THE LIFE HISTORY AND BIONOMICS OF THE RED DEER WARBLE FLY *HYPODERMA DIANA* (BRAUER).

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INTRODUCTION

Three species of warble flies occur in Britain. Hypoderma bovis (de Geer) and H. Lineatum (de Villers) attack cattle and H. diana (Brauer) is a parasite of the Red Deer. H. (Oedemagena) tarandi (Latreille) has been reported on Reindeer at Edinburgh Zoo by MacDougall (1924) and again by the Reindeer Council of the United Kingdom (1951-52) at Rothiemurchus but this species has never become established.

H. bovis and H. lineatum are parasites of considerable economic importance in Europe, N. America and S. Asia (Scharff 1950 and James 1947). The reports of Bishopp et al (1926) and Scharff (1950) show how fully these species have been studied since the beginning of the century. The migrating first instar larvae have been followed along their respective routes towards the back. The majority of H. lineatum migrate in the connective tissues from the point of invasion to the oesophagus (Warburton 1922 and Bishopp et al 1926) after which they proceed to the back. Schmid (1941) reports that H. bovis migrates to the spinal canal along the nerves and then across the muscles to the back.

H. diana whose distribution is limited to parts of Europe has attracted the attention of very few workers compared with the bovine species. While Cameron (1932 and 1937) has contributed much to the knowledge of its life history, he was unable, through lack of material at the proper season, to describe the route pursued by the larva to the back. After examining/

examining twenty deer gullets, he suggested that H. diana resembled H. bovis in omitting this organ on its migration.

Interest in the subject was aroused by a recent paper by Kettle and Utsi (1955). Many Reindeer at Rothiemurchus were found to be attacked by the Red Deer Warble Fly H. diana. A post mortem examination of two reindeer which died revealed gelatinous streaks in the spinal cord, presumably caused by the migrating larva.

It is known that H. bovis and H. lineatum attack other than their normal hosts. While in some of these hosts, e.g. bison (Bishopp et al 1926) the larvae pursue their usual migratory route, in others, e.g. sheep (Dickmans 1948) the course is not always specific. No decisive conclusion can thus be obtained from the migration taken by the Red Deer Warble larva in the reindeer and it was decided to study the appearance of H. diana larva on the carcases of its native host with a view to establishing how it normally reached the back.

Life History

This is a summary of the principal features of the life history as published in two papers by Cameron (1932 and 1937).

He found that the adult flies are on the wing in May and June, the largest numbers appearing in the latter month. He suggested that the eggs are laid on the legs and that the larvae hatch out, burrow into the skin and migrate towards/

towards the back. In late December and January the first instar larvae pierce the skin of the back and moult. The second instar lasts about one month, when it moults to form the final instar. Mature grubs start to leave the host from the middle of March onwards. These black larvae fall to the ground, burrow a short distance into the moss or heather, and pupate. The pupal stage lasts at least a month, when the adults hatch out, mate and reinfest the host.

HOSTS AND DISTRIBUTION

Eichler (1941) lists Red Deer, Roe Deer, Fallow Dear, Elk and Chamois as being hostsof H. diana. Ullrich (1936) describes the warble parasite of the Elk as being H. alcis spec. nov., but further study by Eichler (1938) shows this to be synonymous with H. diana. It was at one time supposed that cattle were infested with H. diana (MacDougall 1934) but this is now considered unlikely. Attempts by Bosworth and Lapage to inoculate cattle with larvae of H. diana proved unsuccessful (MacDougall 1934). Cattle kept by stalkers in the heart of deer forests remain free from warble attack from year to year although the deer in the immediate vicinity are infested with the parasite. As already mentioned, the Reindeer brought to this country were attacked by H. diana (Report 1952) but it is doubtful whether the larvae mature fully in this host. James (1947) recalls the case of man being attacked by H. diana.

H. diana is confined to the Palaearctic Region. There are records of its having been collected in Scotland, France, Germany, Austria and Bulgaria (Dryenski 1933, Eichler 1941 and James 1947). Although species of deer are abundant in North America, there is no mention of H. diana in that continent. At the end of last century and the beginning of this century, several groups of red deer/

deer were shipped to New Zealand from Scotland and England but the warble fly has not become established in that country. The climatic change is probably the decisive factor against its development, as is the case with the related cattle species. Bedford (1926) and Lewis (1933) record that cattle grubs have not succeeded in establishing themselves in Kenya and South Africa after many introductions.

In parts of Europe, Red Deer are also attacked by H. acteon (Brauer), which is specific to them (Eichler 1941), but this species is not found in Scotland.

MATERIAL

Since this study was pursued from the middle of November onwards, the greater number of deer obtained were hinds. However some stags had been doing considerable damage in certain areas and it was possible to examine many of these marauding stags.

The deer were obtained principally from six forests around Newtonmore, Inverness-shire. The supply from each of these forests between 21st November, 1955 and 4th February, 1956 is listed below :-

Forest	Hinds	Stags
Adverikie) Dalwhinnie)	30	1
Coull		14
Gaick	13	
Glenfeshie	46	
Glenshira	7	
Quoich	6	6

Two hinds and one stag from two small estates, Chapel Park and Pitmain, were also sent in during this time. One Roe Deer Hind was stalked in Glen Tromie.

Towards the end of February two more marauding stags were kindly made available to me.

Most of the deer were skinned by the stalkers in the forests and the carcases collected and sent to a centre in Newtonmore. Unlike the cattle grubs, which are removed with the hide, the deer warbles remain on the carcase after skinning/

skinning. Cameron (1932), however, mentions how he collected larvae from several pieces of skin supplied to him, but only one or two larvae were found attached to very few of the many skins which I was able to inspect. Thus all the carcases examined could be taken as bearing their full complement of larvae. Only on one occasion were the larvae removed from the host by the stalker but the pockets left showed the position of the larvae and the number that were present.

In the early part of the season five oesophagi were obtained and throughout the season it was possible to examine the spinal canals of forty-seven deer.

At the beginning of November 1955 the gullets of sixteen cattle were acquired at the abattoir. These were examined for the presence of H. lineatum larvae. Only one oesophagus was infested with seven larvae.

Again at the end of April and beginning of May eight visits were paid to the abattoir for final instar cattle grubs. Approximately ten per cent of the hides were infested and from them forty-eight third instar and five second instar larvae were dissected.

TECHNIQUE

a) Obtaining Larvae

The stage of development, position and number of larvae were recorded on outline diagrams of the carcases and hides. As many larvae as possible were removed and preserved in 70 per cent alcohol.

First instar larvae had to be carefully dissected out but second and third stage larvae, enclosed in a sheath of connective tissue open at one end, were easily withdrawn with forceps.

Second and third instar cattle grubs were not so easily removed from the hides. The only successful method was to puncture the warble cyst on the inside of the hide with a blunt glass rod. Using the inserted rod to protect the larva, an incision was made along the length of the cyst to expose the larva for extraction.

Experience with the gullets of cattle showed that the most satisfactory method of examining this organ for the presence of first instar warble larvae was to "strip" them, that is, separate the muscular part from the inner mucosa. If the mucosa was then inflated by tying one end and filling with water, any larvae could be very clearly seen.

Carcases were split by passing a saw through the centre of the spine from tail to head. The spinal canal/

canal thus exposed suffered little damage and was easily inspected. The nerve cord was carefully stripped out and examined before being discarded. The epidural space and its fatty content thus opened to view could now be closely examined. It is this part that is frequented by the migrating H. bovis larvae (Gebauer 1940). The fat was always lifted and, when thick, teased apart in a glass dish.

b) Morphology

Most morphological features are minute and the use of a microscope is necessary throughout. When 1st instar larvae were kept in 70 per cent alcohol, the cuticle became opaque and this obscured some important details, such as spines and mouth-hooks, ~~in the first instar larvae.~~ If they were placed in very dilute KOH for three minutes the cuticle cleared sufficiently to distinguish the shape of the mouth-hooks. However, for critical examination it was necessary to remove the mouth-hooks and mount them in Euparal on a slide. The posterior spiracles were treated similarly.

c) Rearing adults from the larvae

At first, third instar larvae were placed on top of dry peat moss in a tray and then covered with a thin layer of this moss. After several days in a constant temperature room (65° F. and 35 per cent humidity) the peat moss was moistened slightly when many of the less mature larvae showed signs of desiccation.

The/

The pupae were kept in the same constant temperature room until the adults emerged. In an attempt to make them mate, the adults were removed to another environment of 77° F. and 70 per cent humidity for varying periods.

Test for Presence of Wax Layer in Cuticle

Wigglesworth (1947) described a test for the presence of a wax layer in the cuticle. Polyphenols in a cuticle devoid of wax react positively with 5 per cent ammoniacal silver nitrate (A.S.N.) but show no reaction when a wax layer is present. If the wax layer is dissolved away by boiling in chloroform the reaction with A.S.N. is renewed.

Second and third instar warble larvae were placed in A.S.N. for 30 minutes and the reaction noted. Where the reaction was negative the cuticle was boiled in chloroform for 20 minutes but if there was still no reaction with A.S.N. the boiling was continued for much longer periods. Smaller pieces of cuticle were also boiled in the chloroform to see if a better and quicker reaction could be obtained with the A.S.N.

MORPHOLOGY

The morphology of H. diana has been studied by several workers on a comparative basis with its related species H. bovis and H. lineatum. Brauer (1863), Brumpt (1936, Cameron (1937), Eichler (1940) and James (1947) are perhaps the most prominent contributors in this field. The details of the larval tracheal system are described by Walter (1922) and the external genitalia of the adult by Patton (1936).

