Control of the Human Head Louse with Disulfiram and Benzyl Benzoate Emulsions

A Laboratory Study

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The effects of disulfiram, benzyl benzoate and DDT, all components of a common preparation (Tenutex®) for the control of the head louse, were tested on louse eggs. A significant higher mortality of eggs was evident after treatment with (i) disulfiram, (ii) benzyl benzoate, (iii) these two substances mixed, and (iv) Tenutex®. DDT in this concentration had no effect on the survival of the eggs. Long-time exposure to Tenutex® and Tenutex without DDT significantly lowered the hatching frequency of eggs. Almost no hatching occurred after exposure for 24 hours. The survival of larvae hatched from eggs treated with Tenutex® was lower than for those treated with Tenutex without DDT. DDT seemed to have an effect primarily on the survival of larvae. Newly hatched larvae were more sensitive to the two Tenutex preparations than older larvae. (Received October 4, 1983.)

Man is parasitized by four obligate ectoparasites: the human flea (Pulex irritans), the scab mite (Sarcoptes scabei), the crab louse (Phthirus pubis) and the human louse (Pediculus humanus). The latter is cosmopolitan split into two subspecies, P. h. capitis (head louse) and P. h. humanus ("corporis") (body louse). These two forms are morphologically very similar but easily separable biologically. The body louse is rare in Scandinavia and is occasionally brought in from abroad. It lives mainly on undergarments or garments next to the skin. The body louse is a well known vector of causative agents for e.g. louse-borne typhus (Rickettsia prowazeki), relapsing fever (Borrelia recurrentis) and trench fever (Rockalimaea quintana).

The head louse is on the other hand relatively harmless and lives in the hair of man where it prefers the neck and the hair behind the ears. The direct effects of the head louse, like those of body louse and crab louse, are to cause irritation and consequent scratching. This may lead to secondary infections.

The number of cases of head louse infestations has increased considerably in W. Europe during the last few decades. According to Weidhaas & Gratz (7) there are cities in Britain where the incidence of head lice among children is as high as 15%. Various reasons have been suggested e.g. increased travelling and tourism, immigration, and the fashion of long hair among young people. Whatever the reasons might be, some facts are obvious in all regions of infection: firstly; school children are mostly infested and secondly; lice seem to be well established and difficult to control.

Several chemicals or mixtures thereof are used for the control of the head louse in various parts of the world. Blommers (1) reported for instance that malathion is the active substance used in the Netherlands because the lice populations there have become resistant to lindane and dieldrin. The most frequent product used for control of head lice in
Sweden is Tenutex®. The active components are benzyl benzoate, disulfiram and DDT (5). The use of DDT in direct contact with the skin is not appropriate and has been criticized in Sweden. The aim of this investigation was therefore to test the effect of the different components in Tenutex®. Primarily the effects on eggs were emphasized but effects on adults and larvae were also considered.

**Biology**

The biology of *P. humanus* is well investigated. Extensive studies have been made by Nuttall (6), Wigglesworth (8) and Buxton (3). A later review has been presented by Dittmann & Eichler (4).

A female head louse lives about 3 weeks and produces in general 5–7 eggs per day. Her total egg production is 100–120. The hatching frequency is normally 75–95%. The female fixes the eggs to the bases of hairs with a glue produced in the abdomen. The eggs hatch after 8–10 days; the empty translucent egg shells remain fixed to the hair long after hatching. After three larval stages the louse becomes sexually mature. The whole life cycle from egg to adult takes 12–15 days depending on temperature and relative humidity. The time from egg to egg is about 3–4 weeks. The number of lice per head is often less than ten, but the variance is very great (3).

The head louse is spread primarily by direct contact with infested individuals but also by common use of combs, caps, hats, pillows etc. It should be noted, however, that a louse away from its host will die after 10–12 hours. Already after only a few hours a strayed louse becomes severely weakened and will hardly regain its full vigour.

**METHODS**

** Cultures**

To start a laboratory culture, about a fifty lice were collected from school children. Rearing the lice on man is a standard method (6) and only in investigations of lice as vectors have guinea pigs, rabbits and monkeys been used. A small plastic capsule (24 mm. height 10 mm) was covered underneath with a small-meshed net (we used nylon stockings) through which lice could suck blood but not escape. The capsule, with a small wire frame (17×18 mm) inside wrapped with hair, was placed in a bracelet of leather and fixed to the arm like a watch. The bracelet was worn by the experimentee day and night. Usually 5 females and 5 males were kept in each capsule. The egg production in the capsules varied depending on the fertility of the lice. Every second day lice were transferred to a new capsule and the eggs produced in the first capsule were removed for tests or for incubation.

Newly hatched larvae measure less than 1 mm and could easily escape from the capsules. The first larval stage was therefore kept in Petri dishes and incubated at 32°C and 80–90% RH. The larvae were fed on the underarm of the experimentee twice a day. During feeding (5–10 min) the larvae were kept in a small plastic frame, divided into 8 compartments (Fig. 1). In this way larvae from different tests could be fed at the same time.

