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Short Communication

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New records of three parasitoids, *Pteroptrix chinensis*, *Aphytis hispanicus*, and *Marlattiella prima* (Hymenoptera: Aphelinidae) associated with an exotic scale, *Lopholeucaspis japonica* (Hemiptera: Diaspididae) in Tennessee

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A survey for parasitoids of *Lopholeucaspis japonica* Cockerell (Hemiptera: Diaspididae), an exotic scale of woody ornamentals, resulted in the discovery of 3 species of aphelinid parasitoid wasps, *Pteroptrix chinensis* (Howard), *Aphytis hispanicus* (Mercet), and *Marlattiella prima* Howard. This serves as the first report of these parasitoids reared from a host in the state of Tennessee, USA. Despite routine pesticide applications in the surveyed nursery and directed treatments of the infested plants to control the scale outbreak, the percentage of parasitized scale in privet and euonymus shrubs averaged 7.0% and 7.9%, respectively. These parasitoids may be useful in the natural or managed control of this pest in the United States, but additional research is needed to understand how these parasitoids contribute to the control of *L. japonica* in the landscape and how nursery production practices can be modified to promote parasitoid populations.

Key words: non-native, Japanese maple scale, Diaspididae, parasitoids, Aphelinidae

Introduction

The non-native *Lopholeucaspis japonica* (Cockerell) is an armored scale in the family Diaspididae (Hemiptera). It is reported as a serious pest throughout Asia, Eastern Europe, and South America (EPPO 2016). It was first reported in the United States in Connecticut in 1914 (Miller et al. 2005). Currently, the distribution range of this scale in the United States includes Alabama, Connecticut, Delaware, Georgia, Indiana, Kansas, Kentucky, Louisiana, Maryland, Missouri, Nebraska, New Jersey, New York, North Carolina, Ohio, Pennsylvania, Rhode Island, Tennessee, Texas Virginia, and Washington DC (Frank et al. 2013, García Morales et al. 2016, Jeger et al. 2018, Gilder et al. 2020). It has a host range of approximately 97 plant species from 35 families (Miller and Davidson 2005, Harsur et al. 2018, Jeger et al. 2018, Shrewsbury et al. 2020).

In the eastern United States, this scale insect has been identified as a nursery pest in woody ornamental production regions (Fulcher and Halcomb 2012, Addesso et al. 2016). There were at least 26 recorded shipment rejections of nursery trees from Tennessee in the last 3 years due to its infestation, posing an economic threat to growers and a hit to the nursery industry's reputation (pers. comm, Tennessee Plant Certification Administrator). The survival strategies of this scale include a wide host range, small size, waxy armored covering, and cryptic coloration. The wax repels water and therefore is effective at protecting scales from insecticide formulations applied in water and also acts as a barrier against generalist predators. These features make *L. japonica* challenging to manage. Only adult males and crawlers are mobile and the crawler stage is the most vulnerable stage to predators and insecticides. The crawlers of this scale, however, are reported to secrete their wax covering within a few short hours after settling (Gill et al. 2014) which minimizes the amount of time they are vulnerable. Moreover, this scale feeds primarily on bark tissue, not leaves, which further complicates management as bark feeding can also reduce the efficacy of systemic insecticide treatments against scales (Addesso and O'Neal 2018). The combined attributes of this scale necessitate a multifaceted approach to management, including the use of natural enemies that can overcome the physical and physiological adaptations of this pest.

While outbreaks of this scale have become common in nursery production, population levels do not appear as high in adjacent woods (pers obs.) which led us to hypothesize that scale populations are controlled at the landscape level by natural enemies. We currently know very little about the identity of natural enemies of this scale in landscape or nursery production or how much they contribute to population control. To date, *Marlatiella prima* Howard, an exotic parasitoid wasp native of China and Japan, is the only reported parasitoid in the United States for this pest (Krombein et al. 1979, Gilder et al. 2020). In this context, we surveyed a woody ornamental production nursery infested by this scale with the goal of recovering parasitoids in the middle Tennessee region.

