Are microsatellite mutation rates higher in Orthopterans?



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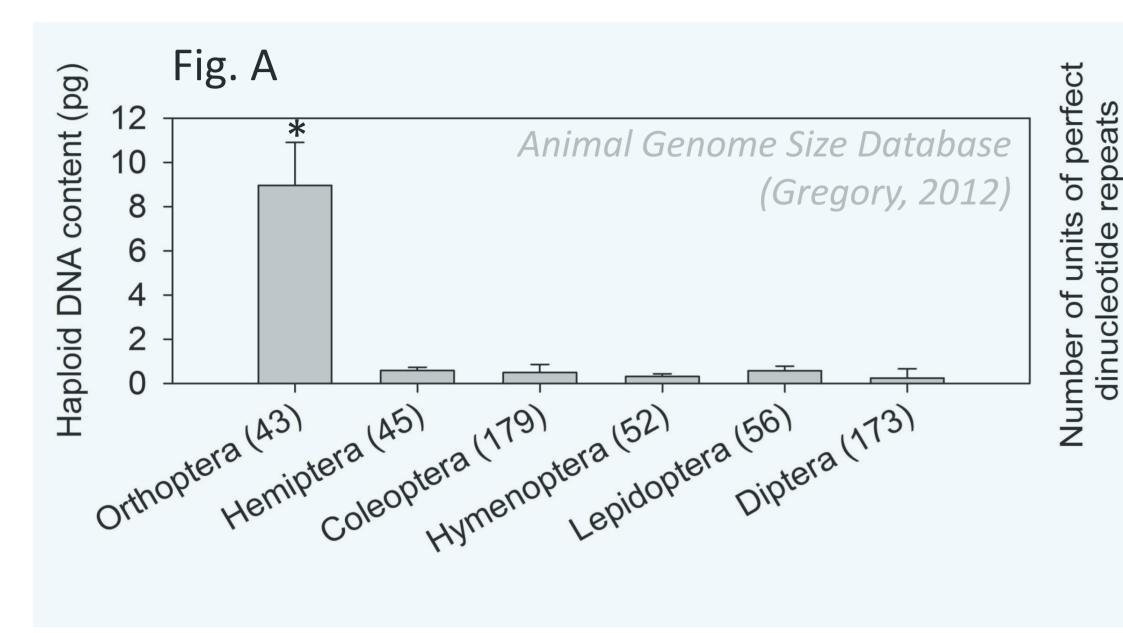


