THE BIOLOGY OF *PEDICULUS HUMANUS*.

SUPPLEMENTARY NOTES.

(1) Colour and Light Reactions. (2) The influence on lice of temperature conditions in clothing and the absorption of radiant heat by cloth. (3) The influence of black, white, and coloured backgrounds and of coloured screens upon pigmentation in lice, proving that pigmentation is not hereditarily transmitted. (4) The relative proportions of the sexes.

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(With Plate X and 1 Text-figure.)

1. COLOUR AND LIGHT REACTIONS.

Only one author has hitherto touched upon the subject of the reactions of lice towards colour, namely, Galli-Valerio (xii. 1916, p. 35), who, learning from a Serbian doctor that black clothing is stated to repel lice, carried out one experiment wherein he placed the insects upon variously coloured paper and counted the number that wandered to each colour. The results recorded permit of no conclusions, no particulars are given regarding the manner in which the experiment was performed, and consequently it remains to be determined if lice react to colour.

Sources of experimental error.

In *Parasitology*, x. pp. 176, 100, reference was made to certain reactions to light that are exhibited by *P. humanus* and how they wander away from a source of light toward the shade. Indeed, I have found that they are so sensitive to different intensities of illumination that if light from a window falls upon them from one side, whilst it is reflected in upon them from the opposite direction by a mirror, the insects wander away quickly from the window side to the mirror side of the experimental cell in which they are confined although the difference in the intensity of the illumination from the two sides is but slightly appreciable to the human eye. It is therefore essential in testing the preference of lice for white, black or coloured backgrounds that the insects should be exposed to *vertical* illumination. Slightly oblique rays will invariably cause them to wander to one side; this being conveniently observed by placing the cell upon a stool with a top that is rotated periodically.

Since experiments conducted with single or but few insects would consume much time, it is best to use many lice, but their tendency to collect in a mass

and to remain clinging to each other requires to be counteracted, otherwise it may vitiate the experiment. The difficulty is met through separating the insects prior to each test and distributing them singly over the floor of the experimental cell.

Methods.

My experiments were conducted on bright sunny days in May, and mostly in the open air on the flat roof of a building. The apparatus employed (Fig. 1) was of a simple improvised character. The lice were confined beneath an

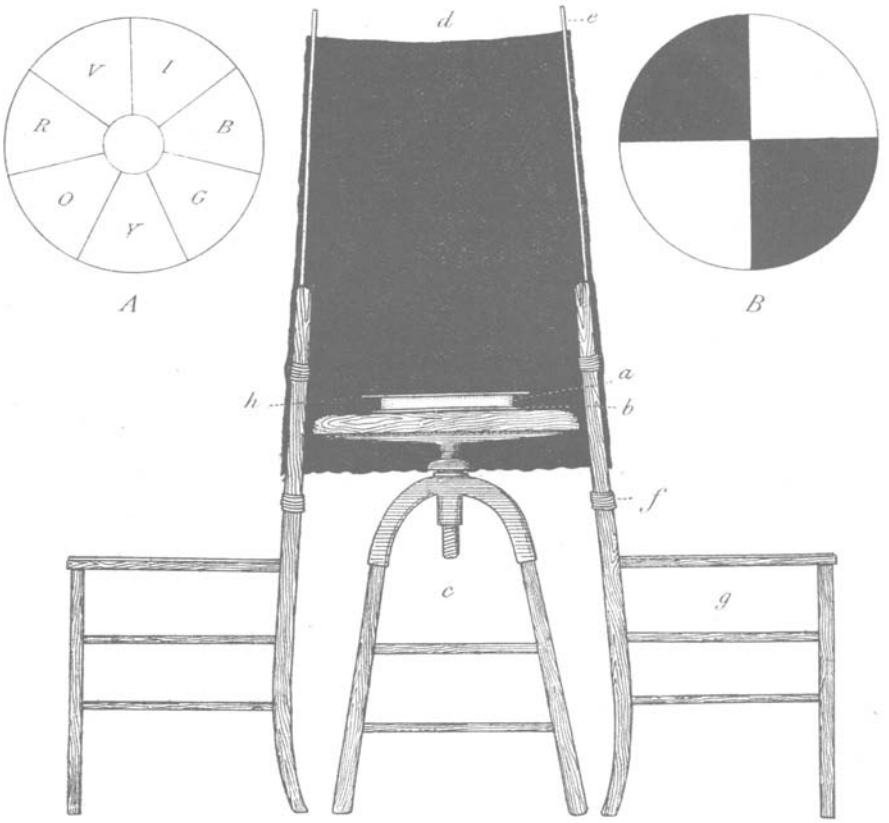


Fig. 1. Apparatus used for experiments on the reactions of lice to light and colour.

inverted, flat, circular glass dish, 22 cm. in diameter, with vertical sides 2 cm. in depth, hereinafter called the cell (Fig. 1 *a*). The cell rested upon a flat-topped stool (*c*) with rotating top. To prevent the light from falling otherwise than vertically upon the insects, the top of the stool was surrounded by a quadrangular funnel of dull black sateen (*d*), the funnel measuring ca. 60 cm. in height and 35 cm. across. The funnel was constructed by tying 4 rods of glass or bamboo (*e*) vertically by means of string (*f*) to the sides of the backs of two chairs (*g*) which were placed back to back on either side of the stool;

the sateen was wound smoothly around the rods and fixed in place with pins. The apparatus was placed in the shade of a low wall so that only bright diffuse light fell vertically into the funnel from the sky.

1. *Light and Shade.* To determine if lice seek the shade under vertical illumination, the cell was carpetted white, and half of it shaded by a black cover.

2. *Black and White.* To determine if lice prefer a black to a white background, a circular piece of white linen, with two quarters blacked with India ink (Fig. 1 *B*) served as a carpet for the cell.

