

Outbreak of Trichinellosis Due to Wild Boar Meat and Evaluation of the Effectiveness of Post Exposure Prophylaxis, Germany, 2013

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Background. Food safety authorities discovered that wild boar meat products contaminated with *Trichinella spiralis* had entered the food chain in Germany in March 2013. Public health authorities issued guidelines for health professionals including post-exposure prophylaxis (PEP) using mebendazole and advised the public to seek medical advice if exposed. Our objective was to identify factors associated with the development of trichinellosis and to evaluate post exposure prophylaxis.

Methods. Persons who reported to local public health departments as exposed were interviewed concerning exposure, symptoms, and medication. Serum samples were tested by an in-house *Trichinella*-specific enzyme-linked immunosorbent assay. Cases were defined as persons presenting with myalgia and/or periorbital edema and *Trichinella*-specific immunoglobulin M and immunoglobulin G antibodies after exposure to implicated products.

Results. Of 101 persons interviewed, 71 were exposed and serologically tested. Antibodies were detected in 21/71 (30%) and 14/71 (20%) met the case definition. Attack rates were positively correlated to the amount of implicated product consumed. Among $n = 37$ persons who received anthelmintics as PEP, 6 persons developed trichinellosis. These cases exclusively occurred among persons starting PEP 6 days or later post-exposure. Exposure to implicated products and delaying PEP were also significantly associated with developing trichinellosis ($P < .01$) in a multivariable analysis.

Conclusions. Concerted efforts by food safety and public health authorities lead to timely outbreak control and facilitated the provision of early PEP. PEP appears to be effective in preventing trichinellosis when given early, preferably within 6 days. We therefore recommend initiating PEP without delay in similar settings and encourage public health professionals to fast-track this intervention.

Keywords. trichinellosis; post-exposure prophylaxis; mebendazole; outbreak; Germany.

Human trichinellosis occurs through consumption of raw or inadequately processed meat or meat products containing larvae of the parasitic nematodes of the

genus *Trichinella*. The course of human infection can be divided into 2 phases: The first or intestinal phase lasts approximately 1 to 2 weeks and is associated with diarrhea and abdominal pain caused by the reproduction of adult nematodes in the intestinal mucosa. The second or muscular phase occurs approximately 1 week post-infection when new born larvae penetrate the mucosa and migrate throughout the body until they reach the cells of the striated skeletal muscles [1]. This phase is characterized by fever, periorbital edema, and myalgia [2]. Possible complications are myocarditis, thromboembolic disease, and encephalitis. Chronic myalgia can persist. In cases following a high

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infectious dose, lethality is estimated to be as high as 5% despite treatment [3].

Pork represents the most important source of human infection worldwide, but horse or game meat also plays a significant role [4]. In the majority of countries in the Americas and Europe including Germany, trichinellosis cases are rare due to controlled housing conditions in commercial swine herds and systematic inspection of meat intended for human consumption [3–5]. Outbreaks however are not uncommon in connection with game meat, especially wild boar [6–9].

Anthelmintics such as mebendazole and albendazole are the principal drugs used for the treatment of trichinellosis [2]. Upon diagnosis, treatment should be initiated as soon as possible to eliminate the parasite in the gastrointestinal tract and thus reduce the generation of the newborn larvae and their migration into striated muscle tissue [10]. Data from animal models suggest that the development of muscle stage larvae can be prevented if anthelmintics are administered within the first few days of exposure [11–13]. Apart from a small case series [14], studies evaluating the effectiveness of post-exposure prophylaxis (PEP) in humans and in an outbreak setting are not available.

Outbreak Setting

On 27 March 2013, the National Reference Laboratory for *Trichinella* (NRL-T) at the Federal Institute for Risk Assessment discovered during routine molecular typing that the carcass of a *Trichinella* infected boar had accidentally been declared fit for human consumption. Eight kilograms of the muscle tissue of this carcass had already been used in the production of 1050 cured raw sausages (“knacker sausage”), and further 30–35 kg were sold as fresh meat intended to be cooked. The fresh meat was on sale on 23, 26, and 27 March at the store of a game-handling establishment and at mobile retail food establishments in 16 cities and towns in the federal states of Saxony and Brandenburg (Figure 1). The sausages, intended to be eaten raw, were on sale on 26 and 27 March only. On 28 March 2013, the Robert-Koch-Institute as national public health authority was informed about the incident and invited to support the outbreak investigation of the local public health and food safety authorities.

