The situation of the Genus *Trogulus* (Arachnida - Opiliones - Trogulidae) in Belgium

Luc Vanhercke[1] & Hay Wijnhoven[2]

[1]Emile Poetoustraat, B-9030 Mariakerke, Belgium.
[2]Groesbeeksedwarsweg 300, NL-6521 DW Nijmegen, The Netherlands.

Summary
Thanks to recent research a more reliable identification of some Trogulus *species, in particular* T. nepaeformis s.l.*, can be done. Specimens from the collection of the Royal Belgian Institute of Natural Sciences have been reviewed and many new samples in our own collection were studied. Three species are recognized:* T. tricarinatus, T. closanicus *and* T. nepaeformis*. Provisional maps are drawn based on the material aforementioned and some observations about the phenology are given.*

Résumé
Grace à des études menées récemment, il est devenu possible d'identifier de manière plus fiable certains espèces du genre Trogulus*, en particulier les* T. nepaeformis s.l*. Matériel des collections de l'Institut royal des Sciences Naturelles de Belgique à été re-examiné et beaucoup d'échantillons supplémentaires en notre possession ont été pris en considération. Trois espèces sont confirmées pour la Belgique:* T. tricarinatus, T. closanicus *et* T. nepaeformis*. Des cartes de distribution provisoires, basées sur le matériel étudié, sont fournies et quelques observations sur la phénologie sont présentées.*

Samenvatting
Dankzij recent onderzoek is het mogelijk geworden om sommige Trogulus *soorten, in het bijzonder* T. nepaeformis s.l.*, betrouwbaarder te identificeren. Materiaal uit de collecties van het Koninklijk Belgisch Instituut voor Natuurwetenschappen is opnieuw bekeken en heel wat nieuwe stalen uit onze eigen collectie zijn bestudeerd. Drie Belgische soorten worden onderscheiden:* T. tricarinatus, T. closanicus *en* T. nepaeformis*. Voorlopige verspreidingskaartjes op basis van de gecontroleerde specimens worden gegeven en er is ook aandacht geschonken aan de fenologie.*

Introduction

Opiliones of the genus *Trogulus* are notoriously difficult to identify. The external morphology is extremely homogeneous. Species were separated by small differences in size, the distance between the eyes, the segments of tarsus II, and the genital morphology. To make matters worse, most species described during the last decennia rather obscured the situation because previously described similar species were not re-characterised carefully enough (SCHÖNHOFER & MARTENS, 2009). That is why we in earlier work (VANHERCKE, 2010; WIJNHOVEN, 2009) choose not to try to guess which species exactly were in play, but rather prefered to indicate that we weren't sure yet by giving specimens, not being identified as *Trogulus tricarinatus* (Scopoli, 1763), the adverb *sensu lato*, i.e. *Trogulus nepaeformis* s.l.

MARTENS (1978) in his magisterial work that covered a huge part of Central and Western Europe, considered *Trogulus nepaeformis* (Scopoli, 1763) and *T. closanicus* (Avram, 1971) as synonyms. He based his decision on the variability in the former species and on the author's drawings of the latter species' description. Soon thereafter, however, and still in 1978, it was concluded, based on material from Romania, that *T. closanicus* was a valid species (SCHÖNHOFER & NOVAK, 2011).

Later on, CHEMINI (1984) agrees with the difficult systematics in *Trogulus* and he managed to differentiate between *T. closanicus* and *T. nepaeformis* based on external morphologic characteristics together with the morphology of the penis. He in fact re-described *T. closanicus* and illustrated the chief morphological characters distinguishing both species.

Schönhofer in his dissertation (SCHÖNHOFER, 2009) gave a key to some specific species groups of *Trogulus* and in an appendix also a preliminary (because of as yet undescribed taxa) key to the genus in general.

During the processing of a batch of pitfall captures originating from the southern part of The Netherlands it was observed that the specimens identified as *T. nepaeformis* probably belonged to two species. It became clear that Schönhofer's work would have repercusions on the situation of *Trogulus* in The Netherlands and that the collected material would need to be revised (WIJNHOVEN, 2008).
Eventually, both *T. closanicus* and *T. nepaeformis* were confirmed for The Netherlands (WIJNHOVEN *et al.*, 2014) and in the same article a key with additional pictures and drawings was given in order to make identification of new finds easier.

