EPIDEMIOLOGY
OF ONCHOCERCIASIS

Report of a WHO Expert Committee
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Geneva, 10–18 November 1975

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EPIDEMIOLOGY OF ONCHOCERCIASIS

Report of a WHO Expert Committee

A WHO Expert Committee on Epidemiology of Onchocerciasis met in Geneva from 10 to 18 November 1975. The meeting was opened by Dr T. Lepes, Director, Division of Malaria and Other Parasitic Diseases, who welcomed the participants on behalf of the Director-General and outlined the reasons why WHO had decided to convene an expert committee on the epidemiology of onchocerciasis.

1. INTRODUCTION

The 10 years that have passed since the second meeting of the WHO Expert Committee on Onchocerciasis* have seen considerable advances in knowledge of this disease and of its epidemiology. Furthermore, there is now a worldwide awareness that onchocerciasis—“river blindness”—is not merely an obscure disease found in a few remote parts of the tropics but a widespread plague that produces grave socioeconomic effects and much human suffering over large parts of tropical Africa as well as in Latin America.

As evidence of this, the past decade has seen the first serious involvement of the international community in an attempt to control onchocerciasis in one of its worst endemic areas. The Onchocerciasis Control Programme in the Volta River Basin Area (OCP), involving 7 West African countries, was launched in 1974. At the request of the governments of the 7 countries concerned, the programme is being executed by WHO, in association with the United Nations Development Programme (UNDP), the Food and Agriculture Organization of the United Nations (FAO), and the International Bank for Reconstruction and Development (IBRD); the Bank administers a special fund for onchocerciasis, the Onchocerciasis Fund, which is financed by a number of donor countries and organizations.

Control of onchocerciasis by killing the parasitic worm *Onchocerca volvulus*, the cause of the disease in man, is not feasible with the drugs at

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present available. Although these drugs are suitable for the treatment of individual patients, their use requires careful medical supervision, which cannot be provided on the large scale that would be required if chemotherapy were to be used to control transmission.

Control of the disease is therefore based at present almost exclusively on control of the vector *Simulium* flies, which can be attacked by the application of insecticides to their localized breeding sites in fast-flowing rivers and streams. Unfortunately this form of control has to be continued for some 20 years before the disease can be expected to die out in the affected communities.

In order to plan such control operations and to assess their effects, both on the transmission of the parasite and on its pathological manifestations in man, it is necessary to have a wide knowledge of the epidemiology of the disease. Such studies embrace a multitude of factors that affect the parasite itself, its definitive host (man), and its intermediate host (the *Simulium* fly). They involve parasitology, entomology, economics, mathematics and many other disciplines, all of which must be considered by the epidemiologist when he tries to assess the distribution and the importance of the disease and to monitor its control. It is for these reasons that epidemiology is of paramount importance and that this report has so wide a scope and includes the views of experts in such a variety of fields.

2. GEOGRAPHICAL DISTRIBUTION OF ONCHOCERCIASIS

Endemic foci of human onchocerciasis caused by *Onchocerca volvulus* exist in tropical Africa, Yemen, Mexico, and Central and South America (Fig. 1 and 2). In Africa the northern boundary of endemic onchocerciasis runs from Senegal to about latitude 15° N in Mali, Niger and Chad and on to Sudan, where it reaches its northernmost point in the Abu Hamed area at 19° 15' N, and then turns south to Ethiopia. Further to the south, foci of the disease exist (moving from west to east) in Gambia, Guinea-Bissau, Guinea, Sierra Leone, Liberia, Mali, Ivory Coast, Upper Volta, Ghana, Togo, Benin, Nigeria, United Republic of Cameroon, Equatorial Guinea, Central Africa Republic, Congo, Zaire, Rwanda, Burundi, and Uganda. The southern limit starts at about latitude 14° S in Angola and runs north-east around the Zambian border to a point as far north as latitude 8° S in Zaire. It then turns again to the south where it reaches its southernmost point in the Cholo district of
Malawi at latitude 17° S and, finally, extends into the United Republic of Tanzania at latitude 12° S. There are no recognized foci of onchocerciasis in Southern Rhodesia or Mozambique. The disease has been eliminated from Kenya, with the exception of a small focus at the Ugandan border.

One of the largest endemic areas occurs in the Volta River basin area, which incorporates parts of Benin, Ghana, Ivory Coast, Mali, Niger and Togo, and all of Upper Volta. This is the area of the Onchocerciasis Control Programme (OCP), of which WHO is executing agency.

In rain-forest areas of equatorial Africa foci of onchocerciasis coexist with endemic infections of another skin-dwelling filarial parasite, *Dipetalonema streptocerca*.

Onchocerciasis is endemic in the southern part of Yemen, in and around Taiz. This focus may be much larger than is at present known and it may extend north into Saudi Arabia. Onchocerciasis has been diagnosed in patients in Democratic Yemen. So far no endemic foci are recognized in this country, and the cases observed are considered to have been imported from neighbouring Yemen.

In Latin America endemic onchocerciasis occurs in Mexico, Guatemala, Colombia, Venezuela and Brazil.

In Mexico there are 3 foci, the first extending from latitude 17° 25’ N to latitude 17° 48’ N and from longitude 96° 12’ W to longitude 96° 40’ W, covering about 1400 km² in the state of Oaxaca. The other 2 foci are located in the state of Chiapas. The northern one extends from latitude 16° 52’ N to latitude 17° 7’ N and from longitude 92° 28’ W to 92° 40’ W, an area of about 700 km². The southern focus lies between latitudes 15° 4’ N and 15° 57’ N and between longitudes 92° 5’ W and 93° 7’ W, embracing an area of about 6800 km² containing many coffee plantations.

The southern focus in Mexico continues eastward to the endemic area of Huehuetenango in Guatemala. Of the 22 administrative departments of Guatemala, 7 have endemic foci. Onchocerciasis is most severe in the area bounded by the Los Esclavos River to the east and by the Nahualate River to the west.

In Colombia there is a relatively small endemic focus in the Micay River area, near the Pacific coast. It is quite probable that onchocerciasis also occurs in other parts of the country where conditions are similar to those found near the Micay River.

In Venezuela 2 major foci are recognized. One lies in the eastern part of the country, in the states of Anzoátegui, Monagas and Sucre. The other, the central focus, includes the states of Aragua, Carabobo, Miranda and Guárico. Isolated cases have been found in other parts of the country.
The map, prepared from information available to WHO at the end of 1975, gives an approximate picture of the main endemic areas as known from publications and reports. White areas
do not necessarily indicate absence of diusasa; they may reflect lack of epidemiological information. Because of the small scale, individual foci within endemic areas are not shown.
FIG. 2. GEOGRAPHICAL DISTRIBUTION OF ONCHOCERCIASIS IN THE WORLD:
MEXICO AND CENTRAL AND SOUTH AMERICA

This map, prepared from information available to WHO at the end of 1975, gives an approximate picture of the main endemic areas as known from publications and reports. White areas do not necessarily indicate absence of disease; they may reflect lack of epidemiological information. Because of the small scale, individual foci within endemic areas are not shown.
In Brazil there is a confirmed focus of onchocerciasis in the state of Amazonas, towards the Venezuelan border. In many of the endemic areas in Central and South America there is likely to be confusion with *Mansonella ozzardi*. The microfilariae of this parasite are concentrated in the upper dermis and they emerge in large numbers from bloodless skin snips (see section 10.2).

The WHO Expert Committee on Onchocerciasis estimated the total number of people infected with *Onchocerca volvulus* at 20 million.* This estimate has not changed since 1947,** despite considerable growth of the populations in the affected areas, the availability of better field methods for the diagnosis of onchocerciasis, and the recognition of many new foci. The difficulty in obtaining reliable figures on onchocerciasis prevalence on a global scale simply reflects the paucity, and also often the poor quality, of available field data. In OCP's recent experience in West Africa, a systematic search for data obtained in previous prevalence surveys of the area, combined with the use of a more sensitive diagnostic test, has doubled the originally estimated number of onchocerciasis cases from 500,000 to 1 million. It is reasonable to postulate that the figure of 20 million for the total number of people in the world infected with *O. volvulus* is a gross underestimate.

3. THE ONCHOCERCIASIS CONTROL PROGRAMME IN THE VOLTA RIVER BASIN AREA, WEST AFRICA

The savanna area of the Volta River basin in West Africa is one of the worst endemic onchocerciasis zones in the world. As already mentioned, the area comprises parts of Benin, Ghana, Ivory Coast, Mali, Niger and Togo, and the whole of Upper Volta. It covers approximately 700,000 km², with about 10 million inhabitants. It is estimated that of these at least 70,000 are blind, mainly from onchocerciasis, while many more have serious visual impairment. The governments of the seven West African countries concerned have recognized that onchocerciasis is the most important single deterrent to large-scale development of the potentially fertile river valleys in the area, which now lie uninhabited and unproductive. Furthermore, the serious effects of drought in the Sahel for 6 successive years have gravely disturbed the delicate socioeconomic balance in the Volta River basin area.

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After a joint meeting on onchocerciasis control, held in Tunis in 1968, had concluded that onchocerciasis control was feasible and would be most effective and economical when applied to a sufficiently large area, the governments of the 7 countries in the Volta River basin area requested WHO and UNDP to study the situation, with the aim of developing a plan of operation to achieve the control of onchocerciasis in the area. After 5 years of preparation the Onchocerciasis Control Programme started at the beginning of the dry season in 1974.

The Programme area lies between latitudes 8° and 14° N. Its northern part is characterized by dry Sudan-type savanna, while the southern region is covered by more wooded Guinea-type savanna with forest galleries along streams and rivers. In the extreme south of the Programme area there are scattered pockets of dense forest constituting a transition zone to the rain-forest. In the whole OCP area there is a great variety of ethnic groups and subgroups with different traditions and languages. Population density is uneven. It is characterized by the concentration of people in the uplands and a relative desertion of the riverine areas.

Onchocerciasis can be controlled by methods directed against the parasite, the vector, or both. Of these, denothing campaigns conducted in tropical America have yielded equivocal results and did not appear sufficiently practical to be recommended for the Volta River basin area. Although two drugs, suramin and diethylcarbamazine, have been used successfully for treating individual patients under medical supervision, and have also been used in mass control campaigns in Venezuela, they are not safe for mass application under the conditions prevailing in West Africa, where many patients with heavy infections develop severe side reactions. Therefore, vector control by systematic destruction of the blackfly (*Simulium damnosum* s.l.) larvae in their circumscribed breeding sites is at present the only practical means of attack to control onchocerciasis in the OCP area.

The Programme's strategy and plan of operation in West Africa have benefited from experience gained in other parts of the continent. This experience includes the *Simulium damnosum* control projects in the Abuja and Kainji regions of Nigeria and in the Ivory Coast, Mali, Upper

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*b Conly, J. Onchocerciasis in Venezuela. In: Research and control of oncho-


*c Simulium damnosum sensu lato is in fact a complex of species; see section 6.*
Volta and Zaire; and the vector control campaigns against *Simulium neavei* in Nyanza Province, Kenya. In the Kenyan programme, follow-up examinations for the continued presence of infection in the local population of the protected area were carried out for a period of 18 years after the interruption of transmission. These studies showed that in an endemic area the parasite may live as long as 15 years, and that the disease in those previously infected will continue to take its course. Therefore, if mass drug administration cannot be used as an additional means of onchocerciasis control, the programme against *S. damnosum* s.l. may need to be continued for about 20 years in order to achieve satisfactory control of the disease in the protected areas.

Because many of the breeding sites of *S. damnosum* s.l. are inaccessible by land the only feasible method of insecticide application is by aircraft. For large, open rivers light fixed-wing planes can be used, but for narrow, twisting waterways and for those overhung by forest, helicopters are needed. After years of research the insecticide finally selected for OCP is a biodegradable insecticide, Abate,* which in suitable formulations combines high effectiveness against the blackfly larvae with very low toxicity for man, non-target fauna and plants.

Monitoring of the effects of insecticide application to the large target area is shared with a specially created, independent Ecological Panel, which advises the programme director and the governments concerned on appropriate measures to ensure the satisfactory protection of the environment.

The control operations are being implemented progressively in 3 stages and, when the complete area is covered, approximately 14,000 km of river will be under treatment. Helicopters and fixed-wing aircraft are being used to apply the larvicide to the rivers weekly in amounts calculated to give an effective concentration of 0.05 mg/l for 10 minutes in the rainy season, and 0.10 mg/l for 10 minutes in the dry season. The insecticide is deposited in a single mass by means of a rapid-release system specially designed for the Programme. Drop points in the large rivers are approximately 30 km apart during the rainy season, when the riverine discharge is sufficient to transport the larvicide downstream. In the dry season applications are made just upstream from each breeding site.

For various purposes, the area is divided into 7 sectors, each headed by an entomologist who has 3 or 4 subsectors under his care. The technician responsible for each subsector supervises mobile teams that go in circuit to a fixed number of catching points, generally located near the

rivers. The *S. damnosum* s.l. adults caught over an 11-hour period during the day are counted and dissected to determine parous rates and infection rates. The results are recorded on prepared forms for short-term evaluation and for long-term computer storage and analysis. The teams also investigate those breeding sites in the area that are accessible to surface transport. Other sites are surveyed by entomologists and operational staff travelling by helicopter. All information on the effectiveness of the control measures and on river conditions is radioed to Vector Control Unit headquarters to permit the planning of subsequent aerial operations.

The success of the control programme is monitored and evaluated by special entomological and epidemiological teams. The entomological evaluation is based on a network of permanent simulid capture stations. Supplementary sampling at additional locations is carried out by mobile entomological teams.

The epidemiological evaluation is made by 2 teams, each with its own epidemiologist, ophthalmologist, and sociologist, which operate in about 150 selected villages throughout the area. There are 2 types of investigations. The first is designed to measure changes in the incidence, prevalence and intensity of infection, as well as in visual acuity, in all the selected villages. These villages will be re-examined at 3-year intervals. The second type of investigation will involve more detailed clinical and laboratory studies of a subsample. The intensive epidemiological follow-up studies are carried out in close cooperation with the entomological teams in order to obtain data on the dynamics of transmission. The information from these combined studies will also be used to test and improve mathematical models of onchocerciasis dynamics, such as those being developed by WHO.

The Programme is now (as at December 1975) in full operation. No adult *S. damnosum* s.l. have been found over several months in most of the northern and central parts of the area covered by the first operational phase. However, relatively large numbers of female flies, many of them infected, have been found in some peripheral parts of this area. It is assumed that these flies come from breeding sites outside the treated area and that most of them will be eliminated as the Programme extends into phases II and III. Special consideration will have to be given to some of the southern river systems, which fall outside the boundary of the OCP area and from which *S. damnosum* may reinvade the protected zones.

The work of the epidemiological evaluation teams is progressing satisfactorily, with excellent cooperation from the local population.\(^a\) As

\(^a\) The epidemiological techniques used in the OCP area are described in Annex I.
the activities of the teams continue, the research programme on the
epidemiology of onchocerciasis will be fully developed in close coopera-
tion with scientists working in other parts of the world. OCP represents
the first longitudinal epidemiological investigation of onchocerciasis
carried out by a multidisciplinary team in a large area under control. The
results will be important for the planning and execution of programmes
for the control of the disease in other areas where onchocerciasis is a
serious public health problem. Provision has been made in the Pro-
gramme for training personnel from the 7 participating countries, and
workers from other countries who are involved in control schemes are
welcome to study the methods being used.

Epidemiological Factors
in Onchocerciasis

4. THE DISEASE

4.1 General features

Onchocercal infection in man produces nodules, a wide variety of skin
changes, lymphatic pathology, and some systemic effects. However the
most important lesions are those of the eye, which lead to serious loss of
vision and blindness. The nodules and lesions of the skin, lymphatics and
eye have been described and illustrated elsewhere.6 Other aspects of the
disease are discussed in section 5 of this report.

In general, similar clinical lesions are found in different parts of the
world. However, the occurrence of regional differences, especially in
the frequency and severity of particular lesions, is well documented.

4.2 Ocular lesions

There is now no doubt that the lesions of the eye listed below are
directly caused by infection with O. volvulus, and that their prevalence
and severity are closely related to the prevalence and intensity of infection,
and particularly to the density of microfilariae found in the head region
and in the eye.

The same ocular lesions are found in Africa and Latin America. They tend to be bilateral but not necessarily symmetrical. The types of lesion are as follows:

(a) "Fluffy" corneal opacities.
(b) Sclerosing keratitis.
(c) Anterior uveitis, and the secondary glaucoma and secondary cataract that may result.
(d) Choroidoretinitis.
(e) Optic neuritis and postneuritic optic atrophy.

4.2.1 Ocular pathology

In histological preparations from autopsy material, microfilariae have been found in all tissues of the eye and in association with all inflammatory lesions, but more material must be examined to determine in detail the relationship between microfilariae and the lesions. There is evidence that microfilariae enter the cornea from the surrounding skin and conjunctiva. There is also evidence suggesting that they penetrate the sclera to reach the choroid by passing alongside the lateral long posterior ciliary vessels and the short posterior ciliary vessels. They may also penetrate the sclera alongside the anterior ciliary vessels. These routes of entry offer a possible explanation for the peculiar distribution of choroidoretinal lesions in onchocerciasis.¹

The finding, in some cases of sowda,² of microfilariae only in the skin of the ankles and in the anterior chamber of the eye suggests that, at least in this form of the disease, microfilariae do not enter the anterior chamber via the skin. Possibly they travel via the bloodstream.

4.2.2 Microfilariae in the eye

Living microfilariae may be seen in the cornea, usually near the periphery, by careful examination with a slit-lamp at a magnification of ×25.³ They are best observed in the reflected beam by retroillumination. The living microfilariae are frequently coiled and often mobile, in contrast to dead microfilariae, which are usually straight.

³ The local name in Yemen for the most important clinical manifestation of onchocerciasis.
Dead microfilariae in the cornea and living microfilariae in the anterior chamber are easily seen at a magnification of ×16. The number of microfilariae visible in the anterior chamber increases if, before examination, the head is bent down for a minute and/or the eyeball is massaged digitally.

