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## INTRODUCTION

THE PRESENT epizootic of rabies in Ontario was introduced by the red fox (Vulpes vulpes) from Quebec into northern Ontario in 1954– 55 and reached the southern portion of the province in 1956. An extensive outbreak of rabies in foxes occurred in southern Ontario during 1958, in which approximately 1,200 cases of fox rabies were diagnosed.

In late summer of 1957, two rabid skunks (Mephitis mephitis) were reported from the Parry Sound area on the eastern edge of Georgian Bay. This was the first diagnosis of skunk rabies in Canada. During early winter, an additional case was reported from Huntsville (east of Parry Sound) and two cases were also reported from the province of Quebec in an area just north of Ottawa. Isolated cases were reported during the spring and summer of 1958 from widely separated areas in Ontario; Huntsville in the Muskoka district, Bowmanville on Lake Ontario, Vankleek Hill in the lower Ottawa valley and from several localities in an area between Georgian Bay and Lake Simcoe. Beginning in July, skunk rabies was reported more frequently, and by the end of 1958, two main areas of skunk rabies were recognized. The first area roughly followed the Ottawa valley from Pembroke in the west to Hawkesbury in the east, with an extension into the Rideau Lakes area through Westport. The second large area was one lying between Georgian Bay to the north and Lake Ontario to the south, bounded by Toronto and Bowmanville.

During 1958, 113 skunks from Ontario were submitted for diagnosis and 80 of these were rabid. Since that time, areas of skunk rabies have extended to cover most parts of southern Ontario. During 1972, a total of 316 skunks were diagnosed rabid from 478 submissions. The persistence of skunk rabies constitutes a continuing threat to domestic animals and man. Almost all of the skunk submissions to this laboratory were made because of some type of domestic animal or human involvement. Of the three laboratory confirmed human fatalities which have occurred in eastern Canada (Ontario and Quebec) as the result of known direct contact with a rabid animal, two were caused by skunks.

The prevalence of rabies in Ontario bats has been reported by Beauregard (2) and Beauregard and Stewart (4). Johnston and Beauregard (9) investigated the age-sex structure of the rabid fox population in southern Ontario. No similar studies have been published for skunk rabies in this area.

Over the past few years, numerous studies in the United States (5, 13, 15, 16, 20) and statistics published by the U.S. Public Health Service (12) have indicated that an annual peak in the numbers of rabid skunks occurs during the second quarter of each year; i.e., April–June. Examination of records at this laboratory has shown that the main annual peak of skunk rabies in southern Ontario occurs in December.

The present study was undertaken in an attempt to explain this difference and to elucidate the general picture of skunk rabies in southern Ontario.

### MATERIALS AND METHODS

All skunks used in this study were submitted to this laboratory by federal veterinary officers, practitioners or the public, as rabiessuspect cases. None of the animals was obtained through special trapping projects.

Information on the general seasonal prevalence of skunk rabies in Ontario from 1963 through 1970 was obtained from the records of this laboratory. During this period, a total of 2,375 skunks were examined from Ontario. Impression smears of brain tissue were stained either with William's modification of Van Gieson's stain (10) or, commencing in 1964, with the fluorescent antibody staining technique (3). Where there was a history of human involvement coupled with a negative result with either of the staining methods, portions of the brain were inoculated into experimental mice. These mice were held under observation for thirty days. Any animal which died during this period was examined by the above mentioned staining techniques.

Five hundred and eighty-one (581) skunk heads received at this laboratory for rabies

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diagnosis during 1971 and 1972 were also examined to determine age and sex. One half of the brain was used to prepare impression smears for the detection of rabies virus by the fluorescent antibody staining technique. The other half of the brain was fixed in 10% buffered formolsaline, embedded in paraffin, sectioned at  $5\mu$  and stained with haematoxylin and eosin. Neurons in the hippocampus were examined for the presence or absence of Barr bodies (11). Comparisons were made with the brains of 66 skunk carcasses, of which the sex was known. Age determinations were made by the microscopic examinations of canine tooth sections (6, 18). Comparisons were made with skunks of known age obtained from a colony maintained at this laboratory.

## RESULTS

During the years 1963 through 1972, a total of 2,956 skunks was examined. Of these, 2,098 (71%) were demonstrated to be rabid. The cumulative monthly distribution of the number of specimens received, specimens positive and the percent positive is shown in Figure 1. It can be seen that during this ten-year period, the largest numbers of skunks were received during the months of December and March and that the least numbers were received during June.