In our overview of the Opiliones of Belgium (VANHERCKE, 2010) we wrote that we most likely also would have to deal with two different species instead of one. We also suggested that all new finds of *T. nepaeformis* s.l. were preserved to allow for a revision later on. In this article we report on the results of such a review.

Material and approach

Our personal database, which, at the time of writing, contains more than 17 056 recordings coming from 202 different UTM squares (10×10 km), contains 640 occurrences of *T. nepaeformis* s.l. which were identified before the time that we were aware that two (or more) species could be involved. As an occurrence is defined as one or more specimens of a certain species found together at a certain place and time, these records in the database stand for many more individuals: 1 994 to be precise.

Back then we did not keep all the finds. Usually we took only a few individuals from each site, or each station when several were installed, and these were then submitted to the collections of the Royal Belgian Institute of Natural Sciences in Brussels. Therefore, at least a substantial fraction of the older samples should be recoverable. When we asked the curator of the collections it did not take long before we had all the specimens which were identified as *T. nepaeformis* at our disposal for review. About 60% (or 388) of the older occurrences equivalent to about half (1 055) of the individuals could be re-identified. The remaining 252 occurrences, which could not be reviewed relate to 17 different UTM-squares. Fortunately we have more recent *Trogulus* material for all of them, except for only three squares (ES36, FR39, FQ78) of the 202 squares for which we do have captures of Opiliones, but no new finds of the species couple under study.

After we learned about the identity problem, all captures of *Trogulus* were kept aside and their identification deferred. In this way we could make sure that all the material would be available once we were ready to tackle the problem. Eventually 711 new occurrences (or 1 945 ind.) could be inserted into our database. This makes for 1 099 occurrences standing for 3 000 individuals (see Table 1) which were reviewed.

Almost all data in our personal database originate from pitfall captures. They stand for 15 193 of the 17 056 recordings. The remaining captures (1 863 recordings) were done with several methods of which coloured pan trap (460 occurrences), captures by hand (312 occ.), Malaise trap (184 occ.) and beating (112 occ.) are the most prominent. Only 7 of those 1 863 recordings of not pitfall captures concern a *Trogulus* species.

For *Trogulus* said database contains 1 092 (out of 1 099, see Table 1) occurrences or 2 987 (out of 3 000) individuals captured with pitfalls. The tiny remaining fraction was captured by hand, with Kaila trap, or coloured pan trap.

For the identification mainly the characteristics given in WIJNHOVEN *et al*. (2014) were used, but also CHEMINI (1984), MARTENS (1978), and SCHÖNHOFER (2009) were consulted in order to make sure that we did not miss

any unexpected other species. The penis of all males was checked and for the females, apart from the eyes, also the tarsus was looked at. Four individuals were damaged and the sex could not be determined. We did, however, tried an identification based on the available characteristics. They are reported as adults in Table 1. Many voucher specimens were prepared for deposit in the collections of the Royal Belgian Institute of Natural Sciences.

The results

Our personal database contains Opiliones recordings from 202 different UTM-squares (10×10 km) of the 384 which cover Belgian territory. When we reported previously about Opiliones in Belgium (VANHERCKE, 2010) we had data from only 170 squares, so our coverage is slowly improving. The number of specimens recorded of all species together has risen from 42 730 back then to 72 007 today.

With 3 000 specimens (see Table 1) distributed over three species *Trogulus* make only a small fraction of the dataset. Because the species are often found together, the total number of squares with members of the genus is less than the sum of the numbers of each species: in only 55 squares (14% of all Belgian squares) did our pitfalls yield one or more *Trogulus* species. Compare this with the most widespread and numerous species in Belgium, *Rilaena triangularis* which was catched in 138 UTM-squares (36%) and present in the database with 10 324 individuals (14%). Of all the other species found in Belgium, 13 are present in more squares than the most common *Trogulus* (*closanicus)* and 11 are more numerous. Therefore as a rule *Trogulus* stand only for a small fraction of any of our pitfall yields.

Occasionally however one pitfall fortnight may yield remarkably high numbers of *T. closanicus* and sometimes of *T. nepaeformis* also. Our personal database contains recordings where the former species was found with up to 104 individuals in one catch. The numbers of the latter species reach never more than 20 but even that is still much more than the usual one or two. The same phenomenon is also reported from The Netherlands (WIJNHOVEN *et al.* 2014).

Because some of the revised identifications resulted into a few new recordings of *T. tricarinatus* we will take this species also into account in our further discussion.