Materials and Methods

We surveyed a nursery in middle Tennessee (35.5574°N, −85.9091°W, Warren County, Morrison, TN) in late June 2023. Scale-infested plant shoots approximately 15 cm long with 1 cm diameter were collected from burning bush (*Euonymus alatus* [Thunb.]) and Chinese privet (*Lingustrum sinense* Lour.). We collected infested shoot samples from 7 privet and 20 euonymus plants grown in 5-gallon containers. The samples were brought back to the lab for evaluation. The total number of scale covers on each shoot section was counted, as well as the number of parasitoid exit holes on each shoot. We determined the percent parasitization of each sample by dividing the number of emergence holes by the total number of scales. Upon confirming the presence of parasitoids at the nursery, we collected additional material in early July 2023 to identify the number and species diversity of the parasitoids present. We collected 26 privet samples and 9 euonymus samples and held them in plastic emergence vials at room temperature. We observed each vial and assessed parasitoid emergence daily for 20 days. The emerged parasitoids were preserved in absolute alcohol and were counted using a stereo microscope (Zeiss Stemi 2000-C, Leica Microsystems, Wetzlar, Germany). Specimens were critical-point-dried at Texas A&M (Gordh and Hall 1979) and card-mounted or slide-mounted in Canada balsam using the methods described in Noyes (1982). Specimens were identified using the keys in Gibson et al. (1997), Rosen and DeBach (1979), and by comparing them with identified specimens in the Texas A&M University Insect Collection (TAMUIC). Voucher specimens have been deposited in TAMUIC under voucher number 761. Morphological terminology below follows (Gibson et al. 1997). Imaging was done using 10× and 20× Mitutoyo Plan-Apochromat objectives mounted in a Macropod system (see macroscopicsolutions.com) and an Olympus BH2 compound microscope equipped with Plan-Apochromat objectives and a Canon EOS 5D camera connected to a Windows 10 computer running Promicra QuickPHOTO software. Focus-stacking was performed in ZereneStacker Version 1.04 and contrast enhancement and minor color correction of images was performed in Adobe Lightroom Version 13.0.1.

Results

Parasitism levels averaging 7.0% were observed in infested privet plant samples and 7.9% in the euonymus samples (Table 1). The range of parasitism among the replicates was 3.8–11.5% for privet, and 0.7–34.3% for the euonymous samples. Three species of parasitoids belonging to the family Aphelinidae were recorded in this study: *Pteroptrix chinensis* (Howard) (Fig. 1), *Aphytis hispanicus* (Mercet) (Fig. 2), and *Marlattiella prima* Howard (Fig. 3).

A total of 94 *Pteroptrix chinensis* emerged from the infested privet samples followed by 29 *M. prima* and 11 *A. hispanicus* (Table 1). A similar trend was observed in the case of euonymus samples, with *P. chinensis* being the most dominant (14) followed by *M. prima* (11) and *A. hispanicus* (9) being the least numerous (Table 1).

Discussion

Pteroptrix species closely resemble those of the genus *Encarsia* but can be differentiated by their 4-segmented tarsi. Our specimens of *P. chinensis* (Fig. 1a–c) agree well with the drawings in Xu and Huang (2004) and with a long series in TAMUIC of *P. chinensis* collected in Japan on *Aulacaspis rosae* Bouché and determined by A. B. Gahan. However, Luke Kresslein (USDA, Systematic Entomology Laboratory, National Museum of Natural History, Washington D.C., USA) noted that our specimens appear to differ slightly in antennal proportions from the type material that he examined at the National Museum of Natural History.

Pteroptrix chinensis is a parasitoid of the California red scale (*Aonidiella aurantia* [Maskell]) on citrus in South China, and it has been regarded as a potential control of the California red scale in California since its discovery in 1906 (Flanders et al. 1958). Currently, it is distributed in countries including India, Italy, Japan, China, Russia, Taiwan, Hawaii, and California of the United States (UCD Curators 2023). It is also known to parasitize *L. japonica* in Japan (Japoshvili et al. 2013). Our current study serves as a range extension in the United States for this parasitoid.

