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Changes in human fasciolosis in a temperate area: about some observations over a 28-year period in central France

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Abstract A retrospective study of 616 patients affected by fasciolosis was carried out to determine the numerical fluctuations of this disease over time, the recruitment of patients, and the modes of infection in a cattle-rearing area under a temperate climate (region of Limousin, central France). The annual number of patients showed irregular fluctuations between 1955 and 1987, after which it showed subsequent decreases until 1998. Before 1980, 66.3% of the persons under study were living in villages, whereas the others inhabited larger towns. An inverse relationship was noted after 1981: 80.8% of the individuals lived in towns consisting of >1000 inhabitants, whereas only 19.2% resided in smaller villages. Watercress was the infection mode in 98% of persons, with infections being attributed to four watercress species before 1980 versus only two species of *Nasturtium* sp. after 1981. The 235 watercress beds investigated in this study were found to contain 1 or 2 *Lymnaea* species as follows: *L. truncatula* only (84% of watering places), *L. glabra* only (6.3%), and both species (6.3%). In the populations of *L. truncatula*, natural infections of snails with *Fasciola hepatica* were irregular and occurred up to six times over the 28-year period in the region of Limousin. According to the authors, the decrease in human cases and the changes in the recruitment of patients might be explained by the demographic movements that occurred over several decades in the region of Limousin, with the moving of younger age groups into towns.

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Introduction

Human distomatosis caused by *Fasciola hepatica* is well known in public health for its endemic status in countries in which the temperature and humidity favor the development and survival of the host snail. The frequency of this parasitosis worldwide is high, since over 300,000 clinical cases may have occurred in more than 61 countries since 1970 (World Health Organization 1995a). The recording of ill persons in France is difficult enough. The single investigation conducted at a national level reports a total of 8,898 cases of human fasciolosis between 1950 and 1982, and the authors indicate that this figure probably represents an underestimation (Gaillet et al. 1983). As the other investigations thus far performed in France have been regional in scope, the total number of patients affected by fasciolosis remains imprecise, with some persons having been counted several times through the successive surveys. Attempts to analyze the changes in population in relation to fasciolosis have been sparse (Dreyfuss et al. 1994). As a result, the following two questions have arisen: how have the numbers of cases of human fasciolosis fluctuated over time in a cattle-breeding area under a temperate climate? Has the disease always affected the same population groups? To answer these questions a retrospective study of human cases of fasciolosis in the region of Limousin (central France) was performed. The choice of this region for the present study was based on the following two points: the greatest part of this cattle-breeding area lay at an altitude of 300–500 m, and its continental climate was modulated by the wet winds that came from the neighboring Atlantic Ocean.

Subjects and methods

Study protocol

As the registration of fasciolosis is not obligatory in France, the recording of patients affected by fasciolosis was performed not only with the aid of doctors and medical-analysis laboratories but also with that of other health professionals. After they had given their informed consent, the subjects under study were requested to answer a list of questions for the precise establishment of the date on which and the conditions under which they had ingested their contaminating meal, their eating habits with regard to vegetables collected in the field, the plant that had been eaten, and its place of origin. Table 1 depicts the number of persons affected by fasciolosis between 1955 and 1998 in the three departments of the Limousin region. The diagnosis of fasciolosis had been established using serology in all 616 persons recovered and subjected to the list of questions. As the metacercaria-carrying plant was often collected at a distance from the main home of these persons, the 616 cases were recorded not according to the address of the patients but in relation to the site at which the plant was collected or bought. This method allowed the retention in this study of some persons who lived in other French regions but were contaminated by the ingestion of vegetables collected in the Limousin region.

Malacology investigations were carried out in the watercress beds mentioned by patients for the identification of snails and determination as to whether they were infected with *F. hepatica*. A 28-year follow-up study was performed in 59 of these beds for determination as to whether the contamination of these watering places by *F. hepatica* was a periodic or irregular process. A sample comprising 50 or 100 adult snails (according to the size of the population) was collected from each bed every year (in June or July) during the 28 years of the study. Snails were dissected under a stereomicroscope for the detection of larval forms of *F. hepatica*.