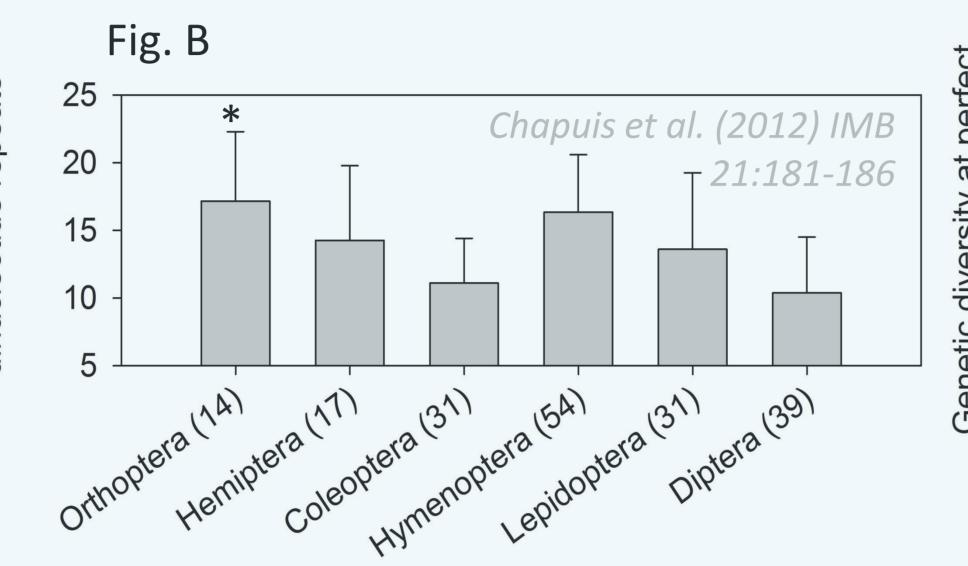
Introduction

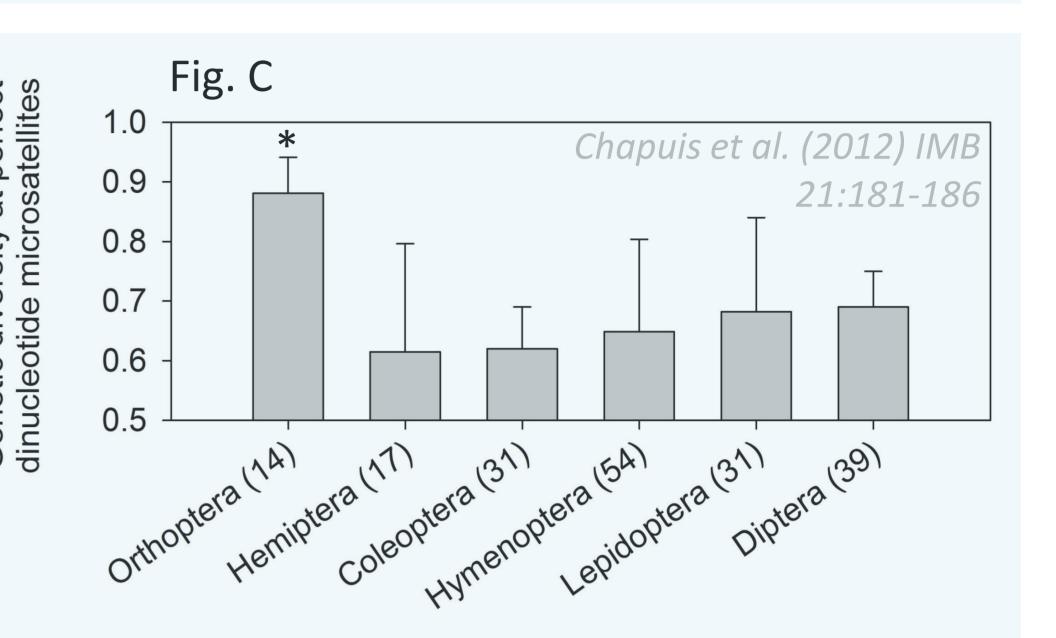


Orthopterans have the largest genome size among all insect orders (Fig. A), the longest microsatellite repeat arrays (Fig. B), and the highest levels of genetic diversity in natural populations (Fig. C).

Besides, some studies showed that microsatellites mutation rates are positively correlated to the length (number of repeats) of the microsatellite loci. All these features tend to presume a high mutation rate at microsatellites loci in Orthopterans.







In corollary to the mutation rate (μ) per se, one can question the correspondence of the mechanisms shaping the size distribution of both genomes and microsatellite repeat regions?

Locus

Hyp. 1: differences in mismatch repair systems lead to differences in relative DNA loss

Hyp. 2: differences in spread of transposable elements (TEs) lead to differences in abundance of TE-derived microsatellites

Methods

A total of 52,000 meioses in *Schistocerca gregaria*: 13 microsatellites; 2000 offspring; μ detection threshold of 2.5x10⁻⁴

- In the top of the range of pedigree studies for non-model species: 75-1615 offspring

2-11 loci

- Mean repeat # is 23:

predicts a mean μ estimate of 4 x10 $^{\text{-4}}$ [1 x10 $^{\text{-4}}$ - 4 x10 $^{\text{-3}}$] , according to the length/ μ correlation observed in previous studies