Table 1: General results of the revision of the recordings of *Trogulus* captured in Belgium. (adults: see Material and approach)

	occurrences	**# males**	**# females**	**# adults**	**# specimens**	**# UTM squares**
T. tricarinatus	80	21	101	1	123	21
T. nepaeformis	330	368	260	1	629	33
T. closanicus	689	1 634	612	2	2 248	40
	1 099				**3 000**	

The number of occasional observations in our database is near to none. Almost all our recordings originate from surveys which lasted from at least one year up to more than 20 years at some places. Most of those surveys are conducted at several stations of which the exact position is known and the environmental characteristics are documented: e.g. 6 stations at De Haan (DESENDER, HUBLÉ & VANHERCKE, 1982), 55 stations for Macro-invertebrates on forest floors (DE VOS, 1998), 9 stations at Viesville (BAERT, VANHERCKE & LIMBOURG, 2014).

In our recordset at least one *Trogulus* species was found in 175 different stations. The exact localisation of the sampling stations allows us to examine where exactly each species was found and what species were catched at the same spot (Table 2).

Table 2: The number of pitfall stations (n=175) where each *Trogulus* species was found put against the number of stations that each species shared with another *Trogulus*.

	no other	***T. tricarinatus***	***T. nepaeformis***	***T. closanicus***
T. tricarinatus	18	30	11	7
T. nepaeformis	40		98	54
T. closanicus	57			113
all three together	6			

In 115, that is 65% of all stations, only one of the species under study was found (Table 2). In the remaining stations, two species and in six cases all three were found together. For *T. closanicus*, *T. nepaeformis and T. tricarinatus* the fraction of pitfall stations where each species was found alone, is about 50%, 41% and 60% respectively. Therefore the first two species where in at least half of the pitfall stations found syntopic with another *Trogulus*. For *T. tricarinatus* a slightly lower fraction of 40% was noted. In 11 of these stations *T. tricarinatus* was found accompanied by *T. nepaeformis* and only in 7 by *T. closanicus*.

Trogulus closanicus

According to our data, this is the most widespread and numerous of the three species. *T. closanicus* was found in 113 of the 175 pitfall stations where at least one *Trogulus* was recorded (Table 2). On a Belgian scale, however, the species is found in barely 40 squares (Figure 1) and we have only 2 248 specimens recorded which is 10% of all squares and 3% of all captures respectively.

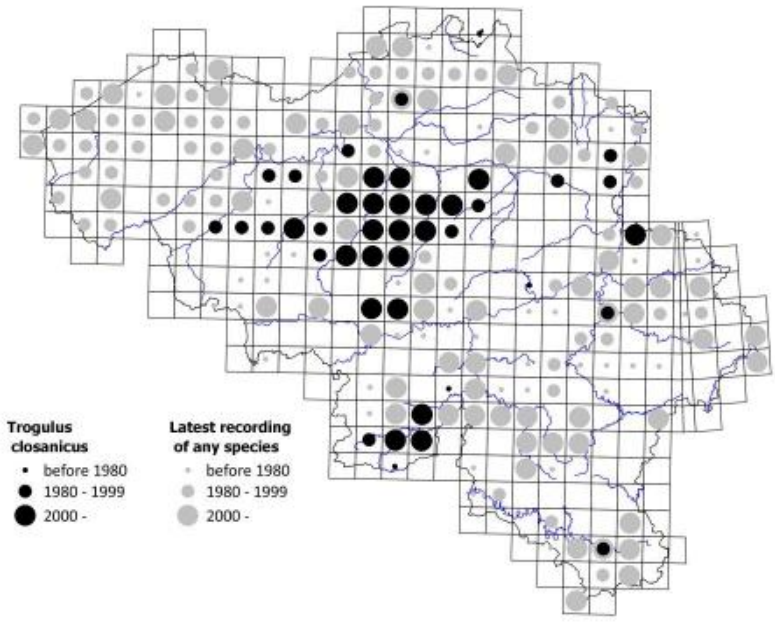

Figure 1: *Provisional map of the occurrence of* T. closanicus *in Belgium. Map is based on the reviewed material only. Grey dots give UTM-squares (10×10 km) where Opiliones were captured.*

According to several authors, *T. closanicus* lives in woods, groves and also in grassland (KOMPOSCH & GRUBER, 2004; MUSTER & MEYER, 2014; WIJNHOVEN *et al*. 2014). For Austria (KOMPOSCH & GRUBER, 2004) the species is

reported to be a trifle more thermophilic than *T. nepaeformis* who tends towards sligthly more colder and more humid conditions. For Luxembourg MUSTER & MEYER (2014) found *T. closanicus* in much higher densities in semiarid grassland than in woodland and they also report that *T. nepaeformis* was exclusively found in woods and copses. In The Netherlands the species is found mainly in deciduous woods, but is also reported from calcareous grassland (WIJNHOVEN *et al*. 2014).

