Unexpected intracranial location of a *Cephenemyia stimulator* larva in a roe deer, *Capreolus capreolus*, revealed by computed tomography

Inesperada ubicación intracraneal de una larva de *Cephenemyia stimulator* en un corzo, *Capreolus capreolus*, detectada mediante tomografía computerizada

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**Abstract**

In this study we describe the finding of a *Cephenemyia stimulator* larva in the brain of a roe deer (*Capreolus capreolus*) after performing a computed tomography (CT) scan of its head. Despite this anatomical location of oestrid larvae could be relatively frequent in other genera, such as *Oestrus*, to our knowledge, this is the first reported case involving the genus *Cephenemyia*. Concretely, a second-instar *C. stimulator* larva was found in the basis of the cranium. The location of a macroscopic hemorrhagic lesion involving the brain parenchyma peripheral to the location of the larva suggests that tissue colonization occurred before the animal was hunted. Since no detectable alterations or damage to the cranial bones were observed, we suggest a possible larval migration route drilling the skull bones. Finally, we propose the use of the term “neuromyiasis” to be referred to the invasion of the central nervous system by dipteran larvae, particularly oestrids.

**Keywords**: *Cephenemyia stimulator*, cerebral myiasis, computed tomography, neuromyiasis, roe deer

**Resumen**

En este estudio describimos el hallazgo de una larva de *Cephenemyia stimulator* en el cerebro de un corzo (*Capreolus capreolus*) tras realizar una tomografía computerizada (TC) de su cabeza. Aunque esta localización anatómica de larvas de Oéstridos puede ser relativamente frecuente en otros géneros, como *Oestrus*, que sepamos, este es el primer caso que involucra al género *Cephenemyia*. Concretamente, una larva de segundo estadio de *C. stimulator* se encontró en la base del cráneo. La localización de una lesión macroscópica hemorrágica que afectaba al parénquima cerebral periférico a la ubicación de la larva sugiere que la colonización del tejido se produjo antes de que el animal fuese abatido. Dado que no se detectaron alteraciones o daños en los huesos craneales, sugerimos una posible ruta de migración larvaria a través de perforaciones de los huesos del cráneo. Finalmente, proponemos el uso del término “neuromiasis” para referirnos a la invasión del sistema nervioso central del hospedador por larvas de dipteros, particularmente Oéstridos.

**Palabras clave**: *Cephenemyia stimulator*, corzo, miasis cerebral, neuromiasis, tomografía computerizada
Introduction

Cephenemyia stimulator (Clark, 1815) causes naso-pharyngeal myiasis in roe deer, Capreolus capreolus (Linnaeus, 1758), throughout the Palaearctic (Colwell et al. 2006). This oestrid species has unusually been found infesting red deer, Cervus elaphus Linnaeus, 1758 (Király & Egri 2004) and an uncommon infection by the moose throat bot fly, C. ulrichii, in a roe deer has also been reported in Finland (Nilssen et al. 2008). Cephenemyia stimulator larvae usually develop closely each other within a single or various pouches in the naso-olfactive area and, with certain frequency, in the oesophageal and respiratory organs of the host, such as trachea and lungs (bronchioles) (Dudzinski 1970, Bernard & Biesemans 1975). C. stimulator larvae have also been found in the eustachian tube, in the arynoid cavity and near the hypophysis of a parasitized roe deer (Ullrich 1938) and might occasionally be swallowed and pass through the digestive tract of the host (Blickle 1956).

Larval migratory routes of first-instar Hypoderma spp. larvae include mainly connective tissues and nerves (Colwell 2006). An intracranial myiasis in a horse caused by a first-instar Hypoderma bovis (Linnaeus 1758) larva was associated with incoordination of gait, circling to the left, head tilt to the right, partial paralysis of the face, impaired vision and, after necropsy, with haemorrhage and oedema in the brain tissue close to the larva (Hadlow et al. 1977).

