وزارت جهاد كشاورزي

مؤسسه تحقيقات شيلات ايران

كميته ملي نظارت بر مبارزه با شانه دار مهاجم درياي خزر

نتايج و دستاوردهاي نخستين نشست فني منطقه اي امكان معرفي *Beroe ovata* به درياي خزر به منظور كنترل جمعيت شانه دار مهاجم *Mnemiopsis leidyi*

3 - 4 اسفند 1382

فهرست مطالب

پیشگفتار

در سال 1378 براي نخستين بار جانداري در درياي خزر مشاهده شد كه قبلا" در درياي سياه مشكلات زيادي را پديدار كرده بود (اسماعيلي و همكاران1378، ضميمه 1). اين جانور كه بصورت شناور و معلق در آب دريا زندگي مي كند و داراي بدني شفاف و ژله اي است و مي تواند بطور سيري ناپذيري از جانداران معلق و پلانكتوني ديگر تغذيه كند (باقري و همكاران 1382، ضميمه 2). جانور ژله اي تازه وارد داراي صفحات شانه مانندي است و از اين روي شانه دار *Mnemiopsis leidyi* ناميده مي شود. اين شانه دار مي تواند با سرعت خيلي زيادي توليد مثل كند، اصولا" موجودي دو جنسي بوده و هر شانه داري مي تواند به تنهايي توليد مثل كرده و در مدت كوتاهي تبديل به 6000 شانه دار گردد (شكل 1).


شكل 1. تصوير شانه دار مهاجم درياي خزر (*Mnemiopsis. leidyi*)

در مدت كوتاهي اين جانور به ظاهر زيبا توانست جمعيت پلانكتونهاي درياي خزر را تحت الشعاع قرار دهد و جانوراني كه از اين پلانكتونها تغذيه مي كنند دچار فقر غذايي شده و شديدا" كاهش جمعيت پيدا كنند (روحي و همكاران 1382، ضميمه 3 و قوليف 1382، ضميمه 4 و كارپيوك و همكاران 1382، ضميمه 5).

اين كاهش جمعيت خصوصا" در ذخاير كيلكا ماهيان خود را نشان داد و كم كم صيادان متوجه كاهش صيد خود گرديدند.

موسسه تحقيقات شيلات ايران كه بطور مرتب مشغول اندازه گيري و پايش ميزان و وضعيت صيد كيلكا ميباشد نيز متوجه تغييرات جدي در محيط زيست درياي خزر در اثر حضور فعالانه اين شانه دار مهاجم گرديد.

در گام نخست مي بايست مشخص مي گرديد كه تاثير حضور اين جانور چگونه بوده است و رابطه آن با كاهش صيد كيلكا چيست؟ در مطالعات اوليه گونه جانور مهاجم مشخص گرديد و با بررسي تاثيرات حضور آن بر روي محيط زيست درياي سياه نقش تعيين كننده شانه دار كاهش صيد روشن گرديد (كديش 1382، ضميمه 6).

در مرحله بعد روش بر خورد با اين مشكل مورد بررسي قرار گرفت. در ابتدا با وجودي كه تاثير آن مشهود بو د، دانشمندان شركت كنند ه در نخستين جلسه گروه مشاوران شانه دار در برنامه محيط زيست درياي خزر (First International Meeting “ The Invasion of the Caspian Sea by the comb jelly *Mnemiopsis*, problem, perspectives, need for action” Baku, Azerbaijan, 24-26 April 2001) ،بر اين اعتقاد بودند كه تا زماني كه ميزان زيتوده شانه دار به 2 كيلوگرم در متر مربع نرسد هنوز نميتوان گفت كه ميزان آن در حد خطر ناك است.

با گذشت يك سال ميزان آن از حد تعيين شده گذشت و در جلسه بعد اين گروه (First CEP Regional *Mnemiopsis* Advisory Group Workshop, Baku, Azerbaijan, 3-4 December 2001) اقدام لازم براي كنترل جمعيت اين جاندار ضروري تشخيص داده شد و مقرر شد كشور روسيه راههاي مبارزه با اين جانور را بررسي كرده و حمايتهاي لازم نيز توسط برنامه محيط زيست درياي خزر ارائه گردد.

در مطالعاتي كه در درياي سياه انجام شد مشخص شد كه امكان مبارزه شيميايي و يا مكانيكي وجود ندارد. همچنين براي مبارزه باكترياي و يا انگلي با اين جانور نياز اطلاعات زيادي وجود دارد كه در دسترس نبوده و ممكن است خطراتي را متوجه محيط كند. تنها راهي كه دانشمندان حاضر در جلسه درياي سياه و نيز در نخستين جلسه گروه

مشاوران شانه دار در برنامه محيط زيست درياي خزر با آن موافقت داشتند مبارزه بيولوژيک از طريق يک جاندار شکارگر بود.

در اقيانوس اطلس که زيستگاه طبيعي شانه دار مهاجم درياي خزر است علاوه بر تعدادي گونه ماهي و شانه داري بنام *Beroe ovata* از آن تغذيه مي کند (شکل 2).

شکل 2. تصوير شانه دار شکارگر (*Beroe ovata*)

از جمله موفق ترين ماهياني که از *M. leidyi* تغذيه مي کنند مي توان از کره ماهي (Butter Fish) نام برد. اين ماهي هم خانواده ماهي حلوا سفيد در خليج فارس است. همه اين ماهيان در کنار پلانکتوهاي ديگر از شانه دار نيز تغذيه مي کنند. به عبارتي تغذيه آنها از شانه دار بطور غير انتخابي است. همچنين امکان سازگار نمودن حلوا سفيد از شوري خليج فارس (42-37 قسمت در هزار) به شوري درياي خزر (13 قسمت درهزار) بسيار بعيد است. از طرف ديگر امکان موثر بودن آن در کاهش جمعيت شانه دار کم است به همين دليل کارشناسان گروه مشاوران شانه دار در برنامه محيط زيست درياي خزر نظر به استفاده از *B. ovata* بعد از انجام مطالعات لازم دادند.

به زودي مشخص شد که تحقيق انجام شده توسط محققين روسي ناموفق بوده و نتوانستند امکان زنده ماندن جانور شکارگر *Beroe ovata* در شوري درياي خزر را فراهم کنند.

چون بيشترين ضرر در خصوص تهاجم شانه دار مهاجم را ايران ديده بود و صيادان ما دچار مشکل جدي شده بودند از سال 1380 با هماهنگي هايي که انجام گرفت وزير محترم جهاد کشاورزي در يک گزارش وضعيت بحراني را به استحضار رياست محترم جمهور رساندند. ايشان به لحاظ حساسيت اين تهاجم موضوع را به جناب آقاي دکترعارف معاون اول رياست جمهوري

ارجاع دادند. بنا به دستور جناب آقاي دكتر عارف موضوع در معاونت هماهنگي امور علمي و اجتماعي معاون اول رئيس جمهوري مورد بررسي و پيگيري قرار گرفت كه متعاقب آن كميته ملي نظارت برمبارزه با شانه دار مهاجم درياي خزر با حضور نمايندگان سطح عالي دستگاههاي زيربط (دفتر معاون اول رياست جمهوري، وزارت امور خارجه، سازمان حفاظت محيط زيست، وزارت علوم تحقيقات و فن آوري، سازمان مديريت وبرنامه ريزي، سازمان بنادر و كشتيراني و وزارت جهاد كشاورزي) بمنظور نظارت و رهبري مبارزه با شانه دار مهاجم درياي خزر تشكيل گرديد. بنا به ابلاغيه معاون اول محترم رياست جمهوري مقرر شد دبيرخانه اين كميته در موسسه تحقيقات شيلات ايران تشكيل و مبارزه با شانه دار مهاجم درياي خزر توسط اين موسسه انجام گيرد و مقرر شد دستگاههاي مذكور نهايت همكاري را با اين دبيرخانه داشته باشند.

از فعاليتهاي مهم انجام شده كميته ملي نظارت بر مبارزه با شانه دار مهاجم درياي خزر علاوه برتشكيل جلسات متعدد براي رهبري ،نظارت، حمايت و پشتيباني فعاليتهاي انجام شده در جهت مبارزه با شانه دار مهاجم، برگزاري نخستين همايش ملي شانه دار درياي خزر در تاريخ 29 و 30 خرداد 1381 در پژوهشكده اكولوژي درياي خزر در ساري و نيز برگزاري نخستين نشست فني منطقه اي بررسي امكان معرفي *Beroe ovata* به درياي خزر در تاريخ 3 و4 اسفند1382 در سالن همايشهاي صدا و سيماي جمهوري اسلامي ايران مي باشد.

موسسه تحقيقات شيلات ايران جهت انجام مطالعات لازم براي مبارزه با شانه دار دست به كار شده و با انتقال *Beroe ovata* به آزمايشگاه پژوهشكده اكولوژي درياي خزر در ساري موفق به سازگار نمودن آن به شوري كم آب درياي خزر شد (روحي و كديش 1382، ضميمه 7).

در اين موقع نظرها و اميدهاي كارشناسان منطقه براي تحت كنترل درآوردن اين جانور مهاجم همه معطوف به ايران شده و حمايت تكنيكي برنامه محيط زيست درياي خزر نيز اين مطالعات را موثرتر نمود.

بعد از مطالعه روي امكان زنده نگه داشتن *B. ovata* در شوري كم آب درياي خزر (13 قسمت در هزار) نسبت به

شوري درياي سياه (28 قسمت در هزار) ، كارشناسان موسسه تحقيقات شيلات ايران با همكاري كارشناسان بين المللي در مورد توانايي تغذيه آن از درياي خزر مطالعه نموده و روشن گرديد كه نرخ تغذيه آن از شانه دار مهاجم درياي خزر مشابه نرخ تغذيه آن در درياي سياه است. به عبارت ديگر انتظار ميرود *B. ovata* در خزر نيز مثل درياي سياه بطور موثر جمعيت شانه دار مهاجم را كاهش دهد.

مطالعات ديگري بر روي تكثير مصنوعي *B. ovata* در آب درياي خزر انجام شدكه با وجودي كه امكان استحصال تخم و لارو ميسر گرديد ولي رشد لاروها مقدور نگرديد (فيننكو و همكاران 1382، ضميمه 8).

جهت بحث و تبادل نظر در مورد شانه دار مهاجم درياي خزر در 29 و 30 شهريور 1381 همايشي در سطح ملي در ساري برگزار شد و نتايج تحقيقات تا آن زمان در اختيار علاقه مندان قرار گرفت.

نتايج اين مطالعات در جلسه بعدي گروه مشاوران شانه دار برنامه محيط زيست درياي خزر CEP
(Second CEP Regional Invasive Species Advisory Group Workshop, 27 June 2002) مطرح گرديد و مورد استقبال قرار گرفت. در اين جلسه مقرر شد در موارد زير مطالعه صورت گيرد:

1- امكان تغذيه *B. ovata* از پلانكتونهاي خزر
2- امكان انتقال باكتري و يا انگل بهمراه *B. ovata* به درياي خزر

همچنين گزارشي در مورد تاثيرات احتمالي معرفي *B. ovata* به درياي خزر (Enivironmental Impact Assessment) تهيه گردد (شيگانوا 1382 ، ضميمه 9).

به همين منظور مطالعاتي در پژوهشكده اكولوژي درياي خزر انجام شد كه در نتيجه آنها مشخص گرديد كه *B. ovata* از جانداري بغير از شانه داران تغذيه نمي كند (جوانشير و شيگانوا 1382، ضميمه 10) و باكتريهاي آن در درياي خزر حضور دارند و جديد نيستند. همچنين انگلهاي *B. ovata* در فرايندسازگار كردن آن با شوري درياي خزر بعلت كاهش شوري از بين مي روند (سعيدي و همكاران 1382، ضميمه 11). در اين مقطع نيز مجددا" براي تكثير مصنوعي تلاش شد كه موفقيتي كسب نگرديد.

نتايج حاصل از اين مطالعات در " نخستين نشست فني منطقه اي بررسي امكان معرفي *B. ovata* به درياي خزر" كه در تهران در 3-4 اسفند 1382 تشكيل گرديد ارائه شد و دانشمندان شركت كننده همگي اظهار داشتند كه اين جانور هيچ خطري براي اكوسيستم درياي خزر نداشته و مي تواند براي تحت كنترل در آوردن شانه دار درياي خزر مورد استفاده قرار گيرد (ضميمه 12 بيانيه نشست و ضميمه 13 اسامي شركت كنندگان در نشست).

در شرايط حاضر تحقيقات در حدي كه مشخص كننده راه مناسب براي كنترل جمعيت شانه دار مهاجم انجام شده است و با برپايي نشست هاي علمي در سطح كشور و منطقه اطلاع رساني در حد مطلوب انجام شده است. براي ادامه فعاليت مي بايست ابتداء نظرات علمي كه به اجماع خوبي رسيده است در اختيار مقامات كشورهاي حاشيه درياي خزر قرار گيرد، كه با همكاري برنامه محيط زيست درياي خزر و كميسيون منابع زنده درياي خزر و وزارت امور خارجه جمهوري اسلامي ايران، عملي خواهد شد. در مقطع بعدي نظر مساعد دولتها براي معرفي *B. ovata* به درياي خزر مي بايست كسب گردد. اين اقدام نيز جنبه سياسي و اجرايي دارد. بعد از موافقت همه دولتها براي معرفي *B. ovata* به درياي خزر مي بايست از سه راه اقدام نمود كه عبارتند از : معرفي در ايران، روسيه و جمهوري آذربايجان.

معرفي اين جانور به درياي خزر به نظر ساده مي رسد ولي مطالعات و بررسي هاي اوليه زيادي را در بر مي گيرد و بعد از معرفي، پايش و بررسي تاثيرات در حال انجام بسيار ضروري مي باشد (نگارستان 1382، ضميمه 14).

نقل از مجله علمي پژوهشي علوم و تكنولوژي محيط زيست، شماره 3 ، زمستان 1378

گزارش مشاهده اولين مورد از شانه داران درياي خزر در سال 1378

عباس اسماعيلي ساري، صابر خدابنده ، بهروز ابطحي ، جعفر سيف آبادي ، هادي ارشاد
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واژه هاي كليدي : گزارش ، درياي خزر، شانه داران ، سلانتره ها، تانتاكولداران

چكيده

طي مطالعات تحقيقاتي درخصوص شناسايي ، بررسي تراكم و پراكنش سانتره ها در سواحل جنوبي درياي خزر، تيم تحقيقاتي دانشكده منابع طبيعي و علوم دريايي دانشگاه تربيت مدرس در بهمن ماه سال 1378 طي نمونه برداري هاي مقدماتي خود در ايستگاه ساحل غازيان و نور به وجود آبزيان متعلق به شاخه جديدي از بي مهرگان در اين دريا پي برد. درابتدا افراد اين شاخه شك برانگير با سلانتره هاي گزارش شده مهاجر و بومي درياي خزر برخورد نمودند، اما بعد از مطالعه و مشاهدات دقيق آزمايشگاهي، مشخص شد كه اين آبزيان متعلق به رده تانتاكولداران 1 از شاخه شانه داران2 بوده و قبلا" وجودشان در درياي خزر گزارش نشده است.

مقدمه

درياي خزر از نظر فراواني گونه هاي مختلف جانوري و پراكندگي آنها، يكي از درياي هاي قابل توجه جهان است. تحقيقات و مطالعات انجام شده روي پلانكتون هاي خزر شمالي و مياني نشان مي دهدكه پنج گونه مهاجروبومي ازشاخه كيسه

تنان [3] در این دریا وجود دارد (1) . مطالعه پراکنش و تراکم افراد این شاخه در سواحل جنوبي درياي خزر، به صید گونه هایي از رده تانتاکولداران متعلق به شاخه شانه داران منجر شد.

شانه داران مذکور که زیستگاه اصلي آنها سواحل شرقي آمریکاي شمالي و جنوبي است، وجودشان دردرياي سیاه وآزوف براي اولین بارتوسط Perelado درسال1982گزارش شد (2).

1-Tentaculata

2- Ctenophora

3- Phylum Coelantrata

براساس گزارش CESAMP [1] پس از ظهور این آبزیان مهاجم در این دریاها، صدمات فراواني به ذخایر ماهیان وارد شد، به طوري که میزان صید شوري سابق از این دریاها از دویست و پنجاه هزارتن به کمتر از سي هزار تن کاهش یافت. کاهش میزان صید ترکیه نیز قابل ملاحظه بود (2).

تحقیقات مشابه در سواحل روماني در درياي سیاه، کاهش جمعیت زئوپلانکتوني را به میزان 80% نشان مي دهد (2 و 3).

مقادیر اندازه گیري شده از بیوماس شانه داران مهاجم در درياي سیاه تا حدود 3 کیلوگرم در مترمربع و در درياي مرمره حتي تا 12 کیلو گرم در متر مربع اندازه گیري شده است (2).

شانه داران تقریبا" از یکصد گونه جانور شنا کننده آزاد با بدن هاي ژله اي تشکیل مي گردند. اینها را غالبا" لرزانك هاي شانه اي [2] مي نامند، زیرا بدنشان داراي صفحات شانه اي شکل مي باشد. برخي از آنها داراي برجستگي طولي هستند و به نام گردوهاي دریایي [3] شناخته مي شوند (4 و 5).

شانه داران قبلا" با کیسه تنان (گزنه داران و سلانتره ها) طبقه بندي مي شدند ولي به علت داشتن خصوصیات ساختماني و بیولوژیك خاص ، چندین سال است که در شاخه جداگانه جاي گرفته اند. این آبزیان در دریاهاي گرم فراوانند و با اشکال مختلف گرد، بیضي و حتي لوزي دیده مي شوند. برخي از آنها در نواحي معتدله یافت مي شوند و بیشتر سطح زي مي باشند. برخي نیز در اعماق مختلف حت تا 3000 متري دیده مي شوند (5).

شانه داران در آب به حالت عمودي بوده و مي توانند به كندي شنا كنند. جريان ها و يا جزرو مدها قادرند آنها را به تعداد زيادي، پهلوي هم جمع و متمركز نمايند. در تاريكي از زير صفحات شانه اي آنها تابش هاي حياتي ساطع مي شود. غذاي آنها شامل جانوراني مانند نرم تنان و سخت پوستان كوچك و همچنين تخم و لاروي ماهيان مي باشد (2 و 4 و 5). اين جانوران با استفاده از كلوبلاست 4 ، جانوران كوچك را شكار مي كنند. گوارش در آنها سريع بوده و در غذا خوردن بسيار حريص مي باشند (5و6).

شانه داران پيوسته در حال تغذيه هستند و به طور مرتب غذاهاي نيمه هضم شده را ازدهان به بيرون رانده و به اين جهت در كاهش غذا، تخم و لارو و ماهيان نقش مهمي دارند (2و4و7).

مواد و روش بررسي

همانطوري كه قبلا" اشاره شد، صيد اتفاقي شانه داران در خزر، در پي انجام نمونه برداري هاي مقدماتي طرح سلانتره (ژله ماهيان) صورت گرفت. براي پيدا كردن روش و ابزار مناسب براي صيد ژله ماهيان، نمونه برداري به طور آزمايشي با تورهاي زئوپلانكتون گير و ترال كف دست ساز در چهار ايستگاه شامل خليج گرگان، سواحل شهرستان نور، بندرانزلي و ساحل غازيان طي ماه هاي دي ، بهمن و اسفند 1378 انجام گرفت.

1-Group or Experts on the Sclantlric Aspects of Marine Environmental Protection
2- Comb jetties 3- Sea Walouts
4-Collobtasts

نمونه برداري در خليج گرگان از عمق دو متري با استفاده از تور زئوپلانكتون، در ساحل نور از عمق 4 متري بااستفاده ازتورفوق وهمچنين تور ترال كف با چشمه يك سانتي متر انجام گرفت. براي نمونه برداري از ساحل غازيان و بندرانزلي تنها از تور ترال كف در اعماق 15 تا 20 متري استفاده شد.

نمونه ها بعد از صيد، بلافاصله به صورت زنده به آزمايشگاه منتقل و مورد مطالعه واقع شدند. پس از انتقال نمونه ها به آزمايشگاه بيولوژي دانشكده علوم

دريايي نور از نمونه ها در درون آب بشر و آكواريوم عكس و همچنين فيلم تهيه گرديد.

براي مطالعه و شناسايي دقيق، نمونه ها بعد از تثبيت در فرمالين4% با استفاده از لوپ دو چشمي و ميكروسكوپ Invert مورد مطالعه قرار گرفتند. شانه داران با داشتن خصوصيات ساختماني و بيولوژيك زير در شاخه جداگانه از ژله ماهيان جاي دارند (9و4و8).

1- هشت رديف صفحه شانه اي
2- عضلات مزانشيمي يا مزودرمي
3- وجود تشكيلات عالي دستگاه گوارش
4- يك ناحيه حسي مقابل دهان
5- نداشتن سلولهاي دفاعي (نماتوسيست)
6- حركت در شانه داران بر خلاف مدورهاي سلانتره ها از طرف دهان انجام مي گيرد.

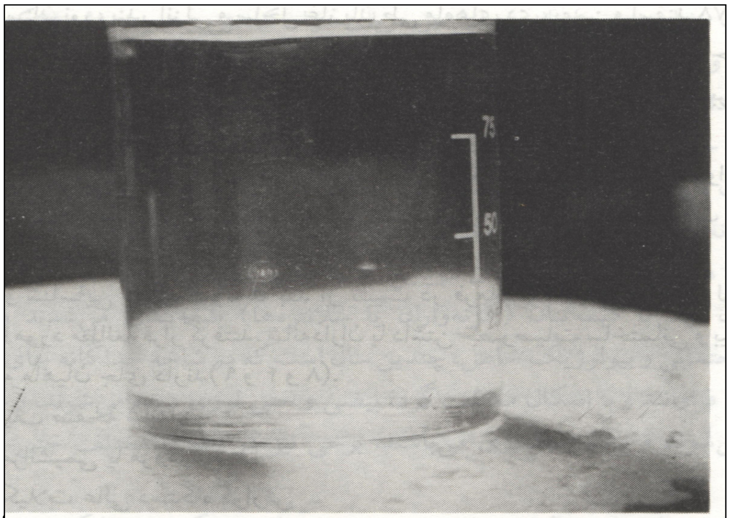
شانه داران داراي رده اي به نام Tentaculata مي باشند. افراد اين رده داراي تانتاكولهاي در اطراف دهان بوده، برخي داراي بدني گرد با تانتاكول هاي شاخه شاخه و قابل جمع شدن در غلاف هستند، مثل Pleurobrachia و در برخي بدن از طرفين اندكي فشرده و داراي دولب دهاني بزرگ با تانتاكول هاي بدون غلاف مي باشند مثل Mnemiopsis.

نتايج

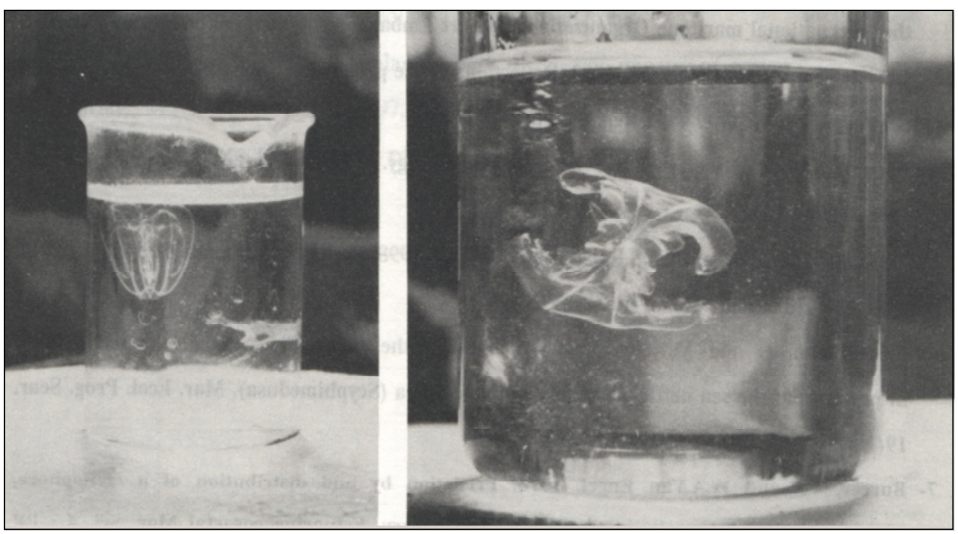
در نمونه برداري هاي دي ماه سا ل1378، تراكم سلانتره هاي صيد شده (عروس ماهيان) زياد بود. اما در ماه بهمن و خصوصا" اسفند همان سال، ميزان صيد بسيار كمتر و در حد چند نمونه بود. حين مطالعه نمونه هاي صيد شده در بهمن ماه از ايستگاه غازيان و نور، جانوران جديدي در بين عروس ماهيان مشاهده شدند كه در ابتدا به دليل شباهت به عروس ماهيان [1] شك برانگيز با سلانتره هاي مهاجر بومي گزارش شده در خزر بود. ولي بررسي هاي بعدي و مطالعه خصوصيات مرفولوژيك آنها مشخص كرد كه اين آبزيان متعلق به شاخه شانه داران بوده و قبلا" در درياي خزر گزارش نشده اند (شكل 1 و 2).

رجوع به منابع درخصوص بي مهرگان و زئوپلانكتون هاي درياي خزر و همچنين تماس و تبادل نظر با كارشناسان علوم آكادمي روسيه و همچنين آستاراخان، عدم گزارش مبني بر وجود شانه داران در درياي خزر را تاييد نمودند. بنابراين وجود افراد اين شاخه در درياي خزر براي نخستين بار توسط اين تيم تحقيقاتي گزارش و فيلم آن از شبكه سيماي جمهوري اسلامي ايران (دوم ارديبهشت ماه 79) پخش گرديد.

1- Jelly fish



بررسي هاي اوليه، تعلق شانه داران مزبور را به جنس گونه هاي مختلف نشان مي دهد (شكل 1و2) اما براي شناسايي دقيق جنس و گونه نمونه ها، تيم تحقيقاتي، مكاتباتي را با كارشناسان موزه تاريخ طبيعي كانادا و آكادمي علوم روسيه انجام داده كه نتايج آن منتشر خواهد شد. پس از انتشار گزارش هاي اوليه پيرامون مشاهده و شناسايي شانه داران در درياي خزر، اظهار نظرهاي پراكنده اي از طرف پاره اي منابع پيرامون ژله ماهيان بومي و مهاجر درياي خزر صورت گرفت كه ارتباطي با شانه داران موجود در شاخه هاي جداگانه از ژله ماهيان ندارد.



پيشنهادها

مورد توجه قرار گرفتن ورود شانه داران مزبور به درياي سياه و آزوف و انجام تحقيقات مستمر و مشترك بين چندين تيم تحقيقاتي از كشورهاي مختلف (GESAMP)، اهميت موضوع را روشن ساخته، مطالعه همه جانبه شانه داران مهاجم درياي خزر، تغذيه، اكولوژي، پراكنش و ديناميك جمعيت و تاثير بر ذخاير آبزيان اقتصادي و همچنين ارائه راهكارهاي لازم براي كاهش صدمات احتمالي را ضرورت مي بخشد. در اين راستا دانشكده منابع طبيعي و علوم دريايي دانشگاه تربيت مدرس ، آمادگي خود را براي همكاري گسترده با نهادهاي اجرايي ذيربط اعلام مي دارد.

اميد است قبل از تكرار تجربه تلخ درياي سياه و آزوف در خزر با ارزش ، تحقيقات لازم و ارائه راهكارهاي مناسب انجام گيرد.

منابع

1-يابلونسكايا. ي. آ و همكاران . 1985. درياي خزر فون و توليدات زيستي آن (آكادمي علوم شوروي) . ترجمه ا. شريعتي 1371. موسسه تحقيقات شيلات ايران . تهران .

2- IMO/FAD/UNFSCO-IOC/WMO/WHO/LAEA/UN/UNEP.(1996) opportunistic settlers the international maritime organization 4 Albert Embakment London SEI 7SR
3- Christensen V., and Caddy J.F. 1993. Relection of the pelagic food web structure in the Black sea, Annex IV in FAO Fish Rep. 495; pp.60.77.
4- Barens, R. and F. Rappert. 1994. Invertebrate zoology, Sixth edition. W. B. Saunders & Co., Philadelphia, PA.
5- Cleveland P. JR Hickman, L. S. Roberts and L. Alian 1998. Biology of Animals, copy right by th MCGraw-Hill companions, inc. pp. 392-415.
6- Feigenbaum, D. and M. Kelly 1984. Changes in the Lower Chesapeake Bay food Chain in presence of the sea nettle chry saora quinquecirrba (scyphimedusa) Mar. ECO1 prog. Sear. 19(1-2): p 39-47.
7- Barrev, V. G. and W. A. Van Eugel. 1976. predation by distribution of a ctenophore, Mnemiopsis leidyi Agassiz, in the york River estnary. Estuarine cosastal Mar, Sci, 4: pp 242-255.
8- Fewkes, J. W. 1880. studies of the Jelly-Fishes of Narragansett Bay, Bull. Mus. Comp. 2001. 8 (8) : pp 141-182.
9- Aria, M.N. 1988. Interactions of fish and pelagic coelenterates. Con. J. 2001.66: 1913-1927

THE STUDY OF CTENOPHORE (*MNEMIOPSIS LEIDYI*) FEEDING IN THE IRANIAN COASTS OF THE CASPIAN SEA

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Key word: Caspian Sea, *Mnemiopsis leidyi* , Zooplankton, phytoplankton

Abstract

The Survey was from 2002 to 2003 in the Iranian Coasts of the Caspian Sea. The gut contents of *Mnemiopsis*in the Caspian Sea include a wide variety of prey, Whit 84 % of items being Zooplankton prey. The maximum Zooplankton prey was Copepoda (34 %) and Minimum prey was Cladocera with 0.75 percentage . In this study we showed fish eggs (% 3) and Mollusca (4.2%). Mnemiopsis consumed Phytoplankton, Maximum items prey was Chrysophyta with 67 % and minimum was showed Pyrrophyta (3.7 %). The highest prey of *M.leidyi* observed young specimens and the lowest prey have been adult *M.leidyi*. In this study, We have Showed Ctenophore cannot digest phytoplankton.

Introduction

In the 1980s, the introduction of a new species (a lobate ctenophore, *Mnemiopsis leidyi*) into the Black Sea radically affected the whole ecosystem (Vinogradov 1989; Kideys 1994). This species had a negative impact on the most dominant fish of the Black Sea, the anchovy *Engralius* *encrasicolus*, though competion for the edible zooplankton as well as consumption anchovy eggs and larvae in the Black Sea. The mass occurrence of *Mnmiopsis* was one of the most important reasons for the sharp decrease of anchovy and other pelagic fish stocks in the Black Sea (Kideys 1994).

Meantime possibility of its introduction into other neighbouring sensitive ecosystems, notably the Caspian Sea, has been mentioned (Dumont 1995; GESAMP 1997). And, as expected, this ctenophore reported from the Caspian Sea by November 1999 (Ivanov et al.2000).

Ivanov et al. (2000) suggested that this ctenophore were transported with ballast water taken aboard in the Black Sea or the Sea Azov (where *Mnemiopsis* occurs in warm months) and released after ballast-loaded ships passed through the Volga Don Canal and the shallow freshwater North Caspian Sea, into the saltier Central or South Caspian.

Since *M.leidyi* is a voracious predator on zooplankton, catches of the main zooplanktivorous fish, Kilka (Clupeonella spp.), are reported to countries since 2001 (kideys et al. 2001a,b). For example, the Kilka (Clupeonella spp.) catches of Iran initially dropped to 64-thousand tones in 2000 and to 45-thousand tones in 2001, from 82 – and 83-thousand tones in 1998 and 1999,respectively (kideys et al. 2001a). So within 2 years, an almost 50 % decrease in the Kilka catches of Iranian fishermen has occurred, with a minimum of 15 million Us dollars in economic loss. Similary, Azerbaijan Kilka catches dropped to 9 thousand tones in 1999 and 18-thousand tones in 2000 (Kideys et al. 2001b). So, for Azeri fisheries, this too represents about a 50% decrease in the Kilka catches between 1999 and 2001. Russia catches have also been reported to have decreased remarkably (Shiganova et al. 2001a) .

Due to damage observed in the Black Sea, there has been a fast response over the presence of *Mnemiopsis* in the Caspian Sea. Since *Mnemiopsis* is a voracious predator on zooplankton, both abundant small pelagic fishes feeding on zooplankton as well as large predators feeding on these fishes such as white sturgeon (*Huso huso*) and endemic Caspian Seal (P*hoca caspica*) would be under significant threat in the Caspian Sea (Kideys et al.2001).

Despite recognition of the problem, there is not published study on the *M. leidyi* feeding in the Caspian Sea.

In this study, Survey Ctenophore (*Mnemiopsis leidyi*) feeding in the Iranian coasts of the Caspian Sea.

Materials and methods

Mnemiopsis leidyi were collected from 5 to 20 m depth in the Iranian coasts of the Caspian Sea, During August 2002 – October 03. *Mnemiopsis*

leidyi was sampling using a 500 micron mesh sized METU net (diameter 50 cm with large bucket suitable for *Mnemiopsis*). Samples were obtained via horizontal towing from the bottom to the surface. At the end of each tow, the net was washed from the exterior ,and the cod end was passed into a container immediately to enumerate ctenophores by naked eye.
The ctenophores were sorted into length groups of 0-5 mm, 6-10 mm, 11-15 mm and so on, for size measurements (total length including lobes) in glass petri dishes using a ruler. A total of 800 individuals were measured and grouped in this way. Animals were fixed with 4 % formaldehyde and transported to the laboratory. Stomach contain of *M.leidyi* was distinguished with microscope and distinguish key of Caspian Sea Invertebrate.
Then, we have taken photo from zooplankton of *M.leidyi* Stomach .

Results

There was stomach contain of *Mnemiopsis* in the Iranian coasts of the Caspian Sea, Zooplankton 84 % and Phytoplankton 16 % (Fig. 1).

The gut contents of *Mnemiopsis* include a wide variety of prey (zooplankton) taxa, Acartia and Acartia Nauplii (Copepoda) 34.17 %, Copepoda egg 7.05 %, Balanus (Cirripedia) 18.6 % , Ciliata 4.2 %, Bivalvia (Mollusca) 2.85 % , Podon (Cladocera) 0.75 % , Branchionus (Rotatoria) 5.99 % , Rotatoria egg 1.35 % , Fish egg 3.15 % and Digest zoo 5.7 % (Fig 2).
The most abundance of phytoplankton in guts were Thalasionema (chrysophyta phylum) 66.97 % , Oscillatoria (Cyanophyta phylum) 2.75 % , Phacus (Euglenaphyta phylum) 5.5 % Prorocentrum (Pyrrophta phylum) 3.7 % and Senedesmus (Chlorophyta phylum) 21.7 % (Fig. 3).

The maximum and minimum prey consumed by *Mnemiopsis* was showed in 0-5 mm and 40-45 mm length group with average 5.82 ind and 1.36 ind (Fig. 4).

The maximum consumed zooplankton was observed with mean 4.42 ind (Rotatoria egg) and the lowest prey consumed was showed (Bivalvia) with average 1 ind in August. In September, the maximum *M.leidyi* feeding was from Bivalvia with average 3.4 ind. The most consumed zooplankton (Rotatoria egg) was observed with average 6 ind in October.The maximum prey consumed was showed fish egg with mean 4.5 ind (Fig. 5).

Discussion

This study showed *M.leidyi* can not prey selection, *Mnemioipsis leidyi* ingests zooplankton and phytoplankton and any organism (Fig. 1).
Prey selection does not appear to be strong in *Mnemiopsis*, since many authors have concluded ther is no selection (Purecell et al.,2001). *M.leidyi* is a macrophage capable of eating rather large prey (at least 1 cm long), It consumes own young ,the eggs and larvae of anchovy and apparently

juveneile medusas (Malyshev et al., 1993). The gut contents of *Mnemiopsis* in U.S. estuaries include a wide variety of prey taxa, with 75-93 5 of the items being copepods, copepod nauplii, Bivalvia veligers. Other common prey items include polychaete larvae, cladocerans crab and shrimp larvae, In Chesapeake Bay including copepod nauplii (59 %), copepods (38 %), bivalve veligers (1.6 %), Cladocerans (0.8 %) (Purcell, unpublished).

In the Black Sea overall, the gastrovascular cavities of *Mnemiopsis leidyi* individuals contained the following food components, in descending oral of frequency: Copepoda (50 %), Mollusca (40 %), fish eggs and larvae (1 %), Cladocera (1 %) and other (8%) (Mutlu 1999). These study confirm with results of *M.leidyi* feeding in the Caspian Sea (Fig .2,3)

Contain Stomach of *M.leidyi* in the Caspian Sea was observed zooplankton (84 %),Thoes were included Copepoda, Cladocera, Mollucsa larvae, Cirripedia, Fish eggs …(Fig. 1,5).

Density, biomass and diversity of zooplankton and meroplankton decreased month after month with expanding *M.leidyi* in all region, Comparing July and August, density decreased some 17 times in the Northern Caspian (this decrease mainly resulted from a drop in the larvae of benthic organisms, which comprised of 74 % of the total in July). In the Middle Caspian, Zooplankton fell to half and in the South to one third of its previous value (Shiganova 2002).

Surveys showed in the Iranian coasts of the Caspian Sea, Copepoda, Cladocera and Rotatoria sharp decreased during August to October and *M.leidyi* biomass was increased during the same (Bagheri and Kideys, 2002).

After the invasion of *Mnemiopsis* a precipitous decline occurred in the numbers of mesozooplankton in the Black Sea, Since the summer of 1989, the abundance of Paracalanus parvus, Oithona similes, Acartia clausi, all species of cladoceras and polycheate and gastropod larvae have decreased, particularly in the upper layer and coastal area Species such as O.nana and representatives of the family pontellidae completely disappeared from samples (Purcell et al., 2001)

The study stomach contain of Kilka (Clupeonella) was showed in the Iranian coasts of the Caspian Sea, Kilka consumed from Copepoda and Cirripedia, Perhaps feed compact M.leidyi with kilka was decreased kilka stock (Bagheri et al.,unpablished)..

References

- Bagheri,K.S; Kideys, E.A., 2002. Oceanography of the Eastern Mediterranean and Black Sea.METU Cultural and Convetion Center Ankara/TURKEY.pp.342

- Dumont, H.J., 1995. Ecocide in the Caspian. Nathre, 377, pp.673-674GESAMP (IMO/FAO/UNESCO-IOC/WMO/WHO/IbAEA/UN/UNEP JoinGroup of Experts on the Scientific Aspects of Marine Enviromental Protection (1997) Opportunistic settlers and the problem of the ctenophore Mnemiopsis leidyi invation in the Black Sea.Rep Stud GESAMP.Vol. 58, pp. 1-84

- Harbison, G.R; 2000. Biology of Mnemiopsis in the coast of American.ICES C.M.pp.74-84

- Ivanov, P.I; Kamakima, A.M; Ushivtzev, V.B; Shiganova, T.A; Zhukova, O; Aladin.N; Wilson, S.I; Harbison, G.R; Domunt, H.J., 2000. Invasion of Caspian Sea by the come jelly fish Mnemiopsis leidyi (Ctenophore), Biologcal Invasion, 2, pp. 255-258

- Kideys,E.A., 1994. Recent dramatic changes in the Black Sea ecosystem: the reason for the sharp decline in Turkish anchovy fisheries. J Mar Syst 5:171-181

- Kideys,E.A; Romanova, Z., 2001. Distribution of gelatinous macrozooplankton in the southern Black Sea during 1996-1999. Marin Biology,Vol.139, pp.535-75

- Kideys AE, Shahram G, Davood G, Roohi A, Bagheri S (2001a) Stratrgy for combating Mnemiopsis in the Caspian Sea waters of Iran. Final report,July 2001, prepared for the Caspian Enviornment Program, Baku, Azerbaijan

- Kideys, AE,Jafarov FM, Kuliyev Z, Zarbalieva T (2001 b) Mponitoring Mnemiopsis in the Caspian waters of Azerbaijan. Final report, August 2001, prepared for the Caspian Enviornment Program, Baku, Azerbaijan

- Malyshev, V.I; Arkhipov, A.G., 1993. The ctenophore Mnemiopsis leidyi in westren Black Sea. Puplished in Hidrobiologicheskiy Zhurnal,Vol. 28,pp.34-39.

- Mutlu, E., 1999. Distribution and abundance of ctenophores and their zooplankton food in the Black Sea.II. Mnemiopsis leidyi. Marine Biology.Vol. 135, pp.603-613

- Pereladov, M.V., 1988. Some observation for biota of Sudak Bay of the Black Sea. The third All-Russia Conference on Marine Biology. Kive, Naukova Dumka, pp. 237-238

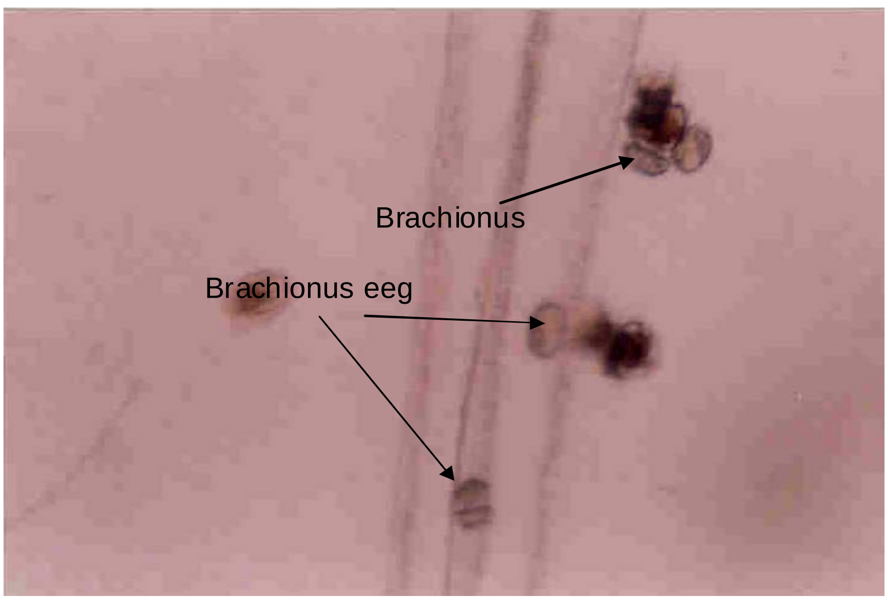
- Purcell,J.E; Shiganova, A.T.; Decker, M.B.; Houde, E.D.,2001. The ctenophore Mnemiopsis in native and exotic habitats: U.S.estuaries versus the Black Sea basin. Hydrobiologia. Vol. 451, pp.145-147.

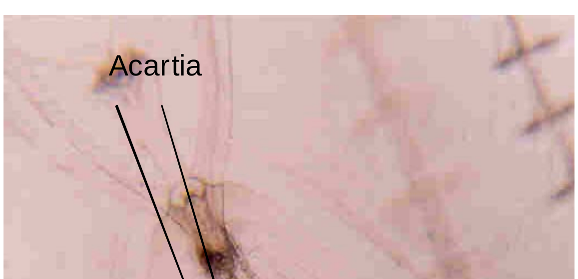
- Sergeeva, N.G; Zaika V.E; Mikhailova, T.V.,1990. Nutrition of ctenophore Mnemiopsis maccradyi (Ctenophore, Lobata) in the Black Sea (in Russian). Zool J Ecologia Morya. Vol. 35, pp.18-22.

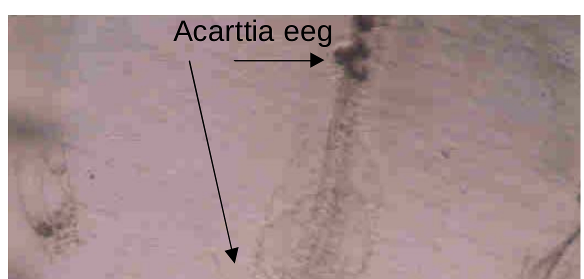
- Shiganova, T., 2002. Enviormental Impact Assessment including Risk Assessment regarding a proposed Introduction of Beroe ovata to the Caspian Sea.Institute of Oceanology RAS,Russia, pp. 1-45

- Vinogradov, M.E; Shushkina, E.A; Musaeva, E.I; Sorokin, P.Y., 1989. A new acclimated species in the Black Sea: the ctenophore Mnemiopsis leidyi (Ctenophora: Lobata). Oceanology Vol.pp. 220-224

Nauplii Acartia







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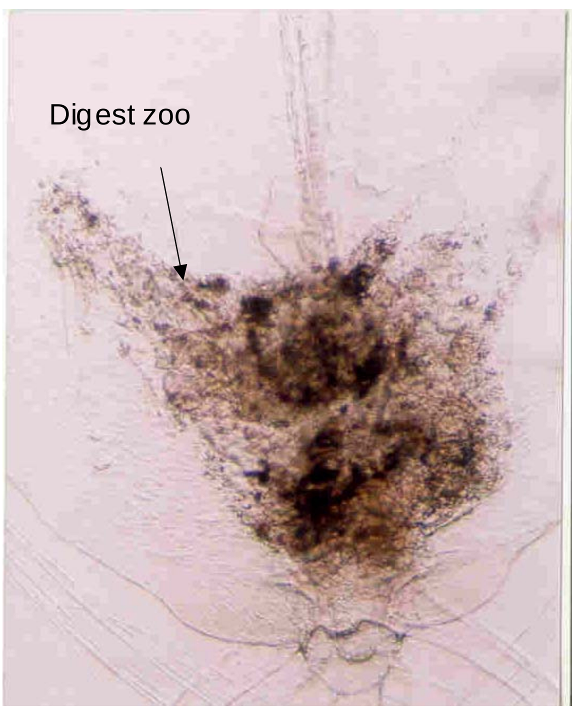

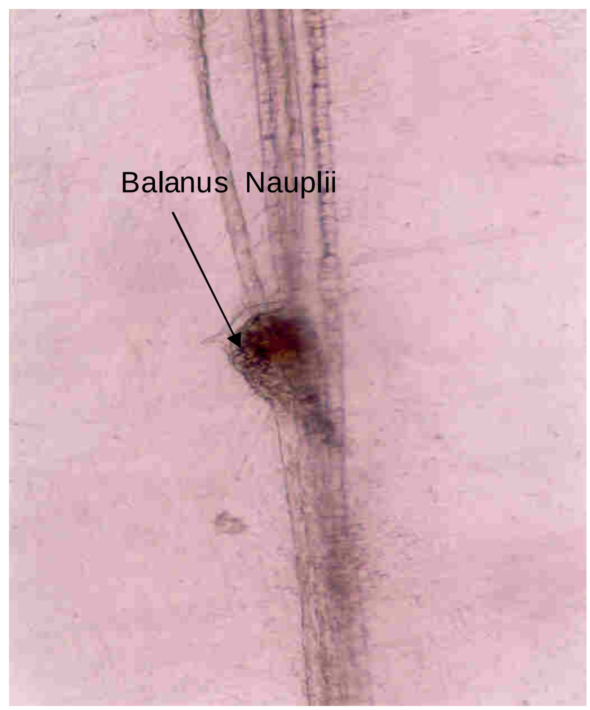

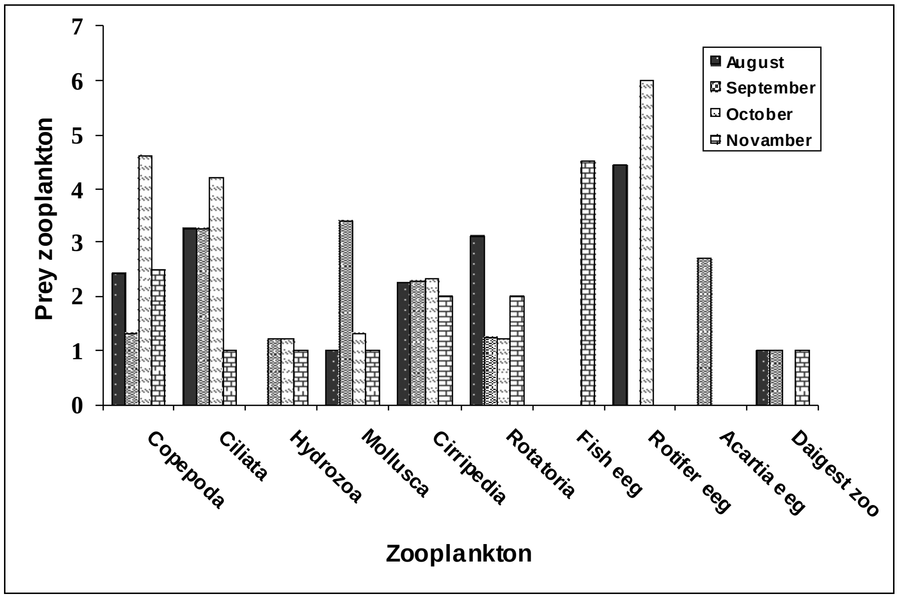

Fig 5 : Average prey zooplanktonin by M. leidyi in diffrened month (2002-03)

(Reproduction experiments of *Beroe ovata*)

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Abstract

The species of *Beroe ovata* has some advantages that can be best candidate for control of Mnemiopsis population size in the Caspian Sea.

This study was done in many sites and was performed at different salinity treatments .

Survival and growing of *Beroe ovata* eggs were studied .Results showed that *Beroe* specimens can produce in Caspian water and the *Beroe* larvae can grow in the Caspian Sea water but the values is low and mortality is high.

Fecundity individual of *Beroe* in the Black Sea water was higher than other experimental that produced a total of 17427 ova .

Results showed a lot of eggs in Caspian sea water were destroyed and could not developed but the results near to Black sea with fresh specimens was better and more values of ova were hatched to larvae.

The results survey of mesocosm tanks showed most of ova and larvae have been obtained from the tanks where individuals *Beroe* were with Mnemiopsis.

The difference between reproduction in Black sea and Caspian sea treatment s could be connected with effect of lower salinity and stress conditions acclimation for the Caspian Sea salinity.

Introduction

Outbreak of *Mnemiopsis leidyi* in the Black and Azov Seas greatly affected their ecosystems and fish stocks. But in 1999 bloom of new invader another ctenophore *Beroe ovata* occurred in the Black Sea. *Beroe ovata* feed exclusively zooplanktivorous ctenophores first of all *Mnemiopsis leidyi*. Resulted from this invasion the Black Sea ecosystem began rapidly recover.

In 1999 *Mnemiopsis leidyi* was introduced to the Caspian Sea from the Black Sea with ballast waters from the ships (Esmaeili et al., Ivanov et al., 2000; Shiganova et al., 2001).

Appearance of predatory comb jelly *B. ovata* consuming *M. leidyi* sharply decreased its population (11–20 fold within 10 days) and considerably enhanced the process of ecosystem recovering on the main components of the Black Sea pelagic ecosystem-zooplankton and fish as well as their eggs and larvae (Shiganova, 2003)

The species of *Beroe ovata* has two outstanding advantages that can be best candidate for control of Mnemiopsis population size in the Caspian Sea firstly, it is highly specific in its feeding, so that even its larval stage feeds on *M.leidyi* larvae. Secondly, its reproductive rate and fecundity are almost as great as that of *M.leidyi*, so that its population can grow at similar rates to its prey (Shiganova et al., 2003).

At 2001-2002 propagation experiments with the aim of mass culture of *Beroe ovata* in Caspian Sea water prepared in the laboratory that the results were unsuccessful since the both the spawning and hatching rates were very low and, none of the larvae developed into adults (Kideys et al., 2001, 2002).

In order to understand the feasibility of *Beroe* introduction, as an effective predator on *Mnemiopsis*, into the Caspian Sea, experiments on survival and reproduction of *Beroe* in Caspian Sea water (12.6 ppt salinity) were performed.

This experiments focus on reproduction experiments to identify probability *Beroe ovata* to reproduce in the Caspian Sea water, to hatch larvae and to develop .

Material and Methods

Experimental was performed three treatment of salinity, the Black sea water 18‰, Mixed water 15‰ and Caspian water 12‰ . *Beroe ovata*, were placed individually in jars, *Beroe* were transferred into the Caspian water after acclimation.

To experiments on growth and survival of *Beroe* larva, the collected eggs from jars were placed in the same treatments and surveyed after 2 and 4 days.

Individuals of *Beroe ovata* were brought from Turkey and were sent to Guilan province for reproduction studies and another part were sent to Mazandaran province for both of reproduction and Mesocosem studies although about half of specimens were died 2 days later arrive to Iran.

Some of this specimens were in the Black Sea water with salinity 18‰ and another part of specimens had been acclimated in salinity 15‰ of the Caspian Sea water in Sinop. After transportation and during acclimation aquariums with *Beroe* specimens were examined for ovae and larvae availability.

The Jars were examined every day for ova and larvae with seawater filtered thought 55 µm mesh. The ova and larvae were collected for hatching and developing studying where Mnemiopsis tissue small pieces were used as food.

Most of individuals were used to mesocosm experiment that each tank was checked for ovae and larvae for five days. Also we continued reproduction study with 3 *Beroe* from Mazandaran and 6 *Beroe* were brought from Caspian Sea bony fishes research center in 3 l aquariums. Some of ovae and larvae were put in Vase incubators.

For control we had 1 *Beroe,* length 30 mm in the Black Sea water that was alive during of study In Anzli and Mazandaran.

Results:

Reproduction studies with freshly specimens in Sinop laboratory have been showed *Beroe ovata* produce ova and larvae in Caspian sea water with 12‰ salinity however it was much less than Black sea 18 ‰ water and the Mixed water 15 ‰ .

Ova can be hatched and developed in Caspian sea water with a little values but it was higher in Black sea water. Survival rate was the highest in Black sea water than another treatments after 4 days with a little value.

At two first day when the specimens arrive to Caspian sea bony fishes research center we obtained 324 ova and 2 larvae from 15 specimens of 6 container. After that during 7 days when specimens were alive we obtained 335 ova and 40 larvae that 47 % of ova were destroyed not achieving development and 40 % of larvae were died.

The specimen with 36 mm length feed for some days and produced total of 235 ova and 8 larvae during of study .

