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on a larva of fire salamander (*Salamandra
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Predatory instance of a horse leech (*Haemopsis* sp.) on a larva of fire salamander (*Salamandra salamandra*)

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Abstract. Amphibians face severe global decline, making a comprehensive understanding of their ecological interactions essential for effective conservation planning. Among these interactions, leeches (Hirudinea) play an intricate role, acting both as ectoparasites and macrophagous predators. While the parasitic interactions between these taxa have been largely explored, the role of leeches as active predators of amphibians remains less documented, especially for caudate taxa. Here, we report a predation event of a horse leech (*Haemopsis* sp.) on a larva of fire salamander (*Salamandra salamandra*) (Linnaeus, 1758) taking place in northern Italy. To our knowledge, this is the first scientific record involving a member of the genus *Salamandra*, and potentially it may represent the first documented case of leech predation on a caudate larva. This observation thus highlights a potentially overlooked trophic interaction that may contribute to larval mortality and population dynamics.

Amphibians represent one of the most threatened groups of vertebrates worldwide (Stuart et al. 2004). In order to conserve current amphibian diversity, in addition to targeting anthropogenic factors, it is necessary to better understand the trophic relationships that exist between amphibians and other taxa (San Sebastián et al., 2015; Wickramasingha et al., 2026).

Among them, leeches (Hirudinea) trophically interact with amphibians in multiple ways across both aquatic and terrestrial habitats (Herczeg et al., 2021). Most commonly, leeches assume the role of ectoparasites, attaching to adult amphibians for feeding purposes (Lunghi et al., 2018). For example, in a survey conducted across five counties in southern England, Seilern-Macpherson and colleagues recorded over 40 instances of ectoparasitism on *Bufo bufo* (Linnaeus, 1758) and *Rana temporaria* (Linnaeus, 1758) over a three-year period, with the introduced *Batracobdella algira* (Moquin-Tandon, 1846) being the most frequently recorded leech species (Seilern-Macpherson et al., 2024). While these parasitic interactions are usually regarded as non-lethal for the hosts (Kutschera et al., 2010; Bull, 2019; Seilern-Macpherson et al., 2024), negative impacts on population trends have been suggested (Stead and Pope, 2010; for review see Merilä and Sterner, 2002), and direct mortality events have been recorded (Merilä and Sterner, 2002). Moreover, the presence of the parasites is likely to impact the amphibians' fitness and may cause the transmission of pathogens (Berven and Boltz, 2001; Raffel et al., 2006; Starzecka et al., 2020; Hernandez-Caballero et al., 2022). Aside from being ectoparasites, different genera of leeches are macrophagous predators of amphibians, known to feed on amphibians at various life stages, such as eggs, larvae, and juveniles (Laurila et al., 2002; Romano and Di Cerbo, 2007; Stead and Pope, 2010; Hettyey et al., 2011; De Luna et al., 2024; Seilern-Macpherson et al., 2024).

While macrophagous predation on anuran and caudate eggs is equally reported, data regarding predation events on caudata larvae or juveniles are currently lacking. Here, we report a potential predation event of a horse leech (*Haemopsis* sp.) on a fire salamander (*Salamandra*

52 *salamandra*) larva. To our knowledge, this represents the first published record involving a
53 member of the genus *Salamandra* and may constitute the first record of predation on a caudata
54 larva.

55 On 23 January 2023, at 22:06 circa, in Carate Brianza, Italy (45.67 N, 9.25 E, 300 m
56 a.s.l.), we observed a large individual of horse leech (*Haemopsis* sp.) in the process of
57 swallowing a larva of fire salamander (*Salamandra salamandra*) (Fig.1). The event took place
58 in a small freshwater rivulet, flowing from the overflow of a spring-fed stone lavoire. When we
59 started observing the scene, the leech had swallowed most of the amphibian, of which just the
60 gills and head were visible. The leech then took around 10 minutes to swallow the rest of the
61 prey. Because we didn't witness the leech attacking the amphibian larva, we cannot exclude
62 that this may represent a scavenging event. However, there are a couple of clues that point
63 against this latter scenario: (i) one of the authors (AVP), accurately surveyed the exact location
64 where the event took place in search of salamander larvae around 30-45 minutes earlier and
65 did not notice any dead animal; (ii) the larva did not show any evident sign of decay compatible
66 with being dead before consumption. Therefore, it is likely that the leech actively preyed upon
67 and swallowed the salamander larva, potentially facilitated by the shallow water level of the
68 rivulet (around 5 cm) and the high density of larvae.