Aberrant migration of Cuterebra larvae into the central nervous system of cats and dogs were also described (Cook et al. 1985, Sartín et al. 1986, Glass et al. 1998). Histopathological findings included presence of parasitic track lesions, superficial laminar cerebrocortical necrosis, cerebral infarction, subependymal rarefaction and astroglisis and subpial astrogliosis. These features were related to the feline ischemic encephalopathy (Glass et al. 1998).

With regards to human hosts, several fatal cases of cerebral myiasis caused by the warble fly, Dermatobia hominis (Linnaeus Jr. In Pallas, 1781), were described (Dunn 1934, Rossi & Zucoloto 1973). Such cases involved young patients (children aging less than 2 years) and Dermatobia larvae, which commonly cause cutaneous myiasis in humans, entered the cerebral cavity through the bregmatic fontanelle. One case of intracerebral myiasis due to a Hypoderma bovis larva was diagnosed in an 8 yr-old child after detecting the hematoma produced by the larva by means of computed tomography (Kaleioglu et al. 1989).

In Europe, Cephenemyia stimulator was described in the early XIXth century and its veterinary importance is known since the first half of the XXth (Ullrich 1938, Zumpt 1965, Dudzinski 1970). It has been reported from Austria (Kutzer 2000), the Czech Republic (Salaba et al. 2013), Estonia (Jõgisalu 2010), Fennoscandia (Norway, Sweden, Finland and Denmark) (Stéen et al. 1998), France (Maes & Bullard 2001), Germany (Nickel et al. 1986), Hungary (Sugár 1974), Italy (Rivosecchi et al. 1978) and Poland (Drozdz 1961).

The first cite of C. stimulator in Spain comes from Ciudad Real and is recent (Notario & Castresana 2001). In northwestern Spain, this parasite was found by the first time in 2005 (Arias et al. 2016). In 2011 and 2012 it was reported parasitizing roe deer in Cataluña (de la Fuente 2014) and Extremadura (Calero-Bernal & Habela 2013), respectively. This oestrid has also been collected from roe deers from Cantabria and País Vasco (Arias et al. 2014). In Galicia, Asturias and León, seropositive animals were detected since 2007 and the seroprevalence for the period 2007-2014 reached 38% (Arias et al. 2016). In an epidemiological survey on cephenemyiosis in roe deer in Galicia, based on direct diagnosis, prevalence was slightly lower: 31% (López-Beceiro et al. 2015). Within this context, the computed tomography (CT), as a no-invasive technique, was adapted to detect Cephenemyia larvae within intact (not necropsied) roe deer heads (Fidalgo et al. 2015).

This work describes a case with an unusual location of a Cephenemyia stimulator larva in the brain of a roe deer, which was first detected with the aid of the CT.

Materials and methods

Material used in this study was the head of an adult male roe deer, Capreolus capreolus, (4 yr-old), which was selectively hunted (trophy) in 27th June 2012 in Santa Colomba de Somoza Council (León province, northwestern Spain). This animal was repeatedly observed during the two previous months. When shot, the animal was in good condition and showed no signs of abnormal behaviour and/or locomotion. The head was removed, introduced in
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Results and Discussion

The morphology and size of collected larvae fitted the descriptions of *Cephenemyia stimulator* given by Zumpt (1965) and Bernard & Biesemans (1975). Sequences obtained from positive clones (GenBank accession numbers: MG763915 and MG763916) were practically identical each other and reached a 99.4 % of identity with regards to the COI sequence for *C. stimulator* available at the GenBank.