In Sari Ecologic academy we found 100 ova 1 stage during acclimation of specimens 16 Beroe in aquarium 9 liters also was found 2 ova and 6 larvae from Caspian Sea water aquarium that had been brought from Anzali.

Reproduction rate in first experiments was very low at temperature 22^0 C in Caspian sea water, we obtained from 2-9 ovae from largest individuals and from 1 to 8 ovae. The individuals less than 48 mm in these experiments did not spawn.

we obtained 20 to 61 ova (mean 37) and 5 to10 larvae from 3 specimens of Anzali and Mazandaran.

In four three litters aquariums we held Beroe without checking reproduction rate for four days and we obtained larvae among them. We found one larvae with 5 mm length, other larvae were at different stages of development but most of them were 1.2–2, 1 mm. Results of ovae developing in Caspian and Black sea water showed the hatching and larvae survival was less much than Sinop, all of ovae were destroyed and 2.7 % larvae were alive in Caspian water. We did not obtained developed larvae from the vase incubator.

Fecundity of *Beroe* in the Black Sea water (control specimen) was higher where this specimen fed very well especially in Anzali and produced a total of 17427 ova during study .

The results survey of mesocosm tanks was shown in table 3. Totally we had 863 ova and 188 larvae from tanks. Most of ova and larvae (79.14 % ova and 68% larvae) have been obtained from the tanks where individuals Beroe were with Mnemiopsis, we had reproduction in these tanks every day (Table 1). We had much lower numbers of ova and larvae, where Beroe was without Mnemiopsis, only with zooplankton (20.86% ova and 32% larvae).

We had ova and larvae only once or two times in each tank (Table 1). Approximately more of observed larvae had not good situation from tanks surveys (Fig. 10).

Table 1 Reproduction of *Beroe ovata* in tanks.

NO Tank	Contents	Initial numbers of Beroe	Initial size mm	Numbers of Beroe ovae and larvae				
				22.09.03	24.09.03	25.09.03	26.09.03	27.09.03

		& (number at beging of reproduction checks)		Ova	Larva	Ova	larva	Ova	larva	Ova	larva	ova	larva
1	Zooplankton +Beroe	7 (5)	35.6±8			90	0	0	0	0	0	0	0
2	Zooplankton + Mnemiopsis+ Beroe	7 (5)	37.7±8.5	263	38	120	0	0	90	60	0	60	0
5	Zooplankton +Beroe	7 (1)	31.3± 9.5			30	30	0	0	0	0	_	_
6	Zooplankton +Beroe	7 (3)	34.2±6			30	0	30		0	0	_	_
9	Zooplankton +Beroe	7 (5)	29.5±13			0	0	0	30	0	0	0	0
10	Zooplankton + Mnemiopsis+ Beroe	7 (4)	39.1±7			180	0	0	0	0	0	0	0

Discussion

These results are showed that Beroe can reproduce and larvae can hatch in the Caspian Sea water. These experiments for reproduction and development ovae and larvae were more successful than in previous two years in Iran by Kideys et al. (2001, 2002). However some larvae did not develop and ovae did not hatch in some of our experiments.

Studies on reproduction of Beroe ovata in Iran last years showed only a few of eggs (<20 eggs) were developed and hatched in Caspian sea water where they did not grow, the results from Marmara Seawater was better and were produced more eggs however more than 90% of them were not fertilized and did not develop that 138 eggs of *B. ovata* from which 7 larvae had been hatched (Kideys et al., 2002).

In these experiments for reproduction and development we obtained as much as numbers of ovae and larvae than experiments conducted in the Caspian Sea water near the Black Sea from fresh collected and immediately acclimated for the Caspian salinity individuals in the Southern branch of Sirshov Institute of Oceanology RAS in Gelendzhik (Shiganova unpublished) however the number of ovae and larvae were less much in Caspian sea bony fishes research center and Caspian sea Ecology Academy.

Egg number depends greatly on temperature and feeding conditions, morphological data shows that upper limit of eggs number in favorable condition is about 2000–3000 per day for large-sized animal (Arashkevich, unpublished) .

In this study hatching success and growth rate of larva had very low values even for ovae and larvae in Black sea where they did not developed .

In generally we can conclude that *Beroe* larvae can grow in the Caspian Sea water though their grow rate is slow and mortality is high.

This difference could be connected with effect of lower salinity and stress conditions acclimation for the Caspian Sea salinity. Examination of Mnemiopsis fecundity showed that in the Caspian Sea its fecundity more than in two times lower than in the Black Sea water (Shiganova unpublished). Finenko (unpublished) supposed to transfer ctenophores to lower salinity at the stages of eggs or early larvae, they die and suggested a long preliminary acclimatization of juvenile animals that can be affect upon the salinity resistance of both adult and embryos and larvae produced by them and as a result the survival range can be shifted towards lower salinity.

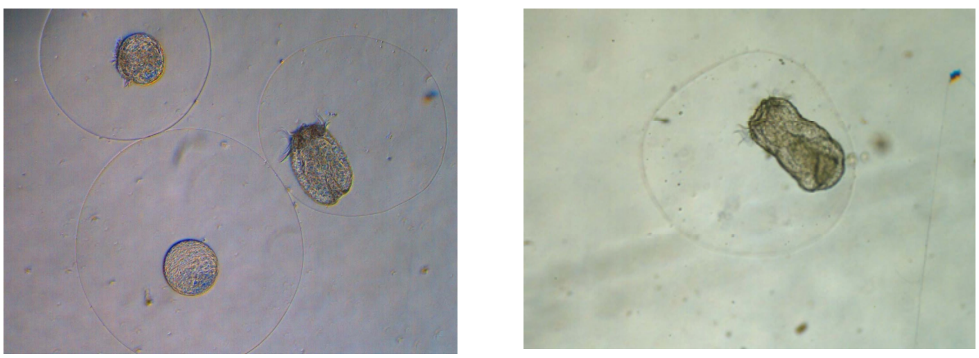

Fig .1A Fig. 1B

Fig 1) Ova and larva of Beroe from Black sea water (A) and larvae (B)in Caspian sea water from Sinop experiments

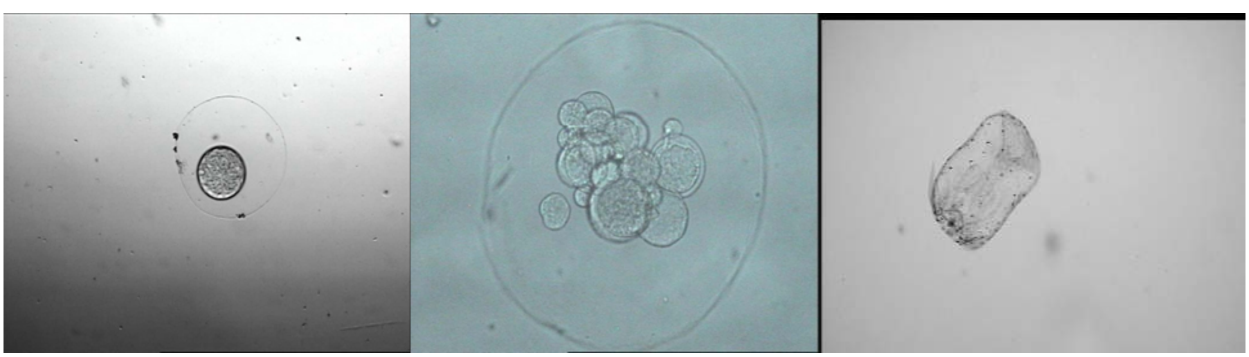

Fig. 6A Fig 6B Fig. 6C (photo by M. Rostamian)

Fig.. 6) New ovae (a) and ova developing (b) and larvae (c) of Beroe ovata from Caspian sea water at Iran experiments

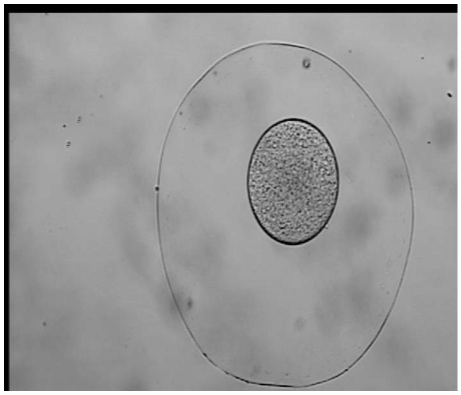

Fig. 8A

Fig. 8B

Fig. 8) New Ovae (a) and not developed ovae after 3 days (b) of Beroe ovata from Black sea

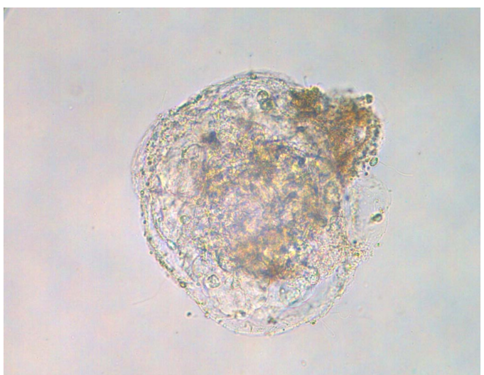

Fig 9) destroyed ovae in Caspian water from sinop experiments

Fig. 10) The died egg from mesocosem tank

Acknowledgment

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Literature.

- Arashkevich E. G. L. L. Anokhina, S. V. Vostokov, A. V. Dritz, T. A. Lukasheva, N. E. Luppova, E. I. Musaeva, A. N. Tolomeev ,2001. Reproduction strategy of Beroe ovata (Ctenophora, Antentaculata, Beroida)- A new invader in the Black sea. Oceanology ; Vol. 41 , No.1 , pp. 111-115 .
- Arashkevich **,** unpublished *Beroe ovata*. Egg production, hatching success and growth rate of larva in the Caspian and Black Sea waters (Results of experiments carried out in Sinop, September 2003).
- Esmaeili, S. A., S. Khodabandeh, B. Abtahi, J. Sifabadi & H. Arshad, 2000. First Report on occurrence of a comb jelly in the Caspian Sea. Journal of Science and Technology of the Environment, 3: 63-69.
- Finenko G. A., unpublished The one of the main purposes from the activity in 2002 was studying survival and tolerance of *Beroe ovata* at lower salinity.
- Harbison G.R. Madin L.P. & Swanberg N.R. On the natural history and distribution of oceanic ctenophores. Deep-Sea Research.1978. Vol.25 p.233-256 .
- Greve, W., 1970. Cultivation experiments on North Sea ctenophores. Helgolander wiss. Meeresunters. 20: 304-317.
- Ivanov, P. I., A. M. Kamakim, V. B. Ushivtzev, T. Shiganova, O. Zhukova, N. Aladin, S. I. Wilson, G. R. Harbison & H. J. Dumont, 2000. Invasion of Caspian Sea by the jellyfish *M.leidyi leidyi* (Ctenophora). Biological Invasions 2: 255-258.
- Kideys A.**,** G A. Finenko, B E. Anninsky, T. A. Shiganova, A. Roohi, M. R. Tabari, M. Youseffyan, M. T. Rostamian, H. Rostami, H. Negarestan Laboratory Studies on Physiological Characteristics of *Beroe ovata* in the Caspian Sea Water.
- Kideys A. E. , G. A. Finenko , A. Arashkevitch , T. A. Shiganova , A. Roohi, M. Roushantabari, M. Russtemian, A. Gevanshir , A. R. Mirzajani , H. Negarestan, F. Parafkandeh , 2002 . Propagation experimentas with Beroe ovata in the Caspian sea water *Iranian Fisheries Research Organization, Tehran, Iran*
- Shiganova, T. A., A. M. Kamakin, O. P. Zhukova, V. B. Ushivtsev, A. B. Dulimov & E. I. Musaeva, 2001b. The invader into the Caspian Sea Ctenophore Mnemiopsis and its initial effect on the pelagic ecosystem. Oceanology T.41. N 4: 542–549.
- Shiganova T. A. , E. I. Musaeva, Yu. V. Bulgakova, Z. A. Minoyan, M. L. Martynyuk, 3. Invaders Ctenophores Mnemiopsis leidyi (A. Agassiz) and *Beroe ovata* Mayer 1912, and Their Influence on the Pelagic Ecosystem of Northeastern Black Sea . Biology Bulletin, Vol. 30, No.2, pp. 180-190.

Распределение, численность, биомасса гребневика Mnemiopsis leidyi в прибрежной зоне Азербайджанского сектора Каспийского моря в 2001 – 2003 гг.

З.М.Кулиев, д.б.н., директор АзерНИРХ

Со времени инвазии гребневика Mnemiopsis leidyi в Каспийское море прошло более 5 лет, если считать началом обнаружения его 1999 год. Весть о появлении Mnemiopsis leidyi в Каспии ученые и специалисты встретили с большой тревогой и, что было не безоснования и совершенно закономерно, так как была хорошо известна информация о широкомасштабном и многплановом влиянии гребневика на фнкционирование всей морской экосистемы Черного и Азовского морей после вселения в них мнемиопсиса.. В этой связи всестороннее исследование нового, весьма опасного для биоразнообразия Каспия, вселенца мнемиопсиса, в настоящее время является одним из важнейших его проблем. Считаем совершенно верным мнение С.П.Воловика (2000г.) о том, что без крупномасштабных и эффективных мер по контролю за развитием популяции мнемиопсиса, которые должны быть срочно разработаны и внедрены, Каспий, как уникальная экологическая система и важный рыбопромысловый регион, будет практически потерян.

Результаты многолетних исследований в Черном и Азовском морях, проводимых после вселения в них Mnemiopsis leidyi, свидетельствуют об обширном и многоплановом влиянии гребневика на функционирование всей экосистемы и в первую очередь на экосистему пелагиали и бентоса (Виноградов, Шушкина, Николаева, 1990; Воловик, Луц 1991; Кучерук, Востоков 2002, Востоков, Ушивцев, и др. 2002, Шиганова 2001). Все проведенные исследования показали, что после вселения Mnemiopsis leidyi биомасса кормового зоопланктона, интенсивно выедаемого мнемиопсисом, в Черном и Азовском морях резко сократилась (в Черном море в 20-30 раз), а некоторые группы планктонных организмов (например Chaetognata) почти полностью исчезли.

Все это незамедлительно сказалось на популяциях планктоноядных рыб – хамсы, тюльки и ставриды, по отношению к которым Mnemiopsis leidyi выступает не только как пищевой конкурент, но и как прямой потребитель их икры и молоди (Цихон-Луконина, Резниченко, Лукашева, 1993). В результате этих и других исследований последствий вселения Mnemiopsis leidyi было зафиксировано катастрофическое падение уловов планктоноядных рыб Азово-Черноморского бассейна (Кидеш, 1994; Виноградова, Шушкина, и др. 1995).

Учитывая, что рыбная отрасль Каспийского моря уже , после вселения гребневика, понесла тяжелые потери, связанные с резким снижением уловов килек, возникла острая необходимость силами специалистов всех прикаспийских стран разработать единый, научно-обоснованный план действий по спасению экосистемы Каспийского моря от пагубного воздействия вселившегося гребневика-мнемиопсиса.

В 2000 г., различными авторами был отмечен значительный рост биомассы мнемиопсиса, особенно в Южном и Среднем Каспии, а также уменьшение биомассы мезопланктона (Шиганова и др. 2000; Сокольский, Шиганова, Зыков, 2001; S.Rezvani,H.Rostami, A.Javanshir, 2002). Стало очевидным, что в первую очередь, биологическая вспышка нового вселенца, может коренным образом изменить видовой состав и биомассу мезопланктона, а затем, последует уменьшение численности наиболее многочисленного планктоноядного промыслового вида – кильки и всех других видов пелагических рыб. Серьезность ситуации было подтверждено на проведенных в 2001-2002 гг. в Баку, по инициативе и поддержке Каспийской Экологической Программы (в апреле, декабре 2001 г. и январе, апреле 2002 г.) Международных семинарах по проблеме мнемиопсиса, с участием ведущих специалистов по данной проблеме из разных стран, в том числе из Российской Федерации, Ирана и Турции. Учитывая большой процент эндемизма среди животных Каспийского моря, реальную угрозу рыбным запасам, и ее биологическому разнообразию, в связи с инвазией гребневика в Каспий, Азербайджанским Научно-исследовательским институтом рыбного хозяйства в апреле 2001г., была разработана научно-исследовательская программа по изучению распределения, численности и биомассы гребневика на западном шельфе Среднего и Южного Каспия

Программой Института предусматривалось, проведение исследований по следующим разрезам: Яламинское взморье (пос.Ялама 6); Сиазань (пос. Зарат); Килязи; Сангачалы (пос.Приморск); Нефтчала (Усть Кура); Ленкорань (порт Лиман). В последующие годы (2002-2003 гг.) исследования проводились по сетке станций, включающих в себя 5 станций в Среднем Каспии и 6 в Южном (Рис.1). Кроме того, проводились исследования и на основных местах добычи кильки в Южном Каспии.

Исследования и сбор проб проводился с помощью судна Министерства Экологии и Природных Ресурсов Азербайджанской Республики – «Алиф Гаджиев» и с килечных судов в районах лова.

Рис.1. Карта-схема Каспийского моря с указанием станций сбора материалов распределению M. leidyi по

При отборе проб на мнемиопсис определялись и абиотические факторы среды: температура, соленость, прозрачность и содержание кислорода в воде на всех разрезах и глубинах.

Для сбора гребневика мнемиопсиса использовали ихтиопланктонную сеть Juday-500 с диаметром входного отверстия 0,5м. Взятие проб осуществлялось вертикальными обловами столба воды 0 – 50 метров с борта судна, на малых глубинах – от дна до поверхности. Пробы обрабатывались на месте непосредственно после их взятия.

За период исследований 2001-2003 гг. всего было собрано и обработано 520 проб мнемиопсиса и свыше 110 тыс. экземпляров гребневика (2001-37620 экзю, 2002-60633 экз., 2003-16117 экз.).

Результаты исследований

В 2001 году почти на всех изученных разрезах был обнаружен гребневик мнемиопсис разных размерных и возрастных групп. Наибольшая биомасса мнемиопсиса была зарегистрирована в 2001 г. летом в Сиазане на глубине 5 м – 151,9 г/м^3 и на станции Лиман (137,0 г/м^3), а осенью в районе Килязи (141,3 г/м^3) и Лиман (121,4 г/м^3). Летом 2002 г. наибольшая биомасса мнемиопсиса зафиксирована на станции килязи (271,0 г/м^3) и Лиман (137,9 г/м^3), осенью максимальная биомасса отмечено на станции лиман (112,0 г/м^3). Зимой отмечалось резкое снижение численности и биомассы M. Leidyi на всех исследованных станциях, особенно при зимних съемках 2003 г., когда биомасса M. Leidyi не превышала 0,3 г/м^3 (Таблица 1).

Таблица 1.

Численность и биомасса гребневика Mnemiopsis leidyi по сезонам и годам

В Азербайджанской акватории Каспийского моря

Года / Разрезы	2001				2002						2003					
	Лето		Осень		Весна*		Лето		Осень*		Зима		Лето*		Осень*	
	экз.	г/м^3	экз.	г/м^3	экз.	г/м^3	экз.	г/м^3	экз.	г/м^3	экз.	г/м^3	экз.	г/м^3	экз.	г/м^3
Лиман	1512	137	1144	121,4	437	58,5	1502	137,9	1335	112,0	45	0,3	196	2,64	70	1,2
Устье куры (Нефтчала)	368	96,2	97	31,35	59	1,1	188	48,7	546	32,4	13	0,1	249	1,63	101	1,1
Сангачал (Приморск)	488	90,4	103	61,68	247	2,1	-	-	398	3,4	41	0,06	-	-	34	1,2
Килязи (Сумгаит)	780	109,2	1699	141,3	685	54,6	1963	271,0	-	-	-	-	257	1,46	15	0,4
Сиазань	139	151,9	368	62,86	113	3,2	524	151,9	-	-	7	0,05	283	2,08	14	0,3
Ялама	345	64,9	15	40,94	67	1,2	683	165,4	251	63,2	1	0,02	280	1,44	13	0,4

* - данные приведены на основании полученных материалов отбора M. leidyi при килечном лове.

Следует отметить, что на различных разрезах соотношение размерного состава сильно варьировало. Самые крупные экземпляры гребневиков, были представлены единичными особями размерной группы 61-65 мм, главным образом в южной части Среднего Каспия и в Южном Каспии. Однако мелкие экземпляры (молодые и ювенальные стадии) практически везде составляли более 70 % от общей численности гребневика.

Такая тенденция сохранялась все годы и сезоны исследования, мелкие фракции M. Leidyi в пробах колебались от 70,7 до 100 % (таб.2).

Таблица 2

Процентное сотношение младших возрастных групп гребневика Mnemiopsis leidyi по сезонам и годам в Азербайджанской акватории Каспийского моря в %

Года / Разрезы	2001		2002			2003	
	Лето	Осень	Зима – Весна*	Лето	Осень*	Зима-Весна*	Осень
Ленкорань (Лиман)	86,3	99,4	97,4	89,1	85,4	94,6	97,2-
Устье Куры (Нефтчала)	86,7	87,7	99,3	68,7	74,2	89,8	89,4
Сангачал	91,0	84,8	98,6	-	76,4	77,9	79,6
Килязи	76,2	71,1	99,7	97,5	-		
Сиазань	82,8	93,7	100,0	78,9	-	78,9	86,5
Ялама	70,7	100,0	100,0	98,8	84,6	100,0	84,3

* - данные приведены на основании полученных материалов отбора M. Leidyi при килечном лове.

Высокие показатели процента молодых экземпляров свидетельствует о неослабевающем процессе размножения мнемиопсиса на протяжении всего года.

Исследования, проведенные в 2001-2003 гг. показали, как было отмечено, повсеместное распространение мнемиопсиса как в Среднем, так и в Южном Каспии. Однако, на отдельных участках Каспия он был распределен неравномерно. На юге Каспия наибольшая концентрация отмечена на разрезе Кюр дили, где она составила 708 экз./м^3. В Среднем Каспии самая высокая численность мнемиопсиса наблюдалась в районе Килязи (Сумгаит), достигая 1153 экз./м^3. Прослеживалась определенная зависимость величины концентраций гребневика от глубин точек пробоотбора. Наибольшая численность была приурочена к мелководным участкам до 25 м (рис2).

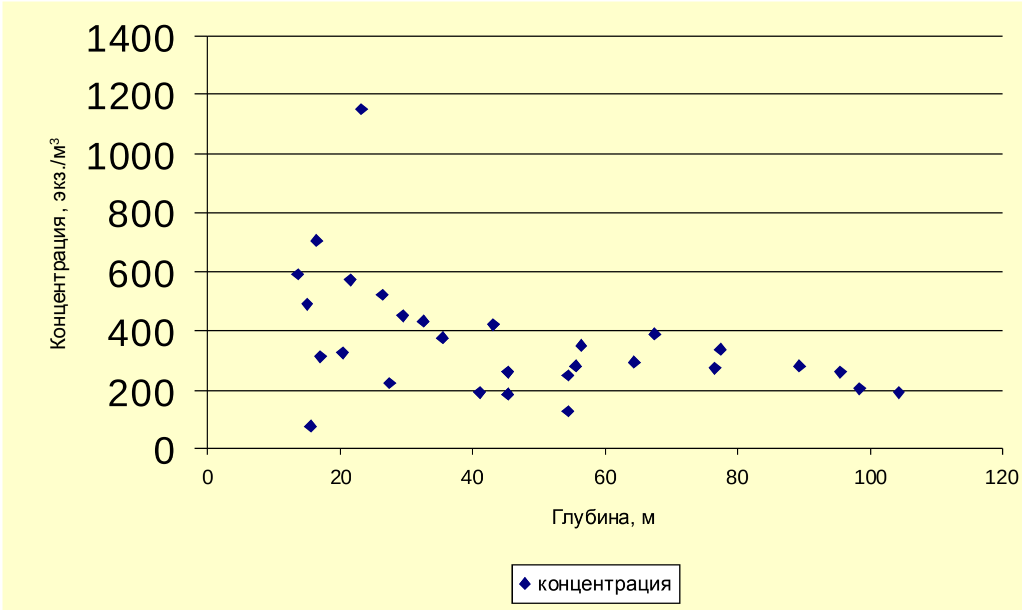


Рис. 2. Распределение M.leidyi по глубинам.

А. Сокольский, Т. Шиганова и др. (2002) указывает на интенсивное размножение M. leidyi в Южном и на западе Среднего Каспия, где концентрация личинок достигает 1000-2000 экз/м^3, тогда как на восточном шельфе Среднего Каспия и в Северном Каспии их максимальная численность составляла не более 100-350 экз/м^3. По нашим данным размножение гребневика протекает в Южном Каспии круглогодично, так как мы обнаруживали мелкие стадии (ювенальные) в декабре, январе и феврале, хотя их численность значительно сокращалось. В этот период взрослые особи практически отсутствовали.

Таблица 3

Количественное (экз.) распределение гребневика M. leidyi
по разрезам и глубинам Азербайджанского прибрежья Каспийского моря

Глубина (м)	Разрезы										
	Шахагач	Ленкорань	Кюр дили	Зюйд-Остов Култук	Норд-Остов Култук	Мыс Бяндован	Мыс Пирсагат	Килязи	Сиязань	Траверз Кубы	Мухтадир
10	22/227	17/1600	18/-	12/2267	6/-	6/994	4/-	6/-	-/1369	3/-	-
25	16/-	16/2412	30/-	10/-	8/2681	7/2540	6/2679	4/5255	5/2722	6/-	5/1293
50	-/3473	37/-	-	12/2826	5/-	24/2478	9/2943	5/3771	6/1552	5/-	4/1303
75	21/-	-/3354	32/-	-	5/2705	11/-	7/-	-/3872	4/-	-	4/-
100	-/2590	-	21/1875	7/2032	-/2789	-	8/-	-	-	-	-
Концен-трация, экз/м^3	-/259	6,5/593	3,7/708	0,78/435	0,84/512	1,0/374	1,03/454	0,96/1153	0,96/524	1,1/220	1,25/323
Числен-ность, экз.	59/6290	70/7366	101/1875	41/7125	24/8175	48/6012	34/5622	15/8643	15/5643	14/-	13/2596

- числитель – зима 2003 г. январь-февраль,
- знаменатель –лето 2002 года июль-август.

Заключение

В результате проведенных исследований можно констатировать, что:

- гребневик мнемиопсис уже прошел стадию «биологической вспышки» и распространился по всему Азербайджанскому прибрежью Каспийского моря;
- наибольшая биомасса и численность гребневика отмечалась в период 2001-2002 гг. и достигала критической для экосистемы Каспия величины;

- во всех пробах, за все годы исследований, в количественном отношении преобладали ювенальные стадии гребневика (размерный ряд до 15 мм летом – 89,1%; до 10 мм осенью – 86,3%); максимальные размеры взрослых особей составили 61-65 мм.;

- преобладание мелких стадий гребневика на всем протяжении весны, лета и осени показывает на интенсивное его размножение почти в течение всего года.
- Наблюдаемая в настоящее время стабилизация численности и биомассы M.leidyi в 2003 г. не обеспечивает восстановления численности зоопланктона и, следует незамедлительно приступить к реализации мероприятий по уменьшению его численности (вселение гребневика Берое).

Литература

1. Виноградов М.Е., Шушкина Е.А., Николаева Г.Г. Массовое развитие гребневика мнемиопсиса, как проявление антропогенного воздействия на экосистему моря. //Практическая экология морских регионов. Черное море. Киев.: Наукова Думка. 1990. 94-102
2. Воловик С.П., Луц Г.Н. Вселение гребневика мнемиопсиса в Азовское море: предварительная оценка последствий // Рыбное хозяйство. – 1991. - №1. : 47-49
3. Воловик С.П. Гребневик *Mnemiopsis leidyi* (A.Agassiz) в Азовском и Черном морях.: Биология и последствия вселения. Ростов -на –Дону. 2000. 490.
4. Кучерук Н.Б., Востоков С.В. Влияние гребневика *Mnemiopsis leidyi на состояние донных сообществ Черного моря. // Океанология, 2001. 227.*
5. Цихон-Луканина Е.А., Резниченко О.Г., Лукашева Т.А. Выедание личинок рыб гребневиком *Mnemiopsis leidyi* в прибрежных водах Черного моря. //Океанология. 1991. Т.33. №6. 895-899.
6. Kideys A.E. 1994. - Recent dramatic changes in the Black Sea ecosistem: The reason for the sharp decrease in Turkish anchovy fisheries. Journal of Marine System 5: 171-181.

7. Тарасов А.Г. Вселение мнемиопсиса в Каспий.: Основные итоги 2001г.// Вестник Каспия. 2001. №5. 120-126.
8. Сокольский А.Ф., Шиганова Е.А., Зыков Л.А., - «Биологическое загрязнение Каспийского моря гребневиком Mnemiopsis и первые результаты его воздействия на пелагическую экосистему», сб. «Рыбохозяйственныеисследования на Каспии». Результаты НИР за 2000 г. Астрахань, 2001г., стр. 105 – 110.
9. Шиганова Т.А., Булгакова Ю.В., Воловик С.П. и др. Новый вселенец *Beroe ovata* и его воздействие на экосистему Азово-Черноморского бассейна в августе-сентябре 1999г. //Гребневик *Mnemiopsis leidyi(A. Agassiz)* в Азовском и Черном морях: Биология и последствия вселения. Ростов- на- Дону. 2000. 560.
10. Rezvani S., Rostami H., Javanshir A. A review of first nacional conference of Caspian Sea invader Ctenophore *Mnemiopsis leidyi. //* Caspian Sea Ecological Academy. 2002. I.I.R.

M.I. Karpyuk, D.N. Katunin, A.S. Abdusamadov, A.A Vorobyeva, L.V. Lartseva, A.F. Sokolski, A.M. Kamakin, V.V. Resnyanski, A.Abdulmedjidov

Results of research into *Mnemiopsis leidyi* impact on the Caspian Sea ecosystem and development of biotechnical principles of possible introduction of *Beroe ovata* for biological control of *Mnemiopsis* population

At the end of the 20th century (1999), during their work under water KaspNIRKh experts V.B. Ushivtsev and A.M. Kamakin detected and identified a new Caspian species referring to the type of comb jellyfish Ctenophora, the order Lobata - *Mnemiopsis leidyi.* From Dr. Nizami's oral presentation (IRI), this representative of the gelatinous was found when the zooplankton was sampled not far from Anzali City in 1995. Thus, the period of development of a mass population of the invader was some 4 years. It may be assumed that the Caspian Sea invasion by *Mnemiopsis leidyi* occurred via ballast waters of tankers which transported oil products from the Black Sea.

Some specific features of mnemiopsis biology (hermaphroditism, eurythermity, euryhalinity, high fecundity and rapid growth rate, a short cycle of reproduction, absence of predators and a broad spectrum of feeding) facilitated the rapid development of an abundant Caspian population. In 2000 it spread nearly through the entire area of the Caspian Sea and in August reached the western part of the Northern Caspian where its explosive development was recorded (up to isohaline of 4‰).

In the following years (2001-2003), the number and biomass of mnemiopsis increased (Table 1).

Table 1
Mnemiopsis leidyi abundance in August 1999-2003

Year	Northern Caspian	Middle Caspian	Southern Caspian
1999	0	0,01	0
2000	1.5-2	2-3	0.5-2
2001	5	12	53
2002	42	375	682
2003	509	920	602

depending on sea area (ind./m^3)

Russian experts' investigations (KaspNIRKh, RAS IO) undertaken in 2000-2002 enabled some conclusions concerning the ecology and biology of mnemiopsis in Caspian waters.

The development of mnemiopsis population depends on seasonal changes in water temperature. In winter (even in 2000-2002 when winters were very warm) the majority of the population inhabits the southern part of the sea and occurs in the boundary area of the Middle Caspian (the Apsheron Ridge). The adult part of the population mainly dies during that period.

In spring (May) the mass development of mnemiopsis larvae and juveniles occurs in the Southern Caspian and a part of the population spreads to the south of the Middle Caspian. The mass development of the invader reaches its maximum in August-September when it spreads into some areas of the western part of the Northern Caspian.

A drop of water temperature in November-December below 4^0 (similar to the Sea of Azov) may cause the mass elimination of mnemiopsis since it does not occur in the Northern Caspian in April.

Thus, *Mnemiopsis leidyi* inhabits the Southern Caspian through out the year, it lives in the Middle Caspian for 8 months (May-November) and in the Northern Caspian (western part) for 4-5 months (July-Novermber).That means that the greatest impact throughout the year is caused by mnemiopsis to the ecosystem of pelagic and benthic communities in the southern and middle parts of the sea.

In the years with extreme heat storage of water, the period of mnemiopsis population development and impact on the sea ecosystem may be extended (warm winters) or reduced (cold winters).

Mnemiopsis form largest concentrations in the zones with higher productivity and heat storage, stable dynamic processes. Within these areas there are the western coast of the southern part of the sea which hydrological regime is under the influence of advection of the Volga and Kura River runoff; the area of cyclonic motion south-east of the Apsheron Peninsular; the central part with southern Caspian cyclone. Such zones along the Iranian coast are as follows: the area with anti-cyclonic processes not far from Sefidrud Cape and in the south-eastern part. In the Middle Caspian, that is the western coast where the continuous input of transformed highly productive northern Caspian waters is recorded, the north-western part which is the zone of convergence of the northern area of Derbent cyclonic process and northern Caspian waters. The eastern part of the Southern and Middle Caspian shows the low productivity and heat storage during summer and, accordingly, the mnemiopsis population is not as abundant there as in the western shelf zone (relatively small abundance, narrow size spectrum, less reproductive activity).

The range of mnemiopsis in the Northern Caspian is limited to the deepwater zone and steep slope area (4-10 m) in the western part of the sea. It has not occurred yet in the east of the Northern Caspian, except for scattered weak individuals.

The major part of the mnemiopsis population inhabits upper water layers, above thermocline, some individuals may descend to the layer of thermocline and even occur below the layer of pycnocline. According to observations carried out under water, the largest densities of mnemiopsis occur in the surface area (down to 2 m) in summer, therefore it may be assumed that it feeds actively on neuston.

As is known, the basis of pelagic ichthyofauna is currently formed by three species of kilka the proportion of which in the total commercial catch of the Russian Federation before Caspian Sea invasion by mnemiopsis reached 70%. In the past three years, Russian catches of kilka were reduced from 150 600 t in 1999 to 17 000 t in 2003 (Table 2, 3). Only the common kilka stock remains rather stable as its reproduction occurs in the Northern Caspian outside the area of mnemiopsis development.

It is known that kilka feed mainly on copepods (Caspian Sea. Ichthyofauna and commercial resources, 1989). From A.A. Lovetski, K.P. Barysheva, L.A. Chayanov, B.I. Prikhodko, R.S. Skobelina's data, the food of adult anchovy kilka, the main species of kilka fishery, consists of 90-97% of the copepods. The main food items of kilka are two species, *Eurytemora* (*grimmi, minor*) and *Limnocalanus grimaldii* (70% of the stomach content).

When mnemiopsis, kilka's food competitor, appeared in the Caspian Sea, the species composition of meso- and macrozooplankton in the Middle and Southern Caspian declined drastically. From the data of the KaspNIRKh Laboratory of Hydrobiology, the gross biomass of the main food species of kilka decreased by a factor of 10, their abundance was reduced to 1/50. The main food species of anchovy kilka, Eurytemora and other copepods, were replaced by another member of copepods, Acartia gp. (tonsa, clausi), presently the leading representative of the mesoplankton.

Kilka stocks and recruitment, abundance of older age groups, their biostatistic characteristics and, as a result, research and commercial catches have drastically declined (Table 2).

Mnemiopsis invasion caused catastrophic disturbances in the Middle and Southern Caspian ecosystems, pelagium in particular.

Mnemiopsis invasion directly or indirectly affected all the trophic levels of the sea ecosystem, including higher ones, sturgeons and seals, as kilka are of great importance for their feeding.

The mass development of mnemiopsis also affected hydrooptical properties. In 2002, during the period of mass development of comb jellyfish in the central part of the Southern Caspian, water transparency decreased nearly by a factor of 2 in comparison with the annual average recorded The euphotic layer also became half as thick. The content of mineral phosphorus and nitrogen, the main sources of biogenic feeding of phytoplankton, was drastically reduced in the layer of 0-100 m (Table 4, 5).

At the same time, the content of silicic acid increased considerably. S.P. Volovik et al. (2000) report that comb jellyfish actively affect the circulation of biogenic substances in water as a result of active feeding on zooplankton and excretion of organic matter which decomposes easily. It may be suggested that because of eating on zooplankton, the mnemiopsis population "accumulates" mineral compounds of nitrogen and phosphorus. As reported by S.P. Volovik, " it was experimentally established that the mnemiopsis excretes a large amount of dissolved organic matter (carbon) and nitrogen, somewhat less phosphorus and very little urea, nitrates and nitrites".

Thus, the development of mnemiopsis population considerably affects the balance of biogenic substances in the eutrophic layer of the Middle and Southern Caspian pelagium.

The mnemiopsis seems to control the development of the most mass diatom, rhizosolenia. The indirect confirmation is a considerable increase in the content of silicic acid in the upper 0-100m layer of water.

Mnemiopsis spread through the area of the Middle and Southern Caspian, deepwater area of the western part of the Northern Caspian and can move with tidal water to the estuarine zone of the Volga River. It does not practically occur in the eastern part of the Northern Caspian, except for scattered weak specimens.

Mnemiopsis development is closely connected with thermal conditions of the sea. The major part of the population hibernates in the south of the sea with maximum densities, most likely, along the Iranian coast, especially in the warmest south-eastern part of the sea.

Table 2

Stocks, recruitment and biotic characteristics of anchovy kilka
(from the data of the Marine Fish Laboratory)

Parameters	1996	1997	1998	1999	2000	2001	2002	2003	1996-1999	2000-2003
Total biomass, thousand t	837	860	944	968	825	698	237	122	902	470*
Commercial stock, thousand t	627	759	765	722	600	643	237	122	718	400*
Annual catch, thousand t	148.7	159.5	215.4	271.4	192.7	74.7	90.0	53.5	199	103
Productivity, ind.	2115	1141	1908	2280	2450	226	2.5	20	1861	675
$W.2^+$, g	7.1	7.1	5.7	6.0	4.8	5.8	6.0	7.1	6.5	5.9
$W.3^+$, g	8.5	9.4	9.4	8.7	6.0	7.3	6.2	7.6	9.0	6.8
$W.4^+$, g	9.7	10.2	10.3	9.8	8.6	8.7	6.6	7.9	10.0	8.0

Table 4

Concentration of biogenic mineral phosphorus in the layer of 0-100 m in the Middle and Southern Caspian (µg/l)

Horizon, m	Middle Caspian									Southern Caspian								
	1964-1981	1985-1991	1998*	2000	2001*	2002	2003	1964-1998	2000-2003	1964-1981	1986-1991	1996*	1999	2000	2001*	2002	1964-1998	1999-2002
0-25	7.4	15.5	25.8	1.6	12.1	0.9	3.2	10.3	4.9	6.9	11.5	31.7	1.7	1.9	12.2	1.4	9.0	4.3
25-50	6.8	13.7	28.9	2.2	12.2	0.8	2.8	9.5	5.1	7.4	12.4	32.4	3.4	1.9	11.5	1.4	9.7	4.6
50-100	9.8	20.2	36.8	6.2	12.3	1.0	4.8	13.3	6.5	10.0	15.6	34.8	7.0	4.0	11.4	0.8	12.5	5.8
0-100	8.5	15.8	32.1	4.1	12.2	0.9	4.4	12.4	5.8	8.6	13.8	33.4	4.8	3.0	11.6	4.0	10.9	5.1

Table 5

Nitrate nitrogen content in the upper layer of water (0-100 m) in the Middle and Southern Caspian during 1985-2002 (μg/l)

Horizon, m	Middle Caspian				Southern Caspian				
	1985-1991	1998*	2000	2000-2003	1986	1998*	1999	2000	2001*-2002
0-25	9.2	2.3	0.3	3.1	5.1	1.5	0.8	1.3	2.6
25-50	17.8	9.8	0.4	6.1	11.4	1.8	4.7	3.0	6.2
50-100	66.4	35.4	27.8	21.9	79.4	24.3	30.8	29.6	13.0
0-100	40.0	20.8	14.1	13.2	43.8	13.0	16.8	15.9	8.7

Table 6

Silicic acid content in the upper layer of water (0-100 m) in the Middle and Southern Caspian during 1964-2002

	Middle Caspian				Southern Caspian				
Horizon, m	1964-1981	1985-1991	1998*	2001*-2002	1964-1981	1986-1991	1998*	1999	2001*-2002
0-25	303	266	117	258	329	180	174	45	219
25-50	336	266	243	408	320	192	252	124	250
50-100	498	399	374	500	435	258	260	296	374
0-100	409	332	277	416	380	222	242	190	304

*- autumn (September, October)

When water becomes warmer in spring, the range of mnemiopsis expands due to the transfer of juveniles to the north by resulting (circular according to Knipovich) current increasing in summer as in the case of anchovy kilka. Similarly, the mnemiopsis moves from the Black Sea to the Sea of Azov with compensatory current. Taking into account the fact that the mnemiopsis enters the Northern Caspian in late July – early August, the speed of its transfer is within 5-10 cm/ sec. which is quite in agreement with A.N. Kosarev's data (1975) on geo-strophic circulation in the Middle Caspian. The largest number of mnemiopsis during the summer period was detected in the southern part of the sea, the smallest in the middle part, its range limit is the deepwater area of the Northern Caspian. From 1999-2003 data, mnemiopsis abundance continues to increase (Table 1).

Caspian kilka harvest in the Caspian Sea (thousand tons) Table 3

States	1995	1996	1997	1998	1999	2000	2001	2002	2003
Azerbaijan	9.3	6.4	5.3	9.5	20.4	18.5	10.4	11.0	5.5
Kazakhstan	10.1	9.1	8.8	6.4	6.1	3.0	0.0	0.0	0.0
Turkmenistan	8.5	8.5	7.8	6.3	8.4	11.5	12.3	12.4	13.0
Russia	80.0	74.4	80.1	111.1	150.5	110.7	46.0	31.6	17.0
Iran	32.8	50.3	57.5	82.1	86.0	30.0	6.0	35.0	19.0
Total	140.7	148.7	159.5	215.4	271.4	182.7	74.7	90.0	54.5

Seasonal research into mnemiopsis population undertaken by KaspNIRKh during the past year was carried out in the entire area of the Caspian Sea to coastal waters of Iran. Its development was affected by rather a cold winter of 2002/2003 as compared to those in 2000-2002 when the mnemiopis population was very abundant in the Caspian Sea.

The winter range covered nearly 100% of the Southern Caspian and the southern part of the Middle Caspian. Comb jellyfish did not occur in shallow water areas (h<20m) of the Southern and Middle Caspian because of low water temperature ($t<6^0$ C). It was not seen in the Northern Caspian and northern part of the Middle Caspian either (Fig. 1a).

In February 2003 its concentration in the Southern Caspian varied widely: from several specimens to 321 ind./m^3 (8.0 g/m^3). Its density was on average 81 ind./m^3 with biomass 2.3 g/m^3.

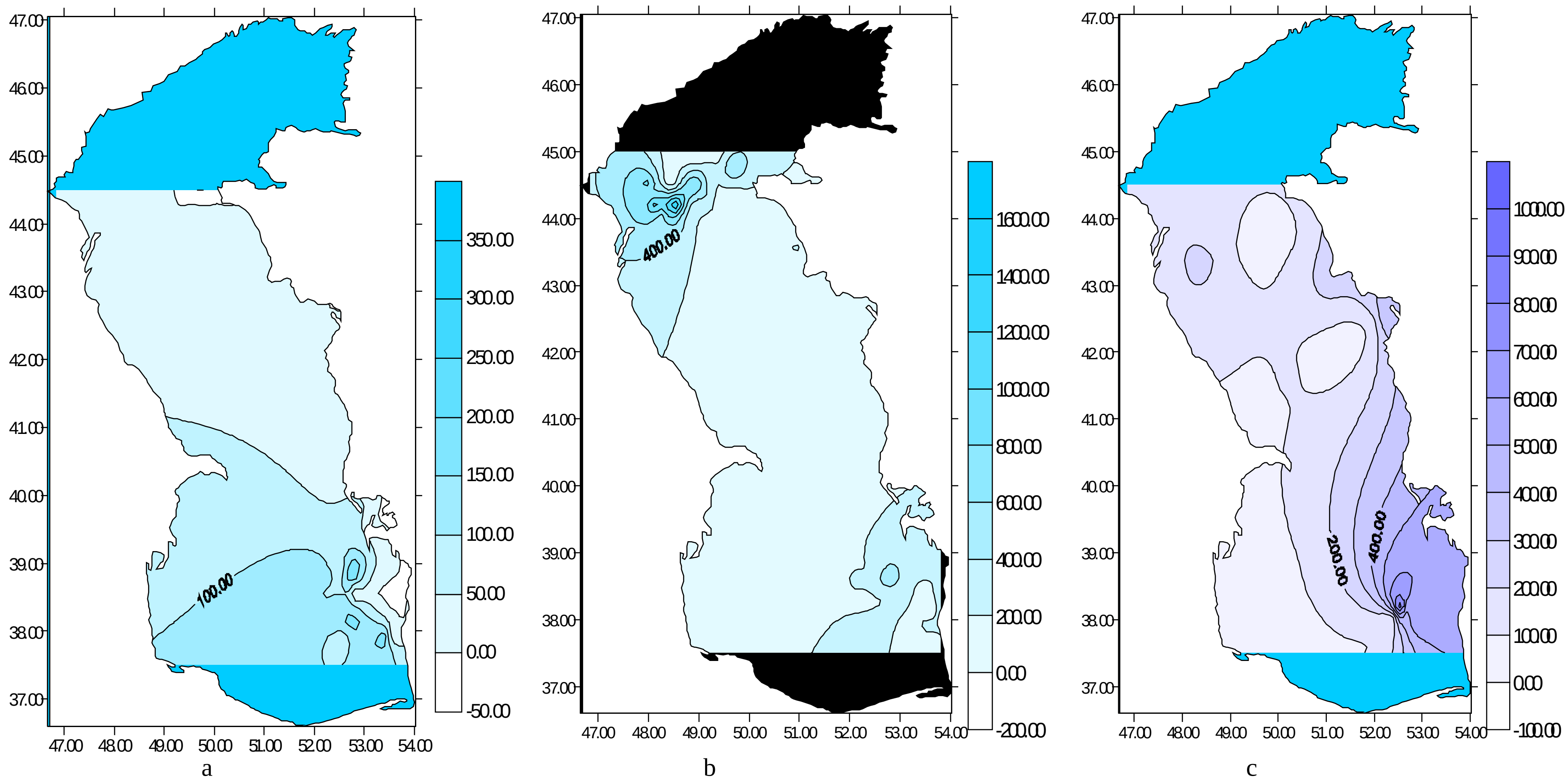


Fig. 1 Seasonal distribution of the Caspian population of *Mnemiopsis leidyi* (ind./m^3) in 2003:
a – February-March; b – August-September ; c – October-November

Juveniles, larvae and eggs (size class < 10 mm) formed the basis of the winter population of *Mnemiopsis leidyi* in the Southern Caspian and in the south of the Middle Caspian. This group accounted for 97.7-100% in the majority of stations while the average number of mnemiopsis in the Middle Caspian was 120 ind./m^3.

Thus, even during the cold winter period at water temperature below 9^0 C this species of gelatinous organisms occurred in large amounts and was represented mainly by juveniles and eggs.

In **spring**, the size of *Mnemiopsis leidyi* population that hibernated in the Southern Caspian was rather large which enabled the reproduction of considerable amounts of larvae and juveniles and their spread throughout the uninhabited areas of the sea.

The spring range of comb jellyfish in April 2002 covered 90% of the Southern Caspian and some 20% of the Middle Caspian. In 2003 its range in early March already covered 95-97% of the Southern Caspian and 40-50% of the Middle Caspian.

At the very beginning of spring season (the first 10-day period of April) mass concentrations of larvae were recorded in all the parts of the Southern Caspian.

In spring the density of comb jellyfish in the Southern Caspian was 154 ind./m^3, but the proportion of eggs, larvae and juveniles (size class < 10 mm) decreased though remaining the major portion of the population - 96.2%. This group accounted for 92.0% in the Middle Caspian.

At the beginning of **summer** (June-July) 2003 as in 2002, mnemiopsis was not seen in the Northern Caspian. The largest concentrations of the population of *Mnemiopsis leidyi* were recorded in the Middle and Southern Caspian. They were detected in the western part of the shelf zone from the coast (above 10-12 m) to the depth line of 650 m. Comb jellyfish occurred sporadically in the eastern shelf zone and in the central part of the sea (Fig. 1b).

By the end of summer, in August, the core of the population moved to the north along the western coastline and reached the boundary between the Middle and Northern Caspian (Fig. 1b). Its density was 12-14 times as large as that recorded in August 2002 (37 ind./m^3 or 12.5 g/m^3).

In August 2003 the average density of gelatinous organisms per unit of volume in the areas of maximum concentration in the Southern Caspian reached 602 ind./ m^3 (36.0 g/ m^3) which is consistent with the same level in 2002 (Table 5). The mean concentration of mnemiopsis in the east of the region was 238 ind./ m^3 (16.0 g/ m^3), i.e. increased by a factor of 2 as compared with the value recorded last year.

By summer, the proportion of the small size group (<10 mm) decreased while the total abundance and biomass of the population increased.

In **autumn** (October) the largest concentrations of comb jellyfish occurred along the western coast above the depth of 50-200 m in the western shelf zone of the Middle and Southern Caspian (Fig. 1c). Its maximum densities there amounted to 442-926 ind./ m^3 (23-28 g/ m^3) which is 1.6-2.6 larger than that during the same period of 2002 (280-355 ind./ m^3 or 9.0-11.5 g/ m).

In comparison with rapidly cooling waters of the shallow water area of the Northern Caspian, waters of the Middle and Southern Caspian remain rather warm (above 15^0C). Therefore, the reproduction of the gelatinous continued. The biomass of the small size group in the Middle Caspian remained at the level recorded in summer (4.2 g/m^3) and accounted for 62%. The biomass of that group in the Southern Caspian continued to increase and reached 12.0 g/ m^3.

KaspNIRKh experts studied vertical distribution of mnemiopsis carrying out underwater telephotography and video shooting. From visual examination and video material of 2001-2003, reliable data were obtained concerning the vertical distribution of comb jellyfish.

During the summer-autumn period, the maximum abundance of *Mnemiopsis leidyi* in the Southern and Middle Caspian was recorded in the warmest surface water layer of 0-2 m. As in the previous years (1999-2002), the lower limit of the major concentration of comb jellyfish coincided with the upper limit of thermocline (25-30 m). Its concentration under thermocline decreased abruptly. Because of lowering and subsequent destruction of thermocline in autumn, mnemiopsis may occur at a depth more than 40-50 m.

Data of 2003 confirmed previous years' findings that the major area of development of comb jellyfish population and of its largest concentrations throughout the year is the Southern Caspian. Large concentrations and biomass of that gelatinous are also recorded during the cold period (2753 ind./ m^3, 115 g/ m^3).

Comb jellyfish can reproduce in that part of the sea during the winter period and begin to spread along the western coast to the north in spring. In 2003 after rather a cold winter, the range limit of the population in the Northern Caspian was reached 2-3 weeks later in the second half of August.

From the data on the abundance of mnemiopsis population in 2000-2003, it is predicted that in the nearest future (up to 2005) its abundance will stabilize in the Southern Caspian and will continue to increase in its range limit in the east of the Middle Caspian and in the Northern Caspian. Based on the 2000-2003 observations, it may be reported that there is no stagnation of mnemiopsis population and it continues to increase (Table 1). Therefore, urgent measures are needed for its regulation (due to beroe acclimatization).

Ecological catastrophe in the Caspian Sea because of its invasion by mnemiopsis in the late 1990s predetermined the elaboration of practical measures to control its abundance.

Considering positive changes in the situation in the Azov-Black Sea basin, international experts' proposal was adopted (Baku, 2001) to introduce an obligatory mnemiopsis predator, comb jellyfish beroe, which was regarded as a main way of solution to the problem.

In this connection, KaspNIRKh started in-depth studies of incidentally "introduced" species and of that planned for introduction.

As literature data are presently based on Azov-Black Sea populations, experiments were made on Caspian mnemiopsis and beroe the offspring of which was produced in the Caspian water.

Experiments carried out during three years (2001-2003) revealed:

a) a method of collecting and transporting *Beroe ovata* for a long distance (2 000 km);

b) technology for adaptation of *Beroe ovata* from the Black Sea water close to oceanic one in composition and salinity (17-19‰) to Caspian water with quite a different ionic composition and lower salinity (8-11%);

c) beroe tolerance to environmental factors (water salinity, temperature, gas regime and pH);

d) feeding habits of comb jellyfish (mnemiopsis and beroe), daily rhythms of feeding and the rate of food digestion at different water temperature, the rate of food intake at its various concentration in habitats;

e) bioproduction potential (fecundity) of beroe adapted to Caspian water.

In addition to that, the technology of beroe cultivation in Caspian water was worked out.

Experiments were performed using samples taken from different zones of the north-eastern part of the Black Sea (Utrish Village, Gelendjik City).

Data obtained were used as a basis for devising a method of cultivation and grounding possible introduction of *Beroe ovata* into the Caspian Sea.

Results

1. Three-year studies undertaken by KaspNIRKh showed that beroe should be caught in the Black Sea using a conic net that excludes the damage of the gelatinous and transported to the experimental base Turali-2 located at the Dagestan coast by motor transport in plastic containers 50-100 l. When beroe are transported for long distances (2 000 km), their density should be rather small: 40 specimens per 50 l to prevent them from being damaged and deliver the maximum amount (92.5%) of viable organisms. The individuals supplied were subsequently used for adaptation, offspring production and cultivation.

When transported at large densities (50 ind./50 l), the number of damaged individuals increases by a factor of 1.5 because of accumulation of products of metabolism in water.

2. When developing the technology of beroe adaptation to Caspian water of different ionic composition and lower salinity (8-11 against 18-19‰), the regime of water desalination was worked through. Water desalination was carried out by different methods taking into account beroe high sensitivity to saline conditions. On the whole, the process of adaptation of beroe of different size classes to the reduction in salinity was almost the same, but unlike large individuals, small and medium ones spent less time for that process (by 2-3 days). In general, the process of adaptation takes some 7-8 days.

