

# Parthenogenesis and bisexuality in the millipede, *Nemasoma varicorne* C. L. Koch, 1847 (Diplopoda: Nemasomatidae). II. Distribution, substrate, and abundance of the bisexual and thelytokous forms in some Danish forests.

HENRIK ENGHOFF

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A large number of *Nemasoma varicorne* was collected in eight SE Danish forests. The distribution of the bisexual and thelytokous forms of the species within each forest was mapped. Although several distinct distribution patterns were found, it could not be decided which factor(s) was/were responsible for the patterns. The kind of tree on which the samples were taken was recorded and the two forms were compared with respect to relative frequency on beech (*Fagus silvatica*) versus other species of trees. A faint tendency for the bisexual form to be relatively more frequent/numerous on beech was found. The sample size, measured as number of females collected per tree trunk/stump, was similar for the two forms within each forest. As, however, the bisexual form has a sex ratio of c. 1 ♂ : 1 ♀, this indicates a larger mean population size (individuals per tree trunk/stump) in the bisexual form.

Henrik Enghoff, Zoologisk Museum, Universitetsparken 15, DK-2100 København Ø, Denmark.

## Introduction

*Nemasoma varicorne* C. L. Koch, 1847, occurs in a bisexual and a thelytokous (female-producing parthenogenetic) form. The sex ratio of the bisexual form is c. 1♂:1♀. In the thelytokous form, males are extremely rare and possibly non-functional. Females of the two forms are readily distinguished: Bisexual females (B-♀♀) have a pear-shaped receptaculum seminis with a coiled stalk. Thelytokous females (T-♀♀) have a vestigial receptaculum, or none at all.

The bisexual form has a large distribution area covering most of Central Europe including south-east Denmark. The thelytokous form has an almost ring-shaped distribution area to the West, North, and East of the area of the bisexual form, including all of Denmark. South-east Denmark thus constitutes a zone of overlap between the two forms (Enghoff, 1976b).

*N. varicorne* lives exclusively under the bark of dead tree trunks and stumps. The development from egg to adult takes two years in nature (Brookes, 1974).

Previous studies (Enghoff, 1976a, b) suggested competitive interactions between the two forms. In the present paper, extensive sampling and mapping of the two forms in the zone of overlap, undertaken in order to elucidate possible competitive interactions, is reported on.

## Methods

Eight forests, all in north-east Sjælland, were investigated (Fig. 1). One forest was investigated twice, with an interval of two years. The investigated area lies within the zone of overlap, close to the north-east distribution limit of the bisexual form.

Collection and examination of specimens were carried out as described previously (Enghoff, 1976b). A sample includes the specimens collected on one tree trunk or stump. The terms “bisexual sample”, “thelytokous sample”, and “mixed sample” are used instead of the longer expressions “sample with B-♀♀, but no T-♀♀” etc.



Fig. 1. The geographic position of the investigated forests in north-east Sjælland. Scale: 10 kms. 1. Jægersborg Hegn, 2. Hareskovene, 3. Rude Skov, 4. Folehaven, 5. Store Dyrehave, 6. Teglstrup Hegn, 7. Danstrup-Krogenberg Hegn, 8. Gammel Grønholt Vang. Inset: Map of Denmark with the study area indicated.

## Results

Figures 2–6 show the distribution of the two forms in Jægersborg Hegn, Hareskovene, Teglstrup Hegn, Folehaven, and Store Dyrehave. In Rude Skov and Gammel Grønholt Vang, the very few samples of one form did not show a distinct distribution pattern. No B-♀♀ were found in Danstrup-Krogenberg Hegn.

The numbers of different kinds of samples taken in each forest are shown in Table 1. The numbers of different kinds of individuals (♂♂, B-♀♀, T-♀♀) taken in different kinds of samples (bisexual, thelytokous, mixed) in Jægersborg Hegn, 1975, are shown in Table 2. Similar tables were constructed for the other forests but are not reproduced. The total number of females in Table 2 is larger than the sum of B-♀♀ and T-♀♀. This is due to the fact that some females could not be assigned to form because of un-

developed genitalia (subadult females) or unsuccessful preparation.