The objective of this investigation was to assess the extent of the outbreak, to identify factors that are associated with the development of trichinellosis, and to evaluate effectiveness of anthelmintic medication administered as post-exposure prophylaxis in an outbreak situation.

METHODS

Public Health and Food Safety Action

Immediate measures included the identification and tracing of implicated meat products, the withdrawal from the market of any unsold stock, as well as informing primary care physicians,

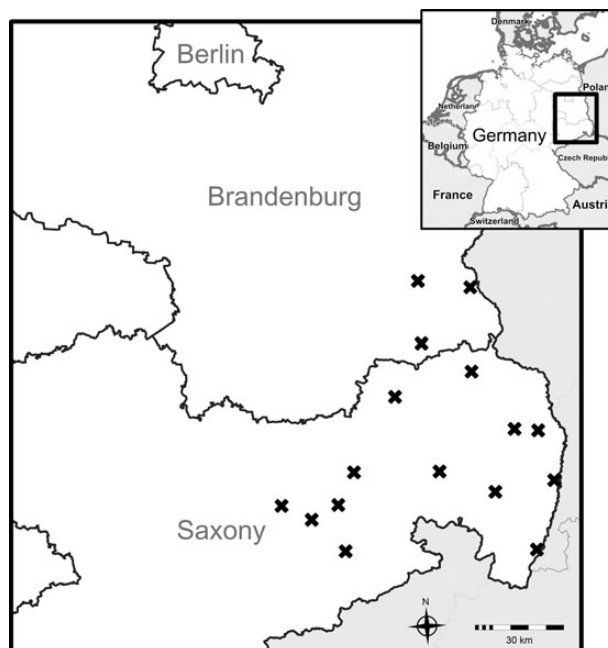


Figure 1. Locations (“X”) within the federal states of Brandenburg and Saxony where the implicated products were on sale between March 23 and 27. Inset shows the location within Germany and neighboring countries. Outbreak of trichinellosis, Germany, 2013.

internal medicine specialists, and emergency departments in the area about the expected outbreak and the possibility of PEP. A press release was issued on 28 March 2013 advising consumers to return or dispose of the implicated products and to seek medical attention as soon as possible if exposed. After ad hoc consultations with clinical infection specialists, recommendations on PEP for asymptomatic persons and treatment were issued as shown in Table 1 [15]. A dedicated telephone hotline was implemented for the public by the local health department (LHD) and ran over the long Easter holiday weekend until 5 April 2013.

Study Design and Epidemiological Investigation

We conducted a prospective cohort study among persons with self-reported exposure to the implicated products using investigator administered questionnaires and serological investigations. A case was defined as a person with *Trichinella*-specific immunoglobulin M (IgM) and/or immunoglobulin G (IgG) and self-reported symptoms of myalgia and/or periorbital edema following consumption of the implicated product.

Participants were recruited among persons contacting LHDs for information or serological testing following the press release of 28 March 2013. All persons who reported having eaten products from the affected batch were invited to participate in a telephone or face-to-face interview with an epidemiologist or public health official and offered *Trichinella*-specific laboratory diagnostics. Our questionnaire included items on demographics, date

Table 1. Recommendations on Diagnostics and Post-Exposure Prophylaxis and Treatment Issued to Health Care Professionals During the Outbreak of Trichinellosis, Germany, 2013 [15]

Scenario	Recommendation
Asymptomatic person presenting within 7 d of exposure	Mebendazole 5 mg/kg body weight, twice daily for 5 d. Contraindication: Children younger than 2 years and pregnant women (Pyrantel may be considered).
Asymptomatic person presenting within 8 to 30 d of exposure	Conduct laboratory testing. If eosinophilia (>500/ μ L), elevated creatine kinase (CK) levels or <i>Trichinella</i> -specific antibodies are detected, treatment should be discussed with a specialized center (serology may be negative initially). If all tests are negative: no further measures. Reassess if symptoms develop.
Asymptomatic person presenting more than 30 d post exposure	No further diagnostics. Reassess if symptoms develop.
Person presenting with symptoms congruent with trichinellosis at any point of time	Conduct laboratory testing (eosinophilia, elevated creatine kinase (CK) levels, and <i>Trichinella</i> -specific antibodies. If negative: repeat laboratory diagnostics after a few days. If positive in initial or follow-up test, a specialized center should be contacted.