It was established that *Beroe ovata* showed high adaptability not only to various salinity of Caspian waters, but also to re-adaptation from low (10‰) to higher salinity (11-12‰).

3. It was reported that the beroe was very particular about habitat conditions. When held in man-made conditions, it needs continuous water aeration and its daily replacement (50%) to prevent the accumulation of metabolic products.
4. In the process of the experiment, considerable differences were recorded in feeding habits and food spectrum of comb jellyfish. Mnemiopsis in the Caspian water feed on
5.

all the members of zooplankton while beroe feed only on mnemiopsis which is in agreement with literature data (Volovik, 2000).

When plankton organisms are abundant in mnemiopsis habitat, it completely fills the gastro-vascular cavity ejecting part of the consumed food through the mouth. Besides, a considerable amount of zooplankters, 2-3 times greater than the quantity of consumed prey, are immersed by mnemiopsis into excreted slime which is held with tentacles. The food ejected from the digestive system and immersed into slime is not already used by comb jellyfish, therefore the loss of valuable high caloric food may be very large (film).

Mnemiopsis display clear daily rhythms of feeding which are closely connected with water temperature. Peaks of feeding at high temperatures (28-25^{0}C) coincide with nighttime. At that, daily food rations are not too large. When water temperature is low (20^{0}C), the feeding activity decreases markedly while the time of digestion increases. Most prey is eaten by mnemiopsis during daytime while during nighttime it does not almost feed (Fig 2,3).

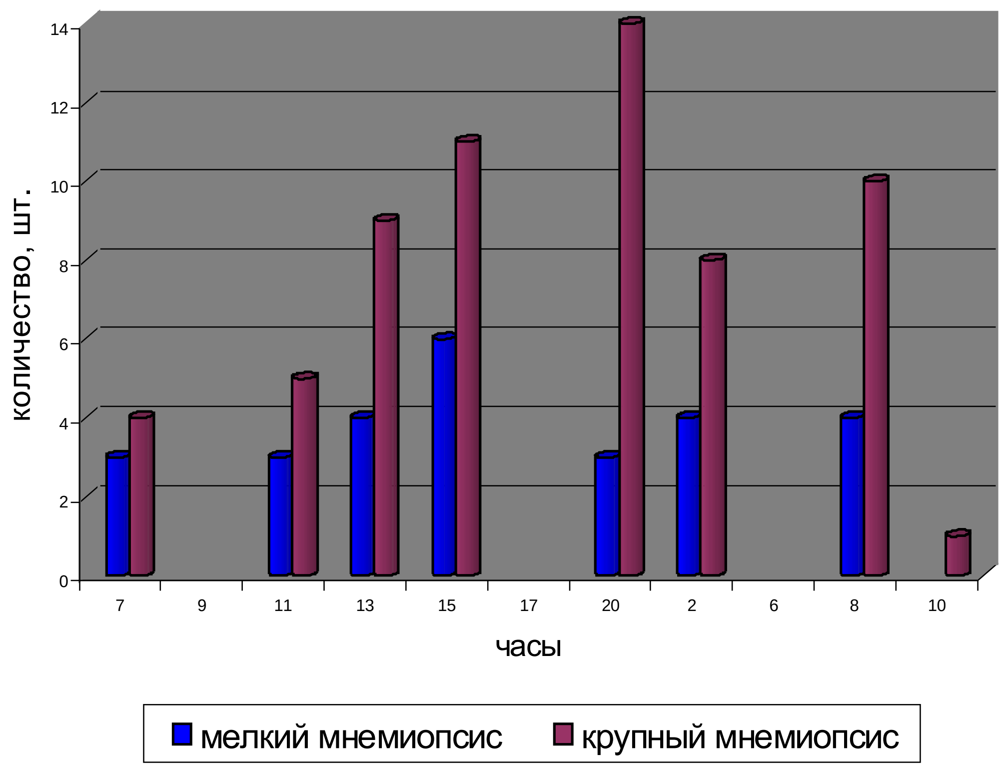


Fig. 2 Daily rhythm of mnemiopsis feeding at a temperature 25^{0}C

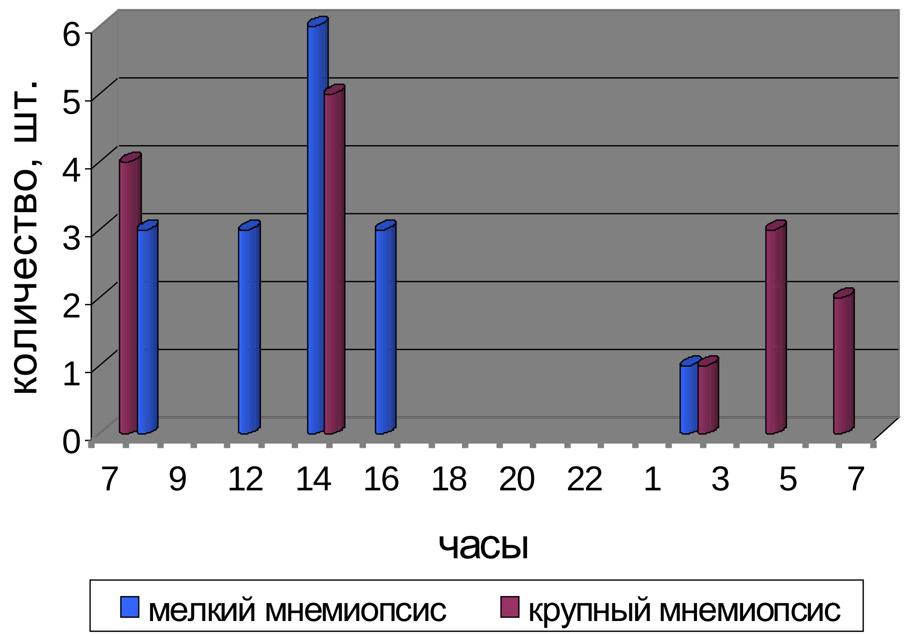


Fig. 3 Dynamics of mnemiopsys feeding at a temperature 20^0C for a day

Feeding habits and food spectrum of beroe are quite different. Even when it starves for a long time (5-6 days), it does not feed on plankton, micro-warms and Ostracoda. When the concentration of plankters is high, it clenches its mouth and changes its usual body shape looking like a ball (film). Beroe's main food is comb jellyfish mnemiopsis which is ingested by beroe mostly as a whole. Daily rhythms of beroe feeding, just as in mnemiopsis, depend on water temperature. When the temperature drops, the feeding activity of all the age classes decreases appreciably. Beroe show two peaks of feeding activity which occur during the day- and nighttime though, in general, beroe feed continuously with some intervals for digestion, but even at that moment they can fill their food reserves repeatedly. The time spent to digest food differs considerably in beroe of various size and depends on the amount of food consumed and water temperature. The amount of daily ration is of great importance for beroe of large and medium size and also depends on water temperature too. When the temperature rises, the amount of the food consumed increases.

6. During their reproductive cycle under favorable biotic and abiotic conditions, the Azov-Black Sea beroe adapted to Caspian water produced 10 times more eggs (some 250) than their first offspring could do. This can probably occur only in man-made conditions when the reproductive success can not be achieved because environmental conditions are worse than in natural water body.

All the size classes display impaired growth when water temperature drops and the concentration of food is reduced. When conditions are quite favorable, 3-cm long beroe produce the second offspring on the 20-th day. In shortage of food, their growth slows down and in order to reach the breeding age, they need more time (by 5-7 days).

7. In breeding practice, there is experience of aquatic invertebrates cultivation (Bogatova et al., 1975, Zhemaeva, 1984, Ivanov, Abrosimov et al., 1975). Similar work on rearing gelatinous organisms was not carried out.

Cultivation suggests recurring production of beroe offspring with its subsequent introduction into the Caspian Sea in order to regulate the number of predator invader - mnemiopsis. Data obtained indicate a real possibility not only of adaptation, but also of regular production of beroe of different size (including mature) which expands their range in the Caspian Sea.

There is no experience of introduction of gelatinous organisms into water bodies including the Caspian Sea.

Summarizing the experience of recent introductions of hydrobionts (Zenkevich, Birshtein,1937a, Zenkevich, 1940, Ioganzen, 1950, Karpevich, 1960, 1965, 1968, Ioffe et al., 1972), having studied the biology and ecology of invaders as well as compatibility of their environmental requirements and conditions in the stocked water body which is quite like maternal by temperature and hydrochemical conditions, it may be concluded that beroe acclimatization in the Caspian Sea is quite possible.

The biotechnique of beroe introduction into the Caspian Sea is thought to consist of its stage-by-stage acclimatization for three years.

During the first year, only a mixed population of beroe (some 5 000 individuals) consisting of 60% of large individuals, 20% of medium and 20% of small individuals adapted to Caspian water should be introduced. The largest percentage of large-size beroe in the mixed population depends on two factors. Firstly, they will begin mnemiopsis extermination more actively thus minimizing its concentration and ensuring an increase in reproductive potential of the introduced species and rapid offspring production. Secondly, that is the shortest way of acclimatization: the phase of naturalization begins one generation earlier than in the case of transplanting juveniles.

The introduction of medium- and small-size individuals may prevent occasional failures.

In developing commercially exploitable populations in the second and third years, it is necessary to use a method of "mixed stocking". Of each portion of beroe adapted to Caspian water, one half (500-600 ind.) of large-size beroe will be released directly into the sea for its population restocking and more intensive extermination of mnemiopsis while the other one will be used for cultivation. Beroe of different size classes which are stored up when rearing in artificial conditions will be released at regular intervals in order to expand the area of beroe introduction into the Caspian Sea.

Releases of beroe adapted to Caspian water should be carried out offshore at a depth of 10-20 m, water salinity of 9-12 ‰ and food (mnemiopsis) concentration not less than 100 ml/l repeatedly. The development of the introduced species may be monitored by divers using underwater video equipment.

Beroe should be introduced into several areas of the Caspian Sea including the north-western coast of Dagestan because:

- the time of mass development of mnemiopsis in that part of the sea (the second half of August-September) coincide with the moment of mass development of beroe in the north-eastern part of the Black Sea. It may be caught and transported to the Caspian Sea in the shortest way;
- water transfer in that area occurs from north to south.

The second promising region is the Southern Caspian including the coast of Azerbaijan and Iran. Favorable conditions for beroe introduction at the coast of Azerbaijan may be in the area south of the Apsheron Peninsular and in the south-eastern part (Islamic Republic of Iran).

Rather a large abundance of beroe may be expected two years after introduction, similar to that process in the Black Sea.

One of the most important requirements that should be complied with before beroe acclimatization in the Caspian Sea is the obligatory conducting of integrated parasitological, microbiological and virologic studies of the members of the family Ctenophora in the maternal (native) water body (the Black Sea) and in experimental conditions according to Russian and international practice.

In the literature available there are no data showing that the family Ctenophora may act as carriers of bacteria or virus. The results of microbial and virologic studies of comb jellyfish (*Beroe ovata, Mnemiopsis leidyi)* and their habitat in the Black and Caspian Seas described in the biological foundation are presented now for the first time.

In compliance with international standards in order to establish the epizootic status of the population, it should be investigated not less than twice a year for four years (Council Directive 91/67/EEC as last amended by Directive 98/45/EC, 1988; 1991).

In connection with indicated above, in the case of positive decision on beroe acclimatization in the Caspian Sea, virologic examination of each batch of comb jellyfish supplied should be performed for the next few years. At that, the examined material should be taken from the batch of beroe prepared for transportation and before the results of examination are obtained, beroe should be placed in quarantine in compliance with home and international standards that regulate hydrobiont transportation. According to published and informal data, it is quite possible that there are viral infection foci of fish in the Black Sea and their invasion of the geo-physically isolated basin of the Caspian Sea may lead to quite undesirable consequences.

Integrated research into comb jellyfish showed no viral agents and parasitic fauna in the material under examination. At the same time, various microflora indigenous to marine ichthyofauna was isolated from mnemiopsis and beroe, these are Aeromonas and vibrioflora common to the Black Sea; Pseudomonas and acinetobacteria common to the Caspian Sea.

Enterobacteria revealed in the microbial spectrum of the Black and Caspian Sea species under examination show that they may act as carriers of opportunistic microflora. Elimination of one complex of microflora by another may be due to different chemical composition and various salinity of Caspian and Black Sea water.

From the results obtained in 2003, the microbial spectrum of beroe originally included 17 species of bacteria of seven genera: Aeromonas, Citobacter, Enterobacter, Hafnia, Pseudononas, Flavobacterium, Vibrio. The family Vibrionaceae (*Aeromonas hydrophila* and *Vibrio alginolyticus*) dominated the bacteriocenosis of Black Sea comb jellyfish (46.4% of the strains of all isolated microflora). Microorganisms of the family Pseudomonadaceae were second in frequency of occurrence (28.6%) with subdominating *Rs. aeruginosa*

Enterobacteria accounted for 21.4% of the strains. The data obtained indicate a significant anthropogenic contamination of the Black Sea ecosystem.

After 7 days of adaptation in Caspian water the beroe under study were contaminated mostly with Pseudomonas and acinetobacteria (52.0 and 28.0%, respectively). The proportion of the families Vibrionaceae and Enterobactericeae in the material under analysis was not large. A similar tendency remained after complete transplanting of comb jellyfish into Caspian water. Consequently, 100% elimination of beroe indigenous microflora by bacteria common to the Caspian hydro-ecosystem occurs in the process of its adaptation.

Analysis of the material using pathogenicity markers showed that the Black Sea microflora exhibited proteolytic, lecithinic and hemolytic activity in 54.7% of the cases while the isolated Aeromonas showed 100% DNA activity. After beroe adaptation to Caspian water, pathogenicity markers of bacteria contaminating that comb jellyfish were at a level of 22.7% or on average 2.4 times lower, which is due to phenotypic characteristics of bacteriocenosis.

The result of bioassay on Caspian fish contaminated with the strains of dominating groups of Black Sea microorganisms (Aeromonas, Vibrio, *Rs. aeruginosa)* turned out positive.

Fish contaminated with Aeromonas and Vibrio were found dead 5-18 hours later while those contaminated with *Rs. aeruginosa* died 2 days after contamination.

The postmortem examination indicated considerable pathologic abnormalities in the internal organs, muscle lysis and a papule in the site of injection. Original strains of bacteria were isolated from affected organs. The group of fish under control remained quite normal.

The bioassay made provided a negative result as it is considered positive only in the case of mortality of 50% of the fish with signs of exterior and interior lesions.

Similar result were obtained from the bioassay on white mice. This implies that the dominating Black Sea microflora is pathogenic both for native Caspian ichthyofauna and for warm-blooded animals.

Thus, from the results of the research done it may be concluded that in acclimatizing Black Sea beroe in the Caspian Sea, it is necessary to carry out adaptation or quarantine for 2-3 weeks to eliminate Black Sea microflora with subsequent control and integrated examination of Ctenophora.

First Regional Technical Meeting on
Possible introduction of *Beroe ovata* into the Caspian Sea,
22-23 February 2004, Tehran, Iran

Lessons to learn from the Black Sea
with respect to invasion of the Caspian by the ctenophore *Mnemiopsis leidyi*

Institute of Marine Sciences
Middle East Technical Univ.
Erdemli, Turkey

Active participants...

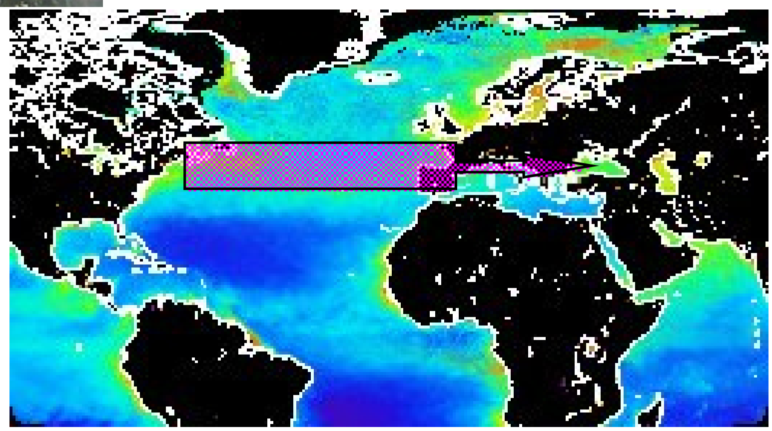
Active participants...

Why talk about the Black Sea

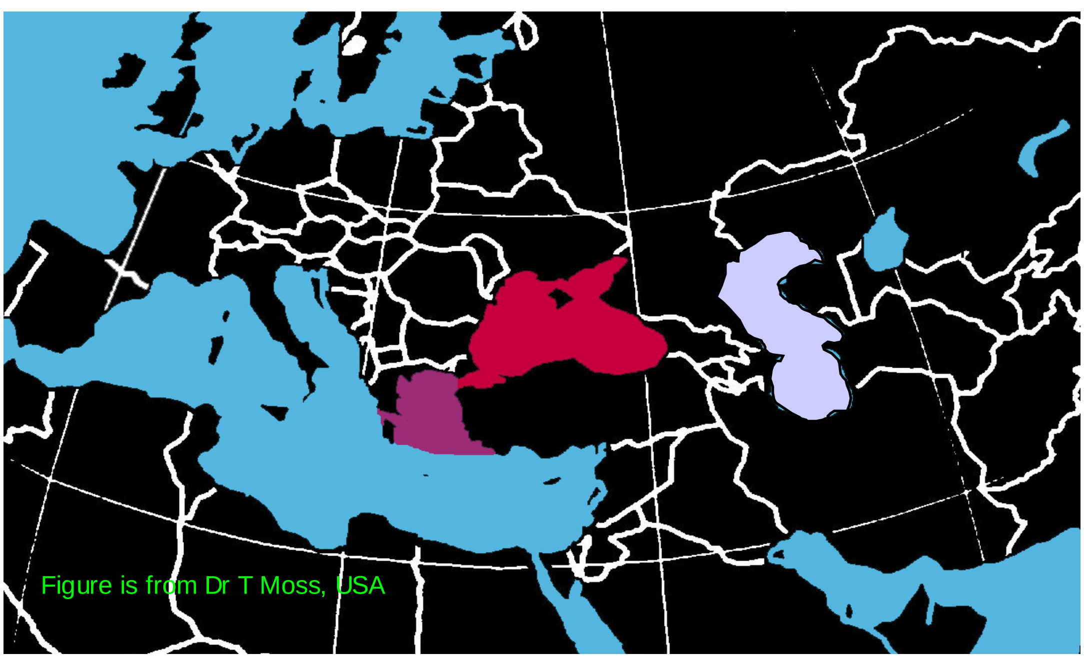
- We learnt alot from similar invasion events occuring in the Black Sea during the last two decades. This information is, at this very time, vitally important in solving the greatest problem ever of the Caspian Sea ecosystem....

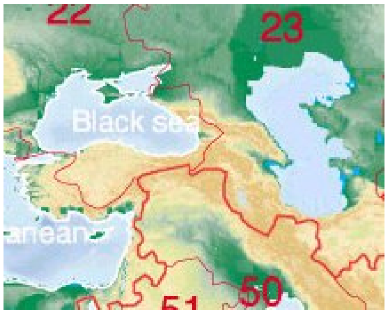
The ctenophore
Mnemiopsis leidyi
transported via ballast water from the western Atlantic into the Black Sea in the late 1980s

This has been one of the most important invasion events of the world seas!



Mnemiopsis is a not problem for “any” ecosystem





Both seas are similar in several aspects…

- Similar surface area (BS 4.2 x10^5 km^2, Caspian 3.9 x10^5 km^2)
- Both have large catchment area (2 million km^2 for BS, 3.5 million km^2 for Caspian)
- Both fed mainly by one large river
- Both have one large shallow region
- Both have one large lagun

Similar in several aspects…

- Both sustain a sizeable pelagic fishery (about half a million tons)
- Both have top mammalian predators
- Both have high number of endemic spp
- Both have high number of invasive

Sensitive ecosystems

- Both seas are sensitive to anthropogenic impacts as they are almost enclosed ecosystems…..

✓ Eutrophication

✓ Invasive species

Large nutrient-rich riverine input as the main reason of eutrophication of the Black Sea

($>2 \times 10^6$ km^2; Zaitsev & Mamaev 1997)

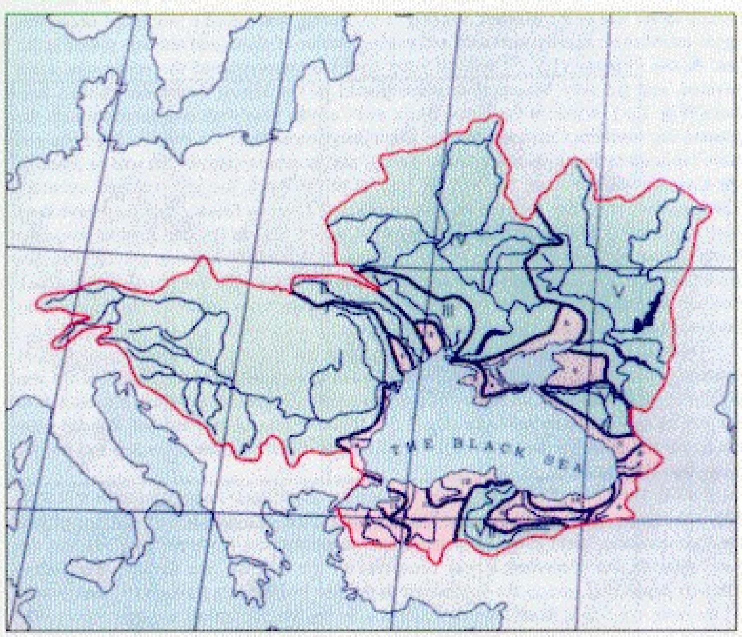
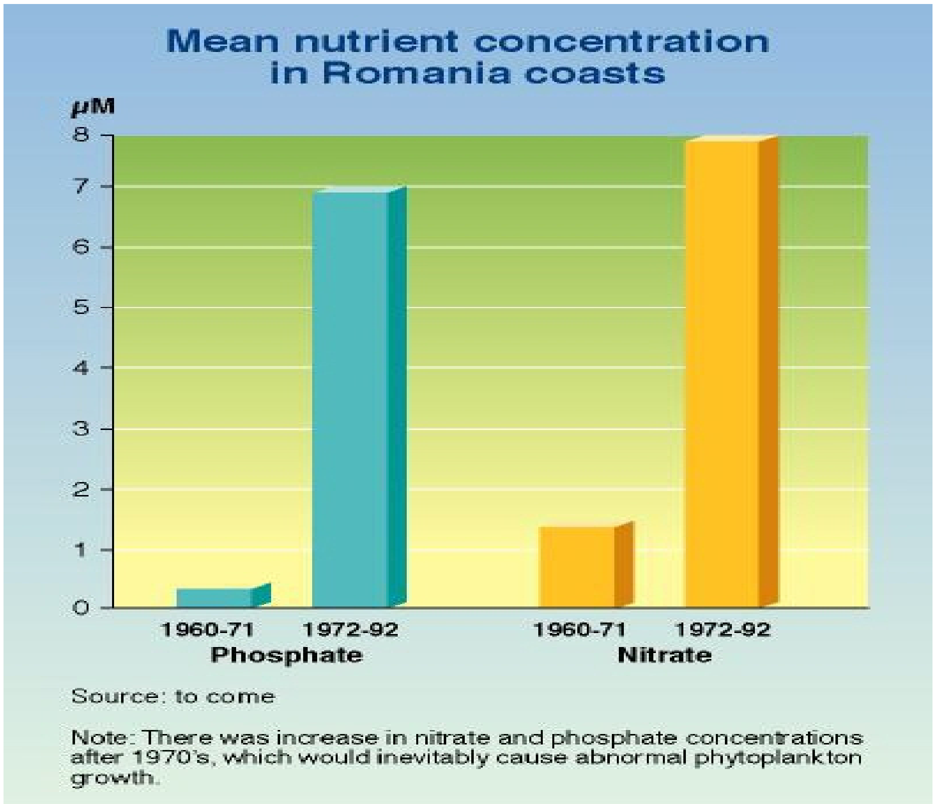
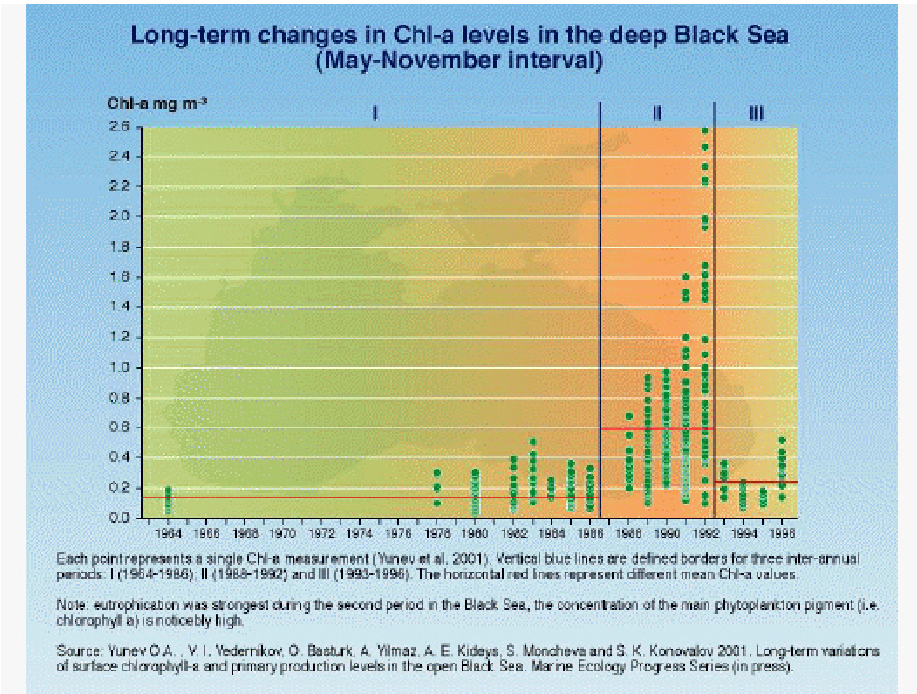


Fig 2. Black Sea Drainage Basin

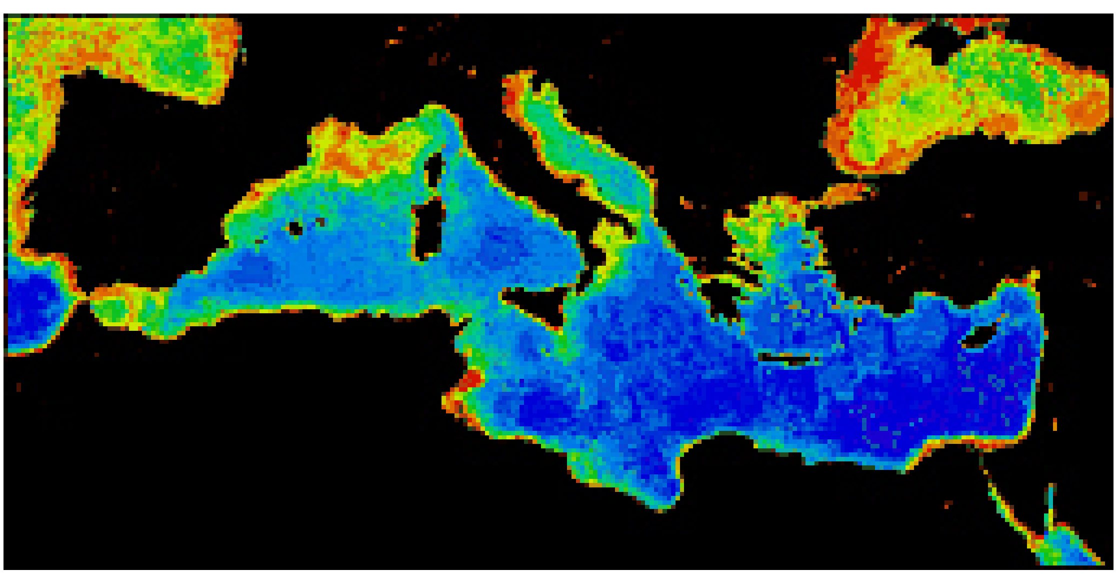
Root source: Increase in nutrients



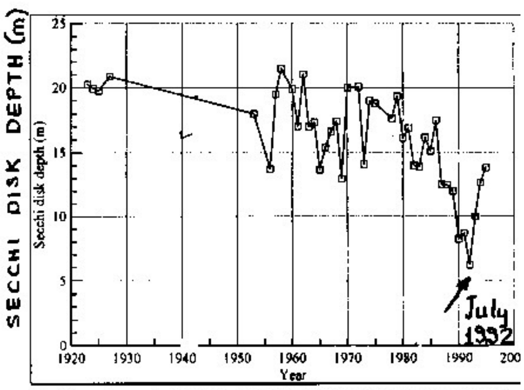
Eutrophication effect:
Increase in chl biomass



Annual chlorophyll distribution from SeaWIFS

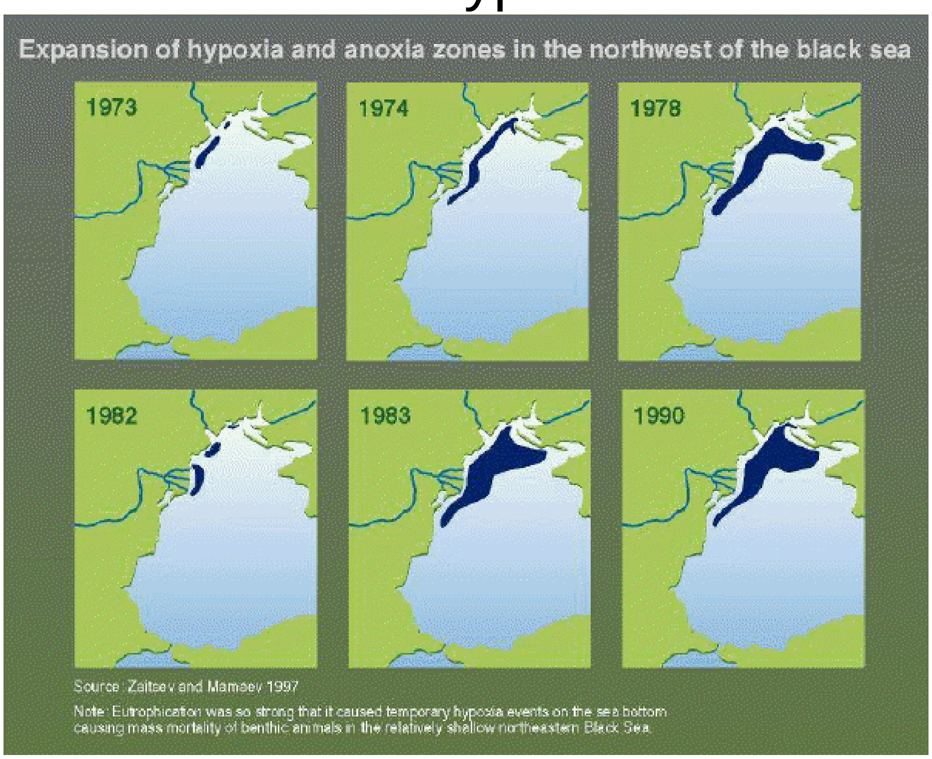


Eutrophication effect:
Decrease in Secchi depth



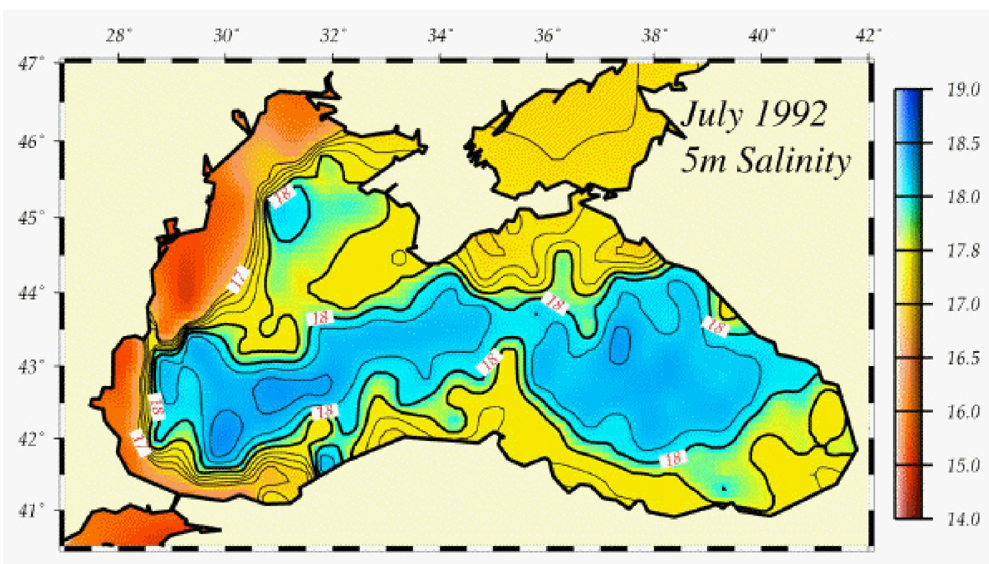
The long-term variability of annual mean Secchi disk depths in the central deep part of the Black Sea in 1922-1995.
Ref: Vladimirov et al., 1997

Eutrophication effect:
Increase in hypoxic areas

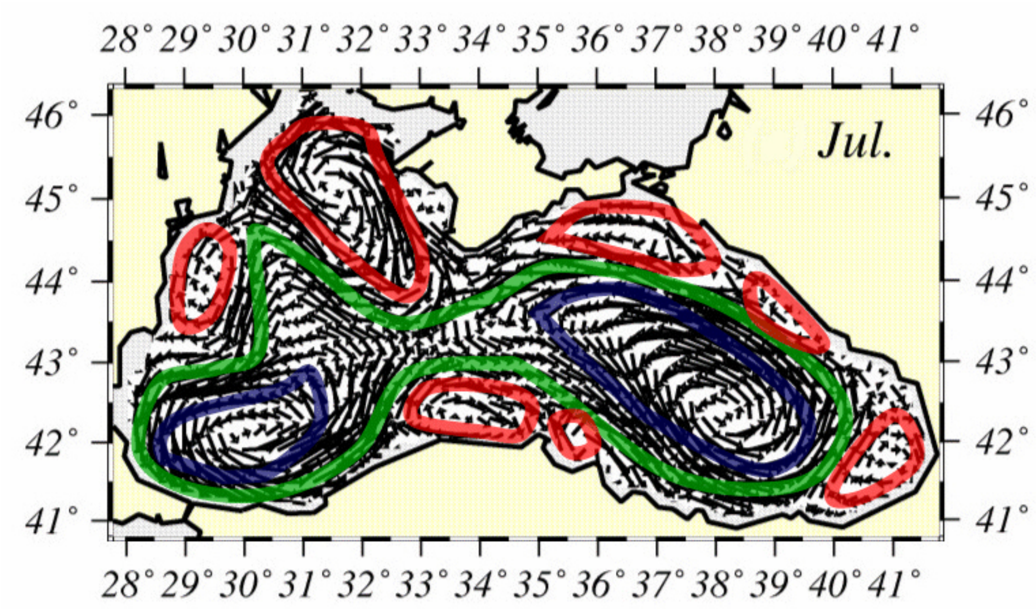


- 1) Eutrophication: affected more the NWS compared to the open sea

Salinity in the Black Sea (from Prof T. Oguz, IMS, Turkey)



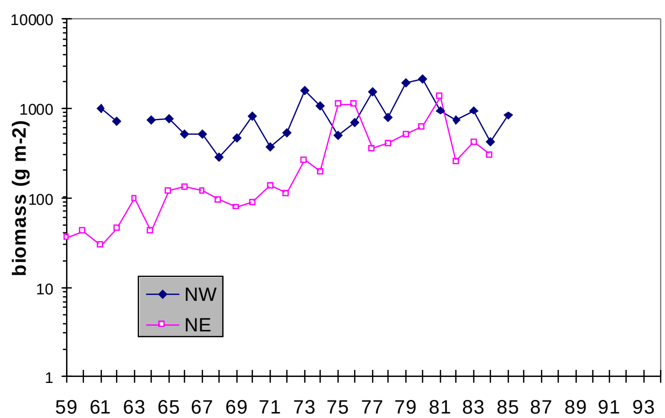
Currents in the Black Sea
(from Prof Temel Oguz. IMS, Turkey)



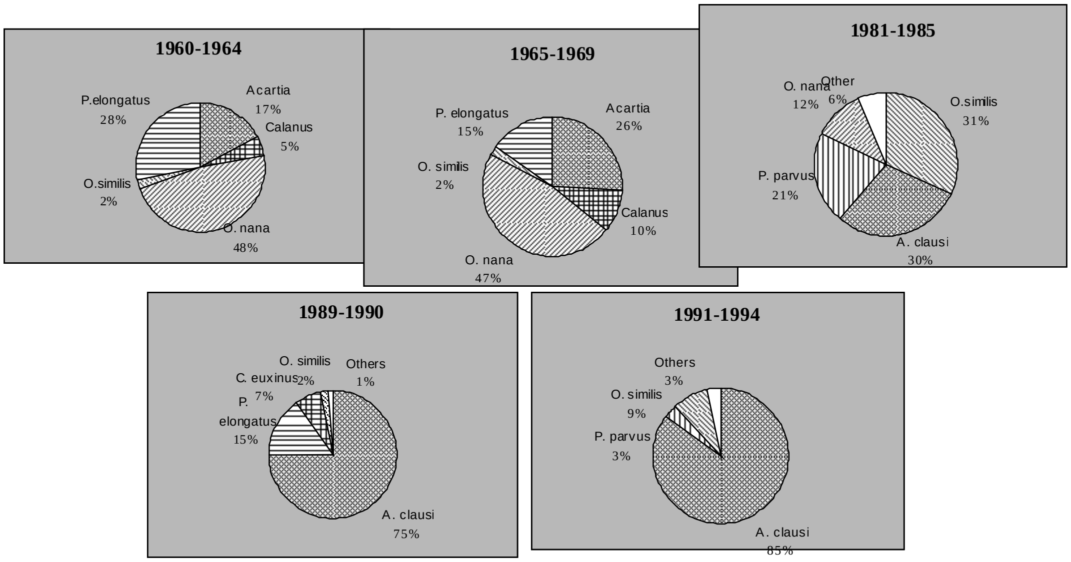
<u>Eutrophication effect:</u>

Increase in phytoplankton biomass

(Kovalev *et al.,* 1998d)

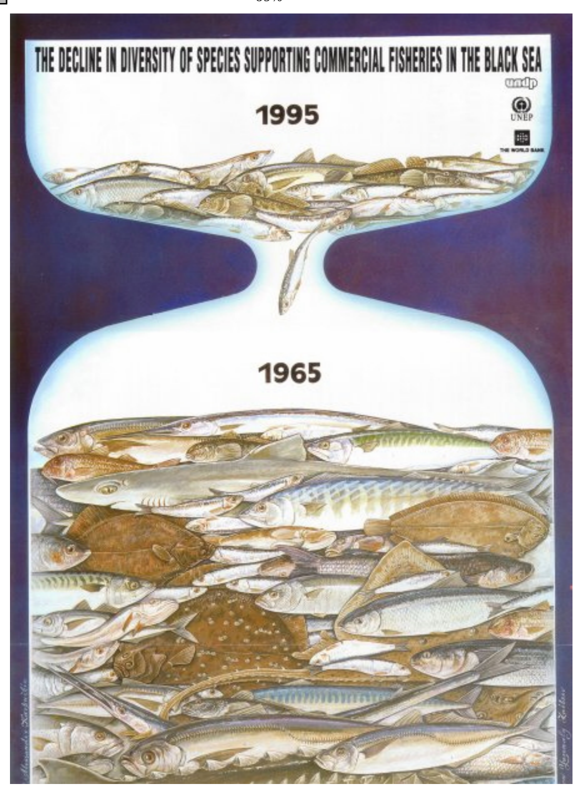


Eutrophication effect: Compositional changes in copepod biomass in Sevastopol Bay



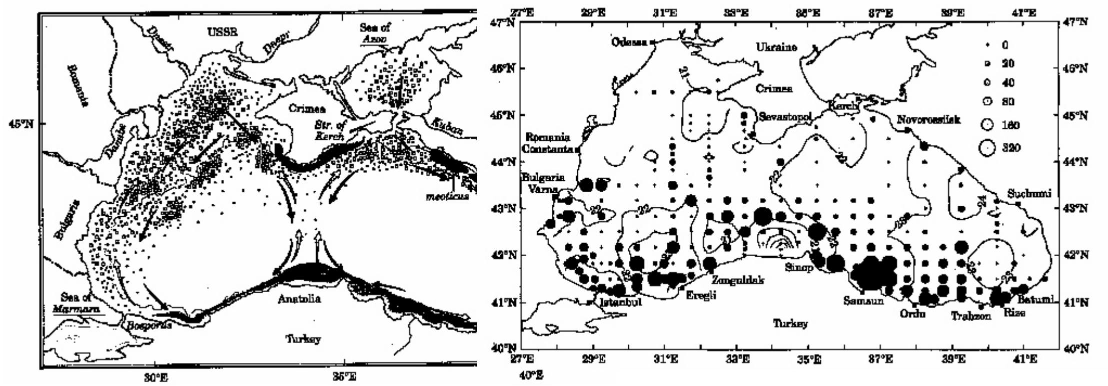
Eutrophication effect:

Decrease in fish biodiversity



<u>Eutrophication effect:</u>

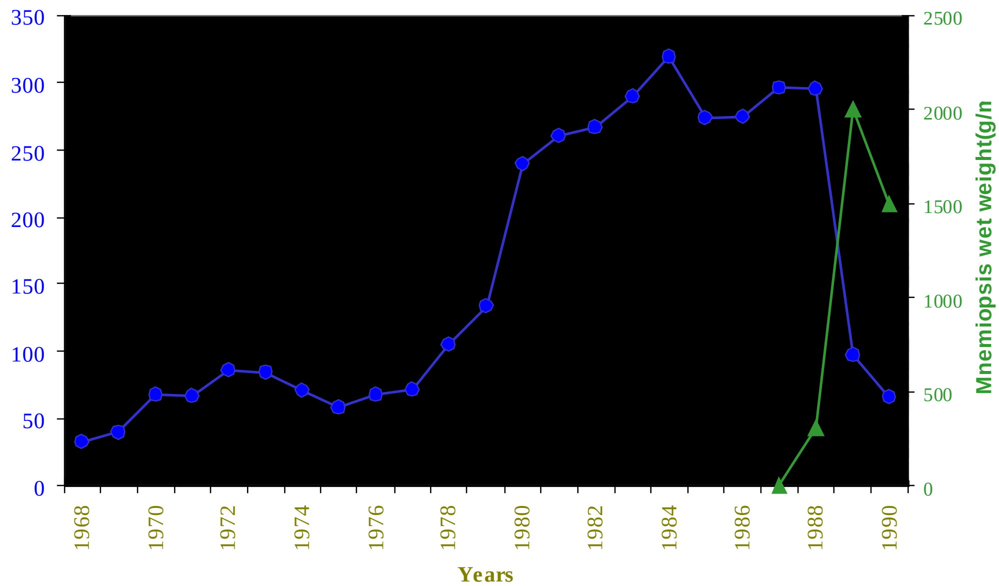
Changes in the spawning grounds of

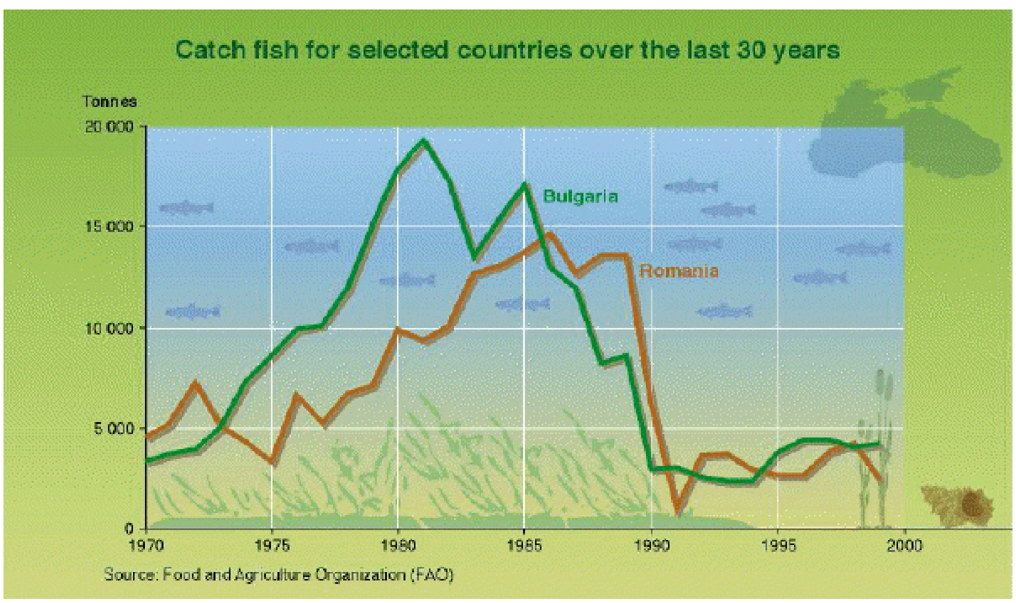


The second of the two main events which greatly affected the ecosystem of the Black Sea is :

- 1) Eutrophication
- **2) Introduced species**
- Notably the alien ctenophore *Mnemiopsis*; in the late 1980's reaching very high biomasses (almost 1 billion tons!) in the Black Sea, immediately affected all compartments of marine life

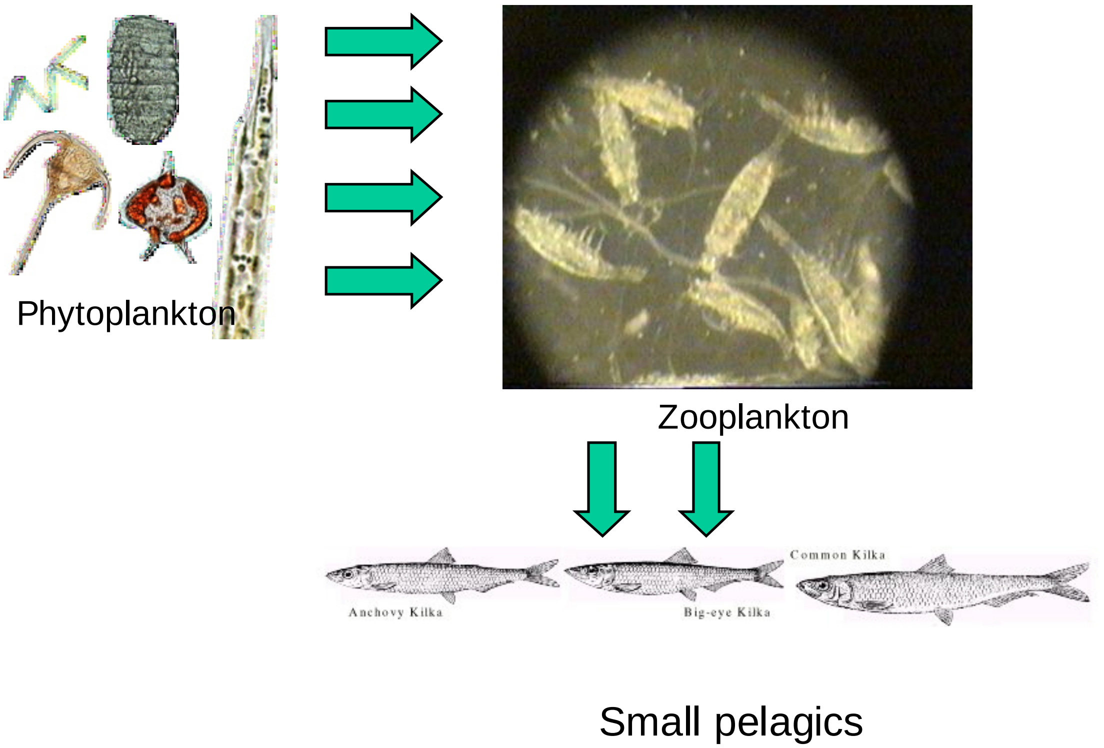
Relationship between the decrease in Turkish anchovy catch (as 10^3 tonnes) and *Mnemiopsis* (WW m^{-2}) outburst



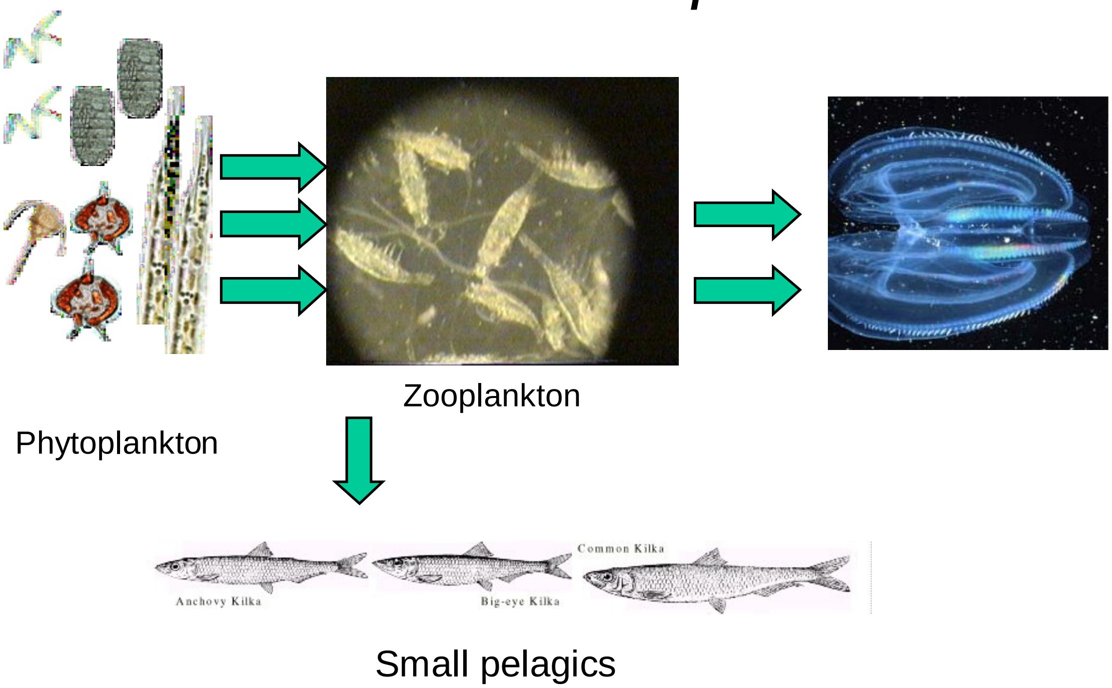


The loss of Turkey alone was roughly 250 million dollars per year due to these low levels of fishery

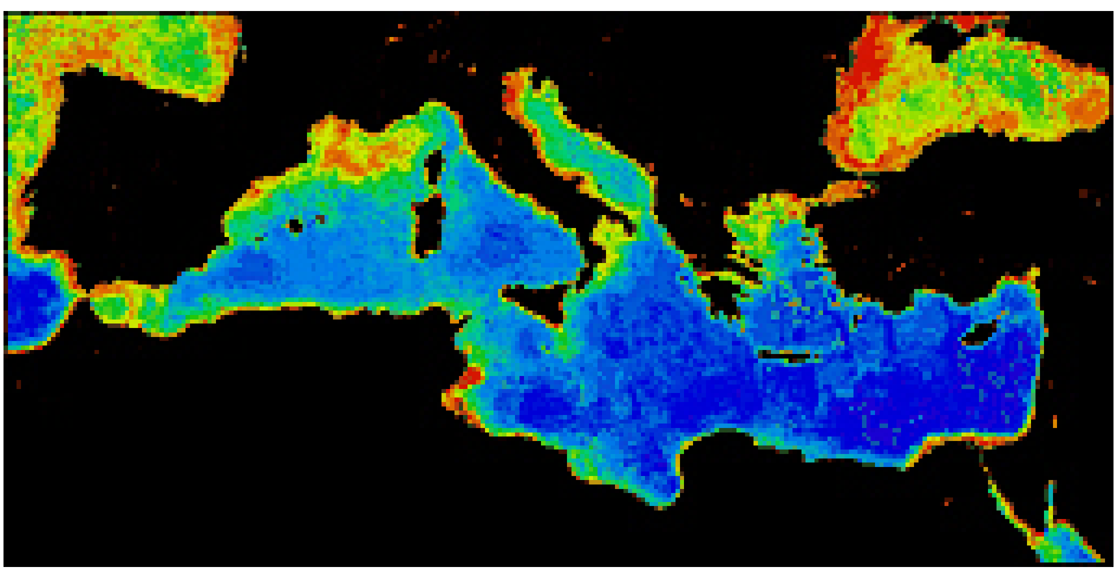
How does *Mnemiopsis* affect ?



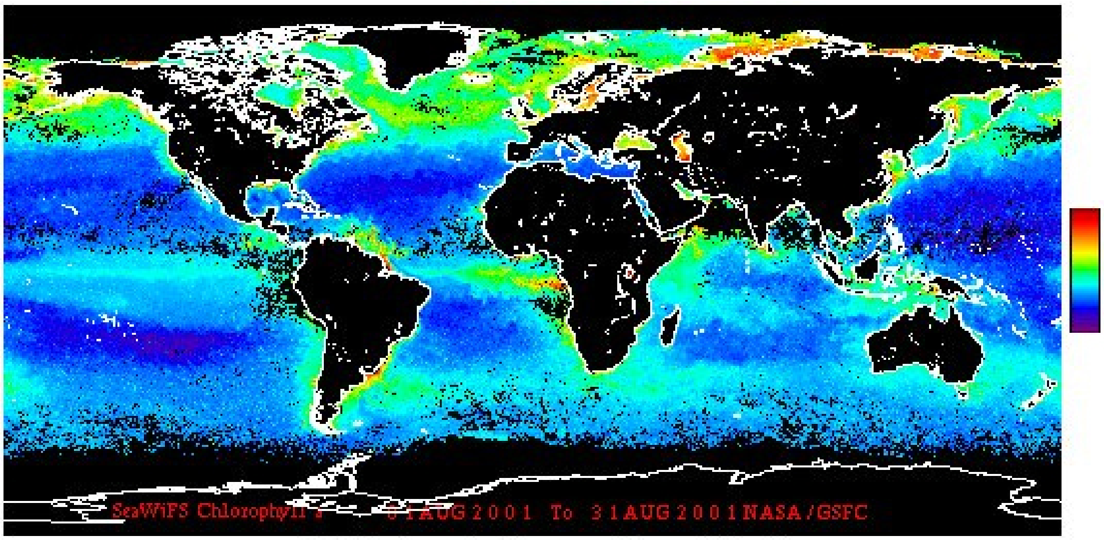
How does *Mnemiopsis* affect ?