A total of 2396 ♂♂ and 3384 ♀♀ was examined.

In Tables 1 and 2, the material has been divided into samples resp. females found on *Fagus silvatica* (beech) and samples resp. females found on other trees. A total of 367 samples was taken on *Fagus*, 70 samples on twelve other tree genera. The most important "others" were *Quercus* spp. (oak) with 21 samples and *Fraxinus excelsior* (ash) with 16. Other tree genera yielded 1–7 samples each.

Assuming that the genus of "host" tree is an important quality of the substrate of *N. varicornis* it was tested whether a statistical difference between the two forms in occurrence on *Fagus* versus "others" was present. Four tests were made for each forest (where possible), employing the data in different ways (Table 3). All tests

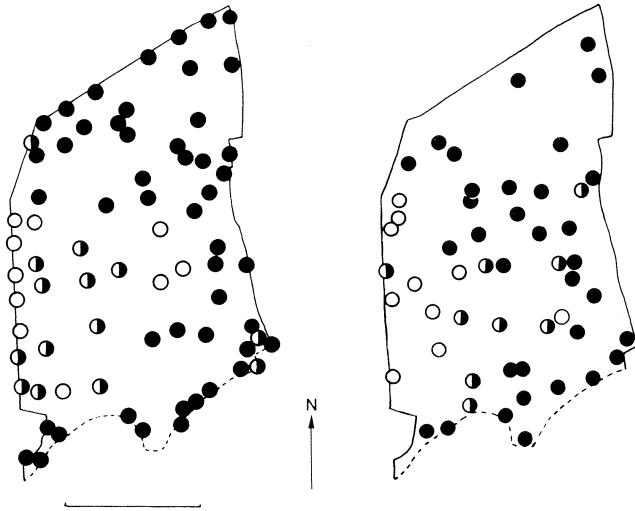


Fig. 2. Distribution of the two forms of *N. varicornis* in Jægersborg Hegn. Left: 1975, right: 1977. Each circle represents one sample. Filled circles: bisexual samples, open circles: thelytokous samples, half-filled circles: mixed samples. Scale: 1 km. Broken line indicates continuity with not investigated forest area.

were in the form of  $2 \times 2$  contingency tables like the example shown in Table 4. A mixed sample is regarded as a bisexual sample plus a thelytokous sample. Therefore, the mixed samples enter the table twice.

Although the difference is not significant in the above example, proportionally more bisexual (incl. mixed) than thelytokous (incl. mixed) samples were found on *Fagus* ( $49:13=3.77 > 17:7=2.43$ ). Hence the "+" in Table 3.

A division of "others" into single tree genera was not practicable due to the small numbers. Some forests do not appear in Table 3 due to too little material of one form.

A comparison between pure bisexual samples

and pure thelytokous samples with regard to sample size revealed no significant differences between the mean number of *females* per sample of each form within one forest. In Jægersborg Hegn, 1975, for example, the means  $\pm$  S. E. were  $5.58 \pm 0.87$  B-♀♀ per sample and  $4.40 \pm 1.75$  T-♀♀ per sample.

## Discussion

### Distribution

There are two levels of distribution patterns inside the investigated area: within-forest patterns and a between-forests pattern.