- Lab environment and genomes are close to nature:

field population (Akjoujt, Mauritania)
T°C= 34 (average during rain months)
no stress regimen imposed
high genetic diversity (*He*=0.84)

		. =		
SgM87	GT	DNA transposon	7	0.83
SgM92	GT	none	14	0.76
SgM74	GT	none	18	0.85
SgM88	GT	none	18	0.91
Sgr36	GT	none	19	0.86
Sgr53	GT	none	21	0.85
SgM66	GT	none	24	0.83
SgM41	GT	DNA transposon	25	0.84
DL13	CT	none	27	0.83
SgM86	GT	none	27	0.87
SgM96	CT	SINE	28	0.91
SgM51	GT	LINE	30	0.89
DL06	GT	none	34	0.75

Mean repeat #

Mean *He*

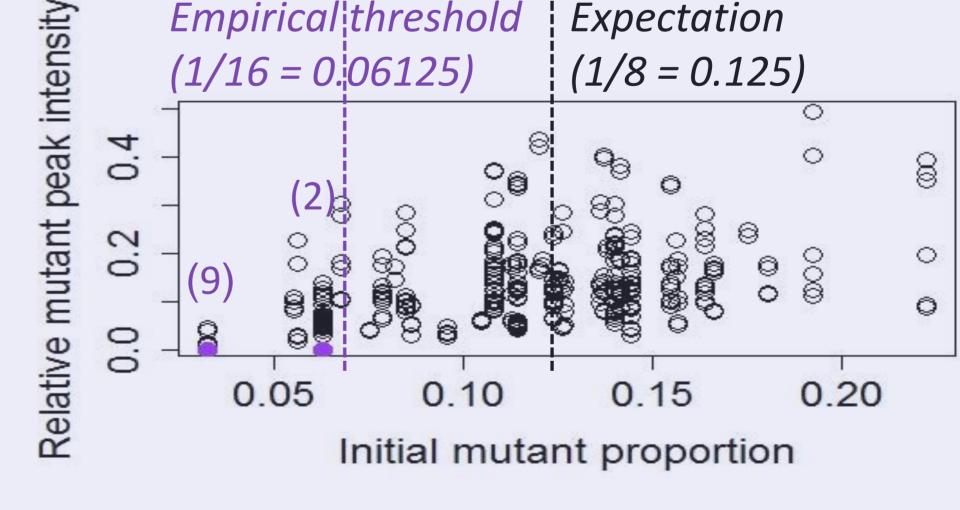
TE hits

Motif

Because of the high number of meiotic events, we developed a pool PCR protocol to detect one mutant in a maximum of 8 segregating alleles

Empirical threshold | Expectation

- Small pools of 4 DNA samples, each extracted from one back-leg of an offspring
- 200 artificial mutant tests to assess the mutation detection ability
- High repeatability of relative intensity of mutant peak between PCR runs ($R^2 = 0.98$)
- Relative mutant peak intensity is affected by initial mutant proportion in test (P = 0.012)
- 11 undetected mutants, all at an observed initial proportion << expectation
- → We adjusted initial concentration of DNA samples above empirical threshold



Results and Prospects

Offspring genotyping is in progress to estimate mutation rates and test for hypotheses of differences among insect orders. Furthermore, our study may identify hypermutable loci, which would be ideal for dissecting mechanisms of instability at microsatellite repeats in the unique genotypic and phenotypic contexts provided by the Desert locust.

Firstly, the high He will allow to confirm the « heterozygosity instability hypothesis » for large differences between the heterozygous allele and its homologue (i.e. up to a maximum of 7-26 repeat units in our loci vs. 4 in previous studies).

Secondly, environmental stress has received little attention as a mutability factor and the Desert locust is polyphenic, with a transition from a solitary form to a gregarious phase induced by high population density. Besides, there is debate over the possibility of increased genetic recombination rate in gregarious locusts.

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