
0.25
0.20
0.15
0.10
0.05
0.00
*
*
*
65-70
62.5-65
60-62.5
57.5-60
55-57.5
52.5-55
45-52.5
Axis One segments

B

Figure 10 [next two pages].

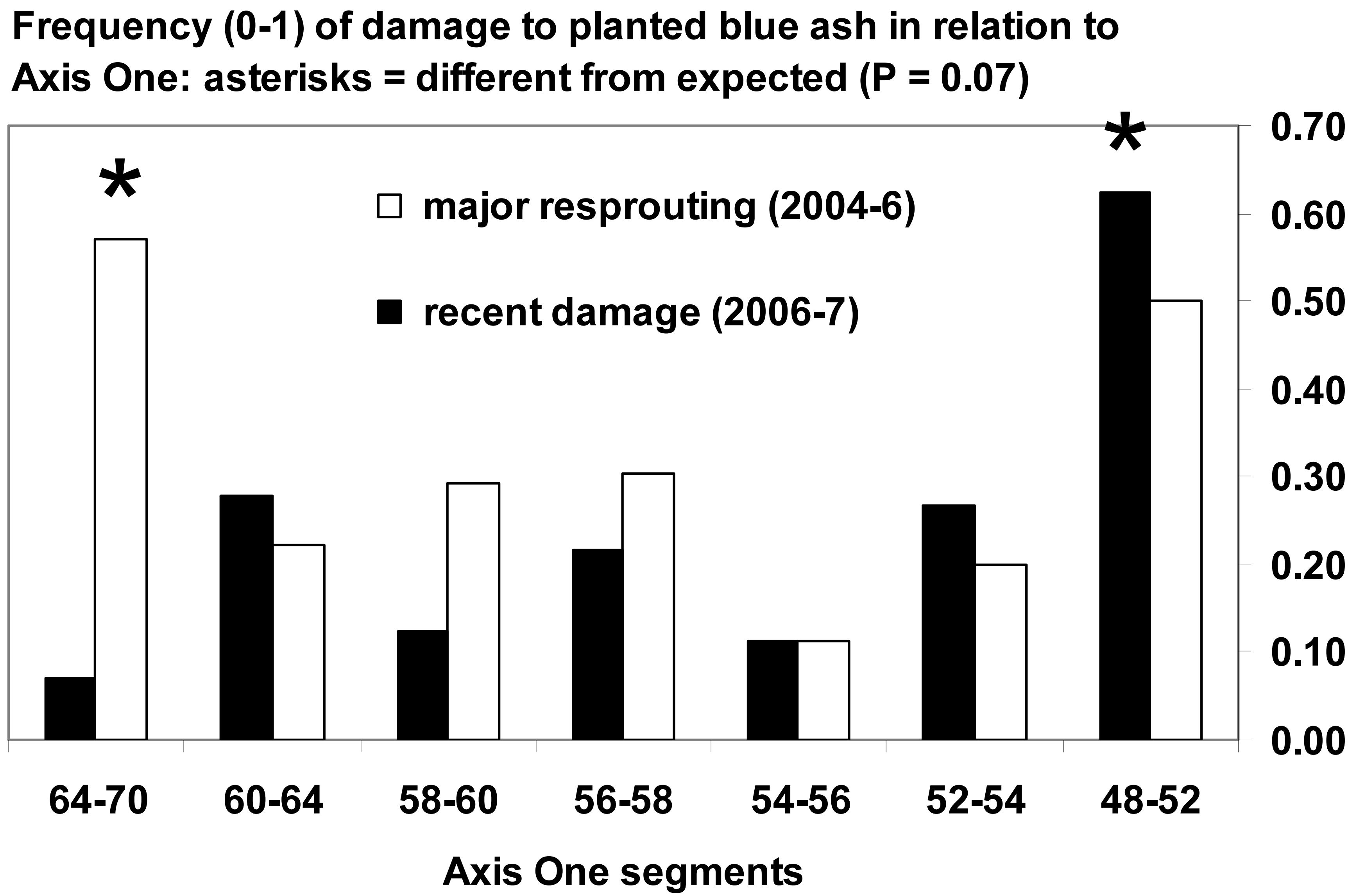
A. Recorded signs of damage to planted blue ash in plots, overlaid on ordination. Note that only about 60% of the 199 plots had planted blue ash; see Part I for details. Plants with "major resprouting" had replaced leaders during 2004-6. Those with "recent damage" appeared to have been browsed by deer or damaged by smaller mammals during 2007. Dashed lines are fitted visually to indicate the consistent tendency for signs of herbivory to be concentrated in the upper right sector; see also Figures 13 and 14. Frequencies in these three zones here are 2/15 (13%), 9/37 (24%) and 12/31 (39%).

B. Trend in frequency of "major resprouting" and "recent damage" along Axis One. The concentrations at extreme positions along the axis have only marginal significance.



70
65
60
55
50
45
50
55
60
65
Damage to planted blue ash
no major damage
major resprouting (2004-6)
recent damage (2007)

A

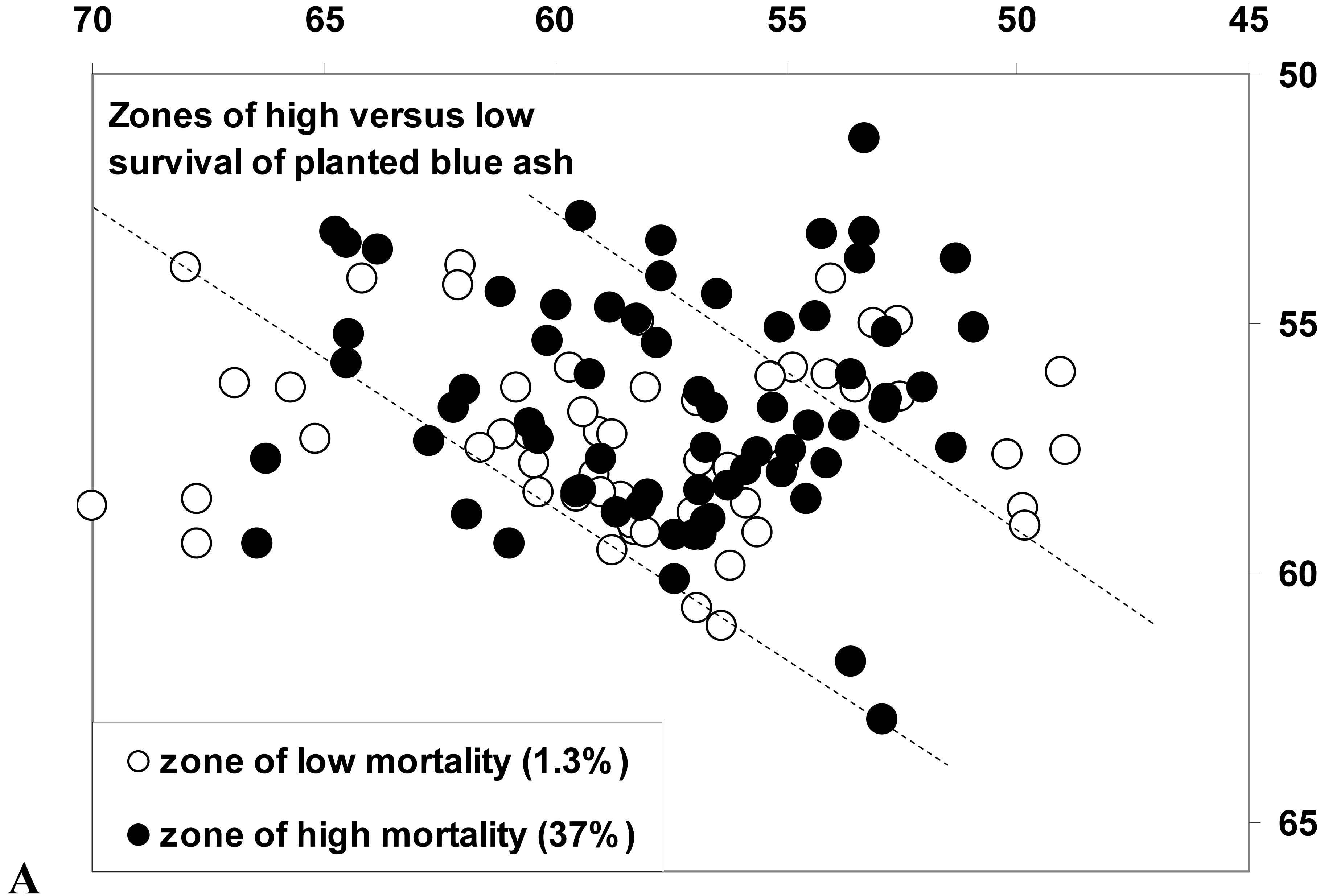

Frequency (0-1) of damage to planted blue ash in relation to
Axis One: asterisks = different from expected (P = 0.07)
*
*
major resprouting (2004-6)
recent damage (2006-7)
0.70
0.60
0.50
0.40
0.30
0.20
0.10
0.00
64-70
60-64
58-60
56-58
54-56
52-54
48-52
Axis One segments

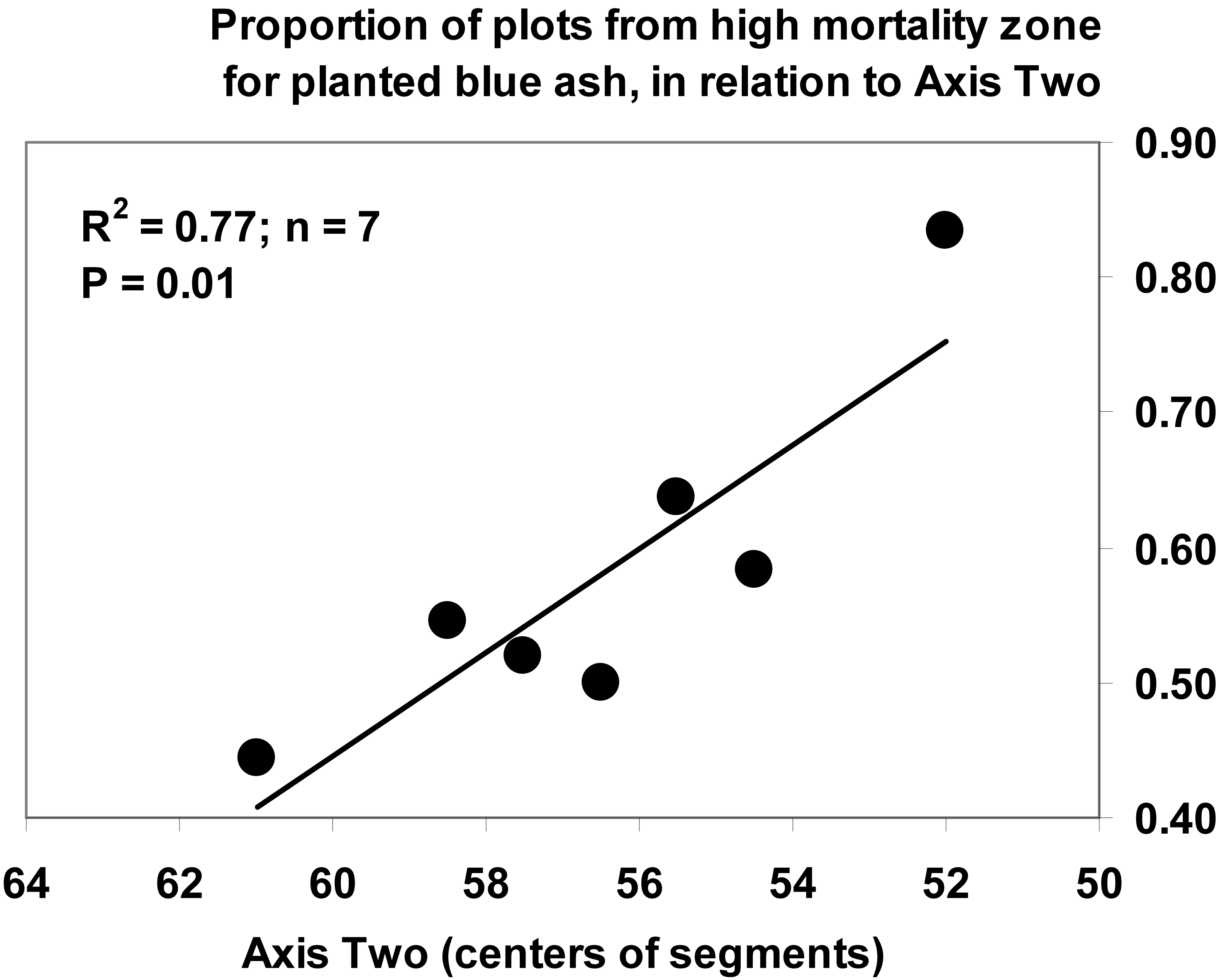
B

Figure 11 [next two pages].

A. Zone of high versus low mortality among planted blue ash, overlaid on ordination. Note that only about 60% of the 199 plots had planted blue ash; see Part I (Figure 10) for details. Despite the apparent zones of concentrated mortality (and a tendency for higher growth rates in zones of low mortality), there was no significant difference in observed damage or resprouting between the zones. Dashed lines are fitted visually to indicate the consistent tendency for signs of herbivory to be concentrated in the upper right sector; see also Figures 13 and 14. Frequencies in these three zones here are 5/15 (50%), 39/65 (60%) and 18/31 (58%).

B. Trend in frequency of high mortality zones along Axis Two.
The concentration at upper positions along the axis has only marginal significance.
With T-test comparing Axis Two scores, $P = 0.09$.


70
65
60
55
50
45
50
55
60
65
Zones of high versus low
survival of planted blue ash
zone of low mortality (1.3%)
zone of high mortality (37%)
A



Proportion of plots from high mortality zone
for planted blue ash, in relation to Axis Two
R2 = 0.77; n = 7
P = 0.01
0.90
0.80
0.70
0.60
0.50
0.40
64
62
60
58
56
54
52
50
Axis Two (centers of segments)

B

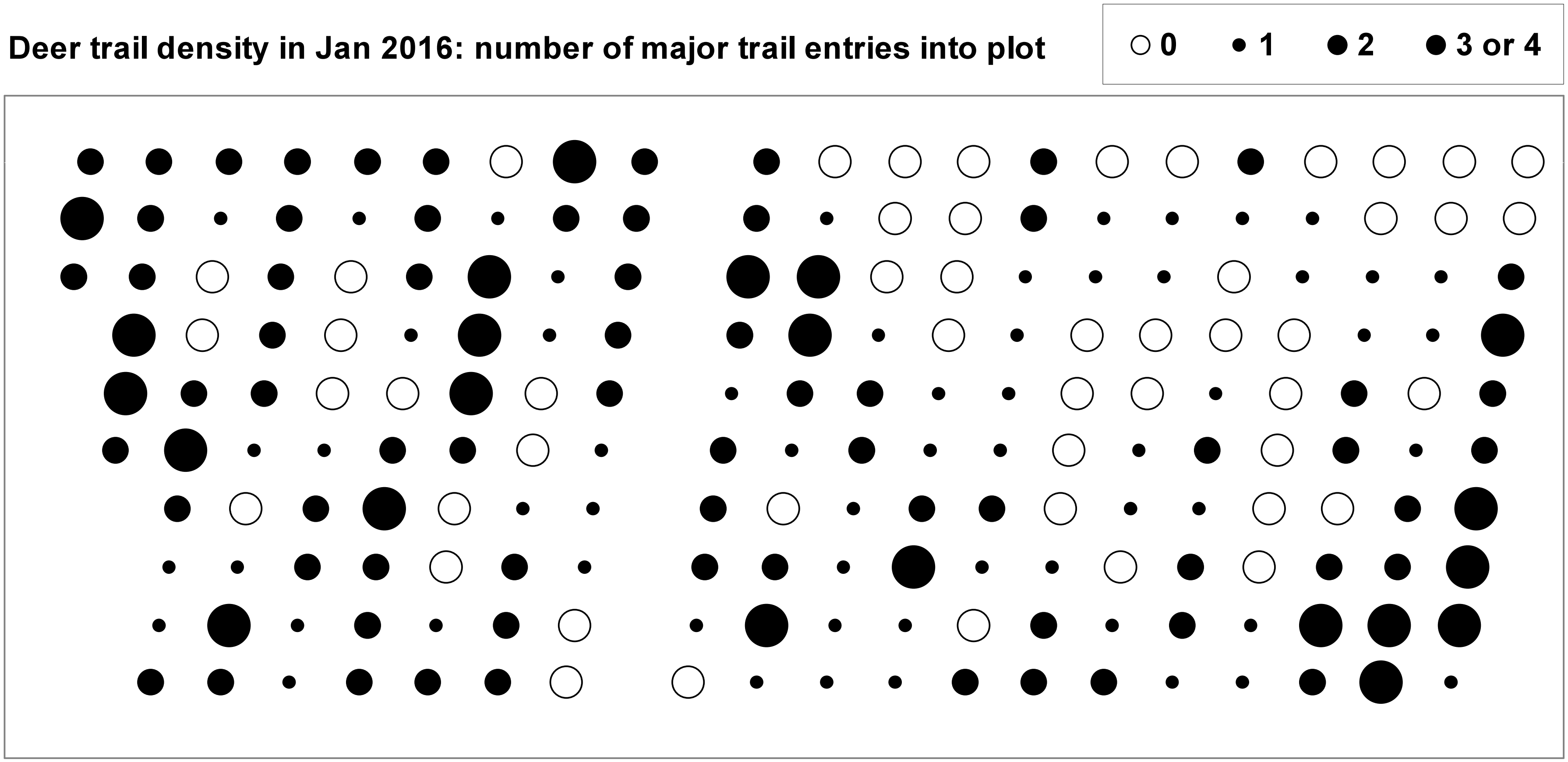


Figure 12a. Density of major deer trails estimated for each plot in Jan 2016.