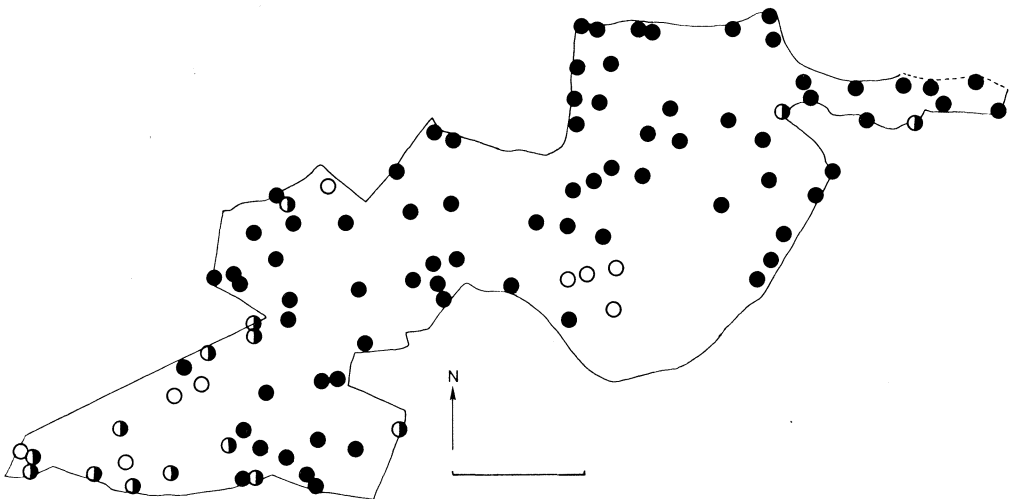


Fig. 3. Distribution of the two forms of *N. varicornis* in Hareskovene, 1975. Symbols as in Fig. 1.

Table 1. Numbers of samples of *N. varicornis* collected in eight Danish forests 1975–1977.

	No. of samples	Bisexual			Mixed			Thelytokous		
		total	<i>Fagus</i>	others	total	<i>Fagus</i>	others	total	<i>Fagus</i>	others
Jægersborg Hegn, 1975	72	48	39	9	14	10	4	10	7	3
Jægersborg Hegn, 1977	53	34	32	2	9	9	0	10	6	4
Hareskovene, 1975	100	76	67	9	15	12	3	9	8	1
Rude Skov, 1976	36	33	22	11	3	3	0	0	0	0
Folehaven, 1975	35	21	15	6	9	6	3	5	2	3
Store Dyrehave, 1976	45	23	21	2	14	13	1	8	8	0
Teglstrup Hegn, 1975	52	0	0	0	9	9	0	43	41	2
Danstrup-Krogenberg Hegn, 1977	30	0	0	0	0	0	0	30	25	5
Gammel Grønholt Vang, 1977	14	0	0	0	4	4	0	10	8	2

Within-forest patterns:

Three (not mutually exclusive) hypotheses may be advanced to explain the distribution pattern within each forest:

*Immigration:* The limited distribution of one form in some forests is due to recent immigration(s) to the forest. Absence is correspondingly due to lacking or failing immigration.

This hypothesis implies a change of the distribution pattern with time. The similarity of the distribution patterns in Jægersborg Hegn in 1975 and 1977 (Fig. 2) contradicts the immigration hypothesis. However, the mode and rate of dis-

persal in *N. varicornis* is unknown (Haacker, 1968 suggests wind dispersal but there is no direct evidence of this). Therefore, the immigration hypothesis remains a possible explanation.

*Competition:* The distribution of the two forms within a forest is governed by the distribution of resources for which the two forms are competing.

Competition might be reflected in the local distribution patterns. For instance, the thelytokous form might prevail in marginal areas due to its superior dispersal abilities. The bisexual form, on the other hand, might prevail in central parts of the forest due to some other factor (analogy with the large-scale distribution of the two forms). However, no correlation between distribution and any of the following environmental factors could be detected: species

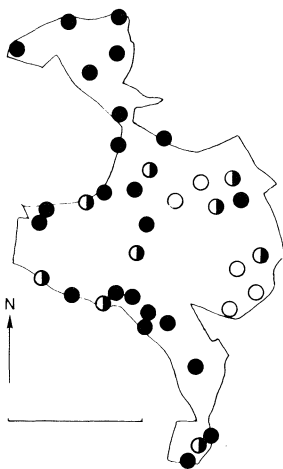


Fig. 4. Distribution of the two forms of *N. varicornis* in Folehaven, 1975. Symbols as in fig. 1.

Table 2. Adult and subadult individuals of *N. varicornis* found in Jægersborg Hegn, 1975 (538 ♂♂, 537 ♀♀).