The following descriptions are of larvae of the various stages collected from deer during the season and preserved in 70 per cent alcohol; of pupae which were formed from the mature third instar larvae in the laboratory and of adults which hatched from these pupae under artificial conditions.

It was at one time thought that Hypoderma spp. had five larval instars (Laake 1921) but it is now certain that there are only three stages (Knipling 1935).

Eggs

No records are obtainable of the eggs of H. diana having been observed. They are very difficult to procure since they are laid on the host at the time when deer are left entirely alone by the stalkers, and by the time the first stag is shot there is little chance of the egg cases still being attached to the coat. But the similarity/

similarity of this species generally to the bovine warble flies would suggest that the eggs were of comparable form. They would then be long and narrow, white to cream in colour and clasped to the hair at the end of a stalk.

<u>First Instar Larva</u> (Plate 1)

The ages of the larvae examined varied from approximately four months to those which were nearing ecdysis. Younger larvae were not obtainable at the commencement of this study.

The larva is transparent, white, cylindrical and of a uniform thickness throughout its length. Just prior to ecdysis it reaches a length of 12 mm. and a breadth of 2·1 mm. at the sixth segment. In the smaller and, presumably, youngest larvae of approximately 6 mm. in length, microscopic spines are evident on the ventral side and the anterior depressions of segments 2 - 6. These spines are not visible on larvae longer than 9 mm. The first instar larvae of other Hypoderma spp. are also noticeably covered with spines early in the instar but appear bare as they mature.

Spines are, however, always present at the oral end and around the posterior spiracles. There is a narrow transverse belt of spines lying at the level of the mouth-hooks, bearing in its middle zone an area of larger spines to form a more dense "band" junction. The length of this "band" in a larva of 12 mm. is 60 - 70 μ

The/

PLATE 1

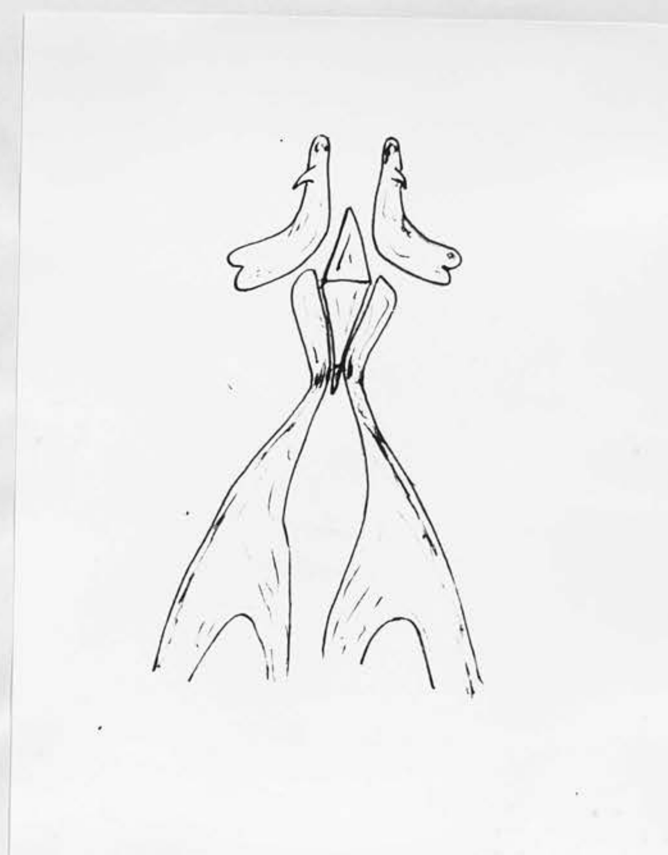
A - 1st Instar Larvae, removed from the carcase and preserved in 70 per cent alcohol.

B - Mouth Hooks of the 1st Instar Larvae.

PLATE I

A.

B.



The posterior spiracles are surrounded by several rings of dark brown spines or discs totalling about 90 in a 12 mm. larva.

The mouth-hooks are dark brown in colour and specific in their form. Their structure is seen in Fig. 1B The anterior end is directed forwards and is relatively blunt. The posterior end is divided by a shallow incision into two blunt lobes. Occasionally there is a small tooth present on the inner angle of the hook. The overall diameter of the mouth hooks is 24 - 30μ.

The transparency of the cuticle reveals the mid-gut occupying three-quarters of the body cavity. This is green in colour due to the presence of an enzyme known as "Hypodermatotoxin", which, according to Ono (1933), has proteolytic and histolytic properties and aids the larva in burrowing through the tissues. It is not found in the later instars.

Second Instar Larvae (Plate 2)

The second stage larva is white, semi-transparent and D - shaped in cross section, the ventral side being convex. The posterior segments are upcurved. The newly moulted larvae are "Indian-club" shaped, the narrow posterior end normally being inserted in a respiratory hole formed in the host's skin by the first instar larva prior to ecdysis.

Bands of blackish spines are distinct on many of the segments. The usual pattern is shown overleaf and any variation/

PLATE 2

A - 2nd instar Larvae. Ventral surface showing the dark bands of spines.

B - Cephalic region of 2nd Instar Larvae showing small chitinous piece in arch of mouth parts and the less distinct sensory tubercles.

PLATE 2.

A.

B.

variation from this is rare and slight :-

Ventral surface (Plate 2A)	:	Anterior and Posterior bands on segs. 2 - 7. Antero-lateral patches of spines on segs. 2 - 6. Postero-lateral patches of spines on segs. 2 - 5.
Dorsal surface	:	Paired patches of spines on each side of mid-line in segs. 2 - 3. Lateral patches in segs. 2 - 4. Posterior bands are undeveloped.

The posterior spiraces are composed of a number of discs which vary in number from 14 - 26.

Scharff (1950) discloses unpublished work of E.E. Knipling, in which he compares the cephalic regions of H. bovis and H. lineatum. Fig. 2B shows the corresponding features of H. diana. It is noticed that there is a small chitinous piece in the arch of the mouthparts similar to H. bovis, but the sensory tubercles correspond with those of H. lineatum in being barely distinct.

When the larvae are preserved in alcohol, the last segment bearing the spiracles turns brown to form a cap at the posterior end. This only occurs in second instar larvae.

Full grown second stage larvae are about 18 mm. long and 4 mm. wide at their greatest diameter.

Third Instar Larvae (Plates 3 and 4)

Immediately following ecdysis, the final instar is white and approximately 18 mm. long and 8 mm. wide. The spines at this stage are scarcely visible but as the larva/

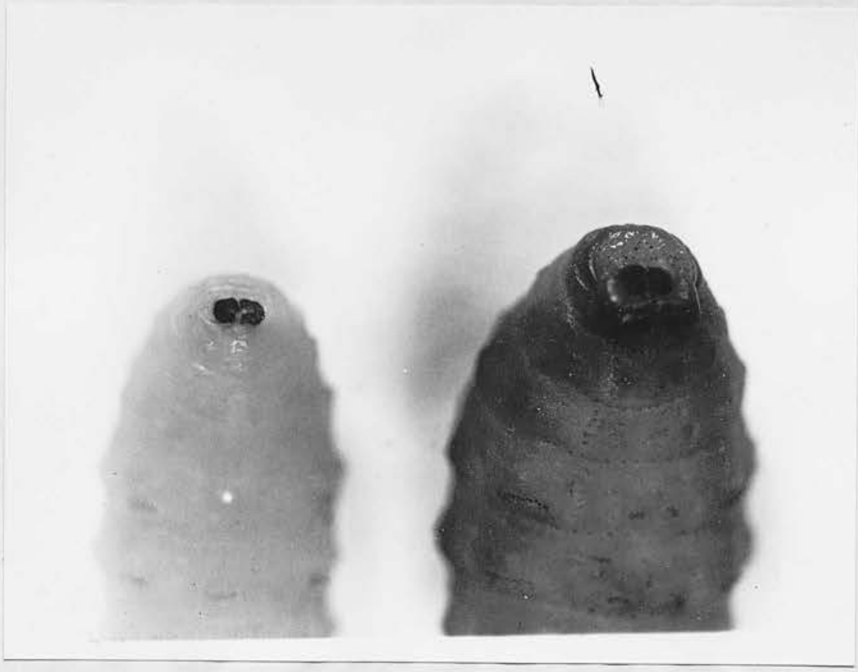
PLATE 3

A - 3rd Instar Larvae at progressive stages of maturity.

B - Spiracles of early and late 3rd Instar Larvae.

PLATE 3

A.



B.

larva matures the body colour merges through yellow and brown to black and the spines become dark brown. The full grown larva has a length of 25 mm. and a breadth of 8 mm.

Its long pear-shape is seen in ~~Fig.~~ Plate 4. The dorsal surface is flat, the ventral rounded and the posterior segments bearing the spiracles are upturned. The outline of the operculum is visible dorsally on the first four segments. There are three lateral ridges of conical shaped tubercles on segments 2 - 10.

The spines are arranged in a definite and specific pattern.