Aphytis hispanicus (Fig. 2a–c) can be identified with the following sets of characters mentioned in Abd-Rabou (2004) and Rosen and Debach (1979). Like other members of the *proclia* species group, *A. hispanicus* has an occiput with a fuscous to black bar on each side of the foramen, but in *hispanicus* this is particularly well marked. The gaster is not uniformly fuscous dorsally, but paler with short fuscous strips on sides, and at most, with complete cross-bands on terga 1–5 (Fig. 2a). The pedicel and flagellum are uniformly fuscous and the apex of club is blackish (Fig. 2b). It is perhaps most similar to *Aphytis comperei* DeBach and Rosen, but it can be distinguished from this species using the characters and diagnosis in Rosen and DeBach (1979).

A. hispanicus is a south Palearctic species that is now widespread in the northern hemisphere. It has been reared from scale insects such

Table 1. Percent parasitism and parasitoid emergence from privet and euonymus samples

Plants	Scales (mean ± SEM)	Emergence holes (mean ± SEM)	% Parasitism (mean ± SEM)	Parasitoids	Total
Privet	294 ± 50	18 ± 3	7.0 ± 1.2	*Pteroptrix chinensis*	94
				Marlattiella prima	29
				Aphytis hispanicus	11
Euonymous	460 ± 68	29 ± 4	7.9 ± 1.7	*Pteroptrix chinensis*	14
				Marlattiella prima	11
				Aphytis hispanicus	9

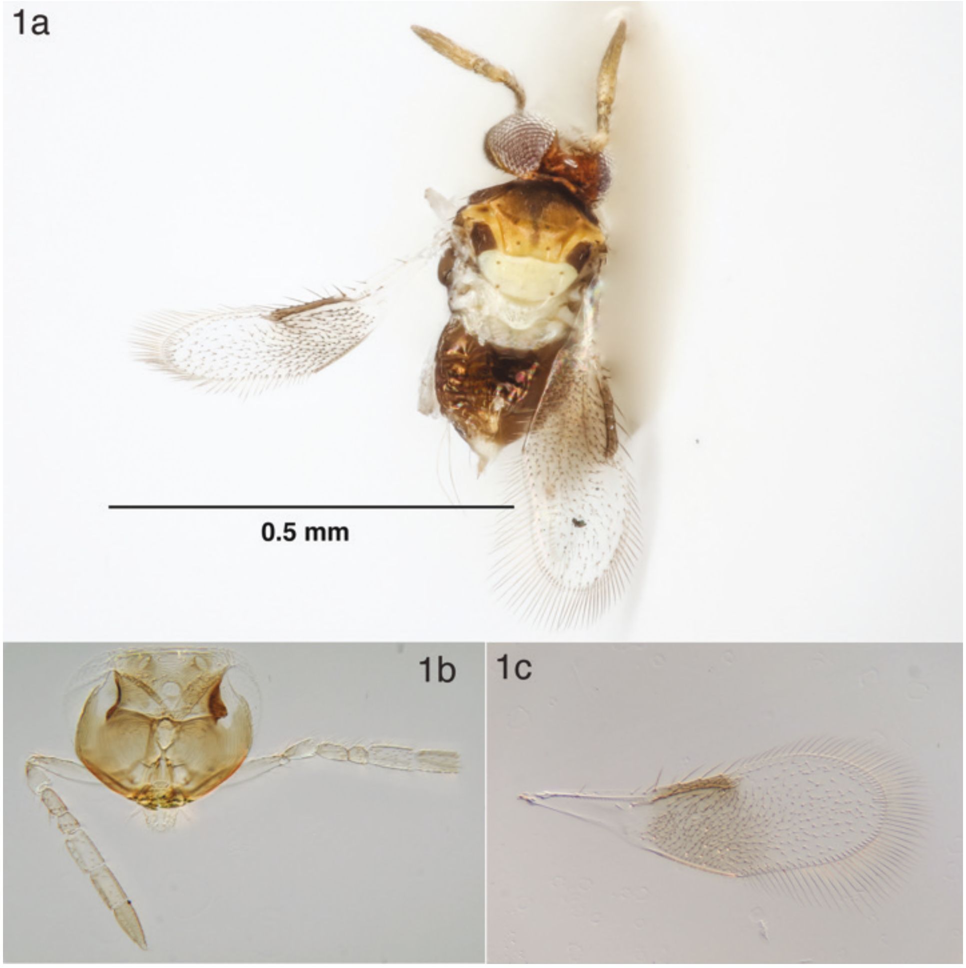

Fig. 1. *Pteroptrix chinensis* (Howard) female: a) dorsal habitus; b) head and antennae; and c) fore wing.

