



NEW BODY METRICS TO DETERMINE ASTEROID SIZE AND WEIGHT DIRECTLY IN THE FIELD

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ABSTRACT

Asteroids are an important group of predators in many marine ecosystems. The measure of body size is crucial in studying asteroid biology as this variable affects both prey selection and predation impact. Current field techniques for estimating asteroid size (i.e. total length TL, total weight TW) include the measure of correlated variables, such as the central disk (CD) and arm length (AL). However, these variables are often time-consuming and require a direct handling of the organism. We tested the accuracy of new asteroid body metrics, the arm height (AH) or arm width (AW), to rapidly estimate body size in asteroids. AH and AW were measured in three of the most common Mediterranean asteroids (*Marthasterias glacialis*, *Ophidiaster ophidianus* and *Coscinasterias tenuispina*), sampled from April to August 2008, along the coasts of Ustica Island off the northern coast of Sicily. We used both linear and exponential regression analyses to compare the performance of AH, AL, AW and CD in estimating size for the three species studied. Results suggest that, in *M. glacialis* and *C. tenuispina*, AH is strongly correlated ($p < 0.001$) with both TL and TW, whereas in *O. ophidianus* it gives a good correlation ($p < 0.001$) with TW only. AW was poorly correlated with both TL and TW in *M. glacialis* and *C. tenuispina*, but not in *O. ophidianus*, where it showed the highest correlation with TW. Thus, only the novel AH measure constitutes a convenient and reliable way of measuring asteroid body size in the field.

Key words: starfish, size, *Marthasterias glacialis*, *Ophidiaster ophidianus*, *Coscinasterias tenuispina*, Mediterranean Sea.

RESUMEN

Las estrellas de mar son un importante grupo de depredadores en muchos ecosistemas marinos. La medida del tamaño del cuerpo es crucial en el estudio de la biología de estrellas de mar dado que esta variable afecta tanto a la selección de las presas como al impacto de la depredación. Las técnicas de campo actuales para estimar el tamaño de las estrellas de mar (es decir, longitud total y peso total) incluyen la medida de variables correlacionadas, como el disco central y la longitud del brazo. Sin embargo, esas variables precisan a menudo mucho tiempo y requieren una manipulación directa del organismo. Hemos puesto a prueba la exactitud de nuevas métricas del cuerpo de las estrellas de mar, la altura del brazo o el ancho del brazo, para estimar rápidamente el tamaño del cuerpo en las estrellas de mar. Estas dos variables se midieron en tres de las estrellas de mar más comunes del Mediterráneo (*Marthasterias glacialis*, *Ophidiaster ophidianus* y *Coscinasterias tenuispina*), obtenidos mediante muestreo entre abril y agosto de 2008, a lo largo de las costas de la Isla de Ustica frente a la costa norte de Sicilia. Se utilizó tanto la regresión lineal como exponencial para comparar el rendimiento de las cuatro variables citadas para estimar el tamaño del cuerpo en las tres especies. Los resultados sugieren que, en *M. glacialis* y *C. tenuispina*, la altura del brazo se correlaciona fuertemente ($p < 0,001$) tanto con la longitud como con el peso total, mientras que en *O. ophidianus* proporciona buena correlación ($p < 0,001$) únicamente con el peso total. La anchura de brazo tuvo pobre correlación tanto con el peso como con la longitud total en *M. glacialis* y *C. tenuispina*, pero no en *O. ophidianus*, donde mostró la mayor correlación con el peso total. Así, sólo la nueva medida propuesta de altura del brazo constituye una manera conveniente y fiable para medir el tamaño del cuerpo de asteroides en el campo.

Palabras clave: estrella de mar, tamaño, *Marthasterias glacialis*, *Ophidiaster ophidianus*, *Coscinasterias tenuispina*, Mar Mediterráneo.

INTRODUCTION

Asteroids, popularly known as starfish or sea stars, are recognized as important predators in most marine ecosystems (Lawrence 2013). Some species are well-known for their crucial role in shaping the structure and functioning of benthic ecosystems such as rocky shores, coral reefs and algal forests (Paine 1974; Fanelli *et al.* 1994; De'ath & Moran 1998; Bonaviri *et al.* 2009; Barahona & Navarrete 2010). Hence, the direct study of starfish populations in the field may have important ecological implications. According to several studies, predator-prey size relationship is perhaps the most important component that affects capture success of asteroids (Lawrence 2013). In this context, it is becoming of primary importance for marine scientists to acquire information about the distribution, population structure, size and weight of asteroids.

While assessing starfish abundance through underwater visual censuses is relatively easy, estimating the individual size (e.g. total length [TL] and total weight [TW], which require the collection of the specimen and its weighing, under dry condition in the laboratory), could prove to be both time-consuming and difficult without a direct handling of the organism.