3. *Colours.* Two methods were employed in testing the influence of colour[1]: (*a*) the cell was roofed by a transparent polychrome gelatin screen, being carpetted white, or (*b*) white light entered the cell from above, the carpet being polychrome. In both cases what appeared to my eye to be the colours of the spectrum were employed, the coloured areas being arranged radially (as shown in Fig. 1 *A*), a circular uncoloured surface, 5 cm. in diameter, being left in the centre. The dyes used for preparing the polychrome screen and carpet were the following:

	Dye	Wave lengths transmitted through the screen[2]
VIOLET	methyl violet	blue to w.l. 4900, and red beyond 6300
INDIGO	toluidine blue	indigo and blue to w.l. 4850
BLUE	methylene blue	indigo, blue and green to w.l. 5500
GREEN	lichtgrün	blue and green from w.l. 4600 to 5750
YELLOW	picric acid	whole spectrum except blue (down to 4700)
ORANGE	orange G.	green, yellow, orange and red below 5300
RED	trypanred	yellow, orange and red below Na line

The colour screen: seven tubes of ordinary 10 per cent. neutral nutrient gelatin, as used for bacteriological purposes, were warmed to melting point in a water-bath, and the requisite amount of each dye was added to each tube. A clean glass plate with lines of demarcation drawn thereon in melted paraffin, was placed on a levelling tripod, and each portion of coloured gelatin was poured over its apportioned area and spread with a glass rod. The paraffin lines set boundaries to the flow of the melted gelatin. After the gelatin had cooled and set, the glass plate was reversed over a shallow circular dish containing calcium chloride, whereby the gelatin became completely dried and presented the appearance of brilliantly stained glass.

The coloured carpet: corresponding lines to those described above were drawn on a piece of linen by means of a brush charged with melted paraffin.

[1] An attempt was made to use the spectrum from an arc light in a dark room, 25 lice being confined in a glass cell measuring 3×2 inches the floor of which was covered with the colours projected by the prism. The insects showed a definite tendency to wander to the walls of the box at the red and violet ends, which, however, may have been due to the obliquity of the rays and the insects seeking darkness. Mr Mills (*f. infra*) very kindly placed the apparatus at my disposal.

[2] I am much indebted to Mr W. H. Mills, M.A., for determining these wave lengths for me from photographs of the spectra kindly taken by Mr Frederick Stoakley at the Chemical Laboratory, Cambridge.

The linen was laid on a glass plate, and the various dyes were applied uniformly thereto by means of a feather upon the desired areas within the paraffined boundaries. The fabric became saturated with the dye, whilst the paraffin set limits to its spread. The surplus dye was absorbed with filter paper and the carpet was allowed to dry.

Experiments.

Light and Shade. Unfed and fed lice, in lots of 50, placed on a white paper carpet and illuminated vertically by a bright light, scattered uniformly on the floor of the cell, but after a time they gathered along the cell wall seeking to escape. When half of the cell was covered with black cloth, they sought the shade, only a few wandering about on the lighted surface.

White and black carpet. Unfed lice, in lots of 50, were placed on the black and white quartered carpet (Fig. 1 *B*) being illuminated from above. 80 per cent. were counted on the black and 20 per cent. on the white. When the apparatus was placed in a room and the light from the sky was reflected down vertically by a mirror, 81 per cent. were counted on the black and 19 per cent. on the white. The total counts in both series of observations were 491 black and 119 white.

Coloured screen. Unfed and fed lice in lots of 50, placed on a white paper carpet and illuminated vertically through the screen, gave the following counts after 15–20 minutes exposure:

	Set I. Lice unfed 16 hrs		Set II. Lice unfed 22 hrs		Set III. Lice fed 2-3 hrs before	
Colour	No. counted	%	No. counted	%	No. counted	%
Violet	104	52	33	22	82	41
Indigo	33	16·5	26	17·3	28	24
Blue	17	8·5	23	15·3	39	19·5
Green	13	6·5	14	9·3	17	8·5
Yellow	13	6·5	21	14	7	3·5
Orange	12	6	13	8·6	11	5·5
Red	5	2·5	20	13·3	16	8
In white centre	3		—		—	
Totals	200		150		200	

Coloured carpet. Two series of experiments were carried out, but they afforded no evidence that the lice were influenced by the colours on the carpet.

Conclusions.

The foregoing experiments demonstrate that lice, when illuminated by rays of light falling vertically upon them, seek the shade, and a black surface in preference to a white one. The experiments were conducted at 17–20° and 20–23° C.

When exposed upon white paper beneath a screen with bright spectral colours, either after fasting 16 hours (Set I) or 2–3 hours after feeding (Set III), the majority (52 per cent. and 41 per cent.) gathered beneath the violet, whilst

indigo and blue gave the next highest figures. In Set I the numbers fell steadily from violet down to red, in Set III the fall was less regular. In Set II, whilst the lice showed a preference for violet and indigo, they behaved irregularly toward the other colours, this being attributable probably to their wandering owing to hunger. The experiments with lice upon a polychrome carpet afforded no evidence of a preference for any particular colour on the part of the insects.

2. THE INFLUENCE ON LICE OF TEMPERATURE CONDITIONS IN CLOTHING AND THE ABSORPTION OF RADIANT HEAT BY CLOTH.

Galli-Valerio (xii. 1916, p. 35) was obviously wrong in attributing the reported inimical effect of black clothing to colour, for black is not a colour. *A priori* it appeared to me most probable that the effect would be found to depend upon the greater power of absorbing radiant heat possessed by black clothing when compared to that made of various coloured fabrics, it being a matter of common knowledge that black clothing may be intolerably hot when worn by persons exposed to the sun. White is worn in the tropics or summer because it reflects the heat rays that are to a greater or less extent absorbed by black and coloured fabrics.

The difference in the heat-absorbing capacity of clothing materials was studied by Krieger[1], who found that white fabrics (cotton, wool, etc.) acted very similarly. When their behaviour was compared to that of various coloured or black materials, a distinct difference was observable. If a white fabric be taken as 100, the following showed an ascending scale of absorbing power for radiant heat: pale sulphur yellow 102, dark yellow 140, light green 155, dark green 168, turkey red 165, light blue 198, black 208.

In this connection, I would refer the reader to *Parasitology*, x. pp. 89–92, 132, wherein the climatic conditions prevailing in clothing are considered and it is shown that a rise in temperature to ca. 35° C. may be distinctly inimical to lice (30–32° C. being favourable), that owing to the high temperature near the body in summer the insects tend to wander out upon ordinary garments, and that for the same reason they wander away from persons in fever. Moreover, it has been noted (*Ibid.* pp. 90, 579) that persons leaving temperate climates for the tropics may become freed from body-lice.

Experiments.