as *Parlatoria pergandii* Comstock in Israel, Turkey, Trinidad, Brazil, Mexico, Texas, and California, from *Parlatoria cinerea* Doane and Hadden in Trinidad, from *Chrysomphalus dictyospermi* (Morgan) and *Acutaspis scutiformis* (Cockerell) in Brazil, from California red scale material (probably chaff scale) in Morocco, and from undetermined hosts in Italy, Jamaica, Taiwan, and Florida. Yasnosh (1972) recorded *A. hispanicus* as a parasite of *P. pergandii, P.oleae* (Colvee), *Aspidiotus nerii* Bouche, *Lopholeucaspis japonica* (Cockerell), *Mytilaspis conchiformis* (Gmelin) *[=Lepidosaphes ficus* (Signoret)], and *Insulaspis pallida* (Green) in the Caucasus. Populations of *A. hispanicus* are commonly uniparental (thelytokous) and the species often occurs in sympatry with *Aphytis comperei* DeBach and Rosen (Woolley and Browning 1987). Yasnosh (1978) noted that introductions of *A. hispanicus* were in progress along the Black Sea coast of the Caucasus, but she provides no further details. Gerson (1967) reported parasitism rates of 4% to almost 30% by *A. hispanicus* on *P. pergandii* in 2 *A. hispanicus* locations in Israel.

Marlattiella prima (Fig. 3a–c) was identified by comparison with specimens collected in Texas during the study of Gilder et al. (2020). This material had been compared with the type material of *M. prima* in the NMNH. The genus *Marlattiella* is a distinctive member of the subfamily Aphelininae (Kim and Heraty 2012), generally characterized by a linea calva in the forewing (lc: Figs. 2c and 3c) and 3 funicle segments and an unsegmented clava in the antenna (Fig. 2b). However, *Marlattiella* spp. individuals can be differentiated from other Aphelininae by a single anellus in females (transverse funicular, anl: Fig. 3b), no anellus in males, i.e., the antenna is 3-segmented in males, Tachikawa (1962a), and a long clava in the antenna. Some species of *Aphytis* share this antennal formula but unlike *Marlattiella*, *Aphytis* species have a longer propodeum that bears marginal crenulae on the posterior margin (Rosen and DeBach 1970). *Eretmocerus* species have a similar antennal formula, but the tarsal formula is 4-4-4 (Kim and Heraty 2012). *M. prima* is originally a native of China and Japan and it is known to parasitize *L. japonica* (UCD curators, 2023). In the United States, *M. prima* was reported in 1979 in Maryland and in 2020 in Texas (Krombein et al. 1979, Gilder et al. 2020). Suh and Evans (2019) and Xu and Huang (2004) mention this species as attacking *L. japonica* in Korea and China, respectively, but neither provide information on its potential impact.

Although *Pteroptrix chinensis* and *Aphytis hispanicus* have been previously reported as parasitoids of *L. japonica*, the current study establishes them as parasitoids of this pest in the United States for the first time. The biology of these parasitoids may contribute to their ability to disperse and establish outside their native range. Some populations of *Aphytis hispanicus* have been reported as uniparental (thelytokous), with populations that do not require a male for reproduction. In our survey, we only reared out female *A. hispanicus*. If the population in Tennessee is thelytokous, this could greatly facilitate the establishment in new areas. Further collections of the species are required to confirm whether males are present in the population. *Pteroptrix chinensis* is a heteronomous hyperparasitoid (Hunter and