	In pure samples	In mixed samples	Total
Bisexual ♀♀			
on <i>Fagus</i>	224	38	
on others	44	13	
total	268	51	319
Thelytokous ♀♀			
on <i>Fagus</i>	29	96	
on others	15	16	
total	44	112	156

Table 3. Substrate difference between bisexual and thelytokous *N. varicornis*. (+ : proportionally more bisexual samples resp. females than thelytokous do. on *Fagus*; - : proportionally fewer bisexual samples resp. females than thelytokous do. on *Fagus*; X: P < 0.05, XX: P < 0.01, XXX: P < 0.005).

	All samples	Pure samples	Females from all samples	Females from pure samples
Jægersborg Hegn, 1975 .....	+	+	+	+ XX
Jægersborg Hegn, 1977 .....	+ X	+ XX	+ XXX	+ XXX
Hareskovene, 1975 .....	-	-	+	-
Folehaven, 1975 .....	+	+	+	+ XXX
Store Dyrehave, 1976 .....	-	-	+	- X
Teglstrup Hegn, 1975 .....	+	-	+	-
Eight forests, pooled .....	-	-	+ XX	+ XXX

and age composition of the tree vegetation at the sampling site (cf. below: substrate); distance from forest edge; altitude (only a few meters' difference within the investigated area); frequency of tree felling (much felling makes more habitat available to *N. varicornis* as stumps and odd pieces of trunks are usually left behind by the foresters).

Other factors not analysed here, e.g. climatic factors, could be important; therefore, the competition hypothesis also remains a possible explanation for the distribution patterns.

*Polyphyly*: The localized occurrence of the thelytokous form in some forests is due to multiple arisal of independent thelytokous offshoots from bisexual ancestors.

The degree of polyphyly of the thelytokous form is at present unknown (Enghoff, 1976b).

Between-forests pattern:

In the southern forests (Jægersborg Hegn, Hareskovene, Rude Skov), the bisexual form occurs all over but the thelytokous form has a limited within-forest distribution. Farther north (Folehaven, Store Dyrehaven), the dominance

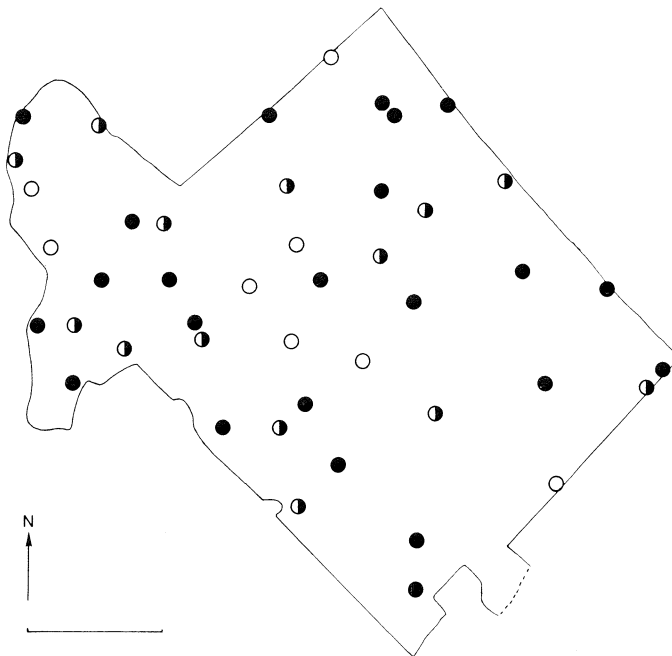


Fig. 5. Distribution of the two forms of *N. varicornis* in Store Dyrehave, 1976. Symbols as in Fig. 1.