T. closanicus is often found to live syntopic with *T. nepaeformis* (CHEMINI, 1984; KOMPOSCH & GRUBER, 2004; WIJNHOVEN *et al*., 2014). In our dataset with occurrences coming from 175 pitfall stations dispersed over the country *T. closanicus* is present in 113. In 54 of the sites (48%) the species was found syntopic with *T. nepaeformis* and in 7 sites (6%) together with *T. tricarinatus.*

To the Northeast its range continues in The Netherlands where the species is restricted to the most southern half of the border with Germany (WIJNHOVEN *et al*., 2014). The species has been reported from Luxembourg, where it is the most common *Trogulus* (MUSTER & MEYER, 2014), and in Germany from the South to the far North, near Hannover (ARACHNOLOGISCHE GESELLSCHAFT, 2017).

It is remarkable that our dataset contains no occurrences of *T. closanicus* west from the River Scheldt. Several pitfall surveys were done at several places between River Scheldt and the Belgian coast. In at least some of them —e.g. 6 stations in WijnendaleBos (POLLET & HUBLÉ, 1987), 11 stations for the macro-invertebrates on forest floors (DEVOS, 1998)— the environmental characteristics match the preferences of *T. closanicus*. Moreover, both other *Trogulus* were present in the catches at those stations (Figure 3 and 6). The habitat preferences of *T. closanicus* and *T. nepaeformis* differ only slightly and both species are in about 50% of our pitfall stations syntopic. Our data suggest that *T. closanicus* is not present west from the River Scheldt. In Belgium, we are probably looking at the north-western boundary of its range.

The ratio male/female is 2.67, which is very close to the 2.5 by MUSTER & MEYER (2014) for Luxembourg. For The Netherlands a ratio of about 5 to 1 has been reported for pitfall captures (WIJNHOVEN *et al*., 2014), but no reason for this remarkable ratio was given.

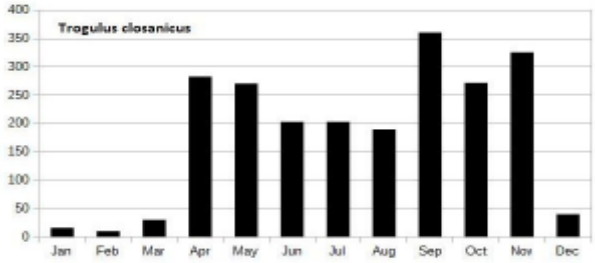

Figure 2: *Seasonal variation of pitfall catches of* T. closanicus *(based on 2 205 adults).*

Trogulus closanicus is year round active (Figure 2). They are not very active during the winter months, but once that temperatures are rising they are in great numbers captured with pitfalls. There is not a clear peak period. It is rather that their activity soon reaches a certain level and then plateaus. There is a slight dip during the summer months.

Trogulus nepaeformis

One of the criteria which is used to distinguish *T. nepaeformis* from *T. closanicus* is the distance between the eyes (WIJNHOVEN *et al*., 2014). The eyes are closer to each other in *T. nepaeformis*. This difference is not, or almost not, found in populations from the Armorican Massif in the Northwest of France (IORIO & DELFOSSE, 2016). Although there is a good fit with the other criteria for *T. nepaeformis s.s*. IORIO prefers to name specimens from that area rather as *T. gr. nepaeformis*. In the specimens from our Belgian pitfalls no deviation from the eye criterium could be found. The eyes of males of both species tend to be slightly

closer to each other than in females, but both sexes of *T. closanicus* have their eyes wider apart than their *T. nepaeformis* counterparts. Therefore we see currently no reason to suspect that our *T. nepaeformis* still could hide another species.