4.2.3 Natural history of ocular lesions

“Fluffy” opacities may appear around the dead bodies of individual microfilariae that have invaded the cornea. These reactions tend to be most marked in light infections (e.g., in immigrants, children in hyperendemic villages, and persons of any age in hypoendemic villages). On the other hand, many intensely infected patients, with large numbers of microfilariae in the cornea, show little or no fluffy reaction.

Permanent ocular lesions (due to sclerosing keratitis, iridocyclitis, chorioretinitis and optic neuritis), unlike fluffy corneal opacities, develop only in response to heavier and more prolonged invasion of the eye by microfilariae. This level of ocular infection can build up only over a number of years but is likely to be achieved at an earlier age in heavily infected than in lightly infected communities.

Lesions due to mild uveitis of the anterior segment, to chorioretinitis, or to optic neuritis, are usually the first to appear. At this stage anterior uveitis is not usually sufficient to impair vision seriously, although chorioretinitis and optic neuritis may cause blindness. As density of infection builds up with age, new lesions develop, frequently in previously unaffected tissues. The complications of anterior uveitis become an important cause of blindness, and in the most heavily infected patients sclerosing keratitis also causes much blindness, particularly in West African savanna villages.

Ocular lesions sufficient to cause blindness usually build up only over a long period. By this time several ocular tissues are almost always involved, but the more posterior lesions may be obscured from view by the more anterior. Although slow development is usual, vision is sometimes lost rapidly—even, occasionally, in childhood. Probably the most common cause of this rapid deterioration is the development of glaucoma secondary to anterior uveitis, but optic neuritis may also lead to rapid loss of vision.

4.2.4 Risk factors of blindness

With the aim of preventing blindness it is necessary to define, as far as possible, those factors that govern the risk of blindness developing in
a community, and those that govern the risk of an individual becoming blind.

(a) **Community risk factors.** The prevalence of onchocercal eye lesions and blindness is associated with the prevalence, and more closely with the intensity, of infection in a community. This in turn is related to the annual transmission potential (see section 6.5.2). However, in West Africa the risk of blindness is higher among communities living in savanna than in rain-forest (see section 7.2).

(b) **Individual risk factors.**

(i) Occupations that bring people (e.g., fishermen, ferrymen, workers in coffee plantations in Latin America, families with farms near breeding sites) into prolonged contact with simulids.

(ii) Age; progress of ocular lesions in hyperendemic areas shows a peak in the 30-39-year age group.

(iii) Males, probably because they are more heavily infected, are at greater risk than females.

(iv) Adult worms in the head region. The presence of head nodules (which are often very small, flat, and only recognized either by the patient himself or after detailed examination of a shaved head) is a risk indicator.

(v) The density of microfilariae in a skin snip taken from the outer canthus. It has been suggested that a density of 10 (or possibly 5) microfilariae per mg of skin at this site indicates serious risk to the eye.

(vi) The presence of more than about 50 microfilariae in the cornea, or of more than about 20 microfilariae in the anterior chamber, indicates a dangerous level of infection. The number of microfilariae in the anterior chamber should be assessed after the patient has sat for at least 1 minute with his head between his knees and with the top of his head as near as possible to the ground (i.e., with his head inverted).

(vii) The appearance of early symptoms and signs of permanent ocular damage, e.g., night blindness, contraction of visual fields (measured by confrontation or with a Goldman perimeter), mild uveitis, and oedema of the optic disc or retina (which may be best demonstrated by fluorescein angiography) all indicate an increased risk of blindness.

4.2.5 Measures to control blindness

The therapeutic drugs now available (suramin and diethylcarbamazine) should only be given under appropriate medical supervision, for they may induce serious side-effects and even death, especially in heavily infected patients. These are the very persons most at risk of becoming blind. Thus, with the treatment schedules at present in use, the risks involved in the treatment of all infected people as part of a programme to control blindness must be very carefully considered before embarking on therapy, especially in hyperendemic areas. However, individual therapy may well be beneficial for patients who can be selected as being at serious risk of becoming blind.

Wherever control measures are planned, consideration should be given to the treatment of patients in whom the development of serious eye lesions may be prevented. It is thought that the numbers requiring therapy would be insufficient to interfere seriously with epidemiological assessment. Furthermore, the arrangement of treatment by surveillance teams will encourage good relations with the population. If possible, treatment should not be limited to onchocercal conditions; other eye diseases, such as trachoma, trichiasis, xerophthalmia and senile cataract, should also be dealt with.

The individual risk factors mentioned above may not always identify patients in whom blindness can be prevented by treatment. Once head nodules have developed, ocular lesions have appeared, or symptoms such as night blindness are present, it may already be too late to prevent progress to blindness. Moreover, treatment with existing drugs may, in some patients, lead to a deterioration in eye lesions. Further controlled therapeutic trials are required to determine which signs and symptoms can be relied on to provide warning early enough for therapy to be truly effective.

4.3 Other aspects of the disease

Onchocerciasis produces serious manifestations apart from those in the eye. The systemic effects of the disease are not fully understood. Microfilariae of *O. volvulus* are found in the urine, blood and cerebrospinal fluid. Onchocerciasis has been shown to be associated with cachexia, where other causes have been excluded. There is possible association with dwarfism among the Nakalanga in the Mabira Forest area of Uganda. The Nakalanga have unusually heavy infections with *O. volu-

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and the dwarfism is sporadic and not familial. Another interesting feature of onchocerciasis, which has so far received little attention, is the relatively high frequency of epilepsy reported from hyperendemic areas of onchocerciasis in Central African Republic, Guatemala, Mexico, Sudan and the United Republic of Cameroon.

Lymphadenopathy, elephantiasis, hanging groin and the hernia sometimes associated with it may be incapacitating. The skin lesions often cause severe and persistent pruritus and loss of sleep. Presbydermia and the depigmentation that is particularly common in African rain-forest are serious social stigmata.

Two types of skin lesion reported from Latin America deserve special mention because they are not described in the study referred to at the beginning of section 4.1, which deals primarily with the African disease. *Mal morado* is a purplish skin discoloration, commonly seen on the ear lobules, cheeks, upper arms and shoulders. It is peculiar to Mexico and Central America. *Eritema de la costa* is a pale red oedema of the skin often associated with fever; it is most common on the face, but may also occur on the body.

4.4 Measurement of visual acuity

The Snellen “E” test, printed on cards for use at 6 metres, is recommended. The 6/60 card may be used at shorter distances (1-5 metres) for patients unable to read 6/60. This test is preferred to the Sjörgen hand test because it is more reliable.

For general evaluation, only two Snellen cards are necessary, the 6/18 and 6/60. The patient is tested with both eyes open, first at a distance of 6 metres.

- If the patient reads 6/18 he is coded 0.
- If the patient cannot read 6/18 but reads 6/60 he is coded 3.
- If the patient cannot read 6/60 but reads the 6/60 card at 3 metres, i.e. his visual acuity is 3/60, he is coded 2.
- If the patient cannot read the 6/60 card at 3 metres, i.e. his visual acuity is less than 3/60, he is coded 1.

Then: 0 = “No visual impairment”, i.e. 6/18 or better

1 = Blind, i.e. inability to read 3/60

2 = Severe visual impairment, i.e. less than 6/60 but better than 3/60

3 = Visual impairment, i.e. less than 6/18 but better than 6/60

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*a* Buck, A. A., ed., op. cit.

For detailed assessment, the complete set of Snellen "E" test cards is required, and visual acuity is recorded for each eye separately in the usual way.

Skill and aptitude are required in order to obtain accurate measurements of visual acuity. The test must be explained to patients in simple terms and must be performed with care and good humour. Attention must be paid to correct lighting and the opposite eye must be effectively occluded with a card.

4.5 Measurement of visual fields

A reliable measurement of visual fields would give useful additional information since many patients with severe fundal lesions and grossly contracted visual fields retain good central vision. Moreover, early lesions might be detected by a loss of field. The confrontation test may give useful information to a skilled worker but cannot give a detailed record. The Goldman perimeter is recommended for trial where accurate records are required.

4.6 Measurement of dark adaptation

Night blindness is a common symptom of onchocerciasis and there is a need for a simple method of measuring dark adaptation.

4.7 Research needs

(1) Further research into the epidemiology of ocular lesions. This should include long-term studies to determine the incidence of these lesions and their progress, in an attempt to assess factors that favour the appearance and development of eye lesions. It is hoped that such work may demonstrate the risk factors of blindness so that patients at risk may be identified at a stage when treatment may be effective.

(2) Research, perhaps using animals, to determine the routes by which microfilariae reach the various tissues of the eye.

(3) Collection of ocular material and its histological examination, possibly by a recognized central pathological reference laboratory interested in ophthalmology. Suitable material might be obtained from enucleated eyes, post-mortem eyes, and ocular tissues (such as iridectomy tissue and corneal discs removed by trephine) obtained at surgical operations.
(4) Assessment of ocular lesions by fluorescein angiography, which should lead to a better understanding of the posterior segment lesions.

(5) Design of a simple method of measuring dark adaptation in the field.

(6) Investigation of practical means of measuring visual field defects in illiterate populations examined in surveys.

(7) Research on new and more suitable drugs for the treatment of onchocerciasis. Entomological control programmes need to be supplemented by treatment of patients whose vision is at risk. Moreover, a safe and effective treatment that could be used on a mass scale would be a valuable addition to entomological control since it would interrupt the life cycle of the parasite.

(8) Determination of reliable signs and symptoms giving warning of impending eye lesions and blindness sufficiently early for therapy to be effective. Controlled therapeutic trials using the drugs at present available according to best dosage schedules (with steroid cover if necessary) should be undertaken to investigate this problem.

5. THE PARASITE

The genus Onchocerca contains several clearly defined species that occur in domestic and wild animals in many parts of the world, including temperate and tropical regions (see section 7). Onchocerca volvulus is believed to be the only species that develops to maturity and produces microfilariae in the skin of man, but there have been occasional records of animal species in a few patients in North America, Europe and the USSR. Animal Onchocerca are particularly common in the tropics in areas where the human disease is endemic, but human infection with these parasites would be difficult to distinguish from infection with O. volvulus.8

O. volvulus is not a uniform species throughout its range and, as can be seen in section 7, recent studies have revealed distinct geographical strains with differences in their vector infectivity and in their pathogenicity. These differences are thought to explain some of the major differences in the epidemiology of the disease in the rain-forest and savanna regions of Africa and in Latin America.

8 See, for example, Buck, A. A., ed., op. cit.; the species illustrated in the aorta of man in Fig. 1 of that publication could be O. armillata, a common parasite with its normal location in the aorta of cattle.
This section gives emphasis to the natural history of *O. volvulus* in relation to the pathogenesis of the disease; it also stresses the epidemiological significance of the various stages of the infection in man.

### 5.1 Immature worms

The routes followed by the infective larvae after entering the skin through the bite-wound of an infective *Simulium* are not known. It seems probable that the immature worms have some chemotactic mechanism by which they locate each other and by which newly inoculated worms can locate those already fixed in the body, for nodules are known to increase in size by accumulation of newly arrived worms.

There is some circumstantial evidence that a "prodromal" pruritus may occur in newly exposed subjects in association with the presence of immature and as yet infertile *O. volvulus* worms. The whole question of the excretory and secretory products of all stages of *O. volvulus* in the human host requires study, especially in its possible relation to pathogenesis.

### 5.2 Adult worms

Most adult worms lie fixed in the tissues and the pre-adult stages probably reach the adult sites within 2 months of the inoculation of infective larvae. Probably all adult worms are encapsulated and enmeshed in a protective coating of the host's fibrous tissue, which may be thin or thick. The manner in which the adult worms derive their nutriment from the host is not known.

The nodules are usually painless, but very large ones, those which lie over pressure points, and those undergoing degenerative changes may become tender or even painful. Mild inflammatory or pressure effects around worm bundles lying deep between the muscles, against the capsules of joints (especially the hip joint), or against the periosteum of the bones, may give rise to deep-seated aches and pains. Penetration of the skull by a nodule has been reported from Central America. The possibility that worms may occur in the orbit or cranium cannot be excluded.

Pain around nodules, both superficial and deep, is especially common during treatment with macrofilaricides. Abscesses around dead worms and extrusion of whole adult worms have been reported during treatment. The release of large quantities of antigen following the death of adult worms under treatment may give rise to a condition resembling serum sickness.

It is probable that in many patients deep-seated palpable worm bundles are more numerous than palpable subcutaneous nodules.
Nodules, particularly those on the head in Africa, may be very small and, as they are poorly encapsulated, they are sometimes difficult to detect. Such nodules may be very dangerous since they produce abundant microfilariae near the eye.

5.3 Microfilariae

Most of the signs and symptoms of onchocerciasis are attributable to the presence of microfilariae in the tissues. Microfilariae are responsible for the itching and inflammatory changes in the skin that can lead to a wide variety of dermatological sequelae. They are also responsible for the lymphatic pathology. In the eye they cause the punctate keratitis and they are also largely responsible for the sclerosing keratitis, anterior uveitis, choroidoretinitis, and optic neuritis leading to postneuritic atrophy.

Systemic onchocerciasis also results from microfilarial spread, particularly in heavily infected patients. The microfilariae are found in the blood stream, and in untreated patients concentrations of up to 30 microfilariae per milliliter of venous blood have been recorded. There is no evidence of a periodicity in the blood. The microfilariae tend to accumulate in the glomerular capillaries of the kidney, the capillaries of the pulmonary alveoli, and the choroid plexuses of the ventricles of the brain. From these sites they may enter the urine, sputum and cerebrospinal fluid.

The presence of microfilariae in the eyes of patients with onchocerciasis in Yemen, where the microfilariae are concentrated around the ankles, suggests that some microfilariae reach the eye via the blood stream. However, in heavily infected individuals the main route into the eyeball and the ocular media would appear to be from the skin of the face and along the sheaths of the ciliary vessels and nerves, rather than directly from the blood. Microfilariae have also been reported from tears and vaginal washings.

The ability of *O. volvulus* microfilariae to penetrate collagenous connective tissue (as in the skin and the cornea) distinguishes them from those of other species, which normally live in the blood. This property, which may depend on their enzyme activity, could account for their presence deep in the tissue of internal organs. Experimental studies suggest that immature microfilariae extracted from nodules are less

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*The term "systemic onchocerciasis" is used to cover the presence of *O. volvulus* microfilariae and all pathological manifestations caused by them in parts of the body other than the skin and eye.*

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pathogenic than mature specimens that have reached the tissues. This period of maturation has been observed in *O. gutturosa* of the cow and *O. cervicalis* of the horse.

Under the influence of diethylcarbamazine, which appears initially to "mobilize" microfilariae, large numbers of them may leave the blood and appear in the urine, sputum and cerebrospinal fluid. These migrations may be responsible for severe reactions, characterized variously by collapse, shortness of breath, coughing, and vertigo. Diethylcarbamazine also greatly increases the migration of microfilariae from the skin, through the lymphatics, into the blood stream, and microfilaraemias of up to 300 microfilariae per ml of blood have been recorded during treatment with this drug. Quantitative studies suggest that the majority of the *O. volvulus* microfilariae that enter the blood stream are destroyed inside the body. It has been estimated that heavily infected adults may harbour some 50–200 million microfilariae in the skin at any one time.

5.4 Epidemiological implications of the life span of *O. volvulus*

The durations of the various stages of *O. volvulus* in the human host are not known with exactitude, but within certain limits they may be inferred. The possibility that the life spans of the pre-adult stages, adult worms and microfilariae may vary with the strain of parasite should not be ignored, for it could have important consequences. Work on experimentally infected chimpanzees has shown that the prepatent interval—the time between inoculation of infective larvae and the first appearance of detectable microfilariae in the skin—varies between 10 and 20 months. Strains from the Cameroonian forest, the Cameroonian savanna, and Guatemala were all similar in this respect. Evidence from human infections in which individuals could be certain of the date of their only possible exposure to infection indicates a prepatent interval of 15 months in the forest zone of Nigeria, but an interval of 3 months has been reported in a case from the savanna zone of Chad. The prepatent interval does not indicate the time of maturation of the worms because it may take many months for sufficient microfilariae to concentrate in the skin at a detectable level.

The long prepatent interval may lead to infections first becoming manifest long after the patient has left the endemic area. It also implies that a recurrence of transmission in a previously controlled area, or the commencement of transmission in a new area, will not become detectable as new infections in the human population until a considerable time later. Likewise, the earliest *Simulium*-transmitted infections in children are
unlikely to be detected before the second year of life. The possibility that a fetus may acquire a direct transplacental microfilarial infection from parasites circulating in the blood stream of its heavily infected mother requires investigation.

The life span of the fecund adult female worms is generally not more than 15 years, a which implies that control operations against the vector must be continued for a period longer than this before the parasite can be expected to die out in the human population.

The life span of the microfilariae has been estimated at between 6 and 30 months. b The average life span certainly varies with the immunological state of the individual host, and may possibly vary as between different strains of the parasite.

5.5 Research needs

(1) The question of transplacental infection is of great interest in assessing the complete interruption of transmission in a given population, as well as in relation to the possibility that a state of immuno-tolerance to O. volvulus may originate from contact with stages of the parasite before birth. It is therefore recommended that:

(a) infants and young children should be included in prevalence surveys, and be examined for microfilariae and nodules;

(b) infants newly born to mothers with heavy infections should be examined and re-examined at 6-monthly intervals to study the changes that occur with time;

(c) the presence of microfilariae in the newborn, and in babies below the age of 2 years, should not be taken as proof that transmission of O. volvulus in a population is continuing, particularly if the mother of the baby is found to be infected;

(d) cord blood and umbilical cord tissues of newborn babies from mothers with severe infections should be examined for microfilariae.

(2) Studies are needed to determine more accurately the range in the life span of adult O. volvulus worms and of the microfilariae.

6. THE VECTORS

Simuliids or "blackflies" (Diptera, family Simuliidae) are the only known vectors of O. volvulus, both in Africa (Fig. 3) and in the western hemisphere (Fig. 4).

In Africa human onchocerciasis is transmitted mainly by members of the *Simulium damnosum* complex, but in East and Central Africa species of the *S. neavei* complex also transmit the disease.