A double thickness of black serge was cut in the shape of a circle, one-half was covered and the other lined with white linen and the whole stitched together to form a mat fitting into a glass dish which rested on a black background. A number of lice were distributed evenly over the bottom of the dish and exposed to the sun's rays after the mat had been placed over the lice. Two experiments were made on a warm day with a total of 54 lice:

[1] Cited by Kratschmer in *Weyl's Handb. d. Hyg.* I. p. 379, Jena, 1896.

Those who regarded whites and negroes as belonging to different species, concluded that the lice found on them were also of different species because of their difference in colour. Murray, like others before him, found that lice taken from negroes were dark whilst those from whites were pale. He remarks that colour is of little value in differentiating species because it "may be derived from the nature of the feeding ground," some observers stating indeed that the negro's lice grow pale when transferred to whites and *vice versa*. The Rev. Hislop, "known as well for his scientific as for his missionary labours in India," informed Murray, "that at Nagpore he thinks he has seen dark Pediculi which have found their way from coloured nurses to white children, after a time becoming white," and another friend told him that "when seated in church behind two lads—the one dark-haired and the other light-coloured, both swarming with vermin—he noticed that those upon the dark-haired lad were darker than those upon the light-haired one."

That *Phthirus pubis* is likewise capable of changing its colour is indicated by Mr G. E. Bodkin (Government Economic Biologist, Science and Agriculture Department, Georgetown, Demerara), who, in a letter addressed to me (8. iv. 1918), states that the insect is very common in Demerara and that he "can speak from personal experience with regard to the chameleon-like qualities of these creatures; when transferred from a negro (where of course they exactly match the colour of the skin) to a white person, they will, in the course of a few generations become almost transparent and extremely difficult to see so closely do they resemble the white skin to which they are attached."

Whereas all specimens of *Phthirus pubis* collected from whites and examined by me have been found to be pale or slightly pigmented, those found on Suaheli negroes (N. 284 from Zanzibar, sent by W. Mansfield-Aders, and other lots) are deeply pigmented in the heavily chitinized regions, the whole integument being dusky.

The accuracy of Murray's statement regarding the variation in colour shown by lice on different races of man has been doubted by several authors. Schjödte (1864) noted the great variability in the colour of lice and so did Neumann (1910), the latter, however, records the presence of dark head-lice on negroes. Piaget (1880, p. 622) collected body-lice in Holland and saw great differences in the coloration of the specimens; he compared lice from Malaya and Europe and found them alike. Blanchard (1890, p. 437) refers to Murray and agrees with him in regarding the colour differences as devoid of specific significance, he nevertheless cites F. A. Pouchet (no reference; probably 1832–1841) as considering lice from negroes and whites distinct species.

Although not referring to Murray, Sikora (viii. 1915, p. 533) notes that variation in colour can be observed in lice taken from one person, three types, white, medium and dark being distinguishable to the naked eye. Peacock (1916, p. 32) examined 1800 lice (source and stages not stated) and found 5·4 per cent. to be "black."

Many authors, in seeking for differences between head-lice and body-lice, lay stress upon the colour. The head-louse is stated to have a dark or cindery gray colour, whilst the body-louse is light or pale coloured (Guérin, 1829–44; Cummings, 1915, etc.) but Schjödte (1864), Piaget (1880), Neumann (1910) and Sikora (1915) reject the colour difference as inconstant. Sikora (1917, p. 172), without giving the reference, cites Fahrenholz as lately having described *Pediculus capitis* var. *maculatus* from negroes, *P. capitis* var. *angustus* from Japanese, and *P. corporis* var. *marginatus* from Japanese, but makes no comments. Judging from Fahrenholz's other publications to which I shall refer elsewhere, this author has also in this instance merely burdened science with three useless names which will fall into the synonymy of *Pediculus humanus*.

Personal Observations.

A survey of some thousands of lice that have reached me from various parts of the world, has revealed that they possess different degrees of pigmentation which can be classified as follows; in the darkest specimens the markings (pleurae, bands, plates, etc.) appear blackish brown and the general integument dusky:

Capitis.

Very dark: N. 257, from Masai negro, German East Africa; N. 76, from negro, Obuasi, Ashanti; N. 244, from East Indian, Madras; N. 240, from Hill woman, Kasauli, India; N. 260, 263, East Indian, British Guiana; N. 231, from Arab, Khartoum, Sudan; N. 249, from native Indian, Lima, Peru.

Very dark to dark (mostly very dark): N. 254, from Suaheli, Zanzibar; N. 243, from East Indian, Madras; N. 229, from Tamils, Federated Malay States.

Dark: N. 235, from Badaga tribe, Nilgiris, India; N. 214, from Arabs, Algeria; N. 268, from Blackfoot Indians, Alberta, Canada; N. 146, from Chinese, Tientsin, China; N. 40, from Eskimo, Frobisher Bay, Baffinland.

Medium: N. 246, from Copper Eskimo, Coronation Gulf Region; N. 176, from Chinese, Szechuen, China; N. 226, from native children, Salonika and Seres.

Medium to pale: N. 208, 282, from grey-haired women (hair partly black or brown), lice mostly medium; N. 211, from soldier's pubic hair (brown), lice mostly medium; N. 181, 209, from children, mostly medium. All of these lice were taken from Europeans.

Corporis.

Dark: N. 225, from negroes and their clothes, Kibondo, Belgian Congo.

Pale to very dark (mostly pale): N. 216, from negroes' clothes, Nigeria; N. 277–281, from negroes, Nairobi, British East Africa.

Pale to dark (mostly pale): N. 217, from Negroes' blankets, Nyasaland; N. 269, from Blackfoot Indians, Alberta, Canada; N. 165, from Chinese, Szechuen, China; N. 213, from Arabs, Algeria. From Americans' and Euro-

peans' clothing: N. 228, Minneapolis, Minn., U.S.A.; N. 33, Liverpool; N. 204, Lambeth; N. 212, London; N. 252, Cambridge.

Pale to medium: N. 259, 261, 222, from East Indians, British Guiana; N. 245, from East Indian, Madras; N. 230, from Sudanese, Khartoum; N. 91, from negro, Biskra, Algeria; N. 45, from Japanese, Yokohama; N. 234, from Budaga tribe, Nilgiris, India.

Pale to slightly pigmented: N. 270, from Suaheli native, Zanzibar; N. 241, from Samper boy, Kasauli, India; N. 232, from Arab, Khartoum, Sudan; N. 90, from Arab, Biskra, Algeria.

