Table 2. Estimates of the parameters of the 'burnt fingers' distribution

Species	$\hat{\delta}$	$\hat{\varepsilon}$	Goodness of fit
Banded plover	0.357	0.765	$\chi_1^2 \simeq 5.28$*$0.05 > P > 0.02$
Spur-winged plover	0.480	0.891	$\chi_1^2 \simeq 0.69$ $0.5 > P > 0.3$
Piping plover	0.336	0.947	$\chi_1^2 \simeq 0.27$*$0.7 > P > 0.5$

* Based on expectations at least one of which was less than 5.

immediately 'learns' enough to lower his chance of having further accidents. This crude model allowed the development of a satisfactory distribution where, in time (0,t), the probability of an accident occurring is δ, but thereafter falls to ε, i.e. $1 > \delta > \varepsilon > 0$.

The probabilities of 0, 1, 2 ... accidents in time intervals of length t are:

$$p(0) = e^{-\delta t}$$

$$p(1) = e^{-\varepsilon t} \frac{\delta}{\varepsilon - \delta} [e^{(\varepsilon - \delta)} - 1]$$

$$p(2) = e^{-\varepsilon t} \frac{\delta\varepsilon}{(\varepsilon - \delta)^2} [e^{(\varepsilon - \delta)t} - 1 - (\varepsilon - \delta)t]$$

$$p(3) = e^{-\varepsilon t} \frac{\delta\varepsilon^2}{(\varepsilon - \delta)^3} \left[e^{(\varepsilon - \delta)t} - 1 - (\varepsilon - \delta)t - \frac{(\varepsilon - \delta)^2 t^2}{2!} \right]$$

The parameters δ and ε may then be estimated, and this has been done for the clutch size of the banded plover, spur-winged plover and piping plover, with the results shown in table 2. Since $\varepsilon > \delta$, these results are not in agreement with the model. (This arises because the mean clutch size $\bar{x} > \delta$).

Table 3. Mode or range of clutch size of 319 species of Charadrii[13]

Modal clutch size	Number of species
4–6	1
4–5	1
4	137
3–5	3
3–4	16
3	25
2–4	19
2–3	32
2	64
1–4	3
1–3	4
1–2	9
1	5

An implication of the model might be that if the clutch sizes of particular species were simply the result of random fixation of genetic variation lowering clutch size, the distribution of modal clutch size among species would also be approximately triangular. Table 3, condensed from table 1 of McLean[13], shows that this is not the case.

Little, if anything, is known of the relationship of the cost of reproduction to variability in reproductive performance, yet this cost is critical[16,17]. An analysis of variability in terms of this cost, possibly using developments of Heyde's methods, is vitally necessary.

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Multiple phosphoglucomutase alleles in *Toxorhynchites splendens* (Diptera: Culcidae)[1]

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Summary. Multiple phosphoglucomutase (E.C. 2.7.5.1) alleles are found in the mosquito *Toxorhynchites splendens.* The sample studied reveals 3 *Pgm* alleles whose frequencies are in good accord with Hardy-Weinberg expectations. The most frequent allele is that controlling a phenotype with an intermediate electrophoretic mobility. Each *Pgm* allele determines a two-band electrophoretic pattern.

Electrophoretic data on gene-enzyme systems in mosquitoes reveal very frequent occurrence of multiple alleles. Of these, phosphoglucomutase (E.C. 2.7.5.1) and esterase (E.C. 3.1.1.1) are the most extensively studied[2,3]. Although many mosquito species have been studied, they belong mainly to 3 genera, *Anopheles, Aedes* and *Culex.* There appears to be no previous report on the genus *Toxorhynchites* Theobald. Members of this genus are large, non-biting mosquitoes. We report here the presence of multiple phosphoglucomutase alleles in *Toxorhynchites splendens.* This and other species of *Toxorhynchites* are considered beneficial to man because a) their larvae are predaceous on other mosquito larvae, and b) the adults may be used for rapid isolation of Dengue-fever virus[4].

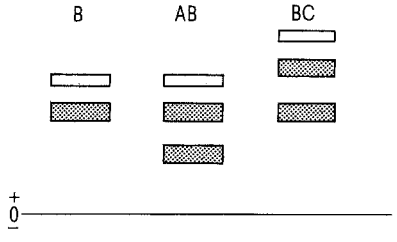
The mosquitoes used for the present study were obtained from laboratory colonies derived from Rantau Panjang, Selangor, Peninsular Malaysia[5]. Adult mosquitoes were used for horizontal starch-gel (12% hydrolyzed starch) electrophoresis employing the 'TEMM' buffer system and the enzyme visualization method of Spencer et al. with slight modification[6].

