

Title: Saprolegnia on Roach (Leuciscus rutilus) and carp (Cyprinus carpio).

Author(s) NOLARD-TINTIGNER, N.

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INTRODUCTION

There is, in nature, as well as in the aquarium, a parasitic disease known as 'mousse' and which attacks predominantly fish. It is caused by Phycomycete fungi, genus Saprolegnia. The fungus causes external lesions and covers the fish with a thick white layer from whence comes the name 'mousse', commonly attributed to the disease, for which the scientific name is Saprolegnia.

This fungus, known for a long time by fishermen, was described in 1882 by the great anatomist Th.J. Huxley, requested by the British government to study the epidemic which attacked the salmon of English rivers in great numbers.

It is surprising to find on perusal of the literature how little is known about Saprolegnia.

In view of its frequency, it seemed interesting to us to study it, from the point of view of epidemiology and the relations existing between parasite and host.

I. INCIDENCE OF SAPROLEGNIA

A. INCIDENCE ON FISH

Saprolegnia infections seen in nature are of the sporadic or epizootic types.

Sporadic cases of infection are reported by oral tradition, fish manuals and occasionally by naturalists from different parts of the world.

Epizootic disease, causing a mortality which is important in the population of edible fish (salmon, perch, trout, carp, pike, eel etc), are described more minutely: Huxley, Th.J. (1882), Murray, G.(1885), Patterson (1903), Tiffney, W.N. (1939), Hoshing, T. (1956), Scott, W. (1964).

Both types of infection are found in pisciculture but the epizootic incidences are more frequent.

All fresh water fish seem to be able to be attacked by Saprolegnia with the help of a trauma or water pollution. What's more, it appears that fish living in pure water with high oxygen levels are more susceptible to pollution and the fungi invades them more rapidly.

B. INCIDENCE IN OTHER AQUATIC ANIMALS

Due to the economic and aesthetic interest in fish, the study of the fungi is limited at present to that class of animal.

Nevertheless, all aquatic vertebrates appear to be able to be infected by it. Different authors report, without naming the species, that amphibians, reptiles and even aquatic mammals can be infected. We have seen it occasionally in a *Xenopus* and probably (isolate not identified) on a seal in the Anver's Zoo. Vanbreuseghen (1966) made a similar observation on a seal in Lake Baikal.

Saprolegnias can parasitize aquatic invertebrates. The genus Aphanomyces is particularly common on copepods, daphnia and other freshwater, brine and even marine crustaceans.

II. GEOGRAPHIC DISTRIBUTION OF SAPROLEGNIA IN NATURE.

The disease is particularly conspicuous in the northern hemisphere: Europe, occidental and oriental, North America, & Asia (Japan, Indes).

One might think that Saprolegnia remains unknown solely in those places where it has never been investigated.

III. PARTICULAR INCIDENCE IN THE AQUARIUM

It is commonly observed that fish kept in aquariums become infected. This fungal disease is actually one of the most feared and most frequent diseases in the aquarium. If one fish becomes ill, one can see all of the fish become infected and die in the following days if no measures are taken.

We are dealing with a disease which always develops from an epizootic form in the aquarium, the more easily as the living space of the animals is limited to a quantity of water which is always relatively small and often unchanged.

IV. SAPROLEGNIA

The saprolegniacea family belongs to Phycomycetes Oomycetes. They reproduce asexually in the form of zoosporangia (photo 1) which emit zoospores which are mobile and biflagellate, and sexually by producing spherical oosporangia containing oospheres which are fertilized by copulatory tubes emitted by the anthrids. From the fertilization, very resistant oospores are formed.

The genus Saprolegnia is characterized by the structure of the sporangia. These are replaced by internal proliferation of new sporangia within the first (Fig. 1). When the first sporangium is empty of the zoospores which it contains, leaving in place only the envelope, a second sporangium forms at the base and grows inside the first. This is emptied in its turn and a third sporangium begins to grow again in the first and so on. Many sporangia (5 or 6) can be formed by internal growth and one can finally observe a succession of empty envelopes nested one inside the other (Fig. 1).

Like the majority of fungi, Saprolegnia exist normally in a saprophytic state on a variety of organic materials (animal and vegetable detritus). It is aquatic or develops in a very damp environment (motile zoospores).