T. nepaeformis prefers an environment that is similar to the preferences of *T. closanicus*, only slightly more humid and colder (KOMPOSCH & GRUBER, 2004; MUSTER & MEYER, 2014). In Luxembourg the species was only found in woods and copses, not in grassland. In The Netherlands the species is reported from woodland and grassland and *T. nepaeformis* is at each site where it is found, syntopic with *T. closanicus* (WIJNHOVEN *et al*., 2014). Our catches show *T. nepaeformis* syntopic with *T. closanicus* in about half (54 with both species out of 98 where *T. nepaeformis* was found) of the pitfall stations (see Table 2). Notice that, contrary to *T. closanicus*, the species is also reported west of the River Scheldt (Figure 3).

In Germany the species gravitates to the southern third of the country (ARACHNOLOGISCHE GESELLSCHAFT, 2017). Most finds are reported from latitudes lower than Belgium. In The Netherlands the species is confined to the southern most corner of the country, near the eastern border of Belgium. *T. nepaeformis* probably reaches here the north-western limit of its range.

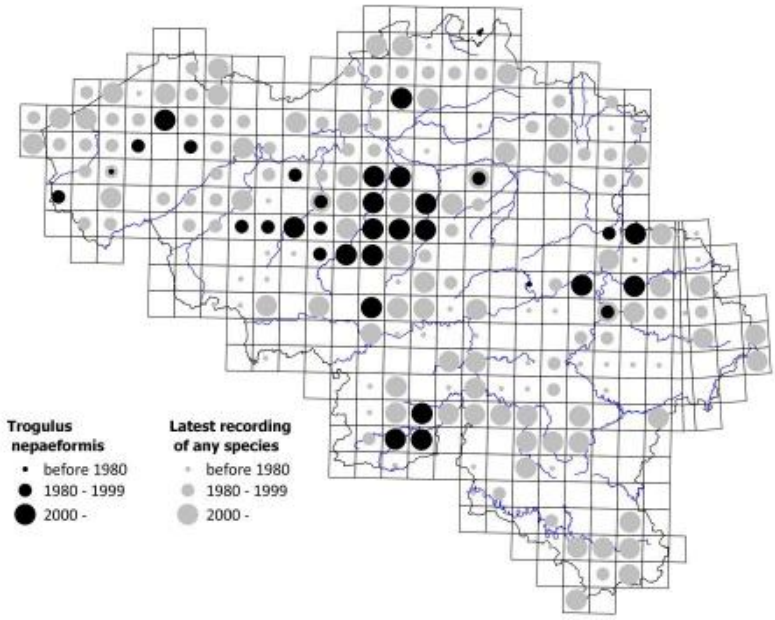

Figure 3: *Provisional map of the occurrence of* T. nepaeformis *in Belgium. Map is based on the reviewed material only. Grey dots give UTM-squares (10×10 km) where Opiliones were captured.*

The ratio male/female is 1.42. In The Netherlands males and females are captured in pitfalls in about the same numbers (WIJNHOVEN *et al*., 2014). A ratio of 3 is reported from Luxembourg (MUSTER & MEYER, 2014) but that is based on not more than 8 individuals, because the species is extremely rarely found in that country.

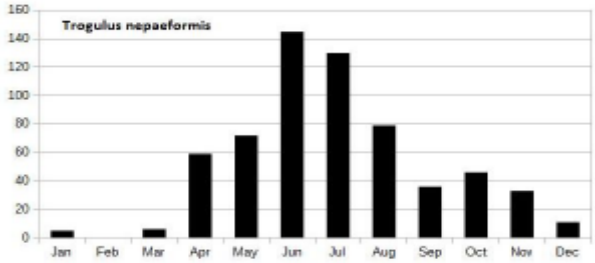

Figure 4: *Seasonal variation of pitfall catches of* T. nepaeformis *(based on 622 adults).*

Trogulus nepaeformis is year-round active (Figure 4). Unlike the previous species, however, they do show a pronounced peak activity period. The maximum occurs in June and July, exactly where *T. closanicus* has its summer recess (Figure 2).

Trogulus tricarinatus

According to the data here presented, *T. tricarinatus* is the most rarely encountered species of the genus in Belgium. It was catched in only 30 of the 175 pitfall stations (Table 2). The number of occurrences is lowest as well as the number of specimens. From The Netherlands, however, *T. tricarinatus* is reported as the most widespread and most common of the three (WIJNHOVEN *et al*., 2014) and is found in the eastern half of the country from the South up to the North. However, no pitfall captures were done in the western part of the country and the species may very well be present westwards also.