Pale: N. 250, from native Indians, Cerro de Pasco, Peru; N. 233, from Arab, Khartoum, Sudan; N. 179, from Chinese, Szechuen; N. 40*a*, from Eskimo, Frobisher Bay.

NOTE. I am indebted to the following gentlemen for collecting or procuring the specimens mentioned in the foregoing list, relatively few having been collected by me personally: Messrs W. Mansfield Aders, G. E. Bodkin, A. J. Chalmers, J. W. Cornwall, J. Donovan, L. Dudgeon, Dr Graham, W. H. Hamer, F. Harker, C. Gordon Hewitt, F. Johansen, W. A. Lamborn, J. W. S. Macfie, J. L. Mitter, A. Owston, R. E. Ribeyro, W. Rose, P. H. Ross, G. G. Sampson, Ed. Sergent, W. R. Sheriffs, S. A. Stericker, C. Strickland, G. L. Tuck, A. M. D. Turnbull.

(*b*) EXPERIMENTS WHEREIN *P. HUMANUS* WERE RAISED ON BLACK, GRAY, WHITE AND COLOURED BACKGROUNDS.

Doubtless acting on the suggestion contained in Murray's paper above cited, Sikora (v. 1917, p. 172) carried out experiments regarding the influence of black, gray, and white backgrounds upon pigmentation in body-lice (*corporis*). In a subsequent paper, Sikora (IX. 1917, pp. 178–179) records similar experiments with head-lice (*capitis*). I have but recently been able to consult this author's papers, which possess considerable interest as throwing further light on the subject. Sikora states that lice maintained on black, gray, and white backgrounds become correspondingly coloured, irrespective of whether they are kept in darkness or exposed to light.

In this connection I have carried out the following experiments in Cambridge:

On 17. v. 1918, Mr A. Bacot kindly sent me some *corporis* (N. 265, 4 ♂, 6 ♀) which had laid some eggs on gray flannel in one of his breeding boxes. The specimens were either pale or moderately pigmented.

Experiment I (Black and White).

As the eggs hatched out (24–30. v. 18) the unfed larvae were placed daily in equal numbers in two pill-boxes so that finally each box contained 46 larvae. The pill-boxes were arranged as described by me in *Parasitology*, X. p. 107, with the difference that the one (*a*) was painted black inside with India ink,

the other (*b*) being left white, whilst instead of a hair-grid, a narrow strip of cloth was wound inside against the sides of the box. The white box contained white flannel, the black box contained black cloth, and its gauze top was blackened. The lice were fed twice daily on my Laboratory Assistant's arm, and, between feeds, were maintained at 31° C. in a glass-doored thermostat facing a window. But little light penetrated the boxes through their upturned gauze fenestra and the insects were disturbed as little as possible. The adults began to emerge in both boxes on the same day (5. vi. 18), and 77 out of 92 reached maturity, the black box containing 40 (18 ♂, 22 ♀) and the white box 37 (16 ♂, 21 ♀). The results in respect to pigmentation were as follows:

(*a*) *Black box*: all the adults (40) were very darkly pigmented, almost as dark as specimens of *capitis* collected from African negroes. The whole integument appeared ashy, whilst the heavily chitinized structures (sides of thorax, pleurae, plates, dorsal bands of male, etc.) were blackish brown and the head and legs dark. (See Plate X, fig. 1.)

(*b*) *White box*: all the adults (37) were pale, whitish or translucent, a few females showed moderately pigmented genital plates, sides of thorax and heads, but almost all had colourless pleurae and legs. (See Plate X, fig. 2.)

The eggs laid by the lice in boxes (*a*) and (*b*) were left *in situ*, hatched out at 31° C., and the larvae raised to adults without being disturbed (see Expt. III). The black adults from box (*a*) were transferred on 11. vi. 18 to white boxes and the pale adults (*b*) to black boxes (see Expt. IV).

Experiment II (Black, white, gray).

The same parent lice that were used in Expt. I, to the number of 2 ♂♂ and 4 ♀♀ were allowed to lay six batches of eggs in as many pill-boxes. Three boxes were placed fenestrum up in the thermostat whilst three stood bottom up with the gauze resting on the floor of the thermostat or on black cloth to exclude the light.

A. Boxes with gauze fenestra uppermost.

(*c*) *Black box*, contained 30 eggs laid 22–23. v. 18. Hatching began 29. v. and 16 adults emerged 8–11. vi. Examined 6–8 days after moulting, 15 appeared dusky and darkly pigmented, and 1 ♀ was moderately dark.

(*d*) *White box*, contained 31 eggs laid 23–24. v. 18. Hatching began 30. v. and 19 adults emerged 9–11. vi. Examined 6–8 days after moulting, 17 insects appeared pale and 2 females showed slight pigmentation at the sides of the thorax and pleurae.

(*e*) *Gray box*, contained 27 eggs laid 24–25. v. 18. Hatching began 31. v. and 19 adults emerged 12–16. vi. Examined 6–10 days after moulting, the colouration of each individual being separately noted with reference to a colour scale, the insects showed various grades of pigmentation. A few were very slightly pigmented, the majority moderately pigmented, but none were as dark as those in black box (*c*).

B. Boxes with fenestra undermost (partial darkness).

(*f*) *Black box*, contained 25 eggs laid 25–26. vi. 18. Hatching began 2. vi., and 7 adults emerged 11–13. vi. Examined 4–6 days after moulting, 6 of the adults appeared dark and 2 moderately pigmented.

(*g*) *White box*, contained 30 eggs laid 26–27. vi. 18. Hatching began 2. vi., and 11 adults emerged 11–14. vi. Examined 3–6 days after moulting, 8 adults appeared pale and 3 slightly pigmented.

(*h*) *Gray box*, contained 32 eggs laid 28–29. vi. 18. Hatching began 3. vi., and 7 adults emerged 14–17. vi. Examined 5–8 days after moulting, the insects (as checked by the colour scale) appeared as a whole slightly darker than those in lot (*e*).

Possibly owing to lack of ventilation, the number of lice raised to maturity in boxes B (*f–h*) was smaller than in boxes A (*c–e*), i.e. 25 from 87 eggs as compared to 54 from 88 eggs. Otherwise there was no material difference in the rate of development. In Expts. III–VI that follow, the boxes were stood in the thermostat with their fenestra uppermost.

Experiment III (second generation on black and white).

