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Original Article

A comparative study on the external parasites of *Oreochromis spp* from wild and cultured setting in Behera, Egypt

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Abstract:

A total one hundred (Tilapia spp) *Oreochromis spp* fishes were collected and examined for the presence of ectoparasites, fifty fishes from each farm and wild environment (River Nile) over a period of five months from February to July 2017. Total infestation rate of ectoparasites from cultured *Oreochromis spp* (100%) was higher than of those recorded in wild *Oreochromis spp* (74 %). The ciliated protozoan parasite *Trichodina spp* was the most abundant ectoparasite recovered from both cultured and wild environment. However, the study revealed two kinds of external parasites, protozoa and monogenetic trematodes. Protozoans showed the most prevalent ectoparasites (100%) in cultured *Oreochromis spp* and (74%) in wild *Orechromis spp*, while the monogenetic trematodes recorded (20%) in cultured fishes only while wild fishes showed no infestation. Furthermore, the load of protozoans in cultured *Oreochromis* differed significantly (P< 0.001) from those of wild *Oreochromis spp*. Concerning, infestation rate of each parasite, the study uncovered that *Trichodina spp*, *Chilodonella spp*, *Apiosoma spp* and *Myxobolus spp* lodged the wild *Oreochromis spp* differ significantly (P < 0.05) from those of cultured type in contrary, the only *Ambiphrya spp* did not differ significantly (P > 0.05). As well as monogenetic trematodes recovered from cultured fishes were differing significantly (P< 0.001) from those recovered from wild fishes but this difference was applied only on *Dactylogyrus spp* since the *Gyrodactlus spp* did not show any significant.

Key words: external parasites, Oreochromis, wild, cultured, Egypt.

Introduction:

Aquaculture is one of the most economically effective policies all over the world. Fishes are one of the most valuable and nutritional resources of human beings. (Banerjee and Bandyopadhyay, 2010). Fish parasites are the major factor of aquatic biodiversity and their monitoring is very essential for health management (Adeogun et al. 2010). Parasites can damage flesh, skin and the condition of the fish resulting in reduction of its production and sale value. (Kayis et al. 2009). Furthermore, synergism between bacteria and parasites represent high potential for high mortalities in the aquacultures where parasites play a crucial role for bacterial invasion (Steigen et al. 2013 and (Saptiani et al. 2017). Fish parasites can affect growth rate by inducing diseases which cause decline in fish production, and so, increase costs for disease control and decrease overall income of fish farms (Pantoja et al. 2012). Therefore, detection of fish parasites has a great importance not only to fish health but also to understand ecological problems of aquatic environment. (Sures, 2001). So that, the question raised, do the quality of aquatic environment (whether cultured or natural sources) affect the parasites population and disease occurrence? Many determents factors are incriminated to make fish prone to diseases. Such as a broad factor, known as environmental stress, any change of water quality consider as stressors such as hardness, ph, dissolved oxygen, ammonia and gas content (Meyer et al. 1983). For instance, some kinds of toxic algae bloom that enforce fishes to escape to better water quality but in farm fish there is a limited space and high density of fish, both together make fish prone to disease (St-Hilaire, 1998). As well as, crowding, confinement, handling and transport of cultured fish are considering a physical stressor, in addition to the skin abrasions and other surface lesions that resulted from these managements make gates for pathogens entrance (Strange et al. 1978). Moreover, the amount of organic materials consider important factor for survival and propagation of pathogens (Paclibare et al. 1994). Generally, the aquatic environment within farm setting is more vulnerable to pathogen than those of wild environment (St-Hilaire 1998). Ectoparasites that live on the body of fish are composed of Protozoa and Monogenea. According to their sites certain species such as Ciliate and Monogenea class found on the gills, while other species prefer to live on the skin and fish fins (Bruno et al. 2006). Most of wild populations of fishes are attacked by parasites without clear drastic harms in majority of cases (Robert, 2001). Parasites of wild fishes are only spotted for consumers or fisherman when they are very evident (Roberts, 1995) therefore, fishes are rejected. On other hand, parasites in cultured system trigger severe outbreak of disease because confinement of fish population under certain environment lead to increase intensity of parasite population (Roberts et al. 2000). Parasites give an indication of water quality while the parasites propagate in number and diversity in polluted water (Poulin, 1992 and Avenant-Oldewage, 2001). Walakira et al. 2014 revealed a high incidence of monogenetic trematodes infesting *Tilapia spp* in farms that particularly use organic fertilizers (animal manure) in Uganda. Meanwhile, they found that *Gyrodactlus* were represented by three worms per each fish as well as they uncovered reverse relationship between size of fish and infestation. In contrary, they discovered a low prevalent of ciliated fauna like *Trichodina spp* and *Ichthyopthirius multifilis* in Tilapia and Catfish. Likewise, (El Amin and Al-Harbi, 2016) reported major factors such as quality of nutrition and water affect the occurrence and diversity of ectoparasites. External parasites are the most common parasites that affect wide range of fishes in our region therefore; external parasites were our choice to study. As well as, we aim in this work to monitor external parasites of both wild and cultured *Oreochromis spp*, in Behera province, and showing the difference of ectoparasites population in both wild and cultured *Oreochromis spp*.