Dorsal surface (Plate 4A) :	As is usual in Hypoderma spp. this is relatively bare. Segs. 2 - 6 or 8 carry a small group or groups of fine forwardly directed spines in their middle anterior border. The hind edge, generally spineless, occasionally bears a narrow row of fine spines on segs. 4 - 6.
Lateral ridges :	Backwardly directed spines are visible on the anterior borders in segs. 2 - 9. Posterior border of dorsal and middle lateral tubercles is naked. On the hind borders of the ventro -lateral tubercles there are one or two groups of forwardly directed spines in segs. 2 - 8 and occasionally in 9.
Ventral surface : (Plate 4B)	One to four groups of backwardly directed spines on the anterior surface of segs. 2 - 10. One group of forwardly directed spines on the posterior border of segs. 2 - 9.

The/

PLATE 4

A - Dorsal surface of 3rd instar larvae showing the arrangement of the groups of spines.

B - Ventral surface of 3rd instar larvae showing spinal arrangement.

PLATE 4.

A.

B.

The colour of the posterior spiracles is light brown turning to black with maturity. They are kidney shaped, divergent and funnelled inwards towards the pseudostigmatic orifice. Radiating furrows are very distinct in the younger specimens. (Plate 3B)

Pupa (Plate 5A)

The puparium has all the features of a stout third instar larva. It is barrel-shaped, black and shiny. The first four segments bearing the operculum are somewhat more expanded and curve upwards. The dorsal surface is flat and the ventral surface rounded. Length 17 mm., breadth 9 mm.

Adult (Plates 5B and 6)

The female is bee-like in appearance. Basically the body is black but it is clothed with yellow hair. This is darker and less plentiful on the middle dorsal part of the abdomen but lighter and more closely set on the head. The mesonotum bears four polished longitudinal stripes interrupted at the suture. The scutellum has deep emarginations at the apex and at the side, thus making it four-lobed. Apart from the femora which are blackish, the legs are generally yellowish brown in colour. The body of the female terminates in a tubular ovipositor consisting of four telescoping sections and is forked at the end. The length of the extended ovipositor is approximately equal to the abdomen and thorax together.

As/

PLATE 5

A - Ventral and Dorsal view of pupae. Operculum has been removed to show the adult insect still intact.

B - Male and Female adult flies - dorsal view.

PLATE 5

A.

B.

As is common with the other species of Hypoderma, the mouth parts are vestigial. The eyes are wide apart and relatively small. The length is about 12 mm.

The male is similar to the female but may be conveniently distinguished by the end of the abdomen, which is rounded, and the eyes which are larger and more closely approximated.

PLATE 6

A - Side and Facial view of Adult Male.

B - Side and Facial view of Adult Female.

PLATE 6.

A.

B.

RESULTS

Oesophagus

In agreement with Cameron (1937), no larvae were found in any of the five oesophagi examined.

The following table describes the deer from which the gullets were obtained.

Source	Ref.	Description	Date	Subdermal Infestation
Dalwhinnie	4D	8 yr. old milk hind	22.11.55	4 first instar 2 cysts
Dalwhinnie	1	1 quarter stag	22.11.55	9 first instar 2 second instar
Gaich	6	75 lb. yeld hind	24.11.55	No larvae
Glenfeshie	19	Very old yeld hind	1.12.55	No larvae
Glenfeshie	20	8 yr. old yeld hind	1.12.55	No larvae

The larvae of <u>H. lineatum</u> are found in the mucous membrane of the cattle gullets from July to November (Schmid 1939). The corresponding stages of the life history are comparable in <u>H. diana</u>, so that if the larvae normally went to the gullet, they would have been expected there in the deer from Dalwhinnie, which were known to be infested.

The other internal organs were examined in as much detail as possible but no larvae were found on them. Seguy (1928) says that second stage (our first stage) larvae live in the gut wall until November, but I was unable/

unable to confirm this.

Spinal Canal

Forty-seven carcases were split throughout the season for examination. In no case was a larva or any sign of larvae visible in any of the canals.

Bishopp et al (1926) mention how the presence of greenish gelatinous streaks betrays the position of H. bovis larvae in the neural canal. Mote (1928) also states how H. bovis larvae pass between the periosteum and dura mater, leaving behind a gelatinous streak. Post mortem reports on the reindeer which died point out that in both cases red gelatinous material was present in the epidural space. Only one of the spinal canals examined showed any sign of irritation. This was in the form of extensive areas of red matter actually enclosed within the dura mater, but the cause seemed to be something other than migrating larvae. The rest bore no unusual markings.

The list below shows that approximately 45 per cent of the deer, whose spinal canals were examined, were infested with warble larvae. In such cases it would be expected that had the migratory route of H. diana larvae to the back been by way of the neural canal, some trace of their former movements in this area would still be visible. But this is lacking and it would appear that, if the larvae do migrate, they must pursue some other course than that followed by either/

either H. bovis or H. lineatum.

A list of the hinds and stags examined in this section is given below to show their actual infestation by larvae in other parts of the body :-

Source	Ref.	Date	Weight or Age		Infestation 1st Instar	2nd Instar	Cysts
Hinds							
Adverikie	3	21.11.55	79	lbs	1	-	-
Glenfeshie	1	"	57	"	44	-	-
"	11-14	22.11.55	av.83	"	-	-	-
Gaick	5	"	99	"	-	-	-
"	6	24.11.55	75	"	-	-	-
Glenfeshie	15-18	28.11.55	av.86	"	-	-	-
Adverikie	7	9.12.55	93	"	-	-	6
"	9	"	95	"	1	1	60
"	10	"	80	"	-	-	4
Gaick	7	12.12.55	75	"	-	1	1
Glenfeshie	21	"	90	"	-	-	-
"	22	"	108	"	-	-	-
"	23	"	78	"	5	2	26
Gaick	8	16.12.55	2	yrs.	2	2	-
"	9	"	75	lbs	-	-	-
Chapel Park	2	23.12.55	80	"	1	4	2
Glenfeshie	24	28.12.55	80	"	-	3	-
"	25	"	85	"	1	1	-
Gaick	10	4. 1.56	65	"	-	3	-
Glenfeshie	31-34	11. 1.56	av.78	"	-	-	-
Stags							
Coull	1	4.12.55	107	lbs	-	-	-
"	2	"	86	"	24	3	2
"	3	"	85	"	10	-	-
"	4	12.12.55	106	"	-	-	-
"	5	15.12.55	113	"	2	2	-
"	6	"	115	"	-	-	-
"	7	"	106	"	-	-	-
"	8	"	109	"	1	-	300
"	9-10	23.12.55	105	"	-	-	-
"	11	"	100	"	-	13	9
"	12	"	110	"	-	8	-
"	13	28.12.55	95	"	-	-	-
"	14	"	88	"	-	3	-
Pitmain	1	4. 1.56	70	"	1	22	1
Quoich	1	"	78	"	-	10	15
"	2	"	99	"	-	4	1
"	3	"	70	"	-	14	4

Migration/

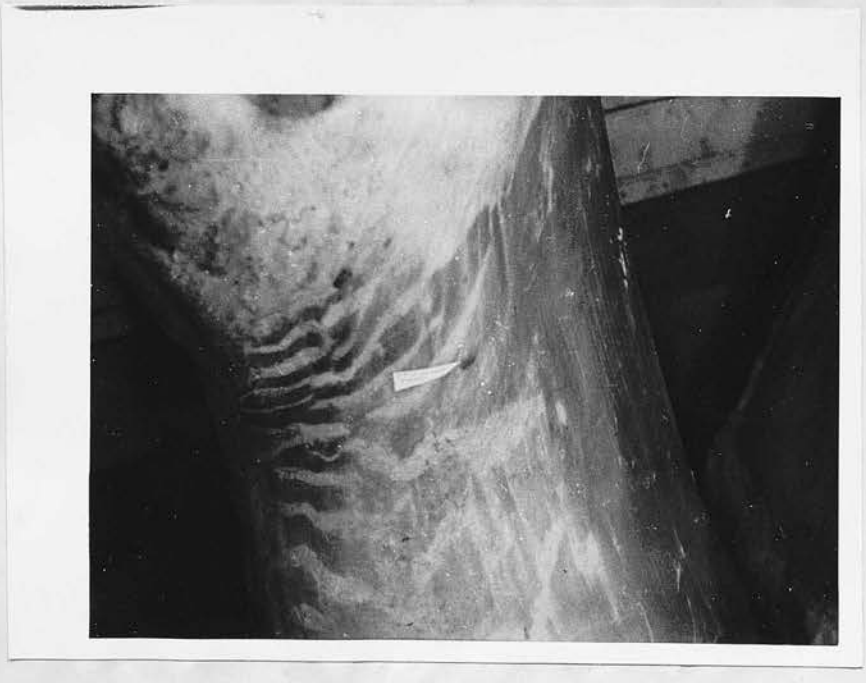
PLATE 7

A - Stag carcase skinned and hung up ready for examination.

B - Close-up of left side rear flank showing 1st instar migrating larva and newly encysted 1st instar larva.

PLATE 7.

A.



B.

Migration

Although deer were not available to me before the middle of November, at which time most of the first instar larvae would be about three-quarters developed, a comparison of the records obtained gave the impression that there was a definite migration of the larvae from one part of the body to another. The route pursued is confined to the subdermal tissues and between the muscle sheaths.