and place of purchase, date of exposure, food preparation and amount eaten, leftover products in the household, clinical symptoms, and contact to the healthcare system including recommendation of and compliance to PEP. Blood specimens were collected at the LHD or, if preferred, at a physician's office of the exposed persons' choice at the earliest 45 days after exposure and sent to the NRL-T for analysis. In case of inconclusive serological results, blood sampling and serological testing were repeated 10 weeks after exposure. Participants were followed up at least once if initial contact had occurred within the first 6 weeks post-exposure to allow for reporting of late symptoms and detection of late seroconversion.

Ethical Considerations

In accordance with article 25, section 1, of the German Infection Protection Act of 2001, a formal ethical review process and approval was not required for this investigation of an ongoing outbreak. Data protection and medical ethics standards were adhered to as regulated by law. Informed consent was obtained by all participants or, if applicable, their legal guardians.

Laboratory Testing

Serological investigations were conducted using an enzyme-linked immunosorbent assay (ELISA) based on the excretory-secretory antigen [16] and implemented for the examination of human sera [17]. The sensitivity/specificity of the ELISA are 94%/85% and 89%/96% for the detection of human anti-

Trichinella-IgM and -IgG, respectively. All positive and inconclusive ELISA results were confirmed by Western blot adapted for examination of human sera [18].

Product samples collected at the meat processing establishment as well as leftover products from private households were analyzed at the NRL-T for *Trichinella* according to regulation EC (No.) 2075/2005 [19]. The number of larvae per gram (lpg) was calculated for each sample. Identification of the larvae at the species level was performed by means of multiplex polymerase chain reaction (PCR) [20].

Statistical Analysis

Attack rates (AR) and exposure-specific relative risks (RR) and 95% confidence intervals (CI) were calculated. Rank sum or *t*-tests were used with continuous variables; Cuzick test was used for trend with data across ordered groups. Prevalence ratios were calculated to investigate the predictive value of self-reported symptoms for seroconversion. To evaluate PEP failure, we analyzed a subset of persons who received anthelmintic medications as PEP. Multivariable regression analyses with case status as the dependent variable (using forward elimination of variables with a cut-off *P*-value of 0.2 and adjusted for age and sex) were performed using a fitted generalized linear model ("binreg") in Stata version 13 (Stata Corp., Texas).

RESULTS

A total of 101 persons or their legal guardians agreed to be interviewed of whom 84 had eaten the implicated products. Follow-up serological results and information on individual PEP regimens was available for 71 exposed persons who thus met the inclusion criteria for this analysis.

Laboratory Investigation

Serological investigations indicated a recent *Trichinella* infection in 21 of 71 (30%) exposed persons, of which 17 (81%) were positive for both anti-*Trichinella*-IgM and -IgG (confirmed by Western blot) and 4 (19%) showed a seroconversion for anti-*Trichinella*-IgM between 6 and 10 weeks post-exposure.

Among 9 persons, with an initially inconclusive ELISA result ≥ 45 days after exposure, 5 (56%) had developed anti-*Trichinella*-IgM and/or -IgG, 2 (22%) were negative by ELISA and further 2 persons (22%) were confirmed negative by Western blot upon retesting 10 weeks after exposure.

The larval burden (larvae per gram, lpg) in the diaphragm muscle of the *Trichinella* infected wild boar was 60 lpg. The larval burden in 10 sausages (weighing 100 grams each) collected at the company ranged from 0.3 to 0.7 lpg (median: 0.5 lpg; standard deviation: 0.13). Three sausages collected from 3 different households contained *Trichinella* larvae with a larval burden ranging from 1.1 to 1.3 lpg. The larvae were identified as

Trichinella spiralis by multiplex PCR. No larvae were detected in the remaining 3 sausages from an additional household.