Eutrophication levels elevated in the BS during early 1990s...



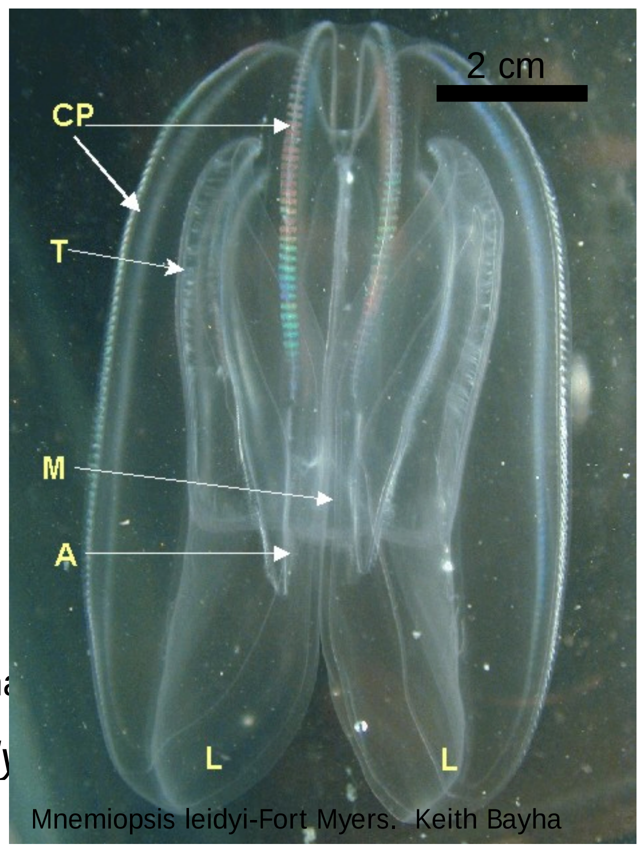
Very high prim. prod. in the Caspian recently (2001 summer)...



Anatomy

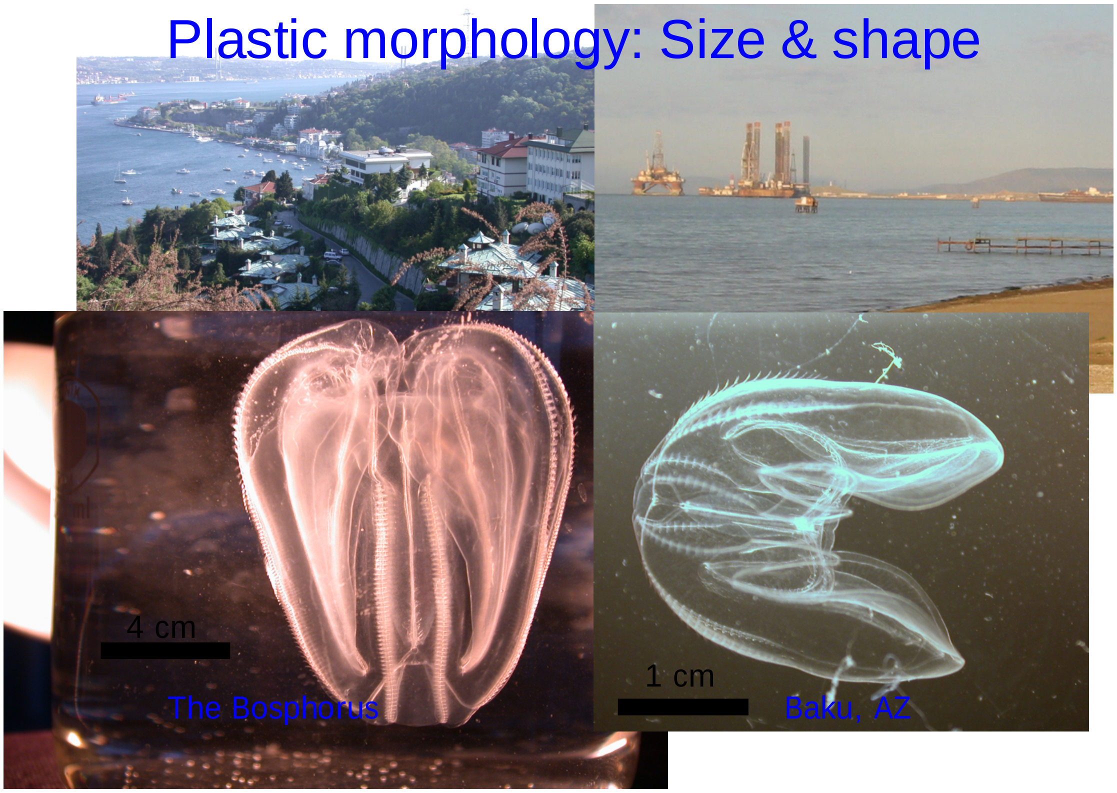
- *Phylum Ctenophora*
- *Class Tentaculata*
- *Order Lobata*
- *Mnemiopsis leidyi*
  - Mid Atlantic
  - 'Smooth'
  - Simple lobular cana
- *Mnemiopsis mccrady*

From Dr T. Moss

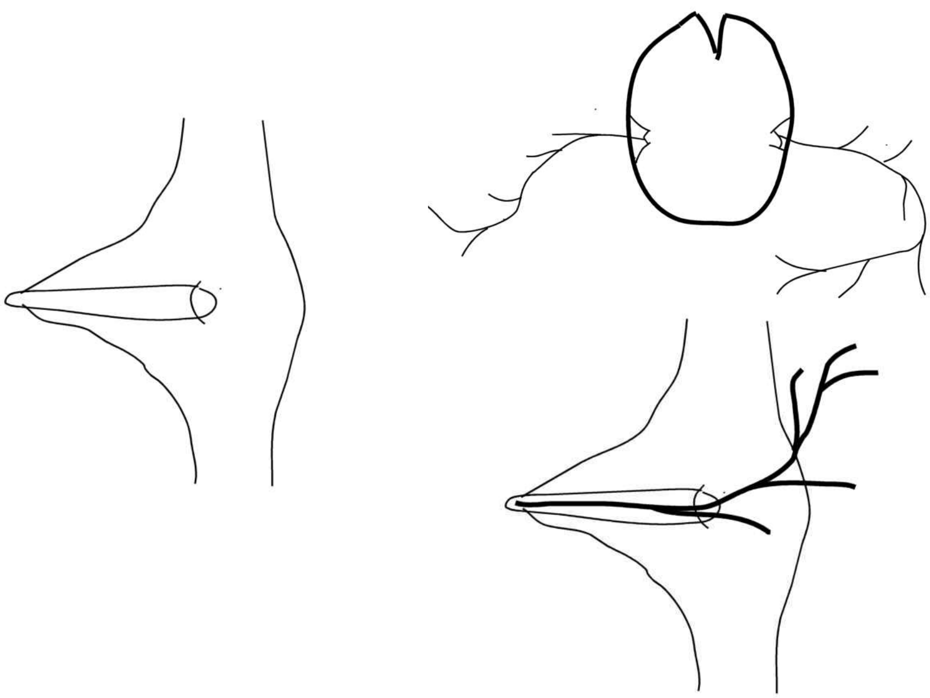


Mnemiopsis leidyi-Fort Myers. Keith Bayha

Plastic morphology: Size & shape



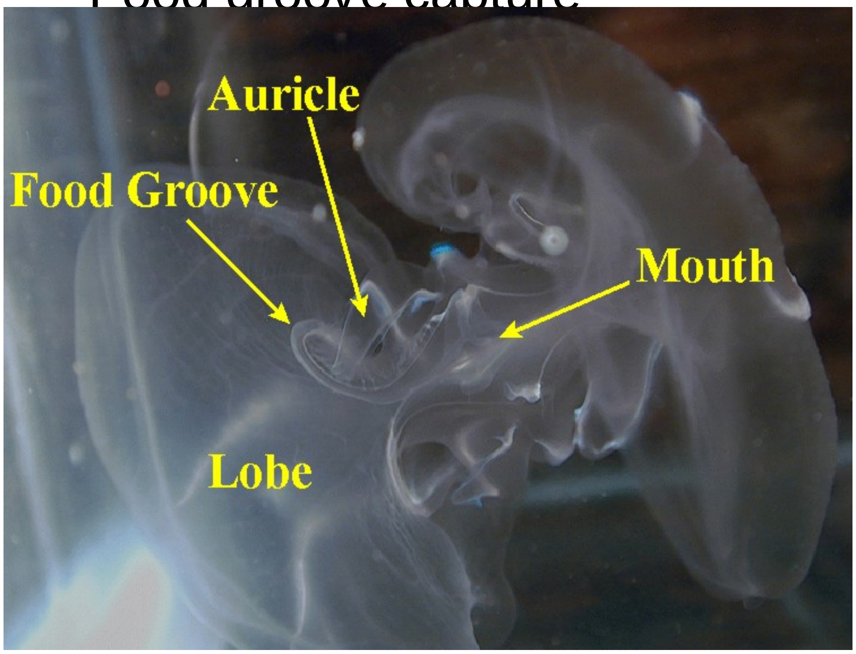
Plastic morphology: Tentacle



Feeding modes in *Mnemiopsis*

(from Dr T. Moss)

- Lobe inner surface capture
- Food groove capture

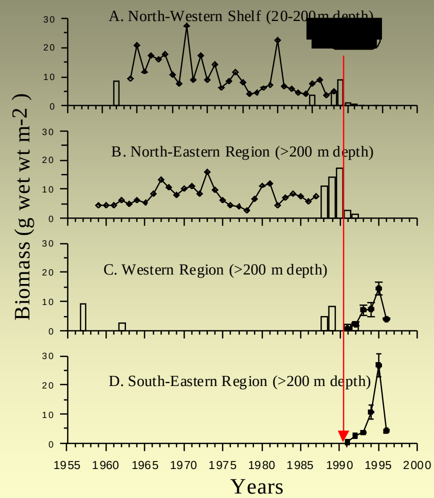


The impact of *M. leidyi* on zooplankton is well documented for the Black Sea both using laboratory and field studies (Finenko & Romanova 2000, Shushkina et al 2000, etc...).

Table 6. Grazing of zooplankton biomass by the population of the ctenophore *M. leidyi* in Sevastopol Bay during different seasons

Season	Mean zooplankton concentration (mg of wet weight per m^3)	Percentage of biomass grazed	Ration (% of energy content equivalent)	Minimum food requirements (% of energy content equivalent)
Winter (December–February)	11.72 ± 18.9	0.10 ± 0.08	0.09 ± 0.17	1.32
Spring (March–May)	24.36 ± 22.6	0.05 ± 0.07	0.055 ± 0.09	2.65
Summer (June–August)	100.67 ± 90.77	21.30 ± 15.82	2.72 ± 3.21	6.78
Autumn (September–November)	117.38 ± 79.0	9.00 ± 6.44	3.17 ± 2.49	4.91

Sharp decrease in the fodder zooplankton biomass of the Black Sea (Kovalev et al., 1998d)

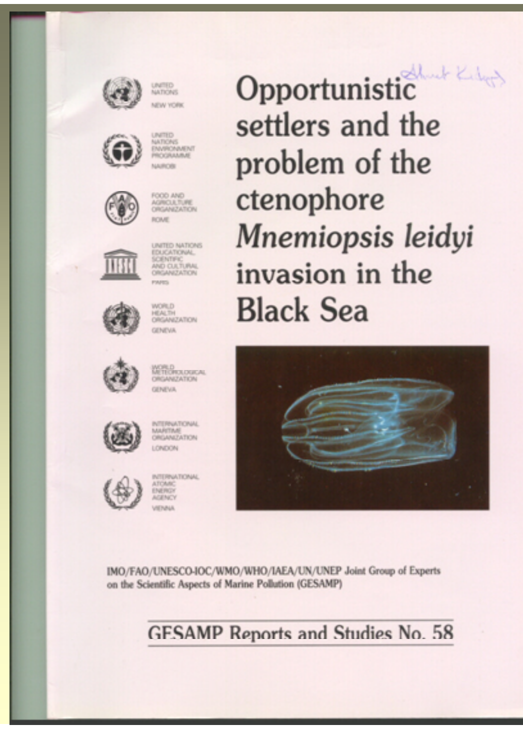


Many more published information on zooplankton decrease during 1988 and 1990….

Eutrophication and *Mnemiopsis* impacts on the Black Sea ecosystem

Kideys 2002, Science 297

In a meeting organised by UNEP in 1994, scientists from several countries discussed strategies for the control of *Mnemiopsis* in the Black Sea


Opportunistic settlers and the problem of the ctenophore *Mnemiopsis leidyi* invasion in the Black Sea

IMO/FAO/UNESCO-IOC/WMO/WHO/IAEA/UN/UNEP Joint Group of Experts on the Scientific Aspects of Marine Pollution (GESAMP)

GESAMP Reports and Studies No. 58

UNEP Working Group had suggested another ctenophore ***Beroe*** to be one of the best candidate for the control of *Mnemiopsis* in the Black Sea

Surprisingly, Beroe appeared in the BS in 1997

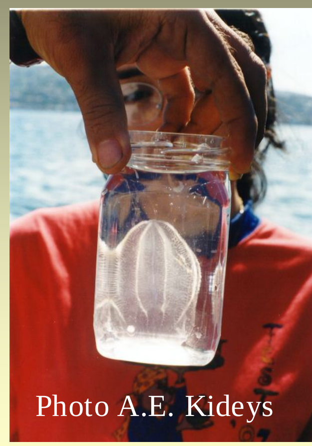


Photo A.E. Kideys

Studies designed to investigate predatory impact of *Beroe* on *Mnemiopsis* in the BS

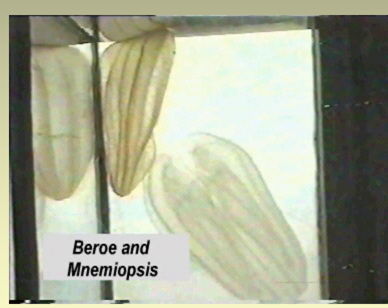


Photo L. Svetlichny

Transport of Caspian sewater to Sinop...

Beroe aquarium...

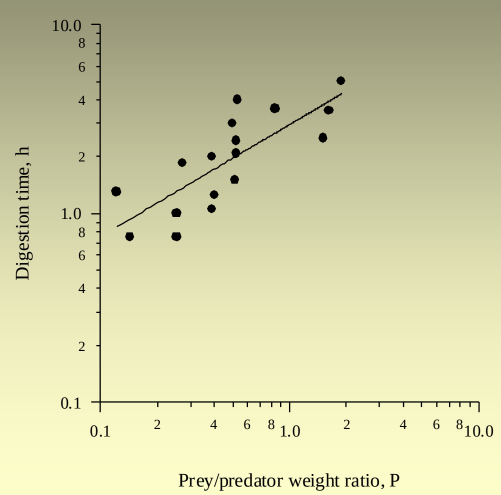
These studies showed that Beroe may control abundance of *Mnemiopsis* population in the Black Sea coastal waters

(Finenko et al., 2000, Hydrobiologia 451)

MOVIE1 MOVIE2

Digestion of *Mnemiopsis by Beroe*

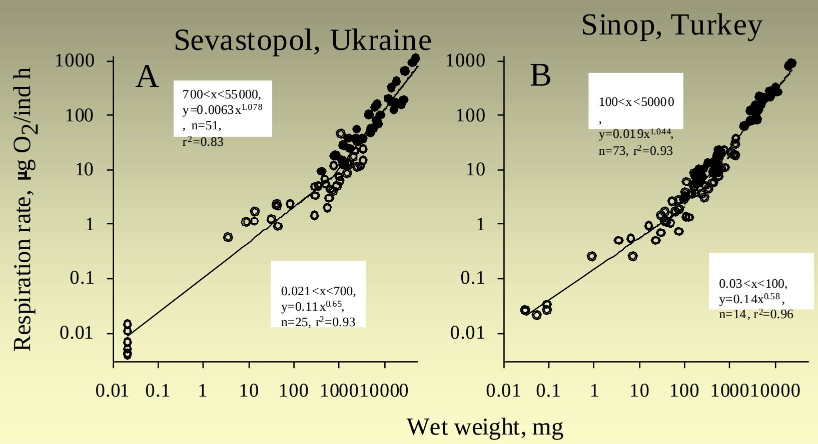
Finenko et al. 2003, J Plank Res



Respiration of *Mnemiopsis by Beroe*

Svetlichny et al. (in press), Marine Biology

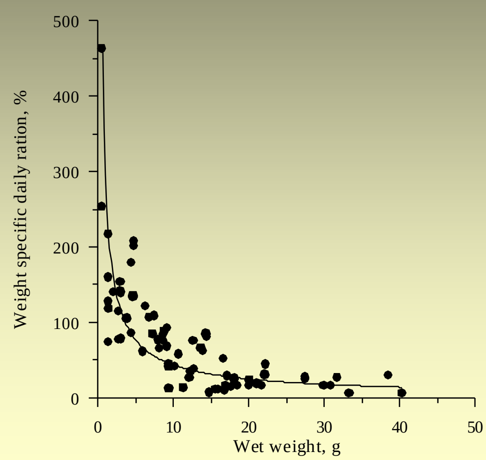
(20 times lower respiration rate for juveniles of 0.4-60 mm; better resistance to low oxygen conditions during transfer..)



polarographic method (O) and by the Winkler technique (●)

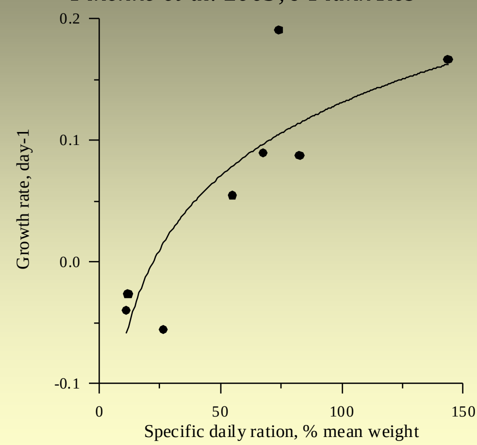
Ration of *Beroe feeding on Mnemiopsis*

Finenko et al. 2003, J Plank Res



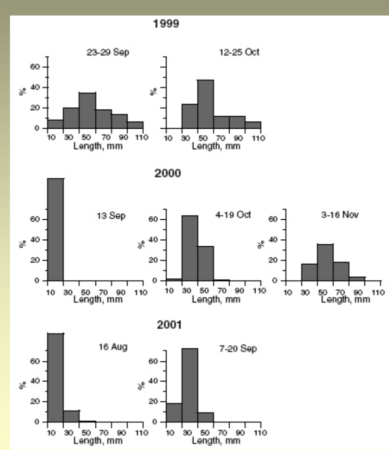
Growth rate of *Beroe feeding on Mnemiopsis*

Finenko et al. 2003, J Plank Res



Growth of *Beroe* at field

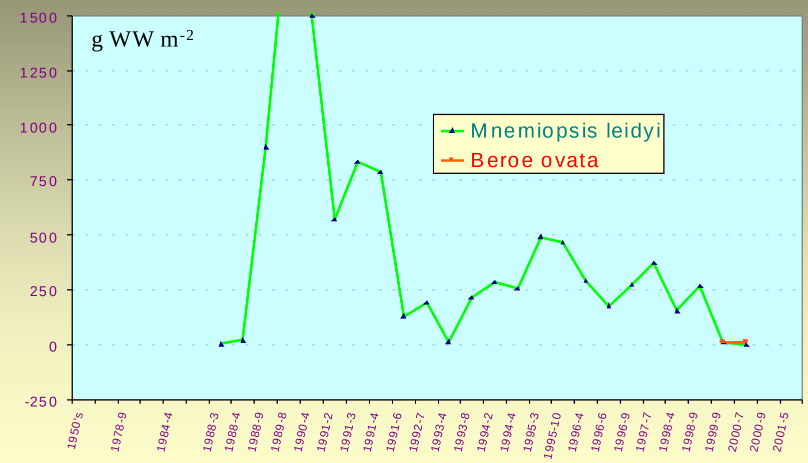
Finenko et al. 2003, J Plank Res



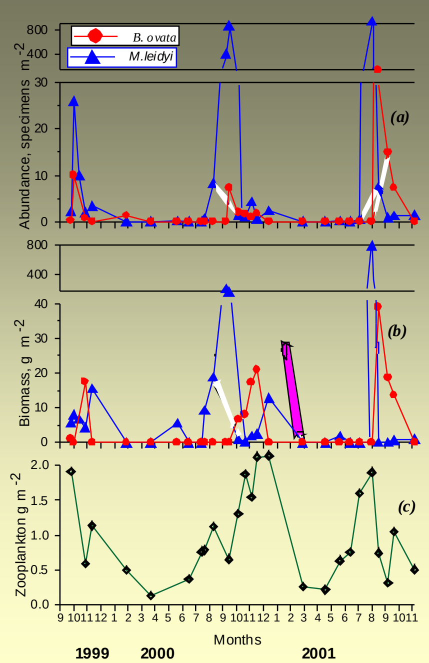
Long-term *Mnemiopsis* biomass

in the southern Black Sea

Kideys et al. 2000, J Mar. Sys 24)

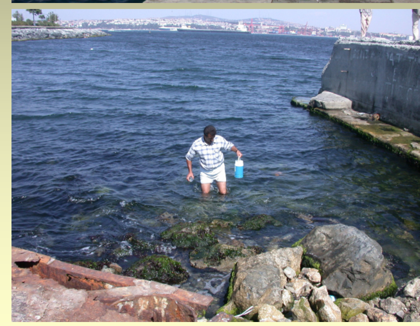


Ctenophore dynamics in Sevastopol Bay (Finenko et al., unpublished data)

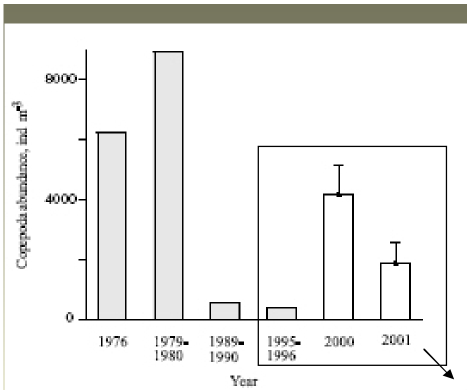


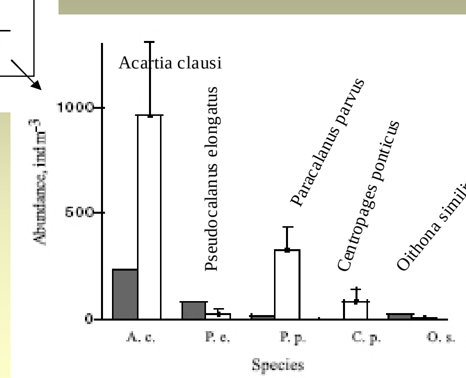
Searching Beroe

(and sometimes Mnemiopsis!)….



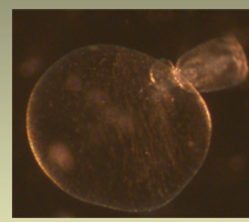
Copepod dynamics in Sevastopol Bay (Gubanova et al. 2001 Finenko et al. 2003)

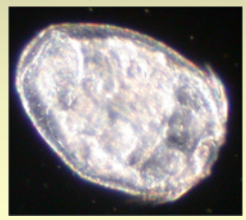




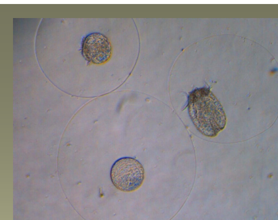
One of the best results ...

- Without exception all the scientists (past and present) working closely with this species know very well that it pose no harm to the other organisms but to ctenophores (Kamyshilov, 1960a,b, Wulf Greve, R. Harbison and others in addition Black Sea scientists).
- We have observed that even the larvae of Beroe feeds on *Mnemiopsis* tissue or larvae...





Reproduction and its rate

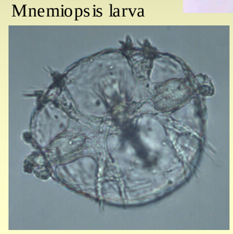


Beroe egg & larva

- During 2001 experiments, only 138 eggs obtained (7 of them hatched to larvae).
- 2003, much much higher numbers....
- Larvae were kept alive over two months in Caspian and Black Sea water...

Photos Zekiye Birinci

Mnemiopsis larva

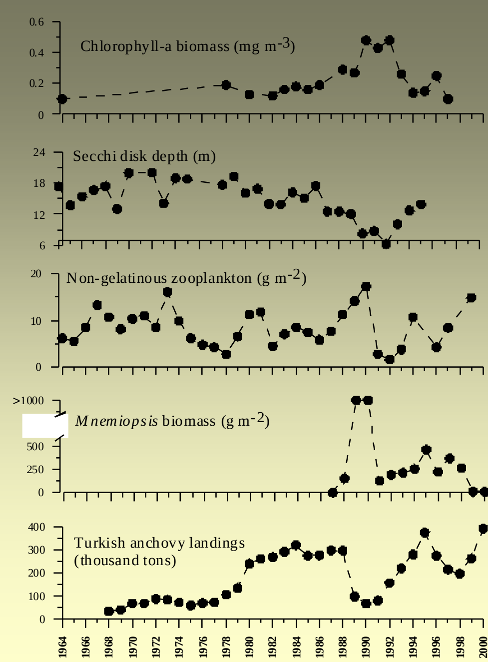


Reproduction rate of *Beroe feeding on Mnemiopsis*

Finenko et al. 2003, J Plank Res

Sample #	Weight, g	Specific daily ration, % WW	Total egg number
1	4.68	108.3±33.0	4007
2	10.7	97.7±43.2	2066
3	17.0	70.1±27.2	4220
4	21.8	20.1±8.4	2525

Recovery of the Black Sea ecosystem over the recent years



Kideys 2002, Science 297

Turkish Anchovy

Results from many new publications present data on recovery of the BS...

- **Lebedeva et al 2003, Oceanology**: After analysing mesozooplankton data concluded that "..Due to the invasion of Beroe ovata feeding on Mnemiopsis leidyi, the structure and concentration of mesoplankton began to be restored..."
- **Shiganova et al. 2003, Biol Bull.:** "..we observed recovering of the main components of the Black Sea pelagic ecosystem-zooplankton and fish as well as their eggs and larvae.."
- **Oguz et al 2003, Vinogradov et al 2003 and others...**

Inter-annual changes in abundance of larval food (<0.5 mm)

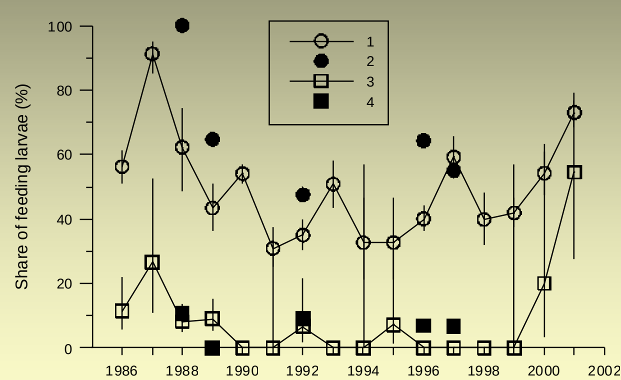
Gordina et al. (in press), J Ichthyology

Period	Nauplii, ind. m^{-3}	Other organisms	Author
1960-69	1074000	13210	Greze et al., 1971
1988	47500	–	Ostrovskaya et al., 1993
1989	21500	2092	The same and our data
1990	29670	708	Our data
1992-93	11670	–	Gruzov et al., 1994
1998	10700	200	Our data
1999	86000	2250	Our data
2000	59670	2353	Our data
2001	382330	4870	Our data

Percentage of larvae with food in intestine

Gordina et al. (in press), J Ichthyology

Larvae of *Gobiidae* and *Blenniidae* in Ukrainian (1) and Turkish (2) waters; larvae of *Engraulis encrasicolus* in Ukrainian (3) and Turkish (4) waters.



Introduction to the Caspian was warned at the mid 1990s..

- GESAMP Report
- Prof H. Dumont in his paper published in Nature

Start of the most important ever anthropogenic problem of the Caspian Sea ecosystem

(or probably the biggest impact from an invasive species in the world seas...)

Unfortunately this invasion occurred by 1997,
First reported by Ivanov et al. in 2000.

Therefore, the sharp decrease observed in fishery is actually "**the tip of the iceberg**" within the Caspian ecosystem

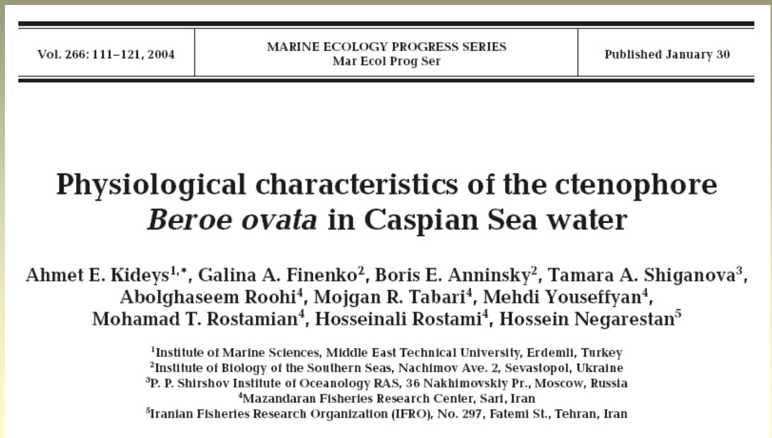
Excerpt from 2001 report by Kideys et al.!
(and further decreases in kilka catch had been foreseen in that report!

We expected that *Mnemiopsis* would damage...

- Not only pelagic fishes and fishermen!
- Entire ecosystem by breaking food web structure
- Perhaps biodiversity including endemic spp
- Top predators such as endemic Caspian Seal (pregnancy) and Beluga sturgeon

Excerpt from 2001!

Conclusion..



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Physiological characteristics of the ctenophore *Beroe ovata* in Caspian Sea water

Ahmet E. Kideys[1,*], Galina A. Finenko[2], Boris E. Anninsky[2], Tamara A. Shiganova[3], Abolghaseem Roohi[4], Mojgan R. Tabari[4], Mehdi Youseffyan[4], Mohamad T. Rostamian[4], Hosseinali Rostami[4], Hossein Negarestan[5]

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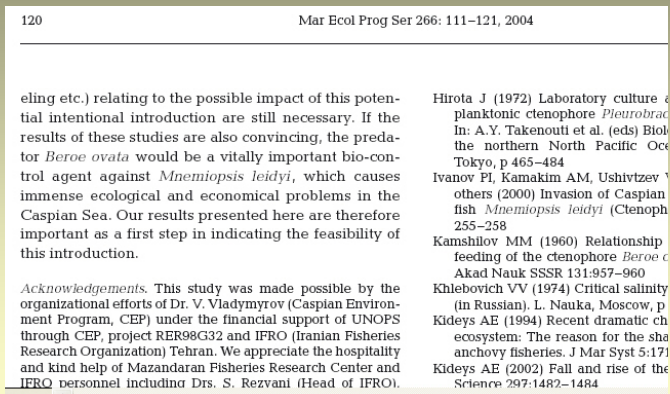
[2]Institute of Biology of the Southern Seas, Nachimov Ave. 2, Sevastopol, Ukraine
[3]P. P. Shirshov Institute of Oceanology RAS, 36 Nakhimovskiy Pr., Moscow, Russia
[4]Mazandaran Fisheries Research Center, Sari, Iran
[5]Iranian Fisheries Research Organization (IFRO), No. 297, Fatemi St., Tehran, Iran

Conclusion..



eling etc.) relating to the possible impact of this potential intentional introduction are still necessary. If the results of these studies are also convincing, the predator *Beroe ovata* would be a vitally important bio-control agent against *Mnemiopsis leidyi*, which causes immense ecological and economical problems in the Caspian Sea. Our results presented here are therefore important as a first step in indicating the feasibility of this introduction.

Acknowledgements. This study was made possible by the organizational efforts of Dr. V. Vladymyrov (Caspian Environment Program, CEP) under the financial support of UNOPS through CEP, project RER98G32 and IFRO (Iranian Fisheries Research Organization) Tehran. We appreciate the hospitality and kind help of Mazandaran Fisheries Research Center and IFRO personnel including Drs. S. Rezvani (Head of IFRO),

Hirota J (1972) Laboratory culture a
planktonic ctenophore *Pleurobrac*
In: A.Y. Takenouti et al. (eds) Biol
the northern North Pacific Oce
Tokyo, p 465–484
Ivanov PI, Kamakim AM, Ushivtzev V
others (2000) Invasion of Caspian
fish *Mnemiopsis leidyi* (Ctenoph
255–258
Kamshilov MM (1960) Relationship
feeding of the ctenophore *Beroe c*
Akad Nauk SSSR 131:957–960
Khlebovich VV (1974) Critical salinity
(in Russian). L. Nauka, Moscow, p
Kideys AE (1994) Recent dramatic ch
ecosystem: The reason for the sha
anchovy fisheries. J Mar Syst 5:171
Kideys AE (2002) Fall and rise of the
Science 297:1482–1484

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Iranian Fisheries Research Center:
S. Rezvani, H. Rustemi, H. Negarestan, Hamid Alizadeh, many others..
for their support of Caspian related studies, and for the invitation.

DISTIBUTION AND ABUNDANCE OF MNEMIOPSIS LEIDYI IN THE EASTERN IRANIAN COASTS OF THE CASPIAN SEA

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Institute of Marine Science, Middle East Technical University, Erdmil, Turkey,
(**kideys@ims.metu.edu.tr**)

Abstract

The alien ctenophore *Mnemiopsis leidyi* which was transported from the Black Sea into Caspian at the end of 1990s has been negatively affecting ecosystem in this new environment. In this study, spatial and temporal distribution of *M.leidyi* were studied from a total of 12 stations located along three transects (Nowshar,Babolsar and Amirabad) in the eastern Iranian coasts of the Caspian Sea (Mazandaran province) during July 2001 to septamber 2002.
M. leidyi achived maximum biomass (1024.5 g m^2 / 102.5 g m^3) in Septamber 2001. Minimum biomass (1.5 g m^2 / 0.27 g m^3) of the ctenophore were measured in March 2002. The highest biomass was at the station with 10 m bottom depth (570.7 g m^2) in autumn and lowest biomass (75.9 g m^2) was obtained at a station with a 50 m bottom depth in spring. The highest average biomass 641.2 g m^2 were measured in Amirabad region, and the lowest biomass (207.5 g m^2) observed in Nowshar region. The young specimens (<5 mm) contributed about 90 % to the total abundance of the population. The maximum length was 51-55 mm which was measured in August. The factors affecting the distribution of *M. leidyi* in the study area were discussed.

Key words : Caspian Sea, Iranian coasts, Distribution, *Mnemiopsis leidyi*

INTRODUCTION

In the 1980s, the introduction of a new species (a lobate ctenophore, *Mnemiopsis leidyi* or *M.maccradyi*) into the Black Sea radically affected the whole ecosystem (Vinogradov 1989; Kideys 1994). This species had a negative impact on the most dominant fish of the Black Sea, the anchovy *Engralius encrasicolus*, though competion for the edible zooplankton as well as consumption anchovy eggs and larvae in the Black Sea. The mass occurrence of *Mnmiopsis* was one of the most important reasons for the sharp decrease of anchovy and other pelagic fish stocks in the Black Sea (Kideys 1994).
Meantime possibility of its introduction into other neighbouring sensitive ecosystems, notably the Caspian Sea, has been mentioned (Dumont 1995; **GESAMP** 1997). And, as expected, this ctenophore reported from the Caspian Sea by November 1999 (Ivanov et al.2000).

Ivanov et al. (2000) suggested that this ctenophore were transported with ballast water taken aboard in the Black Sea or the Sea Azov (where *Mnemiopsis* occurs in warm months) and released after ballast-loaded ships passed through the Volga Don Canal and the shallow freshwater North Caspian Sea, into the saltier Central or South Caspian.

Due to demage observed in the Black Sea, there has been a fast response over the presence of *Mnemiopsis* in the Caspian Sea, since *Mnemiopsis* is a voracious predator on zooplankton, both abundant small pelagic fishes feeding on zooplankton as well as large predators feeding on these fishes such as white sturgeon (*Huso huso*) and endomic Caspian Seal (*phoca caspica*) would be under significant threat in the Caspian Sea (Kideys et al.2001).

In this study temporal and spatial distribution of Mnemiopsis were studied in order to gather information on the levels and ecology of this ctenophore; for evaluation of its impact on the coastal pelagic communities along the southern Caspian Sea.

Materials and METHODS

In this study, spatial and temporal distributions of *M.leidyi* were studied along three transects (Nowshar, Babolsar and Amirabad) in the eastern Iranian coasts of the Caspian Sea (Mazandaran province) during July 2001 to September 2002 . Each transect had four stations located at 5, 10, 20 and 50 m bottom depth contours. Because sampling of each transect is planned to be completed during the day, a speed boat was used for the sampling at sea.
Temperature and salinity of the seawater at depth 5, 10, 20 and 50 m were measured by using an inverted thermometer *in situ* and by using a digital salinometer, respectively. At every station, the water clarity was also measured by using a Secchi Disk. *Mnemiopsis leidyi* was sampled using a 500 micron mesh sized METU net (diameter 50 cm with large bucket suitable for *Mnemiopsis*). Samples was obtained via vertical towing from the bottom to the surface for all stations . At the deepest station, because of existence of thermocline, 20 m sampled for salinity and temprature : (fig 1).

Figure 1. Sampling locations in the Caspian Sea

At the end of each tow, the net was washed from the exterior, and the cod end was passed into a container immediately to enumerate ctenophores by naked eye. The density (both as per m^2 and m^3) of *Mnemiopsis leidyi* was calculated from the diameter of the net and the tow depth.

The ctenophores were sorted in length groups of 0-5 mm, 6-10 mm, 11-15 mm and so on, for size measurements. A total of 144 Samples were measured and grouped in this way. Individual weighing of these animals was not practical at sea. Weights of these animals were therefore calculated from size measurements (269 individuals) using a conversion formula which was obtained individual length (using a ruler for the lobed length) and weight measurements (using a digital balance with a sensitivity of 0.001 g) in July 2001. Length groups were thus converted to weight by using the equation obtained.

Results

The changes in the average temperature of the study area during the course of period study are shown in Fig. 3. It oscillated between 30.2 oC in August to 7.5 oC in March. Within the same period, the salinity values ranged from 12.0 ppt (10 M) to 12.8 ppt (50 M) with an average value of 12.5 (of which standard deviation being 0.7) in the area. The Secchi disk depth values fluctuated between 0.2 and 10 m with an average of 3.0 m during the study period in the southeastern Caspian Sea.

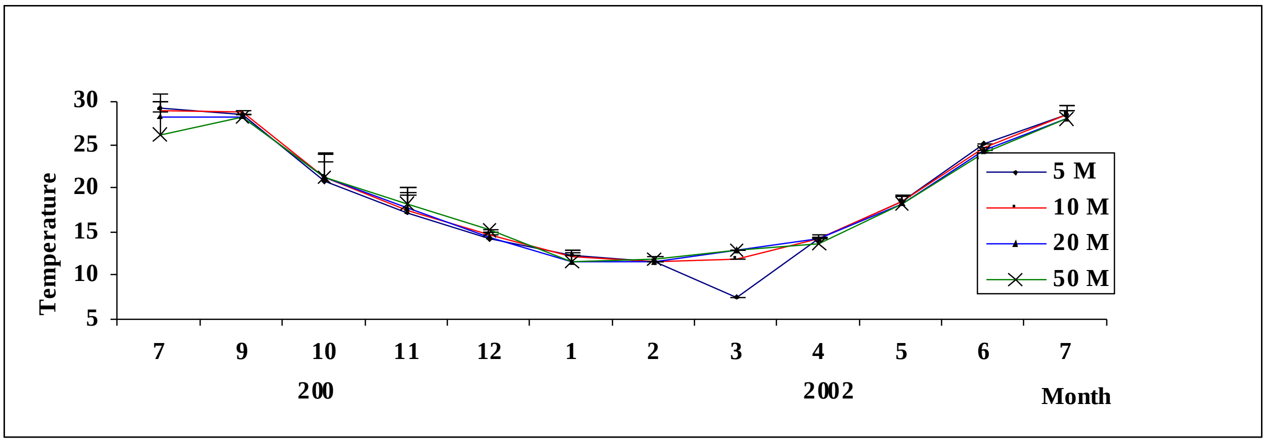

figure 2: Temperature(^{oC})changes at different depths of the Southeastern Caspian Sea .

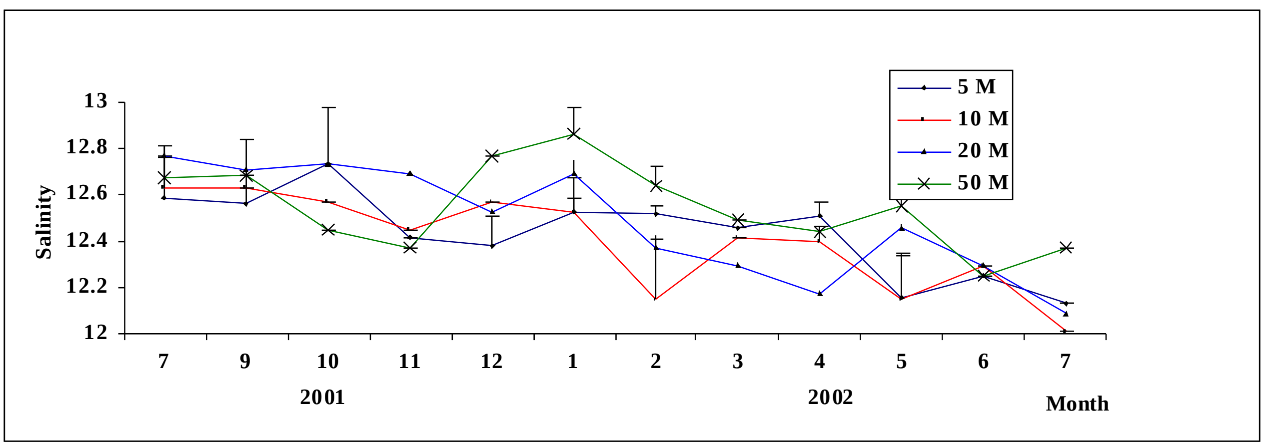

figure 3: Salinity (ppt) changes at different depths of the Southeastern Caspian Sea

The length-weight equation of *Mnemiopsis leidyi* which is used in biomass calculation was found to be: Weight (g) = 0.0013 Length (mm)^2.33 , R^2 = 0.96, n = 269. The power fit of relationship between the total length and wet weight is shown in Fig. 4.

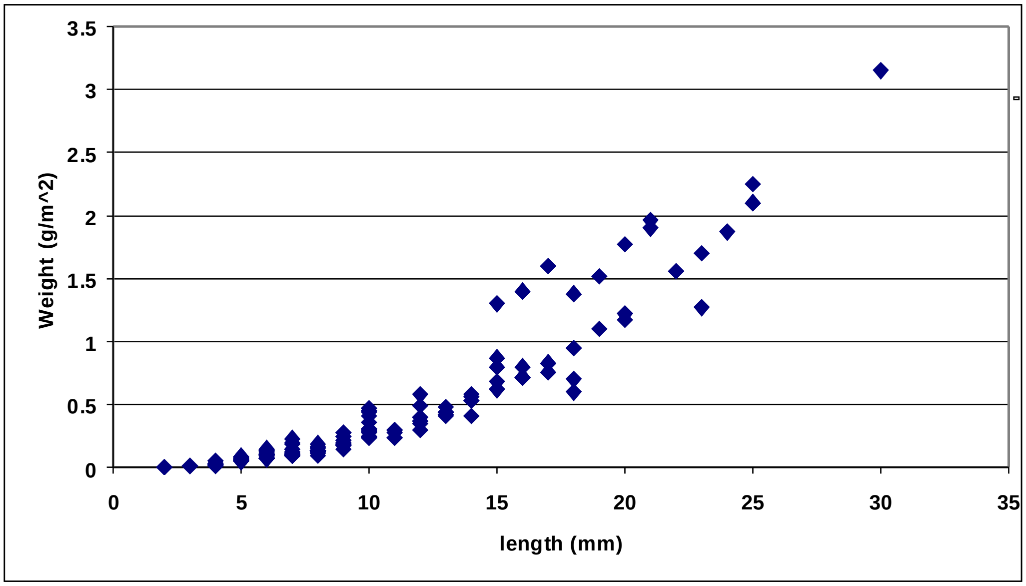

Figure 4. The relationship between the total length and weight of *M. leidyi*

The biomass of *M.leidyi* also fluactuated widely from 12.2 g m^2 m in March 2001 up to 871.1 g m^2 in September 2001 in the southeatern part of the Caspian Sea (Fig. 3). Generally biomass values were low during cold months (i.e. November-March) and high in warmer months. Population increased quite suddenly between August and October 2001 and again in 2002 when it reached to 700.7 g m^2 and sharply decreased by the September 2002. Comparing with the same months of 2001, the biomass values in 2002 were almost the same. The highest biomass values were also observed in September 2001 and August 2002.

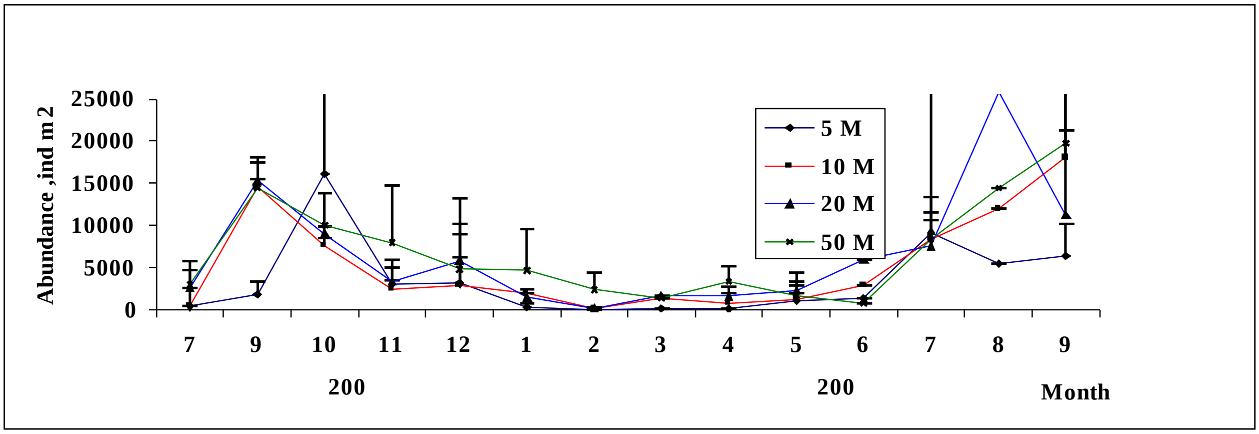

Figure 5. Changes in *Mnemiopsis leidyi* biomass of Mazandaran during sampling period (squares biomass and triangles temperature and lozenges abundance values)

Vertical distribution of *M .leidyi* in difference depth is shown in Figure 6. Except in winter , the highest biomass of *M.leidyi* was occurred in 20 m depth.

The lowest mean biomass of this Ctenephore was observed in 5 m depth in all seasons. From this figure it is observed that generally. The mean biomass of *M.leidyi* was higher in Autumn in compartion to other seasons, and in winter low biomass of ctenphore was encoutend .

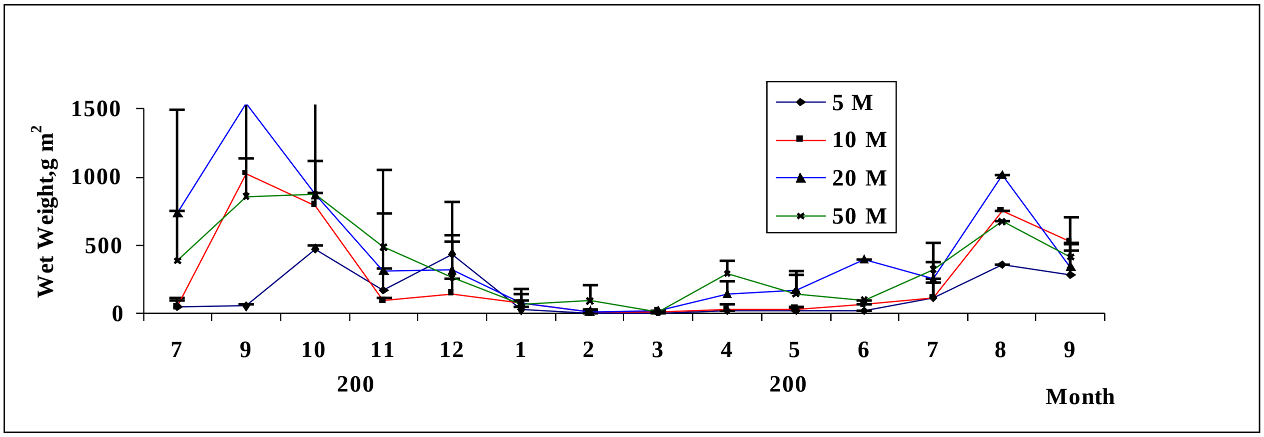

Figure 6. Coastal *Mnemiopsis* Distribution

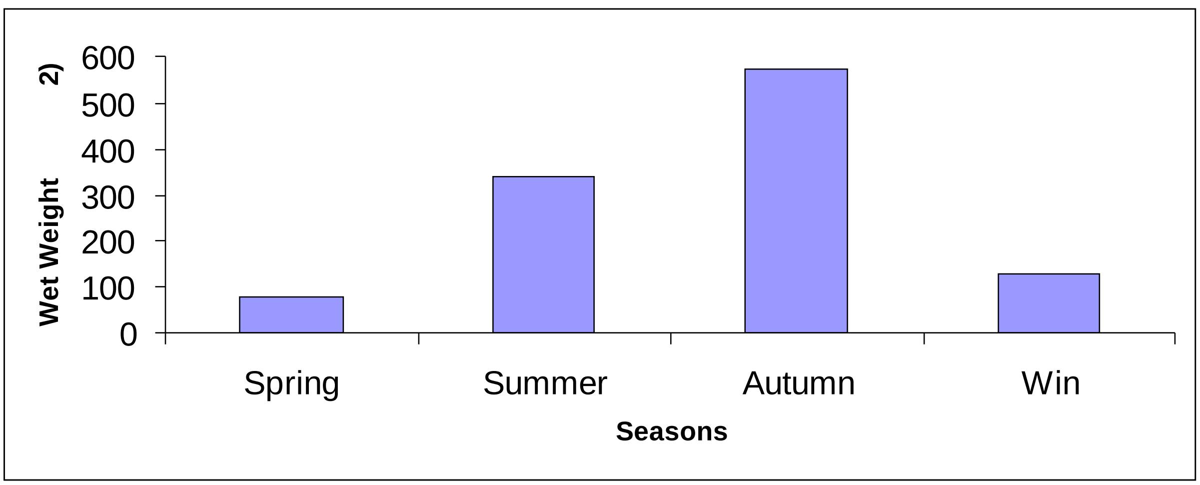

Figure 7: Seasonal changes of *M.leidyi* in the Southeastern of Caspian Sea.

Spatial biomass distribution of *M.leidyi* in 3 transect of Noshahr , Babolsar and Amirabad is presented in figure 10. The high average biomass observed in Amirabad with value of 402.2 g m^2 / 29.4 g m^3 and the lowest was measured in Noshahr with record of 248.6g m^2 / 16.1 g m^3.

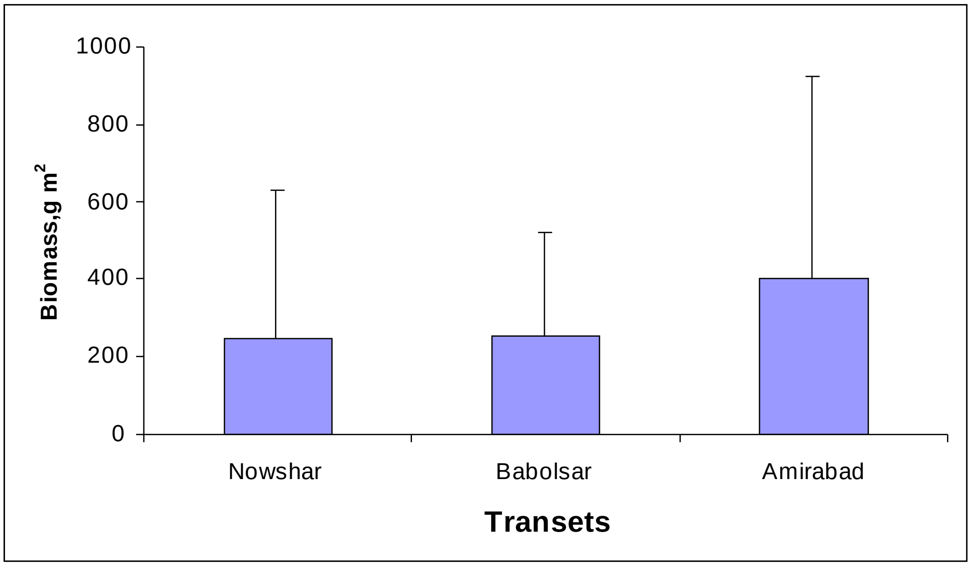

Figure 8. Average weigh of *Mnemiopsis* among transects

Temperature profiles in different season sampled during July 01 – sep 02 cruises are presented in figure 9, as seen in this figure, there are no termoclain in winter and spring . The termoclain observed in summer and in somehow in autumn in 30 m and 40 m depth respectively.