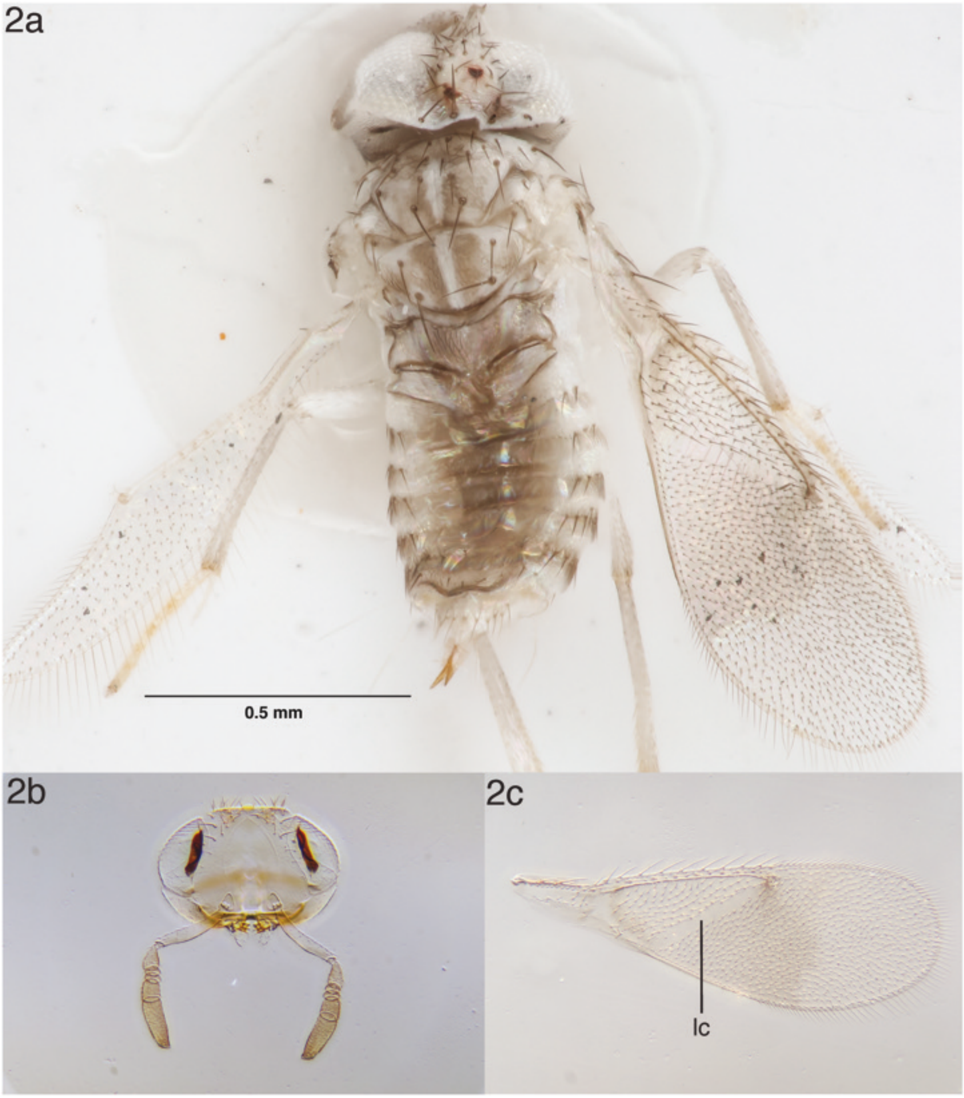

Fig. 2. *Aphytis hispanicus* (Mercet) female: a) dorsal habitus; b) head and antennae; and c) fore wing (lc = linea calva).

Woolley 2001), meaning males are parasitic on conspecific females or on females of other species. Although some have suggested that this life history may lead to failures in colonization efforts in new habitats (Flanders et al. 1958, Rosen and DeBach 1970), many species of heteronomous hyperparasitoid *Encarsia* have been used successfully in biological control programs (Zang et al. 2011).

Due to the severity of the *L. japonica* infestation, the grower treated both plant container species with pesticides before our survey. The parasitization percentages of 7.0 and 7.9 from privet and euonymus samples were surprisingly high, despite the routine and targeted pesticide sprays in the surveyed nursery. These wasps could have some degree of resistance to the insecticides used by the grower and/or were not present as adults during application periods. Prior studies report that the adults of *A. hispanicus* can emerge from underneath the waxy secretions of the scale insect without making an exit hole (Gerson 1967, 1968, Rosen and DeBach 1979). Consequently, parasitization levels could be even higher in the field since we used the exit holes to calculate the percentages.