Epidemiological Investigation

Exposed persons were between 3 and 87 years old (mean: 52 years) and lived in 4 different federal states (Saxony, Brandenburg, Bavaria, Lower-Saxony). Thirty-five (49%) persons were female. Two persons were hospitalized during follow-up; no deaths occurred. Exposure ranged from less than 1 up to 6 sausages (n = 65) or from 1 to 4 or more portions of wild boar roast (n = 5); only one person reported having eaten both sausage and wild boar roast. The median exposure was 1 sausage or 1 portion of wild boar roast. The cured raw sausages were typically consumed on the day of purchase (78%) or the following day (12%) (mean 0.44 days, range 0–8 days). Cooking, frying, or other means of heating above 71°C/160F of the sausages before consumption were not reported.

A total of 44/71 (62%) persons (all adults) had received anthelmintic medication: 42 (95%) persons took mebendazole (Vermox forte) and 2 (5%) took albendazole. Medication was taken for a median time of 6 days (interquartile range: 3.5–9 days). One person discontinued due to adverse effects after the first dose and 1 person took mebendazole for as long as 21 days. Twenty-nine persons (66%) took 2 doses per day, 11 (25%) spread their medication into 3 or more daily doses, and 4 (9%) took medication as single daily dose.

Risk Factors for the Development of Trichinellosis and Effectiveness of PEP

Among persons who had eaten sausages and could specify the exact amount, 14/65 fulfilled the case definition (AR = 21.5%). Seven persons seroconverted without developing myalgia or periorbital edema. No seroconversions occurred among persons

who only ate wild boar roast (n = 5). The AR among those who had eaten less than 1 sausage (1/21 [5%]) increased more than 4-fold among those who had eaten 1 or 2 sausages (10/44 [AR = 22.7%], RR = 4.76) and again almost 3-fold if more than 2 sausages (3/5 [AR = 60%], RR = 12.6) were eaten, demonstrating a strong dose-response relationship (Cuzick test for trend, $P < .006$). The AR among persons who received anthelmintic medication as post-exposure prophylaxis (6/37, AR = 16.2%) was lower than among those who did not (8/26, AR = 30.8%); the difference, however, was not statistically significant ($P > .17$). No association was found between case status and age group, sex, or date and place of purchase.

To further evaluate PEP failure, we analyzed the subset of persons who received mebendazole or albendazole as PEP (n = 37), that is, before developing clinical symptoms consistent with trichinellosis and/or *Trichinella*-specific antibodies (excluding persons who received anthelmintics as treatment). A correlation between the number of days since exposure until the first dose of PEP medication and PEP failure was evident: No cases of trichinellosis were observed among those who had received the first dose of PEP within 6 days post-exposure (n = 21, AR = 0%). PEP failure was observed in 4/12 (33.3%) persons starting PEP within 7–10 days and in 2/4 (50%) persons starting within 11–17 days, respectively (Figure 2, Cuzick test for trend: $P < .002$). In univariable analyses, we did not find differences between cases and noncases in terms of the number of daily doses or the number of consecutive days on anthelmintics. In the final multivariable model (which included all 37 persons who received anthelmintics as PEP) adjusting for age and sex, the factors associated with trichinellosis according to the case definition were the number of sausages consumed (RR = 2.75, 95% CI, 1.52–4.97), the delay (in days) between exposure and start of anthelmintic medication (RR = 1.63, 95% CI,