In having a period of migration it is in contrast to H. crossi (Patton), the three larval stages of which are passed subdermally in the back of goats (Patton 1922), but similar to H. (Oedemagena) tarandi which avoids the body cavities and moves about just below the skin and in the muscles. (Bergman 1917, Hadwen 1926 and Breevui 1946)

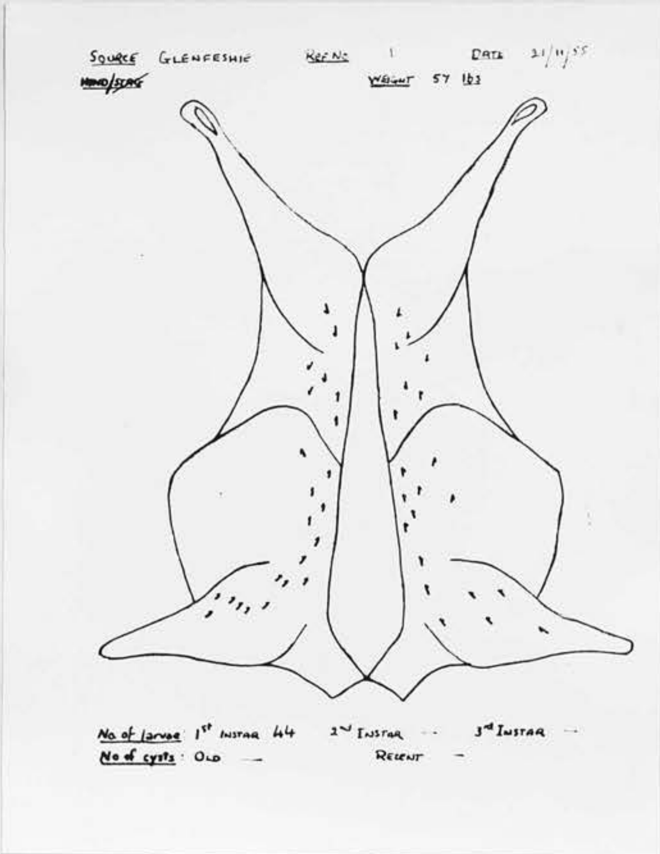
Plates 8A, 8B, 9A, 9B and 10A give examples of what was found. No visible tracks are left by these larvae and there is no way of telling exactly from where they have come. By comparing diagrams such as these, on which is plotted the position of each larva and the direction in which it is facing, a definite route can be established. Most of the larvae plotted are very clearly seen in the external connective tissue, but many of them just appear as thin dark streaks, actually the outline of their mid-gut, between the muscle sheaths. There is the possibility that many may have been contained deeper in the muscles but, when the number of first instar larvae found in the deer/

<u>PLATE 8</u>

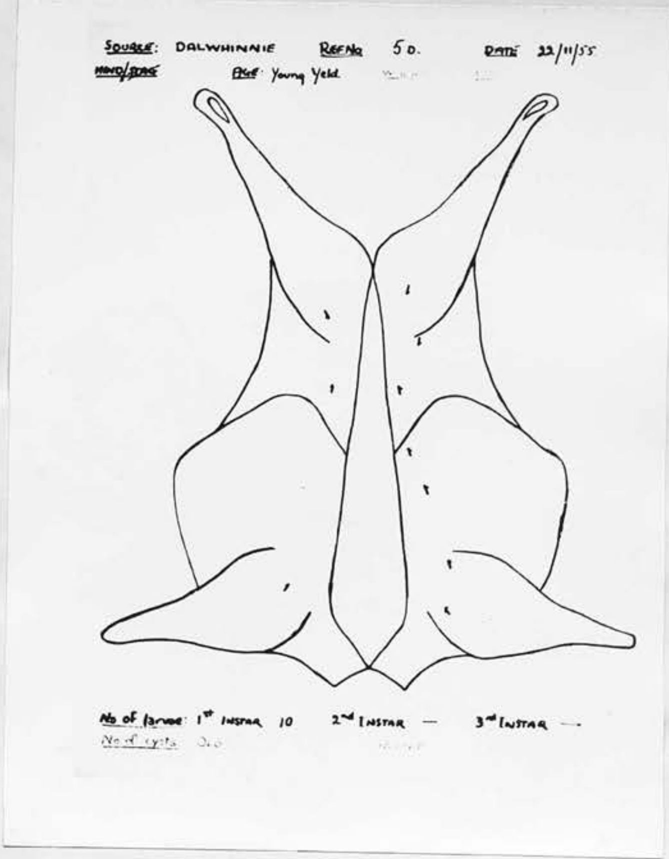
A - Diagram of Glenfeshie hind showing position and direction of travel of migrating lst instar larvae.

B - Diagram of Dalwhinnie hind showing similar condition to A.

PLATE 8.


SOURCE GLENFESHIE REF NO 1 DATE 21/11/55
HIND/STAG WEIGHT 57 lbs
No of larvae: 1st INSTAR 44 2nd INSTAR -- 3rd INSTAR —
No of cysts: OLD — RECENT —

A.


SOURCE: DALWHINNIE REF NO 5 D. DATE 22/11/55
HIND/STAG AGE: Young Yeld
No of larvae: 1st INSTAR 10 2nd INSTAR — 3rd INSTAR —
No of cysts: OLD

B.

deer stalked at the beginning of the season was compared with the total second and third instar present in later hosts, it was decided that only a small number, if any, could have been missed during inspection.

Diagram 8A shows the earliest stage obtained. There are a number of larvae moving up the foreleg, over the shoulder and along the back. Those lower down on the foreleg were actually contained within the muscle sheaths but the remainder were in the connective tissue. The direction of movement of these larvae, apart from the few at the rear end, is, first of all, upwards towards the spine and then along the flanks towards the rear. Those on the thighs appear to have traversed up the hind legs towards the spine and then in a forward direction.

In diagram 8B a similar situation is seen but with fewer larvae. Apart from one larva, all were found within the muscle sheaths.

In diagrams 9A and 9B the larvae have moved down from the spinal region over the flanks of the animal, where they assume their final position, pierce the skin and moult to form the second instar.

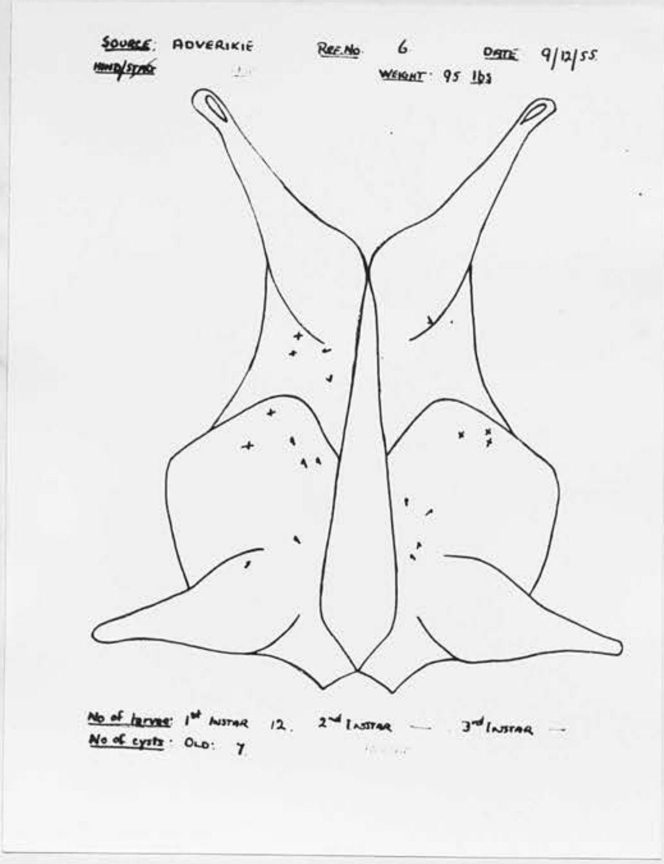
Approximately 75 per cent of the first instar larvae were either found on the forelegs or migrating in an anterior-posterior direction. In no case was a larva noticed to migrate in a dorsal direction over the thorax or abdomen. This would suggest that the flies lay their eggs on the legs of the host, the forelegs being more/

PLATE 9

A - Diagram of Adverikie Hind in which 1st instar larvae are moving down the flanks to their final position. The crosses (X) mark the larvae which have become encysted.

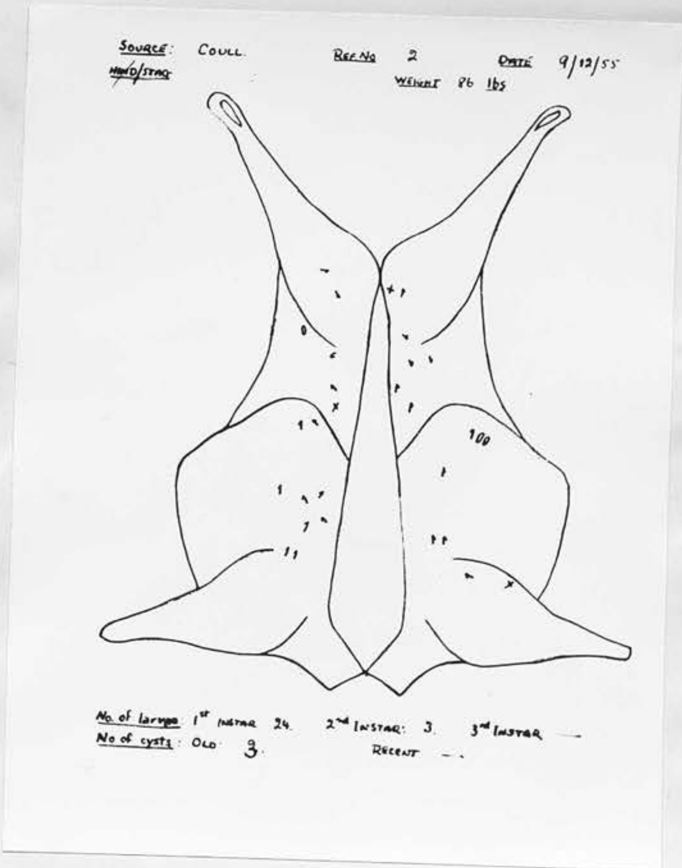
B - Diagram of Coull Stag showing a slightly later stage in which two of the 1st instar larvae have moulted to form 2nd instar larvae (O).