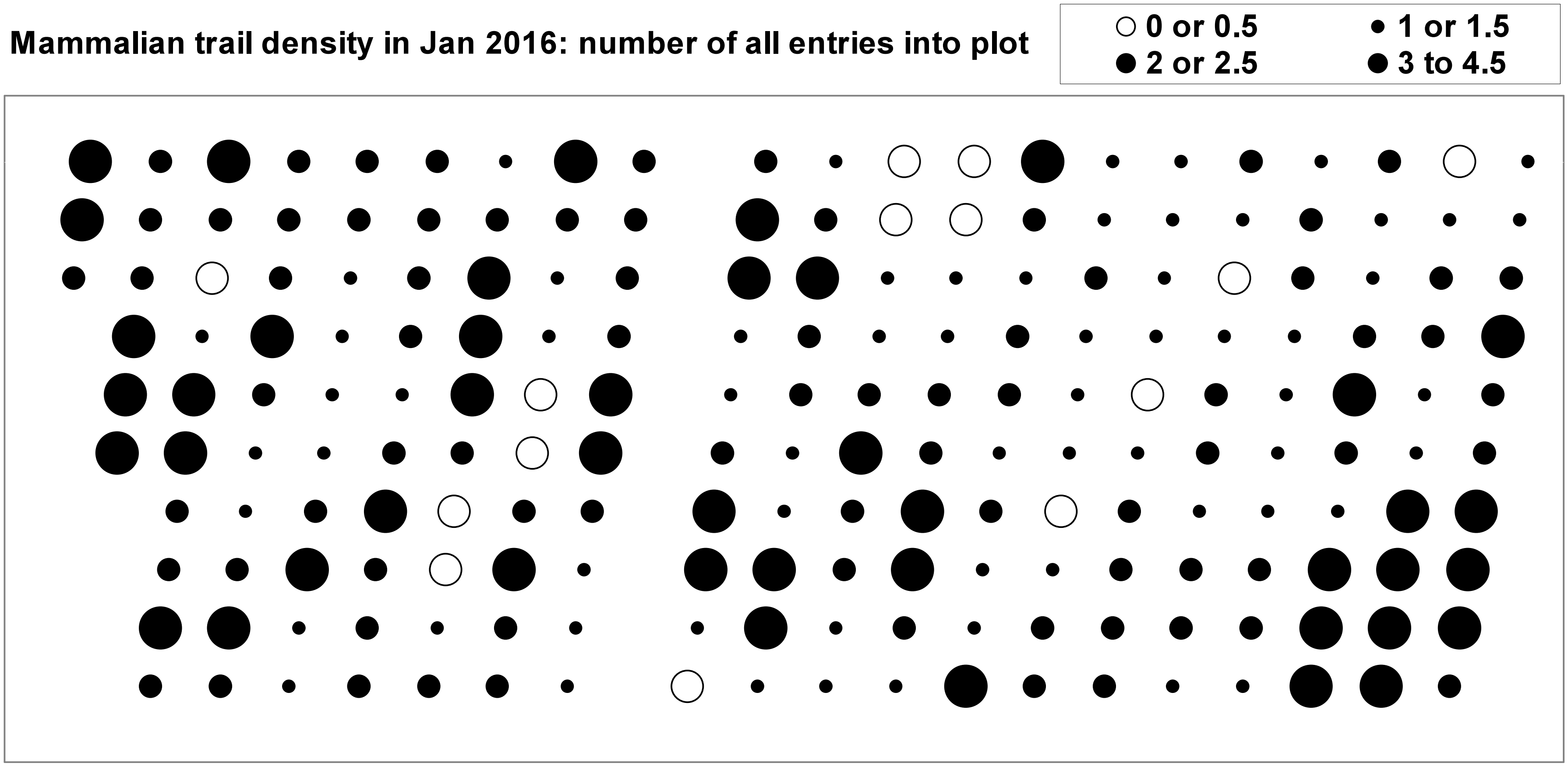


Figure 12b. Density of all mammalian trails estimated for each plot in Jan 2016. Minor trails were added to major trails of Figure 12a, but counting each as 0.5 rather than 1.

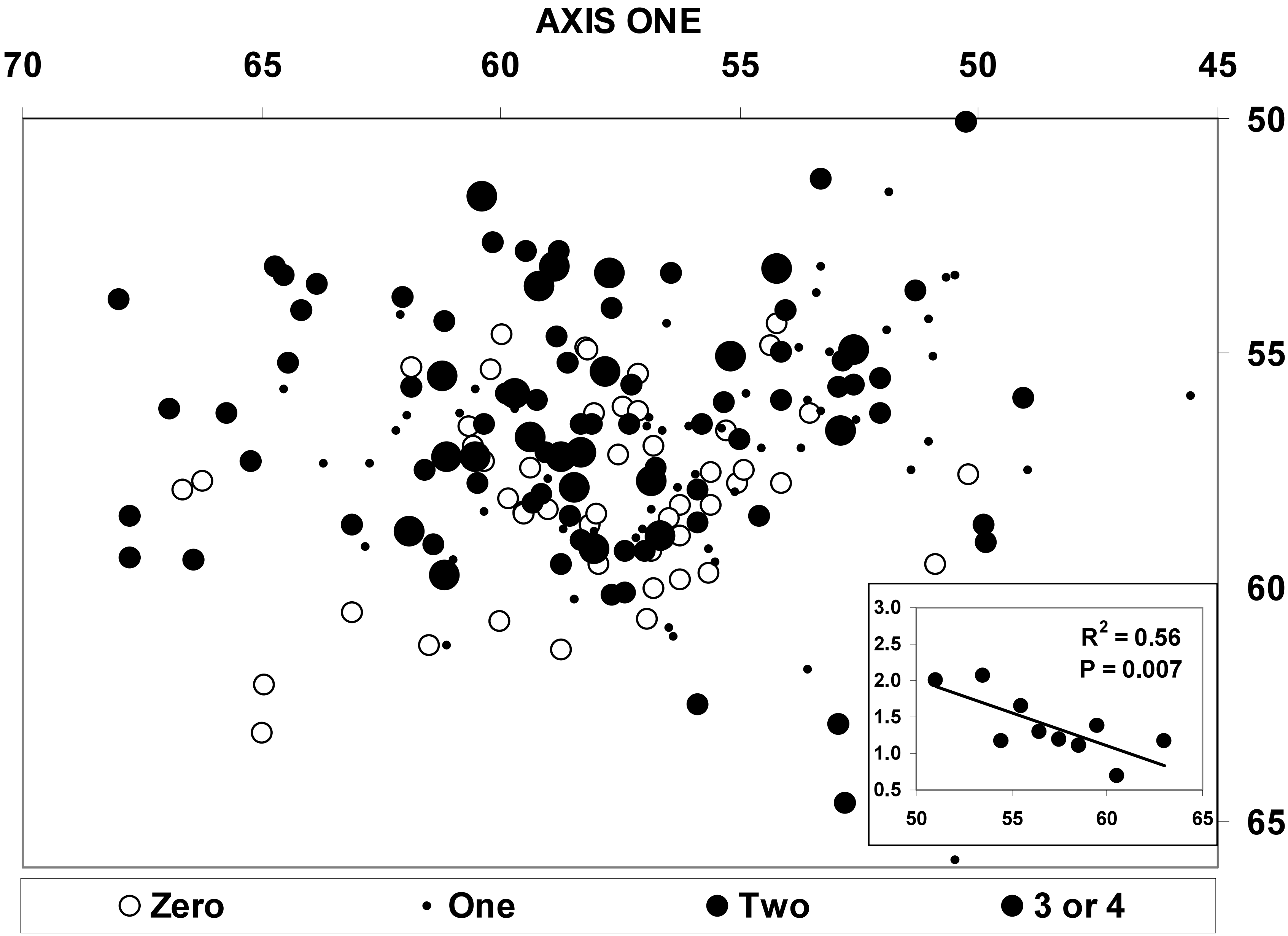


Figure 13a. Density of major deer trails in relation to ordination axes. Inset shows linear trend along Axis Two (Spearman's rho = –0.24; for Axis One, rho = 0.09).

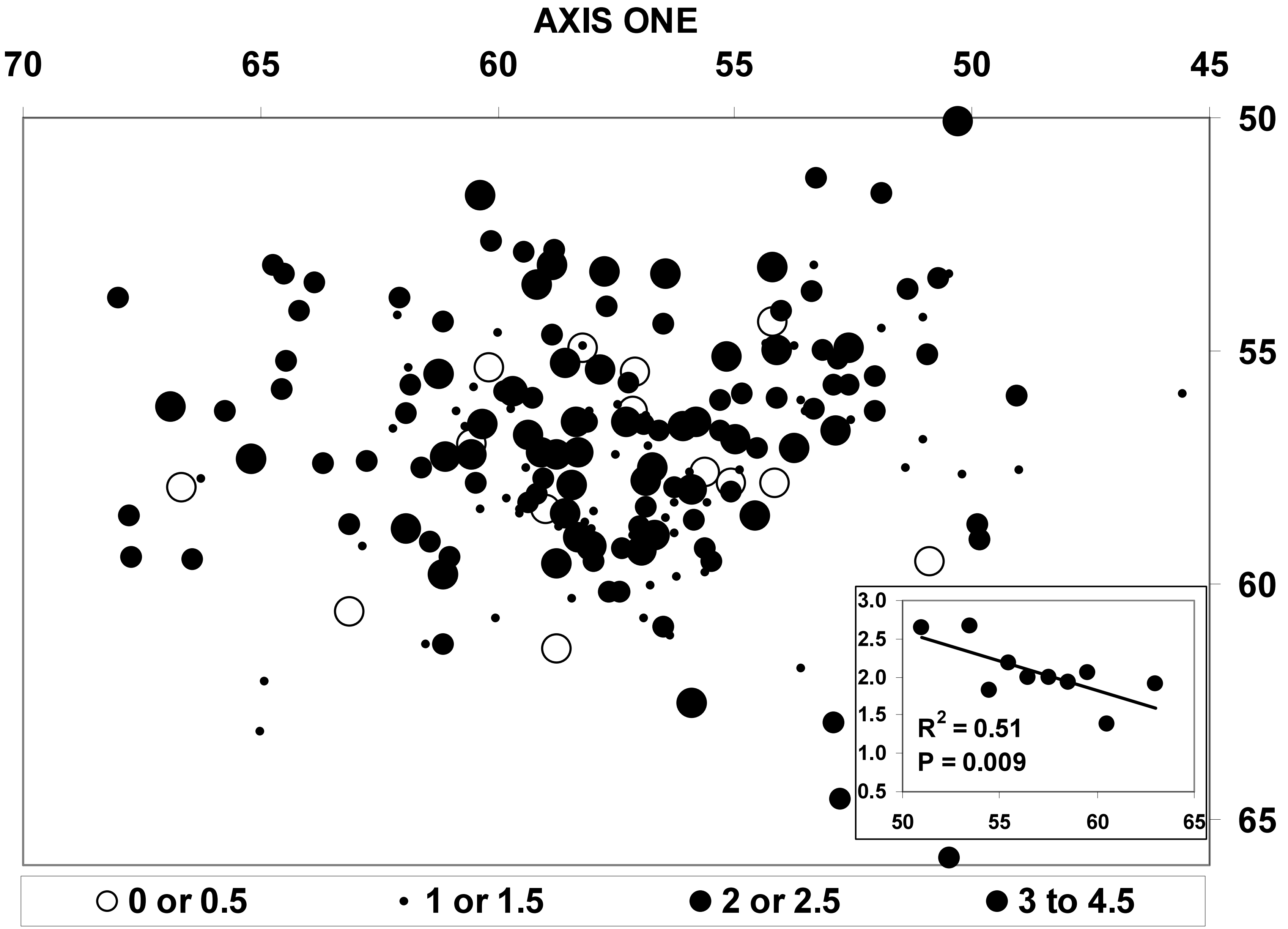


Figure 13b. Density of all mammalian trails in relation to ordination axes. Inset shows linear trend along Axis Two (Spearman's rho = –0.20; for Axis One, rho = 0.02).

Ordination of Species: Relationships with Functional Attributes

When overlaid on the ordination, the major species in each plot reveal a central zone, from upper left to lower right, dominated by two grasses—the alien fescue and the possibly native bluegrass—plus goldenrod (Figure 14). Fescue is more strongly concentrated in a central zone of the ordination than bluegrass or goldenrod (Figure 15). Towards the upper right, there is local dominance of blackberry, trumpet-creeper or asters (*Symphyotrichum pilosum* and locally *S. ontarione*). Towards the lower left, there is local dominance of poison hemlock (*Conium maculatum*) and crown vetch (*Securigera varia*), both aliens. Among all species in the ordination, there are curious concentrations of aliens towards extremes of Axis One (Figures 16a and 17a).

The scores of individual species in the ordination (proportional to weighted means of plots where they occur) can be used to explore patterns in functional attributes. Table 2 presents some attributes for each species based on the literature plus general knowledge of the author. The hydrological associations of species have no clear relationship to the ordination, although there is a weak tendency for species typical of drier sites to increase at low Axis One positions, with greater distance from the road and lower elevation on average (Figure 16b). In contrast , the "basiphilpous index" and "nitrophilous index" have much stronger patterns, both increasing towards to the left, with greater Axis One scores (Figures 16c, 16d, 17b and 17c). Also, species with relatively "nitrogen-rich chemistry" are concentrated at left-central positions, except for most Fabaceae—which fix nitrogen and concentrate at right-central positions here (Figures 16e). Curiously, the "browsing-associated" group that was defined in Part I tends to have lower positions on the ordination (Figure 16f), with marginal significance in the t-test ($P = 0.047$).