There was a pronounced annual peak in December in the number of skunks diagnosed rabid (Figure 1). This annual peak was a fairly constant feature, being found in all of the years in this study except November 1967 and January 1965 which had slightly greater numbers. Following the December peak, the number of rabid skunks declined sharply to a

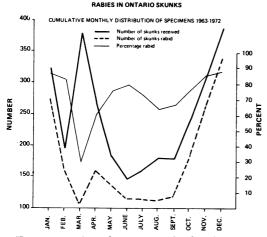
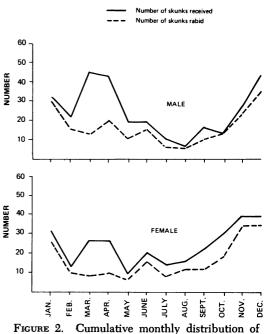


FIGURE 1. Cumulative monthly distribution of specimens 1963–1972.



CUMULATIVE MONTHLY DISTRIBUTION OF SPECIMENS 1971-1972

FIGURE 2. Cumulative monthly distribution of specimens 1971–1972.

low in March. A smaller "secondary" peak occurred during the month of April (Figure 1). This peak was less well defined during individual years and occurred in any of the months of April, May, June or July. However, as indicated in Figure 1, this secondary peak was more likely to occur during April. At no time was this peak of the same magnitude as that of December.

The percentage of those animals received which were rabid varied per month between 60 and 90 with the exception of March (Figure 1). This basic pattern with a low percentage in March was constant during all years of the ten year study. There was also a slight drop in the percentage during the late summer, usually during the months of August and September.

During the years 1971–72, age and sex determinations were carried out on 581 skunks, of which 392 (67%) were diagnosed as rabid. Figure 2 illustrates the monthly distribution of the total number of skunks examined. The largest number of skunks were received in December and March. More females were received during the fall months of August-November, while more males were received during February-May. Slightly more males were received during March than in any other month. A peak in the number of rabid skunks occurred in December with a smaller secondary peak during April and

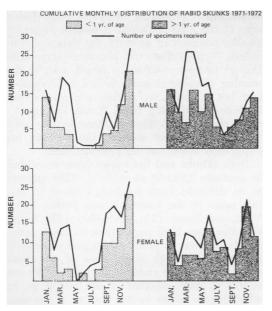


FIGURE 3. Cumulative monthly distribution of rabid skunks 1971–1972.

June. The percentage of rabid skunks in the total received showed fluctuations from month to month with a sharp decrease in March.

During 1971–72, the December peak in the number of rabid skunks was composed predominantly of animals of both sexes less than one year of age (Figure 3). During the early fall and winter, the percentage of rabid skunks less than one year of age increased, and subsequently decreased during late winter and early spring. Very few rabid animals less than one year of age were found during the months of May–July. This pattern followed rather closely the number of animals of this age group which were submitted for examination.

The smaller secondary peak in the numbers of rabid skunks received in 1971-72 occurred in the period April and June (Figures 2 and 3), and was composed of animals older than one year of age. During the month of March, there was a sharp increase in the number of specimens of both sexes received (Figure 2), but only a low percentage (30%) were rabid.

## DISCUSSION

Examination of the records of this laboratory for the years 1963–72 demonstrates a consistent pattern in the seasonal occurrence of rabies in skunks in Ontario. It must be assumed that the results derived from the ten-year period are more representative of the

whole picture of skunks rabies in Ontario than are those of the years 1971–72. The number of rabid skunks is low during the summer months of June to September, increases through the fall and reaches a "primary" peak during the month of December. This number then falls off sharply to a low in March followed by a smaller "secondary" peak in April.

The primary peak is composed predominantly of animals of both sexes less than one year of age. Young skunks leave the parental dens during the fall and begin to search for winter dens. Storm (19) and Bailey (1), in Illinois and Ohio respectively, have shown that young skunks usually become selfsufficient in August. During the period of August to October, the percentage of rabid skunks is often seen to decline. This appears to be due to an increase of young independent skunks being submitted for examination which have not yet contracted the disease. As the season progresses, more animals are found to be rabid until the peak in December is reached, at which time animals less than one year of age accounted for 66% of the submitted skunks and 63% of those rabid. Because of their greater numbers in the total population and because of their basic lack of knowledge of the area (8), young animals as a group would therefore be more exposed to the possibility of infection than would older animals. Thereafter, both the number of animals received and the number rabid declines through January and February when most skunks of all ages are in hibernation. Based upon the results of our studies, it is logical to assume that the primary seasonal peak is the result of increased contact during the fall period of population density of independent high animals.

In the present study, the number of submitted skunks greatly increased during March, although the number (and percentage) of rabid skunks was quite low. The high number of skunks received during March indicates that the animals had just emerged from the winter dens and were searching for and competing for a mate. On the other hand, the low number of skunks found rabid in March can be explained by a low rate of transmission during hibernation, assuming an incubation period of three to six weeks.