Parameters studied

The distribution of 616 persons was studied according to 5 parameters: the date of the contaminating meal (year and month), the sex of the patients, their age group (< 30 years, 31–50 years, > 51 years), their main home address at the time of detection of the disease, and the contaminating vegetable. The figures recorded for each parameter between 1955 and 1980 and between 1981 and 1998, respectively, were divided by the total number of cases recorded in each of these two periods for the determination of frequencies and their subsequent comparison using the χ^2 test.

The prevalence of natural infection with *F. hepatica* was determined in the 235 watercress beds for each species of *Lymnaea* snails. In the case of the 59 beds studied over a 28-year period, the intervals (in years) between 2 natural infections of *L. truncatula* by *F. hepatica* were calculated. The individual values recorded for the latter parameter were averaged and standard deviations were established, taking into account the number of years for which natural infections of snails were found. One-way analysis of variance was used to compare these results and determine levels of significance.

Results

Figure 1 shows the numerical distribution of 616 patients in relation to the date of the contaminating meal. Before 1987 the number of persons affected by fasciolosis showed irregular fluctuations, and peaks were noted in 1958, 1963, 1968, 1969, 1971, 1977, 1981, and 1987. After the latter date a numerical decrease down to two cases in 1998 was noted. There was an appreciable decrease in the number of human cases detected during these last years.

The infection of these persons with *Fasciola hepatica* had occurred via the consumption of a meal of raw vegetables (613 persons), the drinking of spring water

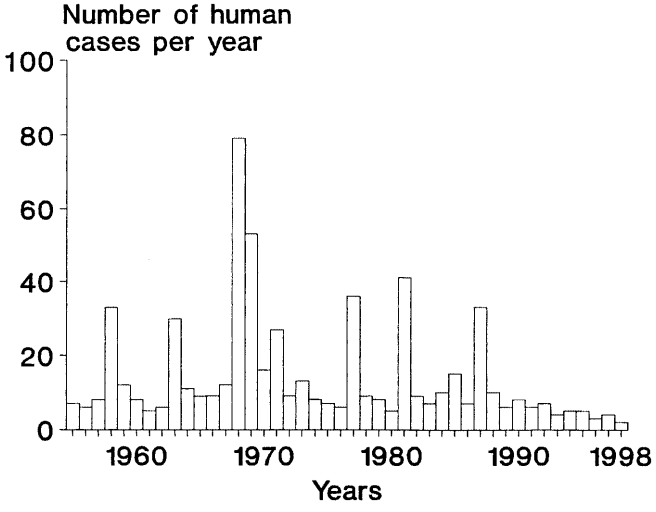

Fig. 1 Numerical distribution over time of 616 persons affected by fasciolosis in the Limousin region

Table 1 Numerical distribution of patients affected by fasciolosis over the period of 1955–1988 in relation to the place in which the vegetable was collected or bought and to the main home address of these individuals at the time of detection of the disease

Site of collection (or purchase) of contaminating vegetables	Categories of persons	Number of persons		
		Affected by the disease	Seen again	Subjected to a list of questions
Limousin region (Corrèze, Creuse, and Haute Vienne departments)	Living in the Limousin region when the disease was detected	701	617	583
	Living in other French regions	72	34	33
	Tourists[b]	11	0	0
	Unidentified persons[b]	3	0	0
Other French regions[a]	Living in the Limousin region	73	67	64
Total values		860	718	680

[a] These persons were not considered in the present analysis due to their contamination with vegetables collected or bought in areas outside the Limousin region
[b] No response to our letter

(2), or the sucking of a grass stalk (1). Most dates of meals were recorded in November (121 cases), in December (176), in January (148), in July (38), or in August (22). All cases recorded in July or in August involved individuals who were contaminated with watercress collected at an altitude of >500 m. The frequency of infection noted for persons infected in January during the period of 1955–1980 did not significantly differ from that recorded after 1981. Similar findings were also noted for the other differences, whatever the month studied.

The sample comprised 323 men and 293 women, whose age was 15–77 and 21–72 years, respectively, when the disease was detected. No significant difference was found between the frequencies recorded before 1980 and those noted after that date for each sex or age class. In a study involving only persons who had collected the metacercaria-carrying plant, drunk spring water, or sucked on a grass stalk (293 persons), 267 individuals were >50 years of age and the 26 others were 31–50 years old. However, no significant difference was found between the frequencies noted before 1980 and those recorded after that date, regardless of the age class involved.