Some morphological trends are also evident. The nine species with thorns or spines are concentrated in a curious band from upper left to lower right (Figure 16f). Species known to be toxic or strongly unpalatable to most mammals are concentrated on the lower left side of this “thorny zone”: with about 15 of the 38 species here versus 5 of the 60 species on the upper right side of the “thorny zone” (Figure 16f). Maximum height of each species also increases from upper right to lower left sector, with vines curiously concentrated in a central band to the right of the “thorny zone” (Figures 16g and 17e). Life-form in general displays little clear pattern, other than this concentration of vines. However, there is an increase the proportion of annuals and biennials (versus perennials) from upper right to lower left (Figure 16h). Also, low running perennials (with potential for rapid clonal spread) are concentrated at intermediate positions on Axis Two (scores all 35 to 70).

When overlaid on the ordination, patterns in woody species are similar to the gradient that was already displayed in Part I of this study (Campbell 2015), comparing species concentrated in the “central” zone with most influence from deer, versus those concentrated in the zone with “little influence”. Figure 18 arranges these species from those concentrated here at upper positions in the ordination (with low scores on Axis Two), to those concentrated at lower positions. Browsing-associates appear to be red cedar (especially), elderberry, nut-trees (walnut, hickories, oaks) and most of the rosalean group (apple, hawthorn, pear, autumn olive, honey locust; also coffeetree). Intermediate species appear to be multiflora rose, black cherry, American elm and hackberry. Browsing-avoiders appear to be white ash (especially), bush-honeysuckle, mulberries and black locust (although vigorous clonal sprouting of the latter makes it difficult to compare). There is much less segregation of these woody species along Axis One; red cedar is the only common species largely restricted to the right sector of the ordination.

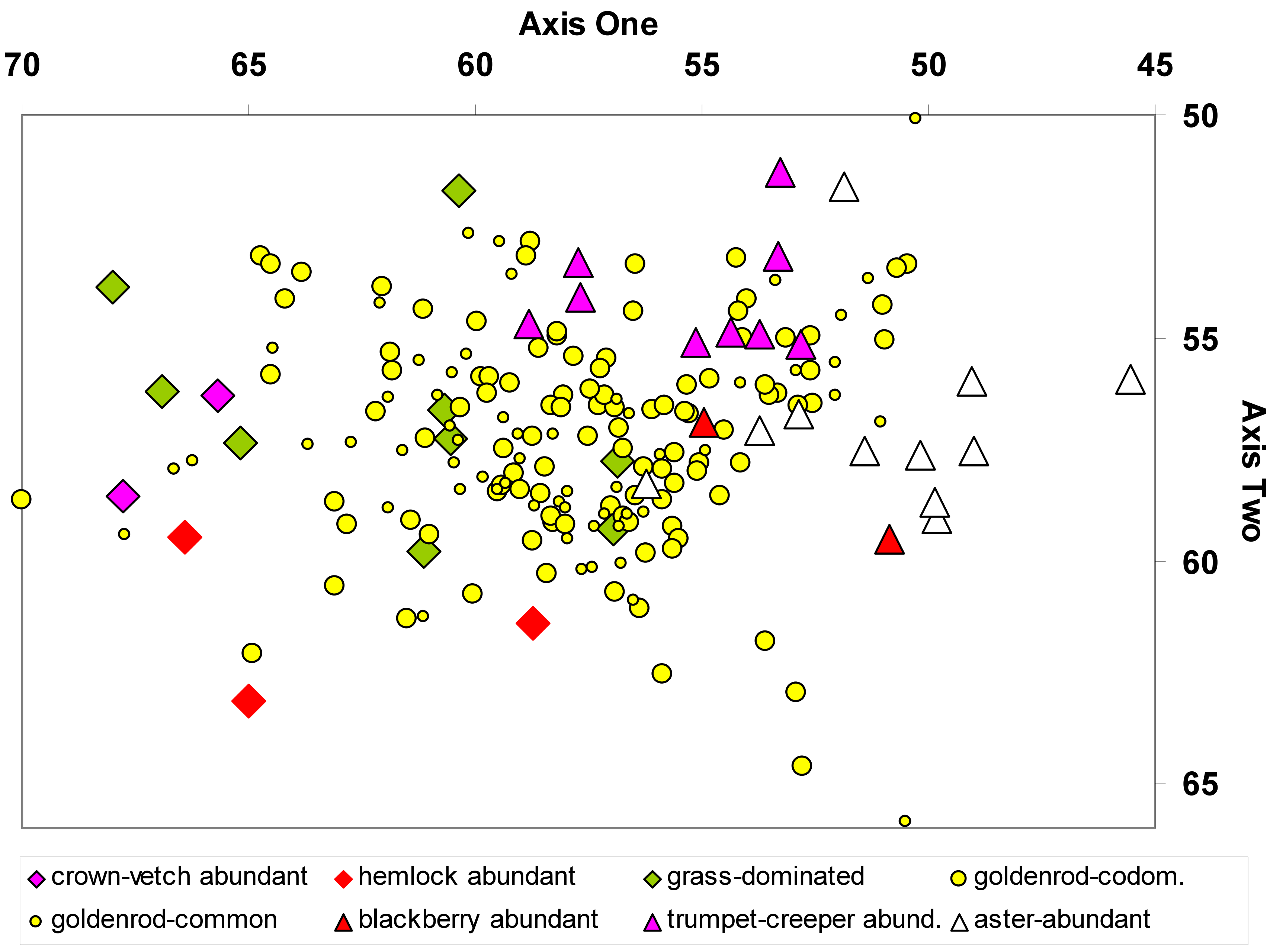


Figure 14. Dominant species in each plot overlaid on ordination (see Part I for details).

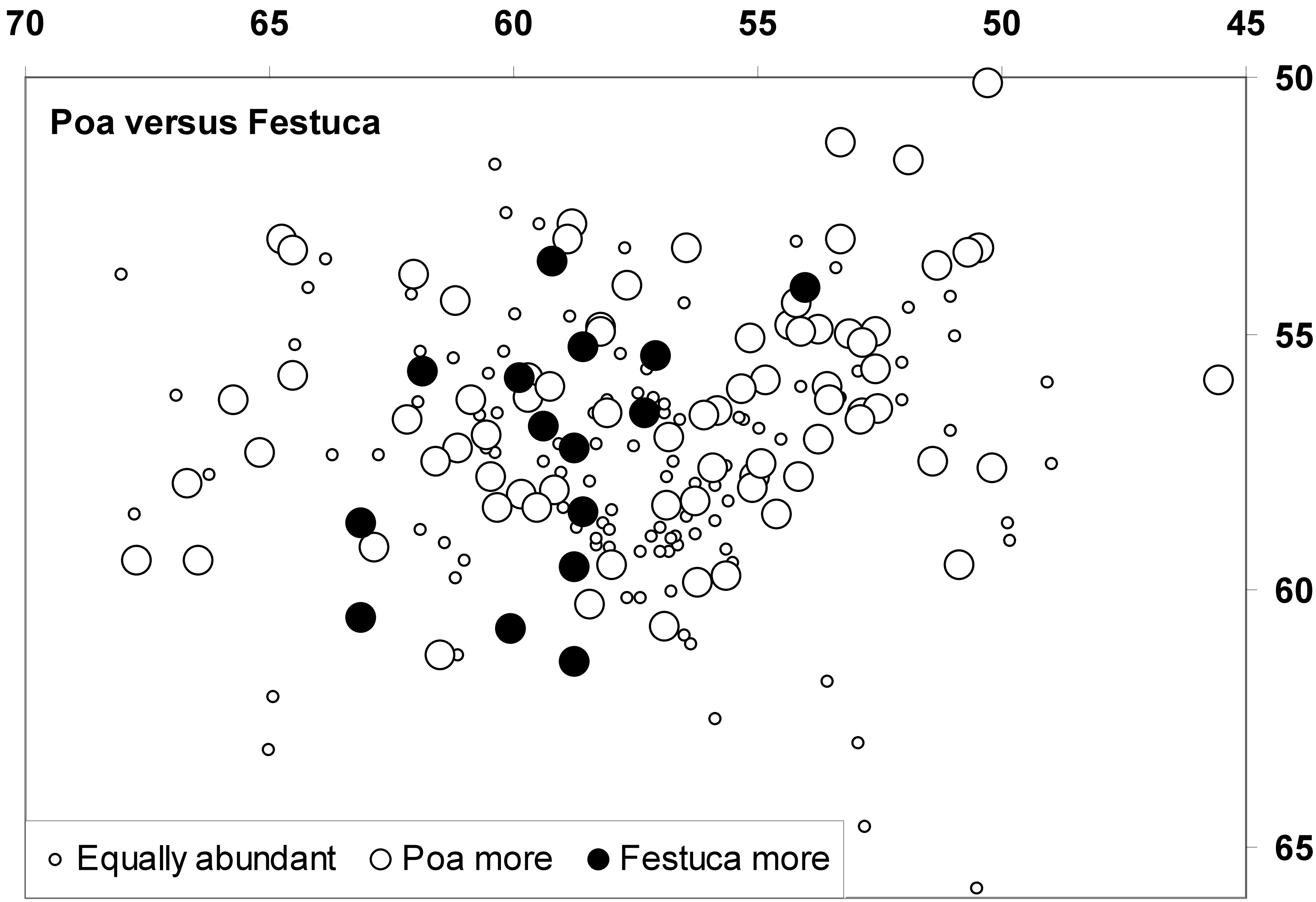


Figure 15a. Abundance of *Poa pratensis* versus *Festuca arundinacea* in each plot, overlaid on ordination. “Equally abundant” = same score assigned in the 8-point scale (see Part I).

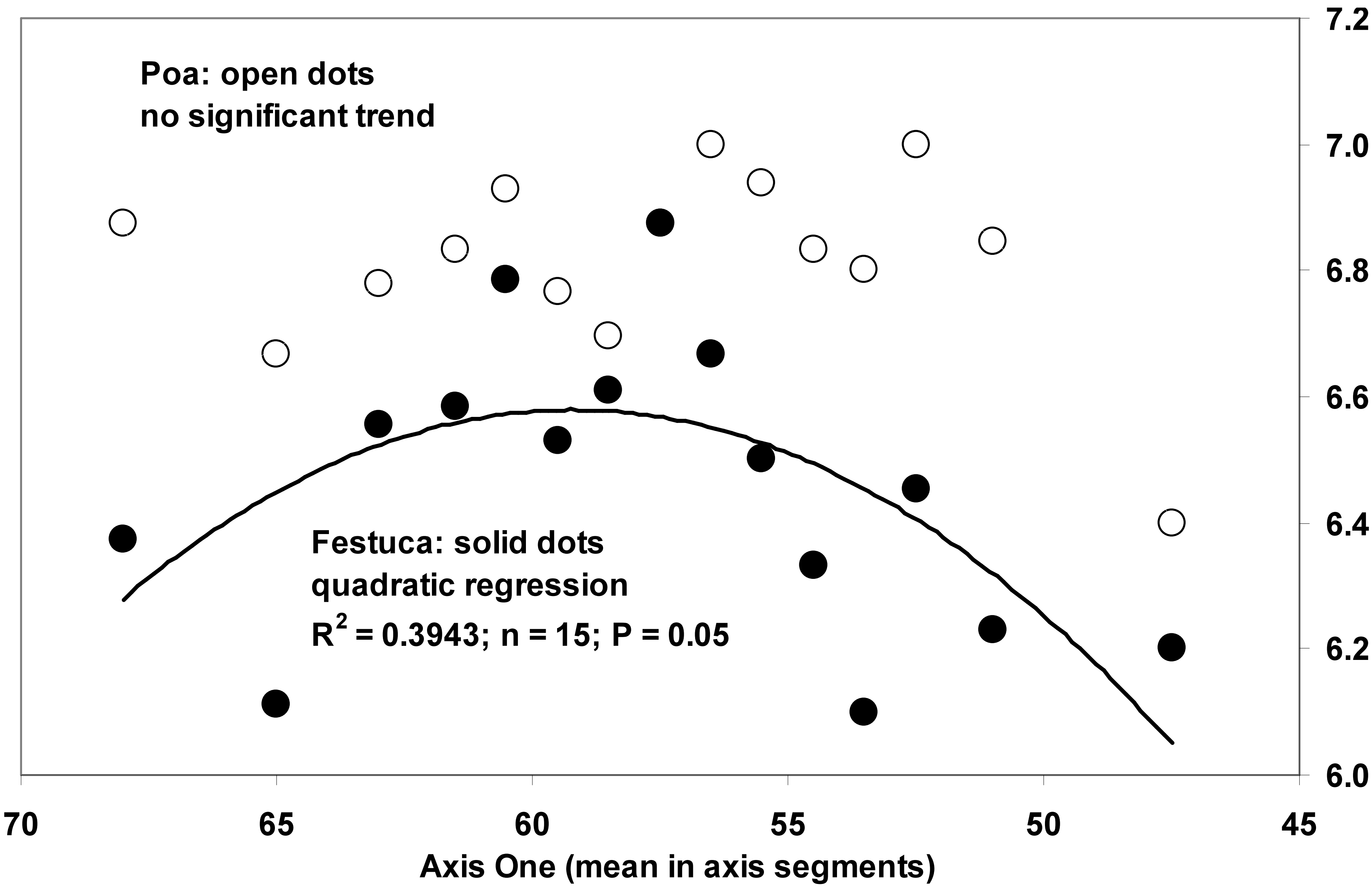


Figure 15b. Abundance of *Festuca arundinacea* and *Poa pratensis* in relation to Axis One: means of the 8-point quasi-logarithmic scale within segments of the axis (see Part I for scale definition).

Table 2. Scores of species in the ordination, and some ecological characteristics. Explanation of columns is as follows.

Species Name. Nomenclature for these 106 species generally follows Weakley (2015), but not using subspecific names. This list includes all species observed in the field during 2003-2015. A few species were not recorded in plots; these are labelled under axis scores as "early" (flowering well before the survey in September) or "trace". Question marks (?) indicate species with somewhat tentative identifications. Asterisks (*) indicate species that spread into this field at least partly due to recent artificial introduction of seed.

Axis One and Axis Two. These are scores from the DCA analysis, standardized along a 0-100 scale in both cases. Plot scores (Figure 1 etc.) are weighted averages of these species scores.

Alien Status. X = native to Eurasia; Z = status uncertain or mixed. Other species (-) appear to be generally native in this region, although a few may have moved in during recent centuries from the south (e.g. *Ipomaea hederacea* with pre-Columbian agriculture) or from the west (e.g. *Rudbeckia serotina*, the more weedy variant in the *R. hirta* complex).

Hydrology. Based on J. Campbell & M. Medley (unpublished database on available at bluegrasswoodland.com). These codes indicate slight to strong association with hydrology not typical of the general rolling uplands of this region. HH = concentrated on hydric or subhydric soils. H = occurs partially on subhydric soils. R = occurs partially in riparian zones, strictly defined (often flooded but without stagnant water). X = occurs partially on subxeric sites. XH = tends to occur on both subhydric and subxeric sites. Other species (-) are concentrated on mesic to submesic sites, with little or no regular extension into these coded habitats.

pH-Index. Based on Campbell & Medley (see above), where a five-point scale is presented for all vascular plants in Kentucky. Species with cores 1 and 2 are not represented here.

3 = Concentrated on medium acid soils (ca. pH 5-6) with medium overall fertility. Typical upland oaks include *Q. alba*, *Q. velutina*, *Q. stellata* and *Q. falcata*. Typical species have widely scattered distribution over the state, including parts of the Bluegrass, or other calcareous regions, as well as the Knobs and Appalachian hills.