**FIG. 3. RECORDED DISTRIBUTION OF THE CHIEF VECTORS OF *O. VOLVULUS* IN AFRICA**

*Map (revised and updated) reproduced by permission of the Trustees of the British Museum (Natural History) from: CROSEKEY, R. W. Simuliidae. In: Smith, K. G. V., ed. Insects and other arthropods of medical importance, London, British Museum (Natural History), 1972, p. 330. *S. damnosum* is considered to be a complex of sibling species (see text) and is not yet known to bite man in Yemen, where onchocerciasis occurs.*
FIG. 4. RECORDED DISTRIBUTION OF THE CHIEF VECTORS OF O. VOLVULUS IN MEXICO AND CENTRAL AMERICA

Prepared from the literature by Dr R. Garmo, Bernhard-Nocht Institute for Naval and Tropical Diseases, Hamburg, Federal Republic of Germany.

In the western hemisphere the situation is complicated by the presence of a large number of man-biting species. The vectorial status of many of them is not yet well established.

6.1 Africa and South Arabia — *S. damnosum* complex

*S. damnosum* was formerly regarded as a fairly uniform species, but chromosomal investigations have shown it to be a complex of forms which cannot be distinguished morphologically except by the banding patterns of the larval chromosomes.

Cytological criteria have been used to separate the West African *S. damnosum* complex into 8 species. A further 11 cytopotypes have been

* See Vajime, C. G. & Dunbar, R. W. *Tropenmed. Parasitol.*, 26: 88 (1975). The descriptions of the new species are based on cytological and statistical criteria and not on morphological characters. They may not be accepted by all taxonomists. The species concerned are: *S. damnosum* "Nile" form; *S. dieguense* Vajime & Dunbar (= "Dieguera" form); *S. sanctipauli* Vajime & Dunbar (= "Bandama" form); *S. sirbanus* Vajime & Dunbar (= "Sirba" form in part); *S. soumbense* Vajime & Dunbar (= "Soumbé" form); *S. squamosum* Enderlein (= "Bille" form); *S. zukanense* Vajime & Dunbar (= "Sirba" form in part); *S. yahense* Vajime & Dunbar (= "Yah" form).
recorded from East Africa but they have not yet been described as separate species.

6.1.1 Distribution
The *S. damnosum* complex is widely distributed in the Ethiopian zoogeographical region. The northern limit of its range reaches latitude 15° N in the rainy season in West Africa and latitude 19° N (along the Nile river) in northern Sudan. The southern limit in Africa is latitude 34° S. *S. damnosum* s.l. is also known from Yemen.

Several of the species have well defined geographical distributions. In West Africa, *S. damnosum* sensu stricto, *S. sirbanum*, *S. sudanense* and *S. diepwereense* are mainly distributed in the savanna areas, while *S. sanctipaulii*, *S. soudrense* and *S. yahense* are typical of the forest zone. *S. squamosum* is widespread in the forest and Guinean-savanna zones of Cameroon, and has also been found in one savanna focus in Upper Volta. *S. damnosum* s.s. is the only species found in both West and East Africa.

6.1.2 Biology and ecology
The bionomics of the *S. damnosum* complex were reviewed recently. 

6.1.2.1 Aquatic stages. Immature stages of *S. damnosum* s.l. inhabit a wide variety of water courses in different bioclimatic zones, from very large rivers to small streams. This spectrum reflects the different ecological requirements of various species.

Among the chief ecological factors governing the existence of breeding places are adequate water velocity (usual range 0.70–1.20 m/sec), which is linked with food supply, and the presence of suitable supports. The duration of larval development depends on the water temperature. In West Africa it usually takes 8 to 10 days. Other factors influencing the productivity of breeding sites are: hydrological variations, species competition, parasites, predators, and man-made factors.

There are indications that different species settle on different kinds of support (rocks or vegetation). Preliminary observations have shown that some hydrochemical factors (e.g., pH and conductivity) may have some influence on the development and distribution of the larval populations of certain cytotypes.

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6.1.2.2 Adult females. Females of the species *S. damnosum* s.l. bite from dawn to dusk, usually outdoors and on the lower parts of the body. Almost nothing is known about the differential behaviour of the members of the complex. Biting densities of the fly populations are primarily governed by the productivity of the breeding sites, which depends on the hydrological conditions and may be subject to considerable seasonal variations. Several types of annual variations in fly populations can be observed.\(^a\) Firstly, the production of flies may vary according to the water level, giving a high density in the rainy season and a low density in the dry season (synchronous variation). Secondly, breeding places may produce flies mainly during the dry season at low water levels (inverse variation). Thirdly, breeding places may be active at both low and high water levels (bimodal variation). Lastly, productivity may be more or less constant throughout the year, as, for example, in small forest streams.

Daily biting cycles are mainly governed by air temperature and humidity. At high temperatures during the dry season the daily biting curve shows 2 peaks, one in the morning and one in the afternoon, separated by a midday trough. At low temperatures during the rainy season the daily biting curve is unimodal, with a single afternoon peak.

A dispersion flight may take place at each gonotrophic cycle before the blood-meal. It is influenced essentially by vegetation, cloudiness and humidity. In the true Sudan-savanna zone, dispersal is linear along the water courses during most of the year and only becomes radial (in all directions from the breeding sites) when clouds provide protection. In the Guinea-savanna zone it is linear in the dry season and radial in the rainy season. In forest areas radial dispersal is possible throughout the year, though it is more pronounced in the rainy season.\(^a\) In the savannas nulliparous flies tend to disperse greater distances from the river than do parous flies;\(^a\) this may be different in the forest.

* S. damnosum* s.l. females have considerable intrinsic powers of flight, which may sometimes be reinforced by the wind. Flight distances of up to 79 km over a period of 24 hours have been reported for marked flies moving along a large river in West African forest.\(^a\) In the OCP area flies

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\(^a\) Le Brez, R. *Contribution à l'étude biologique et écologique de Simulium damnosum Theobald, 1903 (Diptera, Simuliidae).* Paris, Office de la recherche scientifique et technique outre-mer, 1966 (Mèm. ORSTOM No. 17), 204 pp.
have reappeared more than 150 km upstream from the nearest known untreated breeding sites. In some northern savanna areas in West Africa non-permanent breeding sites are repopulated by flies coming from southern permanent breeding sites, possibly by long-distance migration along rivers, or by wind carriage, or by step-by-step colonization via intervening breeding sites. Alternatively, in other areas, the possibility of adult females aestivating during part of the dry season should be considered. The problem of aestivation was discussed extensively in the second report of the WHO Expert Committee on Onchocerciasis. Flight range may be shortened by infection with *O. volvulus*.

Wherever *S. damnosum* s.l. occurs in West Africa the females can be found feeding on man. Anthropophily is facultative and, in the presence of animals and in uninhabited areas, the flies may exhibit a high degree of zoophily. Zoophily has been confirmed: (1) by direct observations of flies feeding on birds and animals; (2) by means of the few precipitin tests made on blood-meals from wild-caught flies; (3) by the occasional findings of remains of avian blood in females; and (4) by the presence of developing filariae, other than *O. volvulus*, in the flies. Zoophily may be indicated also if infection rates of fly populations are extremely low in comparison with those of the human populations, with the provision that the flies concerned are normally susceptible to the local strain of *O. volvulus*. Non-anthropophilic *S. damnosum* s.l. populations have been recorded from East Africa (certain regions of Kenya, Uganda and the United Republic of Tanzania, and at high altitudes in Ethiopia, Rwanda and Burundi). In East Africa, anthropophily is apparently limited to certain cytotypes, whereas in West Africa no correlation has yet been established between species and host preferences.

The maximum longevity of *S. damnosum* females is unknown but there seems to be agreement that the life span does not exceed 1 month. Longevity can be determined by studying the regression of fly populations after the elimination of their breeding sites, or by capture/marking/release/recapture experiments. For epidemiological purposes it is necessary to know the proportion of females within the population that has reached the epidemiologically dangerous age at which they are potentially infective. This can be estimated indirectly by establishing survival probabilities from the age composition of fly populations as expressed by their parous rates. The life expectation of forest flies, as established by parous rates, was found to be shorter than that of flies from savanna areas, but there

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may be considerable regional variations, at least in forest areas. However, it is not certain whether low parous rates are really indicative of short-lived populations. They may result from the differential dispersal or migration of parous females. Longevity is influenced by environmental conditions and possibly depends on genetic factors. It may also be reduced by predators and by infestation with parasites. Parasites commonly observed in *S. damnosum* s.l. females, in addition to *O. volvulus*, include fungi (in the ovaries), ciliates, microsporidia, mermithids, filariae and other nematodes.

6.2 Africa — *S. neavei* group

6.2.1 Species and distribution

The *S. neavei* group (subgenus *Lewisellum*) includes all species with larvae and pupae that attach themselves to riverine crabs of the genus *Potamonauta*. This is usually referred to as a phoretic association. The group is taxonomically difficult. Only a few of its members can be distinguished easily on morphological characters. Cytotaxonomic studies have not yet been carried out, and are urgently required. Within the *S. neavei* group, *S. neavei* s.s. is an important vector of *O. volvulus* in parts of Uganda. Formerly it was responsible for transmission in Kenya, where it has now been eradicated except along the Ugandan border. *S. woodi ethiopiense* bites man in Ethiopia, while in Malawi and the United Republic of Tanzania *S. woodi* is the most important vector within the group. In some areas of Tanzania, *S. nyasalandicum* s.l. bites man, but the species has not yet been found naturally infected with *O. volvulus*.

6.2.2 Biology and ecology

6.2.2.1 Aquatic stages. The sites of oviposition are not known. Larvae at different stages of development can settle on crabs. First-instar larvae have not been observed. Preimaginal development is exceptionally long and may last several weeks. Breeding does not occur in large rivers even when crabs are present.

6.2.2.2 Females. The flight range is much more limited than that of *S. damnosum* s.l. Most biting occurs in forest, including gallery forest, but flies may bite man in relatively open woodland under moist or cloudy conditions. They usually bite the lower parts of the body. Vegetation cover seems to be a prerequisite for species in the *S. neavei* group.

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* Le Berre, R., op. cit.
Deforestation in some areas has led to complete disappearance or a drastic reduction in the flies' abundance. There is evidence from Kenya of *S. neavei* feeding on domestic animals, and the presence of flies in uninhabited areas, together with the occurrence of nonhuman filariae in *S. neavei* on Mount Elgon on the Kenya/Uganda border, indicate a considerable degree of zoophilicy.

### 6.3 Other potential vectors in Africa

Natural infection with *O. volvulus* has been found in *S. dukei* in forest in the United Republic of Cameroon. This species has a phoretic association with prawns. *S. albivirgulatum* is a highly anthropophilic species in Central Africa, but its vectorial significance has not yet been established. Further species reported as occasionally landing on or biting man include *S. ovazzae*, *S. bovis*, *S. griseicollis*, *S. adersi*, and *S. vorax*. Experimentally, *S. adersi* and *S. vorax* support the development of *O. volvulus* to the infective stage.

### 6.4 Latin America

Latin American vector species were extensively reviewed in a recent publication. They will be discussed only briefly in the present report.

#### 6.4.1 Guatemala and Mexico

Three species, in order of importance *S. ochraceum*, *S. metallicum* and *S. callidum*, have been incriminated as vectors by several authors. *S. ochraceum* is considered to be the main vector since it is highly anthropophilic, reaches highest biting densities in the endemic areas, bites the upper parts of the human body where microfilariae are most numerous, and shows the best survival after ingestion of *O. volvulus*. *S. metallicum* and *S. callidum* are more zoophilic. All 3 species can also be found outside the endemic areas. *S. gonzalezi*, *S. veracruzana* and *S. haematopotum* are considered possible vectors under limited circumstances. Feeding experiments have shown that *O. volvulus* can complete

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\[9\] \textit{Dalmat, H. T. The black flies (Diptera, Simulidae) of Guatemala and their role as vectors of onchocerciasis.} Washington, DC, Smithsonian Institute, 1955 (Smithsonian Miscellaneous Collections, vol. 125, No. 1).
its development in *S. ochraceum*, *S. metallicum*, *S. callidum*, *S. veracruza-
num* and *S. haematopotum*.

Natural infections with larvae indistinguishable from those of *O. vol-
vulus* have been found in *S. ochraceum*. However, infectivity rates were
extremely low, in contrast to those observed in *S. damnosum* in West
Africa. This may explain the extraordinary situation in Guatemala,
where onchocerciasis foci have remained small and limited, and have
been stable for the last 40 years, despite large-scale movements of the
human population (seasonal workers in the coffee plantations) and the
fact that *S. ochraceum* also occurs outside the endemic area. This indi-
cates that only in areas with very high fly densities—said to occur in
Guatemala only in the onchocerciasis zones—may the disease be self-
perpetuating.

The more zoophilic behaviour of *S. metallicum*, *S. haematopotum* and
*S. gonzalezi* has been demonstrated by the fact that infections with
developing stages of filariae have been found also in areas where oncho-
cerciasis is not present. The infections were believed to be of bovine
origin. Infective filarial larvae clearly distinguishable from those of
*O. volvulus* have been seen in *S. metallicum*.

*S. ochraceum* shows a preference for rugged mountainous terrain and
is collected most commonly at altitudes between 900 and 1500 m. The
breeding sites are very small streams, usually concealed by a thick over-
growth of vegetation and by a dense canopy. In Guatemala and Mexico
these habitats are found typically in the coffee-growing areas and in
forested country. The duration of larval development under natural
conditions has not been adequately studied but is probably several weeks.

Longevity and flight range have been investigated by means of capture/
marking/release/recapture experiments. The maximum flight range re-
corded was 10 km, and one female was recaptured 62 days after release.
This provides evidence that *S. ochraceum* can live for a long time and may
travel considerable distances, even in mountainous or forested areas. The
species is generally exophagic but, at high densities, may even enter
houses or bite at night.

The other species are less fastidious in their larval habitat requirements
and are more adapted to differences in breeding sites. They also inhabit
larger water courses at lower altitudes. When biting they usually prefer
to feed on the lower parts of the body, unlike *S. ochraceum*.

6.4.2 *Venezuela*

*S. metallicum* seems to be the main vector but *S. exiguum* is con-
sidered to be a secondary vector. *S. metallicum* bites the lower parts of
the human body but *S. exiguum* is less particular as to the site of the
blood-meal. *S. metallicum* is found almost exclusively in medium-sized
streams and *S. exiguum* only in large streams and rivers.

6.4.3 Colombia

*S. exiguum* is assumed to be the vector. It is mainly zoophilic but may
become anthropophilic in the absence of cattle and horses.

6.4.4 Brazil

Natural infections with developing larvae, thought to be those of
*O. volvulus*, have been found in *S. amazonicum*, but as this species is
known to be a vector of *Mansonella ozzardi* the filarial species in the skin
of man in Brazil must be carefully distinguished.

6.5 Factors influencing transmission

6.5.1 Man-fly contact

Contact between man and the vector is an obvious prerequisite for
transmission. The intensity of this contact depends mainly on:

(a) the density of the vector and of the human population;

(b) the bionomics of the vector, its host preferences, dispersal, migra-
     tion, and daily biting activities, which may all vary at different times
     of the year; and

(c) human factors, such as location of communities, daily or seasonal
     activities, and migration, in relation to breeding sites and movements
     of vector females.

6.5.2 Factors influencing the intake and development of the parasite

The main factor determining the proportion of vector females be-
coming infected with *O. volvulus* is the prevalence and intensity of micro-
filaria carriers in the human population.

The microfilariae may be attracted by the biting females, and the
number of microfilariae ingested may be very high (in some instances
several thousand microfilariae have been seen in single flies) in com-
parison with the densities observed in the human skin. Only very few of
the ingested microfilariae survive. The proportion developing shows
regional variations related to the strain of the parasite and the species of
vector. In the West African forest zone, the average number of infective
larvae of *O. volvulus* in infective *S. damnosum* s.l. has been observed to be
about 6, whereas in the savanna areas the average is only just over 2. The
heavy mortality of microfilariae after ingestion is due either to the formation of a peritrophic membrane, for example in *S. damnosum* s.l., or to the buccopharyngeal armature, for example in *S. ochraceum*. There is little mortality of larvae during their intramuscular development in a compatible vector. Very high intakes of microfilariae may be lethal to the flies.

The female flies can transmit only if they survive until the parasite has reached its infective stage. At an average temperature of 25°C the development of *O. volvulus* in *S. damnosum* takes 6-7 days. Low temperatures are limiting factors, as observed during the dry season in the savanna in West Africa and at high altitudes in East Africa and Central America. The minimum critical temperature for the development of *O. volvulus* in *S. woodi* in Tanzania is 18°C.

Flies that ingest microfilariae at the first blood-meal will be carrying developing larvae when they feed for the second time, but usually they will not be infective until they take a third blood-meal. Flies coming for their third blood-meal are therefore potentially infective and have reached the epidemiologically dangerous age.

In order to compare the amount of transmission in different areas, at various seasons or during control campaigns, the transmission potential has been used as a parameter. It is defined as the number of infective larvae of *O. volvulus* carried by flies coming to bite one man during a certain period of time. For epidemiological purposes, the “annual transmission potential” *a* can be used for comparison of transmission in different areas.

In the forest zone of West Africa annual transmission potentials recorded from different places have given values ranging from 50 to 90,000 infective larvae/man/year. In this bioclimatic zone even the highest figures are not associated with high blindness rates or with desertion of villages. In the Sudan-savanna zone of West Africa annual transmission potentials ranging from 300 to 18,000 infective larvae/man/year have been recorded at or near villages with near 100% *O. volvulus* prevalence rates in the human population. Values above 1500 are associated with a high prevalence of blindness, and values of 2500 and over may be associated with the desertion of villages, except in particularly favourable socioeconomic situations.

The annual transmission potential does not indicate the actual number of larvae transmitted to any person in the local population since fly catches are carried out at selected sites, and in reality no person is fully

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exposed 12 hours a day throughout the year. Furthermore, not all third-stage larvae are actually infective, not all of them leave the vector during the blood-meal, and not all those that leave the vector succeed in entering the host.

6.6 Entomological methods for the assessment of the factors related to transmission

For the assessment of the transmission potential the following parameters have to be determined.

(a) Biting density. This is the number of flies coming to bite man in a given period of time (number of flies per man/day or man/year). It is determined by standardized vector catches on human bait. Catches should be made during the whole day from dawn to dusk and repeated at regular intervals for at least 1 year. The selection of catching points is very important. They should be situated at known distances from the breeding sites, and should be selected according to the activities of the human population. Flies may be caught in single tubes, or with sucking tubes in the case of very high biting densities. Flies should be separated into hourly batches. If dissections cannot be done immediately, it is necessary to preserve the flies in low temperature containers.