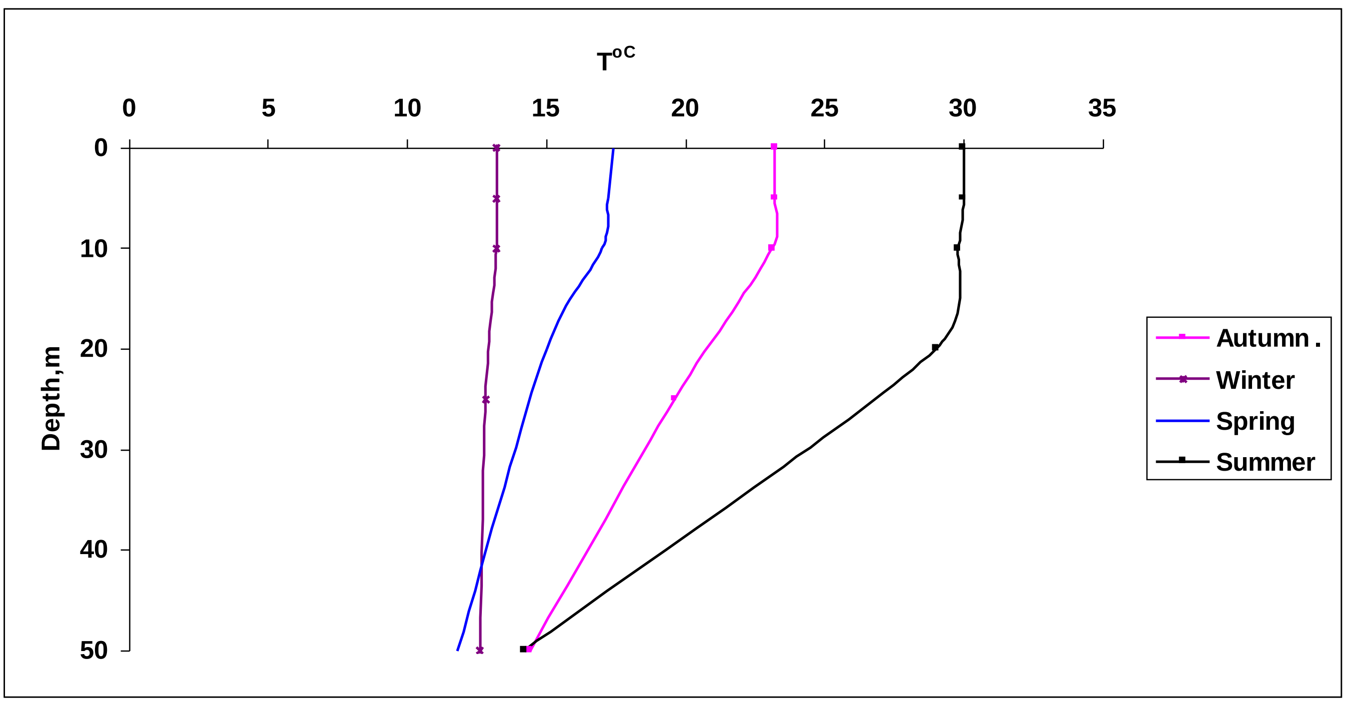

Figure 9. Seasonal temperatures (^{o}C) at the deepest Amirabad station (Am4)

Figure 10 shows monthly percentage of size frequency of *M.leidyi* of Mazandaran coastal water. Small ctenophore (<5 mm) was dominate in all depth (%90).

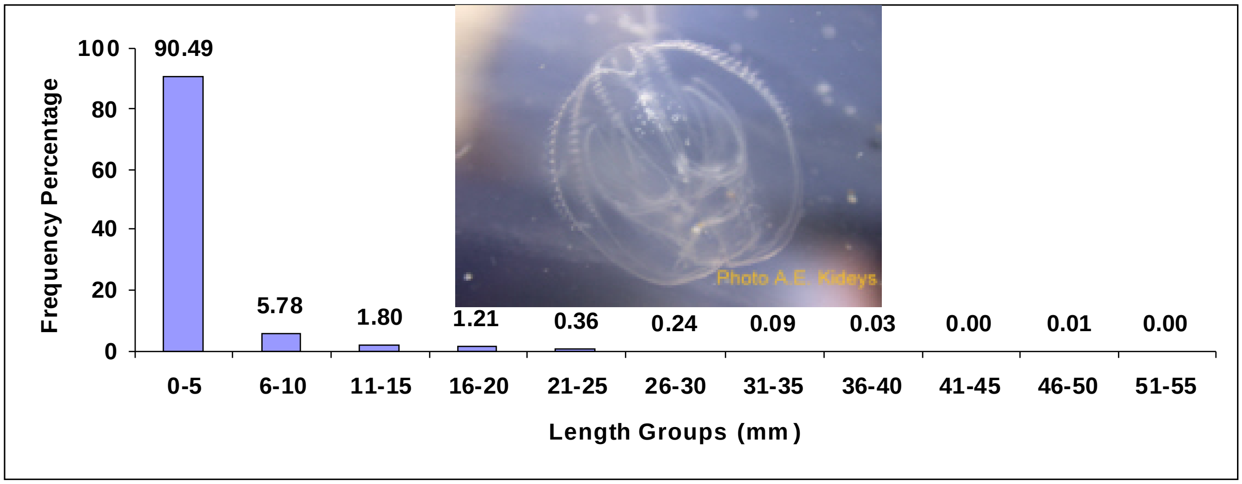

Figure 10. Monthly percentage size frequencies of *Mnemiopsis* off Mazandaran

Monthly changes in the mean size of *M.leidyi* off Mazandaran coastal water is presented in figure 11. As seen there are considance in mean length and mean wet weight fluctuation in different month of 2002 and 2002.

The minimum mean wet weight and mean length was recorded in Dec 01 and Jan 02. These figures increased after Jan 02 and reached to its Maximum in May 02 .

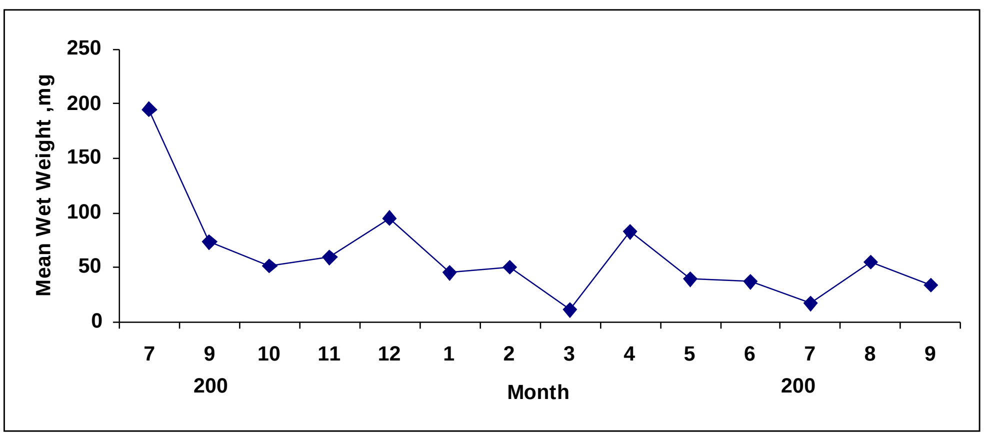

Figure 11. Monthly changes in the mean size *Mnemiopsis* of Mazandaran

Discussion

The variation of the biomass of *M.leidyi* in the Caspian Sea depends on temperature over the month of the year, as was reported for the Black Sea (Shiganova 1998).
Reproduction of this species in the Coastal water of the southern Caspian Sea starts during early of July (as observed in 2001 and 2002) and reach its maximum during August and continue to October which increased the population of *M.leidyi* to its peak in October. It was reported that reproduction of this Ctenophore in some inshore water of the Black Sea, starts during the second half of July or August (which have some delay in comparison to the Caspian Sea due to water temperature after October due to decreasing of the temperature *M.leidyi* biomass decreased), this results is concidence with the Black Sea (shiganova 1998).

Multu (1999) mentioned that *M.leidyi* distribute over a narrow ranges of depth above the thermocline. Also Vinogradov *et al* . , (1998) first observed similar vertical distibution in last September 1987 and this pattern has seen repeatedly (Vinogradov 1990; Bogdanova and Konsoulov 1993).

Based on our results small *M.leidyi* (<5 mm) comprised above 90 % of the total population during the year, and mean length of individuals increased from January to May, but in summer the mean length of individual decrease. In the Black Sea young individuals (<10 m) were abundant in summer due to reproduction, food availability probably plays a more important role in the reproduction of *M.leidyi* (Mutlu 1999). Vinogradov *et al* (1992) and Volovik *et al*. (1993) found similar seasonal changes in size composition of *M.leidyi* , Summer reprodution may be associated with the combined effects of food and higher temperature (>23 0C) on spawing of *M.leidyi*.

Ctenophore body size in the Black Sea in spring increase with temperature, between February and June, somatice growth of population increase (purcell 2001).

The biomass of *M.leidyi* differed among the regions and as seen the Amirabad has much more biomass compared with other area. It seems that nutrient input of the Tajan river and slow slope was responsible for this situation. The influence of rivers such as the Danube, increased input of allochtonous nutrients (vinogradov *et al*. 1992). This increase in food supply may cause population explosion of *M.leidyi* in the Black Sea (Mutlu, 1999).

Conclusions

There is a strong seasonality in Mnemiopsis biomass during the course of the year.
Maximum biomass value of 1024.5 g m^2 / 102.5 g m^3 , though lower than that peak values in the Black Sea in the late 1980s, has already very negatively affected the entire Caspian ecosystem. This is mainly caused by the enormous abundance made up by small invividuals.
The biomass values are almost the same or somehow getting higher in 2002 indicating that it did not reach peak levels in the Caspian Sea yet.
This implies that fishery and overall ecosystem may suffer further losses.

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Reference

D.P.Bogdanova, A.S.Konsoulov. On the distribution of the new Ctenophore species Mnemia Mccradyi in the Black Sea a long the Bulgerian coastlian in the summer of 1990 .Dok B lg Akad, 46, pp.71-77, (1993).

H.J.Dumont. Ecocide in the Caspian. Nathre, 377, pp. 673-673, (1995).

GESAMP (IMO/FAO/UNESCO-IOC/WMO/WHO/IAEA/UN/UNEP Joint Group of Experts on the Scintific Aspects of Marine Environmental Protection) Opportunistic settlers and the problem of the ctenophore *Mnemiopsis leidyi* invation in the Black Sea. Rep.Stud. GESAMP, 58 :pp. 84, (1997).

P.I.Ivanov , A.M. Kamakim, V.B. Ushivtzev, T.A. Shiganova, O. Zhukova, N. Aladin, S.I Wilson, G.R. Harbison, H.J. Dumont. Invasion of Caspian Sea by the come jellyfish *Mnemiopsis leidyi* (Ctenophora),Biological Invasion, 2 , pp. 255-258, (2000)

A.E.Kideys. Recent dramatic changes in the Black Sea ecosystem: The reason for the sharp decrease in Turkish anchovy fisheries, Journal of Marine System ,5, pp.171-181, (1994)

A.E.Kideys, S. Ghasemi, D. Ganinejad, A. Roohi and S. Bagheri. Strategy for combating *Mnemiopsis* in the Caspian Waters of Iran, Final Report, (2001).

E.Mutlu, Distribution and abundance of Ctenophores and their zooplankton food in the Black Sea.Marine Biology ,135 ,603-613, (1999).

J.E.Purcell, T.A.Shiganova, M.B. Decker & E.D.Houde, Hydrobiologia,451, pp 145-176, (2001).

T.A.Shiganova.Invation of the Black Sea by the Ctenophore *Mnemiopsis leidyi* and recent changes in pelagic Community structure, Fisheries oceanography, 7:3/4, 305-310, (1998)

M. E. Vinogradov, E.A. Shushkina, E.I. Musaeva , P.Y. Sorokin. Ctenophore *Mnemiopsis leidyi* (A. Agassiz) (Ctenophora ; Lobata) a new settler in Black Sea. ,Oceanology, 29, 293-298, (1998)

M. E. Vinogradov. Investigation of the pelagic ecosistem of the Black Sea (44 cruise of the R/V Dimitry Mendeleyev), 4July –17 September 1989,Oceanology, 30, 254-256, (1990).

M. E. Vinogradov, E.A. Shushkina. Temporal changes in community struture in the open Sae. Oceanology, 32, pp..485-491, (1992).

S.P.Volovik, Z.A.Myrzoyan. G.s.Volovik. *Mnmiopsis leidyi* in the Azov Sea: Biology, Population dynamics, impact to the ecosystem and fisheries, ICES. CM, 69, pp.12, (1993).

Beroe ovata Egg production, hatching success and growth rate of larva in the Caspian and Black Sea waters.

(Results of experiments carried out in Sinop, September 2003)

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Introduction

The Black Sea is an impressive example of damaging impact of invasive species upon the ecosystem on the whole and on the planktonic community in particular. The introduction and subsequent explosion of the ctenophore *M. leidyi* in the late 80's resulted in dramatic decrease not only the abundance of some species but also temporary poverty in biodiversity of zooplankton community (Vinogradov et al., 1992; Kovalev et al., 1998). *M. leidyi* competition with planktonic fish for zooplankton as a food brought to remarkable decline in the fish stock registered those years (Kideys, 1994).

Appearance of new alien ctenophore *Beroe ovata* in the Black Sea in the late 90-s appeared to be very effective in controlling high levels of *M. leidyi* and reduced its population explosion to the events of short duration. If in the years of *M. leidyi* bloom it was regularly found in abundance in plankton during 7 months, later after *B .ovata* introduction *M. leidyi* predominated only for 1-2 months a year (Finenko et al., 2001, 2003). With the decimation of

M. leidyi levels, zooplankton biomass and hence fish recruitment has been restored. No species appear to have been lost from the Black Sea fauna during the recovery process even if, at the height of the *M. leidyi* outbreak, many fell to levels so low as to make their observation impossible.

A warning that *Mnemiopsis leidyi* might also invade the Caspian Sea had been voiced as early as 1995 (Dumont 1995, GESAMP 1997). Unfortunately, at the end of the 1990s the invasion of *M. leidyi* in the Caspian Sea was reported (Ivanov et al. 2000).

Investigations in the Caspian Sea in 2000-2002 showed that it was found everywhere including the Northern Caspian where salinity was higher than 4ppt. There was an increasing trend in the abundance of *M. leidyi* in 2001 compared to 2000. In August 2001 the average and maximum biomasses of *M.leidyi* over the entire Middle and Southern Caspian Sea were as high as 120 and 351 g wet weight m^{-2} respectively against mean value of 60 g m^{-2} in summer 2000 (Shiganova et al., 2001, Kideys and Moghin, 2003, Shiganova et al., 2003).

Non-gelatinous mesozooplankton in the northern Caspian showed a decrease of density in 5.3 times and biomass in 6 times in October 2001 comparing with July when *Mnemiopsis* was absent; the most considerable was decline in density of Copepoda.The drastic decrease of zooplankton biomass appeared to be in the Southern Caspian where the highest biomasses values of *M. ledyi* have been observed (Shiganova et al., 2001).

Impact of *Mnemiopsis* on the Caspian Sea ecosystem can be much worse than in the Black Sea due to great sensitivity of closed basin to any impact**.** Since *Mnemiopsis* is a voracious predator and a competitor with planktivorous fish for zooplankton, catches of the main of them, particularly kilka (*Clupeonella spp*) for some riparian countries are already reported to have decreased (Kideys et al. 2001a, b). Within two years (2000-2001) almost a 50 % decrease in the kilka catches of Iranian fishermen has occurred, with a minimum of 15 million US dollars economic loss (Kideys, Moghim, 2003).

During the First International Workshop on "The Invasion of the Caspian Sea by the Comb Jelly *Mnemiopsis leidyi*– Problems, Perspectives, Needs for Action", organized by the Caspian Ecological Program (CEP) in April 2001, it was concluded that *Beroe ovata* is the best candidate to control *Mnemiopsis* population in the Caspian Sea.

Regional *M. leidyi* Advisory Group has been organized and has developed an *M. leidyi* Control Strategy. More specifically the Advisory Group has agreed to start studies to introduce *Beroe* as a predator for *M. leidyi*.

In framework of this program special experimental studies were performed in Mazandaran Fisheries Research Center of Iran in 2001- 2002 and these results confirmed that *B.ovata* could live and grow intensively in the Caspian Sea water with a salinity of around 12 ppt (Kideys et al. 2004). However the reproduction was very low and only a few larvae could hatch, which died in 1-2 days.

More detail and intensive study of reproduction of *B. ovata* in the Caspian Sea water should have been carried out to clarify the possibility and feasibility of *Beroe* introduction in the Caspian Sea.

Therefore, in September 2003 such experiments were conducted in Sinop, southern Black Sea by a team of scientists from Russia, Ukraine, Turkey and Iran. These experiments were aimed to study 1) reproduction characteristics (proportion of animals spawned, size of egg clutch, hatching success) of *Beroe ovata* transferred to the Caspian Sea water with and without adaptation and 2) the growth rate of *Beroe* larva in the Caspian water by comparison with those in the Black Sea water.

Materials and Methods

Reproduction characteristics of B*. ovata* in the Caspian and the Black Sea water were studied in four sets of experiments (September, 12-14, 18-20, 21-23, and 24-26). The Caspian Sea water was delivered by plane from Iran, Mazandaran and before the experiments was kept in 20-l containers in Aquaculture Department of Sinop University. In the experiments without previous adaptation freshly collected *Beroe ovata*, 40-50 mm in size, were placed individually in 4-5 l jars filled with seawater filtered through 180-μm mesh. In each set six experimental jars were filled with the Caspian Sea water and six jars – with the Black Sea water. The temperature was 22^{o}C. Animals were kept in the experimental jars for 24 hours, and then were carefully removed and measured. The jars were left without any handling for the time necessary for larva hatching, for approximately 24 hours. Then the water was concentrated to a 100 ml volume and number of larvae and undeveloped eggs was estimated under a dissecting microscope.

In other set of experiments *Beroe* were adapted to lower salinity being transferred into 15‰ water (mixed the Black and Caspian Sea water 1:1) for 24 h. Six jars with the animals were left at salinity of 15‰ and *Beroe* from other six jars were transferred into Caspian Sea water. Six containers with ctenophore in Black Sea water were left as control. The number of eggs and larvae were examined daily until *Beroe* stopped reproduction in the Black Sea, mixed and Caspian Sea water. The conditions of the experiments (temperature, volume and number of animals) were the same as in the first set of the experiments.

150-300 fresh laid eggs were put into 50-150 ml glasses to examine the hatching success.

The experiments on survival and growth of *Beroe* larva were performed during the period from 4 to 26 September. In the first series of experiments for study the effect of lower salinity on survival, new-hatched *Beroe* larvae were previously adapted to lower salinity being transferred into 15‰ water (mixed the Black and Caspian Sea water 1:1) for 24 h. Four jars were left at salinity of 15‰ and the larvae from other 4 jars were transferred into Caspian Sea water. The jars with the Black Sea water served as a control. The larvae were kept at 26^0 C in 100- ml jars at initial density of 40 individuals per jar without any food.

In other set of experiments 160 newly hatched *Beroe* larvae were placed in the Black Sea water, 160 adapted to the lower salinity larvae, which were previously put in water of 15‰ salinity for 1 day, were transferred in the Caspian water. The water was filtered through 0.45-µm glass fiber filter. The temperature was 22^oC. The larvae were cultivated in 100 ml glass jars, 40-60 specimens per jar. *Mnemiopsis* tissue cut to small (1-2 mm) pieces was used as food for larvae.

Additionally the experiments on *Beroe* larvae growth in the Caspian Sea water were conducted using *Mnemiopsis* larvae as a food. Twenty larvae placed in 100-ml jars in three replicates were fed with freshly hatched *Mnemiopsis* larvae with size of 250-300 µm at initial concentration of 30 individuals per jar. In subsequent days the *Mnemiopsis* larvae density was increased to 50-60 specimens per jar.

The larvae were enumerated, measured, transferred into new filtered water, and fresh food was added daily. Wet weight of larvae was calculated from formula: W= Vρ, where W is the wet body weight (mg), V= 1/6 L 3.14 D^2 is the volume of ellipsoid, mm^3, L is the length and D is the diameter of ellipsoid (larvae width), mm, ρ is the body density which is closed to sea water density of 1.012 g/cm^3 for Caspian sea water. 60 *Beroe* larvae in the Caspian water and 60 in the Black Sea water were kept without any food and served as a control. All used larvae belonged to the same brood. To study morphometric changes in the growing larvae, the photos of animals were taken every day using a digital camera connected with a light microscope.

Results

Reproduction characteristics of *Beroe ovata* in the Caspian and Black Sea waters are presented in Table 1. In the period of our work, the proportion of *Beroe* laid eggs decreased both in the Caspian and Black Sea waters from 80 to 0% and from 100 to 50%, correspondingly. Size of their egg clutches also decreased dramatically from 544 to 0 eggs in the Caspian water and from 4498 to 57 eggs in the Black Sea water. It was connected with the end of reproduction period in the sea. Both *Beroe* placed in the Caspian water with and without adaptation showed the lower values of reproduction indexes as compared with those in the Black Sea water. Nevertheless, *Beroe ovata* could reproduce in the Caspian water even placed there without any adaptation and some share of eggs (about 10-15% of total number of the laid eggs) developed into larvae (Fig.1). In the Black Sea water the hatching success was much higher and was equal to 83-87%.

The new laid eggs that were transferred from the experimental jars to another ones to study hatching success did not developed at all, they either stopped their development or disintegrated in both experimental treatments.

The number of laid eggs examined during 4 days in *Beroe* at different salinities decreased but the most dramatic decreasing was observed in the Caspian Sea water (Fig. 2).

Survival of *Beroe* larvae in the Caspian and Black Sea waters at 22 ^{0} C differed significantly. The percentage of larvae survived under starvation and feeding both with larvae and pieces of *Mnemiopsis* during 6-8 days is shown in Fig.3. At the end of experiment, in the Caspian water only 20% of starveling and 30-40% of feeding larvae were alive. Time for 50% survival of the *Beroe* larvae in the Caspian Sea water was about 6 and 7 days for starveling and feeding larvae correspondingly. In the Black Sea water about 80% of larvae survived both under starvation and feeding. The significant difference in survival of starveling larvae in the Caspian and Back Sea water was also observed at 26^{0} C, but survival rate was much lower than at 22^{0} C and time for 50% survival ranged from 1.5 to 2.5 days for all treatments respectively (Fig. 4). Probably low survival rate in these experiments, besides the high temperature, could be result of some special condition of the parents that is an important factor determining survival and development of larvae.

The mean size (length) of *Beroe* larvae cultivated in the Caspian water increased insignificantly during 6 days of both sets of feeding experiments (Fig. 5a, Table 2). The insignificant growth of *Beroe* larvae feeding on *Mnemiopsis* larvae probably was result of lack of food (there was not enough *M.* larvae because it almost stopped reproduction this time). The average daily ration was about 2 ind/ ind/day. However, the maximal growth observed for some larvae was noticeable, from 0.5 to 0.75 mm. There was a great variability in growth rate of larvae, while some of them increased in size, another part stopped their growth and even decreased in size. We divided all larvae feeding on *Mnemiopsis* larvae into two groups: the first one are larvae that grew during experiment (their weight was higher than initial weight) and second group are larvae that had lower or equal of initial weight at the end of the experiment. It was found that the average weight of larvae in the first group was 47.5±2.2 mg, in the second one it was 10.0±0.7 mg against 24.6 ±12 mg of initial weight. It is worth noting that in the Caspian Sea water larvae changed their body form and were more round than in the Black Sea (Table 2). Their growth of width was higher than it was for the length. The changes in size and morphology of larvae are pretty good seen in photos that will be sent later. The growth rate of larvae cultivated in the Black Sea water was higher than that in the Caspian water (Fig.5 b). But there was the same tendency for a part of larvae to stop development.

Thus, we can conclude that *Beroe* larvae can grow in the Caspian Sea water though their grow rate is slow and mortality is high.

It is well known that the narrowest limits of tolerance for salinity are typical for initial ontogenetic stages. In our experiments *Beroe* eggs appeared to be the most sensitive to lower salinity.

During ontogenesis the salinity range is getting wider. According to our previous results larvae *Beroe* were more sensitive to salinity decreasing than adult ctenophores (Finenko et al., 2003) Meantime juvenile individuals were more resistant to reduced salinity from adult *Beroe* (Kideys et al., 2002). It seems possible to suppose that if to introduce *Beroe* to the Caspian Sea at the stages of eggs or early larvae the only small part of them could survive. But rather long preliminary acclimatization of juvenile animals (preferably with size of 10-20 mm) can affect upon the salinity resistance of both adult and embryos and larvae produced by them. As a result the survival range can be shifted towards lower salinity.

Conclusion

Our experiments showed that *B. ovata* can reproduce in the Caspian Sea water even without previous acclimation and some share of eggs laid (10-15%of total number) can develop into larvae. It enables to suppose that *Beroe* could survive in the Caspian Sea (in southern Caspian with salinity about 12 ‰) if it is introduced there occasionally like it was in case of *M. leidyi.*

Beroe larvae can grow in the Caspian Sea water though their growth rate is rather slow and survival is not so high as in the Black Sea.

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References

- Dumont HJ (1995) Ecocide in the Caspian. Nature 377: 673-674
- Finenko G.A., Romanova Z. A.,Abolmasova G.I.,Annindky B.E.,Svetlichny L.S.,Hubareva E.S.,Bat L.& Kideys A.E. Population dynamics, ingestion, growth and reproduction rates of the invader *Beroe ovata* and its impact on plankton community in Sevastopol Bay, the Black Sea. J. Plankton Res., 25:539-549
- Finenko, G. A., Anninsky, B. E, Romanova, Z. A, Abolmasova, G. I. and Kideys, A. E. (2001). Chemical composition, respiration and feeding rates of the new alien ctenophore, *Beroe ovata*, in the Black Sea. *Hydrobiologia,* **451,** 177–186
- Finenko G. A., Bat L., Arashkevich E. et al. 2003. Will the new alien ctenophore *Beroe ovata* control the plankton community in the Black Sea? A report prepared for NATO , EST CLG.976805GESAMP (IMO/FAO/UNESCO-IOC/WMO/WHO/IAEA/UN/UNEP Joint Group of Experts on the Scientific Aspects of Marine Environmental Protection, 1997. Opportunistic settlers and the problem of the ctenophore *Mnemiopsis leidyi* invasion in the Black Sea. Rep Stud GESAMP 58: 84p.
- Ivanov, P.I., Kamakim, A.M., Ushivtzev, V.B., Shiganova, T., Zhukova, O, Aladin, N., Wilson S.I., Harbison G.R., Dumont H.J., 2000 Invasion of Caspian Sea by the comb jellyfish Mnemiopsis leidyi (Ctenophora). Biological Invasions 2: 255-258
- Kideys, A. E., 1994. Recent dramatic changes in the Black Sea ecosystem: The reason for the sharp decline in Turkish anchovy fisheries. J Mar Sys 5: 171-181
- Kideys, A.E., Shahram, G., Davood, G., Roohi, A, Bagheri, S., 2001a. Strategy for combatting Mnemiopsis in the Caspian waters of Iran. A report prepared for the Caspian Environment Programme, Baku, Azerbaijan, Final Report, July 2001
- Kideys, A.E, Jafarov, F.M, Kuliyev, Z., Zarbalieva, T. 2001b. Monitoring Mnemiopsis in the Caspian waters of Azerbaijan. A report prepared for the Caspian Environment Programme, Baku, Azerbaijan, Final Report, August 2001
- Kideys A., Finenko G., Arashkevich A., Shiganova T. et al., 2002. Propagation experiments with Beroe ovata in the Caspian Sea water in autumn 2002. A report prepared for Caspian Environment Program, Baku, Azerbaijan, Final Report, October 2002
- Kideys, A.E., Moghim, M., 2003 Distribution of the alien ctenophore *Mnemiopsis leidyi* in the Caspian Sea in August 2001 Marine Biology 142: 163-171
- Kideys, A.E., Finenko, G.A., Anninsy, B.E., Shiganova T.A. et al. 2004. Physiological characteristics of the ctenophore *Beroe ovata* in Caspian Sea water. Mar. Ecol. Prog. Ser, 266:11-121
- Kovalev, A. V., Gubanova, A. D., Kideys, A. E., Melnikov, V. V., Niermann, U. Ostrovskaya, N. A., Prusova, I. Yu., Skryabin, V. A., Uysal, Z. and Zagorodnyaya, Ju. A ,1998. Long-term changes in the biomass and composition of fodder zooplankton in coastal regions of the Black Sea during the period 1957–1996. In:Ivanov, L. I. and. Oguz, T. (eds.). Ecosystem Modeling as a Management Tool for the Black Sea. Klüwer Academic Publishers, Dordrecht/Boston/London, 1:209–219

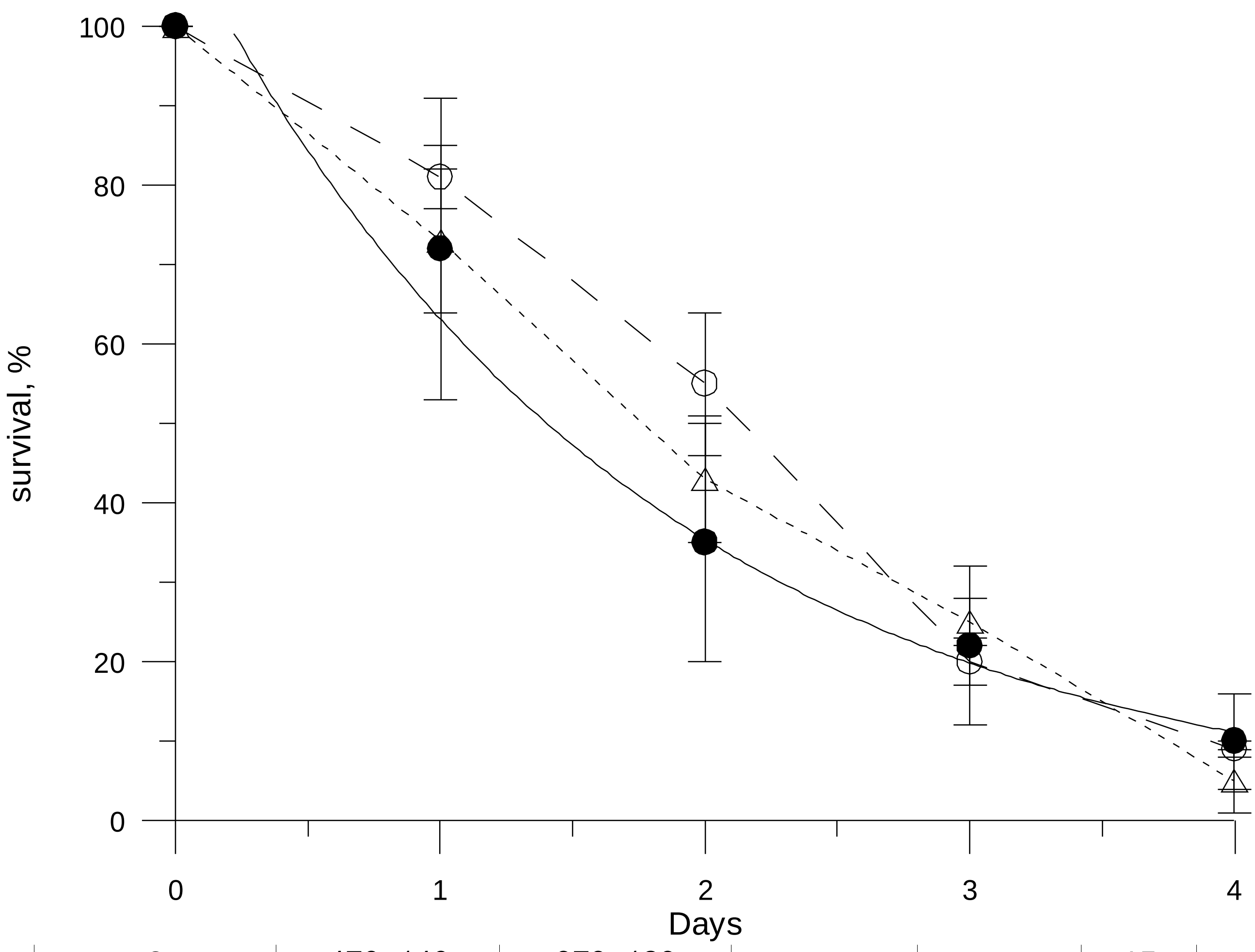
- Shiganova, T.A., Kamakin A.M., Zhukova O.P., Ushivtzev V.B., Dulimov A.B., Musayeva E.I., 2001. The invader into the Caspian Sea ctenophore Mnemiopsis and its initial effect on the pelagic ecosystem. Oceanology, 41: 517-524
- Shiganova, T.A., Sapognikov, V.V., Musaeva, E. I., Domanov, M. M., Bulgakova, Y.V. et al., 2003. Factors that determine pattern of distribution and abundance Mnemiopsis leidyi in the Northern Caspian. Oceanology, 43:716-733
- Vinogradov, M. E., Sapoznikov, V. V. and Shushkina, E. A. (1992). The Black Sea ecosystem. Nauka, Moscow, 112 p. (in Russian)

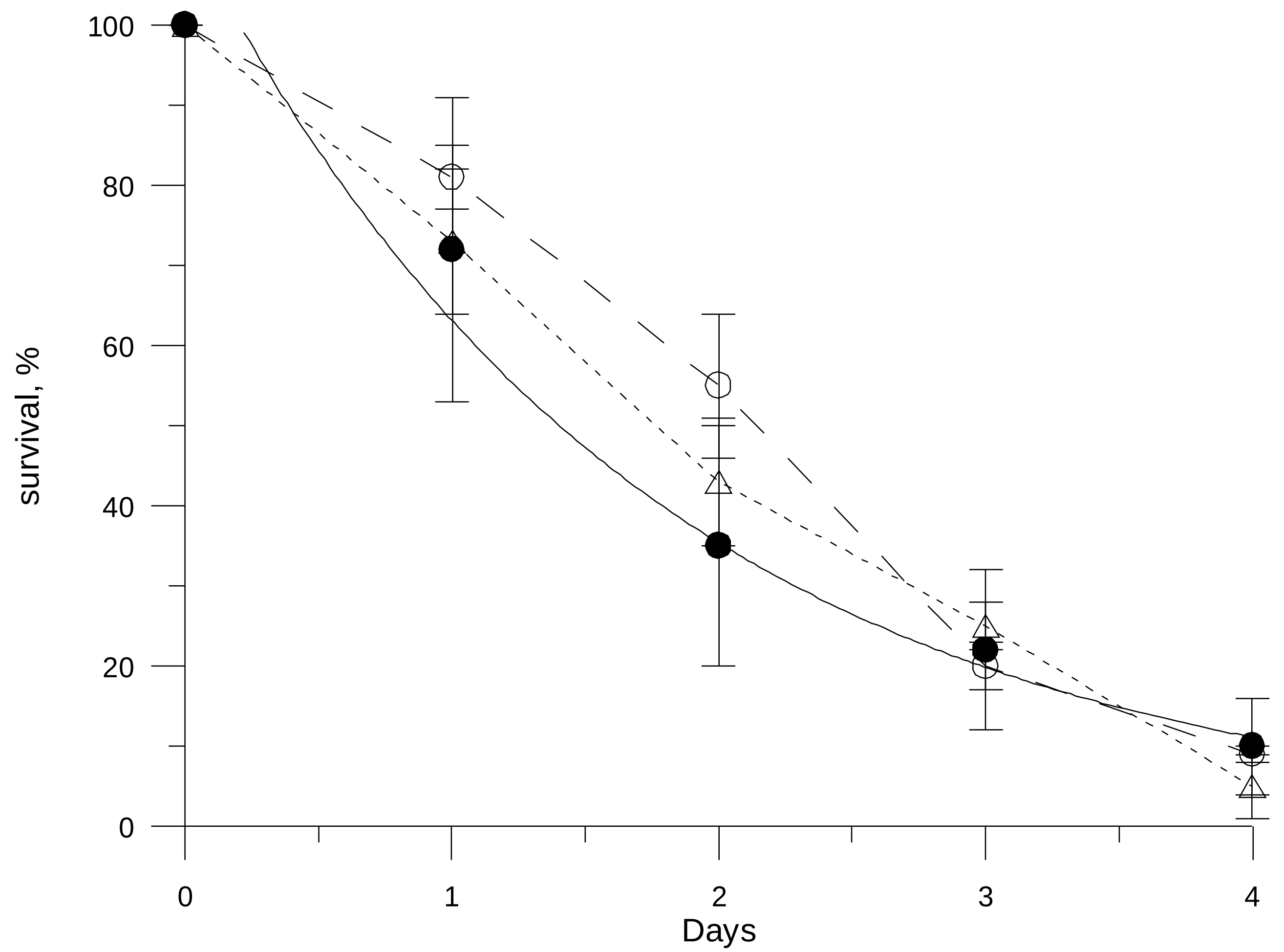
Table 1.
Reproduction characteristics of *Beroe ovata* in the Caspian and Black Sea waters

Reproduction characteristics	Date	The Caspian Sea water	The Black Sea water
Proportion of Beroe spawned (%)	12-14 September 2003	80	100
Clutch size (mean±SD)		544±928	4498±2652
Hatching success (%)		15±3	96±4
Proportion of Beroe spawned (%)	18-20 Sept 2003	71	86
Clutch size (mean±SD)		409±754	1684±543
Hatching success (%)		10±3	95±1
Proportion of Beroe spawned (%)	21-23 Sept 2003	67	67
Clutch size (mean±SD)		55±43	525±386
Hatching success (%)		10±5	87±15
Proportion of Beroe spawned (%)	24-26 Sept 2003	0	50
Clutch size (mean±SD)		0	57±17
Hatching success (%)		0	83±10
Proportion of Beroe spawned (%)	12-14 Sept 2003	80	83
Clutch size (mean±SD)	**adapted Beroe**	103±142	2156±576

Table 2. Size of *B. ovata* larvae at the end of the feeding experiments (food is *M. leidyi* larvae):L is length, D is width, μm. Duration of the experiment is 6 days.

N experiment	**L**	**D**	**L/D**	**W, mg**	**n**
Initial	**440±76**	**310 ±77**	**1.48±0 34**	**24.6±12.0**	**20**
1	478±115	394±138	1.29±0.27		9
2	450±93	413±106	1.15±0.31		8
3	470±140	373±129	1.38±0.58		15
average ± SD	**467±120**	**389±124**	**1.30**±0.45	**28.5±25.2**	**32**





Survival of *Beroe* larvae at salinity 12 (black circles), 15 (triangles) and 18 ppt (open circles) at 26 ^{0}C

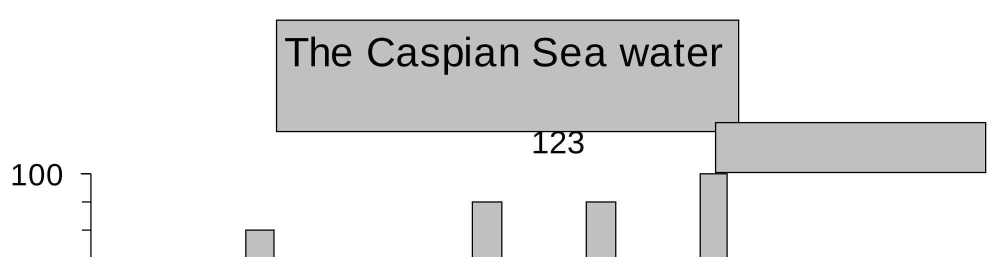


Fig.1. Percentage of disintegrated eggs in *B. ovata* in the Caspian and Black Sea water: A -12-14 September; B, C – 18-20 September 2003

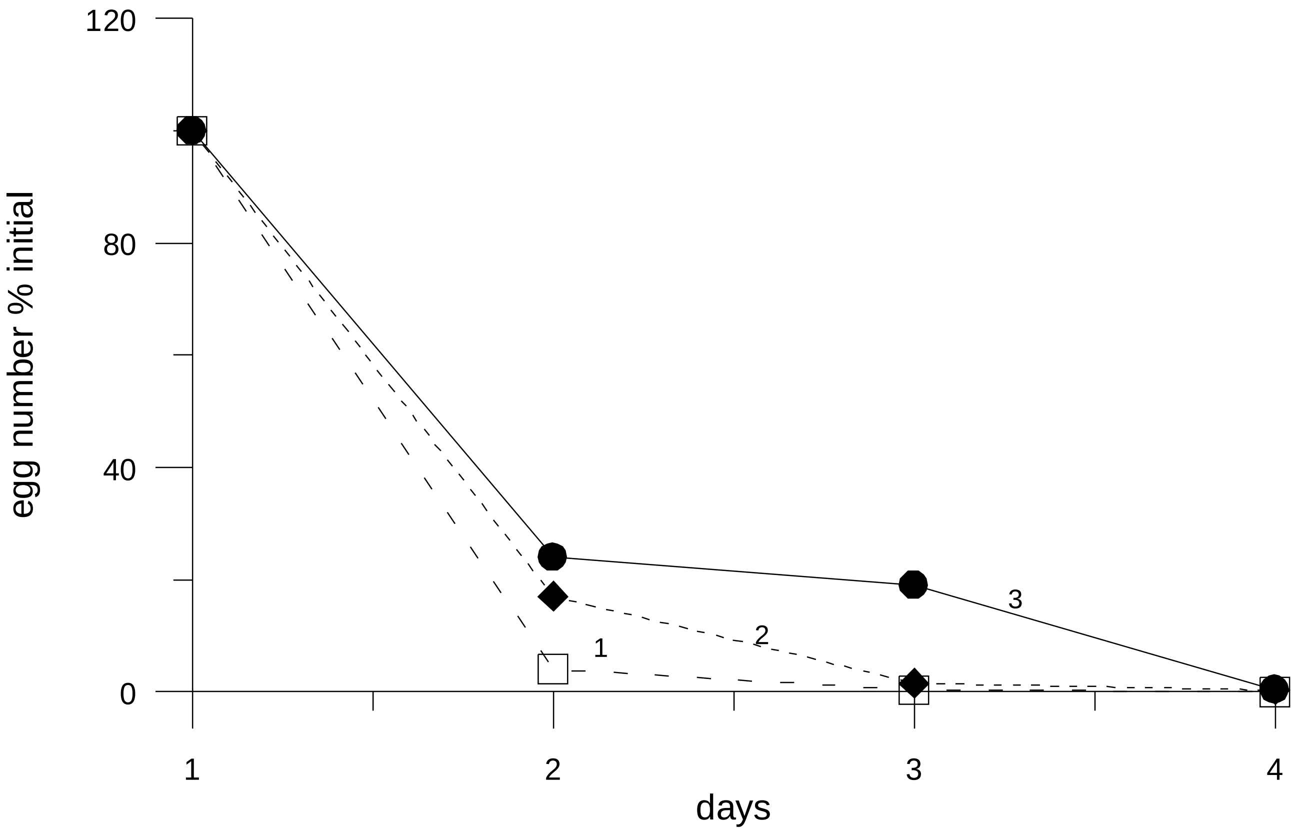
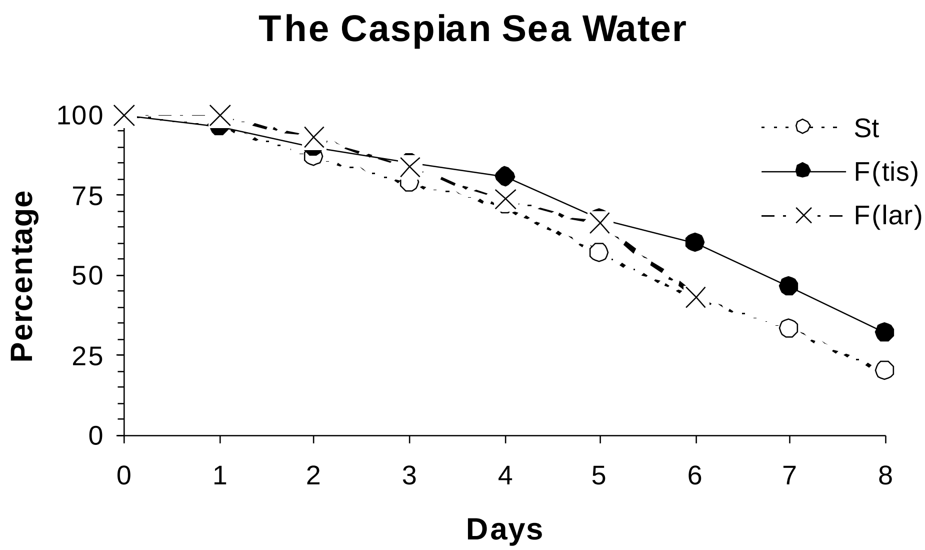


Fig. 2. Dynamics of egg number in *B. ovata* at different salinity: 1- Caspian water, 2- mixed water (Black Sea: Caspian Sea water, 1:1), 3- Black Sea water



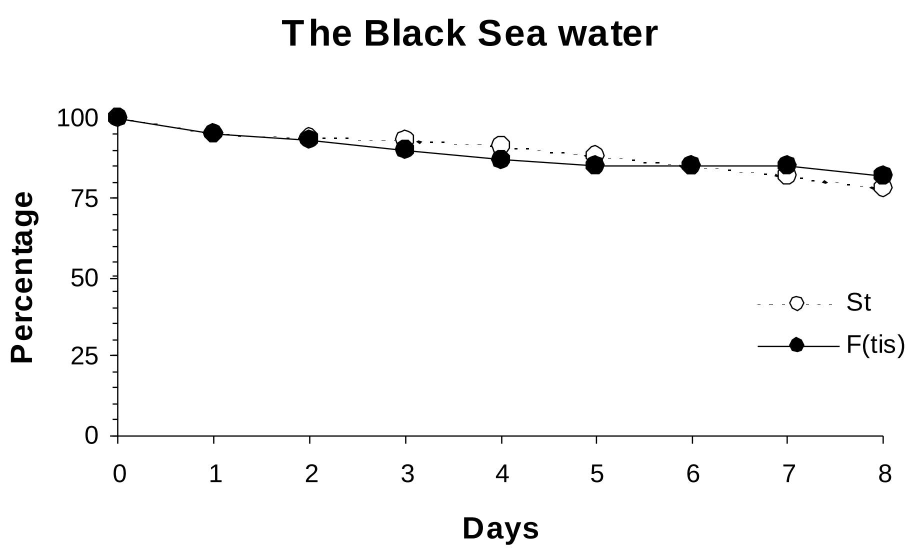
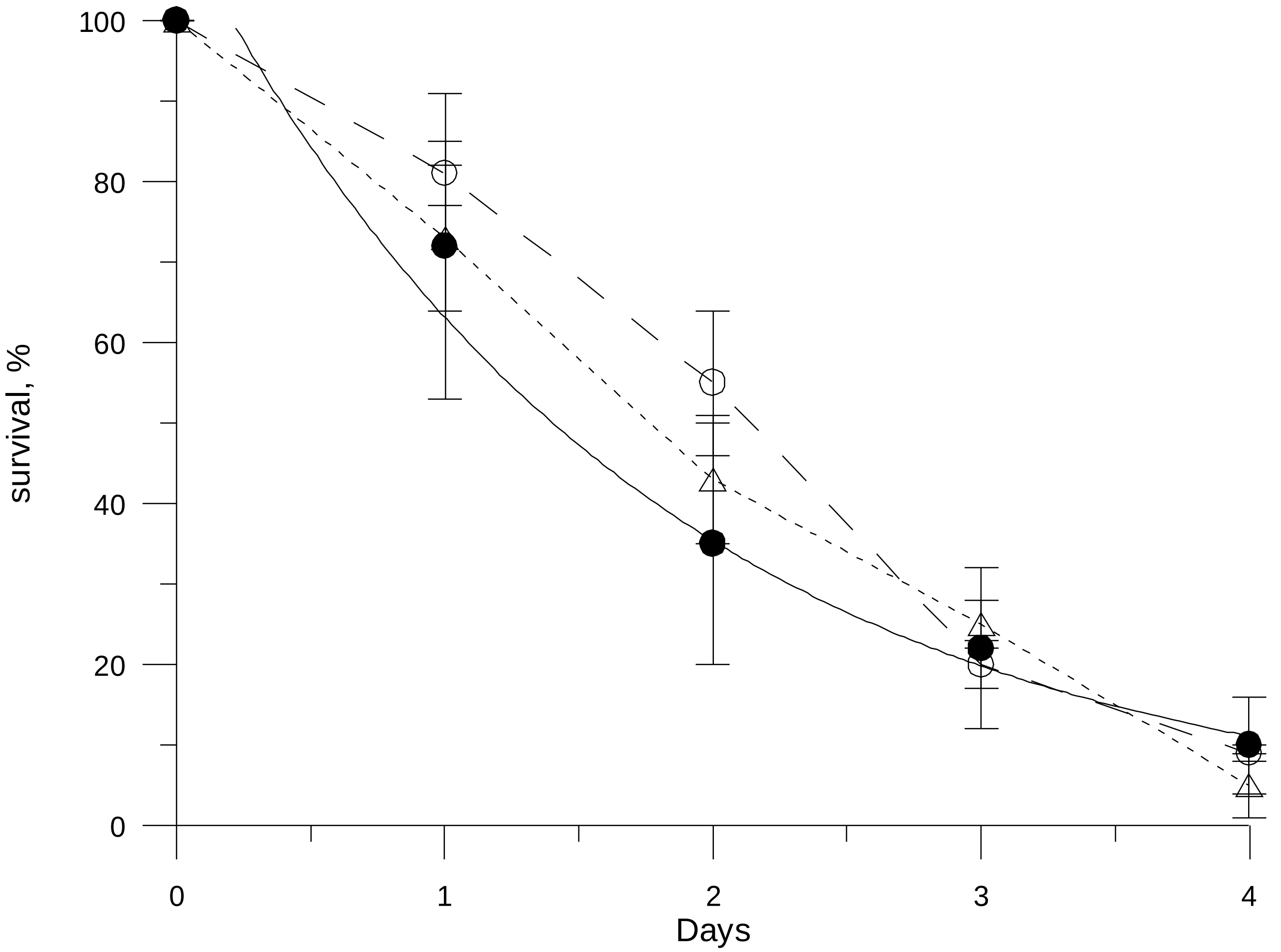


Fig. 3. Survival of *Beroe* larvae in the Caspian and Black Sea water: St is starveling, F(tis) - feeding on *M. leidyi* tissue, F(lar)- feeding on *M. leidyi* larvae



Survival of *Beroe* larvae at salinity 12 (black circles), 15 (triangles) and 18 ppt (open circles) at 26 ^{0}C

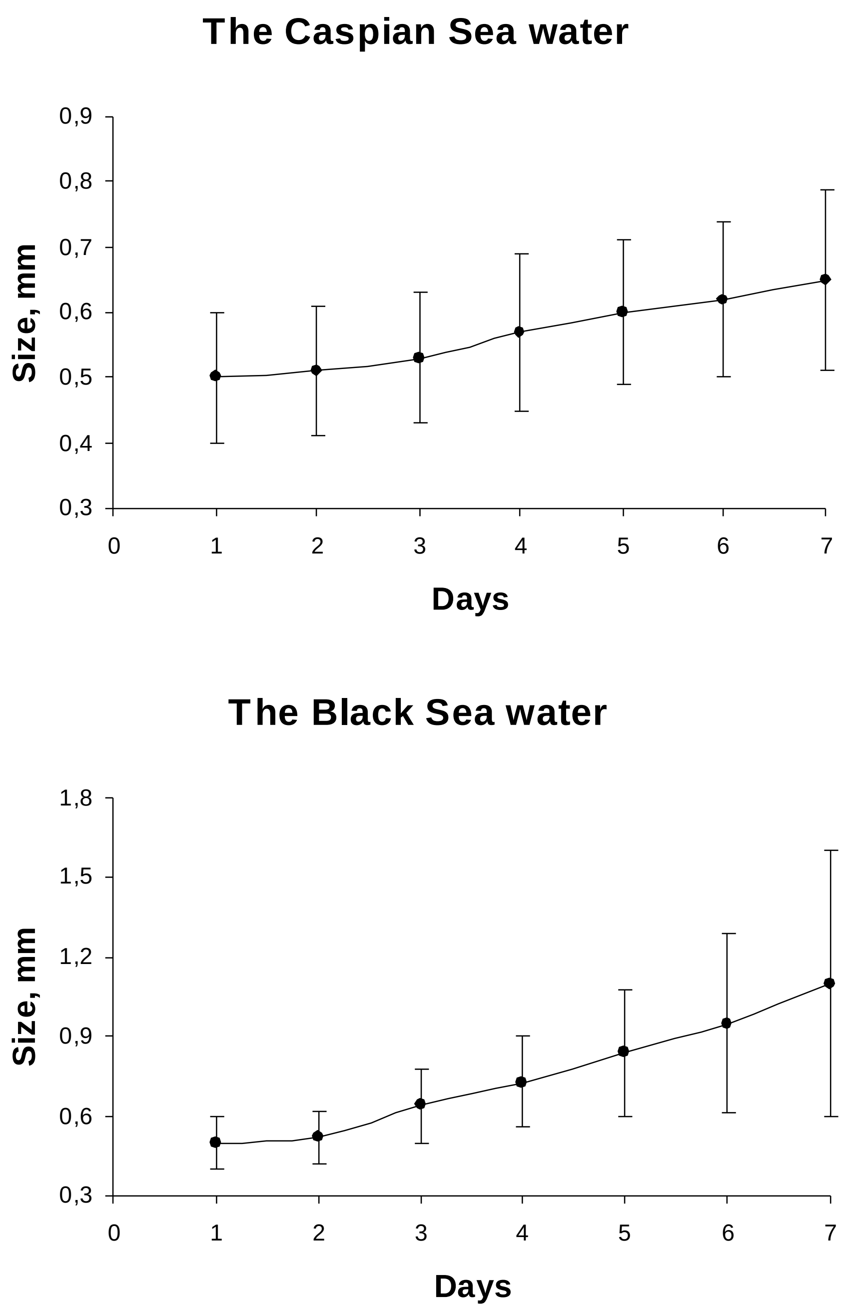
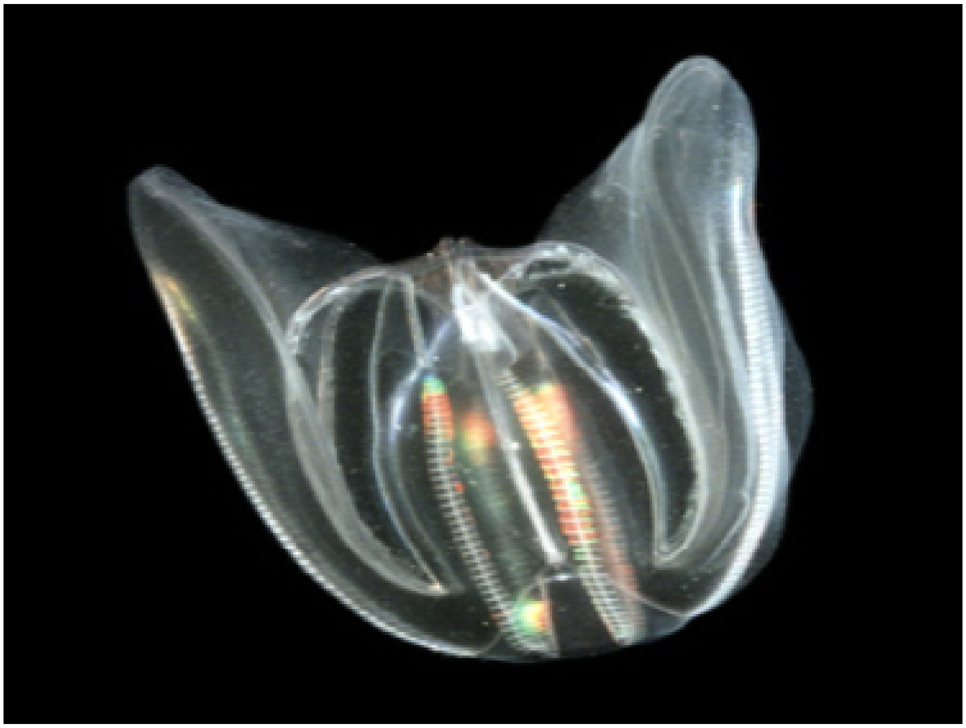