Table 2 shows the numerical distribution of 616 persons in relation to their main home address at the time of detection of the disease. Before 1980, 66.3% of patients lived in the country, whereas the others inhabited larger towns. An inverse relationship was noted after 1981: 80.8% of persons lived in towns consisting of >1000 inhabitants, whereas only 19.2% resided in smaller villages. A significant difference ($P < 0.001$) between the frequency recorded before 1980 and that noted thereafter was found for the persons living in villages consisting of <1000 inhabitants. Another significant difference ($P < 0.001$) was found for the persons living in towns of larger size.

Table 3 depicts the modes of infection for the 616 patients. Watercress was the vegetable most often eaten by these individuals. Before 1980, four species belonging to the genera *Nasturtium* and *Roripa* were incriminated, whereas over the period of 1981–1998, only *N. officinale* and, to a lesser extent, *N. microphyllum* were implicated in human infection. This change in the consumption of watercress species was supported by a significant difference ($P < 0.01$) between the frequencies noted for *Nasturtium* sp. before 1980 and those recorded after that date. The frequencies of infection via the other infection modes (e.g., dandelion leaves, lamb's lettuce) were very low.

The *Nasturtium* and *Roripa* species were collected in 252 watering places or were bought in 47 local markets. Field investigations had allowed the identification of 235 watercress beds as follows: 2 concerns for watercress production on a commercial scale, 87 watercress plantations for familial consumption, and 146 sites ("natural beds") in which watercress grew spontaneously. A wire fence or a wall isolated 52% of these beds from any contact with cattle, sheep, or large wild mammals. Snails living in these 235 watering places formed monospecific (*Lymnaea truncatula*, 199 sites; *L. glabra*, 15; *L. fuscus*, 1; *L. ovata*, 1; *L. palustris*, 1; *L. stagnalis*, 1) or bispecific (*L. truncatula* plus *L. glabra*, 15 sites; *L. truncatula* plus *L. palustris*, 1; *L. ovata* plus *L. stagnalis*, 1) populations. Snail dissections performed for the detection of *F. hepatica* larval forms during the months following the detection of human case(s) were successful in 65 watercress beds (*L. truncatula*, 57 sites; *L. glabra*, 2 sites; *L. truncatula* in bispecific colonies with *L. glabra*, 6 sites).

A total of 59 watercress beds were studied over a 28-year period to follow the natural infection of *L. truncatula* with *F. hepatica*. The results were negative in eight sites. In contrast, infected *L. truncatula* were found in the other 51 watering places, and Table 4 shows their distribution in relation to the intervals between two

Table 2 Numerical distribution of 616 patients over the periods of 1955–1980 and 1981–1998 in relation to their main home address at the time of detection of the disease

Town or village (number of inhabitants)	1955–1980		1981–1998	
	Number of cases (434)	Frequency (%)	Number of cases (182)	Frequency (%)
<1000	288	66.4	35	19.2
1001–10,000	75	17.3	79	43.4
10,001–50,000	44	10.1	42	23.1
>50,000	27	6.2	26	14.3

Table 3 Numerical distribution of 616 patients over the periods of 1955–1980 and 1981–1998 periods in relation to the mode of infection

Mode of human infection	1955–1980		1981–1998	
	Number of cases (434)	Frequency (%)	Number of cases (182)	Frequency (%)
Nasturtium officinale, *N. microphyllum*, or hybrids	262	60.3	180	98.9
Roripa sylvestris, sometimes *R. amphibia*	49	11.2	0	0
"Mixed watercress"[a]	113	26.0	0	0
Taraxacum officinale	7	1.6	1	0.5
Valerianella olitoria	1	0.2	0	0
Others[b]	2	0.4	1	0.5

[a] The interrogation of patients did not allow determination of the species of vegetable eaten, although two or three *Nasturtium* and *Roripa* species were growing in the watercress bed
[b] Spring water (2 cases), unidentified grass (1 case)