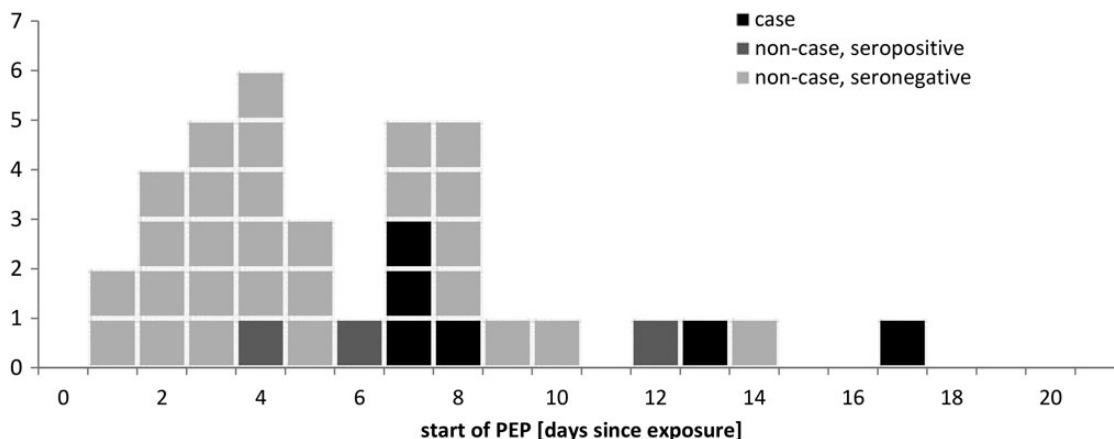


Figure 2. Patient outcome by post-exposure prophylaxis (PEP) delay following consumption of implicated sausages, outbreak of trichinellosis, Germany, 2013 (n = 37).

Table 2. Prevalence of Symptoms Among Study Participants (Total and by Serological Outcome) and Prevalence Ratios Following Consumption of Implicated Meat Products, Outbreak of Trichinellosis, Germany, 2013

Symptom	Prevalence of Symptom Among			
	Total (n = 71)	Seropositive (n = 21)	Seronegative (n = 50)	Prevalence Ratio ^a (Seropositive/Seronegative) [95% CI]
Myalgia and/or periorbital edema	26.5%	66.7%	8.5%	7.8 [2.93–20.97]
Myalgia	19.1%	47.6%	6.4%	7.5 [2.29–24.36]
Periorbital edema	13.0%	38.1%	2.1%	18.3 [2.44–137.10]
Headache	23.2%	47.6%	12.5%	3.8 [1.59–9.12]
Fever >38°C	13.0%	33.3%	4.2%	8.0 [1.81–35.34]
Chills	13.0%	28.6%	6.3%	4.6 [1.26–16.57]
Diarrhea	26.1%	28.6%	25.0%	1.1 [.50–2.63]
Abdominal pain	17.4%	23.8%	14.6%	1.6 [.58–4.56]
Cough	8.7%	14.3%	6.3%	2.3 [.51–10.41]
Conjunctivitis	7.2%	9.5%	6.3%	1.5 [.27–8.46]
Cardiac symptoms (NOS)	1.4%	4.8%	0.0%	∞

Abbreviations: CI, confidence interval; NOS, not otherwise specified.

^a The prevalence ratio is equivalent to the positive likelihood ratio (LR+) in diagnostic testing. Here it denotes how much likelier a positive serological test is in an exposed person when the respective symptom is present compared to when it is not.

1.23–2.16), and the number of consecutive days on PEP (RR = 0.77, 95% CI, .66–.90).

Predictive Value of Self-reported Symptoms With Regard to Seroconversion

Diarrhea, headache, and myalgia were the most frequently reported symptoms among all exposed persons (Table 2). A difference in frequency among persons who did and did not seroconvert was most evident with regard to periorbital edema, fever $\geq 38^\circ\text{C}$, and myalgia as indicated by high prevalence ratios. Symptoms that typically occur soon after infection (in the gastrointestinal phase of the disease) such as diarrhea and abdominal pain were reported almost as often by persons who did not seroconvert as by those who did seroconvert and thus had a low predictive value for seroconversion.

DISCUSSION

We report on a large outbreak of trichinellosis in Germany associated with wild boar meat and products thereof involving at least 14 cases of trichinellosis and possibly several hundreds of exposed persons. We found that among those who received anthelmintic drugs as PEP, cases of trichinellosis exclusively occurred among persons who started PEP later than 6 days post-exposure. The occurrence of (early) gastrointestinal symptoms following exposure carried no predictive value for later seroconversion in our study.

This outbreak confirms that game and meat from animals raised outside of controlled housing conditions continues to pose a risk to consumers [21], especially in cultures with affinity to eat

raw meat preparations and raw sausages. Recent outbreaks due to consumption of such foodstuffs occurred in Poland [22], Romania [23], Lithuania [24], Italy [8], and in the United States in Illinois [7] and Iowa [6]. The present outbreak underlines that meat from a single infected wild boar carcass can contaminate a large batch of meat products and thus put hundreds of people at risk of disease if it is not heated before consumption. Roasting the infected wild boar meat was apparently sufficient to prevent *Trichinella* infections. Diligent food safety inspections of potentially *Trichinella*-contaminated meat are implemented in Germany [19] and would have normally prevented this outbreak. Additional quality assurance measures have been implemented to assure reliable results from meat inspection.