Material and methods:

Collection of samples: A total of 100 fresh caught live fish specimens Tilapia species. (*Oreochromis spp*.). 50 were collected from River Nile resources (wild), Rashid branch, as well as 50 from fish farms at Behera

Province during period from February till July 2017, fishes were transported to the laboratory of Parasitology, Faculty of Veterinary Medicine, Alexandria University in large plastic bags filled with water from the same sources.

Laboratory examination of fish: The specimens under investigation were grossly examined for detection of any external lesion or visible cysts. Then the fishes were scarified by inserting needle or scissor just posterior to head severing the spinal cord. Prepared specimens from skin, fins and gills were examined immediately after fish death to avoid the disintegration of the external protozoa.

Examination of skin and fins: The fish was put on dissecting dish, scraped with cover slide, another slide or scalpel blade from just behind opercula (gill cover) to the tip of the tail, fin, scales and mucus were transferred to the slide.

Examination of the gills: The operculum was removed with scissors, to expose gills. The gills were removed from the body and each branchial arch was best dissected separately with naked eye and by a magnifying lens to detect the presence of protozoal cysts and parasitic crustacae. Gill arch was removed in petri dish containing tape water and examined under the dissecting microscope to detect any monogenea or crustacae. Smears were taken from the body surface, fins and gills, speared on a dry clean slide, air dried and fixed with absolute methyl alcohol for 5 minutes.

The fixed smears were stained with freshly prepared Giemsa stain for 30 – 45 minutes after which the smears rinsed with tap water and left to dry in air according to **(Lucky, 1977).**

The isolated gill worms were stretched between slides and cover slip and fixed by glycerin – alcohol (in ratio of 1: 4). Then cover slip was framed with dense Canada balsam and kept in horizontal positiontill examinations, **(Lucky, 1977).**

Statistical analysis:

Statistical analyses were performed by Chi square test to compare diversity of parasites from wild and cultured environment. A significance level of $p < 0.05$.

Results:

The examination of 100 *Oreochromis spp* fishes, 50 fishes from cultured system and 50 fishes from wild environment (River Nile) during period between February and July 2017 revealed that Out of 50 examined cultured *Oreochromis spp*, 50 samples (100%) were infested with external parasites, alongside 37 (74%) samples of 50 wild *Oreochromis spp* were infested (table 1 & fig 1). These parasites can be identified as Protozoan parasites and Monogenetic Trematodes. Protozoans were *Trichodina spp* (some of Trichodins were identified as *Trichodina califorinica* **(Davis, 1947)** and other were not, *Chilodonella* hexastica **(Kiernik, 1909)**, *Apiosoma spp*, *Ambiphrya spp* and *Myxobolus spheroidalis* **(Abu EL- Wafa, 1988)** whereas the monogenetic Trematodes classified as *Dactylogyrus spp* and *Gyrodactlus spp* (table 2). Regarding each category of parasite in each environment, Protozoan parasites showed full infestation rate in cultured environment while counted 74% in wild fishes. On the other hand Monogenetic Trematodes were not found in wild *Oreochromis spp* but represented 20 % infestation rate from cultured *Oreochromis spp* (table 1 & fig 1). *Trichodina spp* showed the highly prevalent parasites whether in cultured or wild environment, since it recorded 100 % and 74 % in cultured and wild fishes respectively (table 2 & fig 2). Moreover, percentage of each protozoa load on cultured and wild *Oreochromis* fishes as well as Monogenetic Trematodes were displayed in table 2 and analyzed using Chi-square test to discover the significance between each category. It was clear that *Trichodina* is the most dominant parasite found in both environment but moreover, *Trichodina spp* in cultured fishes differed significantly (P < 0.001) from those of wild fishes. In contrary, each of *Chilodonella*, *Apiosoma*, *Ambiphrya* and *Myxobolus* reported a low percentage only on wild environment 8%, 10%, 2% and 14% respectively but there was no evidence for these parasites in cultured fishes. As well as *Chilodonella*, *Apiosoma*, and *Myxobolus* showed significance difference (P < 0.001) but *Ambiphrya* did not show significance among cultured and wild *Oreochromis* (table 2). Concerning the other category of parasites that recovered during examination of *Oreochromis spp*, only two Monogenetic Trematodes were reported, *Dactylogyrus spp* and *Gyrodactlus spp*. These were only found in cultured *Oreochromis spp* with absence of them in wild environment. The percentage of *Dactylogyrus spp* among cultured fishes was 14%, in contrast low percentage of *Gyrodactlus* 6% (table 2 and fig 2). It was clear that the burden of parasites on gills of both kind of environment were greater than of those collected from skin.

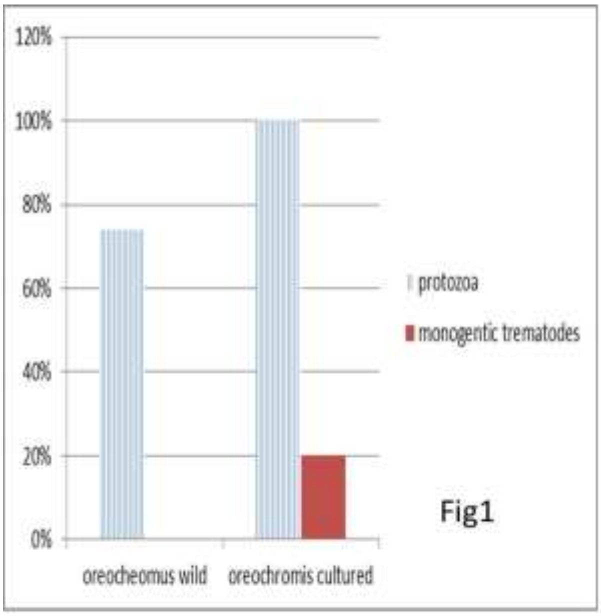
Table 1: Showing the overall infestation rate among cultured and wild *Oreochromis spp.*

Fish	No. of the examined fish	Total infestation Rate	Monogenetic Trematodes	Protozoa
Wild (River Nile) *Oreochromis spp*	50	37 (74 %)	0.00 (0%)	37 (74 %)
Cultured *Oreochromis spp*	50	50 (100 %)	10 (20%)	50 (100 %)

Table 2: Showing the infestation rate for each parasites and their significant difference.

Parasites	Wild (Nile) *Oreochromis spp*	Cultured *Oreochromis spp*	Chi-square significance
Protozoa	37 (74 %)	50 (100 %)	14.94 * (P<0.001)
Trichodina	37 (74 %)	50 (100 %)	14.94 * (P<0.001)
Chilodonella	4 (8%)	0.00 (0%)	4.16* (P<0.05)
Apiosoma	5 (10%)	0.00 (0%)	5.26* (P<0.05)
Ambiphrya	1 (2%)	0.00 (0%)	1.01 NS (P>0.05)
Myxobolus	7 (14%)	0.00 (0%)	7.53* (P<0.05)
Monogenetic Trematodes	0.00 (0%)	10 (20%)	11.11* (P<0.001)
Dactylogyrus	0.00 (0%)	7 (14%)	7.53* (P<0.05)
Gyrodactlus	0.00 (0%)	3 (6%)	3.09 NS (P>0.05)