Fig. 5. Growth of *B. ovata* larvae in the Caspian and Black Sea water

Environmental impact assesment for introduction of Beroe ovata into the Caspian Sea

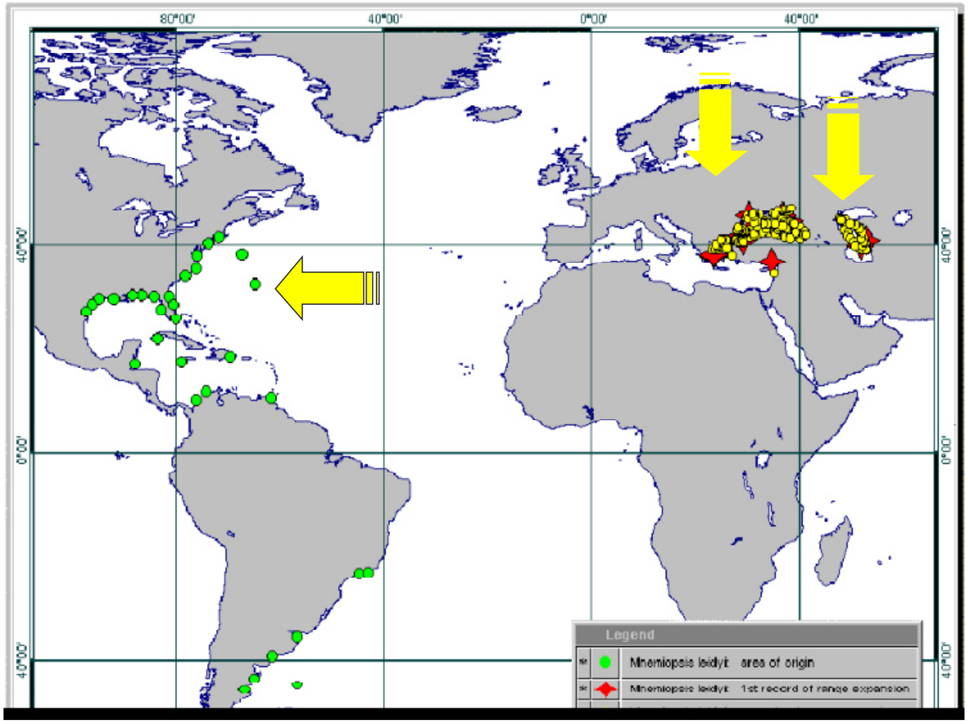
Tamara Shiganova

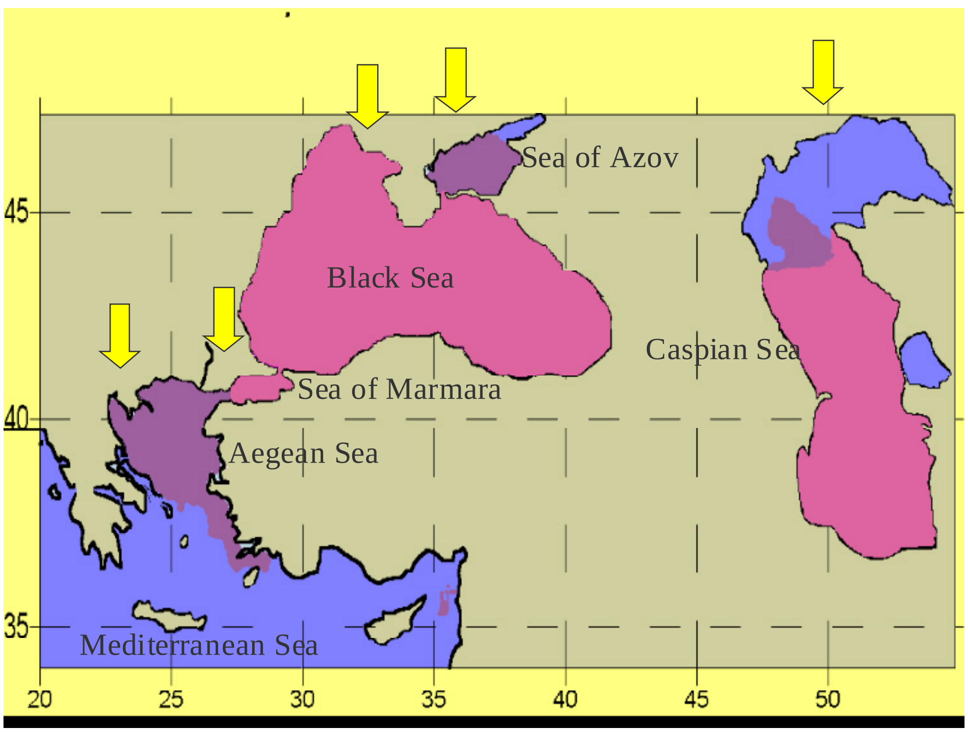
Contents



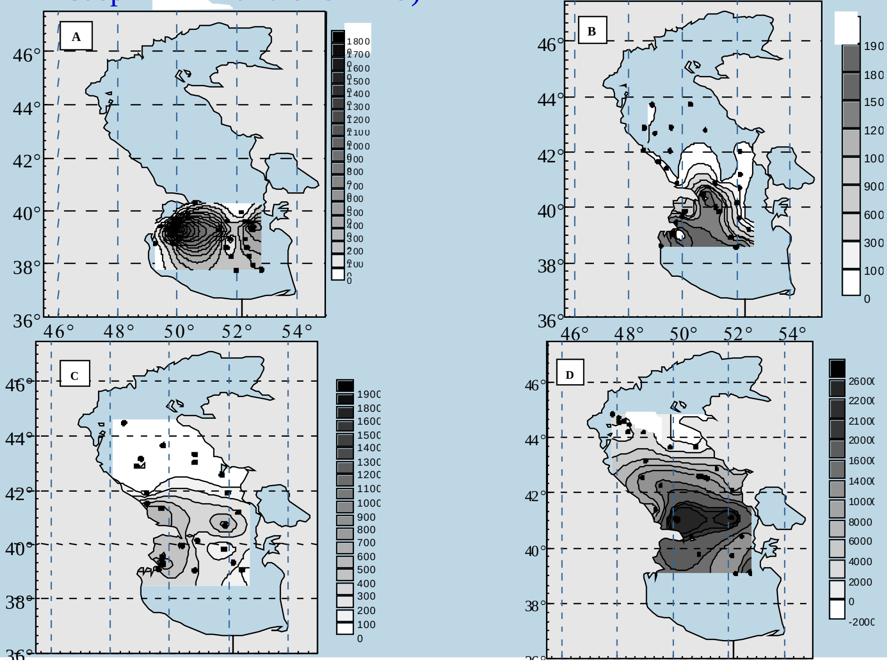
Systematics

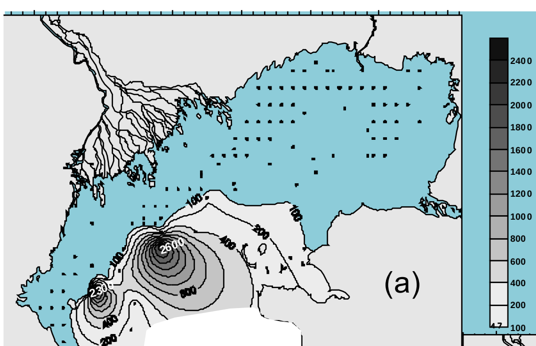
Ctenophora	***Phylum***
Tentaculata	***Class***
Lobata	***Order***
Bolinopsidae	***Family***
Mnemiopsis	***Genus***
leidyi Agassiz,1865	***Species***


80°00'
40°00'
0°00'
40°00'
Legend
Mneniopsis leidyi area of origin
Mneniopsis leidyi 1st record of range expansion

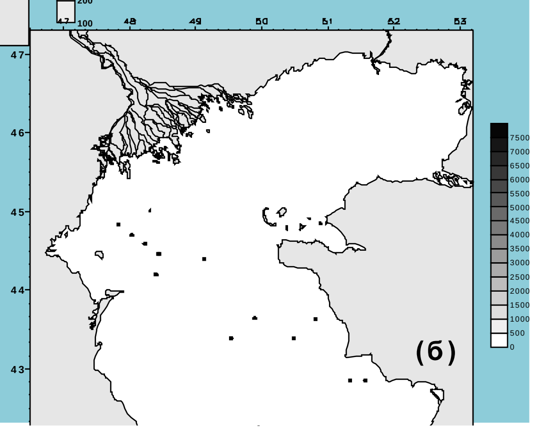

Sea of Azov
Black Sea
Caspian Sea
Sea of Marmara
Aegean Sea
Mediterranean Sea
45
40
35
20
25
30
35
40
45
50

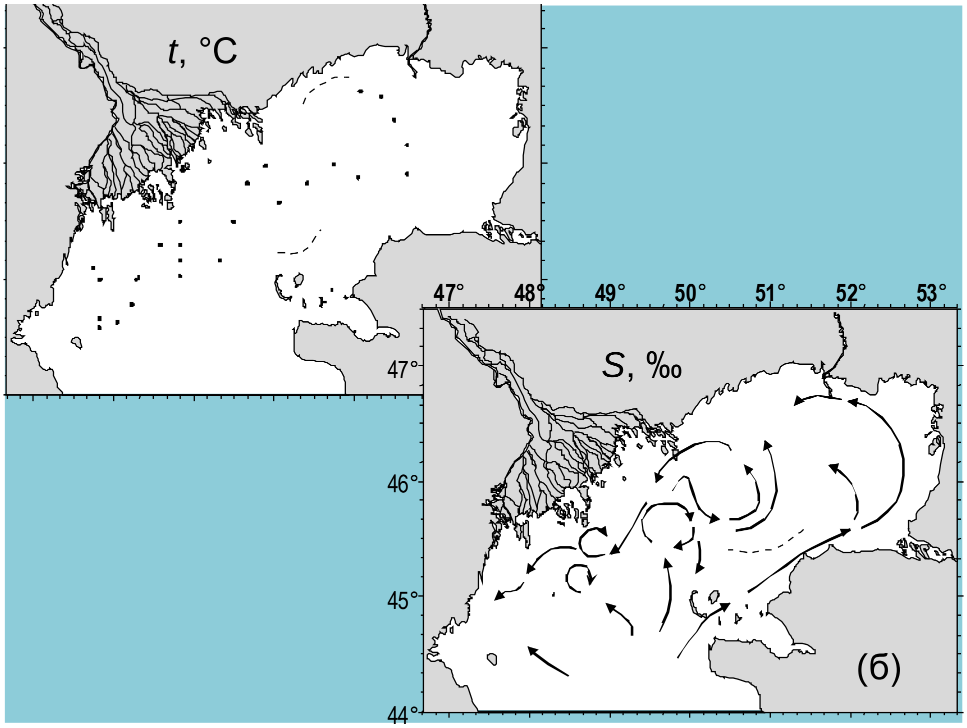
A-in January; B-in May; C-in July; D- in August (data CaspNIRHK and SIO RAS)

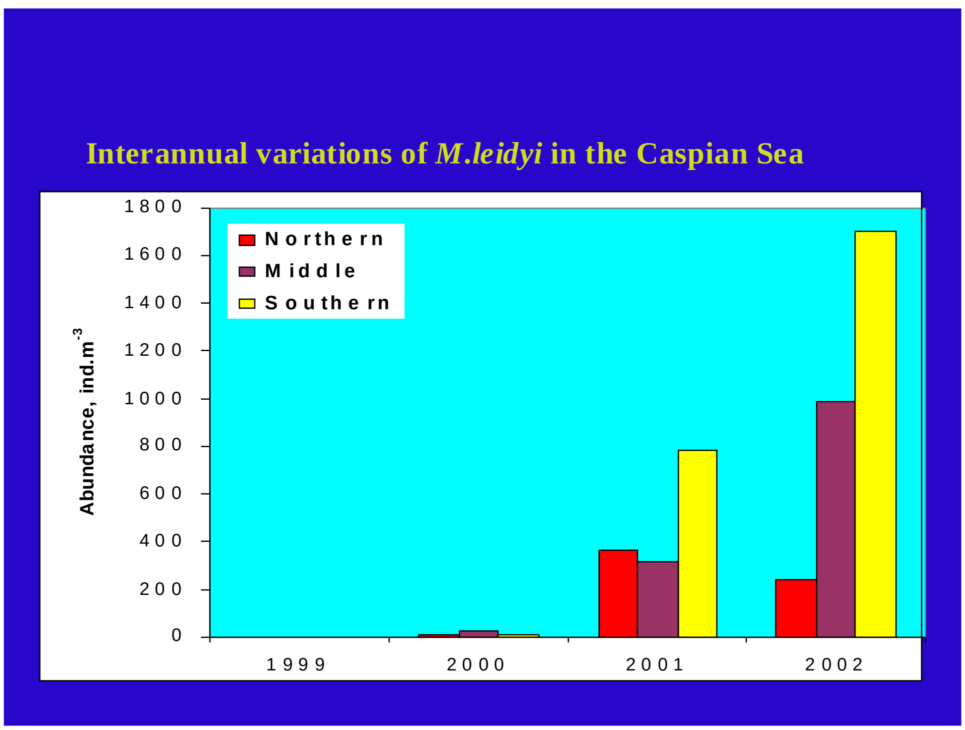




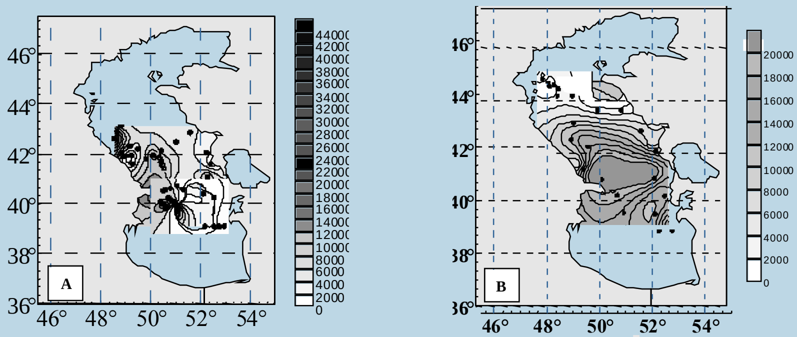
Pattern of *M.leidyi* spatial distribution in the Northern Caspian: a- in 2002, b-in 2001



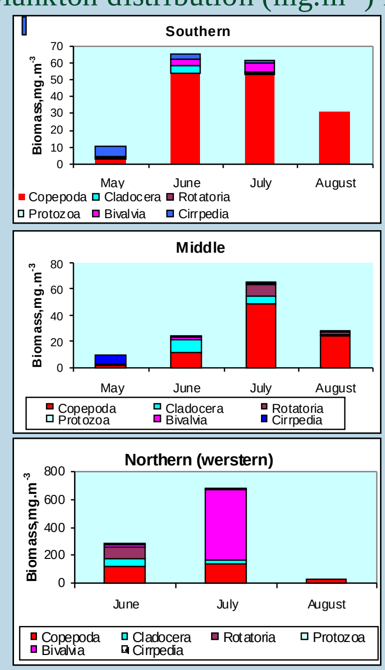

t, °C
S, ‰
(б)
47°
48°
49°
50°
51°
52°
53°
47°
46°
45°
44°


Interannual variations of M.leidyi in the Caspian Sea
Abundance, ind.m-3
Northern
Middle
Southern
0
200
400
600
800
1000
1200
1400
1600
1800
1999
2000
2001
2002

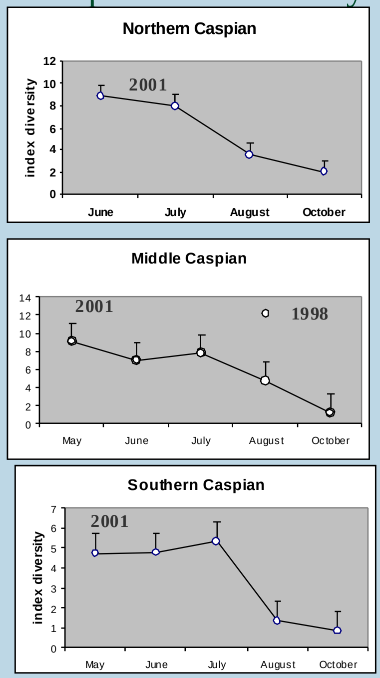
Pattern of zooplankton abundance spatial distribution (ind. m^{-3}): A –in June, B- in August.



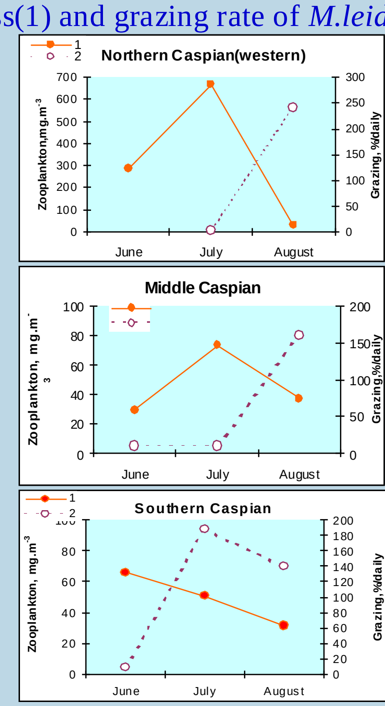
Seasonal zooplankton distribution ($mg.m^{-3}$) in the Caspian Sea



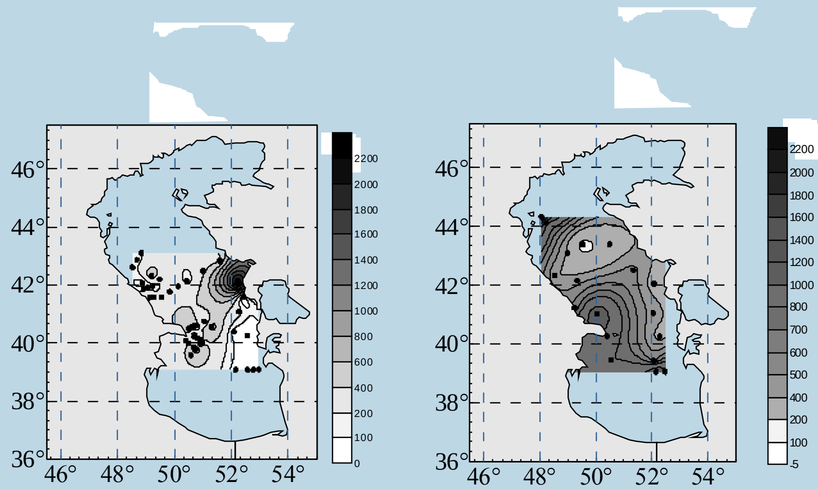
Seasonal changes of zooplankton diversity in the Caspian Sea



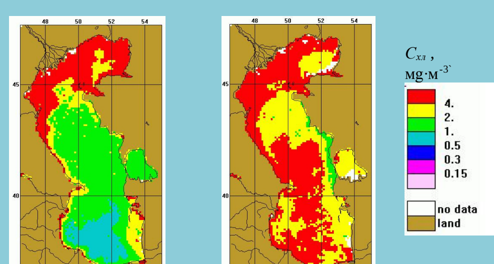
Zooplankton biomass(1) and grazing rate of *M.leidyi*(2) in the Caspian



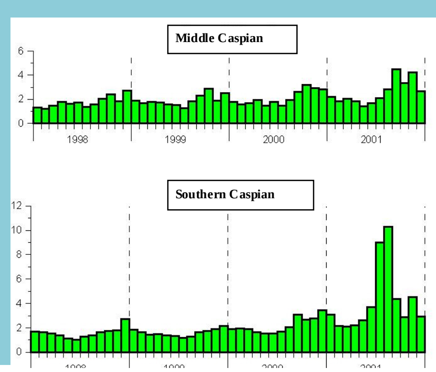
Pattern of phytoplankton biomass spatial distribution (mg. m^{-3}) –
A –in June, B- in August.



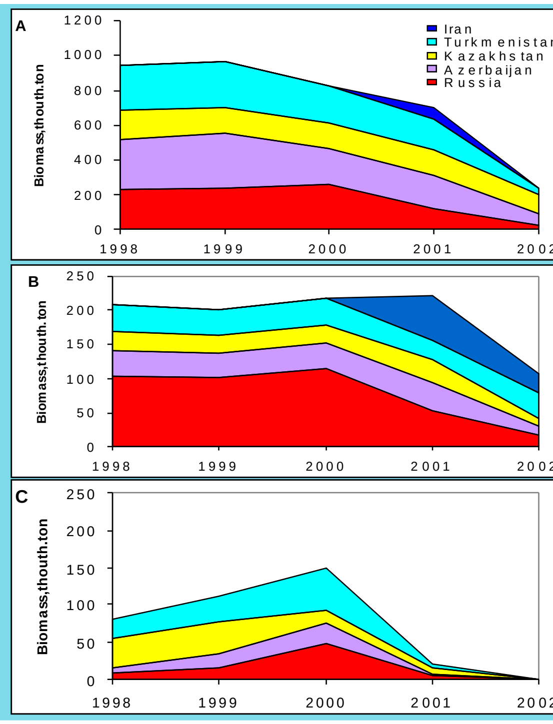
Interannual changes of chlorophyll concentration in the Caspian Sea (Kopelevich et al.2002)



September 1999 September 2001 .



Distribution of biomass of kilka for Caspian countries (after data of kilka surveys, data of lab kilka stocks assessment , KaspNIRKH): anchovy kilka; B- common kilka; C-big-eye kilka.



Predators are known to feed on *M.leidyi,* possible candidate for introduction in the Caspian Sea to combat *M.leidyi.*

•***Chysaora quinquecirrha***

First medusa *C. Quinquecirrha* does not always overcome population of *M.leidyi* because it consumes mainly small size of *M.leidyi* and second, the most important, *C. quinquecirrha* is a dangerous animal for people.

•**The vermform larval sea anemone *Edwardsia leidyi***

Anemone could not live in the low salinity and although tissue damage results, *M.leidyi* can regenerate tissue.

•***Beroe ovata***

It has two outstanding advantages: firstly, it is highly specific in its feeding, so that even its larval stage feeds on *M.leidyi.* Secondly, its reproductive rate and fecundity are almost as great as that of *M.leidyi,* so that its population can grow at similar rates to its prey

Fishes predators on *M.leidyi*.

- **Harvest fish, *Peprilus alepidotus***

- **Butterfish *Peprilus triacanthus***

These both species can eat *M.leidyi* but they are subtropical- temperate coastal species endemic of North America. They did not found in low salinity in the Chesapeake Bay, although in experiments *P. triacanthus* lived two weeks in salinity 4 ‰. Among the disadvantages of the introduction are the facts that its reproductive biology is poorly known, its eggs and larvae may be vulnerable to predation by *M.leidyi* (GEZAMP,1997) and their introduction would be very expensive transcontinental measures.

Some more tolerant for temperate with low salinity basin species were proposed for introduction into the Black Sea by GEZAMP group of experts (GEZAMP,1997).

•**The Baltic cod, *Gadus morhua callarias***

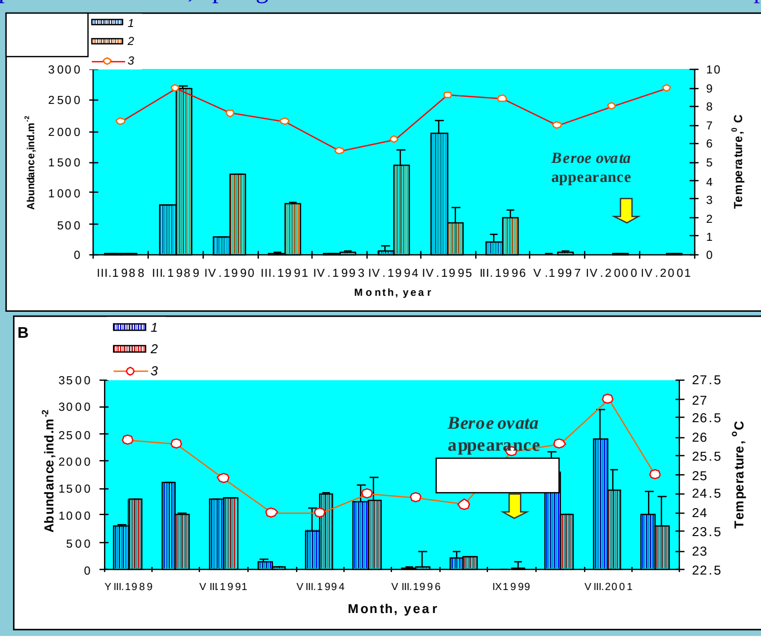
Among disadvantages are the facts that it will also eat commercially valuable small pelagic fish and it lives in cooler waters than Caspian Sea ones.

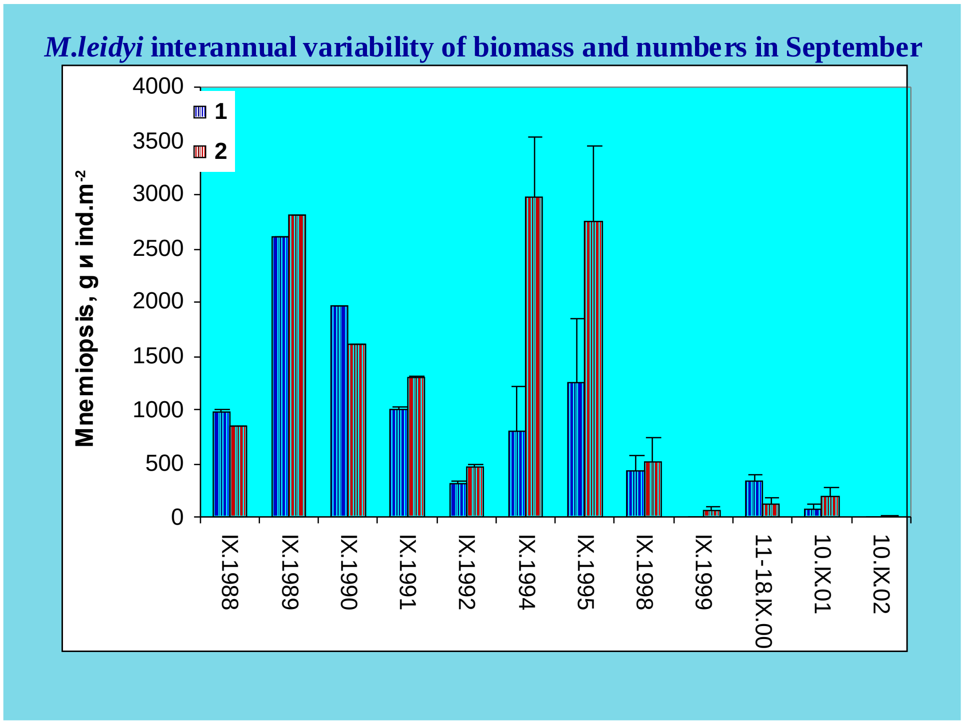
•**The Chum salmon *Oncorhynchus keta***

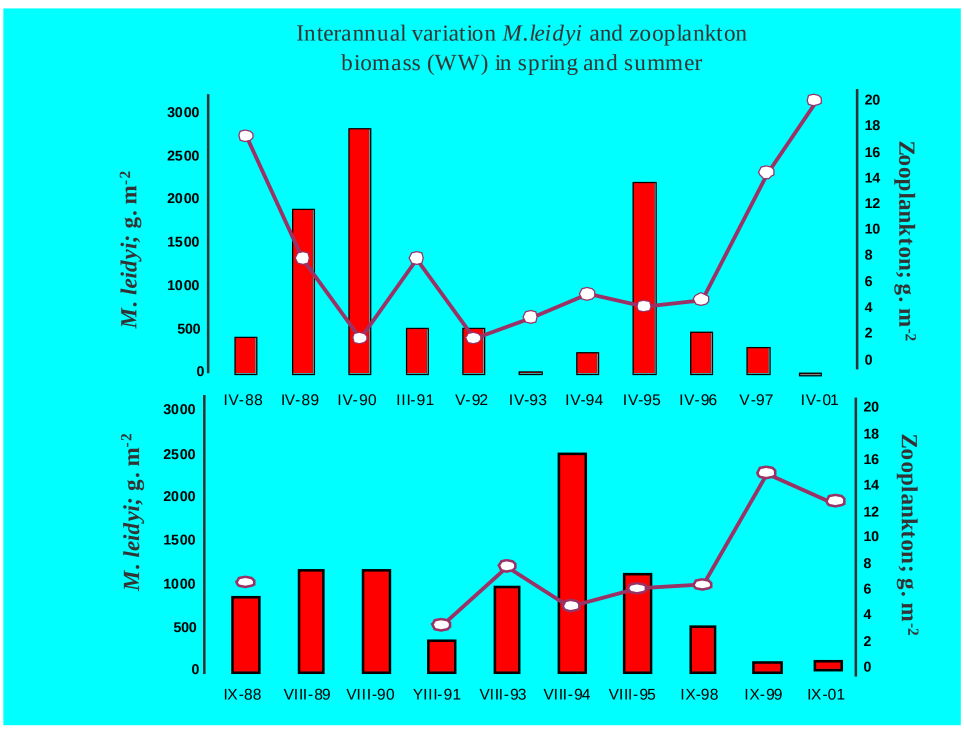
Among potential disadvantages are the facts that it is omnivorous on small pelagic fish, it may not be able to establish itself in the rivers that flow into the Caspian Sea, because of pollution and dams, and it may compete with native sturgeons.

Systematic	
Phylum	Ctenophora Esch
Class	Tentaculata Chun
Order	Beroidae Esch
Family	Beroidae
Genus	*Beroe*
Species	*Ovata Mayer*,1912

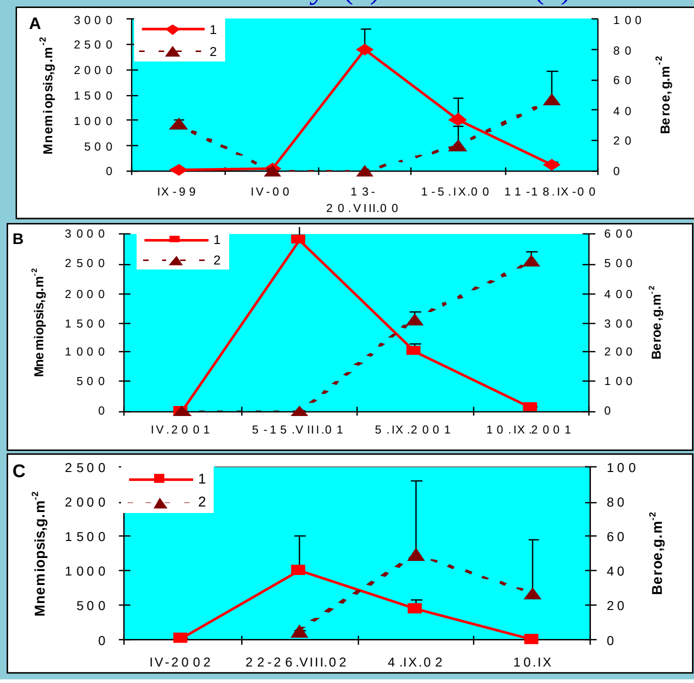
Long term Mnemiopsis leidyi abundance (1-inshore; 2-offshore) and surface water temperature in winter, spring and summer before and after *B. ovata* development



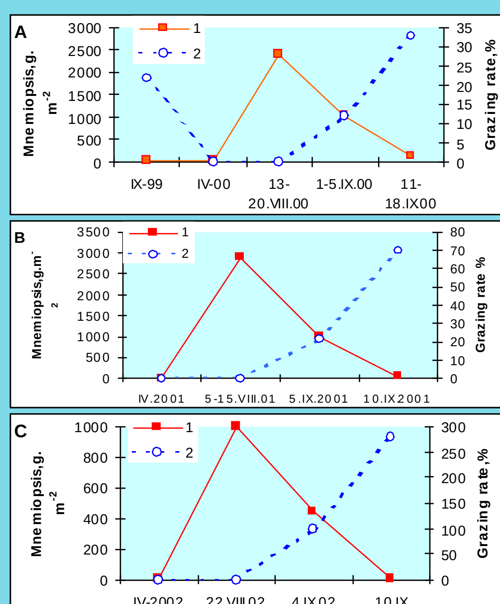

M.leidyi interannual variability of biomass and numbers in September
Mnemiopsis, g и ind.m-2
4000
3500
3000
2500
2000
1500
1000
500
0
1
2
IX.1988
IX.1989
IX.1990
IX.1991
IX.1992
IX.1994
IX.1995
IX.1998
IX.1999
11-18.IX.00
10.IX.01
10.IX.02

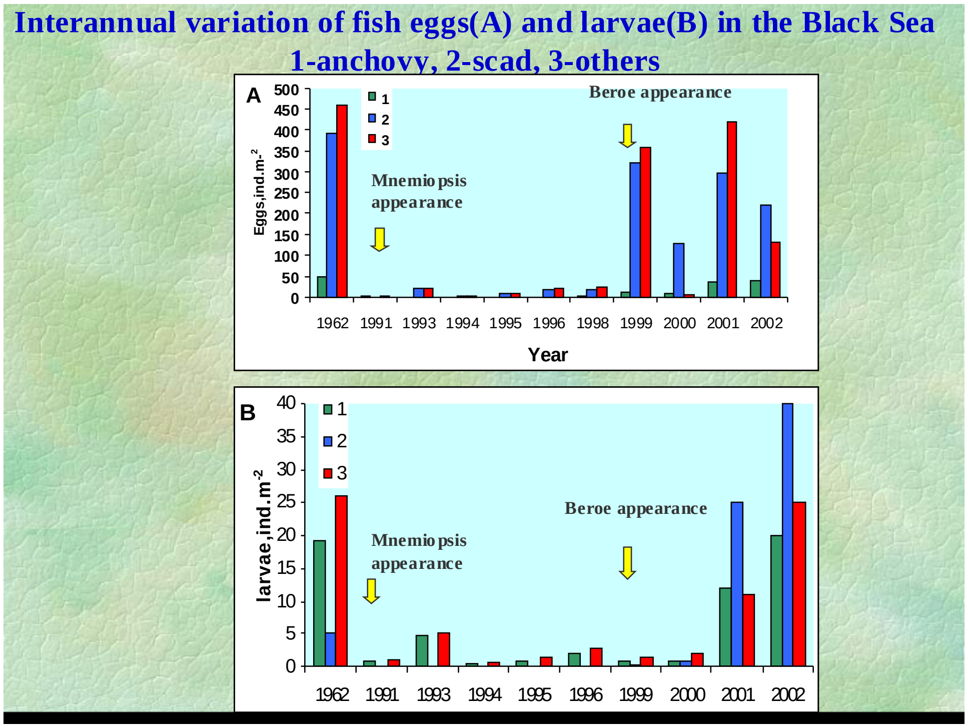

Interannual variation M.leidyi and zooplankton biomass (WW) in spring and summer
M. leidyi; g. m-2
Zooplankton; g. m-2
IV-88
IV-89
IV-90
III-91
V-92
IV-93
IV-94
IV-95
IV-96
V-97
IV-01
IX-88
VIII-89
VIII-90
YIII-91
VIII-93
VIII-94
VIII-95
IX-98
IX-99
IX-01

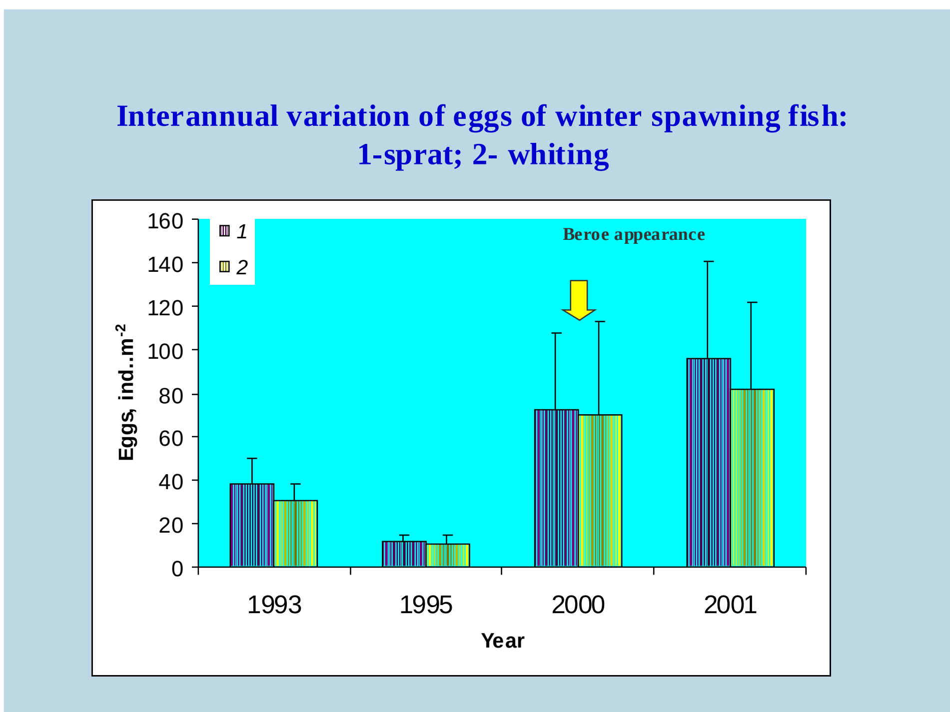
Interaction between *M.leidyi* (1)and *B.ovata*(2) in the Black Sea

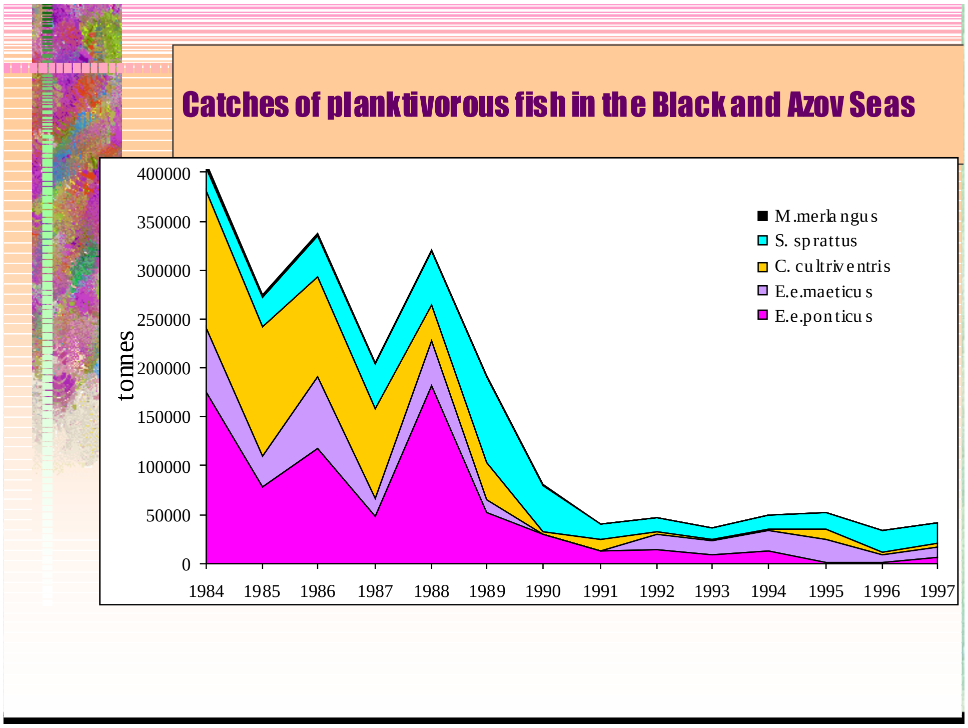


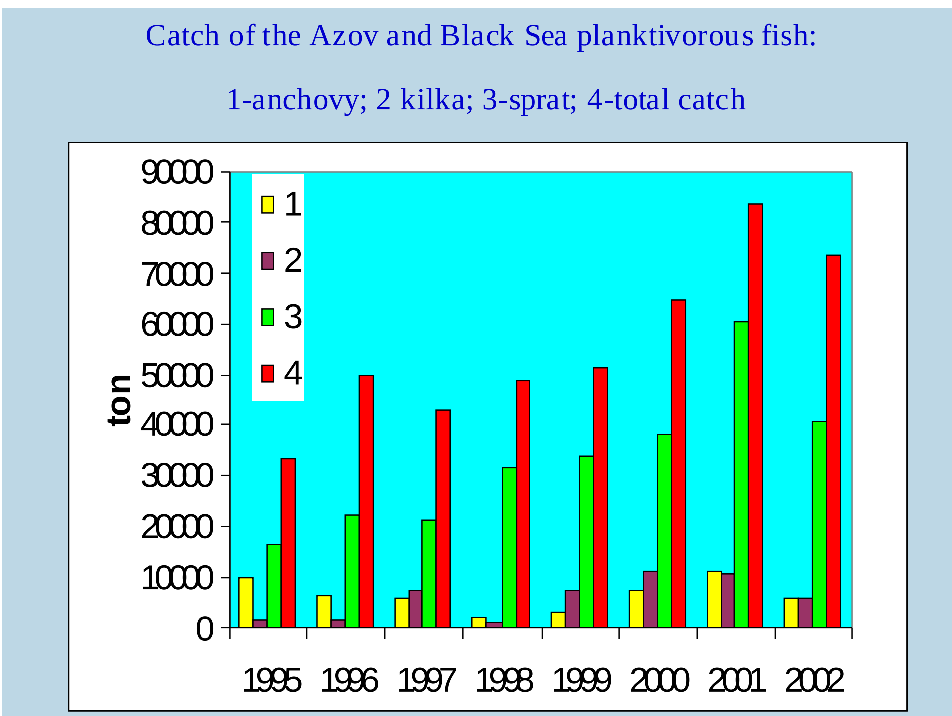
Mnemiopsis leidyi biomass, g.m^{-2} (1) and grazing pressure of *Beroe ovata*,% (2). A- in 1999-2000; B- in 2001; C-in 2002

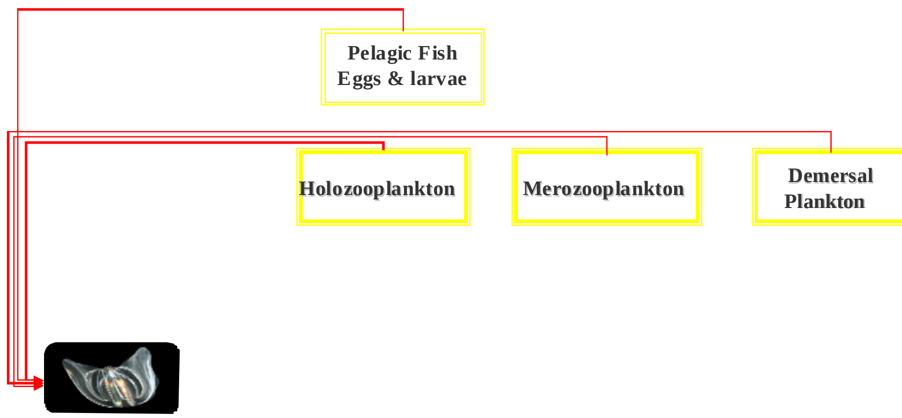


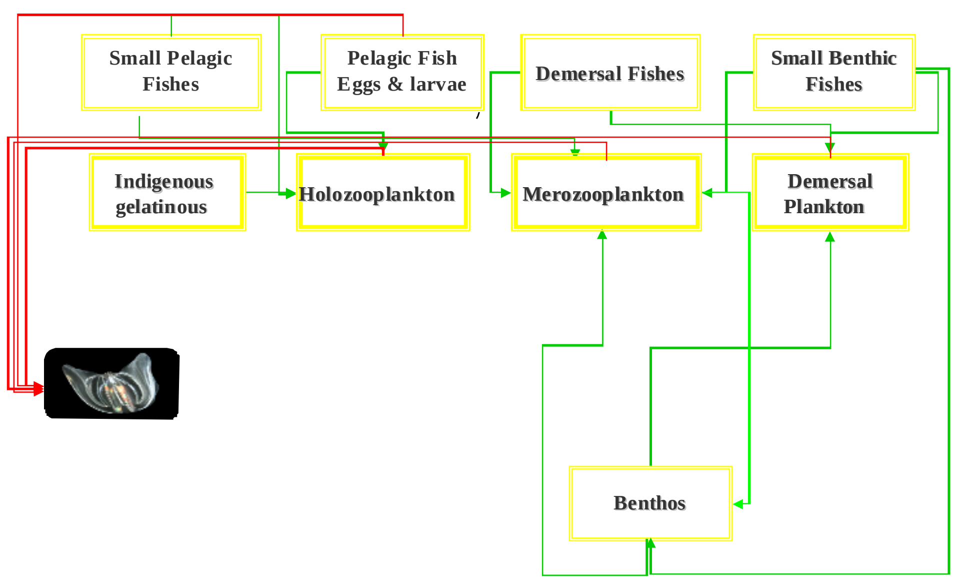

Interannual variation of fish eggs(A) and larvae(B) in the Black Sea
1-anchovy, 2-scad, 3-others
A
Eggs,ind.m-2
500
450
400
350
300
250
200
150
100
50
0
1
2
3
Mnemiopsis appearance
Beroe appearance
1962 1991 1993 1994 1995 1996 1998 1999 2000 2001 2002
Year
B
larvae,ind.m-2
40
35
30
25
20
15
10
5
0
1
2
3
Mnemiopsis appearance
Beroe appearance
1962 1991 1993 1994 1995 1996 1999 2000 2001 2002

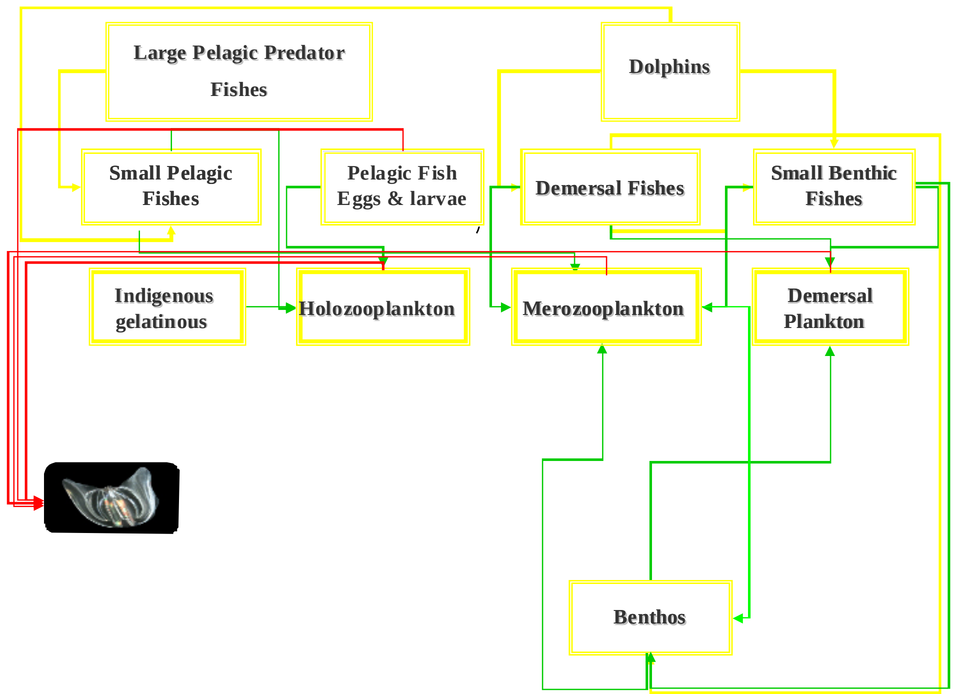

Interannual variation of eggs of winter spawning fish:
1-sprat; 2- whiting
Eggs, ind..m-2
160
140
120
100
80
60
40
20
0
1
2
Beroe appearance
1993 1995 2000 2001
Year


Catches of planktivorous fish in the Black and Azov Seas
tonnes
400000
350000
300000
250000
200000
150000
100000
50000
0
1984 1985 1986 1987 1988 1989 1990 1991 1992 1993 1994 1995 1996 1997
M.merlangus
S. sprattus
C. cultriventris
E.e.maeticus
E.e.ponticus

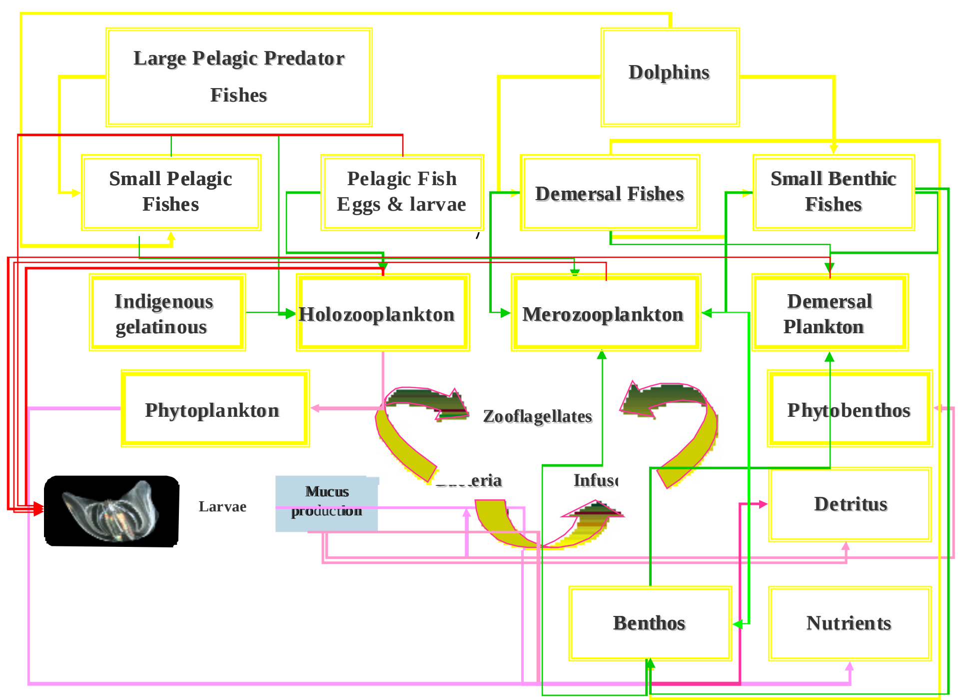

Catch of the Azov and Black Sea planktivorous fish:
1-anchovy; 2 kilka; 3-sprat; 4-total catch
ton
90000
80000
70000
60000
50000
40000
30000
20000
10000
0
1
2
3
4
1995 1996 1997 1998 1999 2000 2001 2002


Pelagic Fish Eggs & larvae
Holozooplankton
Merozooplankton
Demersal Plankton

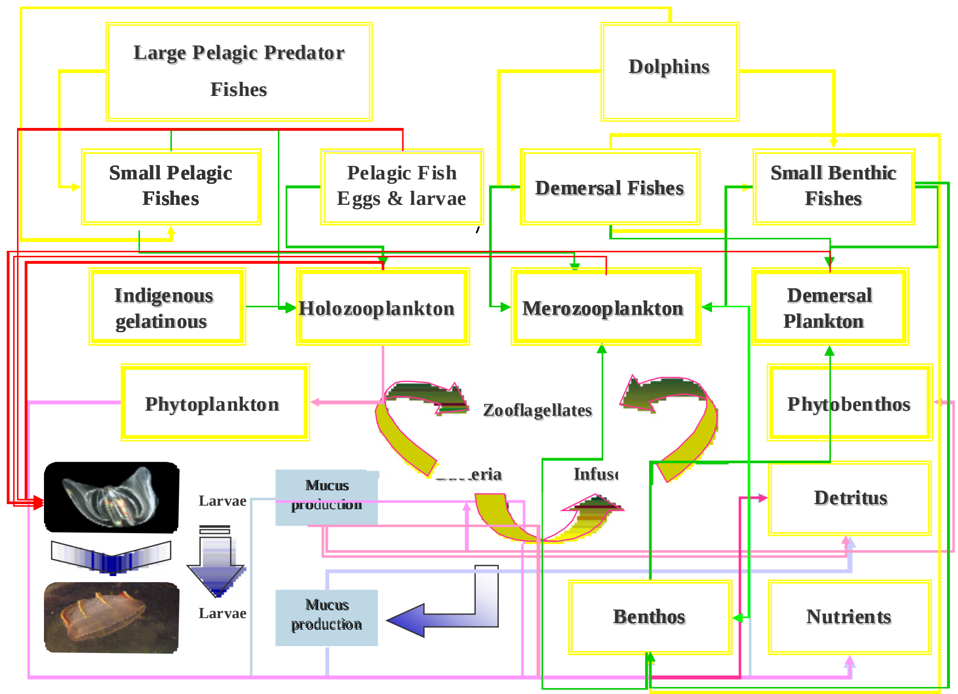

Small Pelagic Fishes
Pelagic Fish Eggs & larvae
Demersal Fishes
Small Benthic Fishes
Indigenous gelatinous
Holozooplankton
Merozooplankton
Demersal Plankton
Benthos

