THE LIGHT THRESHOLD CONTROLLING THE PERIODICITY OF INVERTEBRATE DRIFT

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INTRODUCTION

It is now established that invertebrate drift is a normal feature of the lotic system (Borgh 1927; Ide 1942; Müller 1954; Waters 1961). Also studies on the periodicity of this phenomenon have shown that there is an increase in the level of drift at sunset which is maintained throughout the night (Müller 1963a, b, c; Tanaka 1960; Waters 1961) and analysis of the actual mechanism of this periodicity has resulted in an increased interest in the factors which control it.

It was concluded from a recent laboratory study (Chaston 1968a) that possibly the most important exogenous control factor is light, and that the initial nocturnal increase is a reflection of a rise in the activity level of the benthic animals induced by the decrease in the level of light at dusk. Müller (1965) has examined this light threshold by artificially illuminating a stream and measuring the level of drift. He concluded that the nocturnal increase occurred below 5 lx and in this present study the value of the threshold was examined more fully by (a) field experiments without artificial illumination to measure the variation in the level of drift under the rapidly changing light conditions between sunset and complete darkness, and (b) exposing a number of aquatic insect larvae to controlled light conditions in the laboratory and using the results to calculate the absolute threshold, i.e. the minimum level of light at which the nocturnal increase does not occur.

In addition it has also been suggested that even light levels below the threshold can quantitatively affect drift (Waters 1962) and data supporting this hypothesis have been produced by Anderson (1966) who found, in one sample, a 600% difference between the levels of drift on nights of a full and no moon. Samples were taken during this present study to examine this effect further.

During all this work, measurements of light were made in lux to assist the comparison of the results with those of other workers. Lux, however, is a quantitative estimate of illuminance based on that part of the spectrum visible to the human eye and thus it was necessary to obtain some indication of the action spectra of the animals involved in drift to discover if they are within the same spectral range as man, that is that the animals are most stimulated by light between 500 and 600 mμ. For without this information a statement of the optomotor response is useless as one may be stating a light threshold in terms of wavelengths that may not even stimulate the animals.

METHODS

The field estimation of threshold

The field samples were taken using 'modified high-speed plankton samplers' (Elliott 1967a). Before they could be used for this present work, however, some modifications were required because, with the original method of cone attachment, the samplers had

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to be removed from the water before a new net could be fitted. Thus up to 20 min were required to change three samples (Elliott, personal communication) and in order to overcome this problem two quick-release clips, of the type used as cover-plate retainers on industrial vacuum cleaners, were bolted on to the mouth of each sampler. Also one net was permanently attached to each cone. With these two modifications it was possible to change three nets and cones in $2\frac{1}{2}$ min, whilst the samplers remained in the water.

Sampling, which was carried out in the Walla Brook, a tributary of the East Dart, commenced 15 min before the time of astronomical sunset and continued until complete darkness. This period covers approximately $1\frac{1}{2}$ h and by changing the nets every 15 min, six samples were taken in an evening. Three complete series of samples were taken every month (except December when bad weather conditions damaged the equipment so severely that repairs were not completed until January) between September 1966 and August 1967. During sampling the changing light conditions were measured with a 'probe photometer' (Chaston 1968b) mounted 2 m downstream.

The laboratory estimation of absolute threshold

The controlled light conditions were obtained using a fluorescent strip light and neutral density filters. This type of filter was used because it was then possible to reduce the level of light by graduated absorption without altering the spectral energy distribution of the light source.

The filters, produced by 'fogging' Crystallex (Kodak) X-ray film (plate size 17·8 × 43·2 cm), were made in the laboratory because of the difficulty of obtaining such large filters commercially. After developing, each plate was mounted between two sheets of 50 mm thick plate glass to protect the surface of the negative. (In fact the completed filters were found to be much sturdier than the normal gelatin-type neutral density filters which are easily damaged by handling or damp.) Confirmation that X-ray film was acceptable for neutral density filters was obtained by mounting a test strip in a spectrophotometer, and it can be seen from Table 1 that there is very little variation in the percentage absorption of light at different wavelengths of the visible spectrum.