(b) Species composition. The flies caught must be identified before dissection. It is necessary to separate out species that, though they sometimes land on man, do not bite him.

(c) Age composition. No external characteristics have been found for age-grading of flies. All flies, or representative samples of each hourly batch, have to be dissected in order to determine parous rates. Parous and nulliparous females may be distinguished by the following criteria.

(i) Follicular relics are present in ovaries of parous flies only.
(ii) The ovaries can be spread and stretched easily in parous flies, while those of nulliparous flies are fragile. Furthermore, parous ovaries have a granular appearance and are less translucent than the ovaries of nulliparous flies.
(iii) Retained eggs may be present in a certain number of parous flies.
(iv) Spermatophores are present in young nulliparous flies for some time after mating.
(v) The fat-body is rather large and voluminous in nulliparous flies, while it is reduced or completely absent in parous flies.

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(vi) In most species the Malpighian tubules are rather dark in young flies, and flies with clear tubes are regarded as "old parous".

(vii) Meconium (remains of preimaginal tissues) may be present in young nulliparous flies.

The presence or absence of follicular relics is the only completely reliable criterion. The fat-body is mostly used in routine dissections, but it is not completely reliable (e.g., in S. ochraceum). It is not yet possible to distinguish uniparous and multiparous females.

(d) Infection rates. The head, thorax and abdomen have to be dissected separately. First-stage short sausage forms and second-stage long sausage forms of O. volvulus are present only in the thoracic muscles, but they may be displaced during dissection. Third-stage infective larvae can be found anywhere in the body of the fly, not merely in the head.

To estimate the transmission potential, all third-stage larvae that have left the muscles should be counted, irrespective of their location in the fly.

The number and location of the different stages of O. volvulus in the fly must be recorded. It is necessary to distinguish between flies with (i) developing larvae (first and second stages); (ii) infective third-stage larvae (in any part of the body), which are termed "infective" flies; (iii) 2 broods of larvae (first and third stage) derived from separate blood-meals—referred to as "double infections".

Not all filarial larvae in simuliiids are O. volvulus and in some areas other species predominate. Some species are readily distinguished, even in wet preparations, because of their size or characteristic posterior morphology. All infective larvae should be preserved in glycerol/alcohol for further study. The techniques for preserving and differentiating infective larvae have been described in detail in previous reports of the WHO Expert Committees on Onchocerciasis a and Filariasis. b Records should also be kept of any other parasites found in the flies.

6.7 Research needs

6.7.1 Simulium damnosum complex

(a) Further information is needed on the distribution of various members of the S. damnosum complex.


(b) Members of the *S. damnosum* complex can at present be identified only by examination of the polytene chromosomes of the larval salivary glands. A more practical method of identifying larvae, perhaps by means of diagnostic morphological characters, is urgently needed to accelerate ecological studies on larval populations of the different members of the complex.

(c) More sophisticated methods (serological and biochemical) should be applied to clarify the taxonomic relationship of the species.

(d) Adults of different species of the *S. damnosum* complex cannot be identified cytologically. Their identification is only possible by examination of larvae reared from the eggs of wild-caught females. More practical methods for their identification have to be developed (these may be cytological, morphological, or biochemical).

(e) Information is urgently needed on the bionomics, biting behaviour, host preferences, dispersal, migration, longevity, and vectorial capability of each species in the complex. This will be possible only after methods of identifying females have become available, but valuable information could be collected immediately in areas where only one species is present.

(f) Marking techniques should be used to study migration patterns of females in order to trace the sources of flies reappearing in controlled areas. The possibility of mass-marking of preimaginal stages should be investigated. Such populations may be marked naturally with certain minerals that may be detected in adults by spectrography.

(g) Entomological methods for assessing the effect of control measures are based on vector collections on human bait. Trapping methods should be studied more thoroughly in order to detect very low densities in control areas.

(h) Ecological factors influencing the distribution of the species of the *S. damnosum* complex have to be assessed (physicochemical conditions of the breeding sites, food requirements of the larva, predation, climate and vegetation). Information is needed on how artificial changes of the environment, e.g. deforestation, may alter the distribution and biology of the species. Studies should be designed to determine whether, after elimination of certain cytotypes in control areas, other cytotypes may establish themselves.

(i) The transmission potential of vector populations needs to be studied in more situations in order to correlate it with the pattern of infections and clinical manifestations in man. The results may help to evaluate the critical transmission potential below which serious manifestations do not occur in man.
6.7.2 Simulium neavei group

Cytological methods should be applied to this group to clarify the taxonomic status of the species and to elucidate the biometrics of the different species and transmission patterns in East Africa. Recommendations made for the S. damnosum complex also apply to this species group.

6.7.3 Latin American vectors

The vectorial status of many man-biting simulids has not yet been established except in a few areas and investigations should be made to determine the vectorial importance of the main anthropophilic species in endemic onchocerciasis areas. Specific research recommendations were given in the proceedings of the 1974 PAHO symposium. These are endorsed by the Expert Committee.

6.7.4 Filarial infections other than O. volvulus in vectors

Assessments of Simulium transmission intensities are hampered by the fact that simulids are not only carriers of O. volvulus but may also harbour larvae of other filarial species, some of which cannot yet be distinguished from O. volvulus. The following investigations should be carried out in onchocerciasis areas.

(a) Survey of other filarias, especially Onchocerca species, present in domestic and wild animals.

(b) Assessment of simulid species feeding on these animals, and experimental studies on the possible development of animal filariae in the species concerned.

(c) Comparison of the developing, abortive and infective stages found in experimentally infected flies with those observed in naturally infected flies. Development of techniques for differentiating developing stages of O. volvulus from those of other species.

7. ONCHOERCA VOLVULUS – SIMULIUM COMPLEXES

7.1 Transmission studies

Transmission experiments involving volunteers have been carried out in the United Republic of Cameroon, elsewhere in West Africa, and in

\* PAN AMERICAN HEALTH ORGANIZATION, op. cit.
Latin America. They indicate the existence of a number of different strains of *O. volvulus*, each associated for transmission with a particular species (or form) of *Stomilum*. 

*O. volvulus* microfilariae of the Cameroon forest strain are ingested and develop well to infective larvae in the local forest zone vector (presumed to be *S. squamosum*). By contrast, when the usual vector from the Sudan-savanna zone of Cameroon (presumed to be *S. dannosum s.s.*) is fed on carriers of the forest strain of *O. volvulus*, although microfilariae are ingested in similar numbers, very few or none of them complete their development to infective larvae. Conversely, microfilariae of the Cameroon Sudan-savanna strain of *O. volvulus* are found to develop well in their local vector, but very few or none complete their development in the forest-zone vector.

In a noncompatible vector there are two main ways in which ingested microfilariae are eliminated. They may fail to escape from the peritrophic membrane, or they may so escape only to die later in the course of their migration through the haemocoele, or during their development in the thoracic muscles. Death usually occurs at the time when they attempt to moult from first-stage to second-stage larvae, or when the preinfective larvae attempt their final moult.

Although in Cameroon the forest *O. volvulus-Stomilum* complex, or closely related subcomplexes, extend northwards to cover almost the whole of the Guinea-savanna zone, in more westerly parts of Africa the situation is different. The strain of parasite from the Sudan-savanna zone (normally transmitted by presumed *S. dannosum s.s.* or *S. sirbanum*) can be transmitted well in other zones by a variety of *Stomilum* species (presumed *S. dannosum s.s.*, *S. sirbanum* and *S. squamosum* in the Guinea-savanna zone, and *S. sanctipauli* in the forest-savanna mosaic zone), but it is only poorly transmitted by the local forest vector (presumed *S. yahense*). The local forest parasite is not transmitted by the true Sudan-savanna strain vectors of the same longitude.

It has also been shown, using volunteers, that African strains of *O. volvulus* from the Sudan-savanna and forest zones of the United Republic of Cameroon will develop very poorly or not at all in the main Latin American vector simuliids, *S. oestracum* and *S. metallicum* in Guatemala, and *S. metallicum* and *S. exiguum* in Venezuela. Likewise a Guatemalan strain of *O. volvulus* in an experimentally infected chimpanzee failed to develop in the Cameroon forest or Sudan-savanna vectors (presumed *S. squamosum* and *S. dannosum s.s.*, respectively). These intercontinental experiments have also revealed the presence of a remarkable tropism among skin-dwelling *O. volvulus* microfilariae towards feeding
Simulium of the appropriate vector species. When feeding on human carriers with similar concentrations of microfilariae in the skin, the American vectors (S. ochraceum and S. metallicum) ingested some 10-25 times more microfilariae from a carrier of an American strain than they did from carriers of either of the main West African strains (forest or Sudan-savanna). Similarly, presumed S. squamosum and S. damnosum s.s. ingested 2-4.5 times more microfilariae from carriers of either West African strain than they did from a carrier of the Guatemalan strain with similar concentrations of microfilariae in the skin.

The Onchocerca-Simulium complexes in East and Central Africa and in Yemen have not yet been investigated by means of cross-transmission experiments. The poor transmissibility of certain strains of O. volvulus by species (or forms) of Simulium outside their home-complex area may be a factor limiting the spread of the parasite. However, the existing “barriers” between complexes have evolved over past ages when population movements were limited. In the present context of wider large-scale population movements it must be remembered that mutual nontransmissibility is never absolute. Filarial parasites, subjected to intensive artificial selection, have been induced experimentally to adapt to previously unsuitable vectors.

7.2 Strains of O. volvulus and their pathogenicity

The discovery, by means of transmission experiments, of the existence of distinct strains of O. volvulus has led to investigations into their differential pathogenicity. These investigations are relevant since the pattern of clinical onchocerciasis, especially in regard to eye lesions and blindness, has long been known to differ from one topographical area to another and, most markedly, between the Sudan-savanna and forest regions of West Africa.

The rabbit eye has proved to be suitable for investigating the pathogenicity of O. volvulus microfilariae. It has been shown that microfilariae of the Cameroon Sudan-savanna strain of O. volvulus, when inoculated subconjunctivally, were about 3 times more invasive for the cornea, and produced much more severe lesions therein, than did microfilariae of the Cameroon forest strain. On the other hand, when microfilariae were inoculated into the posterior segment of the rabbit eye (into the vitreous or subretinally), although lesions were produced that closely resembled the fundus lesions of onchocerciasis in man, there was no detectable difference between the pathogenicity of the Cameroon Sudan-savanna and forest strains. Indeed, some of the most severe lesions were associated with the forest strain.
These findings in the rabbit eye, coupled with the results from surveys of ocular onchocerciasis in the Sudan-savanna and forest zones of West Africa, suggest that the ocular pathogenicity and other characteristics of the strain of parasite may be important factors governing the observed differences in the prevalence of blindness due to sclerosing keratitis in the two zones.

However, the importance of the strain of parasite has to be considered against the background of the social and behavioural patterns of the human host, the state of immunity of the individual, the intensity of transmission, and environmental factors, such as nutrition and the degree of exposure of the eye to sunlight.

The comparative pathogenicity of microfilariae of other strains of *O. volvulus*, especially those from Latin America, has yet to be investigated.

### 7.3 Research needs

1. Complementary transmission experiments should be carried out on the natural association between different strains of *O. volvulus* and certain species of *S. damnosum* s.l., and on their ability to develop in other species of the complex, or in the same species originating from other areas. Special attention has to be paid to the question how the transmission pattern may be altered after massive immigration of human populations infected with a different strain of *O. volvulus*, and whether the imported strain can adapt itself to the local *Simulium* populations. In particular, in parts of West Africa where there are now established villages of savanna immigrants who have been living in forest areas for more than 1 generation, the transmissibility of their *O. volvulus* parasites should be investigated in the local *S. damnosum* vector, as well as in the vectors from their villages of origin in the savanna.

2. Further work should be done to distinguish the strains of *O. volvulus*, using biochemical and biological techniques, and to compare the clinical, pathological and epidemiological patterns of onchocerciasis in different geographical and bioclimatic zones. It is necessary to relate the patterns of infection to the strains of *O. volvulus* involved and to the dynamics of transmission by the different *Simulium* vectors concerned.

### 8. HUMAN HOST FACTORS

The frequency of nodules in persons with microfilariae of *O. volvulus* may vary widely between endemic areas from a reported minimum of 21% in Colombia to a maximum of nearly 100% in some villages in West Africa.
In most areas more men than women have nodules, and children with positive skin snips are less likely to have nodules than are adults. Racial factors may be of significance; for example, some ethnic groups in Africa having a fibroblastic diathesis are more prone to develop keloids and lichenification of the skin, and they have a lower frequency of exogenous dermatitis than Caucasians. It has been speculated that hormonal factors may be responsible for the observed differences between males and females.

In African communities with borderline nutrition, the skinfold thickness in women of child-bearing age is about twice that of men in comparable age groups. Skin snips taken with a punch biopsy instrument by a single investigator may vary in weight and surface area, and from one individual to another. Specimens are more easily obtained from people with thin, dry skin than from those whose skin is thick and oily. Constitutional and acquired differences in skin thickness and texture may affect the sensitivity of the skin snip method.

Other host factors that can influence microfilarial densities in skin snips are those that regulate the periodic diurnal variations of the numbers of microfilariae in the upper layers of the skin. There are considerable interpersonal variations in amplitudes between the daily maxima and minima of microfilarial densities in the skin. There are also differences in the apparent attractiveness of individuals to *Simulium* bites. These could be of considerable importance in relation to transmission and severity of infection and disease.

Geographical differences in the frequency and types of the lymphatic manifestations of onchocerciasis are reflected in various conditions, including: lymphadenitis, involving most frequently the superficial regional lymph nodes draining areas of onchocercal dermatitis; hanging groin; and elephantiasis. Lymphatic complications, other than circumscribed lymphadenitis, are said to be absent in the onchocerciasis foci of the western hemisphere. In Africa, inguinal and crural lymphadenopathies, hanging groin and elephantiasis are much more frequent in the West African rain-forest and East African regions than in the West African savanna. Elephantiasis is found associated with endemic onchocerciasis in the Central African Republic, Chad, Sudan and Zaire. Independent etiological factors may coexist with endemic onchocerciasis in some areas but may be absent in others.

Nonfilarial elephantiasis of the lower limbs is now recognized as a prevalent disease with focal distribution in Burundi, Ethiopia, Kenya, Rwanda, Tanzania and Uganda. The present hypothesis about its etiology postulates that an accumulation of inorganic materials in lymph
nodes draining affected areas may lead to blockage of lymph flow, and ultimately to elephantiasis. It is thought that toxic silicates contained in certain types of tropical soil enter the body through abrasions of the feet and lower legs.

Other important contributory factors must also be considered in the complex pathogenesis of onchocercal elephantiasis, namely secondary infections of the limbs in cases with pruritic dermatitis, genetic factors involving the anatomy of the lymphatic system, and co-endemic infections with lymph-dwelling filariae.

Cases of lymphadenitis seen in different endemic areas of onchocerciasis also show characteristic histopathological differences—such as those between souda in Yemen and the lymphadenitis in Africa—which suggest different host response mechanisms to the microfilariae of *O. volvulus*.

It is known that malnutrition increases host susceptibility to some infectious diseases and that it interferes with host defences based on immunological response. On the other hand, severe infections themselves are known to precipitate malnutrition. Some cases of heavy infection with *O. volvulus* may itself be the cause of wasting having been obtained in Chad, where intensity of infection was quantitatively associated with weight loss.

Impaired immune responses in persons heavily infected with *O. volvulus* have been observed in Chad. These deficiencies were more pronounced in persons with a low weight height ratio and with severe disease than in the lightly infected and clinically inapparent cases. The immunological deficiencies included: anergy to purified protein derivatives of tuberculins; poor response to yellow fever vaccine; absence of complement-fixing antibodies to *S. mansoni* in onchocerciasis patients with schistosomiasis; high frequency of anticomplementary serological activity; low titre of antibodies to *O. volvulus* antigen; and persistence of malaria parasitaemia in adults. It is reasonable to postulate that onchocerciasis is no exception to the general rule and that malnutrition, through its influence on the immune response, can aggravate the clinical manifestations of this disease, while onchocerciasis may itself be a contributing factor upsetting the delicate balance of the nutritional state in the often poorly nourished people of endemic areas.

Information on whether specific genetic population characteristics may influence the distribution and severity of onchocerciasis is not available. In particular, haemoglobin phenotypes, blood groups, red-cell enzyme deficiencies, intestinal disaccharide (lactose) deficiency—to list only some conditions that have received much study in rural Africa—
have never been examined in relation to the pronounced geographical differences in onchocercal disease manifestations.

People living in endemic areas of onchocerciasis are frequently exposed to a great variety of other infectious agents that are known causes of mortality and morbidity in their own right. Their possible importance as modifiers of the clinical manifestations of onchocerciasis, through either direct or indirect interaction, is unknown, despite the high frequency with which combined chronic infections can be observed in the field.

At times Simulium bites may themselves cause disease, as was recently observed in communities in Brazil along the trans-Amazon and Santarém-Cuiabá highways. The illness is characterized by a haemorrhagic syndrome with thrombocytopenic purpura and internal haemorrhages. Children and very young adults among new migrants are most frequently affected, whereas long-time residents are usually spared. The disease is said to be particularly frequent during the rainy season when the Simulium population reaches its peak. Antibodies to Simulium extracts were present in a significantly higher proportion of patients than of controls.

9. ENVIRONMENTAL AND SOCIOECONOMIC FACTORS

Environmental factors affecting the human host are clearly often closely related to the cultural, social and behavioural characteristics of the individual. Exposure to the bites of infected simulids is highest for families that live or work near the banks of a river in which the flies breed. The location of their houses may be related to a number of factors, such as the occupation of the head of household, the traditional patterns of land distribution within a village, and the availability of and easy access to alternative water sources for domestic use.

The kind of clothing and headgear worn in different endemic regions may have important implications for the epidemiology of onchocerciasis. The area of uncovered skin available to Simulium may determine to a considerable degree the site and distribution of adult *O. volvulus* and microfilariae in the body.

Large segments of the rural population of Africa are engaged in seasonal activities that can alter their risk of exposure to infected female simulids.