V. SYMPTOMOLOGY OF SAPROLEGNIA IN THE MIRROR CARP AND THE ROACH

We have assessed a variety of fish species but particularly the mirror carp (Cyprinus carpio) and the roach (Leuciscus rutilus). The mirror carp was chosen because of the small number of scales which it possesses, which allows for greater ease in studying alterations of the skin. The roach was chosen in order to have a comparison between fish with scales and one which has partially lost them.

A. CLINICAL PICTURE OF THE DISEASE.

It seems impossible to us to define a clinical picture for the disease which covers all the cases of Saprolegnia. We have, nevertheless, been able to group the fish disease into the following two types of symptomology. All of the observed cases seem to be caused by Saprolegnia parasitica (Coker).

1. Type one: the fungi forms a dense mycelial mass on the lesion.

a). In the carp.

At first, the infected organ was discoloured, it seemed swollen & raised. The mycelium is not externally developed as yet, except for some long and uncommon filaments which come out of the lesion. The hyphae have no reproductive organs.

The distinction between diseased tissue and healthy tissue is very evident. The edges of the regular lesion are surrounded by two zones.

The first edge of the lesion. This seems to be formed of tissue which is perfectly intact but nevertheless raised.

The second zone is characterized by an internal weeping. There seems to be a small hemorrhage.

Apart from the lesion, from which filaments escape, the presence of the fungi is undetectable macroscopically at any other point on the surface of the fish.

During a second period, which corresponds to the most important growth of the lesion (to a diameter greater than approximately 1.5 cm) the appearance changes totally. This was apparent about two days after infection.

The marginal zone keeps the same bulging aspect, surrounded by a strongly vascular area. But the central zone is completely modified. The tissues are discoloured and necrotic. What remains is a white, diffuse mass (what Huxley calls "wet-paper"), from where hyphae, terminating in gross sporangia (150 - 200 μ long) emerge.

Other parts of the fish are not altered and the skin is perfectly normal. (photo 2).

In some cases, secondary infections appear in the vicinity of the first. But, whether there is one lesion or many, the infection extends in an outward manner.

b). In the Roach.

No matter what degree of infection, the parasitized region is always covered by a matt of mycelia, mixed with filaments, various impurities and unicellular organisms (photo 3). The sporangia emerge from it. This area no longer has scales. When scraping the fungi, the muscle is exposed as if the fungi was implanted in it.

The near-by region at first glance is made up of healthy skin, covered in scales. But the scales are arranged abnormally. Actually, they are reset nearly perpendicularly in relation to the body and even seem to be loose. In fact they are inserted into the dermis as are normal scales. Observation with binocular microscope allows one to see that they are intact; that the mass of filaments is situated under those which are reset.

Additionally, that region is surrounded by a zone with a great deal of blood circulation at a greater distance from the edge of the lesion than in the carp. This is the centre of the hemorrhage visible at the base of the scales.

2. Second type: the fungus does not form a mycelial covering on the lesions

a). In the carp.

Here the fungus never develops the typical 'mousse' form. The lesion shows the same characteristics as for the first type of infection

in that it is completely denuded. One only sees some filaments on the edges.

b). In the Roach.

At first, the parasitized region is colonized by a light mycelial mat. But, after a day or two at maximum, the mass of the mycelium is eliminated, generally in one lump, and the muscle is exposed. The fish shows no sign of the former parastism, only the edges of the lesion testifying to a small extent, to the presence of the fungi. The mycelium forms a thin ring around the open muscle, from where longer mycelial hyphea protrude into the water. Sporangia are very rare.

The disease proceeds very rapidly. The flocculate structure does not appear again. The surface of the exposed muscle enlarges more and more, only the edges supporting filaments (photo 4). The parastism spreads more and more and finally causes the death of the fish.

3. Other symptoms.

Additional exterior signs of saprolegnia, other symptoms which always precede death.

First, the fish is much more nervous, more excitable. A small jolt to the side of the container in which it is held, makes it leap from the water. Then, four to five days after the beginning of the infection, equilibrium trouble appears. The fish lies on one side or the other for longer and longer periods. During the periods, the comportment again seems normal. Meanwhile, the majority of fish cease to eat. This stage lasts for one or two days. The illness grows worse and finally, the animal, which does not respond to any stimulation, cannot re-establish its equilibrium. Death follows in the two or three hours which follow.