A thorough scrutiny of the literature reveals that several methods are used to evaluate starfish size and these consist in measuring, with vernier calipers, the length of 1) the maximum tip-to-tip diameter (arm-span) (Barker & Nichols 1983; Gaymer *et al.* 2004; Gianguzza *et al.* 2009a, b; Tuya & Duarte 2012); 2) the radius from the center of the disk to the tip of the longest arm (Penney & Griffiths 1984; Frid 1992; Gaymer & Himmelman 2002; Ganmanee *et al.* 2003; Gaymer & Himmelman 2008); 3) the radius from the edge of the disk to the end of a normal arm on the opposite side (Minchin 1987); 4) a normal arm (Scheibling & Lauzon-Guay 2007; Urriago *et al.* 2011);

5) the mean linear distance from the tips of each arm to the opposite inward pointing angle (Sommer *et al.* 1999; Temara *et al.* 1999); 6) the major radius (Campbell *et al.* 2001); 7) the longest arm (Bernstein *et al.* 1981). A critical examination of the seven metrics above listed reveals that some of them were not clearly described by the authors (e.g. the definition of major radius and normal arm) and that some contain a certain degree of subjectivity so that the reader cannot grasp exactly how the measurements were done and how to reproduce them.

Here we propose two novel and practical metrics for the estimation of asteroid size and weight from field measurements. We used three common Mediterranean species as models: *Marthasterias glacialis* (Linnaeus, 1758), *Coscinasterias tenuispina* (Lamarck, 1816) and *Ophidiaster ophidianus* (Lamarck, 1816) (Fig. 1). The relations among two traditional metrics (diameter of the central disk CD and arm length AL) and two novel ones, arm height (AH) and arm width (AW) *versus* tip-to-tip diameter or total length (TL) and total weight (TW) were examined by two regression models. We predict that the two new metrics, as the traditional ones, are strongly correlated with TL and TW and therefore constitute convenient and practical variables to measure body size in starfish.

MATERIALS AND METHODS

Study area and species investigated

The study was carried out at Ustica Island Marine Protected Area (MPA) in the Southern Tyrrhenian Sea (Western Mediterranean, 38°42'20"N -10°43'43"E), 60 km north of the Sicilian coast. This MPA was chosen as a study area mainly for the high density of asteroids, especially *M. glacialis* in barren grounds, thus facilitating animal collection (Bonaviri *et al.*, 2009; Gianguzza *et al.*, 2009a, b; Gianguzza *et al.*, 2010).

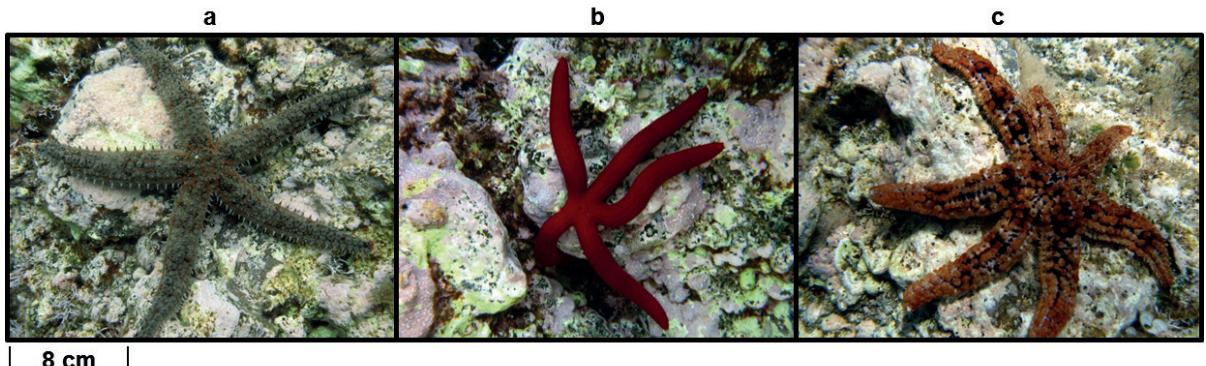


Figure 1:
Asteroids species investigated: *M. glacialis* (a), *O. ophidianus* (b) and *C. tenuispina* (c).