Table 1. *The results of the spectrophotometer test to check if 'fogged' X-ray film was an acceptable material for the manufacture of neutral density filters*

Wavelength to which strip was exposed (mμ)	% absorption of light by test strip
400	5·8
450	6·5
500	6·95
550	5·0
600	5·5
700	5·3
750	5·2

Approximately 150 insect larvae of one genus were placed in a model stream (for construction details see Chaston 1968a) 24 h before the experiment commenced in order that the animals could become settled in their new environment. A box containing the light source and filters was mounted 1 m above the water's surface. The next day, at 14.00 hours, the animals were illuminated by an overhead light (100 W, 240 V AC bulb) and at 15.00 hours the electric water-circulating pump was started, its revolutions being built up to a maximum over a 15 min period with a Variac. (At maximum revolutions the pump circulated 300 litres of water through the model stream per hour.) At 16.00 hours the overhead light was extinguished and the animals were exposed to the controlled light conditions for 3 h. Any larvae in the water column were caught in a net mounted at one end of the stream, and after counting, the contents of the net were returned to the tank, the pump of which had been stopped at the end of the 3 h period. Again the animals were exposed to the overhead light and an hour later the pump was restarted. All lights were extinguished at the end of this second hour and the animals were left in complete darkness for a further 3 h, after which the net was again emptied. The number of animals appearing in drift during the first part of the experiment under controlled light conditions was expressed as a percentage of the number in drift under complete darkness.

The effect of five different light levels on drift was examined and the experiments were repeated three times with each genus of larvae under every light condition. The values of the controlled light conditions were purely arbitrary during the experiments and dependent upon the light absorption characteristics of the various filters. After all the experiments had been completed the five light levels were measured with a probe photometer mounted just above the substratum of the model stream (see Table 2).

Table 2. *The controlled light values to which the five genera of larvae were exposed during the laboratory experiments on threshold*

Experiment no.	Level of light (lx)
1	8·75
2	3·90
3	1·85
4	0·19
5	0·115

The various genera of larvae used in the experiments, *Isoperla* (Plecoptera), *Baetis* (Ephemeroptera), *Ephemerella* (Ephemeroptera), *Protonemura* (Plecoptera) and *Simulium* (Diptera), were all chosen because they could easily be obtained in large numbers.

Samples to investigate the effect of moonlight on drift

Samples were taken from the Walla Brook during nights of a full moon using three Elliott high-speed plankton samplers. The experiments commenced an hour before sunset, the nets being changed every hour, and continued until an hour after sunrise next day. For a comparison, a sample was taken 2 weeks later on a night of no moon. Three of these comparative samples were taken; one in spring, another in summer and a third in early winter.

Investigation of the action spectra

Investigation of the action spectra of the insect larvae was based on the same procedural principles as used in the laboratory threshold experiments except that the animals were exposed to various known wavelengths of light instead of a number of known intensities. The number of animals appearing in drift under a known wavelength was expressed as a percentage of the number in complete darkness.

The various wavelengths of light were produced with a 15·25 cm 'graded-strip' interference filter mounted at the base of a box containing a 100 W, 12 V DC tungsten iodine lamp. This type of filter, which employs light interference to produce the required spectral component, was used in preference to the normal glass or gelatin filter which relies upon absorption and mixture to produce a colour of a much less pure nature.

The action spectra of the same five genera of larvae used in the absolute threshold experiments was investigated by exposing them to light at 400, 450, 500, 600 and 700 mμ.

RESULTS

The field estimation of threshold

In order to estimate the threshold for an evening's sample, a plot was made of the number of animals in drift (i.e. the number of animals caught in the drift sampler) in each 15 min sample against time, and above this a plot of the logarithm of light measured every 15 min against time. The start of the nocturnal increase was taken as the mid-point between the last sample showing the 'day' level of drift and the first sample exhibiting the 'night' level. (If the increase continued over more than two samples, the mid-point was taken as halfway along the slope of the plot of the number of animals in a 15 min sample against time.) Thus, knowing the time of the nocturnal increase, one could read off the level of light at that point and the figure was taken as the threshold for that evening's sample. The plots of 1 month's samples are given in Fig. 1 to illustrate this method of estimating graphically the threshold.

Table 3 shows the calculated thresholds and from these results it was estimated that the mean threshold for the period September 1966 to August 1967 was 1·584 lx.