4 = Transitional or uncertain assignment between scores 3 and 5. Widespread species that are common on farmland or alluvial soils with relatively high fertility (especially in N and P) are generally assigned here.

5 = Concentrated on weakly acid to neutral soils (ca. pH 6-7), with medium to high overall fertility, especially in bases (Ca, Mg, K). Typical upland oaks include *Q. muhlenbergii*, *Q. shumardii*, *Q. macrocarpa* and locally *Q. imbricaria*. Typical species have higher frequency in the Bluegrass or other calcareous regions, compared to the Knobs and Appalachian hills (excluding more fertile valleys and other unusual sites).

Nitrophilous-Index. Developed from the ten-point scale developed in Europe by Ellenberg (1988), Hill (1999, 2000) and others. Application to North American species remains tentative (?), based on experience of the author in Kentucky plus some detailed studies of nitrogen responses elsewhere in eastern states (e.g. Tilman 1987, Wilson & Tilman 2002, Harpole & Tilman 2006). 1 = indicator of extremely infertile sites; 2 = between 1 and 3; 3 = indicator of more or less infertile sites; 4 = between 3 and 5; 5 = indicator of sites of intermediate fertility; 6 = between 5 and 7; 7 = often found in richly fertile places; 8 = between 7 and 9; 9 = indicator of extremely rich situations, such as cattle resting places or near polluted rivers. There is a significant correlation with Maximum Height (see below): $r = 0.37$; $P = 0.0003$ (2-tailed).

Asterisks indicate species with known 'nitrogen-rich secondary chemistry' such as cyanogenic glycosides, alkaloids, nitroproprionates (*Securigera*) or similar compounds.

Maximum Height. Typical mid-points of ranges in reported heights at maturity in decimeters. Sources are mostly standard floras, especially Fernald (1950), Gleason & Cronquist (1991) and Flora of North America (1993-2015). Vines (v) and woody species (w) are excluded.

Toxicity and Spines. T indicates potentially intense mammalian toxity; t = less intense or merely deterrent; see Part I for sources. S/s indicates spines or prickles on leaves or stems.

Life-form. This classification is based mostly on standard floras, as in Maximum Height. A = generally summer annual or winter annual. B = generally biennial or short-lived somewhat monocarpic perennial (often difficult to separate from winter annuals). R = perennial herb with rapid clonal spread by running roots, rhizomes or stolons. V = vines. W = woody plant. Other species (-) are perennial herbs without rapid clonal spread (generally less than 2 dm per year).

Species Name	Axis One	Axis Two	Alien Status	Hydro -logy	pH-Index	Nitro.-Index	Max. Height	Tox. Spi.	Life-form
Acalypha rhomboidea	25.5	85.1	-	R	4	6?	4		A
Achillea borealis	37.4	64.6	Z	-	4	4	4		R
Alliaria petiolata	93.3	72.2	X	R	5	8*	6	t	B
Allium vineale	66.2	68.0	X	-	4	6*	7	t?	-
Amaranthus hybridus	72.7	41.1	-	H	4	7	14		A
Ambrosia artemisiifolia	47.6	68.8	-	X	4	8?	6		A
Ambrosia trifida	75.3	74.6	-	R	4	9?	20		A
Andropogon virginicus	36.1	53.3	-	XH	3	4?	12		-
Apocynum cannabinum	67.8	46.3	-	R	4	7?*	10	t	R

Asclepias syriaca	52.9	48.5	-	-	4	9?*	12	T	R
Barbarea vulgare	64.4	48.3	X	-	4	7*	6	t	A
Bignonia capreolata	42.3	25.3	-	R	4	6?	wv		WV
Brassica rapa	87.1	78.2	X	-	5	6*	6	t	A
Bromus ?japonicus	84.8	33.4	X	-	4	5	5		A
Bromus inermis	95.3	54.4	X	X	5	5	9		R
Calystegia fraterniflora	48.4	44.5	-	-	4	8*	v	t	V
Campsis radicans	43.1	39.8	-	H	4	8?	wv		WV
Cardamine hirsuta	early	early	-	-	4	6*	2	t	A
Carduus acanthoides	67.8	75.3	X	-	5	7	20	s	B
Carduus nutans	40.6	71.3	X	H	4	6	15	s	B
Carex ?aggregata	early	early	-	-	4	7?	7		-
Carex ?blanda	60.3	38.5	-	H	4	8?	3		-
Carex ?conjuncta	72.2	31.6	-	HH	5	8?	6		-
Carex ?frankii	42.3	25.3	-	HH	4	7?	5		-
Carex ?grisea	41.0	57.9	-	R	4	8?	5		-
Carex ?jamesii	early	early	-	-	4	6?	2		-
Carex ?vulpinoidea	28.9	19.2	-	HH	4	7?	7		-
Cerastium glomeratum	early	early	-	-	4	5	2		A
Chamaesyce nutans	82.5	56.4	-	R	4	7?*	4	t	A
Cichorium intybus	32.1	42.2	X	-	4	5	6		B
Cirsium arvense	80.1	36.0	X	-	4	7	12	s	R
Cirsium discolor	48.8	69.6	-	-	4	7?	15	s	B
Cirsium vulgare	39.3	45.9	X	-	4	7	15	s	B
Conium maculatum	82.6	73.4	X	R	5	8*	20	T	B
Conyza canadensis	19.5	72.0	-	-	4	5	15		A
Cruciata pedemontanum	early	early	X	-	4	5?	2		A

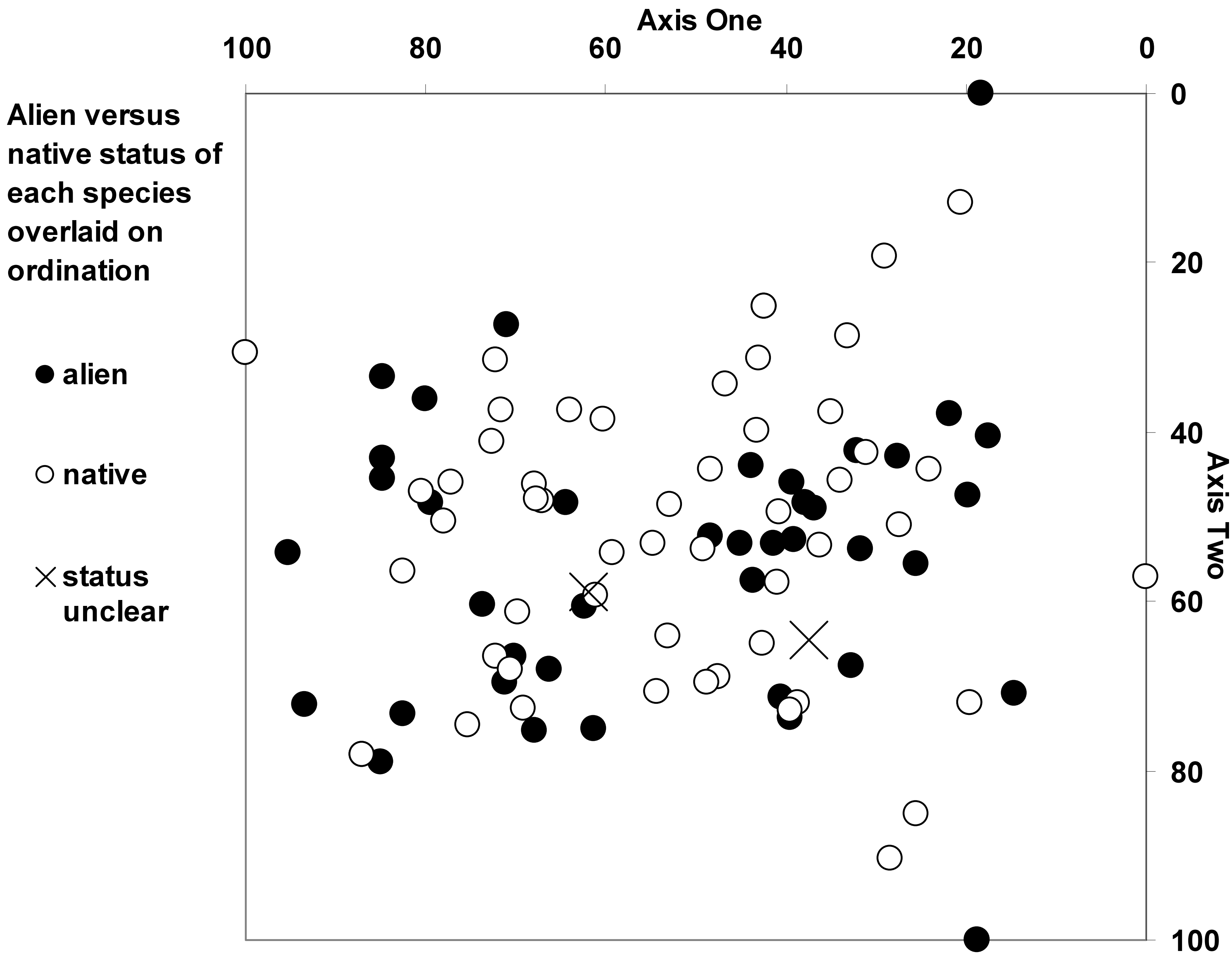
Cynanchum laeve	67.7	47.9	-	-	4	8?*	v	t	V
Dactylis glomerata	39.4	73.7	X	-	4	6	9		-
Daucus carota	41.3	53.1	X	X	4	4*	7	t	B
Desmodium perplexum	31.0	42.4	-	-	4	6?*	9		-
Dianthus armeria	31.7	53.9	X	-	4	3	4		B
Dichanthelium acuminatum	24.2	44.3	-	X	3	4?	6		-
D. clandestinum	33.2	28.6	-	R	4	6?	9		-
Dipsacum fullonum	100.0	30.6	X	XH	4	7	15	s	B
Elymus virginicus	87.1	78.2	-	H	4	8?	8		-
Eragrostis spectabilis	20.6	13.0	-	-	3	6?	5		-
Erigeron annuus	38.8	71.9	-	-	4	7?	13		A
Erigeron philadelphicus	early	early	-	-	4	7?	4		A
Eupatorium perfoliatum	54.3	70.7	-	HH	4	8	10		-
Festuca arundinacea	62.4	60.5	X	H	4	6*	10	t	-
Geum vernum	early	early	-	-	5	6?	4		A
Glechoma hederacea	73.7	60.4	X	R	4	7*	3	t	R
Hypericum perforatum	70.3	66.4	X	-	4	5*	6	t	-
Ipomaea hederacea	trace	trace	-	-	4	9?*	v	t	A
Lactuca saligna	85.0	79.0	X	X	5	6	5	s?	B
Leucanthemum vulgare	36.8	49.0	X	-	4	4	5		R
Lonicera japonica	71.1	27.4	X	X	3	5?	wv		WV
Medicago sativa	37.9	48.4	X	-	4	5*	6		-
Oxalis dillenii	27.4	50.9	-	-	4	5	3		R
Parthenocissus quinquefolia	42.6	65.1	-	XH	4	7?	wv		WV
Paspalum laeve	0.0	57.1	-	-	3	6?	8		-
Pastinaca sativa	79.5	48.5	X	H	5	5*	10	t	B
Physalis heterophylla	70.7	68.0	-	-	4	8?*	5	t?	R

Physalis longifolia	69.8	61.2	-	-	5	7?*	6	t?	R
Phytolacca americana	80.4	47.0	-	-	4	8?*	20	T	-
Plantago lanceolata	39.0	52.7	X	-	4	4	2		-
Plantago rugellii	19.5	72.0	-	-	4	7?	2		-
Poa pratensis / angustifolia	62.0	58.8	Z	-	4	6	5		-
Potentilla recta	25.5	55.6	X	-	4	6?	6		-
Rubus ?flagellaris	28.5	90.4	-	X	3	6?	wv	S	WV
Rubus ?pennsylvanica	39.5	72.8	-	-	4	6?	20	S	W
Rudbeckia serotina	34.0	45.8	-	-	4	7?	6		B
Rudbeckia triloba	63.9	37.4	-	XH	4	8?	10		B
Ruellia strepens	77.1	45.9	-	R	5	7?	6		-
Rumex acetosella	27.7	43.0	X	X	3	3	3		-
Rumex crispus	71.2	69.5	X	H	4	6	8		-
Rumex obtusifolia	45.1	53.1	X	HH	4	9	9		-
Securigera varians	84.7	45.5	X	-	4	6?*	6	T	R
Setaria faberi	18.3	0.0	X	-	4	7?	13		A
Setaria pumila	85.0	79.0	X	X	4	5?	8		A
Setaria viridis	32.6	67.6	X	X	4	6?	13		A
Sisymbrium officinale	43.8	44.0	X	-	4	7*	5	t	A
Solanum carolinense	67.0	48.2	-	-	4	7?*	6	Ts	R
Solidago altissima	61.2	59.3	-	H	4	6	13		R
Sorghum halepense	84.7	43.2	X	-	4	7?*	12		R
Spiranthes gracilis	78.0	50.4	-	-	3	5?	4		-
Symphoricarpus orbiculatus	42.9	31.4	-	X	4	7?	w		W
Symphotrichum pilosum	49.2	53.8	-	H	4	6	7		-
*S. novae-angliae**	69.1	72.6	-	XH	4	8	12		-
S. ontarione	40.8	49.4	-	R	5	7	7		-

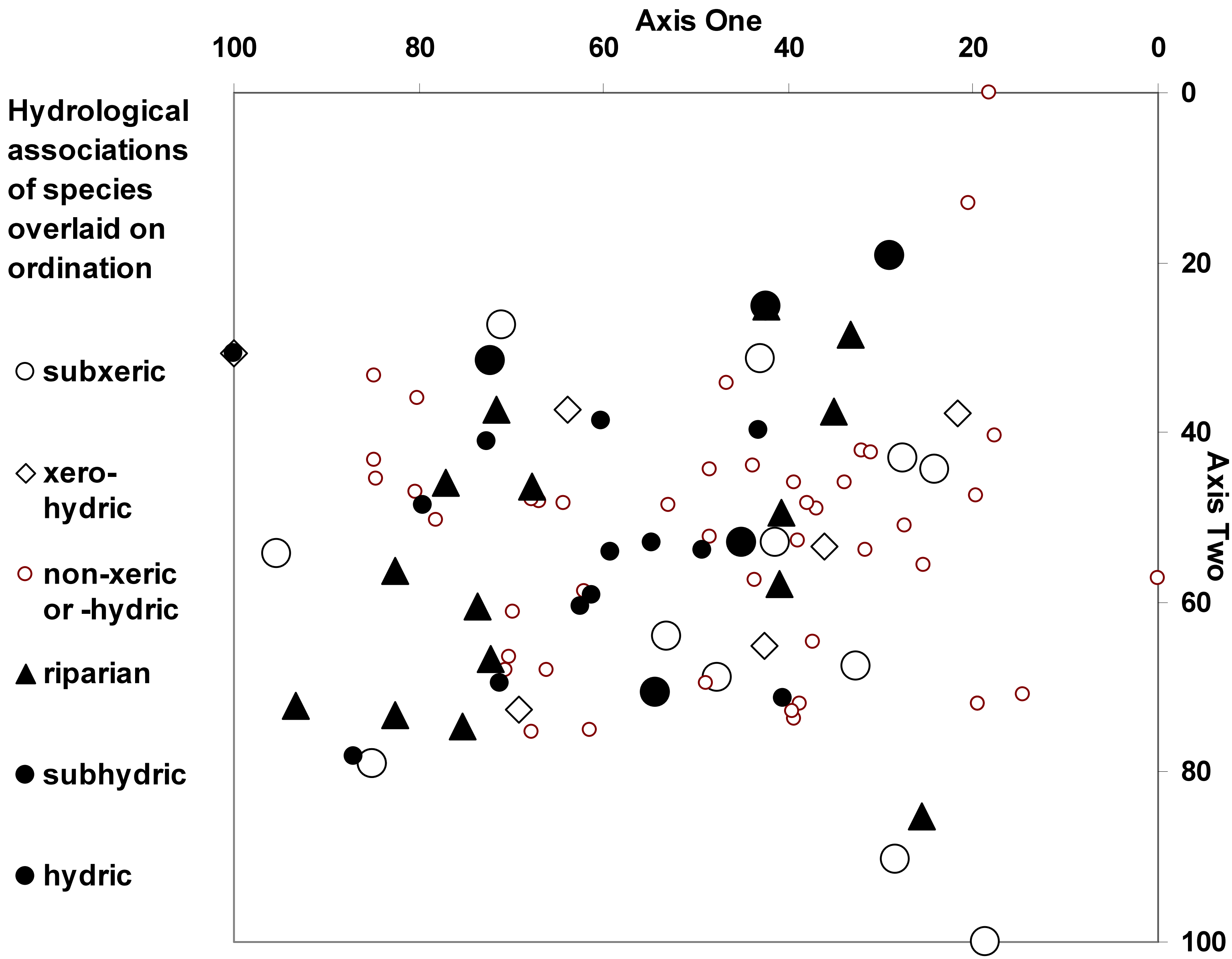
Taraxacum officinale	21.8	37.8	X	XH	4	6	2		-
Torilis japonica	43.6	57.5	X	-	4	7*	8	t	A
Toxicodendron radicans	54.7	53.1	-	H	4	7?	wv	t	WV
Tridens flavus	53.1	64.1	-	X	4	7?	12		-
Trifolium campestre	17.6	40.5	X	-	4	4*	2		A
Trifolium pratense	19.8	47.5	X	-	4	5*	3		-
Trifolium repens	14.6	71.0	X	-	4	6*	2		R
Verbascum blattaria	48.4	52.3	X	-	4	6?	10	t	B
Verbascum thapsus	18.6	100.0	X	X	4	6	15		B
Verbena urticifolia	71.6	37.4	-	R	4	7?	9		B
Verbesina alternifolia	72.2	66.6	-	R	4	8?	15		-
*Verbesina virginica**	46.7	34.3	-	-	4	7?	15		-
Vernonia gigantea	59.2	54.2	-	H	4	8?	15		-
Veronica arvensis	61.4	75.0	X	-	4	6?	2		A
Viola papilionacea	35.0	37.6	-	R	4	7?	2		R
Xanthium canadense	100.0	30.6	-	H	4	7?	11	t	A