Man-made changes of the environment have pronounced effects on the epidemiology of onchocerciasis. The construction of large dams for hydroelectric projects or other purposes is beneficial because of the
definite suppression of breeding sites over long distances upstream. However, new Simulium breeding sites may also be created in the vicinity of dam spillways, and the productivity of existing breeding sites may be maintained at a constant high level further downstream. Other problems of highly intensified rainy season transmission through breeding on the spillways, gates and channels have arisen from the construction of many small earth dams and local irrigation schemes (e.g., for rice-growing) in savanna areas.

Human waste and refuse, resulting from relatively high human population densities in the vicinity of an otherwise suitable breeding site, may cause heavy contamination of the river water and make it unsuitable for Simuliiid breeding. Also, effluents from industrial plants and sewage systems can destroy breeding sites.

Several of the environmental factors relating to the vectors are referred to in section 6. Simulids require running water with a high oxygen content for the development of the larval stages, but many of the ecological factors determining the distribution of the different species are unknown. Recent studies on the physicochemical characteristics of breeding sites suggest that even small chemical differences may account for the presence or absence of different members of the S. damnosum complex. The amount of nutritive material in river water is of considerable importance, and some rivers with low nutritive values, which in all other respects seem suitable for breeding, remain free of S. damnosum. At the same time, rivers containing an excess of organic material, or an excess of algal growth, also prevent breeding. This is exemplified by the Nile, where the very slow reinestation with S. damnosum after treatment with DDT was believed to be due to excessive algal growth on the supports normally used by the larvae.

Water temperature is an important limiting factor for the breeding of some vector species; for example, members of the S. neavei complex are mainly limited to the cooler mountain streams, although the crab hosts with which they live in phoretic association are much more widespread. Temperature also affects the biting behaviour of the adult flies and the development of the parasite in the vector. The nature and profile of the river bed is very important in permitting or preventing breeding. This is well illustrated by studies in West Africa, which have shown that S. damnosum breeding sites are often man-made. Many of the spillways from dams and causeways are ideal breeding sites, but careful design can completely prevent breeding.6

Deforestation can either increase or decrease transmission. In West Africa dense rain-forest acts as a barrier to *S. damnosum* but, with the clearance of forest for cocoa or coffee plantations, breeding becomes intense, and the fly itself (as a biting nuisance) can interfere with development. By contrast, in East Africa the clearing of riverine forest can result in the complete eradication of *S. neavei* and there are several areas where the vector is retreating as a result of increasing human population pressure and the destruction of gallery forest. These changes affect the ecological conditions for both the larval and the adult flies. They may also affect the distribution of hosts other than man. Most vector species are at least partially zoophilic. Absence of the usual blood-meal source may result in excess transmission to man, such as is reported from Colombia, where *S. exiguum* bites man because of the lack of alternative hosts.

Hyperendemic onchocerciasis has serious socioeconomic consequences for family and village life. This is apparent from the age and sex distribution of the rural populations in endemic areas. At first there is selective emigration of young adult males. This is often followed by large-scale retreat of the rural population from the riverine frontiers, where there is heavy *Simulium* breeding and where fly infection rates are high, to more barren upland areas. As a result the river valleys are deserted, the upland areas become overcrowded, and the land is exhausted and eroded by overcropping and overgrazing. This maldistribution of the population has had serious effects on the economy and on development schemes that depend on the better utilization of the few available water sources in the savanna regions of Africa.

The economic development of formerly infested areas, after eradication of the *Simulium* vector, has underlined the harmful role that onchocerciasis previously played in the life of the areas. Social and economic development of endemic areas of onchocerciasis should be preceded by control of the disease. It was because of the significant direct and indirect socioeconomic consequences of onchocerciasis that the 7 countries in the Volta River basin area of West Africa, in cooperation with the international organizations and with the financial support of the international community, launched the first large-scale onchocerciasis control programme in West Africa (see section 3).

10. PARASITOLOGICAL DIAGNOSIS

10.1 The adult worm

In most helminth infections the parasite does not multiply in the body, and the severity of the disease is proportional to the adult worm load which, in turn, depends on the intensity of transmission. Onchocerciasis is unusual in that the most significant pathology is produced by the microfilarial stages of the parasite, and the severe manifestations of the disease usually take many years to develop. There is no information on the relationship between the number of microfilariae in the skin and the number of adult worms in the tissues. There is also no known relationship between the number of palpable nodules and the true worm burden in the host. Many individuals, especially those in the younger age groups, have high microfilarial densities in the skin but no palpable nodules, whereas, at the opposite extreme, considerable numbers of very large nodules can be found in elderly people who may have only a few microfilariae in the skin. In these cases the worms are often dead and calcified, and even in young people a proportion of nodules may contain calcified worms. Many fertile adult worms are impalpable and it is suspected that not all are in the superficial subcutaneous areas; some may be around the capsule of the hip joint, as has been found in the chimpanzee. Nevertheless, the mean number of palpable nodules gives a general indication of the severity of the disease, and the counting and recording of the anatomical position of nodules is of value. Especially valuable are records of head nodules, since there is a close correlation between the position of the nodules and the microfilarial densities in the skin and eye.

Post-mortem studies are needed in order to obtain more precise data on the distribution of the worms in the body in different age groups and in different geographical regions and also to determine correlations between palpable and impalpable worms and between worm loads and microfilarial densities in the skin. This is particularly important in relation to studies on transmission and the host’s immunological response.

10.2 The microfilariae

The microfilariae of *Onchocerca volvulus* are large (270-320 μm long) and unsheathed. They have a characteristic head and a pointed tail. A trained observer has no difficulty in distinguishing them from other species in stained preparations, but care is necessary in examining low-power
wet preparations because of possible confusion with the smaller skin-dwelling microfilariae of Dipetalonema streptocerca in Africa and Mansonella ozzardi in Latin America. Recent observations in Brazil suggest that M. ozzardi microfilariae occur as commonly in the skin as in the blood. M. ozzardi is widespread in Middle America, South America and the Caribbean, so that care should be taken to identify the microfilariae in skin snips in all these areas.

There is no doubt that the overwhelming majority of microfilariae seen in the eye are O. volvulus, but it is possible that other blood-borne microfilariae may enter the eye. The possible presence of microfilariae of Loa loa and other filarial worms in the eye requires further study. The adults of L. loa are usually seen only in the superficial ocular tissues, but there are two recorded cases of adult L. loa being found inside the eye.

The most common diagnostic procedure for onchocerciasis is to examine microfilariae emerging from bloodless skin snip biopsies, but a few workers prefer to examine stained preparations of dermal juices obtained by searification. This method has the advantage that a single stained preparation can be used to detect not only O. volvulus and D. streptocerca but also malaria and blood-dwelling microfilariae at the same time. It is a useful method in general parasitological surveys, but it requires skilled microscopy for differential diagnosis, and it is not suitable for accurate quantitative work because of the loss of microfilariae that may occur during staining.

Bloodless skin snips give more reliable results but there is a need for better standardization for epidemiological studies. The optimum site for the biopsy will vary depending on the geographical strain of the parasite. In Africa the preferred site is below the iliac crest, whereas in Mexico the preferred site is behind the shoulder. However, neither of these sites would be of much value in Yemen, where the highest concentration of microfilariae occurs around the ankles. The anatomical distribution of microfilariae in the skin should be determined in any area where extensive surveys are to be carried out, so as to select the best site for biopsies.

In detailed epidemiological studies extra skin snips should be taken from different parts of the body, including snips from the outer canthus, because this may be associated with eye involvement and the densities of microfilariae there may be a good indicator of future eye disease.

The skin snips can be taken with various instruments, including simple razor blades, sharp scissors, and the various types of scleral punches that produce relatively painless snips of fairly uniform size. Care should be

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*a Buck, A. A., ed., op. cit.*
taken to sterilize the instruments so as to avoid possible transmission of viral hepatitis and other diseases. A. Three types of punch have been proposed. A comparative study of these 3 punches showed that all were satisfactory for diagnosis but that the Holth model was the cheapest and sturdiest.

The snips are usually examined for living microfilariae either in water or in saline, with or without teasing, and the number of emerging microfilariae is usually counted after 10-30 minutes. The rate of emergence is slower in water than saline, but under very arid conditions, if a simple humidifier is not available, the use of water is to be preferred because it avoids the rapid concentration of saline, which interferes with the escape of the microfilariae from the skin and with the subsequent staining of the parasite, if this is required. Emergence is also slower at low temperatures. If these factors are not taken into account false circadian and seasonal rhythms may be recorded. Teasing is necessary only if the microfilariae are to be counted as early as 10 minutes after taking the snip. Otherwise it should be avoided because it can cause difficulty in counting microfilariae.

Simple qualitative information is of little value. For most epidemiological studies it is necessary to express the results quantitatively, either as the number of microfilariae per skin snip or, preferably, as the number of microfilariae per milligram or per unit surface area or volume of skin.

There are often considerable difficulties in examining and counting wet preparations in the field and it is generally admitted that these methods often fail to detect very light infection. It is also impossible to make accurate counts in high density infection. Various compromises have been adopted, including the method used in the OCP area in West Africa of examining the skin snips after 30 minutes in water, with the knowledge that at this stage only 50% of the microfilariae will have emerged.

A much more accurate method has been developed in Togo; this is an adaptation of the membrane filtration method used in filariasis surveys. The skin snips are collected in saline in transparent plastic agglutination trays having the normal 96 celled wells, used in bacteriological

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4 A suggested method is to sterilize the punch in cold 1/2% activated glutaraldehyde for 10 minutes.
5 BUCK, A. A. ed., op. cit.
laboratories. After several hours the deposits are filtered and the membrane filters are stained with Giemsa stain. It is claimed that with this method there is no difficulty in counting even high-density infections, and that in areas of low endemicity the method is almost twice as sensitive as the usual methods.

A modification of this method is at present being tested in the OCP area. The individual snips are placed in 0.3 ml of saline in the wells of the agglutination trays. The microfilariae are allowed to emerge over a 24-hour period, and they are then preserved in the wells by adding 1 drop of 10% formal saline to each specimen. The trays are then sealed with a transparent plastic sealer. The advantage of this method is that no further procedures are necessary in the field, and the preserved material can be examined at leisure or sent to a central laboratory for examination. Filtration is unnecessary. The material from each well is spread on 1 or more ordinary 7.5 cm × 2.5 cm slides (or on a single larger slide, if available), and the microfilariae are counted in the wet preparation using the same technique as that described for filariasis surveys. The snips can be weighed or measured, and the results can then be expressed as the number of microfilariae per mg or per unit surface area of skin. The method has the added advantages that fewer field personnel are required for surveys and that there is a saving on transport and on expensive equipment, such as microscopes and torsion balances.

Further work is necessary to standardize this method, and a careful comparison should be made of its sensitivity and accuracy as compared with those of the techniques at present being used in epidemiological studies, especially in the OCP area.

10.3 Research needs

(1) Further work is necessary to assess the accuracy and the sensitivity of the modified "well" method of examining skin snips for microfilariae so as to standardize this quantitative technique for use in epidemiological surveys.

(2) Thorough autopsies should be done on corpses of people who suffered from onchocerciasis to determine the numbers and distribution of worms in the body and to relate these to the concentrations of microfilariae in the skin. Data should be obtained in this way covering a number of different age groups and geographical regions.

11. IMMUNODIAGNOSIS

Immunological tests are used for both mass and individual diagnosis of parasitic diseases. However, the lack of specific and characterized antigens introduces an element of uncertainty, and onchocerciasis is no exception. Furthermore, the frequent cross-reactivity, which is pronounced in nematode infections, makes it difficult to ascertain that a case is truly positive. Since onchocerciasis can be detected by conventional parasitological techniques (skin biopsy), the role of immunological tests should be considered as complementary, for use where and when these standard techniques cannot be applied.

The immunological tests utilized for the detection of antibodies in onchocerciasis are outlined below (sections 11.1-11.7). Such tests, and particularly those that can be easily performed under field conditions as complementary epidemiological tools, undoubtedly play a useful part.

11.1 Skin tests

Immediate-type hypersensitivity in helminth infections is a common phenomenon and is frequently exploited for immunodiagnostic purposes. The skin test based on this phenomenon is relatively easy to perform, even under field conditions, and has been widely and indiscriminately used for prevalence studies of filarial infections, as well as for onchocerciasis. Because of the frequent and nonspecific false positive reactions observed by many investigators, the test fails to give satisfaction. Unless specific and well characterized antigens are utilized, it has little value for epidemiological purposes. Little information is available on delayed hypersensitivity in onchocerciasis. It would therefore be useful to repeat the reading of the test after 96 hours.

11.2 Gel diffusion test

Precipitin reactions have been used extensively for the detection of antibodies by the agar-gel diffusion technique. The test results provide a certain orientation, but from the diagnostic point of view they are of debatable value since precipitin bands cannot always be observed in proven cases of onchocerciasis.

Immunoelectrophoresis has been frequently applied to the serodiagnosis of onchocerciasis in specialized laboratories, and has been extensively evaluated. Several investigators have used it as the technique of choice and have claimed that it is able to detect specific precipitin arcs in the sera.
of onchocerciasis patients. However, the use of antigens from hetero-
ologous species—Dirofilaria immitis, Dipetalonema vite, Setaria labiato-
papillosa and others—again raises the problem of cross-reactions and the
possibility of concomitant infections.

Both agar-gel diffusion and immunoelectrophoresis represent refined
techniques which, because of the complexity of the precipitin patterns
and the difficulty of interpreting them, can be utilized only as research
tools rather than as practical serodiagnostic procedures. Furthermore, the
quantities of antigen and antibody required for a visual evaluation are
considerable. Such quantities may not always be available.

11.3 Passive haemagglutination test

Although this technique has been used to a considerable extent in
other filarial infections, data on the passive haemagglutination test in
onchocerciasis are rather limited. The test is probably one of the most
sensitive, but its value in the serology of onchocerciasis needs to be further
evaluated as purified and isolated specific antigens become available.

11.4 Complement fixation test

This test has not been used extensively in onchocerciasis and only
limited results have been reported. Some of the difficulties include the
occurrence of anticomplementary activity in sera collected in the
field. The performance of the test requires specialized personnel and good
laboratory conditions.

11.5 Immunofluorescent antibody test (IFA)

Compared to the other serological techniques mentioned, the immuno-
fluorescent antibody test has been the most widely used for both epidemi-
ological and individual diagnosis of onchocerciasis. The results reported
by many investigators are impressive but they vary according to the type
of antigen utilized. Today the technique has been considerably improved
and is based on the use of an antigen composed of frozen sections of
adult O. volvulus recovered from human nodules. Titres of 1 : 20 and
1 : 40 are considered significant and of diagnostic value. On this basis
up to some 90% of positive cases can be identified. The IFA test, as now
applied, seems to be a sensitive serological technique. The correlation
between the results obtained from the test and from skin biopsies has
been satisfactory in most cases. However, it should be noted that the
technique requires elaborate equipment and experienced personnel, as

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well as standardized fluorescein-labelled anti-immunoglobulins. Since characterized and specific antigens are not available, tests such as this, in which intact parasites or sections of them are used, are to be preferred.

11.6 Tests based on in-vitro correlates of cell-mediated immunity

To investigate cell-mediated immunity, a limited number of onchocerciasis cases have been studied by the use of the macrophage inhibition test and the direct rosette quantitation test. The results obtained by these tests were inconclusive.

11.7 Enzyme-linked immunosorbent assay (ELISA)

Efforts in certain laboratories in recent years have led to the development of new serological techniques with promising features. The enzyme-linked immunosorbent assay—commonly known as ELISA—represents a significant addition to the serology of parasitic infections. The test allows quantitative estimation of specific antibody by using alkaline phosphatase- or peroxidase-labelled anti-immunoglobulin in antigen-coated tubes or microtitre plates. Recently the assay has been applied to onchocerciasis, and preliminary results on a small sample of sera collected from an endemic area indicate that it is possible to differentiate between infected and uninfected groups of people. Surprisingly, a crude extract of *O. volvulus* used as antigen gave high values with both categories of sera. This discrepancy was attributed to contamination of the antigen with human material (extract obtained from human nodules containing adult *O. volvulus*) which reacted readily with the anti-human globulin-enzyme conjugate.

The test, with the recent modifications used in malaria serology under field conditions, could offer advantages that will become of more practical interest as better defined specific antigens become available. ELISA has proved to be as sensitive as the IFA test and easier to perform. However, it needs to be further evaluated.

11.8 The antigens

In onchocerciasis, as in many other parasitic diseases, regardless of the serological procedure applied for epidemiological or diagnostic purposes, there is always the problem of the nature and quality of the antigens. Under present circumstances, it is extremely difficult to establish acceptable diagnostic titres for any of the serological techniques available, since these titres will vary according to the type of antigenic preparation
used. The usefulness of serological techniques applied to parasitic diseases in general, and to onchocerciasis in particular, will depend very much on the area where they are applied. It can be said that the success of serology is more a matter of the antigen than of a particular procedure. Collaborative work towards the isolation and preparation of specific onchocercal antigens is in progress in several laboratories supported by WHO.\footnote{Parasite antigens. \textit{Bull. World Health Organ.}, \textbf{52}: 237–249 (1975).}

11.9 Research needs

Work on the isolation of antigens from adults, microfilariae, and infective larvae of \textit{O. volvulus} should be intensified, with a view to improving diagnostic tests.

12. THE MAZZOTTI TEST

The Mazzotti test is a useful aid to the diagnosis of \textit{O. volvulus} infection in persons in whom parasitological evidence of infection cannot be detected. The test consists of giving a small dose of diethylcarbamazine (usually 50 mg for an adult), and then observing the development of the clinical reaction in the skin, which results from the “unmasking” and destruction of hitherto cryptic microfilariae. The reaction in the skin may begin as early as 15 minutes after ingestion of the diethylcarbamazine, or it may be delayed for up to 24 hours. It is usually characterized first by itching, and later by the appearance of a fine discrete papular eruption. The distribution of the developing rash follows the distribution of the cryptic microfilariae in the skin, and in light infection it may therefore be mainly confined to a single anatomical quarter of the body.

In epidemiological work the test should only be employed on persons in whom microfilariae or nodules have not been found by standard parasitological techniques, and the dose of diethylcarbamazine given should not exceed 50 mg for an adult. The test should be read first, if possible, after about 3 hours, and again certainly at 24 hours.