69 The horse leech was identified as belonging to the genus *Haemopsis* based on external
70 morphological features: a dorsoventrally flattened, elongate body up to 10 cm in length, smooth
71 dorsum lacking papillae, a dark brown-black dorsum with scattered black markings, and the
72 absence of the characteristic marginal red-orange stripes typical of *Limnatis*, the reddish
73 colouration pattern of the dorsum characteristic of *Hirudo*, or the turbercles peculiar to some
74 species of the genus *Trocheta* (cf. Nesemann & Neubert, 1999). The anterior sucker was
75 notably broad, and the posterior sucker was well developed, both, coupled with other

characters, consistent with *Haemopsis sanguisuga* and related species. Since the leech was not collected, here we prefer to report it as *Haemopsis* sp.

The prey was identified as a larva of *Salamandra salamandra* based on a combination of diagnostic characters: robust body form and broad head with external gills consisting of three distinct, red-tinged tufts on each side. Multiple years of non-standardised surveys failed to recover any other caudata species in the proximity of the lavoir, however, other caudate species present in nearby areas (e.g., *Lissotriton vulgaris*, *Triturus carnifex*) differ by having more slender larvae, smaller heads, and reduced gill tufts, thus excluding misidentification.

Predatory interactions between invertebrates and amphibians are likely underreported, particularly those involving caudate larvae. Amphibians, including salamanders and newts, are known hosts of a wide range of parasitic and predatory invertebrates, such as nematodes (e.g., *Rhabdias* spp., *Cosmocerca* spp.), trematodes, crustaceans (*Heterocypris incongruens*) and free-living planarians, which may act either as parasites or opportunistic predators (Ottonello and Romano, 2011; Koprivnikar et al., 2012; McAllister and Bursey, 2012). Planarians of the genus *Schmidtea* have been observed preying upon amphibian eggs and early-stage larvae (Segev et al., 2015), while nematodes may directly impair host health and survival, influencing both growth and metamorphic success (Kiesecker, 2002; Johnson and McKenzie, 2009).

Such multi-trophic relationships highlight the ecological complexity of freshwater communities, where amphibians and their larvae are simultaneously prey, host, and competitor. In this context, leeches such as *Haemopsis* spp. may act both as occasional scavengers and active predators, especially under high prey density or restricted water conditions. The observation presented here enriches the understanding of this overlooked dynamic, suggesting that leeches may contribute to natural regulation of amphibian larval populations, as reported for other invertebrate-amphibian systems (Shulse et al., 2013; Arribas et al., 2014). Even further,

understanding the intricate relationship between parasitic leeches and amphibian hosts may contribute in disentangling the complex evolutionary history of amphibians (Torres-Sánchez et al., 2026).

Despite being quite widespread, the fire salamander is currently classified as vulnerable under the IUCN Red List criteria (Crnobrnja-Isailović et al., 2025) and has undergone a strong decline in most of its northern distribution, partly due to the lethal pathogenic fungus *Batrachochytrium salamandrivorans* (Martel et al., 2013; Dondero et al., 2023). In Italy, *S. salamandra* is currently classified as Least Concern under the IUCN Red List's criteria (Rondinini et al. 2022). However, the presence of a new emerging and enigmatic cyst-causing pathogen has been reported in populations geographically close to the one here described (< 7km) (Manenti et al., 2022). Moreover, anthropogenic pressures such as habitat fragmentation and alteration represent critical threats to the survival of salamander populations (Andreone and Luiselli, 2000; Bani et al., 2015; Pisa et al., 2015). For these reasons, a deeper understanding of the trophic relationship which interests this iconic amphibian is fundamental in order to better assess its ecological requirements and thus improve conservation measures.

At the site where the observation took place, one of the authors (XXX) observed over the year an interesting trend regarding both salamander larvae and leech populations. The population density of leeches seems, in fact, to be correlated with the salamander larvae density, with the number of leeches present within the lavoire and the adjacent rivulet dropping exponentially after the larvae have completed their development process and left the water environment. While these observations lack any strong quantitative data, an overlooked ecological relationship between these taxa may occur. For this reason, future research efforts may be necessary for understanding how these invertebrates may be able to impact and regulate salamander populations, as it has been suggested for other amphibian species (Raffel et al., 2006; Stead and Pope, 2010).

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236 **Fig. 1.** Dorsal (A) and lateral (B) view of an individual of horse leech (*Haemopsis* sp.) while
237 swallowing a larva of a fire salamander (*Salamandra salamandra*). The arrows highlight
238 different regions of the salamander and leech bodies.

239