===

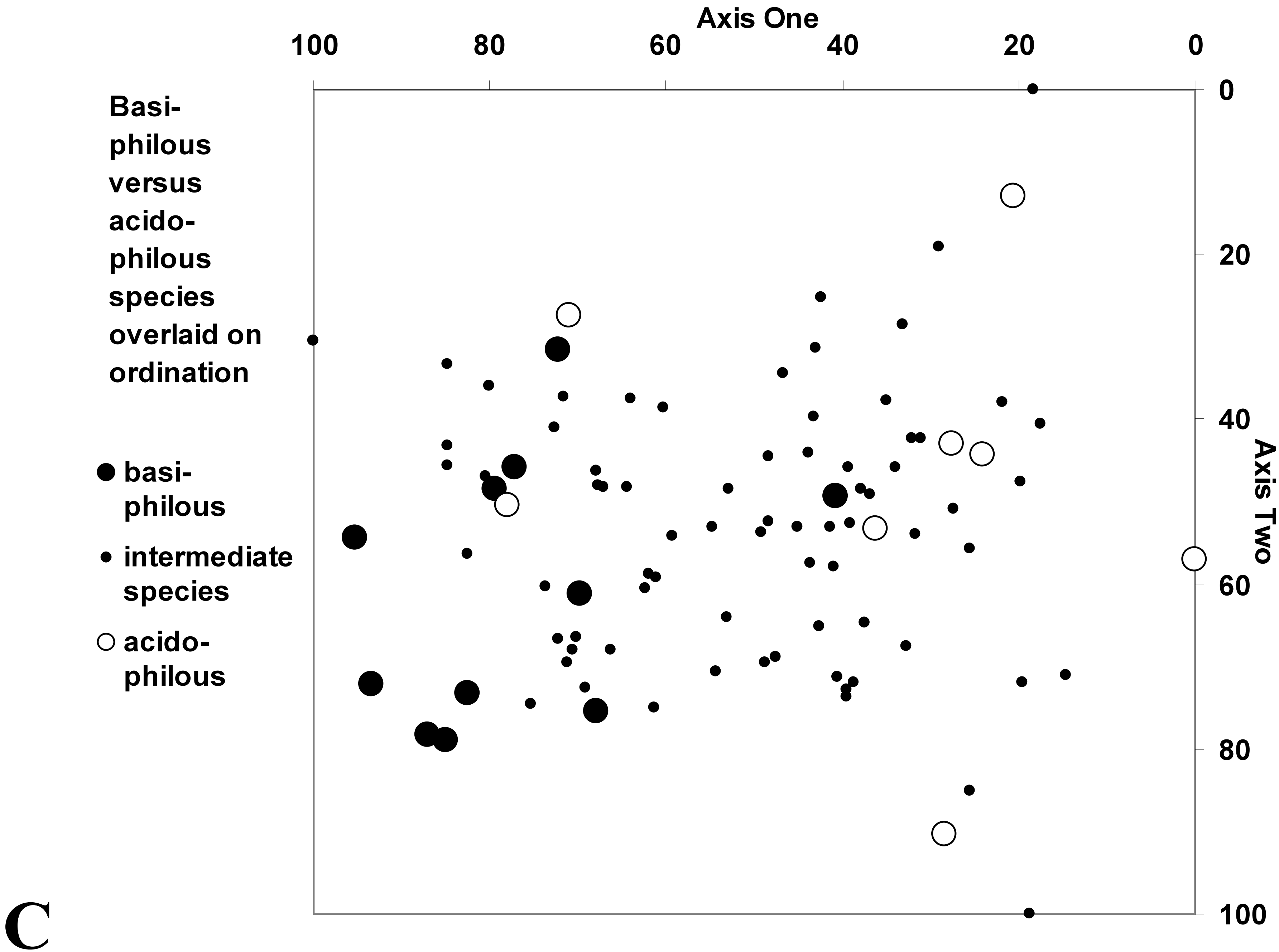
Figure 16 [subsequent pages]. Some characteristics of species overlaid on the ordination. Axes One and Two here are species scores from the DCA analysis, standardized along a 0-100 scale in both cases. Plot scores (Figure 1 etc.) are local weighted averages of these species scores. See Table 1 for details of species' characteristics and sources of information.
A: alien versus native status. B: hydrological associations. C: basiphilous versus acidophilous. D: nitrophilous index. E: species with 'nitrogen-rich' chemistry (alkaloids, cyanogenic glycosides, nitropropionates, etc.). F: indications of herbivore resistance. G: typical maximum height. H: life-form.

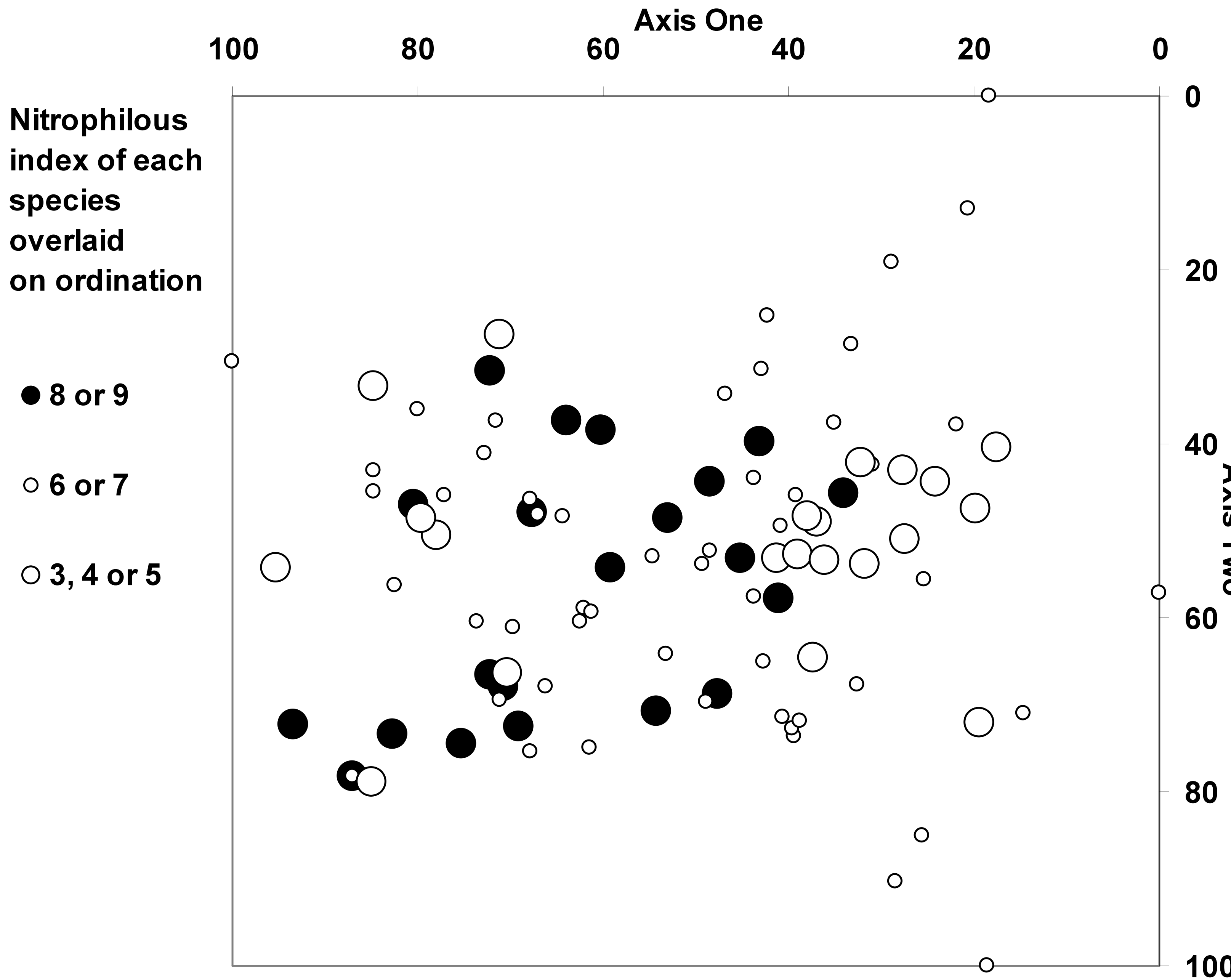

Axis One
100
80
60
40
20
0
Alien versus native status of each species overlaid on ordination
alien
native
status unclear
Axis Two
0
20
40
60
80
100

A

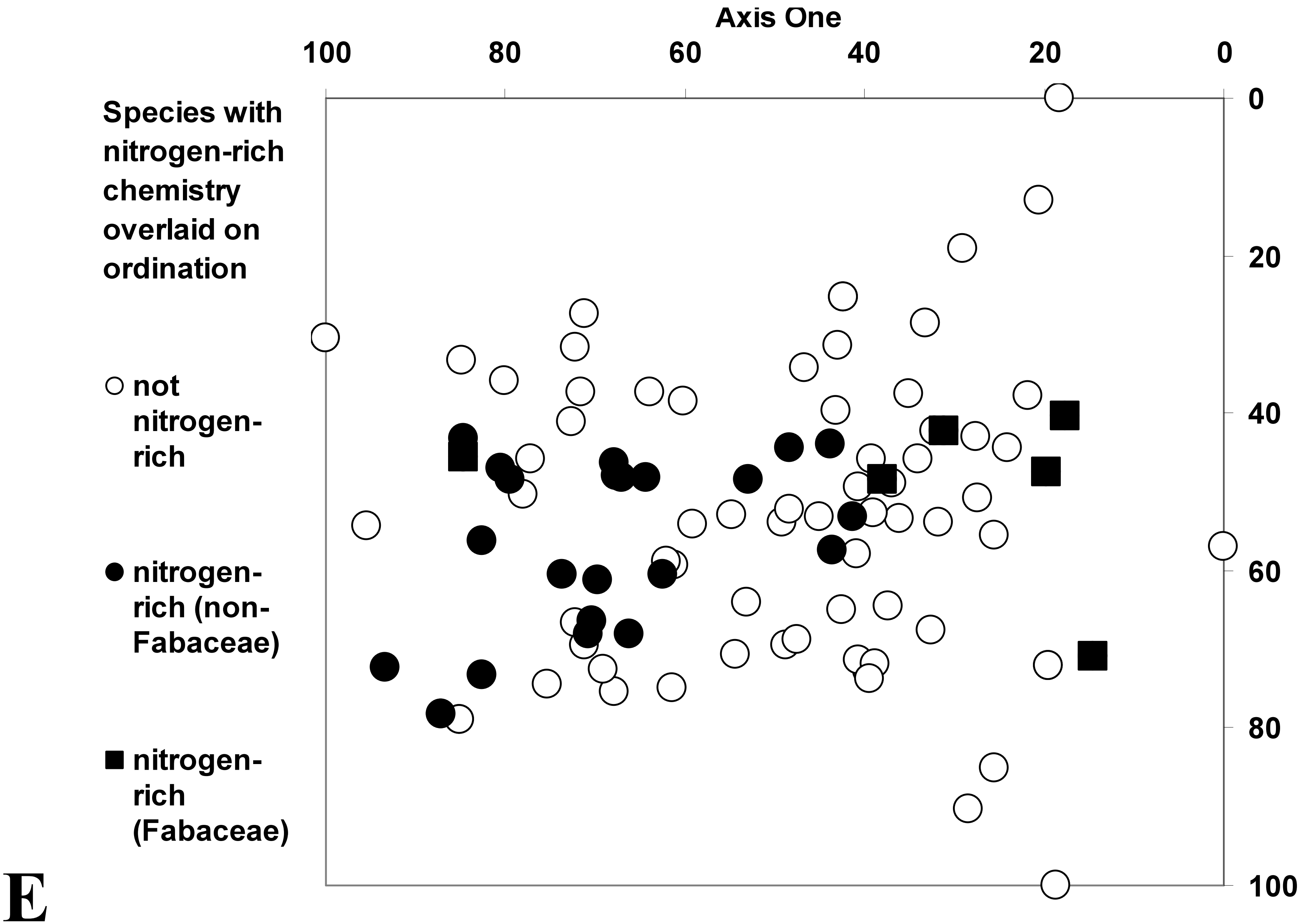

Axis One
100
80
60
40
20
0
Axis Two
0
20
40
60
80
100
Hydrological associations of species overlaid on ordination
subxeric
xero-hydric
non-xeric or -hydric
riparian
subhydric
hydric