PLATE 9.


SOURCE: ADVERIKIE
REF.NO. 6
DATE 9/12/55
HIND/STAG
WEIGHT: 95 lbs
No. of larvae: 1st INSTAR 12. 2nd INSTAR — 3rd INSTAR —
No. of cysts: OLD: 7.

A.

B.


SOURCE: COULL.
REF.NO. 2
DATE 9/12/55
HIND/STAG
WEIGHT 86 lbs
No. of larvae: 1st INSTAR 24. 2nd INSTAR: 3. 3rd INSTAR —
No. of cysts: OLD 3.
RECENT —.

more often chosen than the rear.

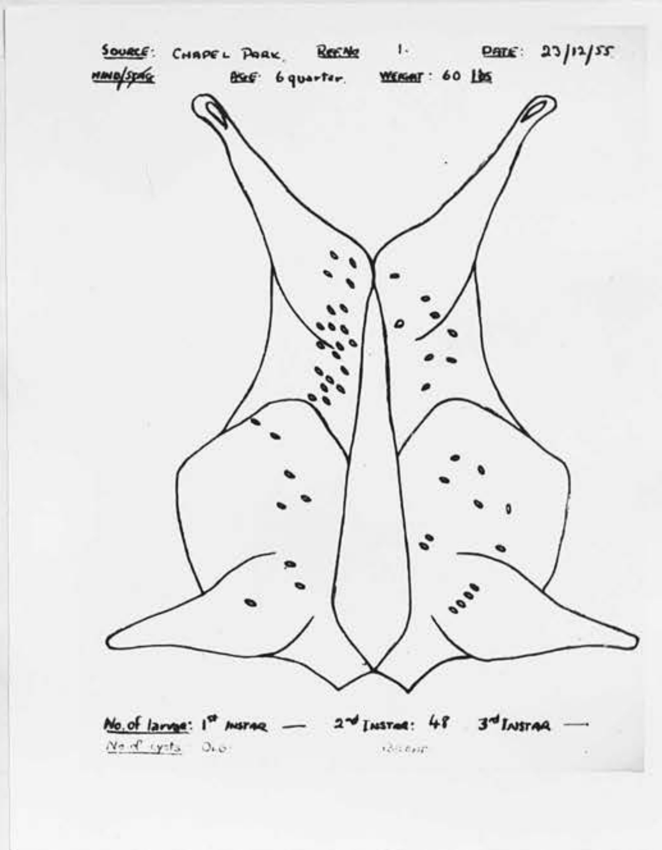
Bishopp et al (1929) state that approximately two and a half months intervene between egg laying and the earliest appearance of young H. lineatum grubs in the gullet. Because of the small distance covered four months after egg laying, it is felt that the young larvae must remain in the lower parts of the legs for a similar time before commencing their migration to the back.

It cannot be assumed that all the larvae on any one carcase are the same age, nor have they a common destination as is shown in diagram 10A, which illustrates the final position taken up by the larvae at ecdysis. Therefore, an analysis of measurements of larvae in different positions is unlikely to be helpful in establishing a migrating pattern.

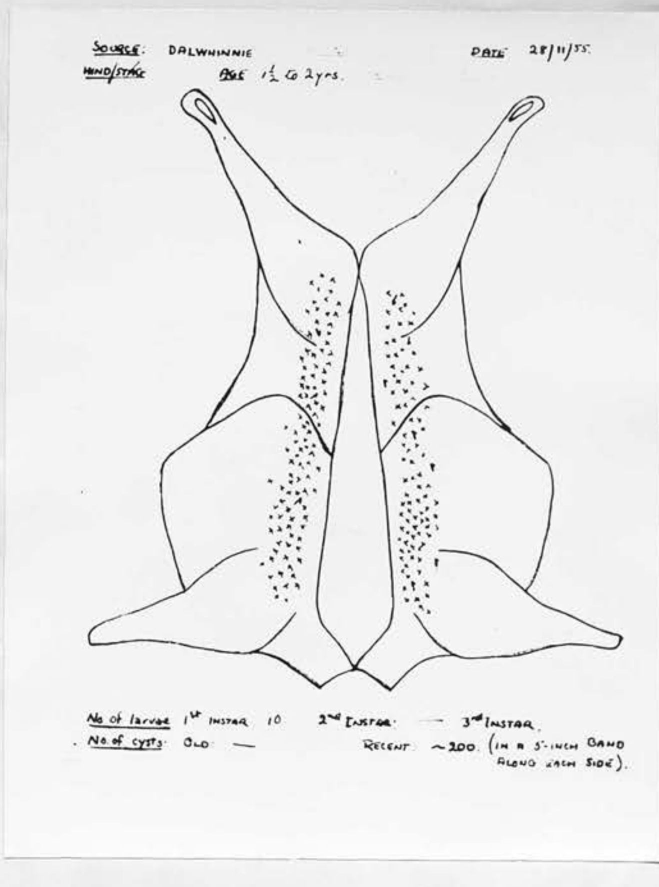
According to Cameron (1937), first instar larvae occur as late as the end of January. The last first stage larva in this study was recorded on the 15th December on a stag and on the 5th January on a hind. The first second instar larva was found on a stag on the 22nd November and on a hind on the 9th December. The egg laying season is principally May and June. In the absence of the eggs, their incubation period can merely be a matter of conjecture, but, by comparison with the eggs of the cattle warble flies, it should be less than one week. It would then follow that the first instar of

H. diana/

PLATE 10.


SOURCE: CHAPEL PARK. REF.No 1. DATE: 23/12/55.
HIND/STAG AGE: 6 quarter. WEIGHT: 60 lbs
No. of larvae: 1st INSTAR — 2nd INSTAR: 48 3rd INSTAR —
No. of cysts: OLD

A.


SOURCE: DALWHINNIE DATE 28/11/55.
HIND/STAG AGE 1½ to 2yrs.
No of larvae 1st INSTAR 10 2nd INSTAR: — 3rd INSTAR.
No. of cysts: OLD — RECENT ~200. (IN A 5-INCH BAND ALONG EACH SIDE).

B.

H. diana lasts about six months.

Summary of Migration

The first instar larva presumably penetrates the skin in the lower part of the leg, the forelegs being more commonly attacked than the hind legs. During its journey to the back it does not approach the gullet, nor yet enter the spinal canal, but follows the line of least resistance subdermally or intramuscularly. It is to be found moving upwards over the shoulder and then posteriorly parallel to the spine or, if on the hind leg, it burrows in an anterior direction, and then down to its ultimate position along the flanks. This migration requires approximately six months, at the end of which time the larva is full grown and moults to the second instar.

Development of Second Instar Larvae

Shortly after the first instar larva pierces the skin using its well developed mouth-hooks, it moults to form the second instar, which inverts itself and applies its posterior end bearing the spiracles to the hole. To keep this hole in the host's skin from healing up, the larva gradually pushes its rear end through the hole until the spiracles are almost level with the outside.

The second instar larva is enclosed by host reaction in a "Warble Cell" (Ormerod 1900) or "Encystment Sac" (Bishopp et al 1926). Simmons (1939) explains that when migration ceases a cyst of dense connective tissue, infiltrated/

infiltrated with leucocytes, is formed about the grub. The larva feeds on the pus and bacteria that accumulate within the cyst (Simmons 1939 a), which is always open to the atmosphere.

Cameron (1937) states that this second instar stage lasts for about one month. Among the deer from Adverikie, second instar larvae appeared on the 9th December, 1955 and the first third instar larvae were found on the 23rd January, 1956. These final stage larvae were already yellow and ecdysis must have occurred several days previously. This would make the length of the second stage to be five or six weeks.

On two stags inspected on the 28th February, second instar grubs were still present. One stag had two second and twenty-one third stage larvae and the other had twelve second and approximately three hundred final instar grubs. Probably by the first week in March only final stage larvae would be present.

Development of Final Instar Larvae

Several days before ecdysis the final stage spiracles appear as pale yellow objects deep beneath the integument of the second stage larva. As the time for moulting approaches the spiracles become more distinct in outline and just prior to moulting the structural details are well defined. Examination of the exuvia left in the cyst after the first and second moults, reveals that the rupture of/

of the old skin does not always occur in the same place.

The integuments of the first and second and newly emerged third instar larvae are more or less transparent and rather soft, but there are definite changes in this final instar as it matures. Growth is very rapid; the integument becomes tougher as it changes colour and the spiracles darken. This darkening of the spiracles begins at the rims and gradually the whole spiracles become dark brown and then black.

The integument of the larva becomes yellow and leathery to the touch. The colour continues to darken through brown, until, when ready to leave the host, it is very dark brown or black. The toughening of the integument does not impede the movement of the larva in any way because it is very obvious that activity increases with maturity. Early final stage larvae were not inclined to move when removed from the warble, but the tendency to crawl was evident with the more mature grubs.