The fact of finding *C. stimulator* larvae of different instars simultaneously in the same host could be explained by the production of several larval generations per year (Dudzinski 1970), by the ability of first-instar larvae to become hypobiotic and overwinter into the host head cavities and/or also by an asynchronous development of each larva (Colwell et al. 2006). In our case, only one larva was found associated with macroscopic hemorrhagic lesions of the surrounding brain tissue, but no clinical nervous signs in the animal before being shot and, as could be expected (Rossi & Zucoloto 1973). This ectopic location without apparent signs of disease could be explained by a very recent migration of the larva or even by a *post-mortem* migration. Dudzinski (1970) suggested that, if necropsy is delayed with regards to the host death, then larvae can move within the head cavities and reach a “random” distribution. In our case, the short time elapsed between sample collection and CT analysis, and the fact that during this period it was maintained at 4 ºC, make larval movement very unlikely. Histologic variation of lesions caused by *Hypoderma lineatum* suggests that larval migration in the horse brain can last several days (Olander 1967). In fact, we found a “normal” location of the 8 third-instar larvae and an absence of larvae in the proximal section of the trachea and esophagus. On the other hand, the presence of surrounding hemorrhagic lesions is associated with active blood circulation through the vascular system (Brooks 2016).

Moreover, the ability of the larva to reach the cerebral cavity without causing damage to cranial bones is noteworthy, in particular taking into account that it was a second-instar larva, with a relatively large size (> 4 mm in diameter). Possible larval migration routes include the labyrinthus ethmoidalis, through the meatus ethmoidalis to reach the cribriform plate of the ethmoides, which delimitates the nasal and cranial cavities each other. This is a very thin plate containing numerous orifices. After passing this plate, the larva could have followed the sub-dural via to reach its final location.
If so, and even in the case of one or several nervous connections become damaged, few or none host behavioural signs would be expected both at short or medium-long term. Anyway, this hypothesis implies certain ability of the larva to perforate soft tissues.

When possible, the methods commonly used to diagnose cephememyiosis in roe deer and other hosts, would benefit from being complemented with observational studies of the host behaviour in order to address the frequency of the cases like that described here (which was the only one of 75 scans made). Finally, we want to emphasize that the brain is not usually sampled in necropsies of ungulate heads for collecting oestrid larvae. Therefore, the use of computerized tomography can become very useful for determining the real frequency of this “aberrant” larval location in this and other hosts, even when still alive.

In conclusion, despite this is the first reported case in which a Cephenemyia stimulator larva has been found within the host brain, the occurrence and implications of the invasion of the cranial cavity by larvae of Oestridae species underline the need for a reappraisal of our understanding of host-myiasis interactions. For cases involving the invasion of the central nervous system by dipteran larvae, we propose the use of the term “neuromyiasis”.

![Figure 1. A) Sagittal computed tomography scan of the roe deer head. Arrow points a well-delimitated ovoid structure (a third-instar oestrid larva). B) Axial computed tomography. Arrow points well-delimitated ovoid structures (third-instar oestrid larvae).](image-url)
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Figure 2. A) Axial computed tomography scan of the roe deer head. Arrow points soft-tissue and well-delimited ovoid structures in the cranium basis, at the level of the middle fossa, with small radio-transparent foci. In this location, a second-instar *Cephenemyia stimulator* larvae was found after necropsy. B) Sagittal computed tomography scan. Arrow points soft-tissue and well-delimited ovoid structures in the right side of the cranium basis, with small radio-transparent foci. In this location, a second-instar *Cephenemyia stimulator* larvae was found after necropsy.

Figure 3. Location of a second-instar *Cephenemyia stimulator* larva in the host brain of the roe deer after necropsy. The parenchyma of the brain peripheral to the location of the larva appeared embedded in hemorrhagic fluid.
Acknowledgements
This study was funded by the Spanish Federation of Hunters (FEC), the Foundation for the Study and Defence of the Nature and Game (FEDENCHCA), and the National Federation of Hunters (France) (Project: FNC-PSN-PR5-2013). The research activities of NC, AS and JMP are partially supported by the Junta de Andalucía, Plan Andaluz de Investigación (BIO-220 and RNM-118 groups). The procedures used in this study were carried out in compliance with Spanish legislation on animal experimentation and welfare.

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Submitted: 5 November 2020
Accepted: 14 February 2021

Associate editor was Francisco Ruiz Fons