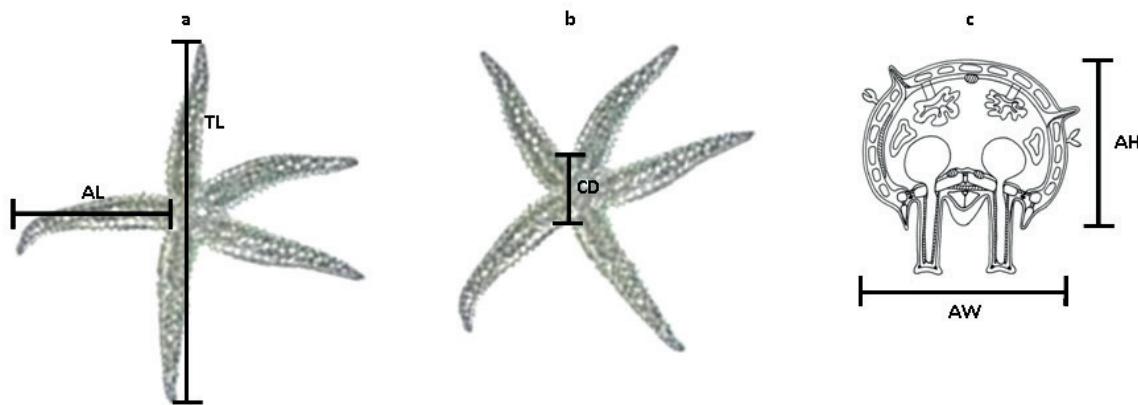


Figure 2:
Biometric variables investigated: total length (TL) (a), central disk (CD) (b), arm height (AH) and arm width (AW) (c, cross-section of a starfish arm).

M. glacialis is a starfish inhabiting the intertidal and subtidal zone of the eastern Atlantic rocky coasts, including southern and southwestern Europe (Guillou 1996), as well as the Mediterranean (Savy 1987) and South Africa (Penney & Griffiths 1984). This species is characterized by five narrow tapering arms and it is a voracious predator capable of exploiting a wide range of prey resources, playing a key role in shaping community structure (Bonaviri *et al.* 2009; Tuya & Duarte 2012).

The thermophilic *O. ophidianus* typically inhabits Mediterranean subtidal rocky bottoms, from 0 to 100 m and also occurs in the Azorean Archipelago (Marques 1983). Little is known about the biology of this species, protected under the EU's Habitats Directive (Relini & Tunisi 2009). It has five long cylindrical, blunt tipped arms, which are narrow at their base.

C. tenuispina is widely distributed in the Atlantic Ocean, from North Carolina to coasts of Guinea along western Africa (Clark & Downey 1992). This species inhabits shallow waters down to a depth of 150 m, and it occurs in a variety of habitats, ranging from the underside of stones on hard seabeds to algae and seagrasses, where it mainly feeds on sea urchins and bivalves (Clark & Downey 1992). The number of arms in this species ranges from 6 to 12 (typically 7), often showing different lengths.

Data collection and analyses

M. glacialis, *O. ophidianus* and *C. tenuispina* individuals were haphazardly collected by SCUBA diving from April to August 2008 along the coasts of Ustica Island. Collections were made in the upper infralittoral, excluding starfish with regenerating arms. Since starfish show diurnal feeding activity (Ebling *et al.* 1966), individuals with intact arms were preferentially collected around midday.

We analyzed 45 individuals of *M. glacialis*, 75 of *O. ophidianus* and 29 of *C. tenuispina*. For each individual, the following morphometric variables were measured with vernier calipers to the nearest 0.1 mm in the field: total length (TL), or tip-to-tip diameter (Fig. 2a); arm length (AL), measured from the edge of the central disk to the tip of the arm (Fig. 2a); diameter of the central disk (CD, Fig. 2b); arm height (AH) and arm width (AW) adjacent to the central disc (Fig. 2c). After field measurements, every starfish was placed individually in a numbered landing net and immediately transported, in large sealed plastic bags containing oxygenated seawater, to the MPA Laboratory of Cala S. Maria. Labeled starfish were then weighed (TW) using a portable balance (1g accuracy), after removing mucus secretions and seawater by gently squeezing them so as to increase weighing accuracy. Immediately after weighing, starfish specimens

Table 1:
Measurements (min-max, mean±s.d.) of starfish biometric variables (CD=central disk. AL=arm length. AH=arm height. AW=arm width). All variables are expressed in millimeters.

	<i>M. glacialis</i>	mean±s.d.	<i>O. ophidianus</i>	mean±s.d.	<i>C. tenuispina</i>	mean±s.d.
CD	18-43	31.0±7.5	15-38	22.9±3.1	4-45	18.2±9.1
AL	55-170	112.4±31.9	42-158	97.5±24.4	11-98	51.1±18.2
AH	6-33	17.1±6.9	6-20	13.0±2.6	4-15	8.2±2.9
AW	8-25	15.4±4.8	5-21	13.3±2.7	3.5-16	7.9±2.9

Table 2:

Regression analysis of starfish biometric variables (TL=total length. CD=central disk. AL=arm length. AH=arm height. AW=arm width). $P<0.001$ for all regressions.

