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Introduction into arrhenotokous genetics

This morning we will hear a series of discourses and various views on genetic investigations relative to spider mites. It is notable that all these subjects deal with insecticide resistance, or are grafted upon resistance. The primary purpose of such investigations is practical: Insight in resistance make-up, causality of related characters etc. But I believe that the yield of researches dealing with the genetics of these mites has even a broader horizon: also for fundamental questions concerning evolutionary dynamics these studies are imperative. The
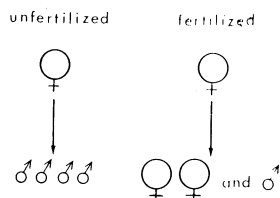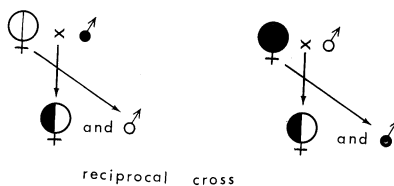

Fig. 1

reciprocal cross

Fig. 2

importance arises from the peculiar mode of reproduction. I guess that when a geneticist was asked before the war, which pests would be the champions of resistance in future, spider mites were not enlisted. And yet we know to-day that spider mites make a good chance to be first in this respect.

The sex-determination of spider mites is from a haplo-diploid mechanism, since males develop from unfertilized (haploid) eggs, and females from fertilized (diploid) ones (fig. 1).

It is my experience that this manner of reproduction causes short circuits in the minds of many of us. Therefore, before we comment with the program, I would like to infiltrate you with some of the

peculiarities from arrhenotokous genetics. The first example I want to show you deals with reciprocal crosses (fig. 2).

You see here that reciprocal crosses are not identical with respect to the male progeny, as the males will have the genotype of the mother. This is especially important to realize when resistance to ovicides enters
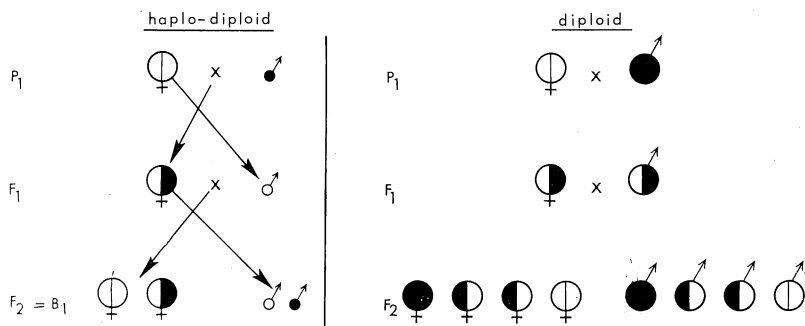
Fig. 3

the picture. Eggs cannot be « sexed » and in reciprocal progeny the haploid fraction of eggs will make the difference in outcome. For adulticides this is not so, because we can sex the progeny and make our toxicological determinations with diploid animals.
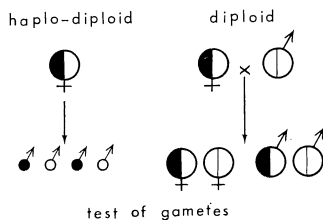
Fig. 4

Continuing a mendelian cross to the F_2, we must realize that here we make a backcross with the ancestral female genotype. In fact we make a test of gametes. Therefore, in the F_2 of spider mites we do not see all genotypical combinations as should be exposed in the F_2 of a « normal » cross (fig. 3).

The use of unfertilized females for gametic testing seems astute, because you will get gametes walking on a leaf (fig. 4).

However, it is a pity that most of the toxicological tests on males are not useable, except in the use of ovicides.