Table 4 Distribution of 59 watercress beds in relation to the intervals between 2 natural infections of *Lymnaea truncatula* with *Fasciola hepatica*

Number of times for which natural infection of snails was found	Number of watercress beds	Intervals[a] (in years) between 2 successive infections of snails with *F. hepatica*			
		1st/2nd	2nd/3rd	3rd/4th	4th/5th
1	7	–	–	–	–
2	22	9.2 ± 7.7	–	–	–
3	11	6.3 ± 4.1	7.5 ± 5.2	–	–
4	7	5.5 ± 3.1	6.4 ± 4.2	4.5 ± 2.2	–
5	3	4.3 ± 2.1	5.5 ± 2.2	3.5 ± 2.1	5.2 ± 3.0
6	1	6	11	3	4

[a] Data represent mean values ± SD

infections of *L. truncatula* populations with *F. hepatica*. The natural infections of these *L. truncatula* were not continuous and occurred at more or less brief intervals. Over the 28 years of the study, 1–6 infections of snails were noted, separated from each other by mean intervals of 3.5–9.2 years. Analyses of variance did not show any significant difference between the mean values.

Discussion

The results obtained in this retrospective study of human cases of fasciolosis in a cattle-breeding area under a temperate climate clearly demonstrate that the annual number of persons affected by the disease has steadily decreased since 1987 and that the recruitment of patients has often occurred in towns. An explanation for these findings is difficult enough to find. Synchronism between the frequency of human fasciolosis and that of the disease in domestic ruminants (Rondelaud 1980; Rondelaud et al. 1982) did not previously exist in central France, as human cases became scarce, whereas the animal reservoir for this parasitosis was always significant, with 13% of cattle (among the 12,724 examined) harboring adult *Fasciola hepatica* in the Limousin region between 1983 and 1985 despite repeated anthelminthic treatments (Mage 1989). Another factor must be found to explain this recent decrease in the number of human cases. As attempts to heighten public awareness of the risks incurred by the consumption of raw watercress after its collection in the field have not aroused the attention of the media, the most logical explanation would involve consideration of the demographic movements that have occurred over several decades in France. The moving of young people from the country into towns and the changes in their eating patterns might be the cause of the fall noted in the number of human cases.

Most of the other characteristics associated with human fasciolosis were in concordance with the data usually reported in reviews of this disease (World Health Organization 1995b; Mas-Coma et al. 1999), particularly those involving the modes favoring the infection of persons. The four species of watercress incriminated (*Nasturtium* sp., *Roripa* sp.) and the changes in their frequency over time suggested that the consumption of these vegetables by inhabitants of the Limousin region must have been of importance before 1980 and that the eating preferences of consumers had changed with time to the selection of *Nasturtium* sp. only. More unusual were the other two modes as sources of infection. The drinking of spring water contaminated with *F. hepatica* had previously been reported in two epidemiology surveys (Sadaillan et al. 1949; Bargues et al. 1996), and this finding could easily be explained by the formation of metacercariae that floated on the water (Esclaire et al. 1989). The development of fasciolosis in a man who had sucked on a grass stalk demonstrated that the metacercariae of *F. hepatica* could encyst even on species belonging to this plant family.

The freshwater snails found in watercress beds comprised six species of *Lymnaea*. If *L. truncatula* was the main host snail, it was not the only species, since larval forms of *F. hepatica* were found in *L. glabra* from two watercress beds. The role of the other four species of *Lymnaea* is more difficult to explain. Indeed, the juvenile snails of these species could sustain the full larval development of *F. hepatica* under experimental conditions (Kendall 1950; Busson et al. 1982), but natural infections were never found in these snail species over the 28 years of the present study. This finding raises the question as to whether it is possible for these species to be intermediate hosts of *F. hepatica* in the watercress beds of the Limousin region.

The irregular infection of *L. truncatula* in watercress beds raises the question of the definitive hosts contaminating watering places, since 52% of beds were isolated from any contact with domestic or wild ruminants. Under these conditions it would be interesting to determine the different wild species that might be responsible for the infection of these isolated places. This question might be resolved only by the capture of small mammals in these sites and their subsequent dissection to determine the presence of a natural infection with *F. hepatica*.

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