**Tests**

Test A: placebo. DDT 0.5 g, disulfiram 2 g, benzyl benzoate 22.5 g, disulfiram + benzyl benzoate (Tenutex without DDT), disulfiram + benzyl benzoate + DDT (Tenutex®).

Test B-D: placebo, Tenutex without DDT, Tenutex®.
Fig. 2. Hatching of eggs and survival of larvae which hatched from eggs treated for 1 h, 4 h, 8 h, and 24 h with a placebo, Tenutex® and Tenutex without DDT. Only fertile eggs were used (100% hatching in the control group). The hatching frequency of treated eggs has been related to the expected hatching of untreated eggs. The time for the first observed hatching has been considered as “day 1”. 
--- hatching of untreated eggs; ----, frequency of living larvae from untreated eggs; ---, hatching of treated eggs; ----, frequency of living larvae from treated eggs.

(a) Hatching frequencies of eggs treated with Tenutex® and its various components
Half the number of eggs from one capsule was treated with the test substance, the other half constituted the control group. The test substance was applied as a thin layer on the eggs. The eggs of the control group were not treated with any substance but were in all other respects handled identically with the test group. All eggs were incubated at 30°C ± 1°C 16 h.

(b) Hatching frequencies at different exposure times
The eggs were treated as above. After exposure times of 1 h, 4 h, 8 h, 16 h (experiment A) and 24 h the substance was carefully washed off with a diluted solution of hair shampoo. The eggs were then rinsed in water and again incubated. Hatching eggs were checked twice a day.

(c) Survival of larvae hatched from treated eggs
The survival of larvae hatched from treated and untreated eggs (control group) was studied. At each check (twice a day) the larvae were fed for 5–10 min on the arm.

(d) Survival of larvae and adults treated with test substances
The following groups of lice were treated with test substances:
1. Larvae, 1–2 days old.
2. Larvae, 7–9 days old; these originated from hatched eggs in the control group.
3. Adult lice used in the production of eggs and larvae. As the number of adult lice was small, only Tenutex without DDT was used as test substance. Blommers (1) gives a method for testing the effect of different chemicals on louse larvae. His method is, however, designed for fluids and not applicable for unguents. Instead, the test substance was applied as a small droplet on the back of the lice.
work was carried out with a brush under a microscope. The insects were incubated and checked for survivors after 4-5 hours and again after 23-24 hours. The mortality of untreated eggs was estimated from survival curves from earlier tests (Fig. 2).

RESULTS AND DISCUSSION

(a) Hatching of eggs treated with Tenutex® and its various components

The survival of the eggs in the control group varied between 53 and 100%. A significant higher mortality was found for four of the tested components. They all contained benzyl benzoate and/or disulfiram (Table I). Consequently these two preparations seemed to be the most active substances for the control of head lice among the tested components. DDT at this concentration had little or no effect at all on the survival of louse eggs. In fact our results were consistent with those reported by Borglund et al. (2) working on the effect of Tenutex® on human scabies.

(b) Hatching frequencies at different exposure times

All three preparations killed fertile eggs (Fig. 2). Treatment with placebo caused a mortality 10-30% higher than in the control but this difference was not statistically significant. No increasing effect due to longer exposure could be found (Fig. 2). There was, however, a clear correlation between the time of exposure and egg mortality for both Tenutex preparations. It was also shown that the results better fit a rectilinear model ($r=0.83$ and $r=0.92$) than a log-normal ($r=0.81$ and $r=0.89$). Especially for Tenutex® the correlation between time exposure and egg mortality is clear while the same correlation for Tenutex without DDT is less obvious (Fig. 3). Tenutex without DDT seemed to be more effective than Tenutex® at shorter times of exposure. The variance is, however, often very great in materials of this type (cf. 3, 6). At longer exposure times no significant difference between the preparations could be detected.

(c) Survival of larvae hatched from treated eggs

The survival of larvae hatched from eggs treated with a placebo and Tenutex without DDT, did not differ from expected. For Tenutex®, however, the larval survival was constantly lower than expected and in one case this was statistically significant ($p<0.001$).

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<th>Table I. Percentage of head louse eggs hatched after treatment with Tenutex® and its various components</th>
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<td>Disulfiram + benzyl benzoate</td>
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(d) Survival of treated larvae and adults
Larvae treated with any of the substances died in a significantly (p<0.001) higher frequency than untreated larvae (Fig. 4). No difference between newly hatched and older larvae was observed. Neither was there any significant difference between the two Tenutex modifications (Fig. 4).

Fig. 3. The frequency of killed fertile eggs in relation to the exposure time of placebo (O), Tenutex® (A), and Tenutex without DDT (*).

Fig. 4. Survival of newly hatched larvae (1–2 days), and older larvae (7–9 days) and adult lice treated with placebo, Tenutex® and Tenutex without DDT.
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REFERENCES