B

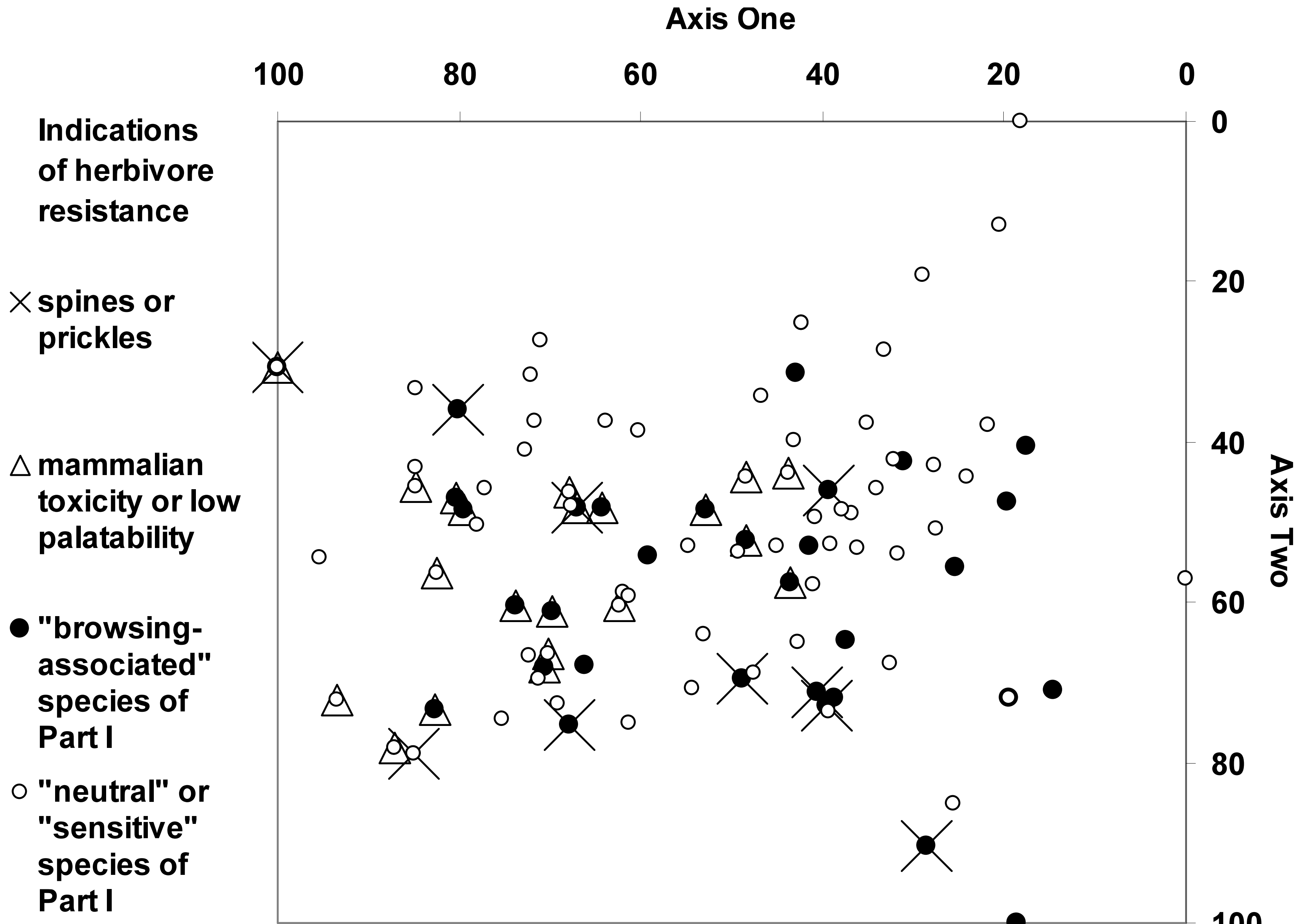

Axis One
100
80
60
40
20
0
0
20
40
60
80
100
Axis Two
Basi-philous versus acido-philous species overlaid on ordination
basi-philous
intermediate species
acido-philous
C


Axis One
100
80
60
40
20
0
Axis Two
0
20
40
60
80
100
Nitrophilous index of each species overlaid on ordination
8 or 9
6 or 7
3, 4 or 5

D

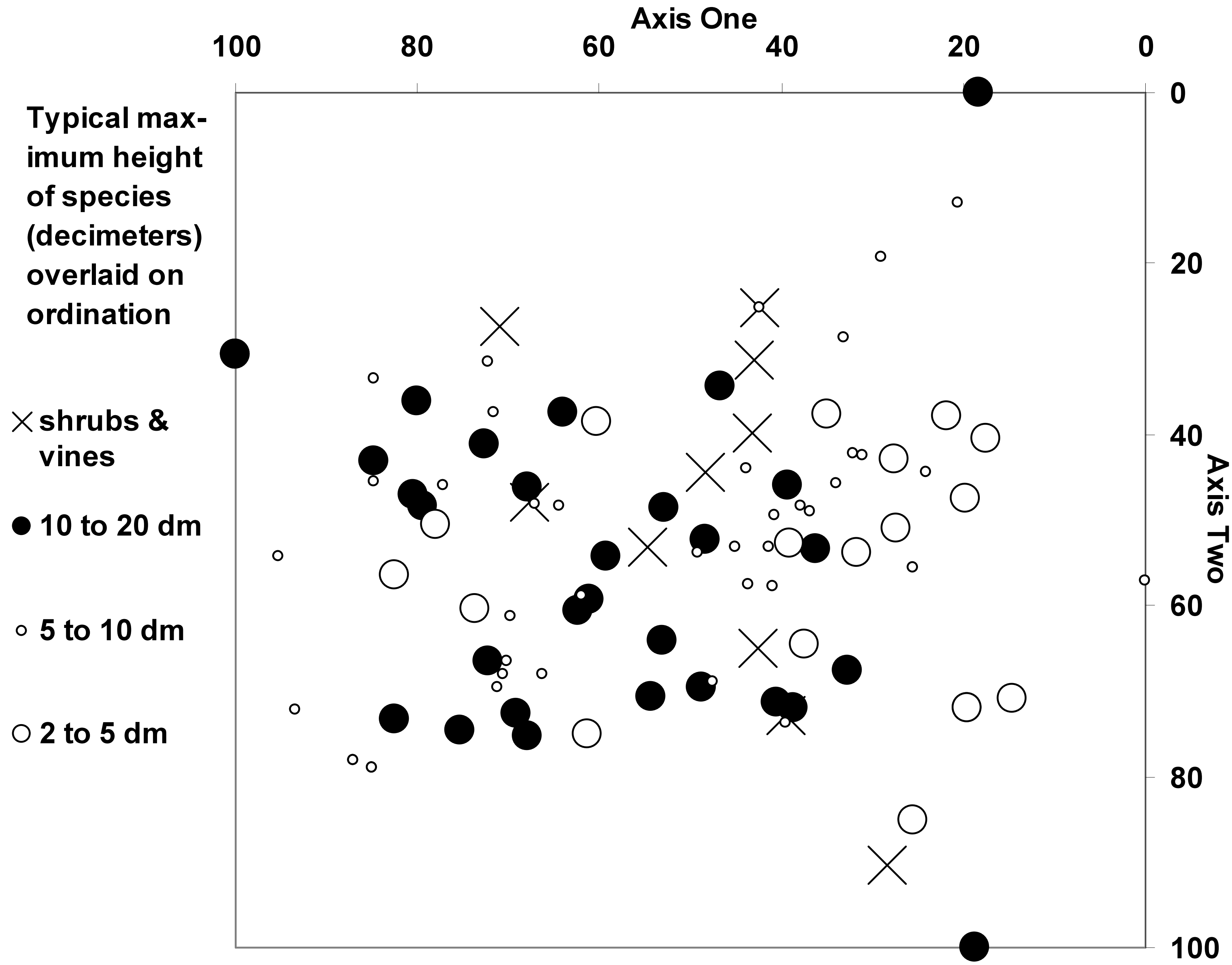


Axis One
100
80
60
40
20
0
Species with nitrogen-rich chemistry overlaid on ordination
not nitrogen-rich
nitrogen-rich (non-Fabaceae)
nitrogen-rich (Fabaceae)
Axis Two
0
20
40
60
80
100
E

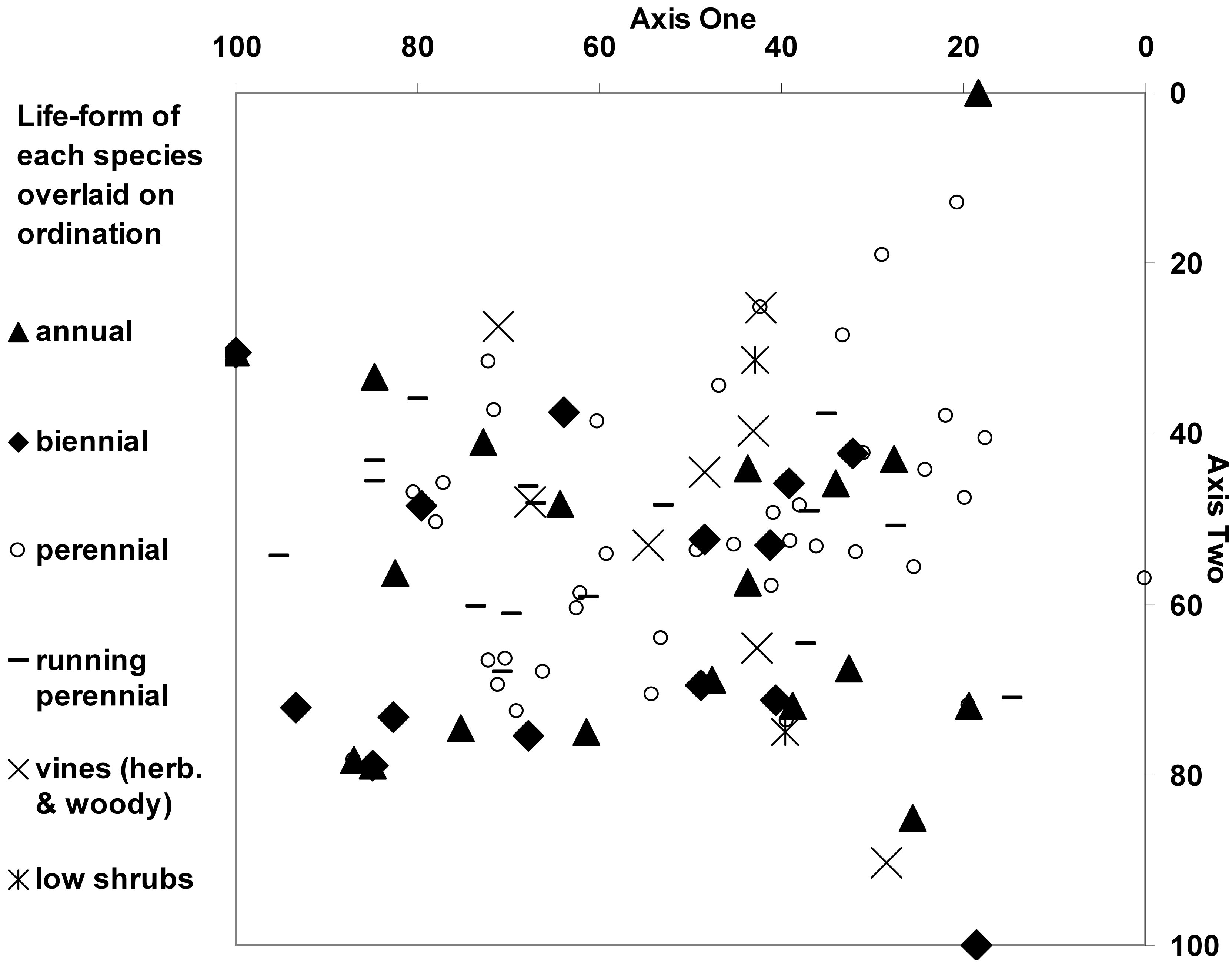


F

The “browsing-associated” group has higher Axis Two scores with t-test but P = 0.047.


Axis One
100
80
60
40
20
0
Axis Two
0
20
40
60
80
100
Typical maximum height of species (decimeters) overlaid on ordination
shrubs & vines
10 to 20 dm
5 to 10 dm
2 to 5 dm

G



Axis One
100
80
60
40
20
0
Axis Two
0
20
40
60
80
100
Life-form of each species overlaid on ordination
annual
biennial
perennial
running perennial
vines (herb. & woody)
low shrubs

H

Figure 17. Trends in some species' characteristics along Axis One.
These are summaries of most significant trends in Figure 16; x-axis is reversed Axis One score.
A: proportion of alien species (excluding the two with unclear status); see Figure 16A.
B: mean basiphilous index; see Figure 16C.
C: mean nitrophilous index; see Figure 16D.
D: mean of log [maximum height in decimeters]; see Figure 16G.