As with most culicine mosquitoes, each *Pgm* allele in *Toxorhynchites splendens* determines a two-band electrophoretic pattern (figure). In the present sample 3 codominant alleles are present. Their frequencies are $Pgm^A = 0.037$, $Pgm^B = 0.908$ and $Pgm^C = 0.055$. The distribution of the various phenotypes is summarized in the table. Only 3 phenotypes are detected in the present sample but they are in good accord with Hardy-Weinberg expectations ($\chi^2 = 1.10$). The high frequency of the Pgm^B allele also agrees with earlier reports for other mosquitoes that the most frequent allele is generally the one controlling a phenotype with an intermediate electrophoretic mobility[2]. This has been taken as supporting evidence for the idea that protein polymorphism is not primarily influenced by random genetic drift acting on a number of neutral isoalleles[7,8].

The present report helps to fill a gap in our knowledge of those mosquitoes which have not been genetically studied. Crossing experiments will be carried out to provide linkage and other data when other polymorphic gene-enzyme systems have been found in these mosquitoes.

Frequencies of *Pgm* phenotypes (= genotypes) in a laboratory colony of *Toxorhynchites splendens*

	Homozygotes			Heterozygotes		
	A	B	C	AB	AC	BC
Observed number	0	89	0	8	0	12
Expected number	0.15	89.87	0.33	7.32	0.44	10.89



Electrophoretic phenotypes of phosphoglucomutase in *Toxorhynchites splendens*.

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Binding of antimalarial drugs to hemozoin from *Plasmodium berghei*

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Summary. Chloroquine, quinacrine and mefloquine bind to *Plasmodium berghei* hemozoin, hemin, heme, protoporphyrin IX and protease digested methemoglobin. This binding may be the basis for drug accumulation and action in the parasite.

Studies on the molecular mechanism of action of schizontocidal antimalarial drugs are of current interest and of great relevance in the present situation of rapidly rising incidence of malarial infection and widespread drug resistance[2], e.g., to chloroquine. It is well established that chloroquine, quinacrine (mepacrine or atebrin), quinine and some similar blood-stage antimalarials bind well to DNA and can inhibit DNA, RNA and protein synthesis[3]. However, their lack of specificity for parasite DNA and the recently-observed lack of strong DNA intercalation by mefloquine, an extremely effective quinolinemethanol antimalarial, make DNA less likely as the main or only target for these drugs[3,4]. Here we report some preliminary results showing that hemozoin, the content of the parasite's 'autophagic vacuoles', is capable of binding all the above-mentioned drugs lending support to hemozoin as a more likely common site for drug uptake and concentration in the parasite than DNA.

Materials and methods. Bovine methemoglobin, hemin, trypsin and pronase were purchased from Worthington, and chloroquine, quinacrine and quinine from Sigma. Mefloquine was a gift from Dr W. Wernsdorfer of WHO. Protoporphyrin IX was from Calbiochem. Human heme (II) was prepared under nitrogen from fresh hemoglobin by the acid-acetone method[5] and also by dithionite reduction of hemin. *Plasmodium berghei* hemozoin was obtained from enriched infected mouse red cells as described by Yamada and Sherman[6] after the first-step treatment in a Parr cell disrupter bomb. A Beckman Acta V and a Perkin-Elmer (Coleman 55) spectrophotometer were used for absorbance and spectral studies; Hellma 2-chambered tandem cuvettes were used for difference spectroscopy, and a Packard scintillation counter was used for measuring ^{14}C-chloroquine (New England Nuclear) radioactivity.

Results and discussion. Our hemozoin preparation from *P. berghei* was similar to a previous preparation[6] from *P. lophurae* in terms of UV-visible spectrum and the main protein bands (mol. wts ~10,000 −20,000 and ~40,000) in the stacked gel. Upon addition of chloroquine, quinacrine and mefloquine to the hemozoin solution, spectral changes could be observed in the solely iron-porphyrin generated region from 450 nm to 700 nm clearly indicating binding. UV-visible difference spectra due to drug-hemozoin binding interaction are shown in the figure; the main longer