Habitat requirements of this species are less specific than for the other two *Trogulus* (MARTENS, 1978). In Luxembourg *T. tricarinatus* is reported from chalk grasslands, hedges, copses and deciduous forest (MUSTER & MEYER, 2014). In Great Britain *T. tricarinatus* is considered mainly a species of calcareous woodland that also occurs in scrub and open grasslands, especially in taller swards with well-developed litter layers (ALEXANDER, 2003). In The Netherlands however the species is also reported from forests on acidic sandy soils (NOORDIJK *et al.*, 2012). In the South of that country *T. tricarinatus* is always found syntopic with at least one of the other *Trogulus* species (WIJNHOVEN *et al*., 2014). Our samples from pitfalls were catched in forests (19 stations), in stone quarries (6 stations) and on calcareous grassland (5 stations). All 12 stations where *T. tricarinatus* was catched syntopic with another species (see Table 2) were located in forests, except for one station in calcareous grasland where all three species where found together. In the other stations, on grassland and in quarries, only *T. tricarinatus* was captured.

Its range in Belgium extends to the West and connects with its distribution in Great Britain where the species is mostly found in the South at "Belgian" latitudes. It is the only *Trogulus* reported from Great Britain (BRITISH ARACHNOLOGICAL SOCIETY, 2017).

T. tricarinatus has the most northernly distribution of the three species at hand. In Germany the species can be found in the whole territory except for the extreme North (ARACHNOLOGISCHE GESELLSCHAFT, 2017).

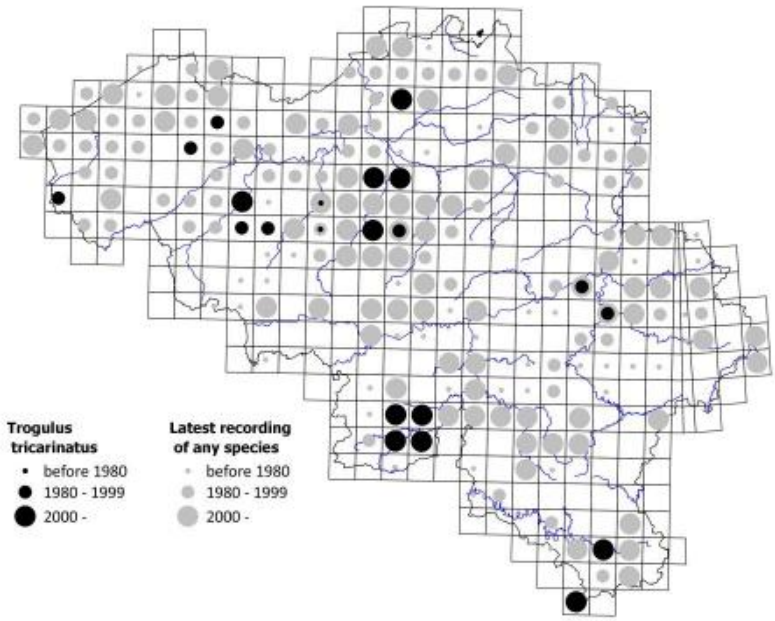

Figure 5: *Provisional map of the occurrence of* T. tricarinatus *in Belgium. Map is based on the reviewed material only. Grey dots give UTM-squares (10×10 km) where Opiliones were captured.*

The ratio male/female is 0.21. Many more females than males are found. In The Netherlands very few males were captured (WIJNHOVEN *et al*., 2014). No males were reported from Luxembourg (MUSTER & MEYER, 2014). For Germany and Austria, MARTENS (1978) paints a more diverse picture from parthenogenetic populations to populations where males prevail. The fraction of males in Belgium tends to be at the higher end of the range.

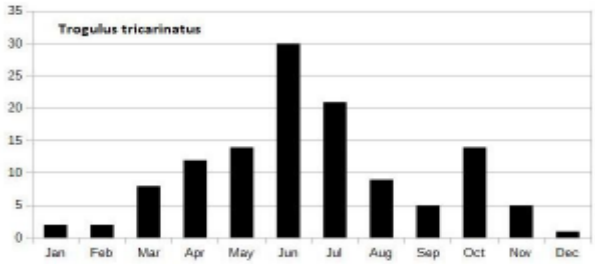

Figure 6: *Seasonal variation of pitfall catches of* T. tricarinatus *(based on 123 adults).*

Trogulus tricarinatus is year-round active with a peak activity in June (Figure 6). For Luxembourg MUSTER & MEYER (2014) report a peak in the same month.

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