*=means significant NS= means non-significant



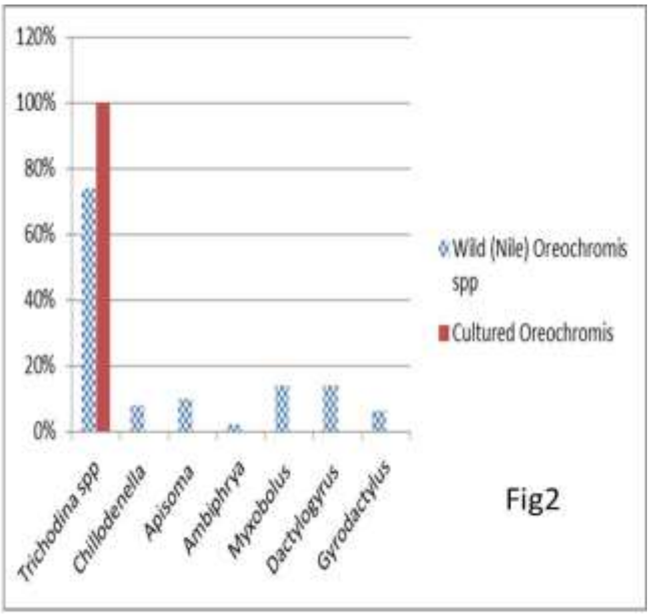


Figure 1: Showing the overall infestation rate among cultured and wild *Oreochromis spp.*

Figure 2: Showing the infestation rate for each parasite.