Another point I want to stress deals with the observation that in reciprocal crosses the descendants incline genotypically to the mother. It is not necessary to make complicated theories about this, since the genetic contribution in reciprocal crosses is different in quantitative respect. It is not 50 %-50 %, but 67 %-33 % (fig. 5).
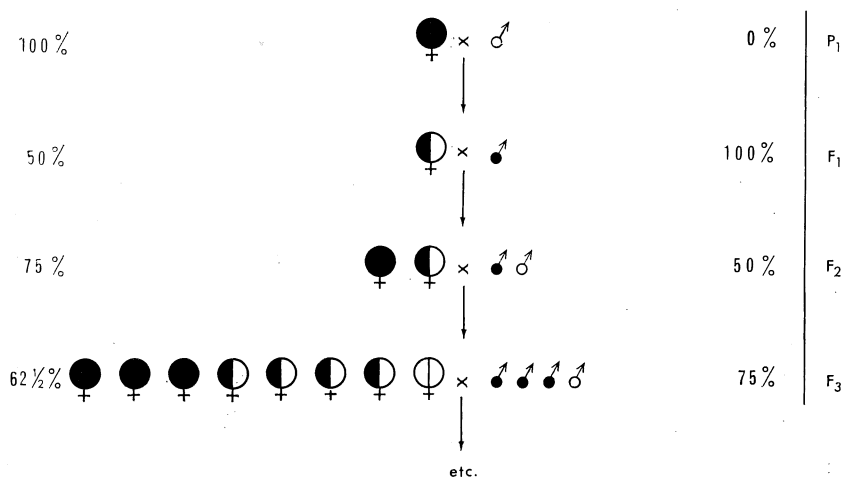
Fig. 5

It is good to emphasize that, when crossing different genotypes, the frequencies go up and down in subsequent generations and come to an equilibrium. We have the same situation as we know from sex-linked factors in diploid species.

With respect to insecticide selection experiments it is also impor-tant to recognize that after relaxation of selection this criss-cross oscillation of frequencies of R-genes can come into the picture.

Also in quantitative genetics arrhenotoky dictates different rules. Especially the role of deleterious alleles seems problematic, because of the male haploidy. The different factors, however, will be exposed in an apart lecture.

SUMMARY

In order to prevent short circuits in our minds during this section some peculiarities of arrhenotokous genetics are considered:

1) Sex-determination of spider mites is from a haplo-diploid mechanism since males develop from unfertilized (haploid) eggs, and females from fertilized (diploid) ones.

2) Reciprocal crosses, therefore, are not identical with respect to the male progeny. The F_1-males will have the genotype of their mother.

3) Gametic analysis is very simple with respect to resistance to ovicides by using unfertilized hybrid females.

4) In a cross the effect from the maternal genetic component is greater than that from the male.

5) Lethals, semilethals and subvitals are in a special position in spider mites, because of the male haploidy. They cannot be concealed into the hemizygous males. This leads to fixation of favorable alleles and genetic loss of unfavorable ones.

RIASSUNTO

Dall'esame di alcune caratteristiche della genetica arrenotoca dei Tetranichidi, l'A. formula le seguenti considerazioni:

1) la determinazione del sesso avviene per mezzo di un meccanismo aplo-diploide, poiché i maschi si sviluppano da uova non fecondate (aploidi) e le femmine da quelle fecondate (diploidi);

2) gli incroci reciproci non sono perciò identici per quanto si riferisce alla progenie maschile. I maschi F_1 hanno il genotipo della madre;

3) l'analisi gametica circa la resistenza agli ovicidi è molto semplice se si impiegano femmine ibride non fecondate;

4) nell'incrocio l'effetto della componente genetica materna è superiore a quello della componente paterna;

5) i caratteri letali, semiletali e subvitali occupano una posizione particolare nei Tetranichidi, a causa dell'aploidia maschile. Essi non possono rimanere celati nei maschi emizigoti. Ciò conduce alla fissazione di alleli favorevoli ed alla perdita genetica di quelli sfavorevoli.

DISCUSSION

MATHYS: This question is not in the line of what has been presented in such a striking way by Dr. Helle. I would like to know the significance of spanandry in *Bryobia* populations.

HELLE: Since nothing is known about the sex-determination with respect to these species, karyological study must be initiated.

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Karyotypes of Tetranychids

Observation of « giant » males in a strain of *Tetranychus urticae* Koch initiated a karyological research. Karyotypes of four *Tetranychus*-species were studied. Eggs were fixed in Na-citrate 1 % for 1-2 min; a squashing technique was used with 2-3 % aceto-orceïn stain.

A basic number of n = 3 chromosomes was observed for *T. urticae*, *T. cinnabarinus*, *T. hydrangeae*, and *T. pacificus*. (cfr. SCHRADER, 1923).

Fig. 1 - Metacentric metaphase chromosomes from a diploid egg of *T. urticae* Koch.

L'osservazione di maschi « giganti » in una stirpe di *Tetranychus urticae* ha dato inizio ad una serie di ricerche cariologiche, con le quali sono stati presi in esame i cariotipi di quattro specie di *Tetranychus*.