The smaller secondary peak in numbers of rabid skunks occurs usually during the month of April and is composed mainly of animals older than one year. The number of rabid males of the older age group increased during the period April–June (1971–72) indicating infection during the breeding season which

occurs in mid-February to early March (20). Verts (20) states that there is considerable fighting between males during this period. The number of rabid females increased during June (1971-72). Sanderson et al. (14), Schurrenberger et al. (16) and Verts (20) have attributed an increase at this time in mature rabid females to the activation of a latent infection by the stress of parturition in May. These authors found that females formed a larger percent of the rabid skunk population during the months of April and June. In the present study, however, there is no imbalance in favour of females during these months; in fact, there were slightly more rabid males during these months. It would appear, therefore, that this secondary peak in April is due primarily to cross infection during the breeding season and possibly subsequently in June to the activation of disease in females by the stress of parturition.

A major discrepancy exists between this study and those of other workers. In our studies, the primary peak of numbers of rabid skunks has been demonstrated in December, whereas studies on skunks in New York, Illinois, Iowa and other north-central states have shown that the annual peak of numbers of rabid skunks occurs during the months of April-June (5, 13, 15, 16, 20). Statistics compiled from the records of the CDC Rabies Surveillance Reports (12) for the years 1971 and 1972 also illustrate the April–June peak in numbers of rabid skunks in those states forming a broad belt of skunk rabies west and south of the Great Lakes (North and South Dakota, Minnesota, Iowa, Illinois, Indiana, Ohio). The implication of those studies is that the majority of skunk rabies is due to interaction between animals during or shortly after the breeding season. Verts (20) suggested that because no peak occurred following a period of high population density of inde-pendent skunks (i.e. in the fall or early winter), therefore epizootics of skunk rabies must involve something more than high population levels.

Unfortunately, most of the published material from the U.S. fails to indicate the number of skunks examined during any given month. Trends in the percentage of rabid animals cannot, therefore, be established. Verts (20) collected 99 skunks during the years 1961 and 1962, but only three were collected during the months of expected high population density; i.e. October-December. It is of particular interest to note that the provinces of Saskatchewan and Manitoba form the northerm-most extension of that broad belt of skunk rabies extending from the Great Lakes area of the United States. Although involving smaller numbers of animals, the data (unpublished; 7) from these two provinces agree with that of Ontario; that the primary peak in the number of rabid animals occurs in December and not in June, as is the case in the immediate neighboring state, North Dakota.

At the present time, a three to five year cycle such as that seen in the Ontario fox population could not be demonstrated.

Both skunks and fox rabies have co-existed in southern Ontario since 1957. This appears to be different from conditions found in certain areas of the United States. Parker (13), Verts and Storm (21), and Sikes (17) found that there are high populations of foxes (nonrabid) in areas of skunk rabies and conversely that there are high populations of non-rabid skunks in areas of endemic fox rabies. We are unable to state whether or not and if so, to what degree rabies in one species at the present time is influenced by rabies of another species. Although the two species do occur together geographically, there may be other factors which would tend to separate them. It must be assumed that in 1957, the initial outbreak in skunks was due to transmission of rabies to this species from foxes.

## Summary

An examination of the records of 2,956 skunk specimens submitted to this laboratory from southern Ontario during the years 1963 through 1972 shows that there is an annual cycle in the incidence of rabies. There is an increase in the number of rabid skunks through the fall and a yearly peak occurs in December. Thereafter, the number decreases. A smaller secondary peak occurs during the month of April. The number of rabid skunks is proportional to the number received except during the month of March when a large number of animals are received, but only a small number are rabid.

Sex and age determinations were done on 581 skunks during the years 1971 and 1972. Results indicate that skunks of both sexes less than one year of age are primarily responsible for the major December peak, while skunks of both sexes greater than one year of age are responsible for the secondary peak in April.

### Résumé

Une analyse des dossiers relatifs à 2,956 mouffettes du sud de l'Ontario, reçus au laboratoire de Hull, de 1963 à 1972, a révélé l'existence d'un cycle annuel concernant le nombre de cas de rage chez ces animaux. Ils augmentent à mesure que progresse l'automne et atteignent un sommet annuel en décembre. Ils diminuent ensuite, jusqu'à ce qu'un second sommet moins élevé survienne en avril. Le nombre de mouffettes enragées est proportionnel à celui des cas suspects référés au laboraoire, sauf durant le mois de mars où le contraire se produit.

La détermination du sexe et de l'âge de 581 mouffettes, au cours de 1971 et 1972, a de plus révélé que les mâles et les femelles de moins d'un an sont les principaux responsables du sommet observé en décembre. Par ailleurs, celui d'avril est attribuable aux mouffettes de plus d'un an.

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