		Linear			Exponential			N
		α	β	R^2	α	β	R^2	
<i>M. glacialis</i>	CD/TL	-29.79	8.55	0.82	67.52	0.04	0.77	30
	AL/TL	-3.80	2.13	0.92	75.91	0.01	0.87	30
	AH/TL	53.49	12.00	0.76	99.10	0.05	0.71	30
	AW/TL	53.27	11.78	0.63	99.30	0.05	0.58	30
	CD/TW	-212.74	11.65	0.85	5.96	0.10	0.90	27
	AL/TW	-155.55	2.66	0.84	9.45	0.02	0.89	27
	AH/TW	-149.20	18.92	0.90	18.64	0.11	0.90	27
	AW/TW	-92.96	15.60	0.65	16.77	0.13	0.65	27
	TL/TW	-133.55	1.18	0.85	11.41	0.01	0.88	34
	CD/TL	-51.42	11.80	0.43	47.19	0.06	0.44	75
<i>O. ophidianus</i>	AL/TL	19.24	1.88	0.76	71.07	0.01	0.75	75
	AH/TL	1.24	15.46	0.60	61.44	0.09	0.65	75
	AW/TL	7.85	14.57	0.58	63.49	0.08	0.63	75
	CD/TW	-146.83	9.70	0.80	2.71	0.14	0.84	26
	AL/TW	-55.16	1.33	0.82	11.22	0.02	0.78	26
	AH/TW	-109.26	13.06	0.82	4.80	0.19	0.85	26
	AW/TW	-138.35	15.08	0.83	3.03	0.22	0.88	26
	TL/TW	-54.10	0.63	0.80	10.95	0.01	0.80	26
	CD/TL	62.26	2.49	0.39	67.34	0.02	0.37	29
	AL/TL	40.90	1.30	0.43	53.98	0.01	0.44	29
<i>C. tenuispina</i>	AH/TL	17.31	11.04	0.80	44.99	0.10	0.74	29
	AW/TL	31.07	9.68	0.62	49.91	0.09	0.61	29
	CD/TW	-5.78	1.30	0.56	4.47	0.07	0.55	15
	AL/TW	-5.59	0.48	0.43	4.51	0.03	0.43	15
	AH/TW	-12.91	3.86	0.74	3.35	0.19	0.67	15
	AW/TW	-2.31	2.72	0.45	5.37	0.14	0.45	15
	TL/TW	-11.42	0.29	0.77	3.42	0.01	0.74	15

were returned to the sampling locations. No individuals lost arms during the handling procedure.

Each morphological variable measured (AL, CD, AH, AW) was contrasted, through linear and exponential regressions, versus total length (TL) and total weight (TW). We applied both linear and exponential regressions, since the strength of the relationship between the two metrics investigated and the other metrics was not known. Since we performed repeated analyses, the Bonferroni correction was applied, setting $\alpha=0.0125$. Data were analyzed using SAS Enterprise Guide 4.3 software.

RESULTS

In *M. glacialis*, TL ranged from 109 to 350 mm, in *O. ophidianus* from 68 to 340 mm and in *C. tenuispina* from 45 to 185 mm. TW ranged from 17.9 to 483 g for *M. glacialis*, 26 to 155 g for *O. ophidianus* and from 8 to 36 g for *C. tenuispina*. Ranges of other variables are shown in Table 1.

In *M. glacialis*, the two novel metrics did not perform better than the traditional ones. AL vs TL linear regression ($R^2=0.92$, Fig. 3) and CD vs TW exponential regression ($R^2=0.90$, Fig. 4, Table 2) showed the highest R^2 values.

Overall, AH showed higher R^2 values and performed much better than AW in estimating TL ($R^2=0.76$, Table 2).

In *O. ophidianus*, one of the traditional (AL vs TL linear regression, $R^2=0.76$ Fig. 5) and the two novel metrics (AW vs TW, $R^2=0.85$ and AH vs TW, $R^2=0.88$ exponential regressions, Fig. 6) showed the highest R^2 values when regressed against TL and TW (Table 2).

In *C. tenuispina*, the novel variable AH (AH vs TL, $R^2=0.80$ and AH vs TW, $R^2=0.74$ linear regression) performed better than any other variable, registering the highest R^2 values (Fig. 7, 8; Table 2) when regressed against TL and TW.