Le uova, sulle quali è applicata una particolare tecnica di schiacciamento, sono fissate in citrato sodico all'12% per 1-2 minuti; come colorante viene impiegata l'aceto-orceina al 2-3 %.

In tutte quattro le specie (*T. urticae, T. cinnabarinus, T. hydrangenae* e *T. pacificus*) il numero base dei cromosomi è n = 3.

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Population genetics of arrhenotokous mites

Most of us grow up in a *Drosophila* tradition in quantitative genetics. Yet, this position seems to be tied too much to a particular animal, as soon as we deal with spider mites.

Population genetics of spider mites is set forth in a peculiar framework. Since the males of spider mites are haploid, it is easy to comprehend that it must have its consequences in a quantitative genetical respect. But the area of these consequences are difficult to estimate. In this paper I want to illustrate you the impact of male haploidy for the genetic variability of a population.

Populations of cross-fertilizing diploid species can be loaded up with recessive variants, even when deleterious. These variants come about by mutations. The possibility of storage is realized by the diploidy of both sexes: recessive mutants will occur in heterozygous conditions and are sheltered from natural selection. They are subject to dispersive processes, because of their scarcity, but not to natural selection. At any time their frequency in a population is increased and homozygous genotypes will occur. At this moment natural selection enters into the picture. A equilibrium condition will be obtained between mutation pressure and natural selection. The equilibrium level approximates the equation $q = \sqrt{\dfrac{u}{s}}$, in which $q =$ equilibrium frequency of the recessive mutant, $u =$ mutation rate, and $s =$ selection coefficient, acting against the mutant (DOBZHANSKY, 1951; FALCONER, 1961). This equation can be considered as a projection of the genetic storage potencies of a population.

In spider mites, storage of deleterious variants must be more restricted, because of the immediate interference of natural selection.

In the hemizygous males a recessive with low adaptive value is exposed to selection, therefore the storage is impeded by a leak.

But what is the quantitative outcome of this leak? For arrhenotokous mites is calculated (see appendix of the end of the paper), that equilibrium between mutation and selection will obtained at:

$$q = \frac{u}{s} (R + 1) \quad \text{(approx.)}$$

in which R represents the sex-ratio.

A comparison with the preceding equation illustrates the important effect of male haploidy. In bisexual zygogenetic animals a lethal ($s = 1$) at a mutation rate $u = 10^{-6}$ can increase to 10^{-3}. In an arrhenotokous mite with a sex-ratio of 3) equilibrium is already reached at a frequency of $4 \cdot 10^{-6}$. It is revealing to substitute different values for s. For a population with limited size storage of semilethals and subvitals is beyond scope. Only variants whose adaptive values are close to unit, present any change for storage.

However, from data about inbreeding depression (HELLE, 1965) and from consecutive selection experiments it appeared that the concealed genetic variability in spider mite populations is greater than one would expect.

A high mutation rate would not be peculiar as one of the explanations. It has been stated for *Drosophila* that mutability is often under genetic control. Since for spider mite populations a high mutability would have selective advantages, it supports the hypothesis. But there is an arithmetic drawback against this mutability hypothesis: a higher mutation rate u results in an insufficient increase of q.

A hypothesis more palatable to me is the existance of sex-limited variants. Variants which affect the females, but not the males, have an excellent opportunity to be stored in an arrhenotokous population. In fact, the equilibrium level of such variants is even higher as compared with normal variants in species with diploid sexes. We find:

$$q = \sqrt{\frac{u\,(R + 1)}{s \cdot R}} \quad \text{(approx.)}$$

There is a fine example of the existance of sex-limited variants in spider mites.

The incidence of diapause in *Tetranychus urticae* is induced by photoperiod and temperature. There is a wide variation between

populations from different geographical origin with respect to their diapause pattern. BONDARENKO and KUAN (1958) demonstrated that we even can distinguish photoperiodical races in *T. urticae*.