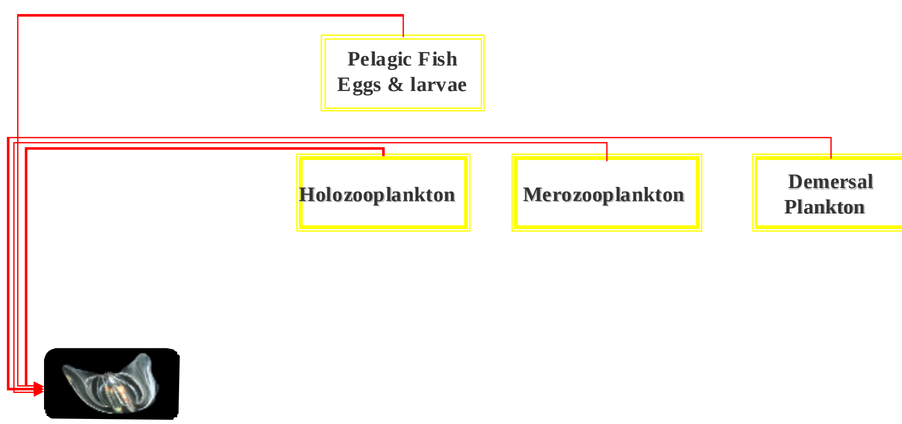


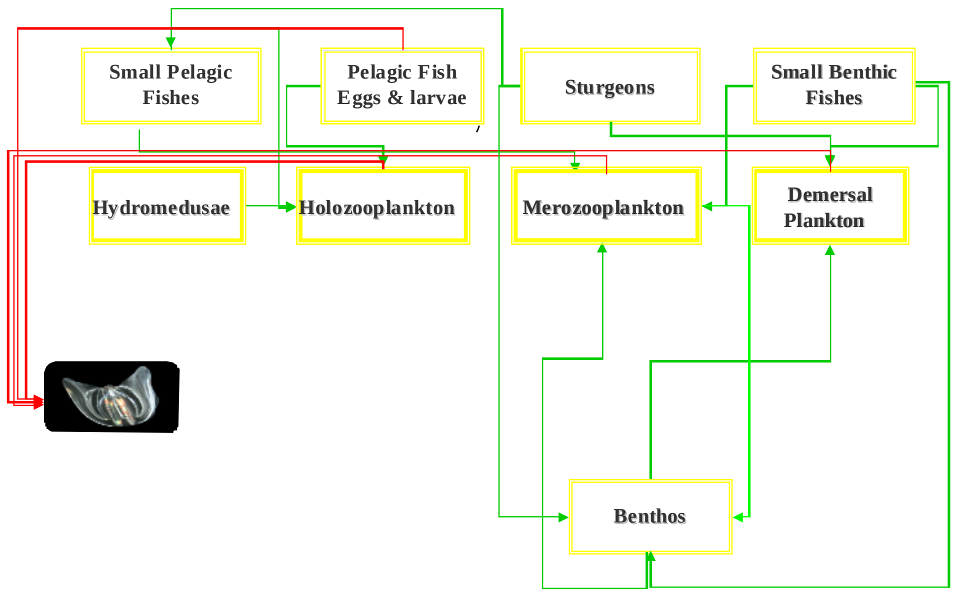
Large Pelagic Predator Fishes
Dolphins
Small Pelagic Fishes
Pelagic Fish Eggs & larvae
Demersal Fishes
Small Benthic Fishes
Indigenous gelatinous
Holozooplankton
Merozooplankton
Demersal Plankton
Benthos

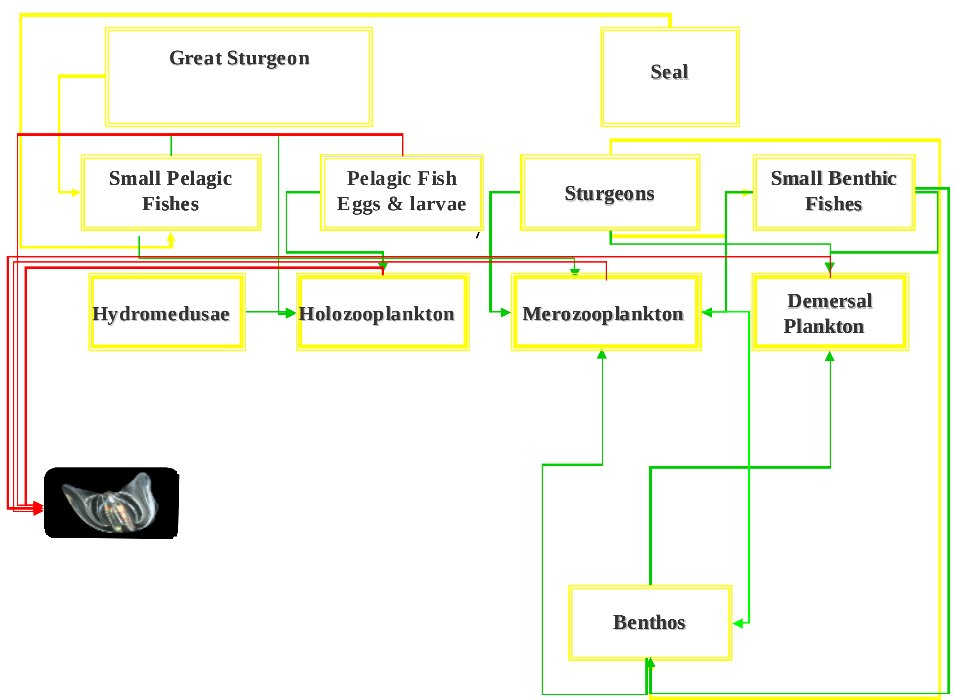


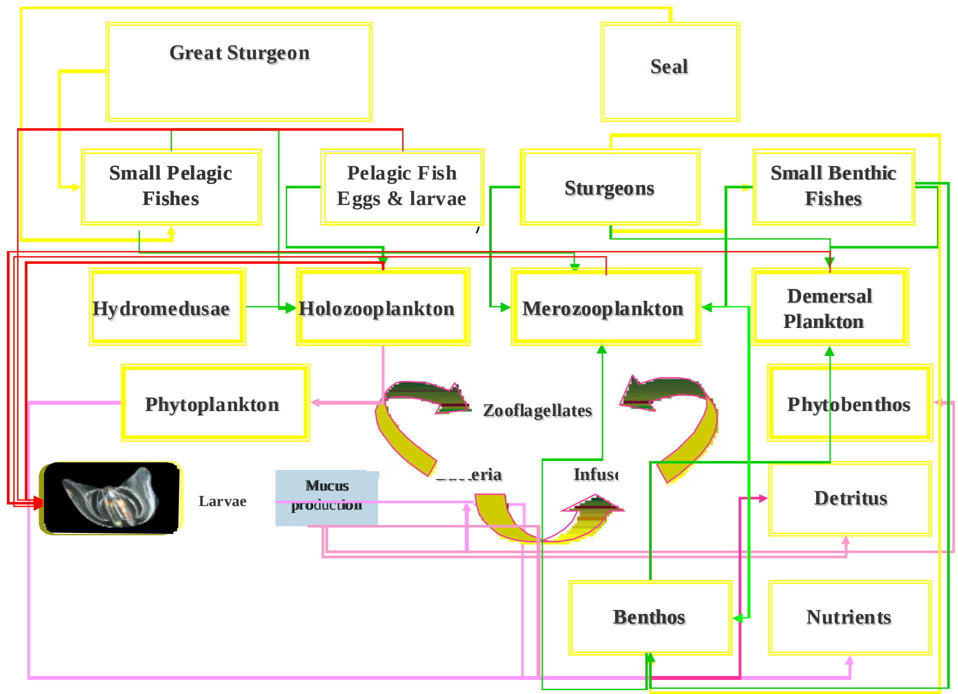
Large Pelagic Predator Fishes
Dolphins
Small Pelagic Fishes
Pelagic Fish Eggs & larvae
Demersal Fishes
Small Benthic Fishes
Indigenous gelatinous
Holozooplankton
Merozooplankton
Demersal Plankton
Phytoplankton
Zooflagellates
Phytobenthos
Larvae
Mucus production
Detritus
Benthos
Nutrients


Large Pelagic Predator Fishes
Dolphins
Small Pelagic Fishes
Pelagic Fish Eggs & larvae
Demersal Fishes
Small Benthic Fishes
Indigenous gelatinous
Holozooplankton
Merozooplankton
Demersal Plankton
Phytoplankton
Zooflagellates
Phytobenthos
Larvae
Mucus production
Detritus
Larvae
Mucus production
Benthos
Nutrients

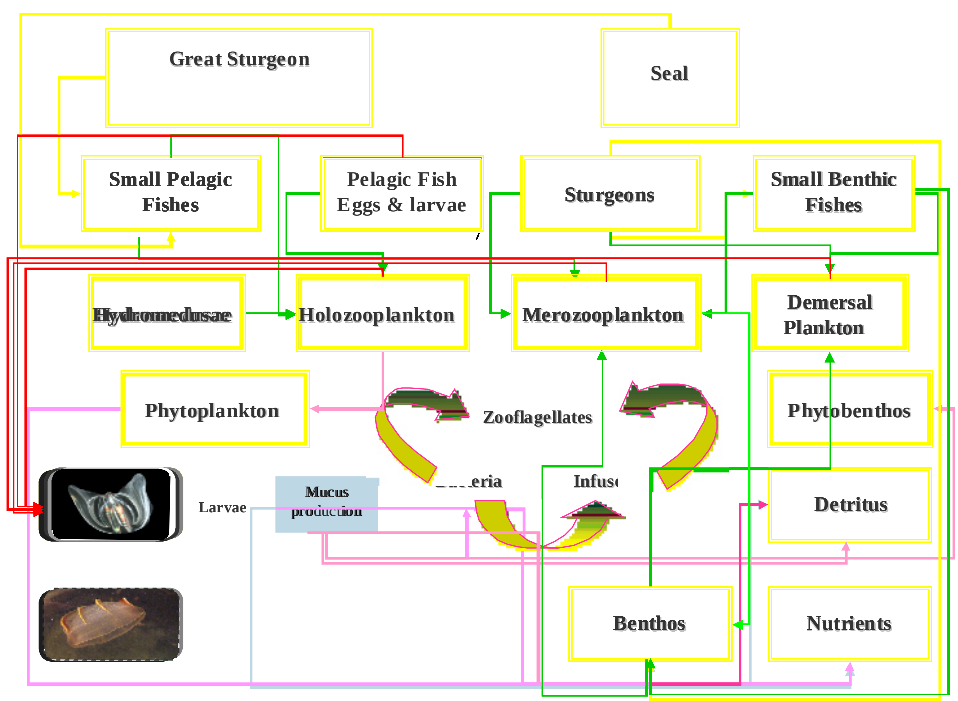

Pelagic Fish Eggs & larvae
Holozooplankton
Merozooplankton
Demersal Plankton


Small Pelagic Fishes
Pelagic Fish Eggs & larvae
Sturgeons
Small Benthic Fishes
Hydromedusae
Holozooplankton
Merozooplankton
Demersal Plankton
Benthos


Great Sturgeon
Seal
Small Pelagic Fishes
Pelagic Fish Eggs & larvae
Sturgeons
Small Benthic Fishes
Hydromedusae
Holozooplankton
Merozooplankton
Demersal Plankton
Benthos



Great Sturgeon
Seal
Small Pelagic Fishes
Pelagic Fish Eggs & larvae
Sturgeons
Small Benthic Fishes
Hydromedusae
Holozooplankton
Merozooplankton
Demersal Plankton
Phytoplankton
Zooflagellates
Phytobenthos
Larvae
Mucus production
Detritus
Benthos
Nutrients



Great Sturgeon
Seal
Small Pelagic Fishes
Pelagic Fish Eggs & larvae
Sturgeons
Small Benthic Fishes
Hydromedusae
Holozooplankton
Merozooplankton
Demersal Plankton
Phytoplankton
Zooflagellates
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Larvae
Mucus production
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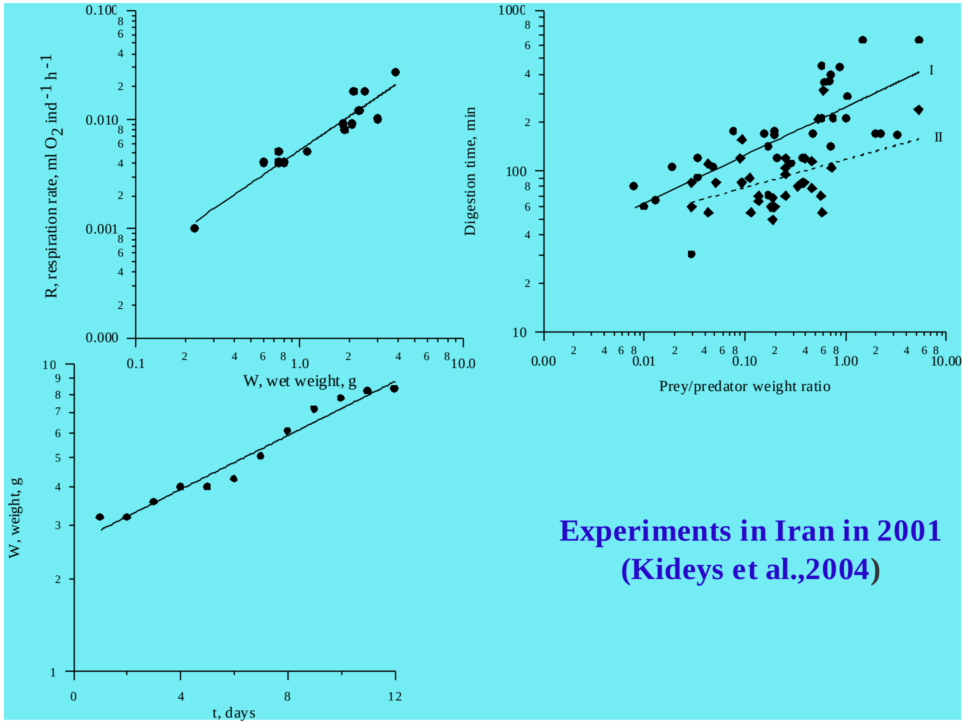
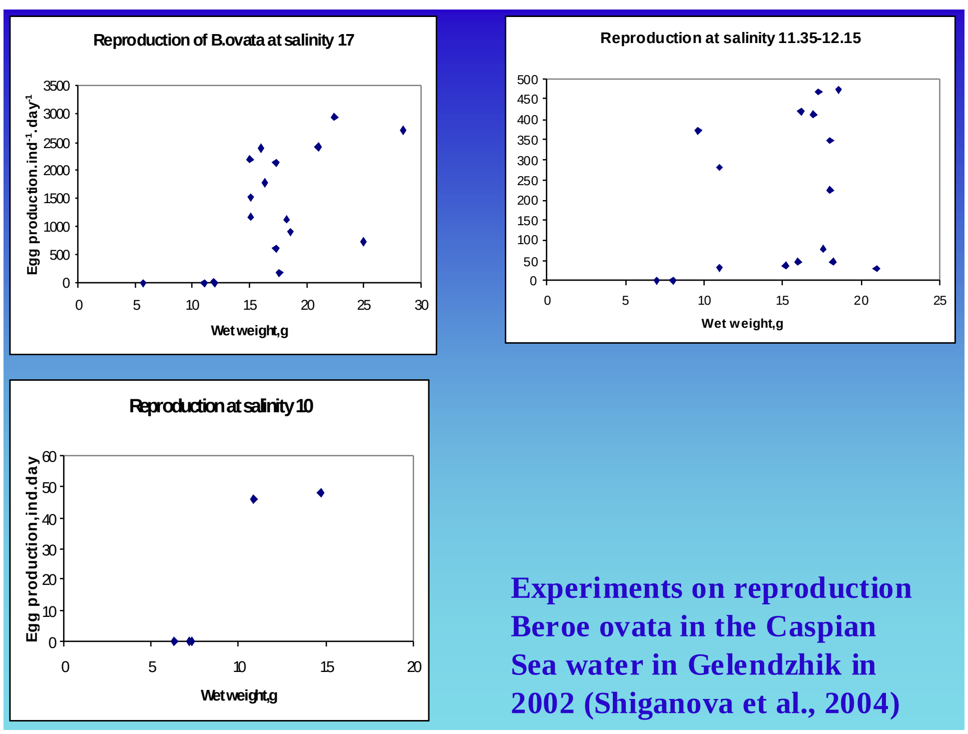
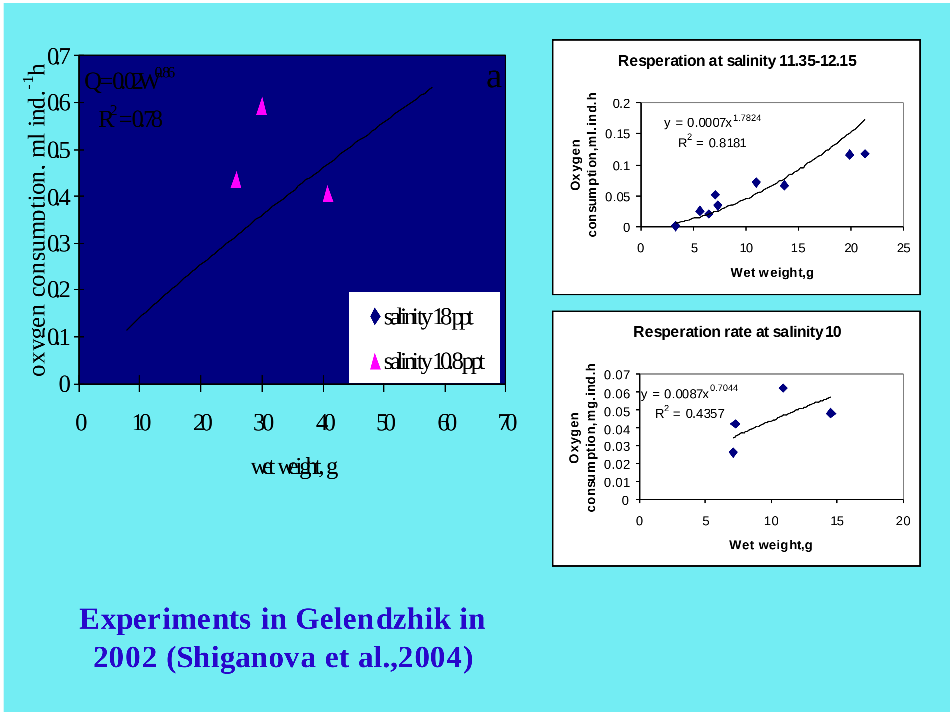
Benthos
Nutrients

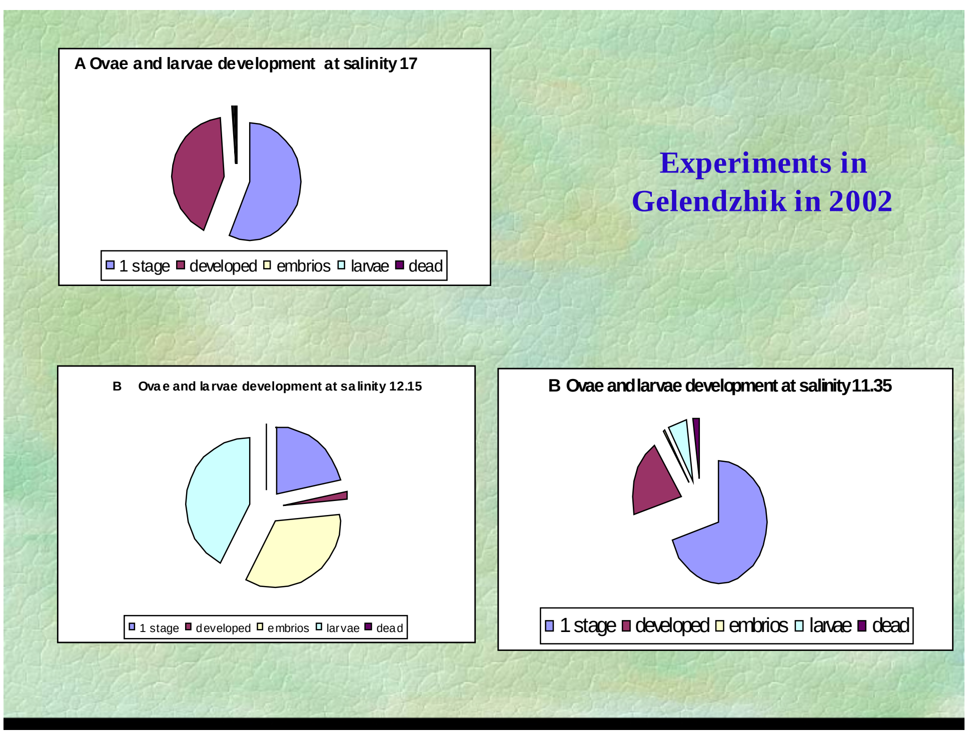


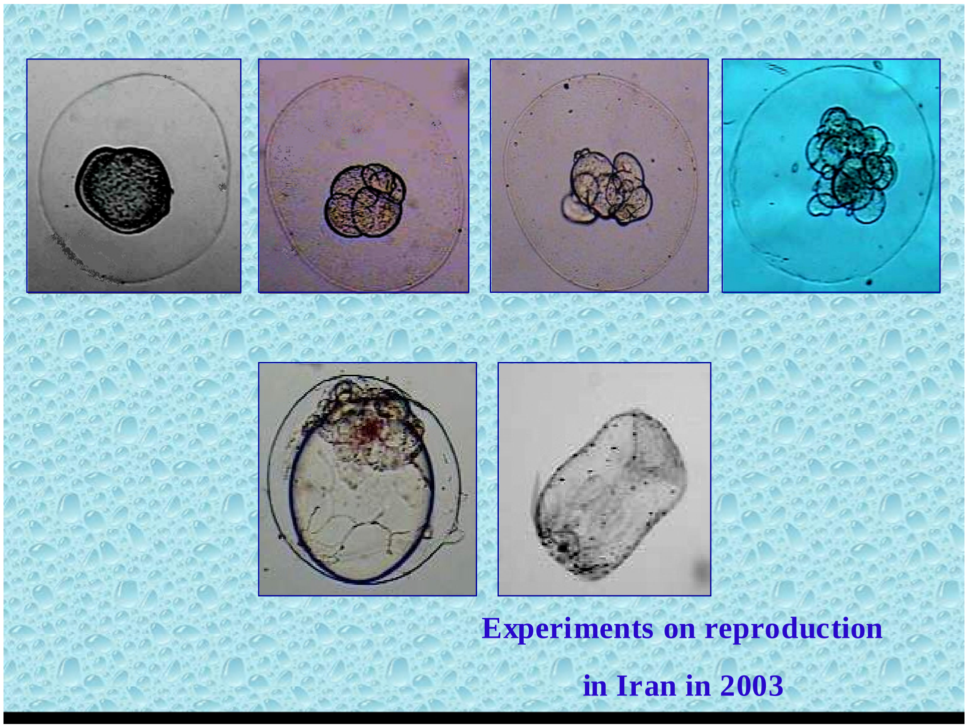
R, respiration rate, ml O_2 ind^{-1} h^{-1}
W, wet weight, g
Digestion time, min
Prey/predator weight ratio
I
II
W, weight, g
t, days
Experiments in Iran in 2001
(Kideys et al.,2004)



Reproduction of B.ovata at salinity 17
Egg production.ind^{-1}.day^{-1}
Wet weight,g
Reproduction at salinity 11.35-12.15
Wet weight,g
Reproduction at salinity 10
Egg production,ind.day
Wet weight,g
Experiments on reproduction
Beroe ovata in the Caspian
Sea water in Gelendzhik in
2002 (Shiganova et al., 2004)


Q=0.03W^0.65
R² =0.78
a
oxygen consumption. ml ind.⁻¹h⁻¹
wet weight, g
salinity 18ppt
salinity 10.8ppt
Resperation at salinity 11.35-12.15
y = 0.0007x^1.7824
R² = 0.8181
Oxygen consumption,ml.ind.h
Wet weight,g
Resperation rate at salinity 10
y = 0.0087x^0.7044
R² = 0.4357
Oxygen consumption,mg.ind.h
Wet weight,g
Experiments in Gelendzhik in 2002 (Shiganova et al.,2004)


A Ovae and larvae development at salinity 17
1 stage developed embrios larvae dead
Experiments in Gelendzhik in 2002
B Ovae and larvae development at salinity 12.15
1 stage developed embrios larvae dead
B Ovae and larvae development at salinity 11.35
1 stage developed embrios larvae dead



Mesocosm experiments in Iran showed:

Beroe ovata **can not eat zooplankton and in absence of *M.leidyi* in the Caspian Sea water conditions just starves and shrinks in size**

Conclusions

- *B.ovata* effectively controls *M.leidyi* population size; it responds by an increase in numbers at high *M.leidyi* availability and maintains a high predation rate as long as prey is abundant. At decreasing *M.leidyi* density, *B.ovata* gradually stops reproducing and finally disappears from the water column.
- *B.ovata* is a specialized carnivore, which consumes exclusively zooplanktivorous ctenophores, in the case under examination *Mnemiopsis leidyi* and *Pleurobrachia pileus*, and cannot digest zooplankton, fish larvae or other groups of gelatinous animals found in the Black and Caspian Seas. Special investigations were conducted to study digestive enzymes of *B. ovata*. It was found that digestive enzymes of *B.ovata* and *M.leidyi* are greatly differ. *B.ovata* does not have chitinaza, which enable to digest carapaces of Crustacea, while *M.leidyi* does have this enzyme .

Beroe ovata can live in the water with salinity less than in the Black Sea and probably lowest salinity for survival is salinity more than 7 .

Physiological evidence suggests that in Caspian water with salinity 10-13‰, *B. ovata* reproduces, grows and ingests *M. leidyi*. Released to the sea, it is therefore expected that it will decrease *Mnemiopsis* abundance sharply.

Beroe will be able to control *Mnemiopsis* population in the most abundant its habitats – in the Middle and Southern Caspian Sea.

The positive example of the recovering of the Black Sea ecosystem should be taken into consideration to save unique for fish resources the Caspian Sea.

Positive impacts to be expected

•Annual biomass and abundance of *M. leidyi* will decreased. Duration of the *M. leidyi* impact will be shorten to not more than two months (July-August) and the impacted area will decrease; probably *M. leidyi* will no longer reach the North Caspian, except in isolated individuals.

•If the start *of B. ovata* development in the Caspian turns out to be sooner than in the Black Sea, its effect on *M. leidyi* may even be faster.

The following key results could be expected from a successful *B.ovata* introduction:

•-*B. ovata* will only feed on *M. leidyi*, because no other ctenophore species is present

•-In the short term, the depletion of zooplankton, including meroplankton, ichthyoplankton and demersal plankton sufficiently decrease to allow a restoration of its density, biomass and species diversity, especially of copepods.

•-The chain of events will continue: because their zooplankton food is restored to exploitable levels, in two years one can expect improvements in small pelagic planktivorous species, first of all the short cycle kilka stock. Caspian seal, in its turn, if not extinct, will benefit from restored kilka stocks, and recover its previous food sources.

Risk assessment in the case of introduction

- **1. *Beroe* is not able to develop in the Caspian Sea**
- An unlikely eventuality, because the experiments described above convincingly showed that *Beroe ovata* can live in the Caspian Sea water, feed on *M.leidyi* with high ingestion rate, growth and reproduce.
- **2. *Beroe* will shift prey and feed on edible zooplankton, fish eggs and larvae**

Mesocosm experiments proofed that it will not happened.

- **3.*Beroe ovata* will spread to rivers and international waters**

The Caspian is a closed brackish water body, from which there is no escape for *Beroe*, since it dies at salinities below 4‰ , and rarely survives at 7 ‰

- **4.Diseases and parasites of *Beroe* spp**

Thus microbial fauna of Beroe in the Black Sea is very poor and harmless. Most of representatives of them inhabit also in the Caspian Sea. *Trichedina ctenophora* which was found in Beroe tussue is harmful only for Ctenophore species.

Estimations of economic loss of Russian fishery in the Caspian Sea resulted in Mnemiopsis invasion for 2000-2002 (Data of CaspNIRKH)*

Drop kilka catches (thous.tons)	Decrease of income of fish meal, mln US dol .	Decrease profit, mln US dol	Cutting of payings in the budget mln US dol.
254,2	185,8	43,2	46

* data were compared with level of 1999 .

Acknowledgment

Creation of EIA was supported by CEP and IFRO

Experimental work in Iran was support by CEP and IFRO in 2001,2002 and by IFRO in 2003

Experiments were successful due to organization of transportation individuals of *B.ovata* by Dr. A.Kideys in Iran

Most of data on the Black Sea were obtained in the joined cruises with the Azov Sea Institute for Fisheries

Most of data on the Caspian Sea were obtained in the joined cruises with the Caspian Sea Institute for Fisheries

Some of investigations were supported by Russian Ministry for Science and Technology Federal target program "The natural resources of the World Ocean"

Development of the Open Information System on Aquatic Invasive Species for the Ponto-Caspian Region

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Development of the open databases and information systems on alien invasive species is needed for support for management and control efforts. Internet-based information systems may serve as main tool of wide dissemination of information on taxonomy, biology, environmental impacts and possible control measures of invasive species. International legal regime requires governments and other relevant organizations to support the creation and maintenance of the databases, information systems and interoperable distributed network of databases for compilation and dissemination of information on alien species for use in the context of any prevention, introduction, monitoring and mitigation activities (Decision VI/23 of the 6th Conference of the Parties of the Convention on Biological Diversity 2002).

There is an urgent need of the open information on aquatic invasive species for the Ponto-Caspian area, specifically for the invasive ctenophore *Mnemiopsis leidyi* and its predator *Beroe ovata.* This region is particularly heavily affected by species invasions: the list of exotic species includes 59 names for the Black Sea, and 39 for the Caspian Sea (Zaitsev & Öztürk 2001). In the early 1980s, the Atlantic ctenophore *Mnemiopsis leidyi* was introduced with ballast water into the Black Sea, and by the late 1990s it had spread to the Mediterranean and Caspian Seas. The invasion of *Mnemiopsis* resulted in a drastic decline in the anchovy fishery in the Ponto-Caspian region with huge economic losses, estimated in hundreds of million of US dollars per year. Currently, the unique biodiversity of the Caspian Sea is also under serious risk, with numerous species facing extinction (Caspian Environment Programme 2002).

On the other hand, the Ponto-Caspian region is serving as an important donor area of harmful aquatic organisms to other parts of Europe and worldwide. During the last two decades, several invasive species of the Ponto-Caspian origin have been introduced into the Baltic Sea with the ballast water of ships, including cladoceran *Cercopagis pengoi* which is considered harmful in the Baltic (Leppäkoski et al. 2002a). The biodiversity of inland running and stagnant waters of Europe and the Great Lakes of North America too is seriously endangered by the introduction of Ponto-Caspian species. Some of the most harmful of those include the zebra mussel *Dreissena polymorpha*, a number of amphipod and mysid species, and the fishhook waterflea *Cercopagis pengoi* (Leppäkoski et al. 2002b).

During 1st Phase of the Caspian Environment Programme (CEP), the members of the CEP Regional Invasive Species Advisory Group had undertaken first efforts in development of the relevant information system. At present the information system, hosted by the Regional Biological Invasions Centre (RBIC) at St.Petersburg, Russia (Regional Biological Invasions Centre 2004), includes detailed profile on *Mnemiopsis leidyi* with information on taxonomy, biology, invasion histories, environmental impacts and illustrated by the distribution maps, generated by the Geographic Information System “INVADER” (Shiganova & Panov 2002).

RBIC-based information system on *Mnemiopsis* is linked to the other on-line sources of information on this species, including those available on the web sites of CEP (Caspian Environment Programme 2002). These informational resources may serve as a basis for the development of the regional information/early warning system on aquatic invasive species.

Also, a demonstration version of the Caspian Sea Biodiversity Database (CSBD) has been
developed during 2001-2002 in the framework of the Caspian Environment Programme, and posted on the CEP web-site since June 2002. The CSBD exists in English and Russian language versions, and currently includes entries on 36 aquatic species, both native (30 species) and alien (6 species) in the Caspian Sea ecosystem (Caspian Sea Biodiversity Database 2002). Entries on alien species in the database include information on the species' taxonomy, their distribution and biology, bibliographic references, and illustrated by figures of the organisms involved. Entries on *Cercopagis* and *Mnemiopsis* include Internet-links to entries on these species in the Regional Biological Invasions Centre Information System (Panov 2004).

The Regional Biological Invasions Centre Information System *(*RBIC*),* hosted by the Zoological Institute of the Russian Academy of Sciences in St. Petersburg, is a new development of the Group on Aquatic Alien Species (GAAS) web-site, which initially was opened in 1999, and at that time already included first versions of entries on two Ponto-Caspian invasive species, *Dreissena polymorpha* and *Cercopagis pengoi* (Panov 1999). Currently RBIC is serving as the pan-European Clearinghouse on invasive alien species (both aquatic and terrestrial), and as a web portal, providing access to the Internet-based information resources on invasive species research and management in Europe and worldwide (Regional Biological Invasions Centre 2004). The development of the Geographic Information System "INVADER" as an international database on the Internet is one of RBIC's priorities. Currently, online version of this GIS "Invasive Species of the Baltic Sea", with comprehensive geo-referenced information on the distribution of some Ponto-Caspian invasive species in the Baltic region is a part of the HELCOM-supported project on development of the open informational resources on alien species for the Baltic Sea (Regional Biological Invasions Centre 2004).

Online geo-referenced distribution maps of selected invasive species, including *Mnemiopsis leidyi*, *Dreissena polymorpha* and *Cercopagis pengoi*, along with detailed descriptions of their taxonomy, invasion histories, biology, and environmental impacts are available at the RBIC Illustrated Database of the Aquatic Invasive Species of Europe, interlinked with the Baltic Sea Alien Species Database, the Global Invasive Species Database and the Caspian Sea Biodiversity Database. The entry on *Mnemiopsis leidyi* provides an example of a comprehensive and user-friendly online information system on the invasive species, linked to other Internet-based sources of information (Shiganova & Panov 2002). The entry on *Mnemiopsis* in the RBIC Illustrated Database is already serving as an open information system on *Mnemiopsis* for the Ponto-Caspian Region, and is updated on a regular basis (Panov 2004).

At present, open information on aquatic invasive species alien to the Ponto-Caspian Region (or introduced from the Ponto-Caspian Region), is located in several online regional and international databases and information systems, linked, within the World Wide Web, with the Regional Biological Invasions Centre serving as web portal, and providing links to these sources (Fig. 1). However, at present available online information is not sufficient for management purposes, such as the prevention of introductions, control or eradication of invasive alien species established in the Ponto-Caspian Region. The development of the regional online information system on aquatic alien species (all alien species in novel ecosystems should be considered as potentially invasive) as a principal management tool should be considered as one of the regional priorities. Considering the significance of the Ponto-Caspian Region as an important donor area of invasive species for the Baltic Sea region and worldwide, such a regional information system should be a part of developing European and global invasive species informational networks. Integration of the Ponto-Caspian regional information system on aquatic alien (invasive) species into the global network of relevant databases will ensure its effective service as an early warning system for other regions and as a tool for risk assessment of harmful species introductions from the Ponto-Caspian to the potential recipient regions. In order to ensure inter-operability, the regional Ponto-Caspian information system on aquatic alien organisms should be build on the basis of the information technologies developed for the adjacent Baltic Sea Region, including online version of GIS "INVADER" (Panov 2004).

Acknowledgements

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References

Caspian Environment Programme, 2002. *Mnemiopsis leidyi* in the Caspian Sea. http://caspianenvironment.org/

Caspian Sea Biodiversity Database, 2002. V. N. Belyaeva (ed) http://www.caspianenvironment.org/biodb/eng/main.htm

Decision VI/23 COP6 of the Convention on Biological Diversity (2002) http://www.biodiv.org/decisions

Leppäkoski, E., S. Gollasch, P.Gruszka, H. Ojaveer, S. Olenin & V. Panov, 2002a. The Baltic—a sea of invaders. Can. J. Fish. Aquat. Sci. 59: 1175-1188

Leppäkoski, E., S. Gollasch & S. Olenin (eds), 2002b. Invasive Aquatic Species of Europe. Distribution, Impacts and Management. Kluwer Academic Publishers, Dordrecht, 583 pp

Panov, V. E., 1999. GAAS: Group on Aquatic Alien Species at the Zoological Institute in St.Petersburg, Russia. Biological Invasions 1: 99-100

Panov V. E. 2004. Internet-based information resources on aquatic alien species relevant to the Ponto-Caspian Region. In: H.J. Dumont, T.A. Shiganova and U. Niermann (eds), Ponto-Caspian aquatic invasions, Kluwer Academic Publishers: 257-269.

Panov V., & S. Gollasch. 2004. Informational resources on aquatic alien species in Europe on the internet: present developments and future perspectives. In: Vanden Berghe E., M. Brown, M.J. Costello, C. Heip, S. Levitus and P. Pissierssens (Eds). Proceedings of "The Colour of Ocean Data" Symposium, Brussels, 25-27 November 2002, IOC Workshop Report 188 (UNESCO, Paris). X + 308 pp. (p. 115-124).

Regional Biological Invasions Centre, 2004. V. Panov, M. Dianov, V. Shestakov & A. Lobanov (eds) http://www.zin.ru/rbic/

Shiganova, T. A. & V. E. Panov, 2002. *Mnemiopsis leidyi* A. Agassiz, 1865. Entry to the Illustrated Database, Regional Biological Invasions Center Information System. http://www.zin.ru/projects/invasions/gaas/mnelei.htm

Zaitsev Y. & B. Öztürk (eds), 2001. Exotic species in the Aegean, Marmara, Black, Azov and Caspian Seas. Published by Turkish Marine Research Foundation, Istanbul, Turkey, 267 pp

Figure captions

Figure 1. Present links between available regional and international online informational resources on aquatic alien species, relevant to the Ponto-Caspian Region (1 - Regional Biological Invasions Centre Information System, 2 - Caspian Sea Biodiversity Database, 3 - CIESM Atlas of Exotic Species, 4 - Baltic Sea Alien Species Database, 5 - Directory of Non-native Marine Species in British Waters, 6 – FAO Database on Introductions of Aquatic Species, 7 - *Caulerpa taxifolia* Database, according Panov 2004)

IMPACTS OF MNEMIOPSIS LEIDYI ON ZOOPANKTON POPULATION IN THE SOUTHERN CAPIAN SEA

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ABSTRACT:

This study was conducted zooplankton population changes in the southern Caspian Sea in the depth of 10 M in 1995-2002. In 2002, the frequency of zooplankton has been decreased, which impacts on Copepoda and some other Meroplankton.

In recent years, introduction of *M. leidyi*, which is the predator of zooplankton has been increased rapidly in the southern Caspian Sea and made up a huge amount due to fast reproduction .Its frequency was 46-18130 ind./m2 and the biomass was 1/6-1829/6 gr/m2. Zooplankton frequency will decrease due to increase of *M. leidyi* in warm months, till November ctenophore will decrease and zooplankton frequency will increase again.

Key words: Zooplankton, Caspian Sea, *Mnemiopsis leidyi*, Copepoda.

INTRODUCTION:

It is obvious that introduction of new species accompany always with changes in ecosystems, and even results to decrease of some the commercial species. *M. leidyi* is one this species that transported by ballast waters from United states coasts to the Black Sea (Vinogradov et al. 1989 , Zaystev et al.1998) and it was introduced suddenly to this aquatic ecosystem in 1999 (Mutlu,1999) and then introduced to the Caspian Sea and recently it spreads in huge amount in the southern Caspian Sea.

M. leidyi effected on the frequency and biomass of many organisms in the Black Sea (Malyshev et al.1992) ,though decreased rapidly on Copepods population as well (Kent,1980). In the Black Sea waters, this new invader decreased rapidly on zooplankton and in 1989-90 due to blooming caused sharp decrease of main fish catch species such as Anchovy *Engrawlis encrosieholcy* (Shiganova, 1998).

In this study, *M. leidyi* and zooplankton frequency will compare and then its effects on zooplankton population during 1994-95,1997-98 and 2001-02 will be investigated.

MATERIAL AND METHODS

Sampling of zooplankton was conducted in 3 regions such as Nowshar (36^{o} 49-$51^{/}$ $29^{//}$) , Babolsar ((36^{o} 44-$52^{/}$ $39^{//}$) and Amirabad ((36^{o} 52-$53^{/}$ $6^{//}$) from July 2001 to April 2002. Zooplankton were collected by vertical tows with 100 Mic mesh and 36 cm mouth area. After gathering , they were preserved in 4% formalin (Wetzel and likens, 1291) and then transported to laboratory for identification and measurements. For counting , we used inverted microscope with Newel modification method (Newel et al. 1997).*Mnemiopsis leidyi* was sampled using a 500 micron mesh sized METU net (diameter 50 cm with large bucket suitable for *Mnemiopsis*). Samples was obtained via vertical towing from the bottom to the surface for all stations. At the end of each tow, the net was washed from the exterior, and the cod end was passed into a container immediately to enumerate ctenophores by naked eye. The ctenophores were sorted in length groups of 0-5 mm, 6-10 mm, 11-15 mm and so on, for size measurements. . Individual weighing of these animals was not practical at sea. Weights of these animals were therefore calculated from size measurements using a conversion formula which was obtained individual length (using a ruler for the lobed length) and weight measurements (using a digital balance with a sensitivity of 0.001 g). Length groups were thus converted to weight by using the equation obtained.

Results

Investigation of zooplankton from 1994 to 2002 in the southern Caspian Sea has shown that zooplankton population decreased rapidly in summer and autumn and its population in 2001 ratio to 1998 decreased 2 to 45 times, respectively. This decrease was mainly on copepods population in which consisted main group of zooplankton in the Sea.

Copepods population in 10 M depth was under influenced of *Acaria spp.* And its frequency in summer 2001 consisted of more then 90 per cent of copepods. Copepods and *Acartia spp.* biomass formed half of zooplankton population in summer and the rest of zooplankton were made up of Bivalvia larvae such as *Lamellibranchiata* larvae, but in 2001 its species composition (diversity) changed and more than 95 per cent of zooplankton population included of copepods and *Acartia spp.*(table 1) *.*

Sharp decrease of zooplankton in recent years was due to introduction of new invader *M. leidyi* in the Caspian Sea, though with increasing of *M. leidyi* in 1998 effects on decreasing of zooplankton population. In 2001-02 *Mnemiopsis* and zooplankton abundance and biomass data shown that with the increasing of Ctenophore in August the biomass of zooplankton has been decreased and in the late of November the amount of *Mnemiopsis* will decrease so the zooplankton biomass as well as its frequency will increase (fig.1).

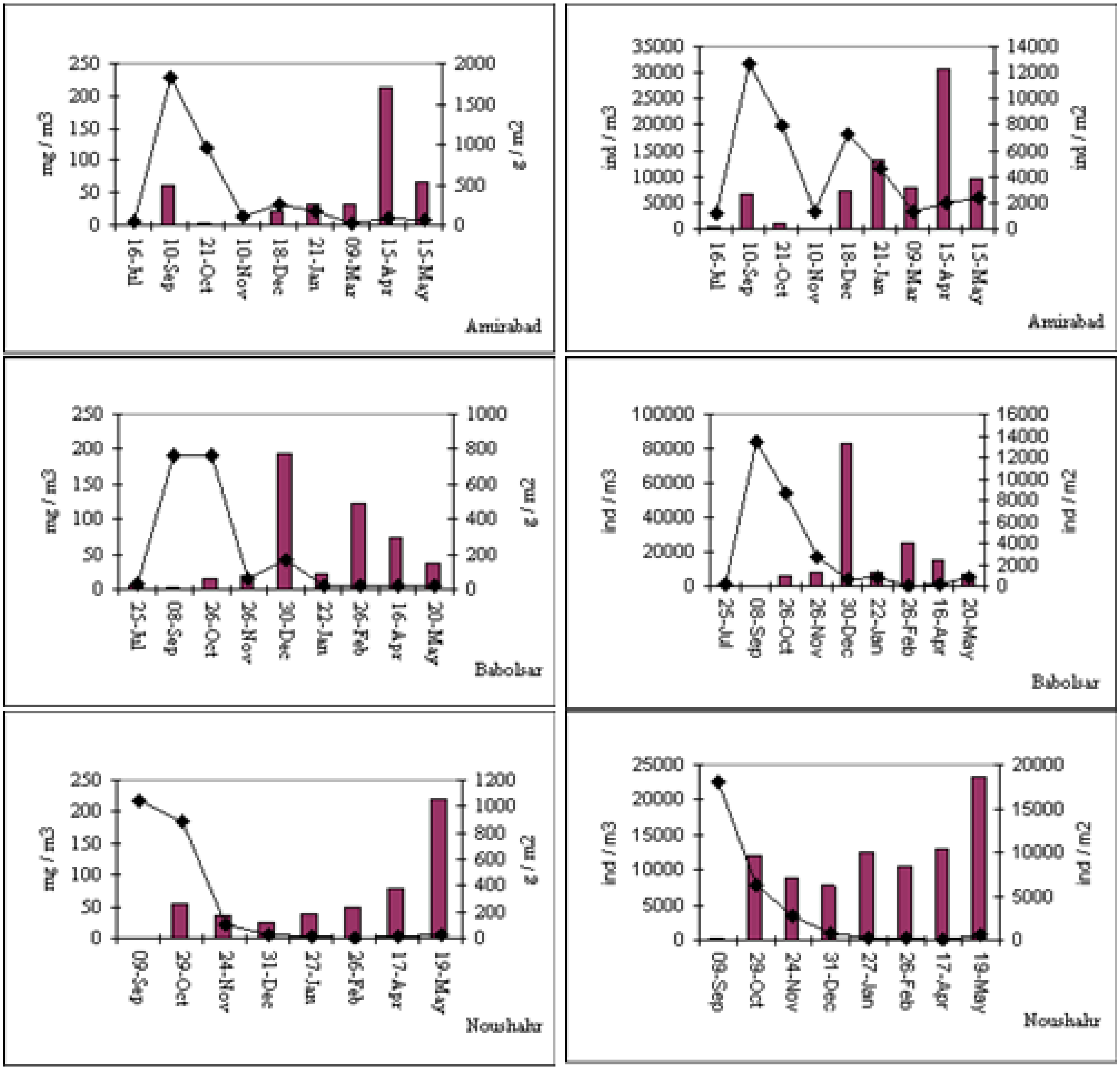

Fig 1: Zooplankton and *M. leidyi* changes at the 10 M depth of the southern Caspian Sea in 2001-02. Zooplankton

Seasonal changes of zooplankton shown that Amirabad had low biomass in 1994-2000 but Nowshar was the minimum in 2000. In previous years, copepods and *Acartia spp.* had the main roles in zooplankton biomass in summer whereas in 2000 almost zooplankton population follows only with *Acartia* which indicates the sharp changes of zooplankton diversity.

M. leidyi not only effected on frequency and biomass of zooplankton but also during 1994-95 , 1996 and 1998-9 decreased the species composition as 26,28 and 26 respectively, which the maximum diversity observed in Cladocereans. But ,in 2000 ,

Table 1: Zooplankton, Copepods, *Eurytemora*, *Acartia* biomass (mgr/m^3) at the different sampling stations of the southern Caspian Sea (10M).

		Summer		Autumn		Winter		Spring	
Organism	year	Average	Std.	Average	Std.	Average	Std.	Average	Std.
Acartia	1994-95	124.67	105.94	67.89	54.66	10.07	10.43	3.54	2.72
	1996	43.76	14.66	129.19	147.69	38.84	29.87	24.98	31.46
	1998-99	26.86	14.48	29.42	14.60	1.21	1.16	1.20	0.51
	2000-01	14.57	13.67	12.05	14.08	16.32	6.17	22.35	10.23
Eurytemora	1994-95	0.08	0.14	1.50	2.47	10.83	17.56	1.52	0.50
	1996	0.05	0.08	2.27	2.86	10.41	16.67	5.88	10.14
	1998-99	0.30	0.49	0.02	0.02	0.01	0.02	0.00	0.00
	2000-01	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
Copepoda	1994-95	133.00	110.01	82.38	63.81	33.77	29.28	10.33	8.08
	1996	49.64	13.55	176.23	124.97	61.70	50.12	40.73	29.96
	1998-99	44.31	11.76	59.97	31.31	4.71	3.68	7.99	3.95
	2000-01	15.93	14.65	23.19	19.43	50.82	34.29	39.59	12.15
Zooplankton	1994-95	238.67	149.50	199.19	163.39	92.39	102.62	42.00	16.52
	1996	158.85	113.78	207.20	116.31	209.10	186.60	119.16	95.06
	1998-99	130.48	114.49	77.28	52.62	26.79	30.56	99.27	34.25
	2000-01	15.96	14.68	23.24	19.42	60.44	45.70	114.65	52.19

only one species of the further order resisted and totally 12 species remained (Table 2).Before introduction of *M. leidyi* into the Caspian Sea , *Eurytemora spp.* consisted a lower group of zooplankton at 10 M depth but in 2000 this species was disappeared and even the biodiversity reached to half as compare to previous years.

Table 2: Frequency of main zooplankton groups in the southern Caspian Sea at 10 M.

Organisms	1994-95	1996	1998-9	2000-01
Protozoa	2	3	4	1
Rotatoria	2	4	4	3
Cladocera	7	12	13	1
Copepods	5	5	5	4
Others	4	5	3	3
Zooplankton	20	29	29	12

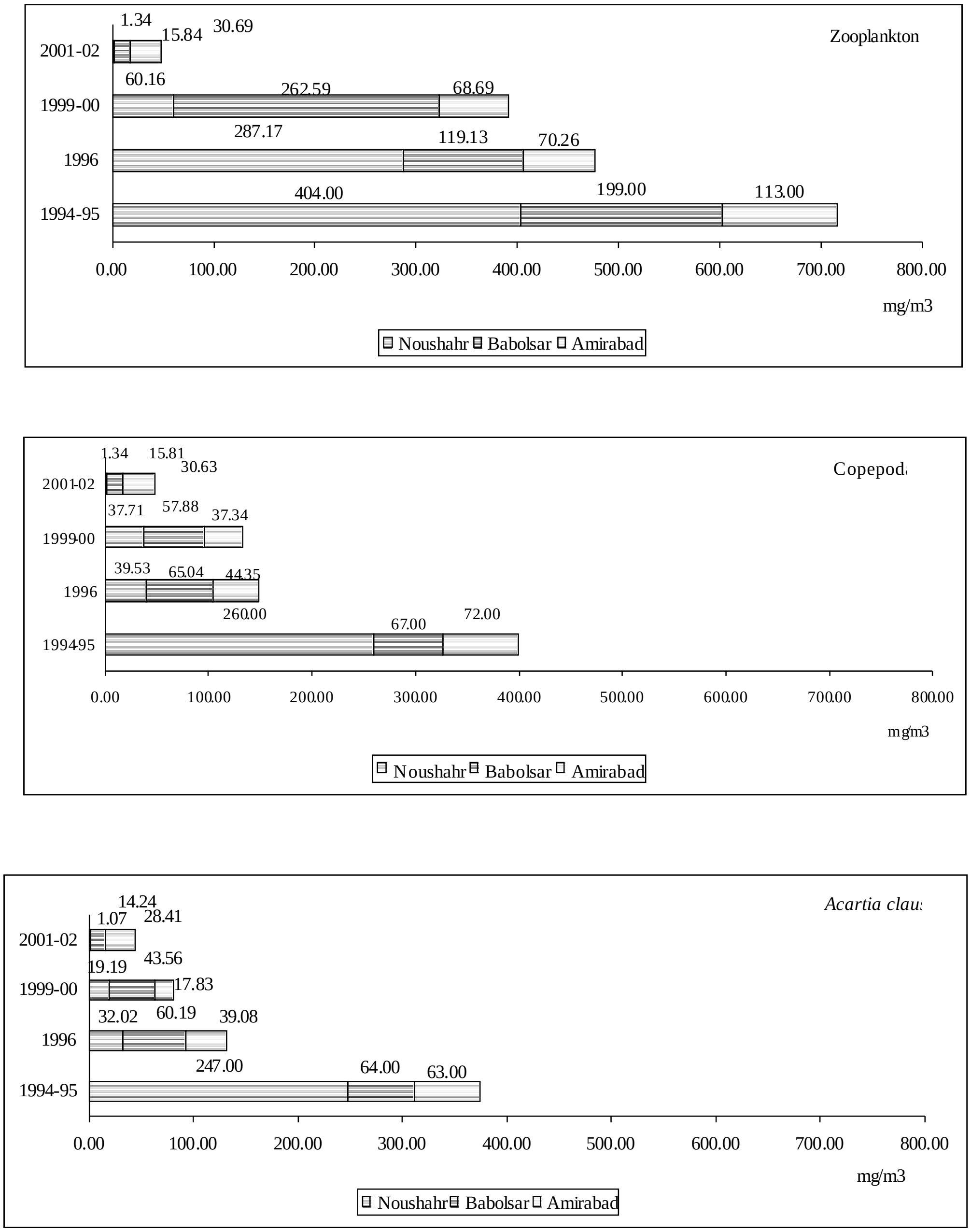

Fig2. The frequency of zooplankton, Copepoda and *Acartia clausi* in summer in the suthern of Caspian sea (10m).