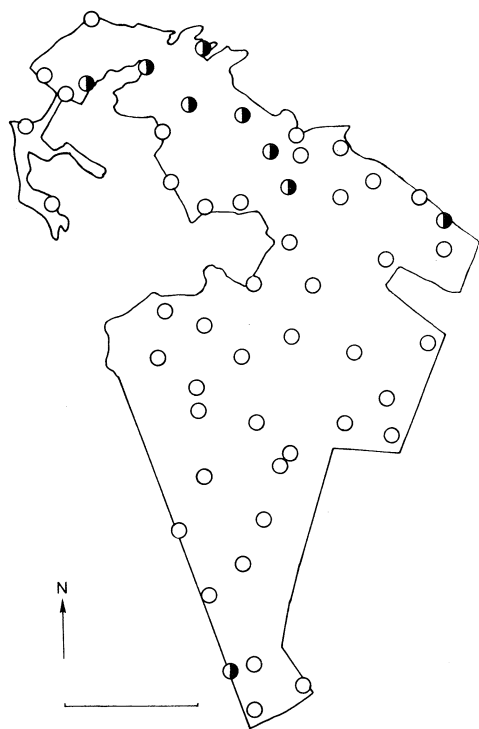


Fig. 6. Distribution of the two forms of *N. varicorne* in Teglstруп Hegn, 1975. Symbols as in Fig. 1.

of the bisexual form is less striking, the two forms being more or less evenly distributed throughout each forest. In the northernmost forests (Teglstруп Hegn, Danstrup-Krogenberg Hegn, Gammel Grønholt Vang), the thelytokous form occurs all over, but the bisexual form has a limited distribution, or is absent.

This geographical trend suggests a gradual decrease in frequency of the bisexual form towards the limit of its total distribution, which is just north of the investigated area.

#### Substrate

The majority of Danish samples of *N. varicorne* has been collected on *Fagus silvatica* (beech), both in the present and in earlier investigations. This apparent preference for *Fagus* is not universal. In other parts of Europe, other tree genera seem to be preferred. However, the frequency of the different trees *per se* would, among other factors, tend to bias the "preference" obtained by simple collecting (Enghoff, 1976b).

In spite of this reservation, the two forms may compete for a substrate resource, and the kind of tree may be an important substrate

parameter. This type of competition could result in one form being relatively more frequent on certain kinds of trees, and the other form, on other kinds.

Few significant within-forest differences in substrate were found (Table 3). Most of the significant differences indicate that the bisexual form is relatively more frequent on *Fagus* than the thelytokous form. Pooling of all eight forests gives a significant result pointing in the same direction, when number of females is considered. The preponderance of "+"s in Table 3 is a further hint of a tendency for the bisexual form to be relatively more numerous (number of females), and perhaps also relatively more frequent (number of samples) on *Fagus* than the thelytokous form. Considering the very rough division between *Fagus* and "others", this tendency is noteworthy.

It is not possible to correlate the substrate differences with geographic distribution, neither within nor between forests. For instance, *Fagus* is not particularly rare in the part of Jæggersborg Hegn where the thelytokous form occurs. The "-"s in Table 3 refer to one southern (Hareskovene) and one intermediate (Store Dyrehave) forest.

#### Abundance

Although sample size is, in the present case, only a very rough measure of population size, the similarity between the two forms in mean no. of females per sample deserves a short comment. The sex ratio of the bisexual form being c. 1 ♂:1 ♀, this finding indicates that, on the average, a bisexual population inhabiting a tree-trunk (or stump) is about twice as large as a corresponding thelytokous population. This difference may be related to the fact that a bisexual female produces, on the average, about twice as many eggs as a thelytokous female (Enghoff, 1976b).

Table 4. Example of the 2 × 2 contingency tables used in testing substrate differences between the two forms. Jæggersborg Hegn, 1975, all samples.

	Bisexual + mixed	Thelytokous + mixed
<i>Fagus</i>	49	17
Others	13	7

$\chi^2 = 0.634, P > 0.1$

## Conclusion

The kind and degree of competitive interactions between bisexual and thelytokous *N. varicornis* remains unsettled in spite of the collecting and mapping project reported on here. Although distinct local distribution patterns were found, they may be explained in several ways. Further research on the dynamics of the distribution patterns and on the genetical interactions between the two forms may restrict the number of possible explanations.