The laboratory estimation of threshold

A mean was calculated for the three experiments under each light condition and the results are given in Table 4. If these raw data were plotted against the controlled light levels, the scatter of the points would not give sufficient information for an estimation of the absolute threshold. Therefore the data were used as the basis of an arithmetic regression of X on Y where $X =$ the level of light and $Y =$ the percentage of drift under a controlled light condition. The absolute threshold was taken as the value of X when $Y = 0$ in the equation $X = bxy.Y + Ax$ and from these calculated estimates for the five genera of larvae (see Table 4) it was calculated that the mean absolute threshold was 7·223 lx.

Samples to investigate the effect of moonlight on drift

Plots were made (see Fig. 2) of the number of animals in drift in each hourly sample against time. Also a t-test was applied to the mean number of animals in drift per hour in each of the comparative experiments. In all three experiments P was more than 5%.

Investigation of the action spectra

The mean percentage of animals in drift under each wavelength was calculated, and a correction of about 2–3% was necessary to allow for the slight variation in the transmission characteristics of the filter at different wavelengths. It was then possible to use the corrected data to obtain an indication of the action spectra of the five genera of larvae by plotting the percentage of animals in drift at a known wavelength against microns. From these plots (see Fig. 3) it can be seen that all five genera are most stimulated by light between 500 and 600 mμ.

CONCLUSIONS

The mean field threshold of 1·584 lx compares favourably with Müller's (1965) estimate, obtained by artificially illuminating a stream, that the threshold for the maximum level of drift lies between 1 and 2 lx. However, there is some variation between the mean monthly figures (see Table 3) and, although the large standard deviations possibly point to experimental error, the drop of the threshold in winter and the rise in summer may not simply reflect faults in the sampling technique. Some possible reasons for this seasonal variation are discussed later.

The absolute threshold of 7·223 lx is higher than the field estimate for two reasons. Firstly, the absolute threshold is an estimate of the level of light below which the nocturnal

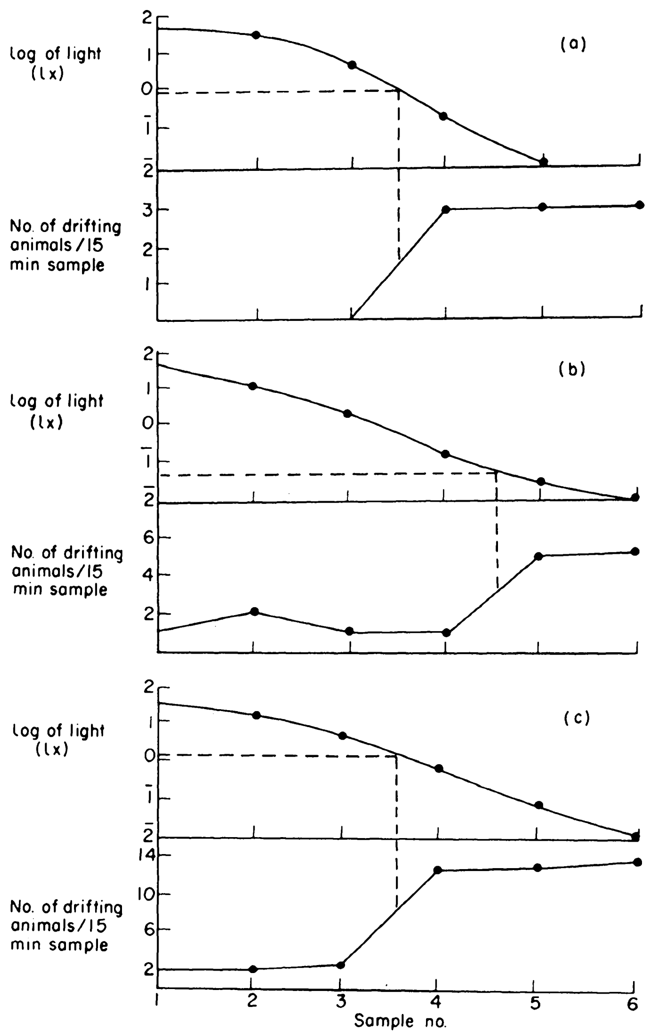

FIG. 1. An example of the graphical method used to estimate the light threshold of 1 month's field samples. (Both the number of animals in drift per 15 min sample and logarithm of light measured every 15 min are plotted against time. The dashed line indicates the time and thus the threshold of the nocturnal increase.) (a) Evening 1; (b) evening 2; (c) evening 3.