The eggs laid (*a*) in the black box by dark adults, and (*b*) in the white box by pale adults in Expt. I, were maintained in said boxes. In the black box, hatching began 15. vi. and adults began to emerge 26. vi. In the white box, the corresponding dates were 16. vi. and 28. vi. The hatching period was one day longer in the white box and the developmental period (1st larva to adult) was two days longer than in the black box. The adults in the black box were very dark, those in the white box pale, the moulted skins of the immature stages being correspondingly dark and pale. These lice, representing the second generation raised on black and on white respectively, were neither darker nor paler than specimens of the first generation similarly raised.

Experiment IV (transferring dark lice to white and *vice versa*).

The dark adults of Expt. I were transferred to a white box and the pale adults to a black box on 11. vi. 18, where they laid eggs that began to hatch 19. vi. The adults began to emerge 31. vi. The progeny of the pale lice raised on black were dark throughout and *vice versa*.

Experiment V (black and white).

Adults raised on black cloth and consequently dark, to the number of 2 ♂♂ and 3 ♀♀, were placed on 2. vii. 18 in each of six boxes arranged as described below. The adults laid eggs which began to hatch 10. vii., and the adult offspring began to emerge 21. vii. The offspring were examined on 23. vii. and all the moults of the six lots collected and separately mounted on slides.

Box No.	Outside of box and gauze	Inside of box	Cloth		Lice found in the box	Pigmentation
1	black	black	black	23	(5 ♂, 5 ♀, 13 L.)	●●●●●
				5	(5 L.)	●●●
					moults	●●●●● to ●●●●
2	black	white	black	22	(7 ♂, 6 ♀, 9 L.)	●●●●
				12	(4 ♂, 2 ♀, 6 L.)	●●●
					moults mostly	●●●●, few ●●●●● few ○
3	black	white	white	25	(6 ♂, 9 ♀, 10 L.)	○
				9	(1 ♂, 1 ♀, 7 L.)	● to ●●●
					moults mostly	○○, few ●●●
4	white	white	white	19	(4 ♂, 1 ♀, 14 L.)	○
				2	(1 ♂, 1 L.)	●
					moults	○, but 2 ●●●
5	white	black	white	17	(6 ♂, 5 ♀, 6 L.)	○
				14	(8 ♂, 1 ♀, 5 L.)	●●●
				3	(1 ♂, 2 L.)	●●●●
					moults mostly	○, few ●●●
6	white	black	black	31	(7 ♂, 7 ♀, 17 L.)	●●●●●
				3	(3 L.)	●●●
					moults	●●●● to ●●●

NOTE. The degrees of pigmentation are indicated as follows in the foregoing and succeeding records: ●●●●● = very dark, ●●●● = dark, ●●● = medium, ● = slight, ○ = pale to very pale.

Experiment VI (red and yellow).

Adults of the same lot as in Expt. V were put to lay (1. vii. 18) in two boxes with the inside, outside, gauze and contained cloth coloured (*a*) pale yellow with picric acid and (*b*) bright red with ink. The eggs began to hatch on the ninth day on yellow, on the eighth day on red, and the adults emerged 11 days later. The examination on 23. vii. revealed the following:

			Pigmentation
Yellow box contained	24	(6 ♂, 7 ♀, 11 L.)	○ or ●
		moults	○ or ●
Red box contained	35	(13 ♂, 7 ♀, 15 L.)	●●●●●
	2	(2 L.)	●●●
		moults	●●●●● to ●●●●

Experiment VII (lice on white, in complete darkness).

Dark adults of the same lot used in Expt. V, were put to lay on 2. vii. 18 in two boxes: (*a*) all black (like box 1, Expt. V) and (*b*) all white (like box 4, Expt. V). These boxes were enclosed in thick, tightly closing wooden boxes blackened inside and outside with India ink. The boxes were opened twice daily in the dark for the purpose of feeding the adults on three successive days during which they oviposited. The adults were then removed, and the boxes with eggs were returned to the thermostat for six days. After this, the hatching having commenced, the young lice were fed twice daily, the boxes containing them being applied to the arm in the dark room, the arm being swathed in

several thicknesses of camera cloth (black, lined red) during the half-hour feeding periods, when my laboratory assistant left the dark room. The lice were exposed to diffuse daylight for only a few minutes as nits (when the parents were removed), and once for 3–4 minutes when the box was inspected to see if the adults had emerged. The result was as follows when the insects were removed on 23. vii. 18:

			Pigmentation
(*a*) Black box (kept in partial darkness) contained	8	(3 ♂, 5 L.)	●●●●
	11	(3 ♂, 4 ♀, 4 L.)	●●●
	1	(♂)	●
		moults	●●●● and ●●●
(*b*) White box (kept in complete darkness) contained	34	(5 ♂, 14 ♀, 15 L.)	○ to ○○

N.B. Expt. VII *a* was vitiated by the entrance of light. It was intended that it should be carried out in complete darkness. Owing to the dry atmosphere in the thermostat, a minute pin-point crack developed in the lid of the outer wooden box, and light entered in sufficient amount to affect a small central area on photographic paper placed in the bottom of the box.

Experiment VIII (rate at which lice darken on black).

Second and third stage larvae that had been raised from the egg in a white box on white flannel and which were consequently pale or but slightly pigmented, were transferred on 25. vii. 18 to a black box containing black cloth. After 36–48 hours had elapsed, 5 adults had emerged and they were dark or moderately pigmented. On the day following some more dark adults and dark third stage larvae were found in the black box. Diffuse light entered the box whilst it was in the thermostat.

Experiment IX (black and white. Lice in absolute darkness).

On 29. vii. 18, seven pale third stage larvae were placed on black cloth in a black pill-box enclosed in a black wooden box as in Expt. VII, it being first ascertained that the latter was light tight. As an extra precaution, these boxes were enveloped in black cloth and the bundle placed in a third box. The lice were fed twice daily in a photographic dark room and were kept at 31° C. during the intervals between feeds. The insects were maintained inside the triple boxes except when being fed in the dark room through the gauze top of the innermost box.

The adults emerged on 30. vii. and all of them from the black as well as the white box were pale.