In our laboratory we made several selections in two different strains, in order to chance the critical combination of photoperiod and temperature. In both strains the selection for nin-diapausing mites resulted in an alteration of the diapause pattern.
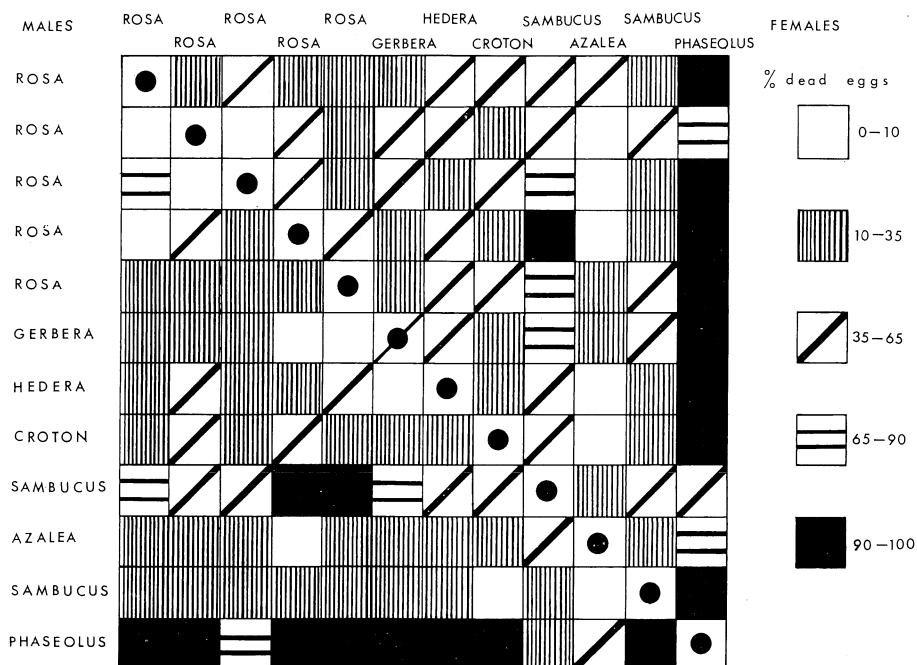

Fig. 1 - Percentages of unviable eggs produced by hybrids from mass crosses between populations from different hostplants.

It must be emphasized that the existance and maintenance of variability for this character is of paramount importance for a population, since genetic plasticity guards against unpredictable onset of winter. Since in *T. urticae* only females enter into diapause (after fertilization), the whole character is sex-limited. Fixation or loss of any allele is thwarted, despite of the haplo-diploid reproductive system.

In *Panonychus ulmi* the eggs hibernate, haploid and diploid both.

But also in this species the situation is genetically similar, because the diapause induction affects only the females.

So far my considerations have been related to the intrapopulational variability. But the composition of the adaptibility of arrhenotokous mites would be incomplete if the interpopulational variability was excluded. Previously mentioned, rare mutants (and also chromosomal alterations) are subject to dispersive processes. As long as these mutants occur at very low frequency, the chance on extinction is considerable. If a mutant has a good adaptive value, the haploidy of the males enables natural selection to dash to the rescue. In spider mites the interaction between mutation and selection operates promptly after their origin, also in case of recessivity. The result will be a higher mutational yield for arrhenotokous mites, as opposed to animals with diploid sexes. Since fixation of a favorable mutant, and loss of the ancestral allele is also a peculiarity of arrhenotoky, genetic differentiation and divergence of populations will occur. I think that the mosaic of genetic incompatibilities between different populations of spider mites can be explained along these lines (HELLE and PIETERSE, 1965). It is quite conceivable that isolated populations will drift off rather soon, because of their high reproductive potential in addition to the mentioned aspect of male haploidy (fig. 1).

Upon considering these peculiarities arising from haploid parthenogenesis we arrive at an adaptive pattern for spider mites, in which adaptedness seems to arise from interpopulational selection rather than from intrapopulational variability. The opportunistic effectiviness of such a system can be compared with an investment trust, in which the risks are scattered in numerous enterprises. In fact, the situation in spider mites remind us of a description of WRIGHT (1932) of the most favorable adaptive type for progressive evolution.

But I think our knowledge of the genetic variability in spider mites is too fragmentary for the presentation of any structure today. The primary intention of this paper was only to arouse apathatic viewpoints to the possibilities and impossibilities within the arrhenotokous framework of spider mites, e.g. the odd position of lethals, semilethals and subvitals.

APPENDIX

(deduction from equation for equilibrium condition between mutation and selection for an arrhenotokous species).