As already mentioned, the first record of final instar larvae was 23rd January. A mature grub had already emerged from a cyst on the back of a shot stag on the 29th February. Cameron (1937) recorded that mature grubs begin to abandon the host at the end of March and by the middle of April the host is practically free of warbles. But since the pupal stage lasts approximately one month, depending on the temperature, and the adult flies, which live only for a few days, are active at the end of June, third instar larvae should still be present,in much reduced/

reduced numbers, of course, in May.

Table summarising occurrence of the three larval stages on host

Larval Stage	Host	Forest	First Record	Last Record
First	Hind	Glenfeshie		28.12.55
		Adverikie		5. 1.56
	Stag	Coull		15.12.55
Second	Hind	Adverikie	9.12.55	
		(Gaich (Glenfeshie	12.12.55	
	Stag	Dalwhinnie	22.11.55	
Third	Hind	Adverikie	23. 1.56	

Larval Emergence

An opportunity did not present itself to observe a final instar grub leaving the host, but Cameron (1932) relates "as is the case with the two bovine species of Hypoderma, the full grown larva of H. diana voluntarily emerges from the warble, forcing its way through the aperture by actively constricting its body". Although data are not available for H. diana larvae, it may be noted that the emergence of warble larvae from cattle most commonly occurs between 5 a.m. and 10 a.m. on bright sunny mornings (Scharff 1950).

Pupating Larvae

The larvae used were obtained from two stags shot on the 29th February. One larva which emerged naturally from one of the stags pupated before the 2nd March. Seventy -nine of the most mature larvae were grouped according to their/

their colour and maturity and placed on trays on dry peat moss.

Tray 1(a)	1 pupa. 4 black larvae. 15 dark brown-black larvae.
Tray 1(b)	20 dark yellow-brown larvae.
Tray 2(a)	20 yellow larvae.
Tray 2(b)	20 white-light yellow larvae.

The larvae in trays 1(a) and 1(b) were quite active and attempted to burrow into the substrate. When covered with peat moss they then proceeded to burrow out on to the surface. The larvae in trays 2(a) and 2(b) showed very little activity.

After being left in a constant temperature room of 65° F. and 35 per cent humidity, the condition of the larvae on the respective trays was noted three days later, on the 5th March.

Tray 1(a). Apart from the one pupa originally present, fifteen larvae had pupated. Of these, eight were black in colour but seven were dark brown. Two of these brown pupae were rather soft.

Tray 1(b). Eleven larvae exposed and desiccated. Of the remaining nine still buried in the moss, only three were still alive of which one was black.

Tray 2(a). Sixteen larvae exposed and desiccated. Two had turned black and five dark brown. Only two of the/

the remaining four were still alive.

Tray 2(b). Twelve exposed and desiccated. Two of the remaining eight alive.

A dry substrate was chosen for these pupating larvae since Soni (1949) found that the eventual emergence of adults was greatest from pupae reared in an environment with a humidity of 5 per cent to nil. But it would appear from the number of larvae which had shown signs of having lost moisture, that a very dry habitat is not conducive to the continuous development of the larvae after leaving the host.

The moss was moistened slightly and the larvae and pupae left as in the original tray 1(a). The remaining seven live larvae were transferred to tray 1(c).

6th March - Two larvae in 1(c) had come to the surface but were replaced in a hole in the moss. One pupa removed from 1(a).

7th March - Larvae in 1(a) showed signs of pupation. In 1(c) one larva was exposed and turning brown.

9th March - No larvae exposed.

19th March - Peat moss dry. Some larvae in 1(c) examined and found to be infected with a fungus. All larvae in 1(a) had pupated - probably about 8th or 9th March.

On 6th March one pupa and four larvae, ranging in colour from yellow to dark brown, were placed in a petri dish in the laboratory.

8th March - The most mature larva turned black and pupated.

12th March - The remaining larvae darker in colour.

23rd March - Larvae all black but flaccid.

From these results, only larvae which are dark brown or black in colour when removed from the host are likely to pupate.

Cameron (1937) describes how he observed mature larvae emerging from the host and crawling along the ground to the base of a tuft of grass or among mossy vegetation, to find a sheltered place to pupate. No data are available on how long the larvae take to pupate naturally. While a larva which emerged of its own accord was in pupal form two days later, some of the others took eight or nine days to pupate.

A black larva of H. lineatum placed on wet peat moss on the 24th April pupated six days later on the 30th April. Pfadt (1947) watched twenty-five larvae of H. lineatum which had dropped to the ground and records the time for transformation as varying from six hours to seven days, with an average of 2·3 days. According to Scharff (1950), pupation may take place in twelve hours if temperatures are favourable, or may be delayed for several days. Gebauer (1939) reports larvae of H. bovis boring small grooves in the soil under grass and pupating about twelve hours after leaving the host.

Formation/

Formation of Pupa

The process of puparium formation is twofold. First of all there is the darkening of the cuticle and, secondly, there is a change in its structure leading to a toughening and hardening of the integument. These reactions are independent.

A number of third instar larvae changed from yellow to black in colour after being preserved in alcohol but the cuticle of these larvae remained soft and did not acquire the firmer texture of the larvae which matured naturally.

The physical change that normally occurs with the darkening process is described by Fraenkel and Rudell (1940). "It is apparent that chitin is more dispersed in larval cuticle than in the puparium. Whereas the chrystallites can be rotated in any direction in larval cuticle, this freedom of rotation is lost in the puparium where the chitin appears to be aggregated into a more rigid framework."

Duration of Pupal Stage

Apart from the pupa kept in the petri dish, the other pupae were placed in moist peat moss, which became progressively drier. From the twenty pupea only four flies emerged.

Pupa formed	Fly emerged	♂ or ♀	Conditions
2nd March	23rd March	♂)	Constant temperature
5th March	24th March	♀)	65° F. 35 per cent
5th March	25th March	♂)	humidity.
6th March	6th April	♀	Laboratory.

The/

The duration of the pupal period is dependent upon temperature. Ono (1938) showed that the pupal stage of H. lineatum under dry conditions lasted twenty-two days at 53·6° F. and ten days at 85° F. Under moist conditions it was thirty-one to forty days at 64° F. Gebauer (1940) found the pupal period of H. bovis to be twenty-four to seventy days, and of H. lineatum to be fourteen days less. Pfadt (1947) records that the pupal period of larvae placed in the field averaged about forty-six days.

The pupal stage of H. diana is then probably considerably longer than the nineteen to thirty-one days found under experimental conditions, and will correspond with the four to six weeks given by Cameron (1937).

Emergence of the Adults

The fly emerges by pushing open the operculum at the dorsal fore-end of the puparium. No observations have been made on the time of emergence of H. diana adults, but Carpenter et al (1914) report that in nature the cattle warble flies generally emerge early on bright sunny mornings. They add that they are sexually mature and may mate within one hour after emergence.

Behaviour of the Adult Flies

Shortly after emerging on 23rd March, the male fly was very active, walking about continuously but flying little. It climbed the gauze at the side of the cage freely but was unable to scale the glass. Each time it tried/

tried to do so it fell on to its back and lay motionless except for sporadic attempts to assume its proper position. No definite response was shown to lamp light.

The female which hatched out on 24th March showed similar behaviour. The male, however, was more active than the female. Whenever they came close together activity greatly increased but mating was never seen to take place. The female would sit motionless for periods of five to ten minutes. Just before moving off she would lower the tip of her abdomen, rub her head with her forelegs then raise and lower her abdomen, extend the ovipositor fully and exude a drop of whitish fluid from the end of it.

On 25th March, when the second male emerged, the first male was already too weak to stand and the female was inactive. The first male died on 27th March.

The remaining flies kept until now in a constant temperature room of 65° F. and 35 per cent humidity, were relatively inactive. When transferred to a room of 77° F. and 70 per cent humidity their activity was much increased, but flight was still rarely attempted. Mating was still not seen to take place.

The female died on the 30th March and the second male on the 31st March. The female which emerged on the 6th April lived only two days.

These observations suggest a definite increase in activity with rise in temperature. Bishopp et al (1926) say that "it seems certain that when the weather is warm and/

and the adults are active their lives are uniformly very short". They record that H. lineatum adults in captivity live from one to twenty-five days. Flies lived from eighteen to twenty-three days in a temperature of 50° F. but only one to three days when in the sun with an average temperature of 80° F.

The mouth parts of the warble flies are degenerate and it is unlikely that they feed during their life time. Almost mature final instar larvae contain a large amount of fat body and Bishopp et al (1926) mention how this supply of food is carried over to the adult stage and appears to be sufficient to meet the needs of the adult insect throughout its life. The rate at which this limited supply of food is consumed must be proportional to the activity of the fly. Active flies will understandably live shorter than inactive adults.

It has been known for a considerable time that cattle warble flies do not travel very far, having little tendency to migrate more than one mile. (Faber 1927, Davies and Jones 1932 and Bishopp et al 1926). This is similarly true of H. tarandi and offers a method of control from this pest by moving the reindeer a considerable distance from where the larvae were dropped (Hadwen 1926). Since deer which are far up on the hill during the time of fly activity are usually free of larvae, there is reason to believe that the flight range of H. diana is also very short.

Factors/

Factors Governing Development

The larval stage occupies by far the greater part of the life history and is spent in the comparatively constant environment of the body of the host. But even here the larvae may be subject to conditions which affect to a varying degree the course of their development. These factors are twofold, (a) the sex of the host, (b) the resistance of the host.