DISCUSSION

In this study, we focused on size-weight relationships of three common Mediterranean asteroid species. Our results confirm, as previously suggested by indirect evidence (Alves *et al.* 2002; Micael *et al.* 2011; O'Gorman *et al.* 2012), that in the three species considered, values for TL and TW, which jointly define the body size of the individuals, are strongly correlated between themselves. In contrast, the hypothesis tested (AH and AW constitute convenient and reliable proxies for TL and TW in starfish)

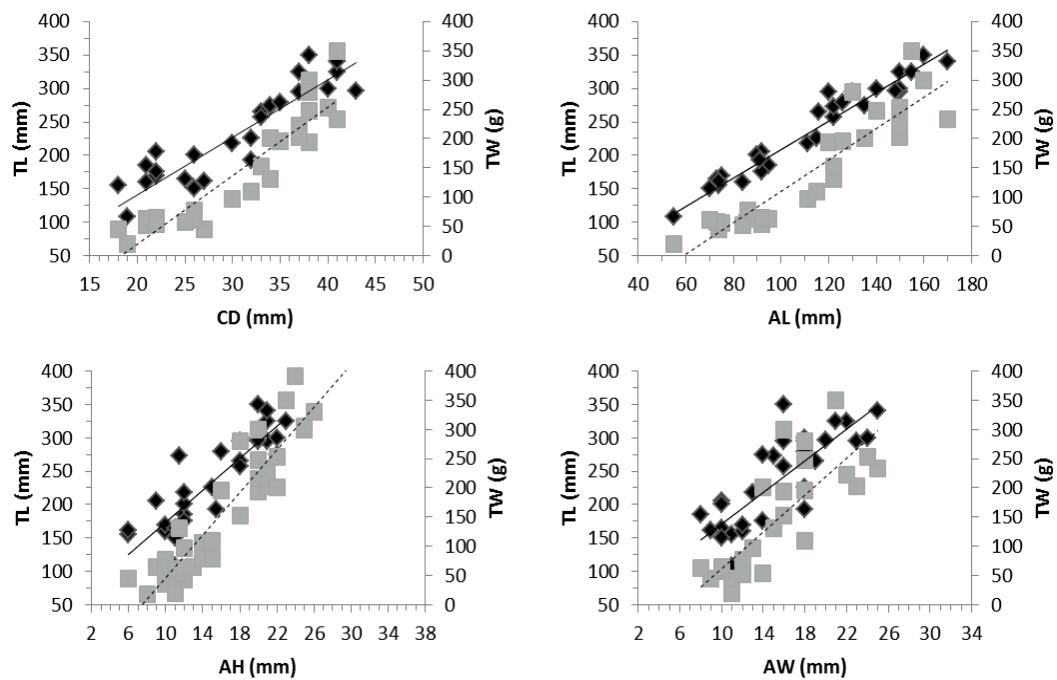


Figure 3:
Linear regression scatterplot of: central disk (CD), arm length (AL), arm height (AH) and arm width (AW) vs total length (TL, black symbols) and total weight (TW, grey symbols) for *M. glacialis* variables.