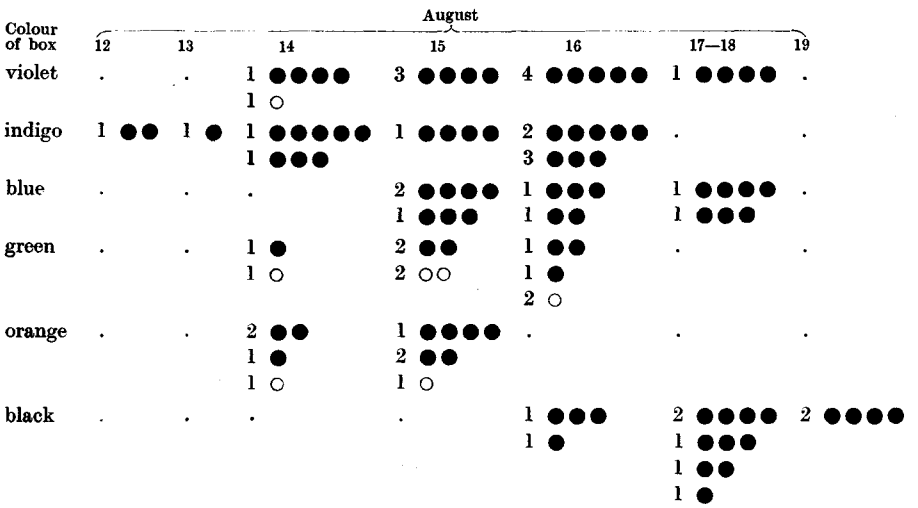
Experiment X (black and white. Lice in a bright light).

A number of first stage larvae were placed in two vertical glass tubes each plugged below by a cork upon which was spanned in the one case white linen in the other black sateen. They were exposed to bright light day and night for about two weeks. (The experiment ran concurrently with Expt. II as

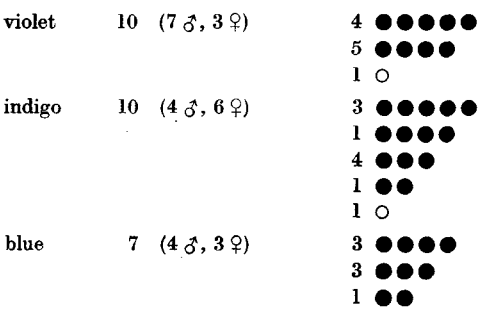
described on p. 216.) Of the seven adults raised on white four were pale, two very slightly and one slightly pigmented. But two adults were raised on black, one was dark and the other pale, the larvae that died whilst being raised were frequently darkly pigmented. The heat absorption by the black affected the insects adversely.

Experiment XI (different colours).

On 10. viii. 18, second and third stage larvae of the foregoing strain of lice were removed from an overstocked breeding box which contained white flannel that the insects had soiled. It was noticed that many larvae were darkly pigmented and this was attributed to (1) the lice being crowded, and (2) the box being soiled. These larvae were distributed in differently coloured boxes (tabulated below) and fed twice daily. The boxes were opened once every 24 hours to remove any adults that had moulted, these insects being isolated in boxes of corresponding colours for 24 hours at 31° C., thus giving them plenty of time to darken. The following results were recorded in respect to pigmentation:



Summing up the results, the different boxes yielded:



green	10	(7 ♂, 3 ♀)	1 ●● 4 ● 5 ○
orange	8	(6 ♂, 2 ♀)	1 ●●●● 4 ●● 2 ● 1 ○
black	12	(6 ♂, 5 ♀, 1 ♀̂)	4 ●●●● 5 ●●● 1 ●● 2 ●

The adults that emerged on green (especially) and on orange, were paler than the rest with few exceptions. Those on violet, indigo and blue would appear to have become progressively darker the longer they sojourned in their boxes prior to moulting. On the other hand the previous sojourn in the soiled stock box appears to have exerted an after effect on the insects kept on orange, if we compare the results with those of the following experiment.

To exclude any possible after effects of a previous sojourn on a background which would induce darkening, the following experiment was carried out.

Experiment XII (different colours).

Lots of 10 first stage larvae, the progeny of pale parents in a clean white box, were put in coloured boxes (tabulated below) on 23. ix. 18. On blue, violet and black, the first adults emerged on 3. x., and the last on 5. x. On green, indigo, yellow and orange, the first adults emerged on 4. x. and the last on 6. x., consequently there was no distinct difference in the rate of development.

Colour of box			Pigmentation in adult lice raised
violet	8	(6 ♂, 2 ♀)	7 ●●●● 1 ●●●
indigo	7	(5 ♂, 2 ♀)	4 ●●●● 3 ●●●
blue	8	(7 ♂, 1 ♀)	5 ●●●● 2 ●●● 1 ○
green	8	(6 ♂, 2 ♀)	1 ● 7 ○
yellow	10	(6 ♂, 4 ♀)	10 ○
orange	10	(4 ♂, 6 ♀)	2 ●●● 8 ♁
red	10	(8 ♂, 2 ♀)	10 ●●●●
black	7	(7 ♂)	9 ●●●● 1 ○ (killed young)

This shows more distinctly than Expt. X that lice raised on green, yellow, and orange do not become pigmented like the others.

THE INFLUENCE OF COLOURED RAYS OF LIGHT ON PIGMENTATION IN LICE MAINTAINED ON WHITE FABRICS.

Having established that lice do not become pigmented when maintained in absolute darkness, and that exposure to a very small amount of light suffices for them to become pigmented if maintained on black and red backgrounds, etc., it seemed of interest to observe if coloured rays of light exert an influence on pigmentation.

Method. Seven cylindrical glass specimen jars, ca. 12 cm. high by 5 cm. in diameter were partly filled with solutions of the seven dyes enumerated on p. 203. The jars were closed by corks of suitable size bored centrally to hold short test tubes which were pushed through up to the rim. The tubes thus became surrounded by a uniform layer of dye solution except at the neck. The jars were stood in alcoves made of cardboard boxes covered with white paper and facing the glazed door of the thermostat opposite a window, the tubes being capped by a piece of white cardboard. Each tube contained a strip of white flannel (ca. 3·5 × 0·7 cm.) standing vertically inside, to which the lice (second and third stage larvae, 10 per tube) that were introduced could cling. The lice were fed in pill boxes whose interior was coloured with the corresponding dye solutions to those contained in the jars, the insects being transferred to the boxes on the pieces of flannel.

Experiment I. Pale lice were maintained for 48 hours in the apparatus; they moulted therein and all of them were found to be pale or very slightly pigmented. Under the conditions of the experiment the coloured rays therefore appeared to exert no effect. The experiment was therefore repeated and varied, the insects being maintained a longer time exposed to coloured rays.

