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Ultraviolet radiation of *Schistosoma mansoni*. II. Post-hatching radiation effect on some aspects of miracidia behaviour and infectivity

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Abstract

Batches of *Schistosoma mansoni* miracidia were irradiated with ultraviolet light for varying time intervals to determine the influence of radiation on the transmission potential of radiated miracidia. There was a decrease in the survival rate of hatched-free swimming miracidia that corresponded with the increasing radiation exposure time. The activity rate of radiated miracidia also decrease with increasing radiation exposure time, showing a 69.3% reduction in the mean rate of movement of miracidia irradiated for 30 minutes and 40.4% reduction in the miracidia exposed for 0.5 minutes. The infectivity rates of the free-swimming miracidia exposed to 5.0, 15.0 and 30.0 minutes uv radiation were significantly different ($P<0.001$, $d\alpha=4.171, 5.07, 5.227$) compared with the non-irradiated miracidium.

Introduction

In the preceding paper, the relationship between dose of ultraviolet radiation, the hatching of eggs, the life span and motility of *Schistosoma mansoni* miracidia obtained from irradiated eggs were reported, with no significant variation in the infectivity rate of miracidia at the various radiation dose between 0.5 to 30 minutes. The ability of the egg stage of *S. mansoni* to resist the radiation influence cannot be insignificant in the desire to reduce transmission from man to snail.

Ghandour and Webbe (1975); Ariyo and Oyerinde (1990), found that the irradiation of *S. mansoni* cercariae had a negative influence on the cercarial infectivity which decreases with increasing radiation levels. Chi and Boeller had earlier reported that the irradiation of spermatozoa of *Oncomelania* species rather than the eggs is advantageous for purposes of snail control (Chi and Boeller, 1968). Under these considerations there is the possibility that irradiating the miracidium which is the first larval schistosome stage could result in a reduction in the efficiency of transmission from man to snail, hence, the present investigation was undertaken to determine possible differences in activity and infectivity of miracidia irradiated after hatching. The result are discussed in relation to aspects of the epidemiology of snail infection.

Materials and methods

Preparation of egg suspension and the hatching of eggs: Eggs of *S. mansoni* were recovered from the livers of infected white mice, 12 weeks post-infection by homogenizing the liver tissue in approximately 50 ml cold normal saline. The number of eggs in 0.5 ml of the thorough mixed suspension was determined.

Irradiation of S. mansoni miracidia: Eggs were induced to hatch in a volumetric flask. The miracidia were made to concentrate in the neck of the flask by darkening the bottom part with carbon. Freshly hatched miracidia were pipetted into a beaker. The mean number of miracidia in 0.5 ml of the thoroughly mixed miracidia

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suspension and hence the number of miracidia in 5 ml was determined after addition of a drop of formalin. Five millilitres of freshly hatched miracidia was pipetted into each of five 90 mm diameter petri dishes. Four of these dishes were exposed to 0.5, 5.0, 15.0 and 30.0 minute(s) radiation levels respectively from a lamp source – (P. W. Allen and Co fluorescent lamp, UV emitter, type A425), according to procedure detailed out by Ariyo and Oyerinde (1990). The control batch of miracidia suspension was not exposed to ultraviolet radiation.

Miracidia life span, motility and infectivity: Suspension of miracidia were obtained from batches of eggs irradiated at different radiation levels. The mean number of miracidia per 0.5 ml of the miracidia suspension was established. At 26°C room temperatures and at one-hour intervals, the number of dead miracidia in 0.5 ml of the suspension was counted. Death of miracidia was determined by lack of motility. A mean number of dead miracidia and hence the percentage of survival was determined.

Table 1: The rate of movement of ultraviolet irradiated S. mansoni miracidia.

Radiation Levels (mins.)	Mean distance in mm/sec	(S.D)	Percent Reduction
0.0	2.18	(0.35)	0.0
0.5	1.03	(0.50)	40.4
5.0	1.12	(0.14)	48.6
15.0	0.79	(0.14)	63.8
30.0	0.67	(0.15)	69.3

Results

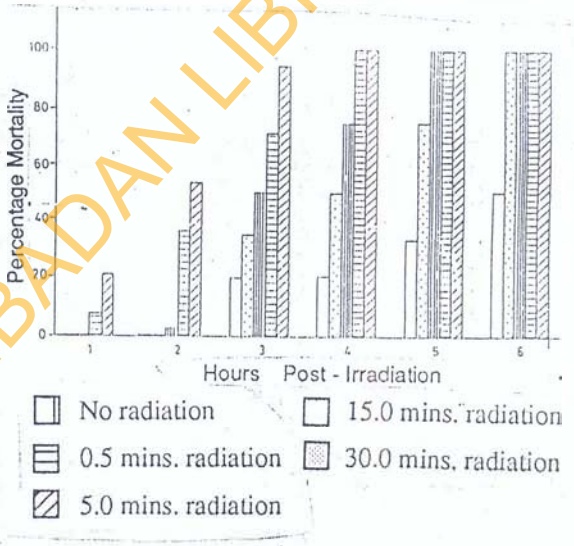
Miracidia motility and life-span: Direct exposure of *S. mansoni* miracidia to uv had an inhibiting effect on the rates of activity and survival of the miracidia. The findings are presented in Tables 1 and 2. The motility rates in the different batches of irradiated miracidia are significantly different from the motility rate in the non-irradiated batch of miracidia, $p<0.001$, ($t=4.573, 8.968, 11.760$ and 12.647 for 0.5, 5.0, 15.0 and 30 minute(s) respectively). The rate of reduction in motility increased as radiation level increased to 30.0 minutes (Table 1). Movement was 69.3 times slower than the movement rate in the non-irradiated control group of miracidia.