The larvae which have emerged from the host, the pupae and the adults, find themselves under different and more changeable conditions. They are without the protection given to the earlier stages and are open to the attack of predators and parasites. They are most definitely influenced by both micro-climatic and general atmospheric conditions.

(a) Sex of the Host

It had been mentioned to me by several stalkers that the larvae developed more quickly in the stags than in the hinds. Results are very few but Table p.27 shows the last record of first instar larvae to be 15th December on a Coull stag, compared with 28th December on a Glenfeshie hind and 5th January on an Adverikie hind. Also the second instar appeared on the 22nd November on a Dalwhinnie (Adverikie) stag and 9th December on a hind from the same forest.

The reason for this advance in development is unknown. Both hinds and stags are most probably infested at/

at the same time. The earlier maturation in the stags may be due to their general loss of condition after the rutting season, which could be conducive to more rapid development.

Breev and Karageena (1953) state that the rate of development of H. tarandi was not affected by the sex of the host.

Host Resistance

The infestation and development of the deer warble larvae cannot be studied from year to year on the same animal as is the case with cattle. The general impression has to be obtained from a comparison of the numbers of larvae, free and encysted, on the carcases of deer of different ages. If the initial number of larvae attacking each host could be ascertained, then a more satisfactory picture of host reaction could be drawn. But it is evident that there is a definite resistance formed by the host to the developing larvae. This resistance appears to be dependent upon the age of the animal and its general condition.

Resistance is found in two forms in deer. First, there is a natural resistance and secondly, an acquired immunity. Scharff (1950) divided natural resistance in cattle into those individuals which are always practically grub free and those which show "delayed resistance". "Delayed Resistance", which describes the reaction to larvae which migrate so far through the body but die and become/

become encysted after reaching the back, definitely occurs in deer. This is seen in diagram 10B of a hind less than two years old. Dead larvae were present in a five inch band along each side. There were ten larvae present but, apart from one, they were deep between the muscle sheaths and possibly escaping host reaction in this position.

Whether there are deer which are free of larvae all their life because of their resistance, is not known.

Acquired immunity is built up over a number of years. Larvae of a first infestation normally all develop, but each successive year of infestation reveals this accumulation of the "resistant factor" in the encystment of larvae of different stages in the back. The proportion of larvae which develop successfully decreases and the number encysted increases as the animal becomes older. It is rare for larvae to develop fully in healthy deer older than five years.

Resistance, however, is very much dependent on the condition of the host. This is clear in the difference of infestation between milk hinds and yeld hinds. Milk hinds which have been suckling a calf and are again pregnant, normally lose much of their resistance, which gives the current season's larvae the freedom to develop. The policy employed in hind stalking in two different forests reveals this fact clearly. In Adverikie the poorer/

poorer deer are being stalked in an attempt to improve the quality of deer in that forest. In Glenfeshie, yeld hinds of prime quality are taken in preference to others. The difference in infestation is shown in the following table :-

Source	Total No.	With Larvae	With Cysts	No larvae or Cysts
Adverikie	30	18 (60%)	9 (30%)	3 (10%)
Glenfeshie	46	5 (11%)	14 (31%)	27 (58%)

Another type of host reaction was seen in the reindeer at Rothiemurchus. Instead of the larvae being encysted they were discarded before they were mature. In the bulls examined many holes were found where larvae had been. In one instance two second instar larvae were found half emerged from the holes. This premature emergence was not encountered in the Red Deer, and may only be the reaction to the parasite of non-specific hosts.

Predators

It is the opinion of several writers that birds destroy a considerable number of cattle warble larvae and pupae. Scharff (1950) and Bishopp et al (1926) report watching robins devouring them. Walton (1924) has observed jackdaws extracting them from the back, as is the habit of the starlings (Goodrich 1940). These birds probably feed on the deer warble larvae as well, but/

but Darling (1937) is of the opinion that the most important winged predator of <u>H. diana</u> grubs is the hoodie -crow. He mentions how he has watched the hoodies fly from birch woods to deer at the time the larvae are leaving the host and "gobble" something. He believes the juicy, newly emerged warbles may be the attraction. Moles and small rodents feed upon mature cattle larvae and pupae (Lucet 1914 and Stegmann 1920) and it is likely that they also play a part in the predation of deer warbles.

<u>Parasites</u>

When the sixteen <u>H. diana</u> pupae which did not produce any adults were dissected open, many were found to be infected with a fungus (Fig.11A). The mycelium coated the inside of the puparium. It was classified as <u>Penicillium glaberum</u> (Wehmer) Westling. Larvae which had failed to pupate were also infected with the same fungus. In this case the larval integument was stretched and grey in colour. At first no hypha was visible but later patches of mycelium formed on each segment. As this spread the larva began to shrink, and approximately two weeks after the fungus was first seen to be present, the outer integument coated with mycelium was all that remained.

Several <u>H. lineatum</u> larvae, placed in moist peat moss, were also infected with the same fungus. In this case/

PLATE 11

A - The fungal attack on the pupae.

B - Reaction of 5 per cent ammoniacal silver nitrate. Left larva untreated. Larva on right was left in solution for about half an hour and developed this characteristic pattern. A similar pattern was visible in untreated more mature 3rd instar larvae.

PLATE 11.

A.

B.

case the hyphae were seen to protrude from the segments after two days, and then the patches of mycelium appeared.

It is quite probable that the larvae and pupae may be parastised by this, or a similar fungus, naturally. It is Lucet's (1914) opinion that certain fungi destroy the pupea of H. lineatum in France.

Effects of Dry and Wet Substrates on Larvae and Pupae

Several second and third instar larvae at different stages of development were immersed in water to find if this had any noticeable effect on them. Second instar and early third instar larvae swelled considerably, but the rest showed no signs of water diffusing into them. Only the most mature larvae attempted to keep their spiracles above the level of the water.

Wigglesworth's (1947) tests were applied to the larvae to see if a wax layer was present.

(1) Second instar larval cuticle turned purple after thirty minutes in 5 per cent ammoniacal silver nitrate (A.S.N.).

(2) White third instar larvae, after thirty minutes in A.S.N., gave a distinct pattern of reaction, as shown in Fig.11B. This pattern is not noticeable on the larvae prior to treatment with A.S.N.

(3) Yellow larvae and later stages gave no reaction with A.S.N. A brown pattern corresponding to that obtained in (2) after treatment could be distinguished in/

an almost mature H. lineatum larva placed in shallow water in a petri dish pupated, but no adult was obtained from it. Soni (1949) found that cattle grubs placed on soil saturated with moisture died without pupating.

The effect of soil moisture has been studied in H. bovis and H. lineatum by several workers. Bruce (1938) states that H. bovis will not mature in soils with moisture content of more than twenty two per cent, and that H. lineatum will not mature if moisture content exceeds ten per cent. Soni (1949) obtained a low adult emergence from pupae kept at ten per cent soil moisture, but a maximum emergence from pupae in containers with soil moisture varying from one to five per cent. He adds that it is the moisture content of the soil that accounts for the concentration of H. lineatum in certain localities and its absence from others. This conclusion is borne out by Scharff (1950) in observations made in the Bozeman area of Montana, where the soils are very wet during the pupal stage of H. lineatum; a probable reason for its absence in the district. A correlation has been found between the distribution of H. crossi infesting goats and H. lineatum infesting cattle in the Kashmir and Kulu districts of India, based on the moisture contents of the soils (Agric. and Anim. Husb. in India 1928-39 Delhi 1941).

Breevui (1946) reports that H. tarandi pupae did not survive repeated prolonged flooding, and their development was retarded by ten days or more by a single prolonged/

prolonged immersion.

It would be reasonable to expect that soil moisture content adversely affects the development of H. diana pupae also. The maximum percentage soil moisture that they will withstand is not known, but it may be assumed that only well drained parts of the deer forests will prove suitable for pupal development. Adult emergence should then be expected mostly from pupae on the hill slopes, since bogs are common on the plateaux and in the valleys.

General Effects of Climate

MacDougall (1925) says that warble flies were less injurious to cattle in 1924 than in the previous year owing to weather conditions being unfavourable to them. Walton (1928) gives tables to show the correlation between the abundance of larvae of H. lineatum and H. bovis and the rainfall of the months April to July of the preceding year. Low infestations in 1921 and 1925 followed heavy rainfall. According to Natvig (1937), flies and grubs are fewer in Norway following cold wet summers, and in the most northern parts of the country they seem to have been totally exterminated in unfavourable years.

The opposite findings prevail in the case of H. diana attack in deer. While no actual figures are available, it is commonly reported amongst the stalkers and venison dealers that infestation is always very heavy in/

in wet seasons but comparatively light following dry mild Springs. My investigations were done following a very warm Spring and Summer and the degree of infestation was very light compared with the previous year, when, after a wet Spring, it was not uncommon to find a deer infested with a hundred or more warble larvae.

Bishopp et al (1926) carefully studied the effects of climate on cattle warble flies and put forth the following facts :-

(1) Wet conditions are unfavourable to the successful development of the pupae. Heavy rains coming at the time the adults emerge may destroy many of them and continued wet weather must limit severely the oviposition of the adult due to its very short life span.

(2) Winds have a double influence. The first is favourable in causing a rapid drying of the surface soil and benefitting the pupae; the second is adverse to oviposition, since the flies will not oviposit freely in a strong wind.