Experiment II. Newly emerged first-stage larvae were placed in the apparatus and raised to maturity whilst *exposed continuously to light.* During the day-time the light emanated from the window and during the evening and night from a 60-candle power electric lamp suspended close to the glazed door outside the thermostat. To ensure a maximum of illumination, a looking glass was placed at the back of the thermostat to reflect the infalling rays into the tubes that contained the lice, whilst the inside of the thermostat was lined with white paper. A piece of starched white cotton netting was placed in each tube for the lice to cling to in lieu of white flannel because the latter afforded a slight amount of shade. The experiment lasted two weeks.

The adults were carefully graded in respect to degrees of pigmentation and classified as follows, the sign × denoting separate individuals:

Colour of tube	Total adults raised	Pigmentation				
		dark	medium	slight	very slight	absent
Violet	9	×	× × ×	× ×	× × ×	.
Indigo	10	.	× × ×	× × ×	× × × ×	.
Blue	8	×	×	×	.	× × × × ×
Green	9	.	× ×	×	× ×	× × × ×
Yellow	11	.	.	×	.	× × × × × × × × × ×
Orange	12	.	.	×	× ×	× × × × × × × × ×
Red	9	.	×	×	×	× × × × × ×

CONCLUSIONS.

Section (a).

The most darkly pigmented *capitis* are derived from dark-skinned black-haired peoples. Head-lice tend to grow paler on yellow or moderately pale races possessing black hair. The palest head-lice are found on white races whose hair is often light in colour.

Whilst the darkest *corporis* are obtained from negroes, the insects in most cases cannot be grouped according to the skin colour of the host. The occurrence of pale body-lice on dark-skinned races is doubtless attributable to their wearing white or light coloured clothing especially in hot countries.

Phthirus pubis derived from negroes are darkly pigmented, whereas those derived from whites are pale for the same reasons that apply to head-lice.

The statement that lice may change colour when transferred from light to dark races and *vice versa* (Murray, Bodkin), in the absence of experiments on man, finds support amounting to confirmation in the experiments herein described wherein lice were raised on differently coloured, black, and white backgrounds.

It appears highly probable that paler head-lice will be obtained from light-haired than from dark-haired individuals among white races. I am at present collecting data in this connection and shall be grateful to any readers who may supply specimens or further information on the subject.

Note. The paper by Sikora (ix. 1917, pp. 278–279) has come to hand whilst this publication is going through the press. According to Sikora, *capitis* derived from black-haired persons are not always darker than those found on individuals with moderately fair hair, but the very fair haired always harbour distinctly paler lice. The contrast in the pigmentation of such lice is never so great as in the insects raised experimentally on black and white respectively. A very blond girl, bearing many pale head-lice, also harboured a few dark lice which were evidently derived from her (verminous) dark-haired mother.

Section (b).

The experiments detailed in this section show:

That pigmentation in lice is dependent upon the nature of the background upon which the insects are raised. If eggs laid by dark lice on white cloth are allowed to hatch and develope on white cloth, they will appear pale in all stages from the first larva to the adult (Expts. IV, V). Conversely, if eggs laid by pale lice on black cloth hatch and the insects be raised on black cloth, they will appear dark in all stages. Lice raised on a gray background show various degrees of pigmentation that are intermediate compared with the results obtained on black and white (Expt. II). It is immaterial if the pill-boxes in which the lice are raised are black or white externally (Expt. IV). The darkest lice were obtained in boxes blackened inside and containing black cloth, they were mostly less dark in boxes that were white inside and contained

black cloth (Expt. IV). Conversely, the palest lice were obtained in boxes that were white inside and contained white cloth, a few showed slightly increased pigmentation when the box was black inside and the cloth white (Expt. V).

Lice raised in the light on violet, indigo, blue and red, were as dark as those raised on black (Expts. VI (red), XI, XII), whilst those raised on green, yellow, and orange (Expts. VI (yellow), XI, XIII) were pale or slightly pigmented in the order yellow (acting like white), green and orange.

Lice kept in absolute darkness on black and white, do not show pigmentation (Expt. IX), but when maintained on black, a very small trace of light admitted to them causes pigmentation to take place (Expts. VII *a*, II *B*).

Pale larvae sojourning on black for 36–48–72 hours prior to moulting, show progressive degrees of pigmentation (Expt. VIII).

Dark adults transferred to a white background and pale adults transferred to a black background do not subsequently undergo modifications in pigmentation. Their coloration is fixed for life.

When many lice inhabit a white box, the colour of their bodies, gut contents and excreta darken the background with the result that the insects kept therein become more or less pigmented (Expt. XI and repeated experience).

Contrary to the experiments with variously coloured backgrounds, those in which lice were exposed on white flannel for 48 hours to the action of coloured rays of light did not show differences in so far as all remained pale or very slightly pigmented.

A longer exposure to coloured rays exerts an influence on pigmentation, for lice exposed continuously to coloured light whilst being raised to maturity were, as a whole, decidedly darker in the violet and indigo tubes than in the others. There were intermediate in the blue and green, and mostly pale in the yellow, orange and red tubes.

The experiments explain the great variation in the pigmentation of lice found on different races of man and variability in the pigmentation of lice occurring on white races, the colour of the skin, hair, and clothing, all exerting their effect on the insects. The statements are doubtless correct (*a*) that dark lice from negroes may turn pale when transferred to whites and *vice versa*, and (*b*) that head-lice on dark-haired whites are darker than on the fair-haired. The latter statement is being investigated.

The change in pigmentation of lice, due to the colour of the background, doubtless affords a measure of protection to these parasites in nature, especially in the case of man, who, like the monkey, uses his eyes in the search for the insects.

The observations recorded in Section (*a*), but especially in Section (*b*) show that Hindle (*Parasitology*, IX. p. 265) was unfortunately misled in his experiments designed to breed pigmented and unpigmented strains of *P. humanus*. Hindle readily reared colourless strains but had difficulty with dark ones, and

concluded that "there seems no doubt that the method of inheritance is alternative, but whether associated with the sex or not is not yet decided."

Pigmentation in *Pediculus* is not an hereditarily transmitted character, its presence depends entirely upon the nature of the background upon which the insect lives and it is a character that may be acquired in a couple of days (see especially Expts. IV and VIII). This phenomenon should be borne in mind by investigators engaged in the study of heredity.