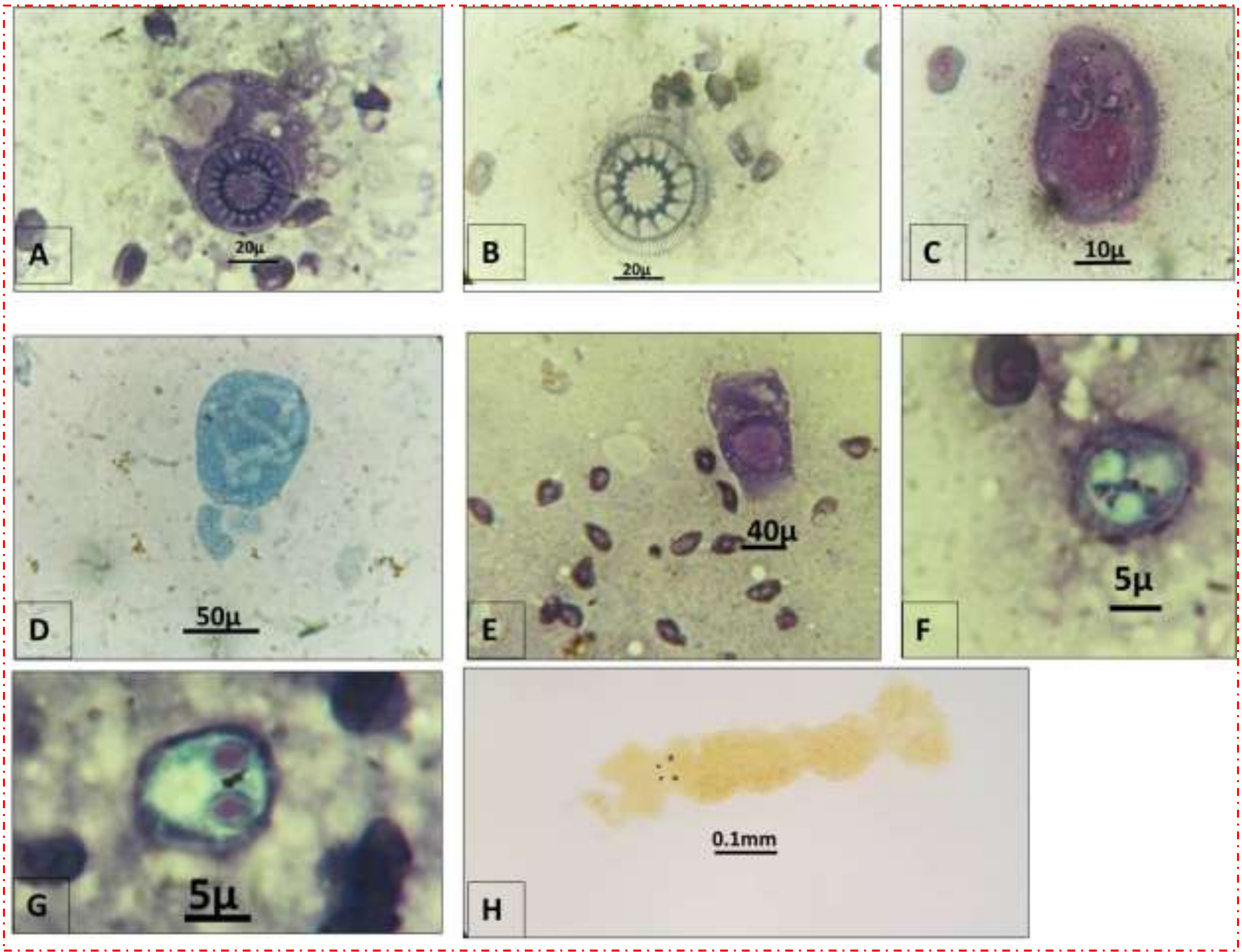


Figure 3: Showing Geimsa stained *Trichodina* californica (A) &Trichodina spp (B); *Chilodonella* hexastica (C); *Ambiphrya spp* (D); *Apiosoma spp* (E); Geisma stained *Myxobolus* spheroidalis (F&G) and non-stained *Dactylogyrus spp* (H).

Discussion:

As a contribution of the current argument of the role of water environment on the parasites burden, this work gave an evident indication of the impact of the environment on parasites population. Where, our study referred to the significant difference of ectoparasites between cultured and wild *Oreochromis spp*. Where, total infestation rate that found in cultured environment was the highest similar to (Alvarez-Pellitero et al. 1993) where they found that prevalence and intensity of ectoparasites were higher in cultured than wild one though the number of parasites species were higher in the wild. This finding also were completely agreed with our work where this study revealed that *Myxobolus, Chilodonella, Apiosoma* and *Ambiphrya* were only present in wild fishes however intensity of total ectoparasites are high in cultured. Individually, *Trichodina spp* was the most abundant ectoparasites in both types but the statistical analysis provided significant increase of this parasite in cultured over the wild, like what mentioned by (El Amin and Al-

Harbi, 2016) this also might be attributed to the parasite is commensal and convert only to pathogenic under stress factor like polluted water (Eissa 2002). In contrary to low prevalence of *Trichodina* and *Ichthyophithrius spp* in *Oreochromis* and catfish were observed (Walakira et al. 2014). However, both results from outside view seem divergent, but both of them agreed with each other while both of them depend on its evaluation of abundance and scarce of *Trichodina spp* the nutritional factor. As well as *Myxobolus spp* was identified only in wild environment similar to the findings of (Alvarez-pellitero et al. 1993) though of some species of this parasites represent a potential pathogen in wild and cultured system (Lom and Dyková, 2013). *Chilodonella* has been recorded in wild fishes only which agreed with (Silva et al. 2011) who have found *Chilodonella sp.* on an amazon wild fish *Oxydoras niger,* but (Eiras et al. 2012) Reported the absence of *Chilodonella species* on Brazilian farmed fish in contrary with (Pádua et al. 2013) that recovered them from cultured fishes in brazil. In addition to theory recommended that *Chilodonella* become dangerous only after subjected to debilitating factor (Schaperclaus, 1935). Abdel-Baki et al. (2014) recorded *Ambiphrya* for the first time in cultured Tiliapia in Saudi Arabia while our work showed low infestation rate of the parasite only in wild *Oreochromis spp* in contrary Infections by *Ambiphrya* and *Vorticella* are common in many cultured fishes (Basson, and Van As, 2006). This might contributed to unsuitable environmental parameters that diminish the growth of the parasites. However, the parasites provide an indication for water quality, several conflicting theories have been emerged each with different opinion. According to (Marcogliese, 2005; Hudson et al. 2006), considered, unpolluted water environment showed a greater intensity of parasites population whereas water with pollution especially chemical diminish the parasites diversity. To great extent our study come in the same way with this theory where we found much intensity of parasites in cultured environment where it assumed free off any chemical pollution in contrast the load of parasites in wild (River Nile) showed less diversity that recommend this theory where River Nile is assumed to be contaminated with industrial refuses. Other field investigation refuted this theory (Dzika and Wyżlic, 2009) where they found that some species of Monogenetic Trematodes react with the pollution and in turn decrease in richness and abundance while some other species react and increase in numbers. In the same context, result obtained by (El Amin and Al-Harbi, 2016) stated that major factors such as water quality and nutritional qualities affect ectoparasites abundance as well as temperature and seasonality and density played a secondary role in the occurrence of ectoparasites in cultured *Oreochromis niloticus*. Likewise, Monogenetic Trematodes incidence was prevalent in farm fish especially that use organic fertilization (animal manure), (Walakira et al. 2014) this come in agreement with our result obtained from cultured environment (20%) since wild environment do not receive fertilizers but in contrary, the diversity and abundance of ciliated protozoa (*Trichodina* and *Ichtyophirius multifilis* was low prevalent dissimilar with our work which recorded 100 % for protozoa especially *Trichodina*. In addition to captivation and density of fish in cultured setting induce drastic outbreak among fish population and increase parasites abundance as well