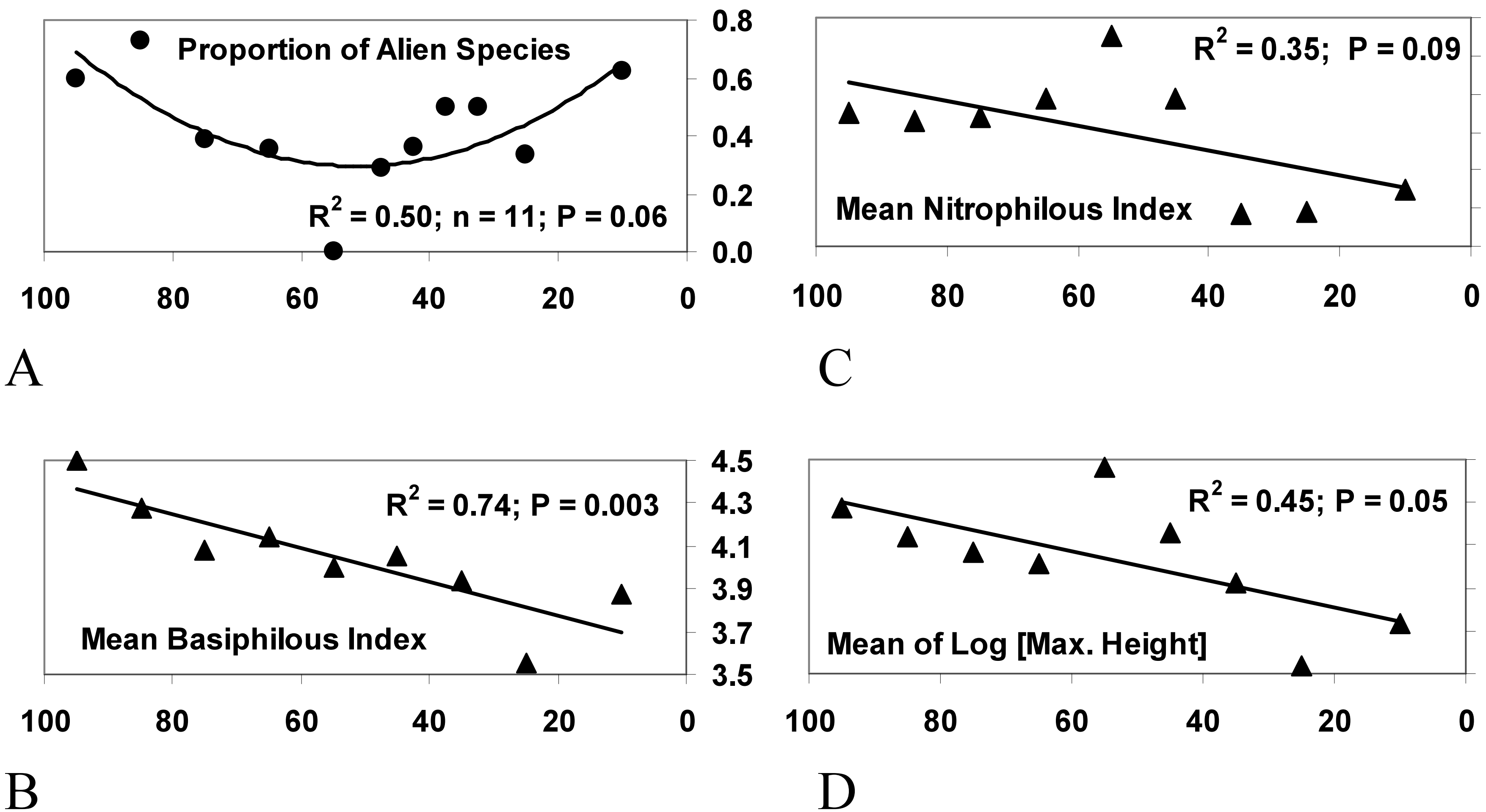
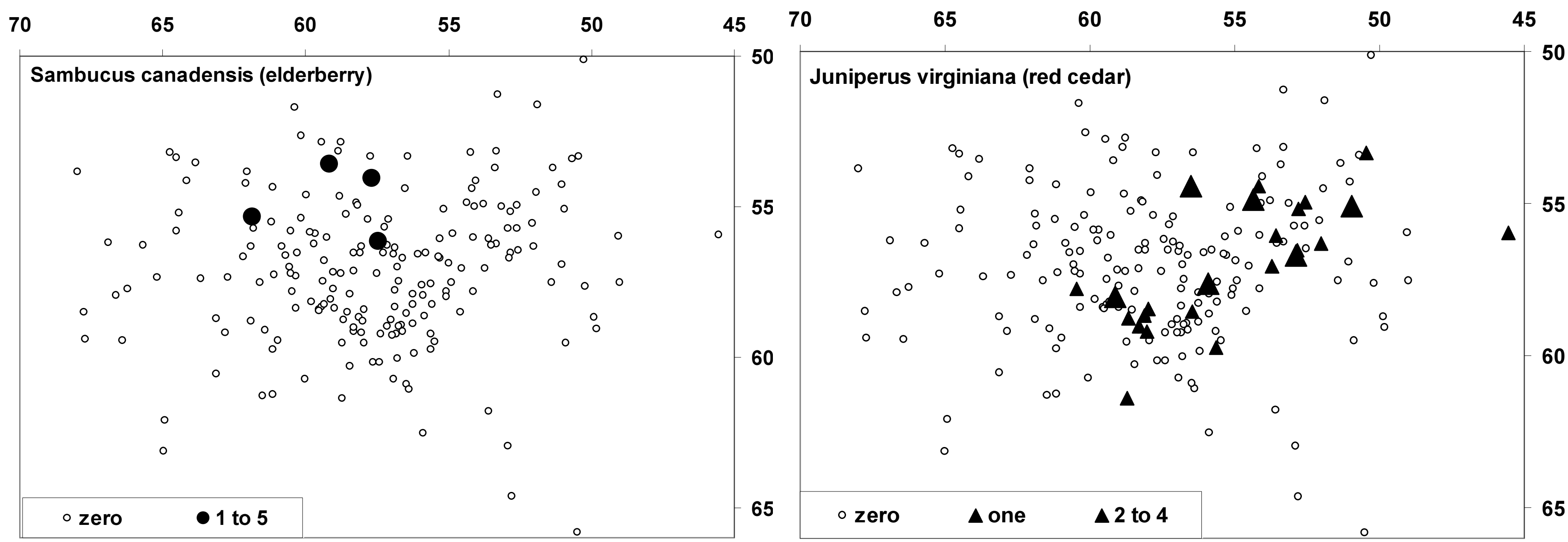
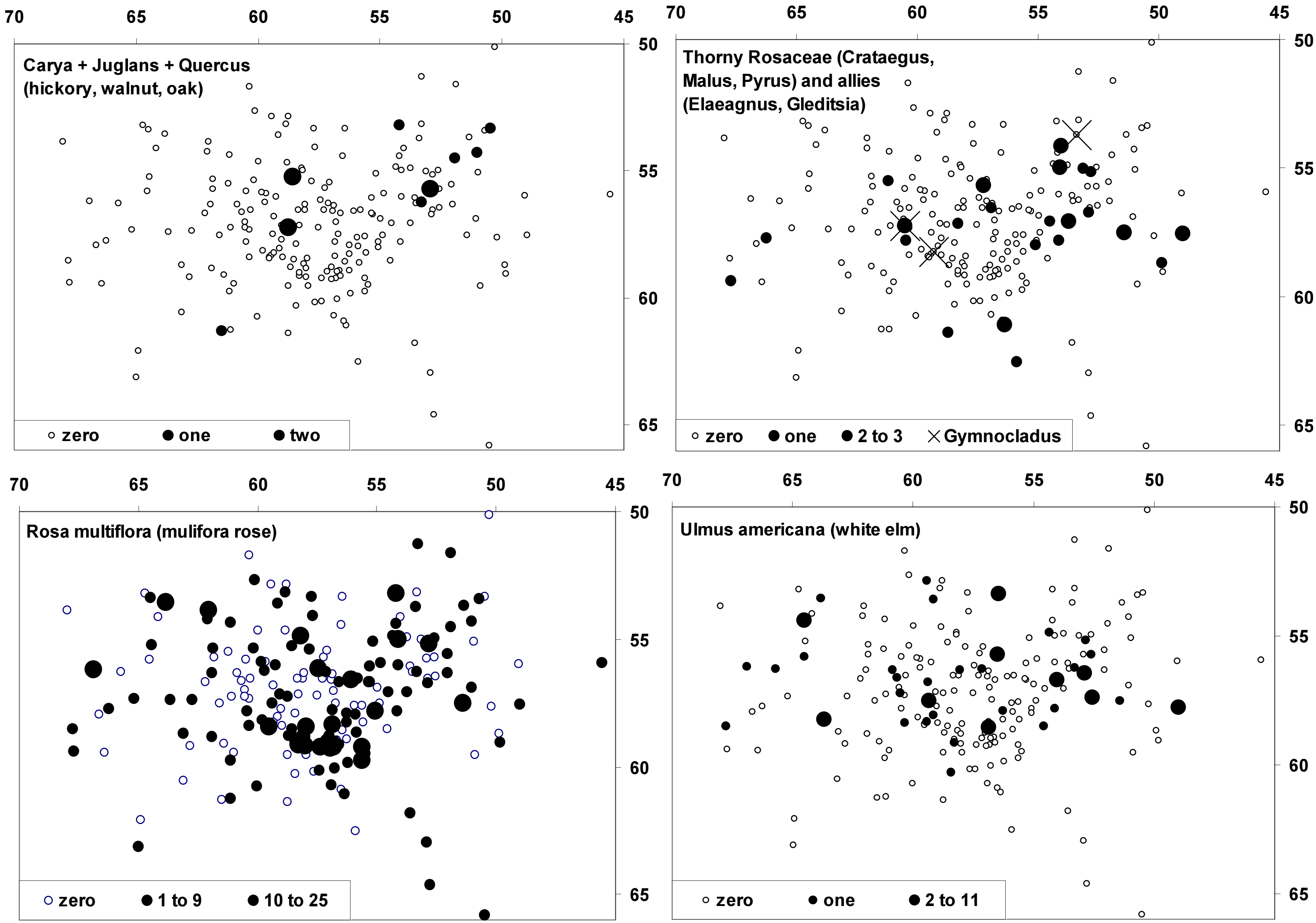
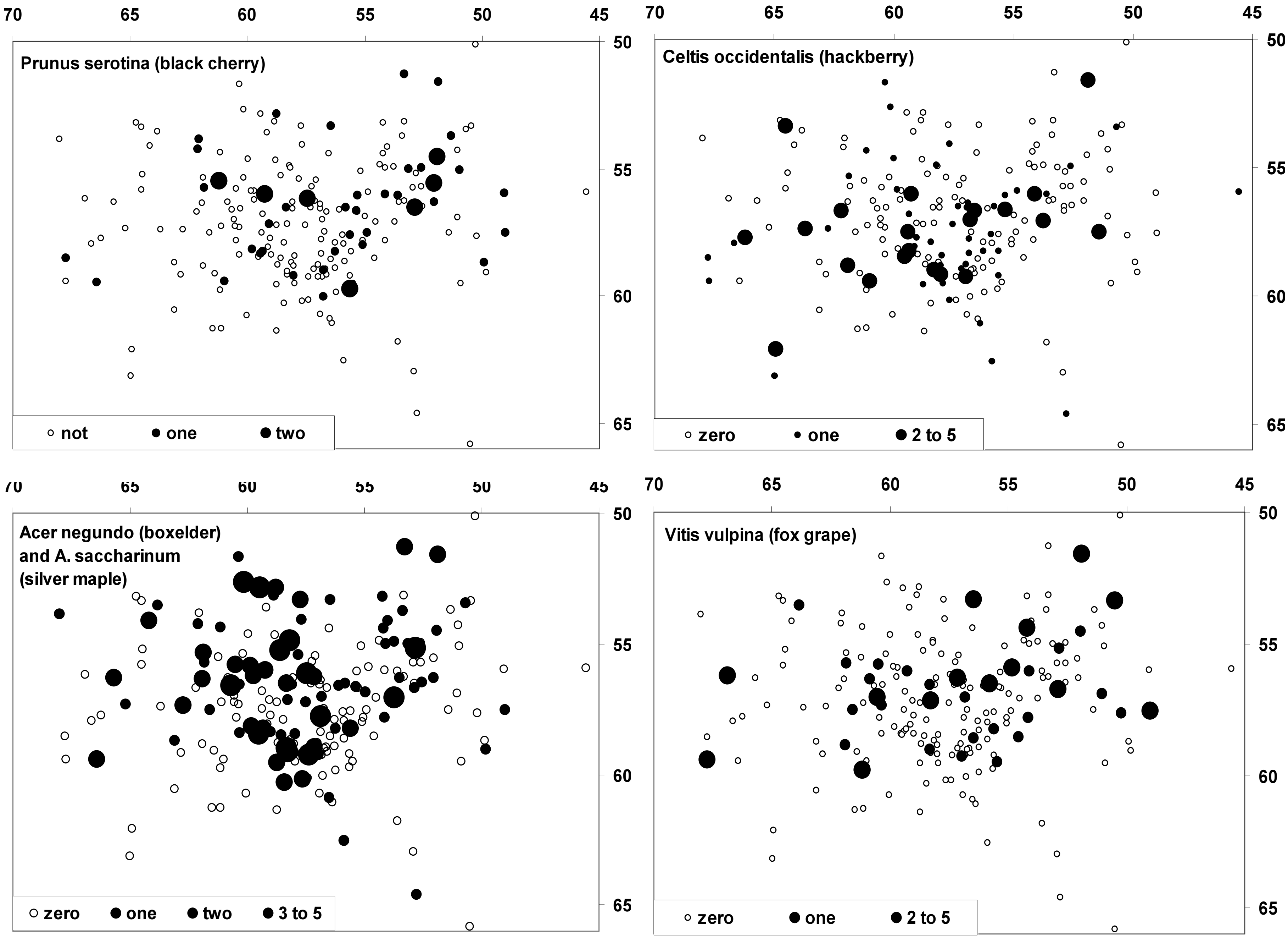


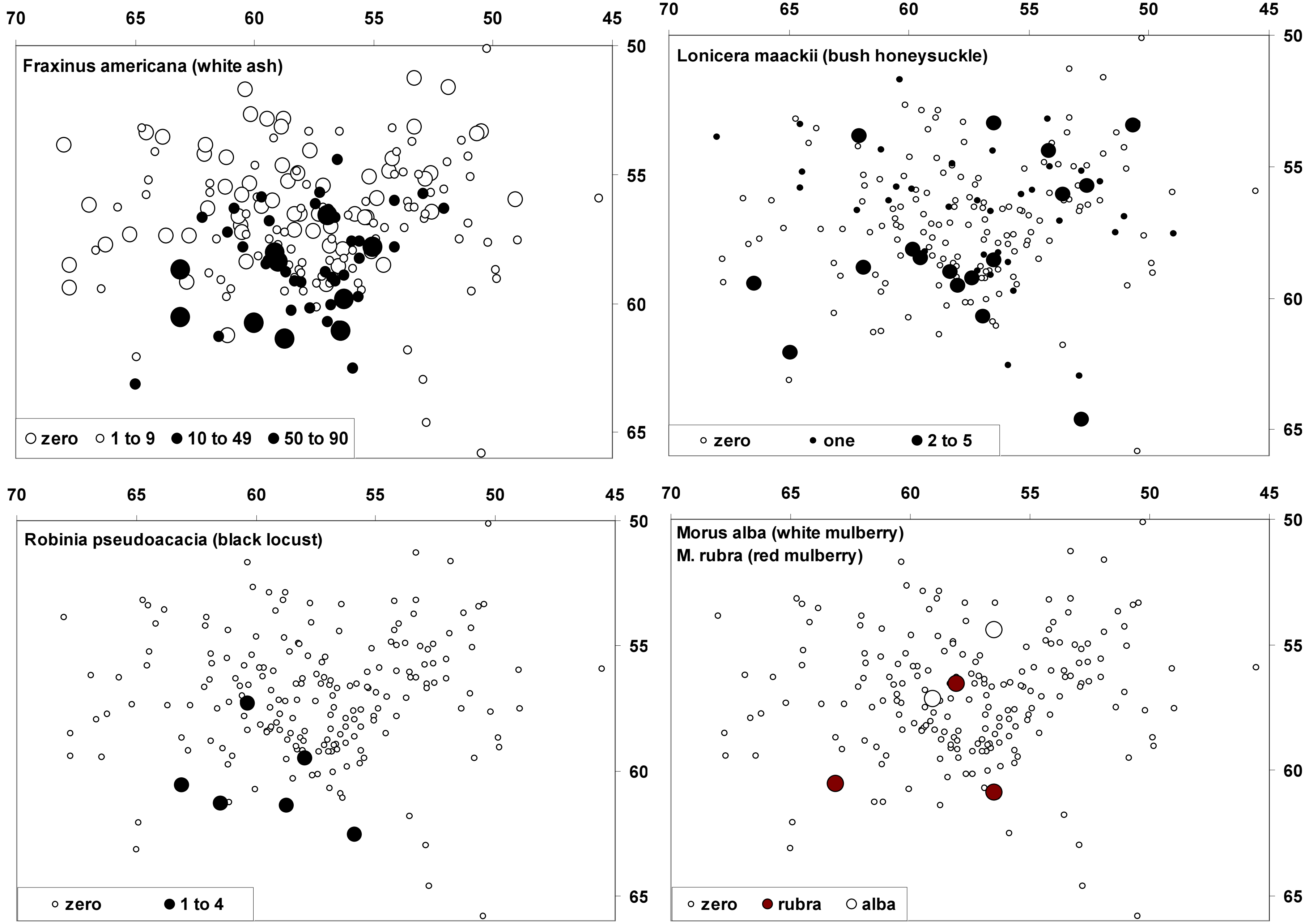
Figure 18. Abundances of self-sown woody species overlaid on ordination. Abundances are measured in the quasi-logarithmic 8-point scale, as detailed in Part I (Table 1). Charts for each species are arranged in approximate order from those concentrated higher on Axis Two (upper) to those lower on Axis Two (below). Vines are excluded except for *Vitis vulpina*, since these are mostly ground covering within this field (< 2 m height). The low shrubs, *Symphoricarpos orbiculatus* and *Rubus pennsylvanicus*, are also excluded since these are mostly < 2 m in height. [Species with 'nitrogen-rich secondary chemistry' include *Sambucus canadensis*, *Elaeagnus umbellata*, *Crataegus* spp., *Prunus serotina*, *Gymnocladus dioicus*, *Robinia pseudoacacia*.]