While only 5% of the 30 minutes irradiated miracidia survived for 3 hours, the control experiment showed that 80% of the miracidia were alive during the same period, post-irradiation, (Table 2). In fig 1, the cumulative mortality rate at interval was shown to increase with the radiation level and age of miracidia.

Table 2: The survival rate of ultraviolet radiated *S. mansoni* miracidia

Radiation Dose (mins)	Time of observation (hours) 1	2	3	4	5	6
0.0	100	100	80	80	67	50
0.5	100	100	65	50	25	0
5.0	100	97	50	23	0	0
15.0	93	64	29	0	0	0
30.0	79	47	5	0	0	0

Fig. 1: Cumulative mortality rate of *S. mansoni* miracidia irradiated post hatching of eggs.



Infectivity of irradiated miracidia: Table 3 shows the number of miracidia that were able to attach to the snail, which was taken to be the number of infecting miracidia and hence the infectivity. The infectivity was found to decrease as the level of radiation increased from 5 to 30 minutes.

Table 3: The infectivity of *S. mansoni* miracidia exposed to varying doses of UV rays

Radiation Levels (min.)	Snail number 1	2	3	4	5	6	7	8	9	10	Mean of attached miracidia per snail (S. D.)
0.0	8	7	6	8	8	7	6	8	7	7	7(1)
0.5	7	7	5	5	5	7	6	8	9	7	6(1)
5.0	4	2	4	6	5	4	3	5	6	3	4(1)
15.0	1	3	4	5	4	2	5	6	3	3	4(2)
30.0	2	2	4	5	3	3	5	4	6	1	4(2)

Irradiation for 0.5 minutes did not seem to have any inhibiting effect on the infectivity of the miracidia as there was no significant difference between infections

produced by non-irradiated controls and those irradiated for 0.5 minutes ($p>0.10$). Irradiation for longer periods, 5.0, 15.0 and 30.0 minutes showed a significant difference in infectivity ($p < 0.001$, da = 4.171, 5.071, 5.227 respectively) compared with infectivity observed in the control group of miracidia.

Discussion

Radiation is expected to influence living organisms to varied degrees (Krakower, 1940; Stowen, 1942; Keeling, 1960). The significance of such influences will be related to the level of inhibition-radiation can have on the ability of parasitic or pest organisms to maintain continuity of life. According to previous observations by these same authors, uv radiation had insignificant influence on miracidia from hatched *S. mansoni* eggs while Ariyo and Oyerinde (1990) reported a significant decrease in worm burden and egg productivity following irradiation of *S. mansoni* cercariae. The present investigation revealed a reduced infectivity of miracidia within a range of exposure to radiation; suggesting the possibility of a significant inhibition to further development of miracidia in their snail host and reduction in transmission with respect to the population dynamics of schistosome development. It is however pertinent to mention that this observation shows a break-point in the life cycle of irradiated schistosome miracidium, for the purpose of reduction in parasite number and transmission, particularly as there is a transmission threshold below which no transmission can successfully take place (Anderson *et al.* 1982). The irradiated larval stages of schistosome, apart from being instrumental to the development of immunity in the host (Lichtenberg and Sadun, 1963), can also be regarded as not being available for transmission from man to snail as the behavioural differences between non-irradiated and irradiated miracidia were significant. Anderson *et al.* (1982) associated a decline in infectivity with age, hence the depletion of a finite energy store. This explains the gradual reduction in the rate of movement and life span of the free-living miracidia stage with time. However, the significant reduction in infectivity, rate of movement, and life span in relation to radiation level exposure could only have been a result of various experimental conditions such as quality and dose of radiation, irradiation temperature, oxygen tension, and so no (Mangold and Dean, 1984; Dean *et al.* 1986). However, the protective effect of the eggshells or the cumulative effect of ordinary light in enhancing hatching and ultraviolet radiation could make it possible for infectivity of miracidia to be unaffected or significantly inhibited as the case may be. The major implication of the conclusion reached concerns the dose dependency nature of uv effect on age-dependent infectivity of irradiated miracidia for control purposes. The possibility that the stage of miracidia at the time of radiation can influence development within the snail is an important area for further investigation.

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