Certain animals put up a greater resistance and stay alive when the fungal disease has almost totally invaded them. Others die when the infection has not exceeded three centimetres in diameter. The environment seems to play an important role. In reservoirs fish die often without more than a small lesion. We have found, in contrast, in nature, individuals almost totally covered with 'mousse' which still live, although they might be so weakened that their capture presents no difficulty.

The lesion in that case is extensive at this point, and is in the form of a thick white mat covering all the body.

4. Other locations.

We have described, up to this point, infections localized on the body of the fish. Other regions are also attacked in carp and roach.

First. Cases of the disease to the eyes are rare. All the head is then covered with a fungal mass. Up to now, we have never seen gills infected by Saprolegnia.

Second. The fins are more frequently attacked. The fungi becomes installed, enveloping them completely and causing necrosis of all the covering tissue. In more advanced cases, the mycelium and the necrosed tissue detach leaving only the rays of the fins which then become completely independent (cfr. histo-pathology).

B. ANATOMOPATHOLOGICAL OBSERVATIONS.

Autopsy of infected fish and coloration of the internal organs with hematoxylin phloxine luminous green and with P.A.S. do not support the suggestion of an invasion of internal organs. All our observations

are consequently carried out on cutaneous lesions and the tissues immediately underlying them.

1. Histopathology of fish infected according to disease type.

a). In the carp.

Infection by Saprolegnia began by implantation of the fungus in the epidermis of the fish. Mycelial filaments infiltrate between the cells of the central layer. They spread in all directions and finish by disorganising all of the epithelium, separating the cells from one another. From then, necrosis is rapid. Saprolegnia occupies the necrosed surface and produces numerous sporangia which sometimes protrude to the surface of the fish. Likewise, the fungi forces the vertical filaments into the superficial layer of the dermis. Certain hyphae follow the blood capillaries and from there go on to directly parasitize the layer of loose connective tissue and muscular tissue underlying it. (fig. 2).

At this point, the invaded epidermis is completely replaced by mycelia (photo 5) and the superficial layer of the dermis is reduced to connective filaments which likewise necrose.

The destruction of the capillaries allows the escape of red corpuscles into the mycelial mat. (fig. 2).

Histological study of the marginal zone shows that the raised tissues which surround the lesion and which seem healthy macroscopically, are already invaded by Saprolegnia.

The filaments penetrate into the central layers of the epithelium (photo 6), separating the cells in their way. This is manifested macroscopically by an increase in the thickness of the skin.

As for the peripheal zone, which macroscopically can be seen to be red, it is made up of a large number of small hemorrhages.

These are localized at the limit of penetration of the mycelial filaments in the dermis. The hyphae disorganize and vitually destroy the capillaries which they encounter.

b). In the roach.

Mycelial filaments infiltrate horizontally between the middle layers of the epithelium of the skin and the scales, and then spread into the invaded tissues.

As the infection spreads, the epidermis is completely destroyed. From time to time, one finds some cells isolated into islands surrounded by fungus (photo 7).

The dermis is very rapidly colonized and the connective fibres disrupted. As a result, the scales become detached, float freely in the mass of mycelial filaments and finally fall away. The marginal zone looks exactly like that of the carp. One finds at the edge of the lesion, a region which is convex in comparison to the normal. The epidermis again is severely infected. The first marginal zone is surrounded by a hemorrhaged region which can be seen very well at the base of the scales (photo 8).

All the region surrounding the lesion for two or three centimetres is covered in scales which are fixed perpendicularly to the body and which no longer seem to adhere. Histological studies show that they occupy their normal places in the fish and are not detached. The mycelia invading the region at the level of the epidermis, initiate an increase in the thickness of the skin at the base of the scales. These last are then compressed and reset in the body of the fish. The dermis is not attacked at that spot again.

2. Histopathology of fish infected with the 'without mycelial mat' type.

The marginal zone has the characteristics described for the preceding type.

The central area is not covered with a thick mat of mycelia. One only finds filaments at the edge of the lesion, in the marginal zone and under the scales which border it. The infection starts first with a spot on the epidermis. The fungus sends out hyphae into the middle layer of the epithelium, the filaments spread in all directions making a path between the epithelial cells, pushing the mucous cells to the surface.