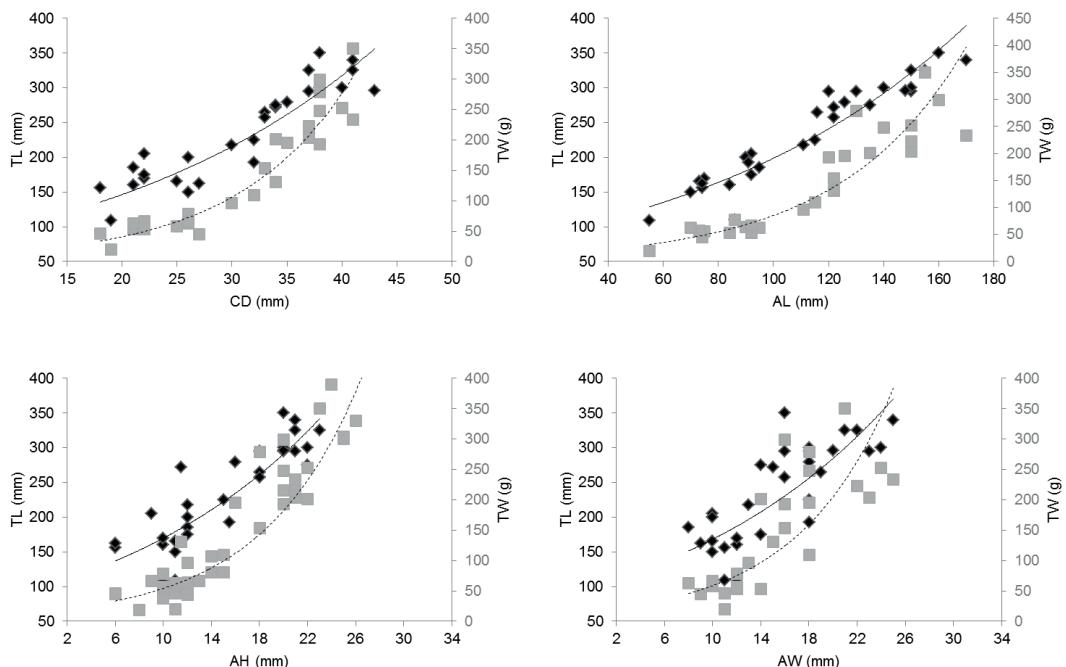


Figure 4:
Exponential regression scatterplot of: central disk (CD), arm length (AL), arm height (AH) and arm width (AW) vs total length (TL, black symbols) and total weight (TW, grey symbols) for *M. glacialis* variables.

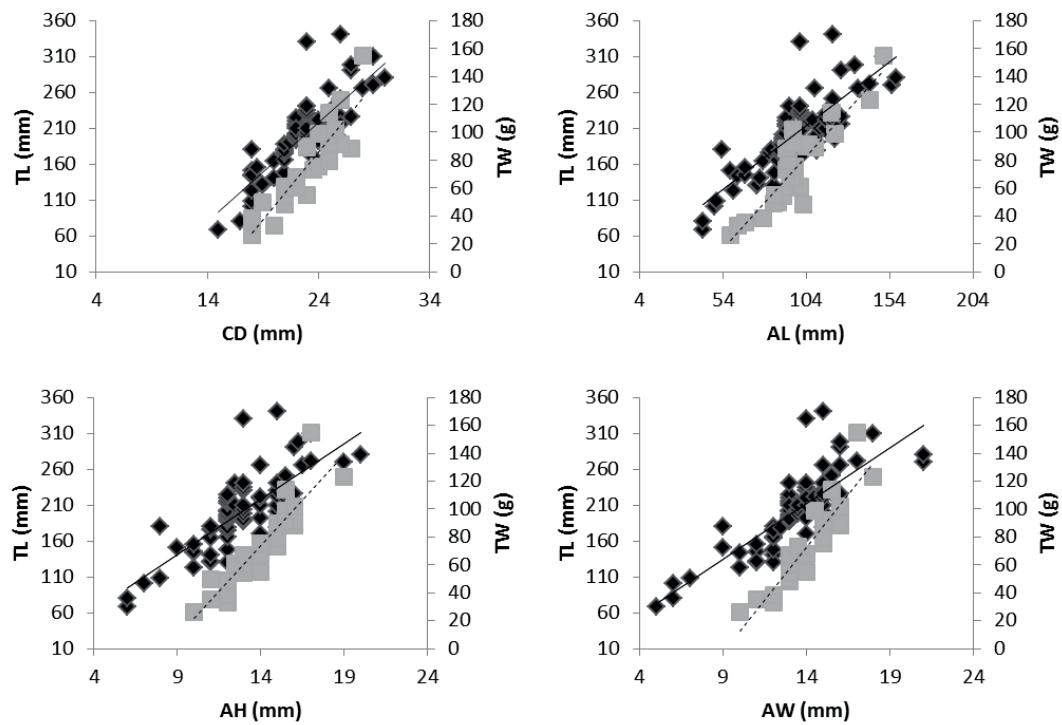


Figure 5:

Linear regression scatterplot of: central disk (CD), arm length (AL), arm height (AH) and arm width (AW) vs total length (TL, black symbols) and total weight (TW, grey symbols) for *O. ophidianus* variables.