12.1 False negative results

Some individuals produce no clinically detectable skin reaction to diethylcarbamazine, despite the presence of microfilariae in the skin. There may be regional differences, and false negatives have been observed
in parts of Ethiopia. Persons with gross sowda-like lesions of oncho-
cerciasis may also give a false negative reaction. Since in these cases
almost all the microfilariae reaching the skin are immediately destroyed
by an exaggerated immune response, it may be presumed that there are
few or none available to be destroyed by the provocative diethylcarbama-
zine used in the Mazzotti test. Infections with *O. volvulus* that are still in
the prepatent phase will also give a false negative reaction to the Mazzotti
test, although the reaction may be expected to become positive as soon
as microfilariae reach the skin.

12.2 False positive results

In Africa, patients harbouring microfilariae of *Dipetalonema streptocerca* in their skins may react to doses of diethylcarbamazine higher than
50 mg. The possibility that microfilariae of *M. ozzardi* may give false
positive Mazzotti reactions requires investigation. Persons infected with
*Loa loa, Wuchereria bancrofti, Brugia malayi*, and *Dipetalonema perstans*
may suffer slight febrile or lymphangitic reactions following provocative
diethylcarbamazine, but apart from the possibility of mild urticaria, false
skin reactions are most unlikely to occur with these parasites. False
positive reactions of “generalized itching” may be reported by hypo-
condriacal persons, who are frightened that they may have “the filaria”,
and who have some limited knowledge of the immediate effects of
diethylcarbamazine in persons with onchocerciasis. The demeanour of
the patient and the absence of scratch marks will put the observer on his
guard.

12.3 Indications

The main uses and indications for the Mazzotti test are as follows.

(a) To confirm the diagnosis of onchocerciasis in persons in whom the
disease is suspected on clinical grounds but in whom the parasite cannot
be found.

(b) To detect lightly-infected cases among persons (especially young
children) recorded as parasitologically negative in epidemiological sur-
veys. The finding of negative Mazzotti reactions in members of the
younger age groups who are parasitologically negative may provide
valuable additional evidence to confirm the absence of new infections in
areas where transmission is thought to have been interrupted.

(c) To assess the effect of treatment. A negative Mazzotti test is a
useful indicator that all microfilariae have been eliminated from the skin.
In most cases, 2 consecutive negative tests separated by an interval of 1 month may be taken as evidence that the adult female worms are dead or sterilized.

12.4 Contraindications

The Mazzotti test should not be used indiscriminately, especially in persons with heavy *O. volvulus* infections. Severe reactions may require treatment with antiserotonins and/or steroids. The test should not be used in patients with heavy *L. loa* microfilaraemia. Clinical judgement must be exercised with very old or debilitated persons, and with pregnant women, before the test is made, even if such persons are parasitologically negative.

In epidemiological surveys, one should bear in mind the possible repercussions of producing positive Mazzotti reactions in persons who previously considered themselves to be in good health.

12.5 Research needs

Further information is required on three aspects of the Mazzotti test.

1. It is not known whether the sensitivity of the test can usefully be improved by searching for microfilariae in the urine or in the blood after provocative diethylcarbamazine.

2. The possibility that over the first few hours after a subject has taken provocative diethylcarbamazine scanty skin microfilariae may be mobilized sufficiently to make them detectable in skin snips requires investigation.

3. The possibility of using diethylcarbamazine applied locally to the skin as an intradermal test, or as a “Mazzotti patch test”, should be studied.

**Epidemiological features of onchocerciasis**

13. Epidemiological methods

There are two kinds of measurement of the occurrence of a disease in a community: incidence and prevalence. Because of the nature of onchocerciasis, its extreme chronicity and slow evolution, the kinds of tools required to make a diagnosis, the inaccessibility of many foci of the disease, and the limited availability of expert medical services, reliance
has, up to the present, been placed almost entirely on the measurement of prevalence. This is the determination of the number of cases or infections existing at a given time, in relation to the size of the population at risk. It is a static index. Incidence, which identifies the number of new or newly discovered cases arising over a standard period of time (e.g., 1 year), again in relation to the number of persons in the population, is a dynamic index. While the direct measurement of incidence in a disease such as onchocerciasis is very difficult, it would be preferable to prevalence in one particular respect. Prevalence at a given time is determined not only by past incidence, but by differential disappearance from the population of different classes of persons, as well as by spontaneous regression or progression of disease. Thus, for example, if the mortality rate of blinded persons is greater than that of others, a prevalence survey will reveal too few blind persons in relation to those having only cutaneous manifestations or only positive skin snips. In other words, it will underestimate the importance of blindness. Excess mortality of the blind as compared with others might help to explain the diminished prevalence of blindness at advanced ages, noted in some surveys; and differential mortality might also be one explanation of the decline in average microfilarial density in the skin that has sometimes been noted in persons past the age of 50 years.

Understandingly, many previous prevalence surveys have been rather incomplete in the enumeration of the local populations, a necessary step in the estimation of prevalence rates. Furthermore, the difficulty of ascertaining the ages of adult persons in many communities where onchocerciasis is endemic makes for uncertainty as to age-specific prevalence rates. It is thus considered that in future prevalence surveys, more attention should be given to the denominators generally, e.g. the completeness of enumeration of the population, the completeness of coverage by the survey examinations, the reasons for omission of examination of those who are missed, and the classification of the subjects (both cases and inhabitants) as to relevant characteristics such as duration of residence in the village, periods of absence, occupation, and place of work if away from the village.

In the classification of villages according to degree of endemicity, it is desirable to record the length of residence of members of the community. One should then record the proportions positive among permanent residents according to age groups, as well as the intensity of infection as measured by numbers of microfilariae per specimen. It is then often desirable to have an index of the occurrence of infection expressed in a single figure, by which to compare the community with other
communities. No index can be satisfactory unless it takes into account the period of time at risk of infection, as reflected in the ages of lifelong residents. There are several possible indices. One is the age-standardized frequency of positives, the age distribution taken as a standard being the same for all communities under consideration. Another is the annual probability of infection, which may be estimated roughly from the age-curve of prevalence. A third method is to determine the so-called AI_{50}, or age at which 50% of persons have become infected. The proportion found positive in a specified young age group (e.g. children aged 5-9 years) is an even simpler index and would also be more discriminating.

The Expert Committee thinks that a method that takes account, not only of years at risk, but also of the intensity of infection as gauged by the number of microfilariae per skin snip, would offer further advantages, and recommends that such a method be developed. Advantage can be taken of the experience now being gained in OCP to evaluate different indices.

13.1 Methods in relation to objectives

Any discussion of epidemiological methods must be prefaced by a statement of the purpose of the study. There are several broad general purposes that may be served by an epidemiological approach. Although a particular study may on occasion serve more than one purpose, it should not be relied upon to do so.

One major purpose of epidemiological study may be termed the programme purpose. Planners need to know the frequency of a disease, its severity, chronicity, and its social and economic cost to the individual and to the community in such terms as lost manpower, crippling, or reproductive wastage. This information is needed so that a rational decision can be made as to the amount of effort, resources and funds to devote to further study and activities to prevent the disease, relative to other problems.

The programme goal requires a knowledge not only of the frequency of the disease but of its natural history, clinical features and other factors, including some that appear unrelated to medicine. As pointed out in an OCP report, the avoidance of areas of highest prevalence has had adverse economic consequences for entire countries, since it has meant the abandonment of the best agricultural land. This is a striking example of the fact that analysis directed toward the programme goal cannot be

* Mission for Preparatory Assistance, op. cit.
limited to a mere counting of cases; a broader approach must be taken. The issue includes, but is not limited to, the securing and use of epidemiological data.

A second purpose, related to the programme purpose, is surveillance. This aims to keep track of the occurrence of a disease, with special attention to its distribution in time and place, in order to know where it is most prevalent, and whether it is increasing or diminishing.

In onchocerciasis, prevalence surveys by medical teams have been the means of getting data for both these purposes. Estimates of total infection rates, with the aid of skin snips, and estimates of blindness have been the two indices most often used. This will continue to be true, at least until practical and specific immunological methods are devised.

There are two considerations in assessing a screening procedure. One is its accuracy in classifying persons. Accuracy, in turn, may be said to have two aspects. One is sensitivity, which is the capacity of the test to identify as positive those who have the condition. The other is specificity, which is the capacity to identify as negative those who do not have the condition.

The second consideration is the consistency with which the test findings can be reproduced, either by the same workers or by a different team. The purpose of prevalence surveys is either to compare villages, districts or subgroups of persons, such as age classes or occupational groups, or else to compare prevalence at two or more times. This being so, it is obvious that the consistency with which test findings are measured must be of paramount concern in epidemiological studies. Equally, of course, gross inaccuracy cannot be countenanced.

A further consideration is the cost in time, effort and money of the procedures used, in view of the great effort expended in examining thousands of people at hundreds of remote locations. What is needed is indices, fairly simple to obtain, not too costly, and comparatively error-free, by means of which the disease burden may be reliably compared between villages and districts. They should be such that different observers can approximately reproduce one another’s findings, and the same observer can on repeated tries approximately replicate his own. Observations from the United Republic of Cameroon indicate that conventional current methods fall short of meeting this goal, even when used by skilled and experienced workers. Upon re-examination of villagers a year after the initial examination, they found sizable differences in the ocular, skin and subcutaneous nodule abnormalities. They

suggested that agreed-upon definitions should be published for the various skin manifestations of onchocerciasis. Skilled ophthalmologists are not always available to participate in surveys, and standardized methods for scoring ocular and visual defects need to be used. Further work is needed on these problems. However, replacement of older methods by new ones should be avoided within one study, unless the new methods are markedly superior, since this causes discontinuity and data are no longer comparable.

A third purpose of epidemiological investigation, related to the first two, is to establish the prevalence or incidence of a condition so as to determine the effects of a disease prevention or eradication programme. For this purpose a series of measurements must be made before, during, and after the operation of the programme. If these steps are not taken, the programme is in danger of being a total failure from the point of view of assessment of the value of the procedures used, so that other areas or future programmes have to start from zero, with no knowledge of what will work. The considerations concerning repeatability of measurements, discussed earlier in this section, are of equal or greater importance here.

It should be noted that epidemiological information of much importance has frequently been discovered as a valuable by-product of field trials of preventive measures against diseases.

The etiological agent of onchocerciasis, O. volvulus, is known and the vectors are likewise known. Nevertheless, there is need to learn other determinants of the disease. Here it may be helpful to use methods developed for the investigation of chronic noninfectious diseases. They permit the study of the distribution of disease in the population in relation to risk factors, and thus the estimation of relative risks.

The methods for doing this are classified as the case-control or retrospective method, and the population-based or prospective method. They could usefully be applied in studies of onchocerciasis, if there are questions they could appropriately explore. The case-control method is the more practical approach. Preferably in a community with a moderate, rather than high, prevalence of the disease, the infected persons would be identified and compared with an approximately age- and sex-matched sample of uninfected controls, with regard to selected attributes such as place of residence (proximity to a stream), duration of residence, and occupation. Persons with ocular disease, or those excreting microfilariae in the urine, could also be compared with other infected persons not having these characteristics. The association between the presence of these complications and the intensity of infection as gauged by skin snips, or by number and location of subcutaneous nodules, could be examined.
Such studies (several of which have been done) can lead to other, more original or more promising hypotheses, which can be tested in a similar fashion. In order to interpret the associations found, it is necessary to take account of so-called "confounding variables," which may be responsible for spurious associations. There are two ways of doing this. One is by matching cases with controls on such variables. The other is by statistical adjustment in the analysis phase of the study.

To give an example of a problem that could be attacked by a combination of these epidemiological methods, a question has arisen whether infants born to women who become pregnant while harbouring a heavy burden of *O. volvulus* microfilariae might exhibit an immune tolerance or some other altered host response to the parasite. It should be possible to examine a series of such infants, together with an equal number of control babies, by appropriate parasitological and immunological methods, and to repeat the examinations on both groups after an appropriate period of time. This would combine elements of the case-control and the longitudinal approaches to epidemiology.

Since onchocerciasis is a highly focal disease, methods of mapping prevalence data with the aid of a computer, as developed and used in studying the spatial distribution of schistosomiasis in South America, would be useful. These methods could provide visual evidence of the relationship of prevalence to various factors, such as altitude, temperature and rainfall. Clearly, migration from heavily infected villages and into villages where transmission does not occur can confuse the picture.

A natural sequence of epidemiological steps in the investigation of a disease begins with the selection of a community for study on the basis of the following considerations among others: suitable size; intermediate or widely varying levels of prevalence of the disease; stability of population; and possession of an appropriate combination of the factors it is desired to study, such as ethnic or occupational factors. Next, a special census of the chosen community is made. In addition to the usual demographic data, information is secured from each person on other characteristics of epidemiological interest, such as, for example, his residence history. Then a prevalence survey is carried out, and prevalence rates are established in relation to the factors identified in the special census. These two steps (census and survey) may sometimes be combined, provided that the investigator is able to determine the completeness of the survey.

The next step is to make a case-control study. This step includes the exploration in detail of the relationship of the disease to particular

attributes that cannot be studied in the whole population. For example, difficult and costly biochemical or immunological tests can be carried out on the cases (or a random sample of the cases) and on an equal number of controls, when these tests could not be done on the whole population.

Finally, repeat visits should be made so that a longitudinal or prospective element can be added to the study. Only in this way is it possible, by determining the number of infected persons who were uninfected at the previous visit, to secure a direct estimate of the current incidence of infection. Likewise, the incidence of new ocular and skin lesions may be calculated, and rates of decline of incidence in areas where control projects are in operation may be determined. Hypotheses that have arisen at earlier stages of the investigation may be explored in further detail, as in the studies of Buck and colleagues in Chad, where they were able to amplify their observations on microfilaruria on a second visit. More broadly, knowledge of the natural history and course of the disease and its manifestations may be enriched. It was in this way that Budden obtained a better understanding of the eye lesions.

Such investigations as have been sketched here may also shed light on problems that are not strictly medical. For instance, the effect of impaired vision upon the individual's ability to sustain himself can be estimated.

13.2 Research needs

(1) In conducting surveys for prevalence of *O. volvulus* infection and disease, more attention should be given to the characteristics of the populations of villages to be surveyed (i.e., the "denominator"), the completeness of coverage, and the estimation of prevalence rates specific for the enumerated characteristics.

(2) The case-control method should be utilized in the study of risk factors for onchocerciasis.

(3) Repeat visits should be made in years following initial prevalence surveys, in order to:

(a) ascertain incidence rates of new infections and new clinical manifestations;

(b) improve knowledge of the natural history of onchocerciasis generally; and

(c) test the validity of suggested relationships or hypotheses.

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(4) Mapping of the prevalence of onchocerciasis according to geographical region should be continued. It should be extended in those areas where present information is inadequate, and where the presence of the disease is suspected but unconfirmed.

(5) Methods used in measuring prevalence should be standardized to achieve maximum comparability between investigations.

14. PATTERNS OF ENDEMICITY

Many data concerning the epidemiology of onchocerciasis have been produced during the last 10 years in West Africa. These data are now of such quality that the main features of the prevalence of onchocerciasis, both in savanna and in forest, can be established.

Very much less is known about the infection in other parts of Africa, and much of the information on onchocerciasis in Central and South America is outdated or incomplete.

14.1 Epidemiological methods and criteria

The general methods of examination and the main criteria used in prevalence surveys to assess the epidemiological features of onchocerciasis in large areas are presented in Annex 1 and will not be discussed here in detail.

14.2 Prevalence and microfilarial density in the skin

Techniques of quantitative skin biopsy now permit an evaluation of the importance of onchocercal infections in the individual and in the community. In addition to an estimate of the prevalence of infection a quantitative evaluation provides information on the intensity of infection and on the pathogenicity of onchocerciasis. The average microfilarial density can be determined as the arithmetic mean of the microfilarial densities or, preferably, as the geometric mean of these values. The histogram in Fig. 5 shows the increase in microfilarial densities by age and sex. As a rule, the intensity of infection is higher in men than in women.

The measurement of the microfilarial density permits the study of correlations between different variables, as for example between the prevalence of microfilariae in the anterior chamber of the eye and the
microfilarial density in the skin. Table 1 shows the frequency of carriers of microfilariae in the anterior chamber of the eye as a function of the microfilarial density in the skin. The frequency of microfilariae of *O. volvulus* in the urine is also closely associated with the microfilarial density in the skin.

14.3 Prevalence and topographical features

(a) Distribution in foci. Endemic onchocerciasis occurs in foci, around *Simulium* breeding sites or a line of breeding sites. The distribution of the foci is determined by the hydrogeographic characteristics of the region.
TABLE 1. CARRIERS OF MICROFILARIAE IN THE ANTERIOR CHAMBER (MFAC) BY AGE AND MICROFILARIA (mf) LOAD  

<table>
<thead>
<tr>
<th>Age</th>
<th>1-10 mf per snip</th>
<th>11-50 mf per snip</th>
<th>51-100 mf per snip</th>
<th>&gt; 100 mf per snip</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>10-19</td>
<td>1/26 = 3.8%</td>
<td>10/31 = 32%</td>
<td>8/13 = 61%</td>
<td>8/12 = 66%</td>
<td>27/82 = 32%</td>
</tr>
<tr>
<td>20-29</td>
<td>1/34 = 2.9%</td>
<td>21/56 = 37%</td>
<td>9/13 = 69%</td>
<td>12/14 = 85%</td>
<td>46/177 = 26%</td>
</tr>
<tr>
<td>30-39</td>
<td>1/30 = 3.3%</td>
<td>10/25 = 40%</td>
<td>11/18 = 61%</td>
<td>7/11 = 63%</td>
<td>29/84 = 34%</td>
</tr>
<tr>
<td>40-49</td>
<td>2/24 = 8.3%</td>
<td>14/31 = 45%</td>
<td>4/11 = 36%</td>
<td>5/8 = 62%</td>
<td>25/74 = 33%</td>
</tr>
<tr>
<td>50-59</td>
<td>1/11 = 9%</td>
<td>7/21 = 33%</td>
<td>6/8 = 75%</td>
<td>6/9 = 66%</td>
<td>20/49 = 40%</td>
</tr>
<tr>
<td>60 and over</td>
<td>1/7 = 14.3%</td>
<td>3/10 = 30%</td>
<td>2/4 = 50%</td>
<td>2/3 = 66%</td>
<td>8/24 = 33%</td>
</tr>
<tr>
<td>Total</td>
<td>7/132 = 5.3%</td>
<td>65/174 = 37%</td>
<td>40/87 = 46%</td>
<td>60/157 = 38%</td>
<td>152/430 = 35%</td>
</tr>
</tbody>
</table>

* Data of Picq & Claveau from a study of 430 case histories (private communication).
(b) The mosaic of endemic areas. The extent and gravity of the foci are quite variable. No more than a hundred or several thousand persons may be involved, and the foci may be separated, contiguous, or overlapping, according to the hydrogeographic features of the region. There exists a mosaic of foci, with much diversity.