The dermis is likewise invaded and the scales, dislodged, drop off.

But instead of proliferating in the necrosed epidermal tissues, the fungus is rapidly rejected along with all the epidermis and dermis which has been invaded and is necrosing. There only remains some muscle stripped of all protective layers and invaded by filaments.

3. Histopathology of infected fins.

The various fins (dorsal, caudal, ventral and abdominal) are also very often covered with fungal disease.

The fungus starts in the epidermis which covers the fin, spreading filaments which reach out in all directions and complete the invasion. Some hyphae develop vertically and enter the dermis. Others grow between connective filaments, disorganize them and finally attack the rays of the fin. All the invaded tissue (epidermis and dermis) necrose completely (photo 9). It results finally in the rays of the fish being free, and independent of each other.

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Fig. 1. Sporangia of Saprolegnia.
 1. Zoospore.
 2,3,4. Successive
 sporangial envelopes.

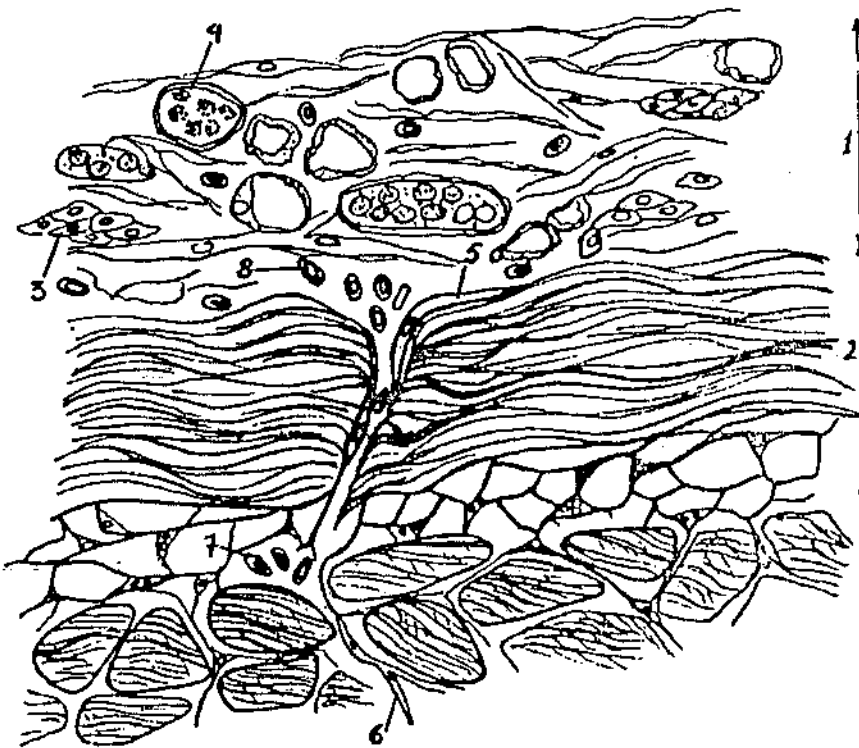
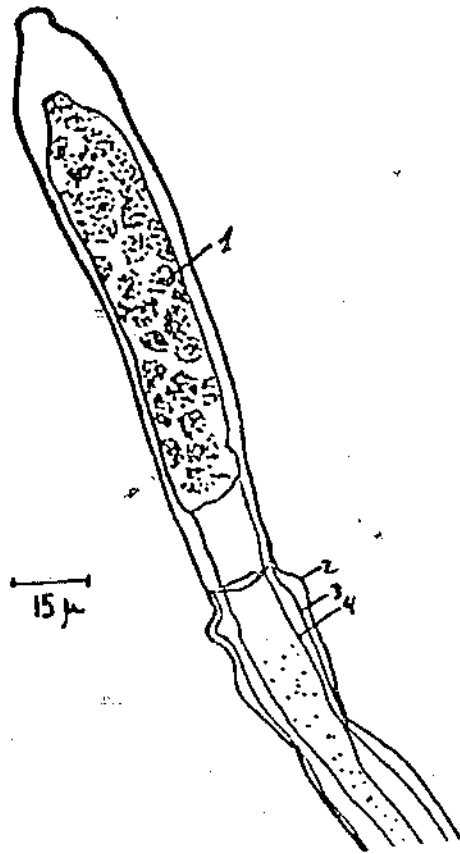


Fig. 2. Section of the skin of a carp invaded by Saprolegnia.
 1. Former location of the epidermis.
 2. Dermis.
 3. Epidermis cells.
 4. Sporangia.
 5. Hyphae following a blood capillary.
 6. Hyphae in the muscle.
 7&8 Red corpuscles.