Our study represents the first occurrence of these 3 parasitoids, *Pteroptrix chinensis*, *Aphytis hispanicus*, and *Marlattiella prima* in Tennessee. This record expands the distribution range of the parasitoids to the southeastern region of the United States, having previously been recorded only from the western United States, California (*P. chinensis*) and the northeastern region of Maryland and the central region of Texas (*M. prima*). Additional surveys are planned to create a comprehensive list of parasitoids of *L. japonica* in Tennessee. Further studies of these parasitoids related to their parasitic potential and conservation will aid us in improving the management of *L. japonica* by promoting their natural enemies.

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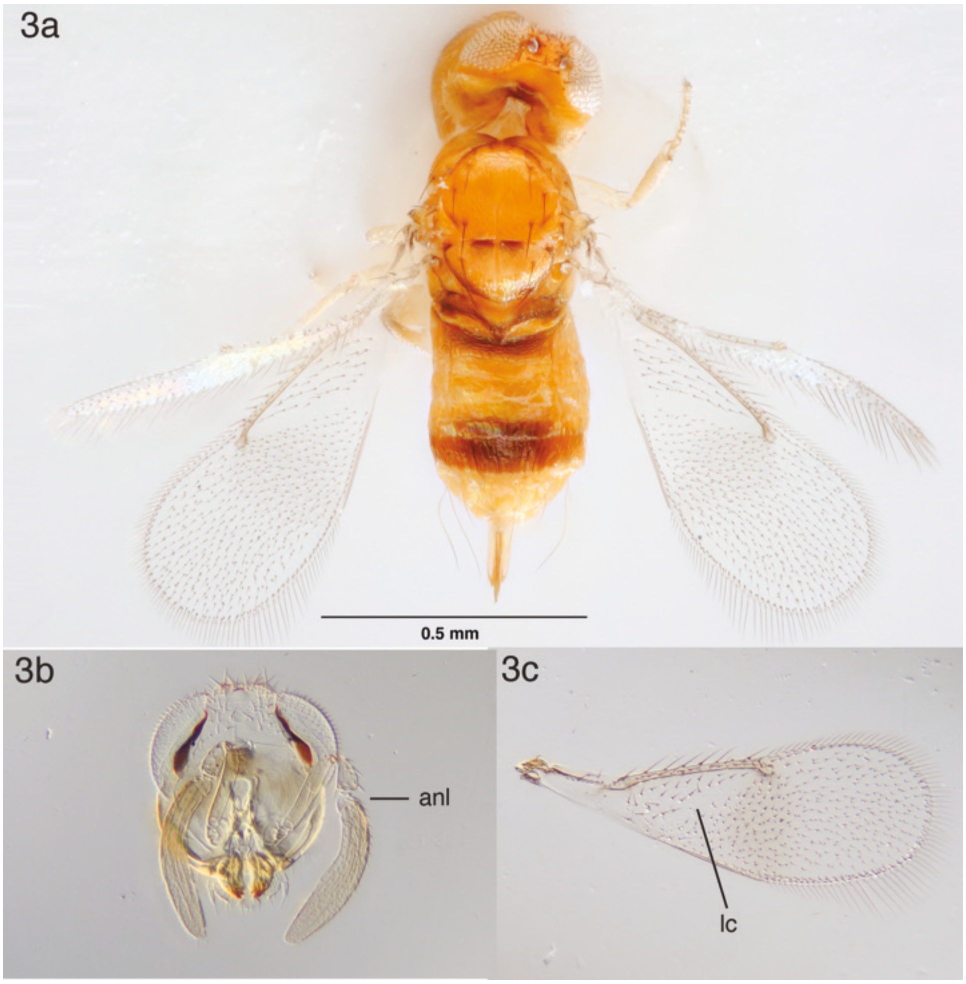

Fig. 3. *Marlattiella prima* (Howard) female: a) dorsal habitus; b) head and antennae; and c) fore wing (anl = anellus, lc = linea calva).

Author Contributions

J. Alfred Daniel (Conceptualization [equal], Data curation [equal], Formal analysis [equal], Methodology [equal], Writing—original draft [equal]), Karla Addesso (Conceptualization [equal], Funding acquisition [lead], Methodology [equal], Project administration [lead], Resources [lead], Supervision [lead], Writing—review & editing [lead]), and James Woolley (Investigation [equal], Resources [equal], Visualization [equal], Writing—review & editing [equal])

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