Table 3. *The values of the field thresholds calculated from the evening drift samples taken from the Walla Brook*

Month	Thresholds (lx) (i)	(ii)	(iii)	Mean threshold
September	10·0	0·502	0·398	3·633 (S.D. = 4·503)
October	3·98	0·398	0·199	1·526 (S.D. = 1·74)
November	2·51	0·126	3·98	2·205 (S.D. = 0·169)
January	1·99	1·0	0·5	1·16 (S.D. = 0·169)
February	0·536	0·1	1·26	0·641 (S.D. = 0·477)
March	0·199	0·708	0·126	0·344 (S.D. = 0·258)
April	0·0795	0·631	6·31	2·34 (S.D. = 0·26)
May	0·795	0·136	0·159	0·432 (S.D. = 0·227)
June	0·0631	1·0	0·159	0·414 (S.D. = 0·421)
July	3·16	0·398	3·98	2·513 (S.D. = 1·593)
August	3·16	0·502	0·795	2·229 (S.D. = 1·403)

Mean threshold based on figures from September 1966 to August 1967 = 1·584 lx (S.D. = 1·034).

Table 4. *The results of the laboratory threshold experiments expressed as a mean percentage of drift from the three experiments under each of the controlled light levels used on the five genera of larvae*

Experiment no. (for equivalent light values see Table 2)	% in drift *Baetis*	*Ephemerella*	*Simulium*	*Isoperla*	*Protonemura*
1	5·49	11·07	0·0	0·0	0·0
2	23·18	31·78	0·0	0·0	0·0
3	37·89	48·18	32·87	21·17	26·12
4	73·88	66·91	54·86	44·81	63·25
5	92·42	78·62	101·85	63·17	80·58
Absolute threshold*	7·709 lx	9·634 lx	7·396 lx	5·677 lx	5·68 lx

* (calculated from an arithmetic regression of X on Y where X = level of light and Y = % in drift)
Mean absolute threshold = 7·223 lx.

increase can occur. But in fact although the increase occurs below this figure, the level of drift continues to rise and stabilizes between 1 and 2 lx (Müller 1965). Thus owing to the insensitivity of the sampling technique employed, the nocturnal rise is not obvious in the field until this maximum level is reached, and the field estimate of threshold applies to this evident rise in drift and not to the maximum level of light at which the increase can start. Secondly, the laboratory technique needed to estimate the threshold necessitates the use of an artificial light source and, although fluorescent light has a similar spectral characteristic to that of sunlight, the respective radiant energy outputs of these two types of light differ. One lux corresponding to 4×10^{-6} gcals/cm^2/min for fluorescent light and 6×10^{-6} gcals/cm^2/min for sunlight (Westlake 1965). Thus if it were possible to repeat the experiments employing controlled daylight values, one would expect the absolute threshold value to be reduced by a third, and this would decrease the difference between the absolute threshold and the level of light at which the maximum nocturnal increase occurred.

There is no statistical difference between the number of animals in drift per hour under full and no moon, and thus no evidence is forthcoming from these samples that would support Waters' (1962) hypothesis that moonlight can quantitatively depress drift. However, although the results of the three comparative samples substantiate Müller's (1965) view that little variation in the level of drift occurs below 1 lx, there is still a theoretical case for moonlight depressing drift because during the laboratory experiments on threshold it was found that there was a mean 40% difference between the number of animals in drift per unit time at 0·19 lx and complete darkness. This effect, nevertheless, would be difficult to demonstrate in the field because variations of 30–40% would probably be lost in sampling error. Therefore it does not seem possible that the 600% difference between drift under full and no moon found by Anderson (1966) can be entirely attributed to moonlight. It is more likely to have been caused by some other variant factor such as the velocity of the water in the stream.

From the results of the action spectra experiments (see Fig. 3) it is concluded that lux is an acceptable unit because it estimates light within the spectral range that stimulates the animals involved in drift.