Photo 1. Sporangia of Saprolegnia (cultured on a half-seed of hemp).

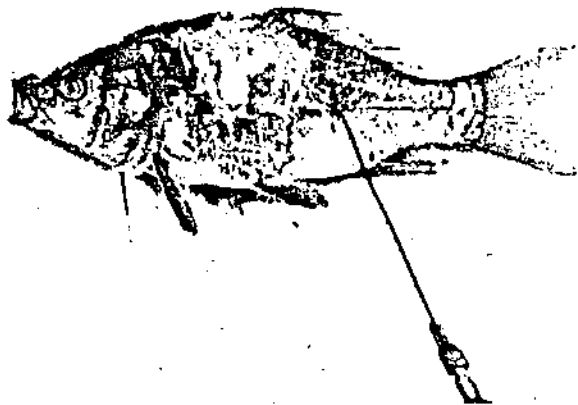


Photo 2. Carp: lesion covered by a mycelial mat.

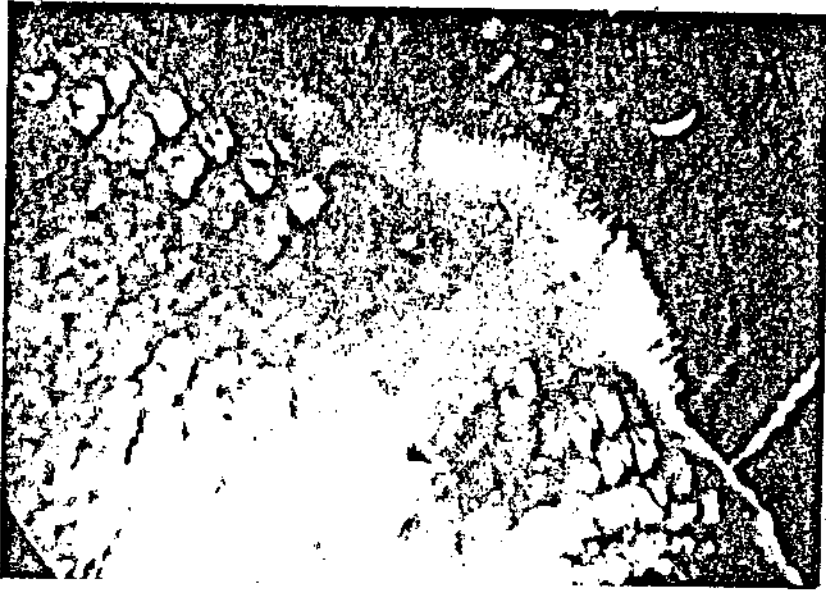


Photo 3. Roach: lesion covered by a thick mycelial mat.

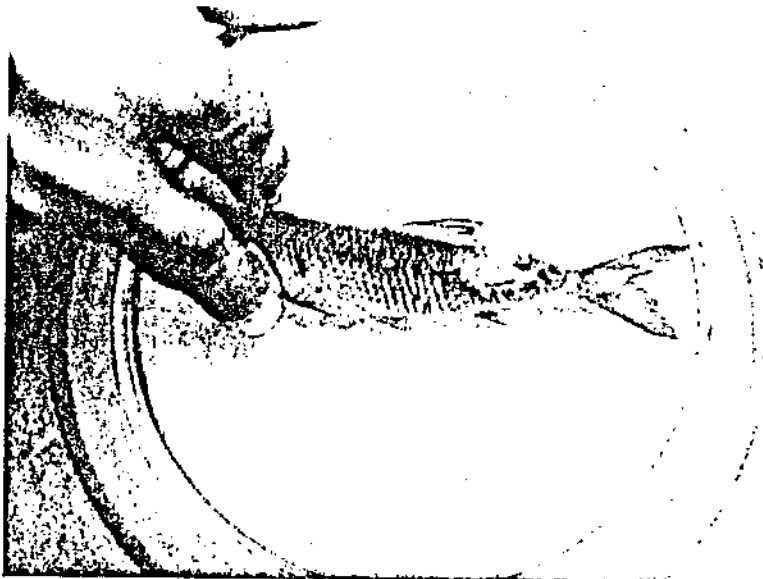


Photo 4. Roach: lesion not covered by mycelial mat.