Suppose gene A mutates to a at a mutation rate u, we will get three female genotypes, and two male genotypes. The selection effect is different in both sexes, therefore the effect of selection is calculated separately for females and males respectively.

genotypes	AA	Aa	aa	A	a
frequencies ...	$(1-q)^2$	$2q(1-q)$	q^2	$1-q$	q
fitness	1	1	$1-s$	1	$1-s$
frequencies after selection	$(1-q)^2$	$2q(1-q)$	$q^2(1-s)$	$1-q$	$q(1-s)$

Decrement in females:

$$-\Delta q = q - \frac{q^2(1-s)+q(1-q)}{1-sq^2}$$

$$-\Delta q = \frac{sq^2(1-q)}{1-sq^2}$$

Decrement in males:

$$-\Delta q = q - \frac{q(1-s)}{1-sq}$$

$$-\Delta q = \frac{sq(1-q)}{1-sq}$$

Here I have modified both denominators upwards to 1, so that the expression become:

$$-\Delta q = sq^2(1-q) \qquad \text{and} \qquad -\Delta q = sq(1-q)$$

The average decrement in the population must be corrected with the sex-ratio coefficient R ($=$ females/males).

$$\overline{-\Delta q} = \frac{R \cdot sq^2(1-q)+sq(1-q)}{R+1}$$

Now, for equilibrium condition the increment of a by mutation must be equal to the decrement of a by selection:

$$u(1-q) = \frac{R \cdot sq^2(1-q)+sq(1-q)}{R+1}$$

$$u = \frac{R \cdot sq^2+sq}{R+1}$$

$R \cdot sq^2$ is very small in relation to sq, and can be neglected:

$$u = \frac{sq}{R+1} \qquad\qquad q = \frac{u(R+1)}{s}$$

SUMMARY

Adaptational dyinamics of arrhenotokous species is set forth in an peculiar framework, that differs in some principal aspects from that of species with diploid sexes. Theoretically we can deduce from male haploidy:
1) *Strong tendency to homozygosity.*

Populations of cross-fertilizing diploid species can be loaded with recessive variants, even when deleterious. The possibility of storage is realized by the fact that at low frequencies recessive mutants occur mainly in heterozygous condition, and are out of grasp of natural selection. It is assumed that this store of concealed genetic variability is important for the adaptive potencies of a population.

On the other hand, in arrhenotokous mites the possibilities for a mutational genetic load must be more restricted. Here, deleterious variants, whether recessive or dominant, remain even at low frequency under the direct influence of natural selection. The presence of hemizygous males gives deleterious variants no opportuny to hide. Therefore, a strong tendency to fixation of favorable and to loss of unfavorable alleles will exist, and consequently positive limitations in genetic variability are expected.

2) *High mutational yield.*

Favorable mutations (and also chromosomal alterations) in arrhenotokous species are less liable to dispersive processes (like genetic drift) as compared to animals with diploid sexes. Interaction between natural selection and mutation operate from the beginning in the haploid males. Consequently the chance of extinction for favorable mutants is much slighter, which results in a greater mutational yield and subsequent genetic divergence.

Upon considering these peculiarities we arrive at an adaptive type for spider mites, in which adaptedness arises from interpopulational selection rather than from intrapopulational variability. Several data, however, indicate that intrapopulational variability is greater than expected. In order to explain the intrapopulation variability, two hypotheses seems attractive:
1) Storage of sex-limited variants.
2) High spontaneous mutation rate.

RIASSUNTO

Le specie arrenotoche di Tetranichidi possiedono una dinamica di adattamento che si manifesta con caratteristiche differenti, in alcuni aspetti principali, da quelle delle specie a sessi diploidi.

Nelle specie a maschi aploidi si nota:
1) forte tendenza alla omozigosi;
2) alta produzione di mutanti.

In questi Acari, l'adattamento sembra dovuto ad un processo selettivo delle popolazioni piuttosto che a fenomeni di variabilità nell'ambito delle popolazioni, anche se tale variabilità risulta superiore alle previsioni.

A proposito di detta variabilità, l'A. formula due ipotesi:
1) conservazione di varianti legate al sesso;
2) frequenza di mutazioni spontanee.

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DISCUSSION

DE PIETRI-TONELLI: According to the fact that males of Tetranychids are most frequently haploid whereas females are diploid, can the process of resistance selection occur at a different speed on males and on females?

HELLE: It seems to me, when recessive factors are involved, the process of resistance selection will be very fast in spider mites, as compared with zygogenetic diploid species