The contaminated sausages were typically eaten on the day of purchase or the day after. The viability and infectivity of larvae was thus likely not reduced due to the passing of time as demonstrated from previous studies [25]. Congruent with this finding, we found a strong dose relationship between the number of sausages consumed and the development of trichinellosis. Seroconversion after infection with *Trichinella* spp. usually occurs between the 2nd and 5th week and is inversely correlated with the infective dose [26]. Most persons who were seropositive 6 weeks after exposure showed a low initial titer for anti-*Trichinella*-IgM and -IgG. This can be explained by a prolonged seroconversion due to the relatively low number of larvae (30–130) per sausage.

This study is to our knowledge the first to prospectively evaluate the effectiveness of post-exposure prophylaxis in an outbreak of trichinellosis. In the multivariable analysis, delaying administration of PEP was an independent risk factor in addition to the number of sausages eaten and a shorter duration of

treatment. The distribution of cases and noncases by time of PEP administration as shown in Figure 2 reveals a sharp cut-off with cases exclusively occurring in the group that received PEP 6 days post exposure or later. Although there is controversy concerning the effectiveness of mebendazole during the muscular phase of trichinellosis [27, 28], our data thus suggests that if taken within the first 6 days of exposure the development of clinical trichinellosis in humans can effectively be prevented. Supporting evidence comes from several animal studies [11–13] and a case series involving 4 persons exposed to a high dose of *Trichinella* larvae treated with mebendazole within 48 hours of exposure [14]. It is assumed, that such early application of anthelmintics kills larvae and adult forms in the gut before new larvae are produced and migrate to other parts of the body. Mebendazole may be particularly suitable for the use in PEP as (a) it is poorly absorbed in the gastrointestinal tract, leading to a high concentration in the lumen [29], and (b) it was shown to be highly effective against intestinal stages of the parasite [14]. Albendazole, on the other hand, may be more useful should the larvae have already migrated and have become established in the muscles [30]. Randomized clinical trials comparing different treatments are scarce, and the small number of persons in our study that were administered albendazole does not allow us to recommend one drug over the other.

We would like to emphasize that the results of this study do not suggest anthelmintics should be withheld if a patient presents more than a week after exposure. In trichinellosis, the severity of extraintestinal disease is proportional to the number of larvae produced by adult worms in the small intestine. Thus, it is imperative to rid the gut of adult parasites even if the patient presents late and with only mild gastrointestinal symptoms [31].

A post-exposure prophylaxis recommendation carries the risk of administering drugs to and causing adverse effects in persons that are not infected and would not have needed treatment. We thus analyzed whether early symptoms could be used as a predictor for seroconversion on which a decision to administer PEP could be based. We found that periorbital edema and myalgia are distinctive clinical signs of trichinellosis, indicated by high prevalence ratios between persons that did and did not seroconvert, yet they only occur after larvae have already disseminated from the gastrointestinal tract. Fever and chills were predictive as well, yet these symptoms are unspecific. Early symptoms like diarrhea and abdominal pain linked to invasion of the intestinal mucosa by the parasite had little or no predictive power for later outcome in our outbreak setting.

CONCLUSIONS AND RECOMMENDATIONS

Mebendazole can apparently prevent the development of clinical trichinellosis if administered within the first 6 days of alimentary exposure. We therefore recommend that PEP is offered to all

persons with exposure to meat containing viable *Trichinella* larvae in a similar outbreak setting. Due to the unspecific nature of the early symptoms of trichinellosis, the decision to recommend or withhold PEP should not be based on symptoms but on potential exposure. The key measures to prevent human trichinellosis, however, remain the prevention of *Trichinella* infection in livestock and the systematic sampling and examination of potentially infected meat intended for human consumption.

Notes

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Potential conflicts of interest. All authors: No reported conflicts.

All authors have submitted the ICMJE Form for Disclosure of Potential Conflicts of Interest. Conflicts that the editors consider relevant to the content of the manuscript have been disclosed.

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