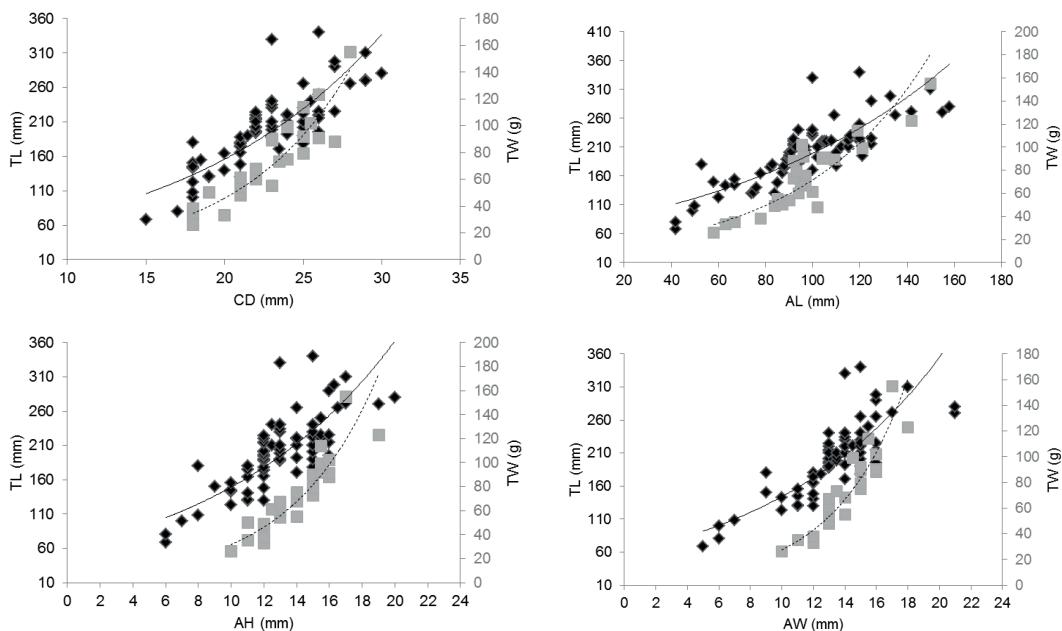


Figure 6:

Exponential regression scatterplot of: central disk (CD), arm length (AL), arm height (AH) and arm width (AW) vs total length (TL, black symbols) and total weight (TW, grey symbols) for *O. ophidianus* variables.

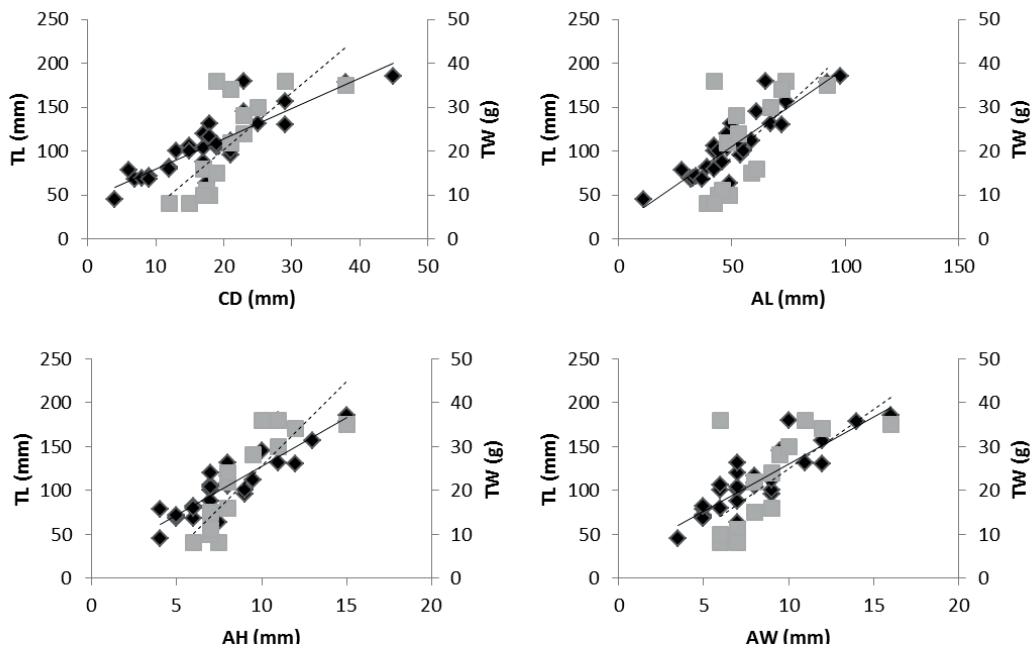


Figure 7:
Linear regression scatterplot of: central disk (CD), arm length (AL), arm height (AH) and arm width (AW) vs total length (TL, black symbols) and total weight (TW, grey symbols) for *C. tenuispina* variables.