(3) High temperatures are most suitable for pupal development and fly activity. Flies normally oviposit on warm days but they will attack in remarkably low temperatures provided the sun is shining. Oviposition rarely takes place when the sky is heavily clouded. Pfadt (1947) showed that mature/

mature larvae and pupae can withstand sub-zero temperatures without being adversely affected.

MacDougall (1935) summarises the reaction of the flies to different conditions briefly, by saying that the warble flies show most activity in warm sunny weather and do not attack in rain or high wind. They dislike shade and the flies will not follow cattle into, or over, water.

A warm dry Spring thus favours the heavy infestation of cattle. The reaction of the deer warble flies is most likely similar in the various weather conditions, and yet infestation is least after a mild Spring. The reason for this seems to lie in the movements of the deer. The fly's flight range is limited, but if the deer trek away into the forests from the place where the flies emerge, they are unlikely to be hampered by these parasites.

Deer Movement and its Relation to Fly Attack

The movement of deer in the months March to June seems to have a direct bearing on the resulting infestation of warble larvae. This season includes the time when the larvae emerge from the host and the period of fly activity.

In March and April the deer are usually found on the lower slopes of the hills. There is little or no fresh growth of herbage on the hill-tops and in March especially, snow is often present. If the weather does become/

become mild, the deer may extend their movements to a greater height, but at night they trek back into the valleys. Several stalkers have mentioned to me that the deer in the poorest condition at this time of year are those which are most heavily warbled, and they come down to a much lower level than healthy animals.

It has already been suggested that the deer warble larvae probably resemble the cattle warble larvae in the time of their emergence from the host. This would be early in the morning when the deer are still very low. The majority of the flies would then be found in the lower reaches of the deer forests.

Darling (1937) records wide daily movements of deer in May and June from 300 - 400 ft. up to 2,000 - 3,000 ft. and back again in the evening. If the weather is good they start trekking upwards very early in the morning. The warm, high sun in June, has the effect of driving the deer into whatever shaded parts they can find, and they stay there for a considerable part of the day.

The flies have a limited flight range. Though their activity is at its peak on bright sunny mornings, most of the deer will have climbed high in the hills and, possibly, out of range. The flies do not like the shade, and those deer lying out of the sun will avoid the females seeking hosts on which to oviposit.

A continuous cold wet Spring, however, restricts deer movement/

movement considerably. It is at this time that they are shedding their winter coats and will be more influenced by the cold and rain than at other seasons. The deer are apt, under such conditions, to restrict their wanderings to the lower slopes of the hills. Only a return to more seasonable weather will take them to the hill-tops again. While a few hours sunshine in the mornings will entice them to climb a little, a continuous mild spell is required to encourage them to go very high. This means that they will most likely be within reach of the flies, which are roused to activity by the morning sunshine.

Some Tabanids are rarely encountered over 1,500 ft. (Darling 1937), and it was suggested to me that the yearly variation in warble attack may be the result of the warble fly having a similar altitude limit, only hosts below this height each year being open to attack. While this may have some significance, it is felt that the behaviour and limited flight range of the adults, and the movements of the deer as influenced by weather conditions, are the main factors determining the resulting infestation from year to year. Cold, wet weather keeps the deer low and within reach of flies roused to activity by a brief spell of sunshine. Bright, warm weather takes the deer to the heights and into the shade, where they are unhampered by the parasites.

PLATE 12

A - Deer hide showing extent of warble damage.

B - Close-up of same hide.

PLATE 12.

A.

B.

CONTROL

Measures of control of the warble flies have been taken with various degrees of success in cattle and reindeer. These have taken the form of removal of the grubs from the back of the animals (MacDougall 1934), the treatment of the animals with insecticides (Scharff 1950) and the driving of the reindeer as far away as possible from the pastures where the larvae have pupated (Nakhlupin and Pavlovskie 1932).

Since no such remedial treatment can be given to undomesticated animals like the Red Deer, it is doubtful whether the warble fly can be controlled. It is of certain economic importance to a small branch of the community - a stag hide in this country is worth only 20 per cent of a similar hide in New Zealand, where the warble fly does not exist - and therefore control would be of considerable advantage to them.

In this respect, several points mentioned in this paper are of interest :-

1) The flight range of the adult is limited.
2) The deer in poorest condition in March and April carry most warble grubs and come further down the hills.
3) Healthy deer remain higher up and are relatively free from/

from attack.

(4) A warm dry Spring favours a low infestation.

Many people are under the impression that the Highlands are overpopulated with deer and that some way of controlling their numbers is long overdue. If this control was to take the form of stalking the deer which are in poor condition and low down in the hills and valleys between May and June, a certain measure of control would also be given to the warble fly, especially if this was practised during a warm dry Spring. While this could not be fully effective, it would be improved throughout the season, because any deer attacked would be of better quality and in better condition, and a healthy host always offers resistance to the developing larvae.

Several successive years of weather conditions adverse to the warble fly attack can themselves offer reasonable control. Natvig (1937) reports that cattle warble flies and grubs seen to have been totally exterminated in unfavourable years in the more Northern parts of Norway.

DISCUSSION

Deer stalking has an unofficial closed season from the end of January until September, when the stag shooting begins. There is then a gap in the shooting from the middle of October until the middle of November, when the rut is in progress. During this second part of the season the stags are in poorer condition and the hinds are then stalked. The closing date is often determined by the stalkers themselves, who find the carcases revolting to handle because of the presence of large numbers of warble larvae at the beginning of the year.

This shooting programme has severely limited the number of deer obtainable throughout the year. The lack of material at certain periods of this study has left gaps in the life history still requiring to be filled. The eggs have not yet been found, nor have the very early stages of the first instar larvae. While it is almost certain that these will be found in the lower parts of the legs, the early parts of the migratory route will not be followed unless there are some deer available before the normal shooting season starts. The antlers of the stags are still in velvet until the end of August and it is considered bad sportsmanship to stalk them before September. Hinds at this time are suckling their calves and must be left alone. So, while a few deer may be obtained/

obtained, it is unlikely that a sufficient number could be available for significant observations to be made.

The eggs of H. diana could probably be found on the Reindeer at Rothiemurchus, but, although several attempts were made in June to contact Mr. Utsi, the Technical Advisor of the Reindeer Council of the United Kingdom, without success, the opportunity did not present itself at the appropriate season.

Many workers have mentioned the stampede of cattle resulting from the continued approach of H. bovis to oviposit (Bishopp et al 1926 and MacDougall 1934). Due to the inclemency of the weather in June 1956, it was not possible to study the response of deer to H. diana adults, but stalkers I have met have never seen Red Deer stampeding. They have, however, noticed deer holding their heads low in the heather, as if reacting to some external stimulus. This is most probably a means of defence against the repeated attacks of Cephenomyia auribarbis, the deer nostril fly, as reported by Darling (1937). Not all warble flies do cause a reaction in the host, as is the case with H. lineatum, and the observations of these stalkers suggest that H. diana approaches the deer unobtrusively to lay its eggs.

It will be noted that many comparisons have been made in this papers with other Hypoderma spp. These species are all very similar morphologically and the biologies/

biologies of Hypoderma bovis, H. lineatum and H. tarandi resemble each other to a marked degree. The known facts of H. diana are generally in common with these other species, and in this respect the few deductions which have been applied to this species from the more voluminous study of the other species seem justified.

The work that has been done, has been sufficient to show that the behaviour of the Red Deer Warble Fly, H. diana is different when it attacks Reindeer than in its normal species. Whereas in its native host, the first instar larvae migrate subdermally and intermuscularly only, H. diana larvae in Reindeer invade the spinal canal for a considerable period before appearing in the back. Also, the resistance to the parasite differs in each host. In the Red Deer the larvae becomes encysted, but in the Reindeer the grubs emerge from the host prematurely. While the presence of H. diana larvae in the Red Deer may lower their general condition, it may prove fatal in the Reindeer.

SUMMARY

(1) The oesophagi of five deer were stripped and inspected but no larvae were present.

(2) Forty-seven carcases were split and the spinal canals examined, but there was no sign of larvae, nor any trace left by the larvae in the epidural space.

(3) Migrating first instar larvae were encountered only under the skin and between the muscles, and by plotting their respective positions on various carcases throughout the season, their route could be established. It was suggested that the eggs were laid on the lower parts of the legs, the forelegs being chosen in preference to the hind legs. After the larvae hatch out and burrow into the skin, they remain in these lower parts for some time before moving towards the back. From the forelegs, their route takes them up over the shoulders towards the spine and then in a posterior direction to assume their position along the flanks. Those on the back legs move over the thighs and then anteriorly to the flanks. This migration requires almost six months, at the end of which time the larva moults to the second instar.

(4) The second and third instars are non-migratory and develop in the one position under the skin. The second/

second instar larvae moults after about six weeks, the third instar larvae being found from the end of January onwards.

(5) The third instar larvae may mature in one month. As they develop they change in colour from white, through yellow and brown to black, and the integument becomes leathery and waterproof to withstand the variable conditions without the host.

(6) The larvae emerge and pupate on the ground in two to nine days.

(7) Flies emerged in the laboratory in three to four weeks. They are actively ovipositing during May and June.

(8) The behaviour of adults in captivity is described. They do not feed and their life span is restricted.

(9) The sex of the host and its acquired resistance governed by its general condition, affect the development of the larvae. The grubs may be parasitised by a fungus. Climatic conditions and their effect on the various stages are discussed.

(10) Deer movement is shown to have an effect on the resulting infestation of warble larvae.

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