The faint indication of a difference in substrate between the two forms calls for further investigation as does the observation that the bisexual form may have a larger mean population size (individuals per trunk) than the thelytokous form. Unfortunately, the patchy occurrence of *N. varicornis* on dead tree trunks and stumps and its two-year life cycle are important obstacles to quantitative field sampling programmes resp. laboratory experiments.

## Acknowledgements

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## References

- Brookes, C. H., 1974: The life cycle of *Proteroiulus fuscus* (Am Stein) and *Isobates varicornis* (Koch) with notes on the anamorphosis of Blaniulidae. – Symp. zool. Soc. Lond. 32: 485–501.
- Enghoff, H., 1976a: Competition in connection with geographic parthenogenesis. Theory and examples, including some original observations on *Nemasoma varicornis* C. L. Koch (Diplopoda: Blaniulidae). – J. nat. Hist. 10: 475–479.
- , 1976b: Parthenogenesis and bisexuality in the millipede, *Nemasoma varicornis* C. L. Koch, 1847 (Diplopoda: Blaniulidae). Morphological, ecological, and biogeographical aspects. – Vidensk. Meddr dansk naturh. Foren. 139: 21–59.
- Haacker, U., 1968: Deskriptive, experimentelle und vergleichende Untersuchungen zur Autökologie rhein-mainischer Diplopoden. – Oecologia (Berl.) 1: 87–129.

## Sammendrag

Parthenogenese og biseksualitet hos tusindbenet *Nemasoma varicornis* C. L. Koch, 1847 (Diplopoda: Nemasomatidae). II. De biseksuelle og thelytoke formers udbredelse, substrat og talrighed i nogle danske skove.

Tusindbenet *Nemasoma varicornis* findes i en biseksuel og en thelytok (hun-producerende parthenogenetisk) form. Tidligere undersøgelser har vist, at de to former har stort set ikke-overlappende udbredelsesarealer. Det sydøstlige Danmark udgør dog en overlappingszone.

Med henblik på at belyse eventuel konkurrence mellem de to former er der blevet indsamlet et stort materiale i otte nordøstsjællandske skove (Fig. 1), og de to formers lokaludbredelse i skovene er blevet kortlagt.

Figurerne 2–6 viser udbredelsen af de to former i fem af skovene. I de øvrige skove blev den ene form kun fundet nogle få gange eller slet ikke. Der er tydelige udbredelsesforskelle i nogle af skovene, men fortolkningen af disse er usikker. På indeværende tidspunkt må både indvandring, konkurrence og polyfyletisk oprindelse af den thelytoke form anses for mulige forklaringer på udbredelserne. Det er påfaldende, at den biseksuelle form er talmæssigt mest dominerende i de sydligste skove (Jægersborg Hegn, Hareskovene, Rude Skov), mens den thelytoke form er dominerende i de nordligste skove (Teglstrup Hegn, Danstrup-Krogenberg Hegn, Gammel Grønholt Vang). Måske afspejler dette en gradvis aftagen i hyppigheden af den biseksuelle form i retning mod dens udbredelsesgrænse, som ligger lige nord for undersøgelsesområdet.

Materialet er blevet opdelt i prøver taget på bøg og prøver taget på andre træarter. Der er en svag tendens til, at den biseksuelle form er forholdsvis hyppigere på bøg end den thelytoke (Tabel 3).

Prøvestørrelsen (antal individer pr. træstamme eller stub) udviser ikke statistisk signifikante forskelle mellem de to former, når man kun tæller hunnerne. Men da den biseksuelle form består af cirka lige mange hanner og hunner, antyder dette, at den biseksuelle form har større populationer pr. stamme/stub end den thelytoke form. Det er muligt, at denne forskel skyldes større ægproduktion hos den biseksuelle form (tidligere undersøgelser viste, at en biseksuel hun producerer cirka dobbelt så mange æg som en thelytok hun).

Alt i alt må det konkluderes, at arten og omfanget af en eventuel konkurrence mellem de to former endnu er ukendt. Det her fremlagte materiale giver kun antydninger. Måske kan yderligere felt-indsamlinger, suppleret med laboratorieeksperimenter, belyse problemet nærmere.