(c) Stratification of endemicity levels. Within the same focus, several degrees of endemicity may exist. This stratification depends upon distance from the breeding sites, which in turn determines the frequency of man-fly contact.

If the breeding sites are productive, the zone bordering on the banks of the river may be hyperendemic; the area of low endemicity is on the periphery of the focus; and the mesoendemic area lies in between. The depth of the hyperendemic zone may vary from 1 to 10 kilometres, depending on the extent of the breeding site. In some places there may be no hyperendemic zone. Therefore, some authors designate villages as "first-line", "second-line" or "third-line" according to their position in relation to breeding sites, without implying a particular endemic level.

14.4 Prevalence and cumulative aspect of onchocercal infection

In terms of epidemiological, clinical and pathogenic considerations the cumulative aspect of the infection is a major characteristic of onchocerciasis, the worm load increasing progressively in the individual with the passage of years. This cumulative aspect can be shown indirectly by the study of populations and by the analysis of the age-specific prevalence of specific disease manifestations (Fig. 6).

In general, the prevalence and severity of onchocerciasis increase with progressing age up to about 50 years. Thereafter, both the prevalence of infection and the microfilarial density show a slight decline. This general pattern is most evident in areas where infection is of high or medium intensity but diminishes in communities with low intensity (Fig. 7).

14.5 Prevalence and differences between the sexes

Differences in the prevalence of *O. volvulus* infection between the sexes have been noted by numerous workers. They are more pronounced in hypoendemic than in hyperendemic areas. However, microfilarial concentrations are higher in males than in females even in hyperendemic areas.

These differences are reflected in the frequency of severe eye lesions and blindness, men being more frequently affected than women in both hyperendemic and mesoendemic zones.

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FIG. 6. AGE-SPECIFIC PREVALENCE OF INFECTION WITH O. VOLVULUS IN FIRST-, SECOND-, AND THIRD-LINE VILLAGES

Source: Preparatory Assistance Mission, 1973 (unpublished WHO document OCP/33.1), eq. es. Overall blindness rates were: first-line village, 15% (114/759); second-line village, 9.9% (33/340); third-line village, 0% (0/156).
The differences between the sexes may not appear until after a certain age. Difference in type of occupation is one reason advanced to explain the difference in infection between males and females, but this does not exclude the existence of other possible factors.

**Fig. 7. Histograms comparing four Cameroon Sudan-Savanna villages, by age-groups, with regard to:** (A) arithmetic mean number of nodules per person, (B) prevalence of a positive buttock skin snip, and (C) arithmetic mean number of microfilariae per milligram of skin per person at the buttock.


### 14.6 Prevalence and blindness

In hypoendemic communities serious visual impairment is uncommon and largely due to posterior segment lesions. In mesoendemic communities blindness is more common. Here anterior uveitis and posterior segment lesions tend to be equally important causes. In hyperendemic communities blindness is a serious social problem (Fig. 8 and 9) and
depopulation may result. In these areas sclerosing keratitis is superimposed on the other lesions as an important cause of visual loss. A clear correlation exists in the West African savanna and in the forest zone between the prevalence of onchocerciasis and the rates of
blindness in the infected villages. It should be noted, however, that in the forest the prevalence of blindness is very much lower, and severe eye lesions occur significantly less frequently than in the savanna (Fig. 10).

14.7 Prevalence of blindness and size of villages

In the rural savanna zones of West Africa the size of villages correlates negatively with the prevalence of blindness. This correlation is illustrated by Fig. 11. Hyperendemic villages frequently have fewer than 300 inhabitants, and usually fewer than 200. The number of inhabitants of many severely infected places may even be considerably lower.

14.8 Prevalence and microfilaria

Microfilaria appears to be considerably more frequent than was previously recognized. It has been found in Central America as well as
in Africa, and in the savanna as well as the forest. The prevalence of infected persons found to be spontaneously excreting microfilariae in the urine varies from region to region and with the techniques of examination employed. The percentage of those excreting microfilariae seems to increase with the level of endemicity.
14.9 Prevalence and skin manifestations

The cutaneous manifestations of onchocerciasis are manifold. They include pruritus, lesions due to scratching, *gale filarienne*, lichenification, cutaneous atrophy, depigmentation, xeroderma, pachyderma and lizard skin. Depigmentation is a comparatively frequent sign in more heavily
infected communities, where it may be encountered in 15-30% of patients. In the savanna zone it is frequently found in blind people and those with severe ocular lesions.

In West Africa depigmentation is usually found more frequently in the forest zone than in the savanna. More detailed information about the differential diagnostic criteria of these lesions is needed.

14.10 Prevalence and bioclimatic zones

There are clear epidemiological differences in West Africa between the patterns of onchocerciasis in the savanna and in the forest. This difference appears particularly clearly in hyperendemic communities. It does not show up in the microfilarial prevalence rates, nor in the prevalence of persons with nodules or with punctate keratitis, but is very clearly apparent in the prevalence of severe ocular lesions and blindness, the rates of which are very much higher in savanna than in forest communities.

15. DYNAMICS OF TRANSMISSION

15.1 Vectorial capacity

The concept of vectorial capacity, developed for malaria, is, in principle, applicable to all vector-borne infections. It is a contact rate, defined by the product of the following factors: the flies' biting density on man; the proportion of flies surviving for a period at least equal to the incubation period of the parasite in the vector; their remaining expectation of life; and the frequency at which the individual vector feeds on man. It is distinct from the physiological capacity to transmit.

The application of this concept to onchocerciasis calls for adaptation. The definition supposes that the infection does not affect the mortality of the vector, and that contact between the vector and human populations is random. Neither is true in the case of onchocerciasis, and this is likely to have a significant effect on transmission.

Most of the factors involved in vectorial capacity in onchocerciasis can be, and have been, estimated. The biting density on man is the common measure of vector density (see section 6.6). It varies by hour and season, and a yearly total is usually estimated from a sample of whole days distributed over the year. It is usually measured at a fixed station, or at a small number of such stations, and the estimate obtained is probably a maximum. The relationship of this maximum to the
average, and the distribution of individuals around the average, may vary from place to place. The infective biting potential is an estimate (expressed as a number per 1000) of the proportion of vectors surviving for a period equivalent to the incubation period. This proportion has also been calculated from the expectation of life and the incubation period. The incubation period varies with temperature. The expectation of life has been estimated from the proportion of parous flies. It could also be estimated from a more detailed age distribution of the biting population, when better techniques become available for subdividing the age groups (see section 6.6).

The differential dispersal of _Simulium_ by age introduces a bias, varying from place to place, in the estimation both of the proportion surviving the extrinsic cycle and of the expectation of life. The frequency at which the individual vector feeds on man is the product of the frequency of feeding multiplied by the proportion of meals taken on man. The frequency of feeding has been estimated from the age of larvae of _O. volvulus_ found in man-biting _Simulium_ vectors. The proportion of meals taken on man cannot be estimated directly. In certain places, the vector is relatively zoophilic, as is shown by direct observation, the results of precipitin tests, or the finding of infective larvae of non-human filariae. In other places, the _Simulium_ vector is presumably predominantly anthropophilic.

As already noted (section 6), a vector population may be composed of several species, reliably identified only by the larval polytene chromosomes. The geographic distribution of the members of such a species complex, and their relative importance as vectors, are not yet completely known.

It is desirable to study transmission in a relatively closed system, where a parasite population is maintained in defined human and vector populations that have limited exchange with other human and vector populations. In onchocerciasis, it may be particularly difficult to identify such transmission units of manageable size.

The vectorial capacity is, at best, estimated over one or a very small number of years. The corresponding parasitological situation is the cumulative effect of vectorial capacity over a lifetime. In relating the two this limitation has to be kept in mind.

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* Le Berre, R., op. cit.

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15.2 The parasite in man

The probability of successful development and mating of the infective larvae of *O. volvulus* inoculated into man is unknown. The mating behaviour of *O. flexuosa* in deer has been deduced from the age and number of male and female worms found in subcutaneous nodules at all stages of development. This species is polygamous and there is probably an efficient chemotropism.\(^a\)

The prepatent period of *O. volvulus* in man has been estimated from the youngest age at which children are found positive, but this may need qualifying in view of the possibility of transplacental transmission. Records of people who have visited endemic areas for short periods of time may provide more reliable data. Also, records of infection in new immigrants from non-endemic areas could be useful. The longevity of microfilariae in the skin has been estimated from their rate of disappearance in patients treated with macrofilaricides or from their rate of reappearance in patients treated with a microfilaricide. The longevity of the (fertile) adult female has been estimated from the rate of disappearance of microfilariae after interruption of transmission by eradication of the vector in Kenya. It could also be estimated from the rate of approach to the plateau of microfilarial densities that is reached in old age groups. The number of microfilariae reaching the skin from 1 fertile female is unknown.

The distribution of microfilariae over the body varies with the geographical area, and is usually positively correlated with the distribution of bites by the local vector. The microfilarial density in the skin shows a circadian fluctuation, of low amplitude, positively correlated with the biting cycle of the vector.\(^b\)

The existence of a density-dependent regulation of the parasite population in man is suggested by epidemiological findings. Where transmission is intense, the microfilarial density reaches a plateau in adults. In a study of 4 villages in the West African forest, an increase in transmission above a certain level did not increase the level of this plateau, but only the rate at which it was reached. Between villages, the microfilarial reservoir in man was found to be largely independent of the number of infective bites, beyond a certain level.\(^c\) In villages of the West African Sudansavanna zone the plateau was probably at a higher level, and the level was

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apparently increasing throughout the range of transmission observed, although this range was smaller than the range observed in the forest. Here also, between villages, the microfilarial reservoir in man varied much less than the number of infective bites or infective larvae offered. These findings suggest that the density-dependent regulation in man may be more efficient in the forest than in the savanna.

15.3 Transfer from man to the vector

The vector usually concentrates the microfilariae of the same parasite-vector complex. This phenomenon varies in intensity between complexes, and it is not known whether it is density-dependent. The number of microfilariae ingested by the vector increases with the density of microfilariae in the skin, up to a maximum after which it may decrease, probably because of the skin changes resulting from the presence of the microfilariae themselves. There seems thus to be a density-dependent regulation both of the number of microfilariae in the human host (see section 15.2) and of their accessibility.

The presence of the skin lesions of onchocerciasis has been found to prolong the feeding time, and the number of microfilariae ingested increases with the feeding time. However, this is not sufficient to compensate for the apparent decrease in accessibility.

The proportion of vectors infected per blood-meal has been estimated from the proportion of biting parous flies (i.e., those having taken at least 1 previous blood-meal) with developing infections. This is justified by the clear-cut bimodal age distribution (i.e., half-developed larvae and infective larvae) of the parasites in the biting fly population. In the comparative studies of forest and savanna villages mentioned in section 15.2, the proportion increases with the intensity of infection in the human population in the forest, but not in the savanna. It depends on the intake and survival of microfilariae, and on the survival of the vector (see section 15.4).

15.4 The parasite in the vector

Only a proportion of the microfilariae ingested are successful in penetrating the haemocoel and starting their development. This proportion has been estimated directly as 15–44% in the West African forest (S. damnosum s.l.), but only 0.3–7% in the West African Sudan-savanna zone (S. damnosum s.l.) and Guatemala (S. ochraceum). The proportion

decreases as the number of microfilariae ingested increases, at least in the West African Sudan-savanna. It is likely that the density-dependent regulation in the vector is stronger in the West African savanna than in the forest.

There is apparently no significant mortality of the larvae while developing in the vector. However, the ingestion of a large number of microfilariae increases the mortality of the vector.

15.5 Transfer from the vector to man

Given the very uneven spatial distribution of the infective biting density, and the variable mobility of man (by age, sex and occupation), exposure must vary greatly from person to person. It is possible that a relatively simple index of exposure, based on history and/or observation, could be developed. How many of the epidemiological findings can be explained by differential exposure is not known. For example, the usually lower prevalence and density of the infection in females is probably due to a lower exposure, but intrinsic factors cannot be excluded.

From a comparison (in the Cameroon forest) of flies caught immediately before feeding with flies caught immediately after completing their blood-meal, it has been estimated that during the meal 80% of infective larvae escape and that 40% of the infective flies become non-infective. The proportion of escaping larvae actually penetrating man is unknown. The possibility of vector mortality associated with the exit of the infective larvae has been considered, by analogy with *Chrysops silacea* and *Loa loa*, but no definite conclusion has been reached.

15.6 Mathematical transmission models

A dynamic model is being developed by WHO which takes into account some of the factors referred to above, e.g., the differential exposure of the human population to *Simulium* bites, the maturation period of the parasite in man, the chance of a female worm being fertilized, the density-dependent production of microfilariae, the density-dependent development of infective larvae in the vector, and the differential dispersal of nulliparous and parous flies. Although most of the necessary parameter estimates are not yet available, qualitative conclusions may be drawn. For instance, it is possible to analyse the relationship between the minimum vectorial capacity necessary for the maintenance

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of a stable endemic level and the strength of density-dependent factors. As far as possible, the model should be tested with data from the literature.

15.7 Research needs

(1) The planning and evaluation of control programmes would benefit from a better understanding of the dynamics of transmission. Efforts to construct and test a mathematical model, using available information, should therefore be pursued.

(2) A multidisciplinary team should be instructed to draft field study designs regarding the dynamics of transmission. The team should identify specific gaps in knowledge, the filling of which would have practical consequences for the control of onchocerciasis, and its proposals should include an estimate of the cost at which this can be done with a reasonable chance of success.

16. ANIMAL MODELS

The only reliable record of a natural infection in an animal host other than man is from Zaire, where *O. volvulus* was recorded from the gorilla. Several attempts have been made to transmit the parasite to laboratory animals, including the gerbil, which is susceptible to a wide range of other filarial parasites. However, so far the only animal that has produced patent infection is the chimpanzee. This animal has been used extensively to study strain differences of the parasite (see section 7), and for chemotherapy studies. However, nothing is known about the role of the chimpanzee as a reservoir host in nature, so that this host system is of little value for epidemiological studies. Even if a more convenient animal proved susceptible to *O. volvulus* there would still be difficulties in maintaining the parasite by cyclical transmission in the laboratory because of the failure to establish colonies of vector simulids.

An alternative approach is to use other species of *Onchocerca*, which occur in domestic and wild animals in many parts of the world. These include *O. gutturosa*, *O. gibsoni*, *O. armillata*, *O. ochengi*, *O. dukes*, and *O. dermatophi* in cattle, *O. cervicalis* in the horse, *O. sweat* in water buffalo, *O. flexuosa*, *O. tenuigen* and *O. taenia* in deer in Europe, and *O. cervipedia* in North American deer. It is perhaps signifi-

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8 Subcutaneous nodules similar to those in *O. volvulus* infections are found in these species.

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cant that most of these species have been found in animals of economic importance. It is likely that further studies will reveal other species in wild animals, especially in the tropics. If they are transmitted by the same vectors as those transmitting *O. volvulus* to man this could result in considerable confusion in the interpretation of transmission indices (see sections 5 and 6).

The observation in England of the transmission of *O. gutturosa* by *Simulium ornatum* and the observation in the Federal Republic of Germany of the transmission of *O. tarsoeula* by *Prosimulium nigripeps* and *S. ornatum* suggest that these host-parasite systems could be exploited to obtain a deeper understanding of many aspects of the natural history of the parasites and their vectors. Such work would include studies on transmission, on the development of the parasite in the host, on the pathogenesis and immunity of the infections, on the development of the parasite in the vector and, in the case of the parasites transmitted by simulids, on the bionomics of the vectors, their laboratory maintenance, and the effect of vector control on their transmission. *O. cervicalis* and its *Culicoides* vectors are probably cosmopolitan in their distribution and provide a useful model, with the added advantages that the vectors are easily reared in the laboratory and the associated disease in the horse includes ocular lesions closely resembling those seen in man.

Most of the *Onchocerca* of domestic animals escape detection in routine examination. They are very rarely recorded by meat inspectors or veterinarians, even in countries where there is a very high prevalence in livestock. Some species may be of economic importance, but nothing is known about the transmission of many of them, including *O. armillata*, which causes extensive damage to the aorta of cattle in Africa and Asia.

There is an urgent need for more effective and less toxic drugs for the treatment and control of onchocerciasis. Filaricidal drugs that are discovered using the rodent filarial systems may not be effective against *Onchocerca*. The species in domestic animals could provide systems for secondary screening, and in areas where detailed studies have been carried out on the transmission of these parasites it might be possible to use the domestic animal systems to determine the value of chemotherapy as a control measure.

### 16.1 Research needs

Further studies are necessary on *Onchocerca* species in domestic animals in order to:

(a) understand better the development of the parasites in their definitive hosts and vectors;

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(b) study the transmission of these infections under natural conditions;

c) provide systems for the evaluation of chemotherapeutic agents.

RECOMMENDATIONS

1. Better systems should be established for gathering essential information on the prevalence and incidence of onchocerciasis throughout the world, since this disease has been assigned a high priority by the World Health Assembly and there is evidence that better control methods are becoming available. Particular attention should be paid to collecting basic entomological and epidemiological data in areas where control schemes are likely to be implemented.

2. In long-term epidemiological surveys, involving repeated ophthalmological examinations of the same populations for *O. volvulus* infection, such as in the Onchocerciasis Control Programme in the Volta River Basin Area (OCP), mobile treatment teams should be set up to deal with ocular onchocerciasis and other eye diseases that can be treated under field conditions (e.g., acute and chronic eye infections, xerophthalmia, trachoma, senile cataract, and trichiasis). If this is not done it may prove impossible to secure the continued cooperation of the populations involved in repeated epidemiological assessments. It is essential that the clinical work should not interfere with the work of the epidemiological teams. WHO should encourage governments to seek the cooperation of the International Agency in the Prevention of Blindness or other similar organizations in the formation of such teams.