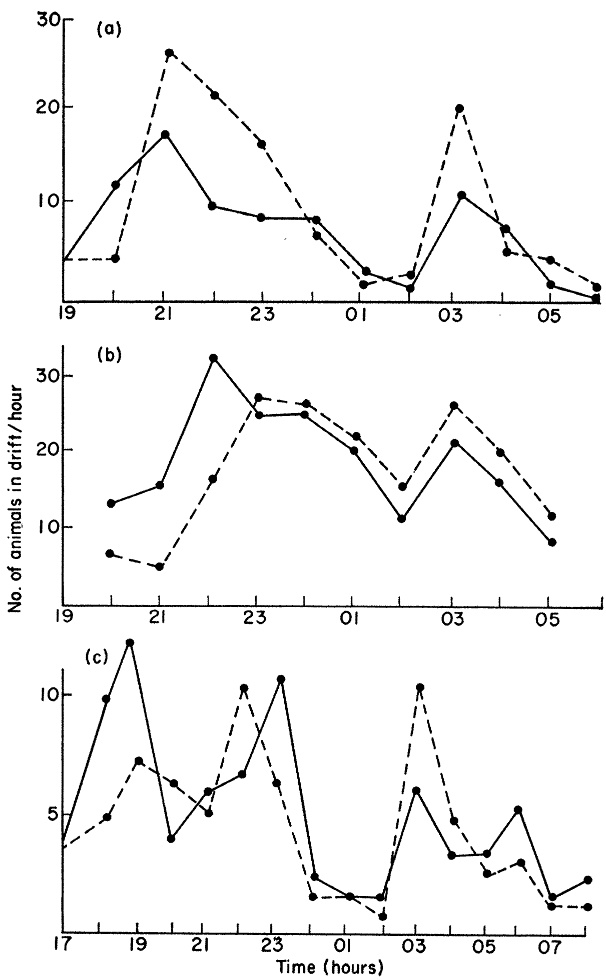

FIG. 2. Plots of the number of animals in drift per hour against time under full and no moon during (a) spring, (b) summer and (c) autumn. (The dashed line represents drift on a night of no moon.)

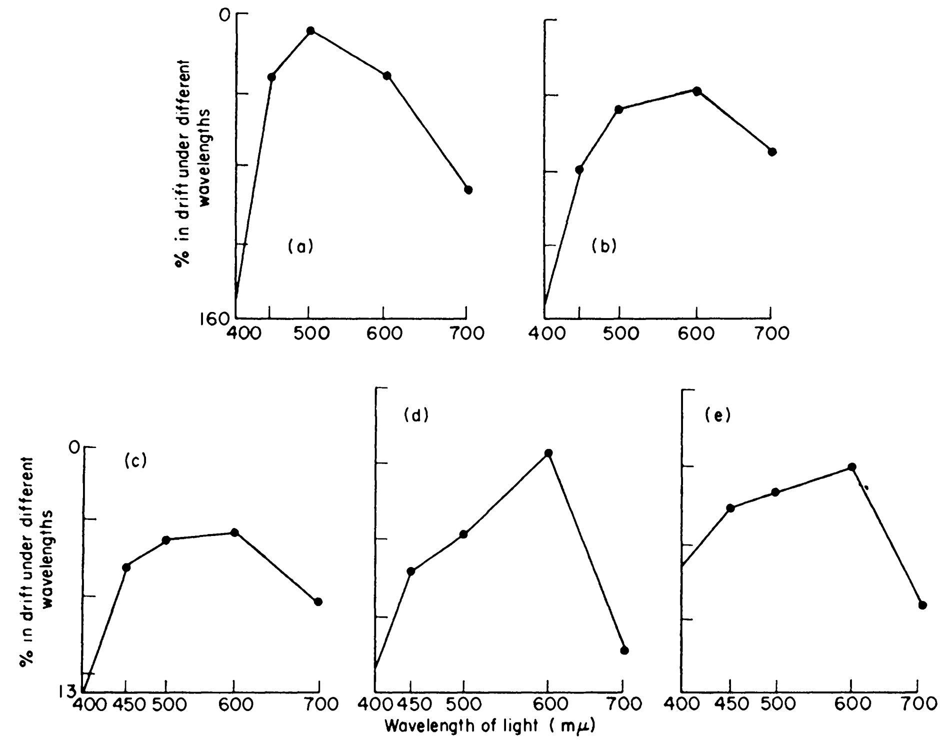

FIG. 3. Plots of the percentage of animals in drift under various known wavelengths to obtain an indication of the action spectra of animals involved in drift. (a) *Isoperla*; (b) *Baetis*; (c) *Protonemura*; (d) *Simulium*; (e) *Ephemerella*.