4. THE RELATIVE PROPORTION OF THE SEXES.

In *Parasitology*, x. pp. 114–115, reference was made to the relative proportion of the sexes in *P. humanus* encountered (*a*) naturally upon the host and (*b*) in broods raised experimentally, and it was stated that further enumerations were required before reaching final conclusions on the subject. An examination of Bacot's hybrids of *capitis* and *corporis* has revealed the presence of a large number of hermaphrodites amongst them, a discovery that vitiates the figures cited (*loc. cit.*) from that author. A detailed study of these and other hermaphrodites will form the subject of another paper by Dr Keilin and myself.

ENUMERATION OF SEXES IN *PEDICULI*:

(*a*) *in insects encountered naturally upon the host.*

The following enumerations are based on specimens which have reached me from various parts of the world. The different lots contained numerous immature stages together with the adults.

No. of lot	Description	Total adults	♂	♀	⚥	% ♀
273	*corporis*, Sydney, N.S.W.	196	62	134	19	68
278	,, Nairobi, B.E.A.	364	141	223	1	64
279	,, ,, ,,	350	131	219	0	63
277	,, ,, ,,	245	95	150	2	61
274	,, Sydney, N.S.W.	103	40	63	0	61
257	,, Tanga, E.A.	173	71	102	2	59
236	*capitis* + *corporis*, Nairobi	358	148	210	0	59
237	,, ,, ,,	214	94	120	2	56
252	*corporis*, Cambridge	446	220	226	0	51
208	*capitis* ,,	208	104	104	0	50
282	,, ,,	35	6	29	0	*
209	,, London	39	14	25	0	*
285	,, Bogota, Colombia	38	7	31	0	*
	Totals	2769	1133	1636	26	59%

* These figures alone are too small to afford a reliable percentage. 0·5 % or over is reckoned as 1 % so as to give a round figure in the percentage column.

(*b*) *found in broods raised experimentally.*

The following enumerations are based on one lot (N. 265) of *corporis* raised in successive broods in Cambridge during August–October 1918 from a few parents kindly supplied by Mr A. Bacot. The breeding boxes contained black cloth, the insects were fed twice daily and maintained at 31° C. in the thermostat between feeds.

Date of emergence	Total adults	♂	♀	% ♀
Prior to following	357	167	190	53
" "	149	66	83	56
21. x.	210	136	74	35
22	71	44	27	*
23	31	18	13	*
28	80	46	34	*
30	54	24	30	*
Totals	952	501	451	47 %

* See footnote to previous table.

CONCLUSION.

In my previous paper (*loc. cit.*) it was stated that of 208 adult *capitis* collected by me from a woman in Cambridge 50 % were female; of 100 *capitis* raised by me 57 % were female, whilst of 944 *corporis* raised by Hindle roundly 60 % were female. (The first of these records is included in the table on p. 219.)

Of 2769 adult *corporis* and *capitis* collected for me in different parts of the world 59 % were females. Of 952 *corporis* raised by me experimentally in Cambridge 47 % were females.

It would appear from the tabulated figures that the proportion of the sexes varies, this depending on conditions which remain to be determined.

SUMMARY.

Refer to the conclusions stated at the end of the foregoing sections on pp. 204, 206, 217–219 and the foregoing paragraph.

REFERENCES.

See Bibliography in *Parasitology*, x. pp. 1–42, 582–586.

SIKORA, H. (ix. 1917). Zur Kleiderlaus-Kopflausfrage Vorläufige Mitteilung. *Arch. f. Schiffs- u. Tropen-Hyg.* XXI. 275–284, 3 figs.

CORRECTIONS.

Pp. 467, 468, 475, Pl. X, figs. 1-2. *The "Newman" Disinfector* was designed and an illustrated description of it published by Dr John C. Thresh in 1902 (see The "Emergency" Disinfector, *Brit. Med. Journ.* I. pp. 1606–1608, figs. 1, 2). Dr Thresh informs me that the perusal of my paper (*loc. cit.*) gave him the first intimation that his apparatus was being sold under the Trade name of the "Newman." The vendors of the latter might with advantage give credit to the designer in the circulars dealing with this apparatus.

P. 426, footnote. *Read* "Semon" *in place of* "Lemon." The error is due to a misprint in the publication quoted.

P. 581. "duddie" should be defined as meaning "ragged."

EXPLANATION OF PLATE X.

Pediculus humanus corporis. Specimens treated with caustic potash and mounted in balsam. Illustrating Experiment I (see text, pp. 209–210).

Fig. 1. 3 ♂ and 7 ♀ (10 out of 40 raised) maintained in a black pill-box. All the insects were darkly pigmented.

Fig. 2. 8 ♂, 7 ♀ (15 out of 37 raised) maintained in a white pill-box. Nearly all the insects are pale or but slightly pigmented.

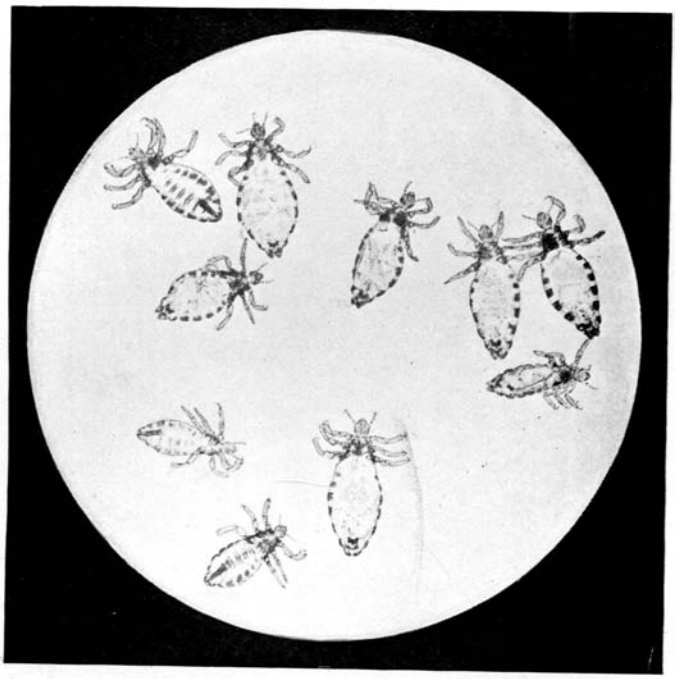


Fig. 1

Fig. 2