3. In any areas of Africa where control of onchocerciasis transmission has been achieved, and where repopulation of riverine valleys is likely to ensue, the governments should be advised to take adequate precautions—by means of surveillance, treatment and control—to prevent outbreaks of other diseases, such as human trypanosomiasis and schistosomiasis, that may occur in the repopulated and development areas.

4. A WHO reference centre for the study of histopathological material from onchocerciasis cases should be established.

5. Centres should be established in all areas where major onchocerciasis control programmes are in operation for the following purposes:

(a) to undertake detailed clinicopathological studies on patients with onchocerciasis so as to gain a better understanding of the natural history of the disease and to evaluate diagnostic procedures;
(b) to carry out controlled trials of drugs for the treatment of onchocerciasis;

(c) to undertake research on the treatment of patients at risk of developing eye lesions and blindness.

6. In view of the importance of onchocerciasis and other blinding eye diseases in tropical Africa it is recommended that special attention should be given to tropical ophthalmic diseases in the undergraduate training of doctors and medical auxiliaries in this region. It is further recommended that WHO should organize postgraduate training courses in tropical public health ophthalmology for doctors working in areas where onchocerciasis is prevalent, especially where control programmes are in operation.

7. The need to establish laboratory colonies of African and Latin American *Simulium* vectors is stressed. The establishment of such colonies would greatly help to clarify the taxonomic status, bionomics, and vectorial characteristics of the various species, and would also allow more detailed studies of *Onchocerca* spp. The recommendations made at its meeting in October 1975 by the OCP Scientific Advisory Panel working group on the rearing of *Simulium* species and establishment of a laboratory colony of *S. damnosum*\(^a\) are endorsed by this Committee.

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Annex 1

TECHNIQUES USED IN THE OCP AREA
FOR THE EPIDEMIOLOGICAL EVALUATION OF CONTROL OPERATIONS

1. ENTOMOLOGICAL EVALUATION

The entomological evaluation in the Onchocerciasis Control Programme in the Volta River Basin Area (OCP) is based on:

(a) examining a sample of treated breeding sites each week to determine the effectiveness of larviciding operations, and

(b) catching and dissecting adult *S. damnosum* s.s. to measure the density of the residual population and to assess the transmission potential.

A different sample of breeding sites is surveyed each week, whereas the adult catching stations are fixed and are visited on a routine basis, either weekly or fortnightly, depending on the situation. The location of catching points and the number in each area have been selected according to the following criteria:

(a) extent and complexity of the breeding sites along a stretch of river;

(b) accessibility throughout the year;

(c) potential risk, e.g., the periphery of the Programme area, or stretches of river difficult to control.

Wherever possible, the catching points are near the rivers where the highest number of flies can easily be caught.

1.1 Methodology of the surveys

1.1.1 Breeding site surveys

The following information is recorded on a standard form:*a*

(a) the date of examination of the breeding site;

(b) the name and location of the river and site, by established code;

(c) the situation regarding larvae and pupae.

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*a For reasons of economy the forms used for epidemiological evaluation have not been reproduced in this report. Specimen copies of these forms are available on request from Onchocerciasis Control Programme, World Health Organization, 1211 Geneva 27, Switzerland.
The following procedure is adopted:
(a) because of the general inaccessibility by other means, the sites are reached by helicopter;
(b) natural supports—e.g. rocks, vegetation trailing in the water—are examined across a breeding site wherever possible;
(c) because no standard has been established for estimating larval or pupal density quantitatively, only the presence or absence of the immature stages is recorded.

These surveys provide an immediate check on the effectiveness of larvicidal control and indicate whether immediate remedial action or modification of control operations is required.

The data collected, which are coded and stored, are a necessary complement to those collected on adult density and distribution.

1.1.2 Adult blackfly surveys

The whole OCP area is divided for purposes of entomological evaluation into 7 sectors. The sectors are further divided into 3 or 4 subsectors, each of which has 2 or 3 mobile catching teams responsible for visiting established catching points regularly. Each catching team has 3 vector collectors working in turn so that someone is always on duty throughout the day catching flies that come to bite. The information obtained is recorded on the following four forms.

**Form 1**

On this form the person in charge of the catching team records the numbers of flies caught each hour between 07 h 00 and 18 h 00 at a particular catching point.

**Form 2**

The flies caught are dissected. Either this is done on the spot or the flies are preserved on ice and dissected later at a central laboratory. Dissections are done by a technician or entomologist. A separate form is used for each catching point and the following information is recorded for each fly:

(a) time of catching;
(b) whether a blood-meal has been taken;
(c) parity;
(d) number of 1st and 2nd stage *O. volutus* larvae in thorax and abdomen;

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(e) number of 3rd stage *O. volvulus* larvae in head, thorax and abdomen;

(f) degree of infection according to *O. volvulus* larval stage and location;

(g) presence of meconium;

(h) other parasites found, e.g., Mermithidae, fungi.

*Form 3*

The main purpose of this form is to relay concisely to the OCP Vector Control Unit headquarters the current situation in each sub-sector each week. This information enables the subsequent week’s operations to be planned. Each line of the form contains information from a different catching point, the data all being transcribed from form 2.

*Form 3* includes for each site the particulars of numbers of hours worked by the vector collectors and total numbers of:

(a) females caught, females dissected and females found to be parous;

(b) females with 1st, 2nd and 3rd stage larvae of *O. volvulus* in the head, thorax or abdomen, and the numbers of larvae found;

(c) flies infected with other parasites.

*Form 4*

*Form 4* includes all the information collected from a particular catching point during 1 month. This form not only reveals at a glance the trend at that station as a result of the control measures undertaken, but is coded so that all data thereon can be readily stored in the computer and analysed.

Other data collected under the Programme include riverine discharge rates, location of larvicide dosing points, and quantities of insecticide applied. When this information is combined with the observations made during breeding site surveys and with that provided in form 4, the short- and long-term effectiveness of vector control operations can be monitored.

2. MEDICAL EVALUATION

As mentioned in section 3 of the main report, medical evaluation in the OCP area is based on simple and detailed surveys. In both instances, after the size of the sample for the whole area has been decided by the
statistician, the selection of the clusters is usually based on the following
criteria:
(a) size of the village (about 300 persons);
(b) level of endemicity (villages are considered hyperendemic with
75% infection, hypoendemic with 25% infection);
(c) geographical dispersal (representation of all main onchoecerciasis
foci);
(d) social homogeneity and stability;
(e) economic importance of certain areas;
(f) relationship with entomological evaluation (close to and away
from catching points);
(g) random selection after all the above-mentioned characteristics
have been considered.

2.1 Methodology of the surveys

2.1.1 Simple surveys
These include:
(a) exhaustive census of the population;
(b) parasitological examination (2 skin biopsies, one above each
iliac crest);
(c) simple measurements of visual acuity;
(d) examination for head nodules;
(e) skin biopsy at the outer canthus, when head nodules are present
or at the request of the ophthalmologist.

2.1.2 Detailed surveys
These comprise the following:
(a) exhaustive census with personal identification and recording of
migratory status;
(b) simple medical examination covering:
(i) measurement of weight and height;
(ii) degree and extent of dermatological involvement: excoria-
tions from scratching, papular onchodermatitis, pachyderma,
lichenification, atrophy of the skin, local melanoleucoderma;
(iii) involvement of the lymphatic system: elephantiasis, hyper-
trophy of the inguinal glands and hanging groin;
(iv) genital lesions: hydrocele with or without hernia;
(c) parasitological examination: counting of microfilariae from 2
skin biopsies taken above the iliac crest, staining of some specimens for
possible identification of other filarial species;

(d) ophthalmological examination aiming at:
   (i) continuous clinical and epidemiological evaluation of the total
       amount of blindness and impaired vision in the population;
   (ii) estimation of the importance of onchocerciasis as a blinding
disease, in relation to different levels of endemicity and different
populations;
   (iii) estimation of the influence of other diseases on the rates of
visual impairment, and their future role in relation to a change in
onchocerciasis transmission;
   (iv) continuous clinical study of the different ocular onchocer-
ciasis lesions and their development in relation to various degrees
of transmission or medical treatment;
   (e) other biological investigations whenever required.

2.1.3 Analysis of data

All data collected in the course of simple and detailed surveys will be
stored in a computer for any further analysis. Some calculations can,
however, be made at once. Among those most commonly used in the
OCP area are:

(a) calculation of prevalence by sex and age group;
(b) calculation of the total prevalence of onchocerciasis by village,
   adjusted by age and sex;
(c) measurement of intensity of infection by age and sex groups;
(d) study of the frequency distribution of the microfilarial load by
logarithmic conversion of the results of quantitative biopsies;
(e) calculation of the geometric mean of microfilarial loads by sex,
age group, and village (for the village the representative load is adjusted);
(f) a statistical evaluation of ocular lesions in the population, viz:
   (i) rates of ocular involvement due to onchocerciasis;
   (ii) pre-blindness rates, which are divided into 2 categories, visual
impairment and severe visual impairment;
   (iii) blindness rates, as defined in the report of the WHO Study
Group on the Prevention of Blindness.9

Blindness rates are calculated on the basis of the census population and not just of the people examined, i.e. allowing for absent people who may be blind. All these rates are weighted for age and sex to make them comparable from village to village.

The interval between surveys, for both simple and detailed evaluations, will be 3 years. The changes taking place in a single year do not appear to be large enough to warrant more frequent visits, nor do the survey techniques seem sensitive enough to detect them. To check this point, several clusters will be surveyed annually.

3. AQUATIC AND CHEMICAL MONITORING

In the Onchocerciasis Control Programme the objective is to control the populations of *Simulium damnosum* through the use of insecticides so as to interrupt the transmission of onchocerciasis. At the same time it is necessary to determine the immediate and long-term effects that the insecticide may produce on the other aquatic life forms in the rivers concerned. The insecticide being used in this programme is Abate, at total volumes of 20,000 litres of 20% emulsion concentrate in the first year, 80,000 litres in the second year, and 120–150,000 litres in the third year. This will amount to 1 kg of active ingredient for 25 km² of land area controlled. Abate has been selected for its high toxicity to *Simulium* larvae, its low vertebrate toxicity, and its relative safety to other non-target organisms. As the control programme is planned to continue over a period of 20 years, it is necessary to ensure that the prolonged application of this insecticide (and others that may be used in the future) will not have a deleterious effect on the aquatic environment.

The monitoring protocol that follows is designed to assess possible changes in the aquatic environment by measuring the populations of a number of non-target organisms and certain physical parameters of the environment. It is necessary to monitor (a) the short- and long-term effects of the chemicals used on the aquatic biota and (b) the physical and chemical fate and possible accumulation of the insecticide. Sample collections are made monthly at sites on selected rivers under regular treatment.

3.1 Aquatic monitoring

3.1.1 Invertebrates

(1) Drift nets. Standard nets are used either singly or in groups of three. The nets should be placed in several positions across the river and
arranged so that the top of the frame is 2 cm below the surface. Samples are taken (a) commencing 1½ hours before sunset and (b) commencing 1½ hours after sunset.

For daytime drift samples three nets should be used for half an hour. For night samples either six nets in two sets of three are used for a minimum of three minutes per set or, alternatively, two individual drift nets are used three times to produce a total of six nettings.

The rate of river flow is measured at the time of taking the daytime samples. It is done by measuring surface velocity with a float ten times over a distance of 10 metres. The mean of the ten readings is recorded on the data sheets. Alternatively, current meters may be used for this purpose.

(2) Artificial substrates. Standard containers are used and filled with lateritic stones 6–7 cm in diameter. The containers are placed in rivers and anchored in such a way that they may be retrieved at all river levels. At least two artificial substrate containers are used on each river, each being retrieved once per month (two months on the Bago River). The organisms are removed by rinsing the containers and stones in a bucket containing a dilute solution of formalin and acid (e.g., a 10-litre bucket will contain 100 ml of formalin and sufficient acid to give a pH below 4). The substrates are rinsed until the animals cease to emerge. The containers are then replaced in the river.

(3) Surber samplers. Standard Surber samplers are employed once at each sampling station during the dry season. Samples should not be taken until the dry season river regime is well established but should not be delayed until so late in the season that there is a danger of river levels beginning to rise. From each station 10–20 samples are taken once a year.

3.1.2 Fish

(1) Drift nets. Standard drift nets are used. Samples are taken once a month, at least two nets being put in position at sunset and retrieved 2 hours later. The nets should be placed in rapidly flowing water (1–2 m/s). The fish caught are killed, separated from any debris, and the sample preserved by fixing in formalin for later examination in the laboratory.

(2) Gill nets. Five gill nets of various mesh sizes (i.e., a battery of nets) are left in the river overnight. At least two nights’ fishing is essential to obtain representative results, but if this is not possible the battery of nets should be doubled for one night. The fish caught are
identified, measured, and weighed. Gill nets are used every 3 months in
the different stations.

3.1.3 Phytoplankton

There are two sources of phytoplankton in rivers—the river-bed
periphyton that contributes algal cells to the open water and real phyto-
plankton that develops in sluggish reaches of the river. The importance
of phytoplankton in river ecology is not yet known but it is probably
insignificant; nevertheless, investigations should be carried out. Samples
should be taken each month from each monitoring station, stored until
such time as they can be concentrated, and sent to an appropriate
authority for study. The sample taken at each site should be a composite
of five 100-ml dipper samples from the surface of the river. The sample
is preserved by the addition of 1 ml of 40% formalin, 0.5 ml of house-
hold detergent, and 5–6 drops of Lugol's iodine. On no account should
cupric sulfate (CuSO₄) be added to the samples.

3.1.4 Classification of organisms

Initially, invertebrates should be identified according to the taxo-
nomic level indicated on the record sheets and fish should be identified
by species wherever possible. As expertise and knowledge of the fauna
increase in any area, the level of identification and recording will
improve. Classification of organisms is reviewed periodically.

3.1.5 Equipment

Sampling equipment is supplied by WHO and is of a standard form,
specifications being as described below.

(1) Drift nets (invertebrates). The frame of each net is 25 cm ×
25 cm, the net being 3 m long and made of 300 μm mesh. The net
terminates in a collecting jar, the outer end of which is covered with
standard mesh. The nets may be used either singly or in groups of three.

(2) Artificial substrates. Containers of approximately 500 cm³
capacity are filled with laterite stones of diameter 6–7 cm.

(3) Surber samplers. The modified Surber sampler, which is placed
on the bed of a river, consists of a metal box measuring 15 cm on each
side. The upstream side is covered with removable gauze, and the
downstream side is attached to a removable net about 1 metre long
made of 210 μm mesh. The sides of the box parallel with the stream are
enclosed, and the top and bottom are open to permit agitation of the
river bed with a stick. The bottom edge is faced with rubber 1 cm thick to provide a good contact with the substrate. There is a container at the end of the net to collect the sample.

(4) Drift nets (fish). Drift nets for fish have a rectangular opening measuring 40 cm × 70 cm, are 3 m long, and have a mesh of 1.5 mm.

(5) Gill nets. Gill nets are 25 m long and 2 m deep. They exist in five mesh sizes—15, 20, 25, 30 and 40 mm. A battery of nets is a composite of all five sizes.

3.1.6 Storage and analysis of data

Standard forms for recording invertebrate monitoring data are used at all stations. The format has been adopted to facilitate comparison of records for storage and analysis. The completed forms are airmailed to Geneva at the end of each month, a copy being kept at the monitoring station.

Information on fish is similarly recorded on standard forms. When further information is available on the main species of fish present in each river throughout the year, the forms may be revised.

The data are analysed in Geneva and the results distributed to the monitoring teams. The information collected is reviewed at joint meetings of the teams, assisted by WHO staff and advisers, held twice a year. An annual report based on the data from the monitoring programme will be prepared for the Ecological Panel of the Onchocerciasis Control Programme.

3.2 Chemical monitoring

3.2.1 Water samples

In order to determine what happens to the insecticide in the river, water samples are taken at an appropriate site downstream from the point of application and sent to a laboratory for chemical analysis. The extraction procedure may vary according to the insecticide used; that for Abate is described in section 3.2.4 below.

3.2.2 Mud samples

Mud samples are taken from the bottom of a river by a method that permits the removal and collection of the top 2–3 cm of the area sampled. The samples, weighing 100–200 g, are transferred to a glass bottle and the area of the surface sample is recorded on the label. Procedures for extraction and analysis of mud vary according to the insecticide used.
3.2.3 Fish samples

At least two species of fish (one top-feeding and one bottom-feeding) are taken from the rivers at appropriate sites and prepared for shipment to a residue laboratory. Each sample of fish should weigh at least 200 g. Procedures for extraction, analysis and storage of fish vary according to the insecticide used.

3.2.4 Procedure for handling water samples

The sample, comprising 300 ml of river-water, is collected in a glass bottle, which is sealed and labelled with the time and place of collection and sent to a field laboratory for extraction within 24 hours if possible. In extracting the sample the water is transferred to a 500-ml glass-stoppered volumetric flask. A 4×1 cm stirring bar is placed in the flask and 1 ml of concentrated hydrochloric acid and 10 ml of n-hexane are added to the sample, which is then stirred at high speed for 15 minutes on a magnetic stirrer. A sufficient quantity of pure water is then added to force the hexane into the neck of the flask, from which 5 ml are pipetted into a small vial. The vial is labelled and forwarded to the gas chromatography laboratory.

3.2.5 Equipment required for chemical monitoring

The following supplies and equipment are required for the collection and dispatching of samples for chemical analysis.

- magnetic stirrers 3 per base station
- magnetic stirring bars 6 per base station,
- volumetric flask, 500-ml glass-stoppered 6 per base station
- n-hexane 4 litres per base station
- acetone 8 litres per base station
- formalin solution
- hydrochloric acid 1 litre per station
- vials, screw-cap 5-ml 1 gross per station
- glass bottles, screw-cap 500-ml 1 gross per station
- glass bottles, wide mouth, with screw cap (for fish and mud samples)
- wash bottles, polyethylene, 500 ml.