Carya + Juglans + Quercus
(hickory, walnut, oak)
zero
one
two
Thorny Rosaceae (Crataegus,
Malus, Pyrus) and allies
(Elaeagnus, Gleditsia)
zero
one
2 to 3
Gymnocladus
Rosa multiflora (mulifora rose)
zero
1 to 9
10 to 25
Ulmus americana (white elm)
zero
one
2 to 11
70
65
60
55
50
45



Prunus serotina (black cherry)
not
one
two
Celtis occidentalis (hackberry)
zero
one
2 to 5
Acer negundo (boxelder)
and A. saccharinum
(silver maple)
zero
one
two
3 to 5
Vitis vulpina (fox grape)
zero
one
2 to 5
70
65
60
55
50
45
50
55
60
65


Fraxinus americana (white ash)
zero
1 to 9
10 to 49
50 to 90
Lonicera maackii (bush honeysuckle)
zero
one
2 to 5
Robinia pseudoacacia (black locust)
zero
1 to 4
Morus alba (white mulberry)
M. rubra (red mulberry)
zero
rubra
alba
70
65
60
55
50
45

DISCUSSION

“This Old Field”

How did this particular old field come to be studied? How relevant are results to the region? How do they contribute to basic ecological understanding? Answers are not yet satisfying. The field was selected for serendipitous reasons unrelated to the ultimate themes of this paper. Current results may be relevant at a broader regional or ecological level, but a broader program of research is needed to test implications of these results. As reviewed in Part I, there has been virtually no extended work on old field vegetation in Kentucky, despite much general ecological theory having been developed from old fields in other states further north and east (KS, MN, WI, NJ, NC). It is reasonable to suppose that succession and other basic ecological processes could be rather different on the eutrophic soils of this Bluegrass region, as well as on similar soils elsewhere in the Ohio and central Mississippi Valleys (Campbell 2013).

There is a growing body of literature on the diverse effects of eutrophication or other pollution from roads on adjacent vegetation, especially in Europe (e.g., Pagotto et al. 2001, Blok 2005, Truscott et al. 2005, Legret & Pagotto 2006, Bignal et al. 2007, Coffin 2007, Thorpe & Harrison 2008). In the eastern U.S.A., some relevant recent research has been published (e.g., Councell et al. 2004, Trammell et al. 2011, Redling et al. 2013). More research would have practical relevance to agricultural systems and native vegetation. For example, staff at the University of Kentucky’s Soil Analysis Laboratory (D. Hunter, pers. comm.) are well aware that zinc and other obvious pollutants are often increased in soils close to roads. Moreover, zinc showed the greatest increase among edaphic attributes from back to front of this field (Table 1). However, there appears to have been little or no published research in eastern states that documents the spatial extent of pollution from roads into adjacent soils and vegetation.

Effects of deer and other herbivores on vegetation have of course received much scientific attention, especially in eastern North America. But again, there has been little published research on spatial patterns in effects at the scale of 100 m^2 plots. The more relevant studies from eastern states have already been noted in Part I (Campbell 2015); none of them have mapped out indications of herbivory over a whole field, as done here.

Old fields can often be convenient starting points for basic ecological research, and for restoration of native vegetation. The current paucity of research in old fields of Kentucky, and elsewhere in the Ohio Valley, reflects the rather low degree of useful interaction between academics and conservationists across this region.

Conclusions and Limitations of this Intitial Research

The ordination clearly reveals a predominant gradient (Axis One) that is related to decreasing overall soil fertility away from the road (e.g. Figure 4a), and to increasing evidence of herbivory from deer or other mammals (e.g., Figure 13). This pattern in soil chemistry (especially pH, Ca, Zn, organic matter, P and total N) is probably attributable to 200+ years of eutrophication from the road, U.S. 62, which is currently a busy two-lane highway with 1000s of vehicles per day. Elevation also declines away from the road, in general, but it does not appear to be a major independent factor influencing the vegetation (Figure 6). Although more detailed survey of soil and moisture conditions would be desirable, it is relevant that there is no general increase along Axis One in species with more hydric or riparian associations (Figure 16b). Soils within this field are all mapped as a typic argiudoll ("Loradale silt loam") in two phases: 2-6% slopes and 6-12% slopes (Odor et al. 1968). Initial inspection of soil samples revealed no significant differences in color or texture between front and back rows of the field.

Axis Two of the ordination is interpretable in terms of the somewhat independent effects of herbivores in this field. Effects of deer appear to be generally more intense at greater distances from the road, but this spatial trend is probably less consistent than the spatial trend in soil fertility (Table 1). In 2007 a diagonal zone of maximum influence from deer was indicated between road-crossings near the southwest corner and the back of the field (Figure 2 in Campbell 2015). Subsequent mapping of animal trails in January 2016 revealed a more complex pattern but still with greater densities in the southern part of the field as well as a general increase away from the road (Figure 12).

Evidence for a partially independent gradient related to herbivory can be summarized as follows; see Results for details.
1. There is slightly more overall segregation of the three 2007 zones along Axis Two than along Axis One: “central pathway” versus “transitional” versus “little influence” (Figure 8)
2. Recorded signs of browsing by deer in plots during 2007 (excepting data from the planted blue ash) are virtually absent from the lower left sector of the ordination (Figure 9a). These signs are strongly concentrated in a central zone of the ordination (Figure 9b).
3. Signs of recent browsing on the planted blue ash during 2007 increase from lower left to upper right sectors of the ordination, generally away from the road (Figure 10a, b).
4. Plots in the high mortality zone for planted blue ash (Campbell 2015) are concentrated in a central zone along Axis One, and the proportion of these plots increases into upper sectors of the ordination, with lower Axis Two scores (Figure 11a, b).
5. Animal trails mapped during January 2016 are more frequent in upper sectors of the ordination, with lower Axis Two scores, and there is no trend along Axis One (Figure 13a).
But despite these trends (1-5), “browsing-associated” herbaceous species of Part I tend to have higher Axis Two scores, and toxic herbaceous species higher Axis One scores (Figure 16f).

Although there is overall evidence of pattern here in herbivory, the potential for dynamic variation in effects of herbivores continues to present challenges for interpretation, and for plans to deepen the research. In Part I of this study (under Discussion), some complexities in results were used to suggest temporal shifts of effects by deer across the field. To these can be added the observation here that, in contrast to 2007 signs of browsing on planted blue ash being concentrated in the upper right sector of the ordination, evidence of damage in 2004-2006 was most frequent in the lower left sector (Figure 10a,b). Moreover, the varied patterns in different indicators of herbivory on the ordination, as just summarized, could be due to non-linear relationships and shifting spatial patterns. And the January 2016 survey of animal trails does suggest a more widespread, intense and complex pattern than existed in 2007.

Relevance to Spatial Dynamics of Vegetation-Herbivore Relationships

In order to deepen investigation of Vera's (2000) concept, together with its application for woodlands of the Ohio Valley (Campbell 2012), we will need eventually to develop predictive models of patterns in movements by animals between different patches of land, integrated with models of interactions between animals and different states of the vegetation (e.g., Bowler & Benton 2005). Patches of land could be imagined on the landscape in a cellular fashion, or in some other elemental form amenable to analysis. It may become useful, in a fundamental approach, to conceive of factors that influence optimal paths by large animals like deer in three simplified categories, as follows.

1. Factors determining the regular route between two or more areas, by the easiest path with little relationship to species in the traversed vegetation. Such factors would include the routines of daily rounds between areas with superior forage, physical constraints on travel (such as

cliffs, streams, fences or roads), and safe refuges from predators or other threats (such as nearby human residences). In and around the field studied here, it appears that the deer for many years have preferred a broad route across the field: from adjacent woods on the west side of U.S. 62; crossing the highway most easily near the southwest corner; avoiding the northwestern corner close to adjacent houses; and then moving into the old-growth woods on the east side (Campbell 2015). It is notable that the "central path" for deer had concentrations of less palatable woody species in 2007, and that signs of browsing were less in this zone than in the adjacent "transitional zone" (except on the planted blue ash; see Figure 2g in Part I).

2. Factors involved in active changes to the vegetation by the animals, with two important potential outcomes: (a) persistent browsing at a site that leads to increase in forage of lower quality; or (b) increase or maintenance of relatively nutritious plants at a site, as in the "grazing-lawn" concept (McNaughton 1984). In this field, spatial relationships suggest that relatively toxic or unpalatable woody plants are associated with more intense effects of deer on the vegetation. But there are some curvilinear relationships of browsing signs to overall forage quality (e.g., Figure 8 in Part I). Also, herbaceous plants may display an opposite trend on the ordination (Figure 16f). The "grazing-lawn" concept can be attractive where high densities of white-tailed deer appear to have created relatively homogeneous, graminoid-dominated ground-vegetation (Rooney 2009). But to what extent is such vegetation truly productive for deer, as opposed to smaller mammals or others? And were canebrakes with *Arundinaria gigantea* our major original "grazing-lawns" in this region, when maintained by overwintering mastodons?

3. Factors leading to avoidance of areas with less nutritious or more toxic vegetation, and to preference for areas with better forage. As just noted, some complexities in results of this study might be interpreted in terms of deer shifting their foraging away from areas with lower quality. As well as evidence summarized in Part I, it is reasonable to presume that deer reduce their

effects in areas where the lethal poison hemlock increases. That tall nitrophilous biennial has increased to become locally dominant in some parts of the Griffith Woods WMA, but within the Collection Field poison hemlock remains strongly concentrated along the front two rows.

Such ecological models will eventually aid understanding of evolutionary relationships, when comparing species with respect to their morphology, chemistry, nutrition, and responses to herbivory. In particular, most species with 'nitrogen-rich chemistry' are concentrated at left-central positions of the ordination, where soils have relatively high nitrogen content and the herbaceous species are generally taller (Figures 7 and 16g). This trend could be interpreted in terms of the generally repellant or toxic value of such chemistry (Table 2). The species include members of Apiaceae, Apocynaceae (sensu lato), Brassicaceae, Euphorbiaceae, Fabaceae (*Securigera*), Hypericaceae, Lamiaceae, *Phytolacca*, Poaceae (*Festuca*, *Sorghum*), and Solanaceae. They are all considered highly unpalatable or toxic for mammals, except perhaps the wild onion (*Allium vineale*)—which has marginal palatability. In contrast, the 'nitrogen-rich' species at right-central positions in the ordination are nitrogen-fixing Fabaceae that remain relatively palatable or at least non-toxic (*Desmodium, Medicago, Trifolium*).

Although these chemicals have generally been shown to defend against insects (e.g., Halkier & Gershenzon 2006), they can also contribute to interactions with mammals. For example, glucosinolates in Brassicaceae have strong taste (attractive or repellant) and potential medicinal value (at moderate doses) in mammals, contributing to a 'love-hate' relationship (e.g., Smith et al. 1991, Tripathi & Mishra 2007, Traka & Mithen 2009). Crown-vetch (*Securinega varia*) is an unusual legume, containing nitroproprionates that are toxic to small mammals (Smolenski et al. 1981). Tall fescue (*Festuca arundinacea*) often has a fungal endophyte, which leads to synthesis of alkaloids that are toxic to mammals, but the endophyte can be infrequent to absent on dry or infertile soils (Hall 2011). In the ordination here, tall fescue was most abundant in a central zone along Axis One, contrasting with the uniform distribution of bluegrass (Figure 15). It would be interesting to determine if tall fescue is more infected and unpalatable in this zone or at the eutrophic extreme of Axis One.

When comparing plant species, theoretical interpretation of chemical and morphological trends that appear related to herbivory can be elusive (e.g., Grubb 1992), and details are beyond the scope of this paper. Trends here are complex. The ordination of herbaceous species suggests that those with thorns or spines, as well as vines, are concentrated in a central zone between relatively unpalatable to toxic species at lower left and more palatable species at upper right (Figure 16f). However, overlays of woody species have a somewhat opposing pattern: more palatable species (especially white ash) are concentrated at lower positions, more thorny or spiny, toxic or unpalatable species (especially red cedar) are concentrated at upper right positions (Figure 18). This contrast might suggest some form of correlated shifts in herbivory between herbaceous versus woody vegetation. It is possible that smaller mammals than deer are also involved in these patterns. Many mysteries remain in “this old field”.

The “Herbivore Hypothesis” can be tested to a limited extent by deeper analysis of historical information, especially statistical associations between different species of witness trees in early land surveys (Campbell 1985). But further research here and elsewhere at Griffith Woods should explore the fundamental processes in interactions of vegetation and herbivores. As well as continued surveys of changing patterns in vegetation, together with related signs of herbivory, we need to conduct more experimental manipulations. In addition to using exclosures of varied size, arrangement and seasonal timing, it would be interesting to study effects of removing old fences that appear to reduce local movement of deer.

Relevance to Conservation, Restoration and Management

Part I provided a general discussion of issues related to management, and only a few notes are added here. The ordination has confirmed that species composition in this field, with relatively uniform topography and soils, has a surprising degree of variation related to soil fertility and to browsing by deer. However, much of this pattern appears to have originated from effects of the adjacent road on local eutrophication and on movements of deer. It would be useful to extend related research across the Griffith Woods WMA, at least with survey of carefully selected parameters, for estimating more general patterns in interactions of deer with the vegetation, and for gaining a better concept of what should be naturalistic processes.

It is important to determine the extent to which patterns of herbivory can enhance native biological diversity in the vegetation. Spatial variation among woody species within the Collection Field provides some initial encouragement: the gradient from white ash and associates to red cedar and associates does appear to be caused partially by patterns in browsing by deer. However, this gradient also involves invasive aliens, from relatively palatable species (especially bush-honeysuckle) to more browsing-resistant species (such as Bradford pear). It is likely that appropriate combinations of intense seasonal browsing (perhaps including livestock), plus further development of shade (excluding intolerant aliens like the pear), will be needed to help with reduction of aliens and other ecological restoration.

The potential ecological role for more labile components of soil fertility, especially nitrogen, deserves a lot more research. While phosphorous, calcium and other bases tend to have high natural levels across much of the central Bluegrass, it is likely that local variation in nitrogen was associated with significant variation in vegetation before settlement. There were remarkably few native nitrogen-fixing plants (Campbell 1985, 2013); the only common species appear to have been black locust, “peavine” (or hog-peanut, *Amphicarpaea*) and perhaps old-field tick-trefoil (*Desmodium perplexum*). Other common legumes here have little or no nitrogen-fixation: coffee-tree, honey-locust, senna (Bryan et al. 1996), and running buffalo-clover (Morris et al. 2002). Therefore, the thin woods and openings suitable for black-locust, peavine and tick-trefoil, which are relatively intolerant of shade, may have been important sources of nitrogen for the landscape, dispersed outwards by herbivores, especially large mammals. Based on patterns at Griffith Woods and elsewhere in the region, these plants may have been generally associated with moderate nitrogen level and moderate browsing intensity—the animals regularly removing nitrogen from associated vegetation, perhaps in a seasonal rhythm. Cane (*Arundinaria*), which appears to be a nitrogen-demanding plant like most bamboos, was probably associated with black locust and peavine (Campbell 2012b, 2013). The rhizomatous spread of cane could have been fueled by nitrogen-fixation in these legumes and by nitrogen transported through larger animals, especially along trails. But herbivory may, alternatively, reduce nitrate in some soils if unpalatable or toxic plants are selected and if animals move elsewhere in diurnal to annual cycles (e.g., Ritchie et al. 1998).

The potential value of fire in restoration of Bluegrass Woodland remains unclear. One aspect that deserves much deeper research is the potential interaction of fire with nitrogen dynamics. Fire generally increases soluble nitrate levels in soils, which can then be flushed by rain or snow (Wan et al. 2001). Thus, frequent prescribed fire to maintain openings, especially attempts to burn back clonal thickets of black-locust, could artificially reduce nitrogen levels in the vegetation. And black-locust is notoriously difficult to reduce with fire alone, since it resprouts with great vigor. Moreover, fire does not appear to have been a significant factor in this region before settlement (Campbell 2013, McEwan & McCarthy 2008).

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The Acknowledgements of Part I also apply here, and will not be repeated. But, in addition, I am most grateful to Kim Daehyun (Department of Geography, University of Kentucky) for providing positive feedback and cooperative plans for future research along these lines. Also, Diane Hunter and her staff (Soil Analysis Laboratory, University of Kentucky) provided a friendly rapid response to the request for processing soil samples.

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