DISCUSSION

The seasonal change in threshold is probably related to variations in environmental conditions and/or changes in the physiology and behaviour of the various genera involved in drift. The sampling site was above any areas of dense human habitation and thus the chemical composition of the water was reasonably constant throughout the year. Also the site was chosen because of the sparse cover of macrophytes and therefore the benthic animals were not shaded from the direct effects of illumination even in midsummer. Thus probably the only two important seasonally varying environmental factors are temperature and the velocity of the water. A plot of the mean monthly air temperatures for Dartmoor (from figures kindly supplied by the Meteorological Office and taken at Hessary Tor) shows that the temperature is at its lowest in January (see Fig. 4), whereas a plot of the mean monthly field thresholds against time shows that the lowest threshold occurred in March. It would therefore appear that there is little possibility of the lowest temperatures affecting the threshold, although this does not eliminate the possibility of a relationship existing between high temperatures and maximum light thresholds in the summer months. A plot of the mean monthly water velocity (see Fig. 4) shows little relationship to any of the seasonal variations of threshold. Thus there is insufficient correlation between fluctuations in the physical environmental conditions and the light threshold to explain entirely the seasonal changes in the latter's value.

It is, therefore, necessary to examine the possibility of variations in the physiology and behaviour of the various species found in drift affecting the threshold, but this is beyond the scope of this present study. However, two lines of possible investigation are suggested. Firstly, that variations in the threshold are a reflection of differences between the thresholds of the genera involved in drift, and secondly that the activity patterns of the benthic fauna are seasonally modified. Evidence to support this latter hypothesis has been found by Elliott (1967b) who noted a correlation between the magnitude of drift and periods of maximum growth. He suggested that this relationship was perhaps a result of the animals being more active during periods of maximum growth.

Thus a much fuller sampling programme and complete measurements of all environmental conditions, coupled with more information on the interspecific variations in