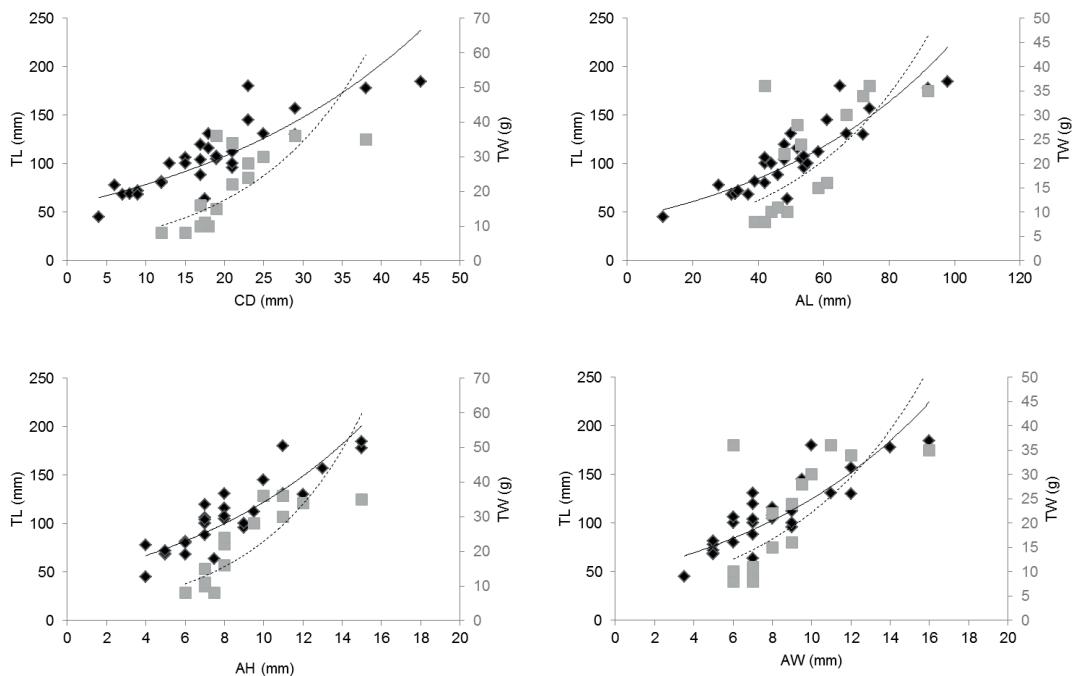


Figure 8:
Exponential regression scatterplot of: central disk (CD), arm length (AL), arm height (AH) and arm width (AW) vs total length (TL, black symbols) and total weight (TW, grey symbols) for *C. tenuispina* variables.

has to be partially rejected. Results obtained are in fact conflicting and vary among the three investigated asteroid species, probably as a result of their different morphology. In *C. tenuispina* and *M. glacialis*, AH performed better than AW, which exhibited weak correlations with both TL and TW, whereas in *O. ophidianus*, both AH and AW were strongly correlated with TW and weakly with TL.

Therefore, only one of the two novel proposed morphometric variables, AH, may constitute a practical and reliable method to estimate body size in starfish directly in the field. Furthermore, it is worth noting that AH was the only morphological variable in the three investigated asteroid species to register a good (i.e. $R^2 > 0.7$) correlation with both TL and TW in *C. tenuispina* and *M. glacialis*, two species that behave cryptically, hiding under boulders, in crevices and holes. Although there is no published evidence, we observed that these two species are quite sensitive to handling; for example they are often subjected to arm autotomy when pulled out from the crevices they hide in or when they are stretched (author's personal observation). Autotomy or self-amputation is a common self-defense mechanism designed in marine invertebrates to elude a predator's attack (see Fleming *et al.* 2007 and references therein). For asteroids, arm loss is a process that strongly impacts their biology. Arm loss, in fact, decreases their prey handling capacity, locomotion, growth, reproductive output and energy storage, and causes changes in depth distribution and feeding behavior of the same species (Diaz-Guisado *et al.*, 2006).

Our proposal to estimate the asteroid size directly in the field by measuring AH with the aid of a vernier caliper could be particularly rapid, simple, and not invasive for species characterized by cryptic behaviour. Since only a small portion of the body is analyzed, as compared to the assessment of total length or weight, even individuals exposing only part of one arm close to the disc can be successfully scored, with no harm inflicted to the same individuals.

In general, the new morphological metrics are better correlated with TW than with TL; this result is a fairly surprising and interesting one in many respects. For example, it would not be necessary to collect and move specimens in the laboratory to gather information on weight as the AH- or AW-TW relationship has proved to function reliably in the field. A simple *in situ* AH or AW measurement would be sufficient to approximate asteroid weight, representing a novel and non-invasive tool for estimating starfish biomass. Temporal variation in AH or AW in tagged starfish individuals could also be monitored in this way, allowing workers to analyze the reproductive cycle of asteroid species, in relation to

variations in resource allocation, and thus weight, without any dissection of the same specimens. Furthermore, in species such as *O. ophidianus*, characterized by arms of the same length, AW measurement could be useful for studies using imagery to generate *a posteriori* biomass estimates.

The three asteroid species analyzed in this study differ widely in their morphology and arm number; therefore, we expect that a similar correlation between body size metrics (TL and TW) and AH occurs in other starfish species. In conclusion, the novel morphological metric – AH – proposed as a proxy to estimate size and weight in sea stars, minimizes the operator's impact on asteroid specimens in the field and represents a rapid and reliable method.

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