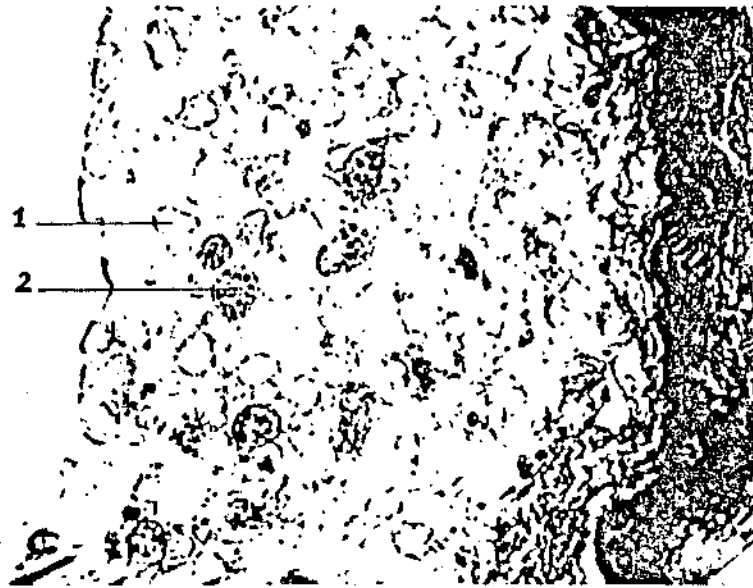


Photo 5. Section of the ectodermis of a carp completely invaded by Saprolegnia.

1. Cross section of a mycelial filament.
2. Sporangia.

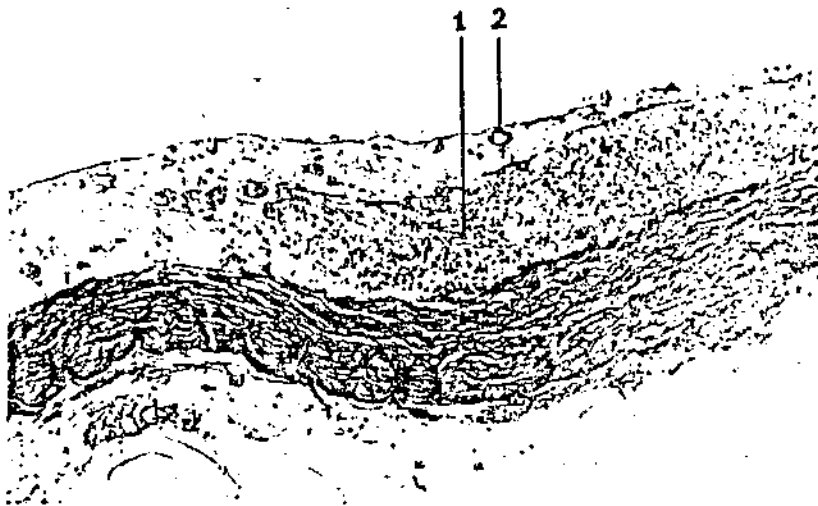


Photo 6. Lesion in a carp; section of the marginal zone.

1. Hyphe^a penetrate to the middle layer of the ectodermis.
2. Sporangia.



Photo 7. Section of the skin of an infected roach.

1. Scale.
2. Islands of ectodermal cells.
3. Saprolegnia.
4. Dermis.



Photo 8. Section of the skin of an infected roach.

1. Former location of the epidermis.
2. Dermis.
3. Muscle.
4. Red blood cells.



Photo 9. Section of a fin of an infected roach, and completely invaded by fungus.

1. Mycelial filament.
2. Connective fibres.
3. Rays.
4. Islands of ectodermal cells.

Notice

Please note that these translations were produced to assist the scientific staff of the FBA (Freshwater Biological Association) in their research. These translations were done by scientific staff with relevant language skills and not by professional translators.