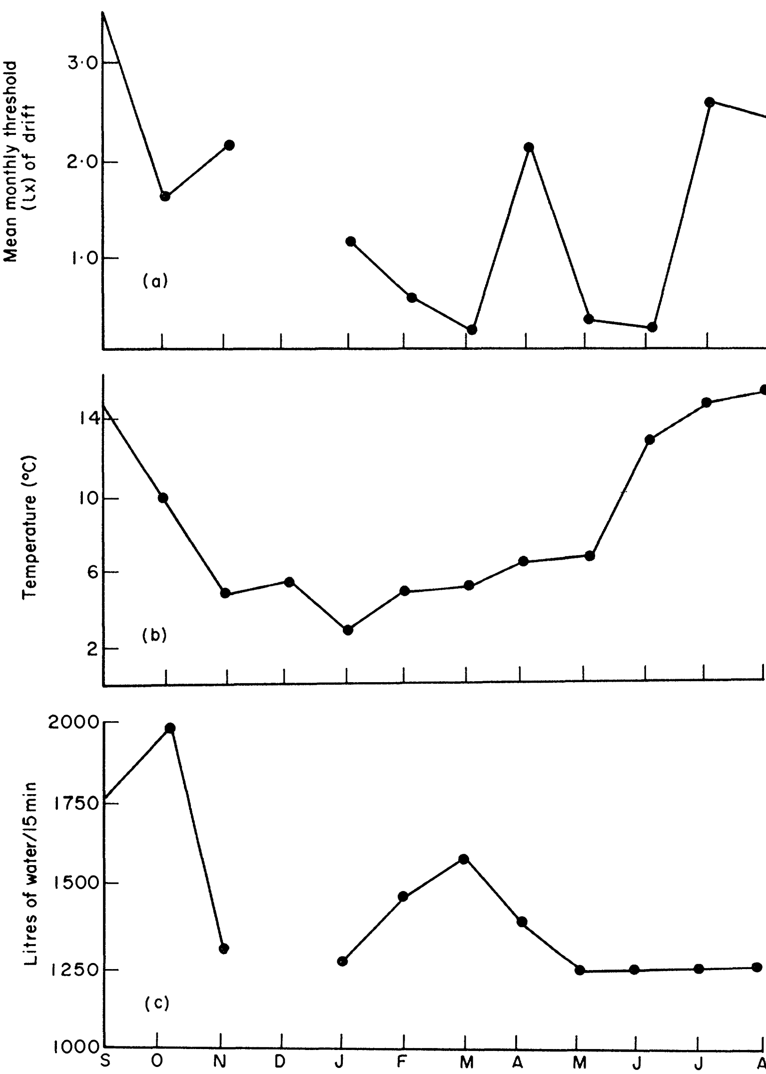

FIG. 4. Plots to compare the monthly variations in the field threshold with changes in the air temperature and water velocity of the stream. (a) Mean monthly light threshold of drift; (b) mean monthly air temperature; (c) mean monthly water velocity of stream.

physiology is required if the seasonal changes in threshold are to be explained completely.

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SUMMARY

1. The light threshold associated with the nocturnal increase in the level of invertebrate drift was investigated by field and laboratory studies.

2. Evening field samples were taken three times a month between September 1966 and August 1967 and it was estimated that the mean threshold was 1·584 lx.

3. The absolute threshold (i.e. the maximum level of light at which the nocturnal increase can commence), estimated by exposing insect larvae to controlled light conditions in the laboratory, was found to be 7·223 lx. The reasons for the difference between this figure and the field estimate are discussed.

4. Samples were taken to investigate the suggestion of other workers that moonlight can depress the level of drift. No difference was found, however, in this present study between the level of drift during nights of a full and no moon.

5. All measurements of light were made in lux and therefore a check was made to see if the aquatic animals were stimulated by light within the spectral range estimated by this unit of illuminance. Five genera of larvae were found to be most stimulated by light between 500 and 600 mμ and thus it was concluded that the use of lux was acceptable.

REFERENCES

Anderson, N. H. (1966). Depressant effect of moonlight on activity of aquatic insects. *Nature, Lond.* **209,** 319–20.

Borgh, T. (1927). The minutes of the 'Zoologiska Seminariet' of 22.XII.27 at the University of Uppsala.

Chaston, I. (1968a). Endogenous activity as a factor in invertebrate drift. *Arch. Hydrobiol.* **64,** 324–34.

Chaston, I. (1968b). A probe photometer. *Oikos,* **19,** 146–8.

Elliott, J. M. (1967a). Invertebrate drift in a Dartmoor stream. *Arch. Hydrobiol.* **63,** 202–37.

Elliott, J. M. (1967b). The life histories and drifting of the Plecoptera and Ephemeroptera in a Dartmoor stream. *J. Anim. Ecol.* **36,** 343–62.

Ide, F. P. (1942). Availability of aquatic insects as food of the speckled trout, *Salvelinus fontinalis*. *Trans. N. Am. Wildl. Conf.* **7,** 442–50.

Müller, K. (1954). Investigations on the organic drift in North Swedish streams. *Rep. Inst. Freshwat. Res. Drottningholm,* **35,** 133–48.

Müller, K. (1963a). Tag-Nachtrhythmus von Baetidenlarven in der 'Organischen Drift'. *Naturwissenschaften,* **50,** 161.

Müller, K. (1963b). Diurnal rhythm in 'Organic Drift' of *Gammarus pulex*. *Nature, Lond.* **198,** 806–7.

Müller, K. (1963c). Temperatur und Tagesperiodik der 'Organischen Drift' von *Gammarus pulex*. *Naturwissenschaften,* **50,** 410–11.

Müller, K. (1965). Field experiments on periodicity of freshwater invertebrates. In: *Circadian Clocks* (Ed. by J. Aschoff), pp. 314–17. Amsterdam.

Tanaka, H. (1960). On the daily changes of benthic animals in streams, especially the types of daily changes observed in the taxonomic groups of insects. *Bull. Freshwat. Fish Res. Lab., Tokyo,* **9,** 13–24.

Waters, T. F. (1961). Standing crop and drift of stream bottom organisms. *Ecology,* **42,** 532–7.

Waters, T. F. (1962). Diurnal periodicity in the drift of stream invertebrates. *Ecology,* **43,** 316–20.

Westlake, D. F. (1965). Some problems in the measurement of radiation under water: a review. *Photochem. Photobiol.* **4,** 849–68.

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