(Roberts et al. 2000). Gills were noticed to be loaded with the greatest numbers of the ectoparasites whether on cultured or wild fishes. This might returned to the function of gill that filter feeding and the site of gaseous exchange (Emere and Egbe , 2006 and Omeji et al 2011). Another theory is adopted according to (Somerville, 1984) who attributed the abundance of protozoa in gills because the sieving ability of gill rakers those prevent the parasites to pass. It concluded that the effect of kind of environment on abundance of parasites is controversial issue therefore we aimed to illuminate on how the ectoparasites in cultured *Oreochromis* differ from wild *Oreochromis spp*. As well as relationship between prevalence of parasites and type of environment is a multifactorial dependent, chemical pollution, organic materials, transportation, handling, crowding, captivity, temperature and other water parameters like ph., ammonia and dissolved oxygen and so on. Unfortunately, there is unclear and mysterious association between parasites and environmental parameters, sometimes the parasites abundance come up with adverse environmental parameters and sometimes it comes down thus there is more than one crossed factor that determine the parasites profusion.

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الملخص العربي

دراسة مقارنة علي الطفيليات الخارجية التى تصيب سمك البلطى فى كلا من المزارع والنيل فى محافظة البحيرة, جمهوريه مصر العربيه

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تم تجميع عدد مائة سمكة بلطى (50 سمكة من المزارع و50 سمكة من النيل) على مدار 6 شهور فى الفترة من فبراير 2017 الى يوليو 2017 وتم فحصهم لتصنيف الطفيليات الخارجية الموجودة. وكان معدل الإصابة بالطفيليات الخارجية فى اسماك المزارع 100%, وكان أعلى من معدل الإصابة فى اسماك النيل والتى بلغت 74%. وسجل طفيل الترايكودينا أعلى نسبة انتشار فى كل من اسماك المزارع واسماك النيل. وبالرغم من أن الدراسه اسفرت عن وجود نوعين من الطفيليات الخارجيه متمثلين فى الأوليات (البروتوزوا) و والديدان المثقوبة وحيدة العائل (المونوجينك تريماتودا), الا ان الأوليات سجلت أعلى نسبة انتشار حيث وجدت بنسبه 100% فى بلطى المزارع وبنسبه 74% فى بلطى النيل. كما وجدت المثقوبات وحيدة العائل بنسبه 20% فى بلطى المزارع بينما لم توجد فى بلطى النيل، كما اختلفت نسبه انتشار الأوليات فى بلطى المزارع اختلاف جوهرى عن بلطى النيل. وأوضحت الدراسه أن نسبه انتشار طفيل التريكودينا, الكيلودينيلا, الابيزوما و الميكزوبولاس قد أختلف جوهريا فى بلطى المزارع عنه فى بلطى النيل. أما بالنسبة لطفيل الأمبيفريا فلم تسجل إختلافا جوهريا. اما المثقوبات وحيدة العائل فقد اختلفت جوهريا فى بلطى المزارع عن بلطى النيل ولكن هذا الاختلاف ينطبق فقط على طفيل الداكتيلوجيرس ولكن لا ينطبق على طفيل الجيروداكتيلاس.