DISCUSTION:

As human effects on recovery of aquatic ecosystems, unfortunately interferes to inlet the introduction of new invader as well, and even prepared the condition for introduction of new species so far. According to the hypothesis, introduction of organisms from the Black Sea to the Caspian had been regarded to 8 thousands years ago which inlet from wooden boats. From that time till now, different species introduced into the Caspian Sea such as N*ematoda, Cerastoderma lamarcki , Mytilaster lineatus , Rhizosolenia calcar avis (*Diatoms)*, Abra ovata ,Nereis diversicolor*(Zaitasev et al. 2001).At present lobate Ctenophore *M. leidyi* comfirmed a big population in the Caspian Sea which effected on other organisms .Much reproduction of this animal occurs in warm months and its biomass depends on growth period.*M. leidyi* population follows temperature so it decreases in late November and early December and the zooplankton population is vise versa.

In this study it shown that when temperature reaches at 20 ^{oC} ,digestive set is able to digest 24 copepods , 24 Cladocera , 12 Lamellibrachiata larvae , 4 fish larvae and 12 of other organisms in 24 h (Zaittasev et al. 2001).Biodiversity investigation in the Caspian Sea shown that species composition of Cladocera in recent years decreased and only one species is being observed in 10 M. So from 29 species in 1994 it reaches to 12 species in 2001-02.

Zooplankton biomass in summer time decreased rapidly and reached to 1.34 mg/m^3 in Nowshar whereas ctenophore biomass was more than 1000 gr/m^2 . *M. leidyi* will increase and reproduce when zooplankton abundance is high(Larson, 1987 , Reeve, 1980 and Kermer, 1976).Though it is obvious that *M. leidyi* effects on zooplankton population (Kermer, 1976 , mayer, 1912).

Due to feed on zooplankton, *Mnemiopsis* abundance will increase as zooplankton population decrease to half in summer and autumn and even in some regions such as Nowshar it decrease to 45 times as compare to last summer (in 2001).

In 2001-02 , zooplankton population shown that it follows 95 percent of copepods and the dominant species of copepoda *Acartia spp.*. This species was dominant at 10M whereas *Eurytemora spp.* was dominant at 20M and also it was much more abundance at 50 and 100M (Rowshantabari, 2000). In spite of this fact, unfortunately we couldnot observe any *Eurytemora spp.* in our samples. *Lamellibranchiata larvae* also decreased in spite of its abundance in previous summer was high. In 2001-02 data from 3 regions, Amirabad, nowshar and babolsar at 10 M shown that frequency and diversity of zooplankton was changed with the introduction of new invader *Mnemiopsis* as compare to previous years.
Figure 2: Seasonal changes of zooplankton ,copepods and *Acartia clausi* in summer of the southern Caspian Sea (10M).

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REFRENCES:

Kremer ,p. 1976. Population Dynamics and ecological energetics of a pulsed zooplankton predator ,the ctenophore *Mnemiopsis leidyi* .Estuarine processes V:N P.197-15

Kent M.1980. Occurrence and predation by *Mnemiopsis leidyi* in Barnegat Bay, New Jersey .Estuarine and Coastal marine science 10, 393-402

Larson, R .J. 1978. In situ feeding rates of the Ctenophore *Mnemiopsis leidyi.*Estuaries. V.10.p.87-91.

Malyshev, V.I., A.G, Arkhipov . 1992. The Ctenophore *Mnemiopsis leidyi*in the Western Black Sea.Hydrobiology.J.28(1). 34-39.

Mutlu ,E.1999. Distribution and abundance of ctenophore and their zooplankton food in the Black Sea. *Mnemiopsis leidyi* ,Marine biology, 135: 603-613.

Mayer A.G.1912.Ctenophore of the Atlantic coast of North America. Carnegie Institute publication, 162.j.b. Lippincott Co, Philadelphia. D.58.

Main, R.J11928. Observation of the feeding mechanism of a ctenophore *Mnemiopsis leidyi* ,biological bulletin 55,69-78.

Newel, G.E. and R.C. Newel ,1997.Marine plankton. Hutchinson of London.

Reeve, M.R.1980. Comparative experimental studies on the feeding of chaetognaths and ctenophores.J.plankton Res. V.2,p.381-393.

Reeve ,M.R.,M.A. Syms and P. Kremer .1989. Growth dynamics of a ctenophore (*Mnemiopsis*) in the relation to variable food supply.J . plankton Res.V,11 No. 3, 535-552.

Shiganova, T.A.1998. Invasion of the Black Sea by the Ctenophore *Mnemiopsis leidyi* recent changes in pelagic community structure.Fisheries Oceanography_GLOBEC special Issue,Ed.Steeve Coombs.,P.305-310.

Wetzel, R.G. andG.E. Likens.1991. Limnological analyses. Springer verlag.

Zaitsev, V.F., A.A. Korapov., A.E.Meliakina ., A.F.Sacolski. 2001. The ecological effects of the enterance of *Mnemiopsis* into the Caspian Sea. The technical and government university of Astarakhan. P.20-60.

Experimental mesocosm : investigation of possibility predation of ctenophore *Beroe ovata* on zooplankton and other preys in addition to ctenophore *Mnemiopsis leidyi* in the Caspian Sea

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1. INTRODUCTION

During the last 30 years, environmental conditions in the Caspian Sea significantly degraded under the impact of various pressures, among which sea level changes and pollution from multiple sources took a prevalent position (Ivanov, 2000; Salmanov, 1999). Additionally, a novel type of anthropogenic impact, that became widespread across the world in recent years, started to affect the Caspian Sea, viz. invasion of the ecosystem by undesirable alien species of animals and plants.

The most dramatic example of such an accidental introduction was the arrival of the ctenophore *Mnemiopsis leidyi* with ship ballast water from the Black or Azov Seas and distribution in the sea in 1999 (Esmaeili et al., Ivanov et al., 2000; Shiganova et al., 2001) .
In 2000 it spread across all areas of the Caspian with a salinity of minimum 4.3 ‰. In 2001 – 2002 it greatly increased in population size, to reach a critical wet mean biomass of about one 1 kg. m^{-2} (55 g m^{-3}) in 2001 (allowing a coefficient 2 to compensate for imperfect catchability), an abundance in excess of the highest values ever recorded in the Black Sea. In 2002 this biomass again doubled (Shiganova et. al., 2003; Shiganova et al., in press).
As showed example of the Black Sea the best candidate that can successfully control Mnemiopsis population size in the Caspian Sea is another ctenophore Beroe ovata
The species of *Beroe ovata* has two outstanding advantages: firstly, it is highly specific in its feeding, so that even its larval stage feeds on *M.leidyi* larvae. Secondly, its reproductive rate

and fecundity are almost as great as that of *M.leidyi*, so that its population can grow at similar rates to its prey (Shiganova et al., 2003).

Representatives of *Beroe* live in the shallows and estuaries of the Mediterranean Sea, and of the tropical and temperate Pacific and Atlantic Oceans. Few species inhabit Arctic Seas (Mayer, 1912; Chun, 1880; Greve & Reiers, 1988; Harbison et al., 1978; Seravin, 1998). All species of beroids are considered to be exclusively feeding on other planktivorous ctenophores; some also consume salps. There is often a trophic linkage between *Beroe* species and planktivorous ctenophores. As a rule *Beroe ovata* and *Mnemiopsis leidyi* form a pair*;* another such pair is constituted by *Beroe cucumis* and *Bolinopsis infundibulum* (a second lobate planktivorous species) (Greve, 1971; Kamshilov, 1960a). Representatives of *Beroe* in their turn serve as food for fish such cod, herring and mackerel (Kamshilov, 1960b).
Beroe is an important link in pelagic food chains, but before its arrival in the Black Sea, comparatively little was known about its biology. *Beroe* significantly affects the population structure of planktivorous ctenophores and thus indirectly modifies the population dynamics of the zooplankters at lower trophic levels. The example demonstrated by *Beroe ovata* in the Black Sea after its introduction and development there is pertinent in this respect.
The Caspian countries had opted to deal with the *M.leidyi* issue by taking strictly biological measures. Specifically, they agreed to introduce *Beroe* ovata as a predator specific to *M.leidyi*, pending the outcome of an environmental impact study of this intended introduction.
In order to understand the feasibility of *Beroe* introduction, as an effective predator on *Mnemiopsis*, into the Caspian Sea, experiments on survival of *Beroe* in Caspian Sea water (12.6 ppt salinity) and on some physiological characteristics (feeding, respiration, reproduction and growth) were performed in Khazerabad laboratory (Mazandaran) on the Caspian coasts of Iran.

Therefore specially designed laboratory experiments were performed in Mazandaran Fisheries Research Center in Iran in 2001-2002.

The main purposes from this research were:

1). Studying survival and tolerance of *Beroe ovata* in the Caspian Sea water
2). Studying ecological physiological characteristics *(*feeding, respiration, growth and reproduction rates) in the Caspian Sea water to determine possibility its introduction into the Caspian Sea (Kideys et al., 2003)
Experiments for investigation of possibility introduction ctenophore *Beroe ovata* to the Caspian Sea from the Black Sea were continued again in 2003 in Iran, Institute for Fishery. A goal of the present study was to test if *Beroe ovata* might feed other preys such as zooplankton in addition to *Mnemiopsis leidyi*.

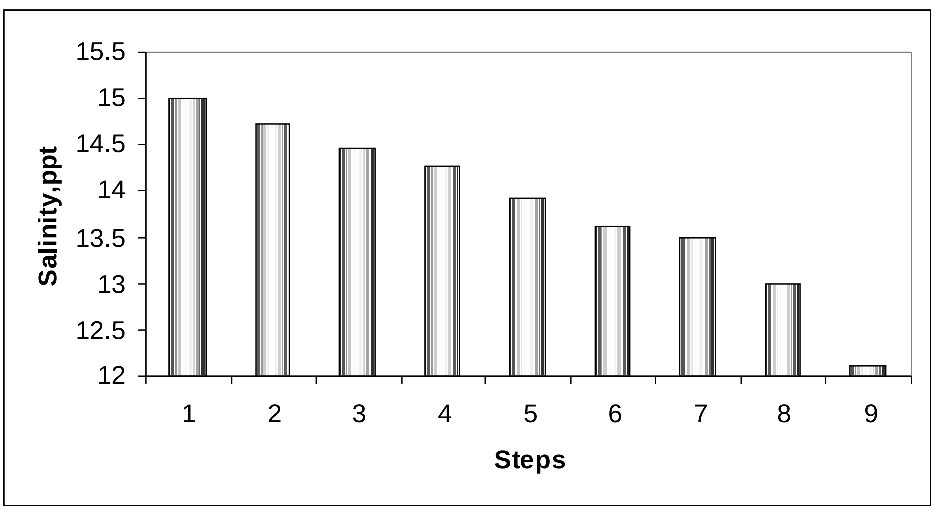
Acclimation to the Caspian salinity.

Individuals of *Beroe ovata* were brought from Sinop (Turkey). One part of individuals were in the Black Sea water with salinity 21. Another one partly acclimated in salinity 15 (partly acclimated in the Caspian Sea water in Sinop). At the moment of arrival we had:
In the Black Sea water with salinity 21:
14 ind. were in good condition, 12 were very bad.

In the sea water with salinity 15.
45 ind. were in very good conditions
14 were worse but still alive.
Acclimation was started in two different salinities.

Individuals in the Black Sea salinity were put in special prepared acclimating system by Mr. Roohi and Dr. A.Kideys. These individuals did not survival.
Beroe in salinity 15 were gradually acclimated in four 9 l aquariums.
First days salinity decreased per 0.2 every 4 hours (Fig.1) when salinity reached 13 all acclimated individuals were placed per 2-3 individuals in 4 liter jags.

Fig.1 Acclimation Beroe for Caspian Sea salinity.



Feeding experiments:

First Beroe were fed in the 2nd day of acclimation, while they were not completely acclimated on 16 September.

Table 1.

Digestion and ingestion at salinity 15-13.7

No Aquarium	Beroe Numbers	Mnemiopsis numbers	Water T (C^0)	Length of fed Beroe (mm)	Number of Disgusted Mnemiopsis	Aquarium disgusted Mnemiopsis (mm)	Duration of ingestion
1	16	5	22	40	1	30	12.30
2	12	5	22	30	1	35	12.30
3	16	5	22	40	1	38	14.00
4	9	5	22	35	1	30	16.00

Table 2.

Digestion and ingestion at salinity 13.0

17 September

No Aquarium 10l	Beroe Numbers	Mnemiopsis numbers	Water T (C^0)	Length of fed Beroe (mm)	Number of Disgusted Mnemiopsis	Length disgusted Mnemiopsis (mm)	Duration of ingestion (hour)
1	16	7	22	40	1	30	2
2	12	6	22	30	1	38	2.30
3	16	8	22	40	1	42	4
4	9	9	22	35	1	35	2.30

Table 3.

Digestion and ingestion at salinity 13.0-12.6

18 September

No Aquarium (4 l)	Beroe Numbers	Mnemiopsis numbers	Water T (C^0)	Length of fed Beroe (mm)	Number of Ingested Mnemiopsis	Duration of digestion (hour)
1	3	10	22	35	1	2.25
2	3	10	22	35	1	2.25
3	3	10	22	40	1	2.25
4	3	10	22	30	1	2.25
				25	1	2.25

Thus we may conclude that Beroe could fed even during acclimation, but time of digestion was much longer of non completely adapted Beroe likely due to stress conditions during adaptation.

Mesocosm experiments.

Mesocosm system was developed in order to estimate as many as possible effects Beroe on the tropic webs of Caspian ecosystem and environment. Therefore we include measurements of

Chemical parameters: O_2 (mg/l) pH, $CaCO_3$, TDS (g/l), Si O_2(mg/l), PO_4(mg/l), NO_3, NO_2, NH_4 (mg/l), EC ms.
Hydrophysical parameters: t ^{0}C, salinity.
Biological parameters:
Phytoplankton
Microplankton (bacteria)
Zooplankton
Mnemiopsis leidyi
Beroe ovata.

Methods.

Mesocosm description.

The mesocosms were conducted in round plastic tanks with volume 300 l. filtrated Caspian Sea water, diameter 90 cm and 60 cm high. Tanks were supply by aeration. Salinity Caspian Sea water was 11. 25 – 12.01‰ in tanks. Temperature during all days of experiment was 23 ^{o}C. Altogether 10 tanks with fresh Caspian Sea water were set up with the following design of testing parameters:

Table 4.

Experimental mesocosm design

	Number of replications	Beroe Numbers in tank	Mnemiopsis numbers in tank	Zooplankton Consentration in tank
Beroe+ zooplankton	4	7		*Acartia tonsa* **Copepodits and adult 2400** Nauplii 7900
Beroe+ Mnemiopsis+ zooplankton	2	7	300	*Acartia tonsa* **Copepodits and adult 2400** Nauplii 7900
Mnemiopsis+ Zooplankton	2		300	*Acartia tonsa* **Copepodits and adult 2400** Nauplii 7900
Zooplankton	2		300	*Acartia tonsa* **Copepodits and adult 2400** Nauplii 7900
Caspian Sea Water	1			

The samples on initial chemical, hydrological parameters were taken immediately after beginning of the experiments. Estimated numbers of biological items were put into each tank (Table 4).

Samples were taken every morning and after estimation concentration itemsin each tank we added zooplankton, Mnemiopsis and Beroe individuals in each tank to keep initial contents of experimental tanks.

Zooplankton and Mnemiopsis sampled with concentration of 8 l water from each tank.

Samples of zooplankton was fixed with 4% formaldehyde and immediately proceeded.

Mnemiopsis individuals were calculated and measured.

Phytoplankton samples were taken in initial concentrations and on third day of experiments.

500 ml was sampled from each tank and fixed with 2% formaldehyde. After homogenization and sedimentation during 24 hours we use 5 ml chambers in order to identification by inverse microscope. We use standard constant indices for estimation of biomass in g/l.

Microplankton (bacteria) was sampled in initial concentrations and on fourth day.

100ml sample was taken from each tank. Than testing solid was prepared with different concentration of bacteria from 10^{-1} to 10^{-4} on the nutrient agar medium and on plate count agar medium. Samples are incubated at temperature 25 –30^0 for 48 hours. The grown colonies of bacteria was calculated.

In addition samples for Beroe reproduction were collected with plastic pipe from the bottom to surface during last five days (even when mesocosm experiment was over).

For chemical parameters examination the following methods were used (Clescert et al., 1986):

1. for oxygen titration after Winkler.
2. T.O.D - titration with EDTA
3. NH^4 - Phonat spectrophotometry method
4. NO_2 - Naphtil spectrophotometry method.
5. NO_3 – Cadmium Reduction, spectrophotometry.
6. SiO_2 – Yellow Complex with Molibdate method, spectrophotometry

Description of mesocosm experiment.

The main focus of our mesocosm was identification of possibility Beroe individuals to feed zooplankton or other items from the Caspian Sea in addition to Mnemiopsis in condition when Mnemiopsis as a prey is not available. Therefore four replications were taken with Beroe and trophic zooplankton (*Acartia tonsa* and its copepodits and nauplii). For control this trophic feedback we had two tanks with only zooplankton. In addition we set up tanks with natural situation: Beroe + Mnemiopsis+ zooplankton, which we expect to have in the case if Beroe includes in the Caspian trophic web (2 replications) to asses possibility Beroe control Mnemiopsis population and as result increase zooplankton abundance . To control these conditions we set up tanks with Mnemiopsis + zooplankton (two replication). For control Mnemiopsis grazing rate and possible Beroe grazing on zooplankton we had also tanks with zooplankton (two replications).

Simultaneously we examined reproduction of Beroe in the tanks with Caspian Sea water.

In addition we include two trophic webs – phytoplankton and microplankton which also indirectly effected by Mnemiopsis.

Chemical parameters was analyzed to identify effect both ctenophores (Mnemiopsis and Beroe) on nutrients and chemical conditions of environment.

Results

1. Beroe + zooplankton

After 5 days analyses of zooplankton abundance in tanks with Beroe and zooplankton and comparison them with abundance in tanks which contain only zooplankton we obtained data which allow us to conclude that Beroe did not consume zooplankton (Fig. 2, 3). The density of zooplankton in all tanks with **Beroe were not lowers than in tanks with only zooplnkton.**

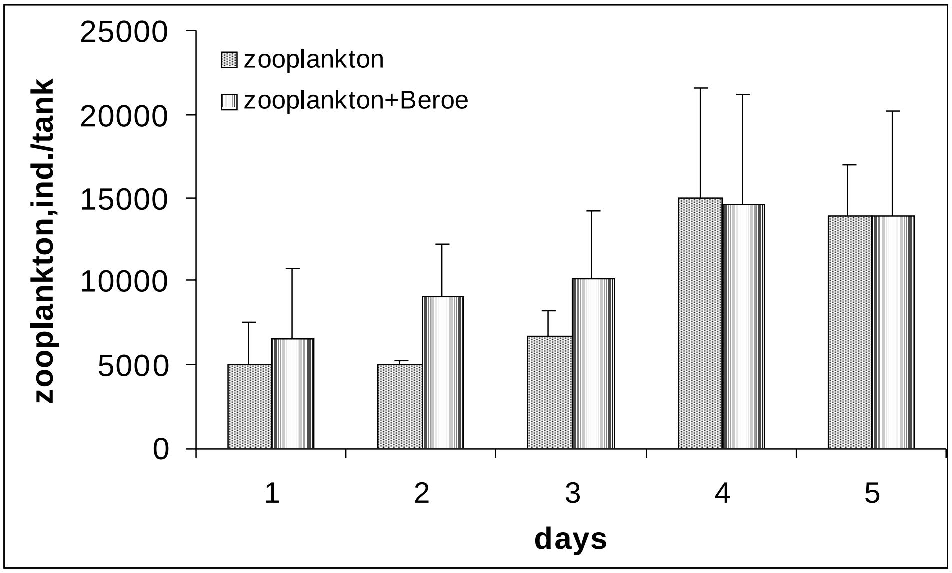


Fig.2. Zooplankton abundance in tanks: 1- with only zooplankton, 2 – with Beroe and zooplankton.

Results of measurements of Beroe individuals length support these conclusions. By the end of experiments size of Beoe individuals decrease in these tanks (Table 5). Beroe mortality was also higher in these tanks (Table 5).

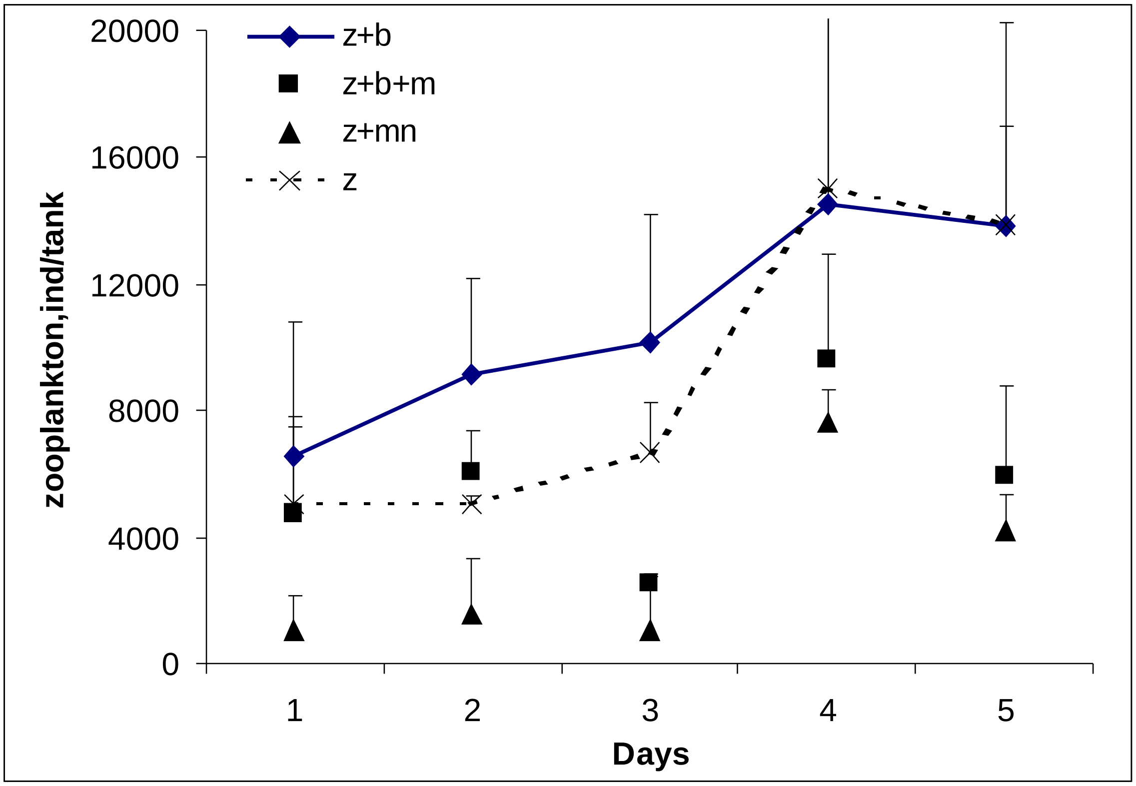


Fig.3. Zooplankton abundance in tanks: 1- zooplankton +Beoe; 2- zooplankton +Beroe+ Mnemiopsis; 3- zooplankton +Mnemiopsis; 4- zooplankton.

Table 5
Numbers and size of *Beroe ovata* in experiments

No Tank	Contents	Initial Numbers of Beroe	Initial size Mm	Final Numbers of Beroe	Final size mm	% Beroe mortality
1	Zooplankton +Beroe	7	35.6±8	7	33.5±8.5	0
2	Zooplankton + Mnemiopsis+Beroe	7	37.7±8.5	7	38.6±8.0	0
5	Zooplankton +Beroe	7	31.3±9.5	6	27.5±9.0	14.3
6	Zooplankton +Beroe	7	34.2±6	6	33.7±8.5	14.3
9	Zooplankton +Beroe	7	29.5±13	7	18.8±6.5	0
10	Zooplankton + Mnemiopsis+Beroe	7	39.1±7	6	39.5±7	14.3

Beroe +Mnemiopsis +zooplankton.

Here we tested situation that we may expect if Beroe would be introduced in the Caspian Sea.

Beroe had high clearance rate Beroe on Mnemiopsis in tanks from 36.2 to 60.6 l/ day or from 36 to 60 % total volume (Fig. 4). As a result zooplankton abundance increased in these tanks comparing with the tanks where we had only Mnemiopsis +zooplankton

(current situation in the Caspian Sea) (Fig.3).

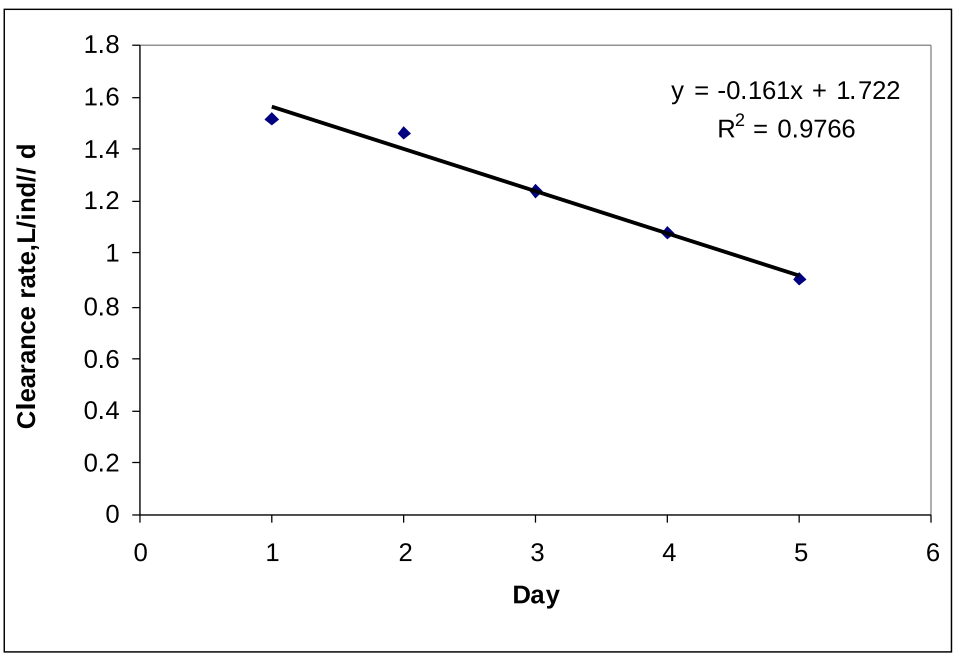


Fig. 4. Clearance rate Beroe on Mnemiopsis (l/day).

Mnemiopsis +zooplankton**. This experiment concerned the current situation in the Caspian Sea and we estimated clearance rate Mnemiopsis on zooplankton (Fig.5.).**

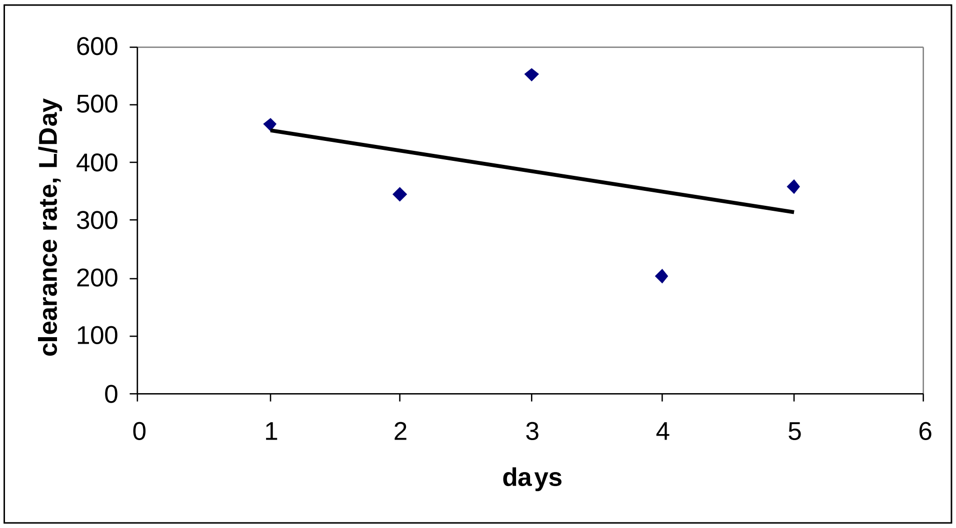


Fig. 5. Clearance rate of Mnemiopsis on zooplankton (l/day).

Clearance rate of Mnemiopsis comprised of 0.064-0.1 l/ind/h and 203-551 l/day and in percents it made up 67-154%. The concentration of zooplankton is in four times lower than real concentration in the sea this year. Thus real grazing rate of Mnemiopsis on zooplankton in the Caspian Sea in four times higher. We recorded also high numbers released ovae and larvae in tanks with Mnemiopsis+zooplankton. It was mean 131±27 ova/tank.

Reproduction.

During days of mesocosm experiments we examined Beroe reproduction in each tank.

Results of our examinations showed in Table 6 .

Table 6
Reproduction of *Beroe ovata* in tanks

No	Contents	Initial	Initial	Numbers of Beroe ovae and larvae

tank		numbers of Beroe	size Beroe, Mm	22.09.03		24.09.03		25.09.03		26.09.03	
				Ova	Larva	Ova	larva	ova	Larva	ova	larva
1	**Zooplankton +Beroe**	**7**	**35.6±8**			**90**	**0**	**0**	**0**	**0**	**0**
5	**Zooplankton +Beroe**	**7**	**31.3±9.5**			**30**	**30**	**0**	**0**	**0**	**0**
6	**Zooplankton +Beroe**	**7**	**34.2±6**			**30**	**0**	**30**		**0**	**0**
9	**Zooplankton +Beroe**	**7**	**29.5±13**			**0**	**0**	**0**	**30**	**0**	**0**
	Total numbers Zoo + Beroe	**7**	**32.65±2.8**	**0**	**0**	**50**	**8**	**8**	**8**	**0**	**0**
2	**Zooplankton + Mnemiopsis+Beroe**	**7**	**37.7±8.5**	**263**	**38**	**120**	**0**	**0**	**90**	**60**	**0**
10	**Zooplankton + Mnemiopsis+Beroe**	**7**	**39.1±7**			**180**	**0**	**0**	**0**	**0**	**0**
	Total Zoo+Mne. +Beroe	**7**	**38.4±1**	**132**	**19**	**150**	**0**	**0**	**45**	**30**	**0**

Altogether we had 863 ovae and 188 latvae. Most of ovae and larvae (79.14 % ovae and 68% larvae) we obtained from the tanks where individuals Beroe were with Mnemiopsis, we had reproduction in these tanks every day (Table 6). We had much lower numbers of ovae and larvae, where Beroe was without Mnemiopsis, only with zooplankton (20.86% ovae and 32% larvae) we had them only once or two times.

Microplankton.

In addition to the main trophic webs we tested microplankton, which abundance and biomass also change in the case of Mnemiopsis introduction. Mnemiopsis realizes mucus in the water and at this substrate bacteria develop. In tanks with Mnemiopsis and zooplankton the bacterial biomass was the highest, it was a little lower in tanks with Beroe+Mnemiopsis+ zooplankton. In the tanks with Mnemiopsis + zooplankton bacterial loop provoked also development Ciliata which consume bacteria. Ciliata reached abundance 9038 ind./tank in these tanks, in other tanks we did not find Ciliata.

Phytoplankton.

We estimated phytoplankton biomass in tanks and took phytoplankton sample in the sea for comparison.

Phytoplankton biomass was the highest in tanks with Mnemiopsis and zooplankton and lowest in tanks with zooplankton . Decline of herbivorous zooplankton abundance due to grazing of Mnemiopsis led to rise of phytoplankton abundance and diversity. In tanks with Beroe and Mnemiopsis phytoplankton biomass and diversity was lower than in tanks with Mnemiosis and zooplankton. This situation should be expected in the case of Beroe introduction in the Caspian Sea.

Biomass of phytoplankton in the Caspian Sea is much higher than in tanks. It might be explained that in our tanks we used filtrated water and tanked were placed in the laboratory, and in addition in the Caspian Sea biomass of phytoplankton is very high due high Mnemiopsis population size (13 400 ind..m-^{3,}, 747 g/m^3)

Conclusions.

Based on the observations presented here, we can conclude that individuals of Beroe in our experiments were in good conditions after acclimation to the Caspian Sea water salinity, they fed on Mnemiopsis and reproduce.

Our estimations showed that Beroe does not consume zooplankton and in conditions of absence Mnemiopsis just starve. (Beroe+Mnemiopsis).

Mnemiopsis grazing rate on zooplankton was very high and zooplankton abundance in these tanks decreased in 4-6 times every day of experiment (Mnemiopsis and zooplankton).

When we include in this trophic web(Mnemiopsis+zooplankton) Beroe, zooplankton abundance increase in 2 times, grazing pressure of Mnemiopsis decreased in the same proportions (Beroe+Mnemiopsis+zooplankton).

We showed that Mnemiopsis indirectly affected phytoplankton and microplankton biomass and species composition.

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Literature.

Clescert, L.S., Grrenberg, A.F., Trussel , 1986. Standard method for water and waste water. American public health Association. Washington, USA.

Chun C., 1880. Die Ctenophoren des Goelfes von Neapel und der Angrezenden Meeres-abschitte. Leipzig: Verlag von W. Engelmann. P. 1-313.

Esmaeili, S. A., S. Khodabandeh, B. Abtahi, J. Sifabadi & H. Arshad, 2000. First Report on occurrence of a comb jelly in the Caspian Sea. Journal of Science and Technology of the Environment, 3: 63-69.

Harbison G.R. Madin L.P. & Swanberg N.R. On the natural history and distribution of oceanic ctenophores. Deep-Sea Research.1978. Vol.25 p.233-256

Greve, W., 1970. Cultivation experiments on North Sea ctenophores. Helgolander wiss. Meeresunters. 20: 304-317.

Ivanov V.P. Biologicheskie resursy Kaspiiskogo morya. (Biological resources of the Caspian Sea).Astrakhan: CaspNIIRKH. 100 P.

Ivanov, P. I., A. M. Kamakim, V. B. Ushivtzev, T. Shiganova, O. Zhukova, N. Aladin, S. I. Wilson, G. R. Harbison & H. J. Dumont, 2000. Invasion of Caspian Sea by the comb jellyfish *M.leidyi leidyi* (Ctenophora). Biological Invasions 2: 255-258.

Kamshilov, M. M., 1960a. Feeding of ctenophore *Beroe cucumis* Fab. Dokl. Acad. Nauk SSSR, 130: 1138-1140 (In Russian).

Kamshilov, M. M., 1960b.The dependence of ctenophore *Beroe cucumis* Fab sizes from feeding. Dokl. Acad. Nauk SSSR 131: 957-960 (In Russian).

Kideys A., G A. Finenko, B E. Anninsky, T. A. Shiganova, A. Roohi, M. R. Tabari, M. Youseffyan, M. T. Rostamian, H. Rostami, H. Negarestan Laboratory Studies on Physiological Characteristics of *Beroe ovata* in the Caspian Sea Water.

Mayer, A. G., 1912. Ctenophores of the Atlantic coast of North America. Publs. Carnegie Inst. 162: 1-58.

Seravin, L. N, 1998. Ctenophora. A. Dobrovol'sky (ed), St.-Petersburg: 1-84. (in Russian).

Seravin, L. N , Shiganova T.A., Luppova N.E., 2002. The history of study of the ctenophore *Beroe ovata* (CTENOPHORA, BEROIDA) and some features of the Black Sea representavive's morphology. Zool. J. T.81. N 10. P.1193-1201. (in Russian).

Shiganova, T. A., V. V. .Sapognikov, E. I. Musaeva, M. M. Domanov , Y. V. Bulgakova, et al., 2003. Factors that determine pattern of distribution and abundance *Mnemiopsis leidyi* in the Northern Caspian. Oceanology. T.43. N5

Shiganova, T. A., A. M. Kamakin, O. P. Zhukova, V. B. Ushivtsev, A. B. Dulimov & E. I. Musaeva, 2001b. The invader into the Caspian Sea Ctenophore Mnemiopsis and its initial effect on the pelagic ecosystem. Oceanology T.41. N 4: 542–549.

In vestigation and recognition of parasites and microbial flora of caspian sea ,M. leidyi and Beroe ovata.

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Abstract:

The M. Leidyi is an unendemic invasive animal in southern coasty of caspian sea and is called as a predator of eggs , larvae and also the compete of kilka feeding .
This case was reported in recent years . by in order to control the M. leidyi population biological method in Black sea the Beroe as a predator animal has been selected .Totally 178 samples of M. Leidyi along of south capian sea (from astara to torkaman) was obtain in depths of 20m and 50m in this case 18 stations have been selected from 3-11.jul . 2003 .
in Marmara sea and black sea totally 12 and 14 samples of Beroe ovata have been obtained respectively .
the results showed that 100% of M. leidyi didn't responsed to parasites but about 83% of Beroe of Marmara sea has been infested to Trichodina-ctenophora with the intensity of minimum 4200 and maximum 21000.about 73% of Beroe samples of Black sea from the different salinities (21.6%. - 12.6%.) have been infested to T. ctenophora with the intensity of minimum 130 (salinity 12.6%.) and maximum 10500(salinily `21.6%).
all of the T. ctenophora which seperated from the salinity less than 14.6%. were degenerated .
for investigation of bacterial felora of caspian sea water ,M. leidyi and Beroe 18 , 36 and 9 samples were obtained respectively.
In this investigation 9 species of gram positive and 11species of gram negative bacteira were seperately recognized .

Introduction

In 1982 a typical Ctenophora (comb fish) belonged to Mnemiopsis genus situated in black sea has been reported (Harbison,1993).

Initially this stranger animal has been entered the black sea from U.S.A coasts by water balance of different ships. In that time this fish has been distributed all over the U.S coast.According to suitable ecological condition of black sea it provided a possibility for propagation and reproduction of Mnemiopsis . In this aspect due to predator nature this animal started to feed from the eggs and other fish larvae and also to compete the other bony fishes for food . this phenomena caused highly reduction of fish stocks in black sea and also fishing was faced a critical danger . In this case the amount of economical damage was estimated about 250 million $ (Harbison,1993) . This predator invasion fish was not reported from Caspian sea till 1999 but it was observed and reported in southern coasts of Caspian sea by the end of 2002. (Esmaely,1999). This invasion animal has been entered the Caspian sea from black sea by water balance of ships and by passing the time it made an adaptation in south basin of Caspian sea .

According to some ecological prevented factors in different parts of Caspian sea particularly less salinity in north parts of Caspian sea that was impossible for Beroe comb fish to enter the Caspian sea because Beroe has not ability for adaptation in the sea with salinity less than 10 ppt . However the south coasts of Caspian sea provided this condition (Esmaely,1999) . Comb fishes normally called as a shelter of some parasites . However they resident in the form of local in Mesogloea portion (crowell 1976) . Amphipoda is also another external parasite which has already been reported . (Harbison,1977). There was not special report and particular definition about virus , Bacterial , fungous, protozoa and trematodae in comb fishes (Harbison,1993).

In fact very few number of pathogenic organism in comb fish were identified. For example multicellular parasites like sea anomene larvae by biologic method can control and reduce the M-leidy population as it was happened in eastern north of U.S.A coasts.

Tricodinae is another ciliated unicellular parasite which can live as a host in different sea animals but usually it is called as parasite in fresh water fishes (Lom,1970). They often exist as a external parasite on the skin of sea bony fishes (Arthur,1984) . More than 28 species of this parasite in Caspian sea fishes has already been recognized and M.mccradyi comb fish can be called as a host for this parasite (Anthony. et all,2001) .

There is very limited information regarding microbial florae in comb fishes (Estes.et all,1997) but a large number of comb fishes carry natural florae and natural pathogenesis located in sea coast like Aeromonas and vibrio . The comb fish M.leidyi is called as a non local animal of Caspian sea but its presence has been reported in 1999.However it may be entered the Caspian sea many years age but in recent years it caused intensive reduction of kilka fish stocks which are consisted in their food chain .The comb fish may also carried some new micro-organisms. like funae parasites and microbial florae to the Caspian sea.

In order to control this stranger animal the selection of Beroe is the only comb fish which can be successfully linked as a predatory invasion to the south basin of Caspian sea in the form of research program . In this case the necessity of research work is to investigate the fauna parasites and microbial florae of Beroe and M.leidyi to beware of probability dangers for Caspian sea ecosystem .

Materials and methods :

in order to obtain samples from sea water and M .leidyi the south basin was divided in to3 regions(Gillan,Mazandaran,Golestan). in each region 6 station were selected in 20m and 50m depths which are showed in below picture . all of the samples were collected from 3rd to 11th of july . 2003.

1.all of the parasite samples were collected by plankton net with the mesh size of 16 micron and the sampling obtained were in a randum method . out of 720 samples of M.leidyi about 178 samples were under consideration.

parasite samples were sedimented by centrifuge (1000-1500rpm) and then were investigated by light microscope .

2.for sampling of bacterial flora of sea water the rotner has been used with the volume of 1000cc . in this connection first rotner was striled by 70% of Ethanol alchol and then washed by normal strille saline 25cc of samples was selected and transfered a strile plastic tube vessel with the volume of 50cc.these samples were finaly preserved in a refregerator.

3.for sampling from the surface and depths of M. leidyi and Beroe body the procedure is as follows :

1.the samples were kept in a plastic vessel (already washed by normal strile saline) .

2.for surface sampling of cetenophora the soap strile was used

3.these soaps were transfered to a 50 cc volume of plastic tube which contained of 25 cc normal strile saline .

4.for depth sampling of ctenophora initialy the surface of ctenophora were striled by 79% of alchol then it was washed by normal strile saline , transfering to a plastic strile tube with 50 cc volume which contaied 25 cc normal saline strile .

5.all of the microbial samples were laboratory cultured and then by the use medium culture (general , specific , selective and differentiea) the species of bacterial were recognized .

Results

Investigation and recognition of M leidyi and Beroe ovata is as fellows:

table 1 :percentage and intensity of infestation M. leidyi and Beroe T.ctenophora parastie

Place of sampling	Intensity of infestation	Percentage of infestation	No.of samples	Ctenophore species
South caspian sea	-	-	178	M.leidyi
Marmara sea	4200-21000	83	12	Beroe

Table 2: percentage and intensity of infestation in beroe to t.ctenophEra parasite in different salinitias

Percentage of infestation	Intensity of infestation					No.of samples	Ctenophore species
73%	12.6 %	14.9 %	19%	21.6%	salinity	14	Beroe
	130	250	1400 - 5000	2600-10500			

In comparison of percentage and intensity of infestation in M. leidyi to T.ctenophera parasites it was observed that all the M .leidyi samples had not any infest but 83% of Beroe samples of Marmara sea were infested to T.ctenophora parasites with intensity of minimum 4200and maximum21000.
73% of Beroe samples in black sea were infested to T.ctenophora parasites which had a maximum salinity 10500 and minimum salinity 130 in number . the samples of T.ctenophora in 12.6%. salinity were degenerated.

3

table 3: different types of recognized bacteria on the surface and in body depth of M.leidyi

Body depth of M.leidyi	Body surface of M.leidyi
Micrococcus	Bacilus circulans
Staphilococcus	B.sphrericoccus
Vibrio metschnokovii	B.coagulans
Burkhiferia mallei	Micrococcus
	M.kristinae
	Vibrio metschnokovii
	Enterobacteria
	Vibrio
	Streptobacillus
	Cytophaga

Table 4: Recognized bacteria in sampling stations of M.leidyi

1- Cytophaga
2- Vibrio
3- Micrococcus
4- Aeromonas salmonicida
5- Aeromonas
6- Erisiplothrix
7- Streptobacillus moniliformis
8- Bacillus fermus

Table 5: Recognized bacteria on the surface and in the body depth of Beroe ovata

1-Agromabacterium tumefaciens
2-Aeromonas
3-Vibrio harvaiy
4-Choromobacterium violaccum
5-Bacillus coagulans
6-Bacillus linens
7-Shewarella

in these bacteriological investigation from surface and depth body of ctenophora 14 species of bactetrial gram negative and gram positive were recogoized which are common in two pactarial species (Micrococcus and Vibrio).
the bacterial flora situated on the surface of M. leidyi ane more variety.
by comparison of bacterial flora surface of M .leidiy to caspian sea water it was observed that most of bacteria were visible in both the samples .
If comare Beroe with M. leidyi all the bacteria are common unless Chromobacterium , Agromobacterium and shewarella.

4

Discussion

Ctenophora act as a host for several parasites namely trematode (stunkard 1980) , Amphipoda (Harbison-etal,1977), protistes (kinne, 1990) and unicellular (crowell,1976).
Most of the parasites inhabit the tissues of animal body namely the tissues of mesoglea ,mouth and alimentary canal more over the most variety of unicellular and multicellular parasites have been reported in M.mccradyi (Anthony .G,2001 and sergio .R,2001). The parasites like Antamoeba, Tericodinae and unicellular flagellated protodonium attack only ectoderm tissues of comb fish to inhibit the animal body for the purpose of nourishment needs. Flagellated unicellular parasite Oodinium inhabits the Beroe comb fish near the portion of comb - rows tissues. Some of the multicellular parasites like sea -anomene larvae inhabits the abdominal pit (groove) , pharynx and mouth ventral of M. leidyi for the purpose of feeding from comb fish . This process prevent the growth and reproduction of the animal which finally caused the reduction of their population . This process can be called as a effective biological control method against M.leidyi population stocks. This is happening as an special case in open seas and oceans (Bumann.D,1996).

Tericodinae parasites more or less observed in 83 % of Beroe sample in Marmara sea and 73 % in black sea . This parasite is generally harmless although in high infections may cause serious damage to the tissues surface . In Caspian sea about 178 sample of M.leidyi during summer season were investigated in the field of parasites but no parasites was observed in animal body . It seems that there are much differences between black sea comb fish and Caspian sea comb fish from the view point of ecological condition particularly salinity amount of both the seas. When the comb fish from black sea with the salinity of 24 ppt enter the Caspian sea through volga canal by water balast of ships , it passes the several salinity changes . These salinity differences caused high changes in osmotic pressure. In this case most of the parasites particularly unicellular which forms dominant parasites Among fauna parasite destroys. In fact the comb fish devoid of any parasite. On the other side comb fish invasion the Caspian sea some years ago ,so in this connection it is possible for some of local Caspian sea fauna parasites to adapt themselves to the physiological and anatomy condition of comb fish tissues and start the new life generation as a parasites in the animal body as a host. For example Marmara Beroe was highly infected to Tricodina parasites but during its adaptation process to Caspian sea it passed through several different salinity to reach Caspian sea . At this time the final salinity in Caspian sea was estimated less than 14.9 ppt which resulted in high reduction of tericodinae parasites but in salinity of 12.6 ppt the tricodinae parasites were going to be destroyed . It means that if even the Beroe adapts to Caspian sea water its faunae parasites due to high reduction of salinity can not adapt itself to ecological condition of Caspian sea . Therefore from the view point of parasitology and importance of ecological parameters the Beroe comb fish is not able to transfer parasites infections to Caspian sea water.

From the view point of microbial flora and comparison of Beroe with M.leidyi , some bacteriae are said to be common namely vibrio and Aeromonas which can act as a factor of primary and secondary out break diseases of fishes . But other bacteria are quite different like Agromobacetrium and chromobacterium and shewarella which seem to be natural flora of water .

These microorganisms can live in high salinity of water condition which has a different environmental inhabit from Caspian sea.

If the microorganisms are successfully able to adapt them several to Caspian sea through water balance of ships , therefore it can be suggested that if Beroe successfully adapts to Caspian sea condition, it can reproduce through propagation and will have ability to feed only on Caspian sea M.leidyi

Hence parasite funae and microbial flora if exist in Beroe body will not threat the Caspian sea from different disasters. So the Beroe comb fish is the only animal which can challenge the M.leidyi by the mecanism of feeding . Before release ,the Beroe should be passed throu quarantine steps for assurance of aquatic community sanitation.

Resources:

1-Anthony.G.etal-2001 protistan epibionts of the ctenophore mnemipsis mccradyi mayer

2-Bumann.D and G.puls 1995 infestation with larva of sea anomene edwardsia lineata affected nutrition and grow of the ctnophore mnemipsis leidyi

3-Crowell.S 1975 on edwardsia larva parasitic in mnemiopsis in mackie G.O(ed).coelentrate ecology and behavour phemam publishing crop.new york 247-250

4-Edmiston-1997 the zooplankton of the apatachyola Bay systm dissertation.Florida-university.tall a ha 6600-florida 104pp

5-Esmaelli sari.A .2002 the invasive of ctenophora (M.leidyi) and future caspian sea tarbiat modarres university

6-Estes A.M,B.S.Reynolds and G.moss 1997 trichodina ctenophpre of the northern coast of the Gulf of Mexico J.euk microbial 44 ; 420- 424

7-Harbison G R 1993 methods for the control of pupulation of the ctenophore mnemiopsis leidyi in black and Azov seas

8-Kinne.O-1990.Disease of marine animals.vol 3 biological institute hdgoland Hamburg.Germany 550 pp

9-Lom.J-1970 observation on trichodinid cilites from fresh water fishes.Arch protistenk.Bd 112 : 153-177

10- Sergio-R.Mortorelli.2001 degenea parasite of jelly fish and ctenophores of the southern Atlantic

11-Stunkard.H.W-1980 the morphology life cycle and taxonomic repation lepocreadiun(trematodes Digenea) Biol-Ball 24:69-76

Population molecular study of *Mnemiopsis leidyi* in the Caspian Sea and Black Sea

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ABSTRACT: In this study genetic variation of *Mnemiopsis leidyi* was investigated in 160 samples of the Caspain Sea and Black Sea. DNA was extracted by phenol – chloroform methods. The specific primer were designed by sequence of 18s rRNA gene in mtDNA molecule. This gene of mtDNA were amplified using PCR techniques followed by RFLP analysis. The PCR products of samples were digested by restriction endonuclease such as *Alu I, Alw 26I, Bcl I, Dra I, Rsa I, Msp I, Eco 47I* and *Hae III*. Digested PCR products were observes by silver staining method followed by polyacrilamid gel electropharesis (PAGE). The results indicated no polymorphism between samples by *Eco 47I, Alw 26I, Bcl I, Rsa I, Msp I and Hae III* enzymes. *Dra I* and *Alu I* showed different patterns on PAGE. *Dra I* and *Alu I* have had different enzymatic digestion patterns and each of them shows two different genotypes. *Alu I* and *Dra I* have shown difference in 5 and 6 samples respectively.

KEY WORDS: mtDNA, PCR-RFLP, Mnemiopsis leidyi, genetic diversity

Introduction

Ctenophores represent a distinct phylum of gelatinous invertebrates that are found in virtually all marine environments (costal and oceanic, from the surface to the deep sea, and from the tropics to the poles). Of the known species of ctenophores, most were described in the 19th century and during the early 1900s (Chun, 1898; Mayer, 1912).

It is currently believed that many of those species are synonyms or represent development stages of others. The number of valid described ctenophores species is between 100 and 150 (Mills, 2001), and it is believed that there are many deep-sea species still to be discovered. Ctenophores are very poorly known, primarily because they are extremely fragile and different to collect and identify, they can not be preserved, and many species inhabit hard–to–reach locations (Harbinson et al., 1978).

Molecular marker (DNA sequences) could greatly improve the accuracy of species identification and will be invaluable for taxonomic and ecological studies. Some species reach high densities in coastal blooms and lobate ctenophore (*Mnemiopsis leidyi*) caused major ecological disturbances after being introduced into the Black Sea, presumably through discharges of ballast water from western Atlantic ports in the 1970s (GESAMP, 1997).

RCR techniques such as RAPD (Williams et al., 1990), KAPS (Konieczny et al., 1993), Microsatellite (Panaud et al., 1996), RFLP (Cronin et al., 1994) have been used in animals and plants for molecular mapping, identification of genotypes associated with genes of interest, and genetic diversity studies. Although development in molecular biology have improved are ability to resolve many problems, identification by DNA sequencing, however, is both costly and time consuming. Restriction fragment length polymorphism (RFLP) analysis, instead, is more economical than DNA sequencing analysis and more reliable than morphology based judgments. The higher rate of evolution of mtDNA in comparison with the single – copy nuclear DNA makes it a good candidate for evaluation studies. In mtDNA, approximately 2% sequence divergence occurs per million years (Rezvani Gilkolai, 2000). This value is five to ten times faster than the rate for nuclear DNA. There is also variation in the evolutionary rate of mtDNA in different taxa in comparison with most higher vertebrates. In this investigation, Genetic variation of Mnemiopsis leidyi had studies in the North and Southern Caspian Sea and also Black Sea.

Material and Method

Mnemiopsis leidyi **samples were collected from Northern and Southern (Astara, Noshahr port and Gomishan) Caspian Sea and Black Sea (Table 1). Total genomic DNA was extracted using the phenol – chloroform protocols (Towner, 1991). Approximately 0.5 – 1 µg of DNA sample were run on a 1% agarose gel and stained with ethidium bromide to check its quality and approximate quantity. The PCR was optimized by changing the reaction, cycle number, annealing temperature and incubation time. 18s rRNA gene were amplified using 1400 f (5′ – TGT ACA CAC CGC CCG TC-3′) and 5 28 Sr (5′- CTT AAG TTC AGC GGG TAG TCT CG-3′) primers (Mircea, 2001) . Thirty cycles of PCR (denaturing at 94ºc for 1 min, annealing at 48-50ºc for 60 Sec, and extending at 72ºc for 90 Sec) were performed in 50 µl reactions containing 67 mM Tris –HCl (pH 8.3), 1.5 mM $MgCl_2$, 0.4 mM each of dNTP, 2.5 mM each of primer, 1-1.5 units of Taq DNA polymerase and 50-100 ng of Target DNA in final volume of 50 µl.**

Table 1 – The number samples used in this study

Caspian sea			Black Sea
South	**North**	**Gomishan**	
70	**30**	**20**	**40**

Digestion of PCR products using restriction enzymes: ***Alu I, Bcl I, Dra I, Alw 26 I ,Rsa I , Msp I, Eco 47I and HaeIII.***

The composition of the reaction mix for digestion was as follows:

PCR product	**3µl**
Restriction enzyme	**1µl**
Reaction buffer	**2µl**
Distilled water	**14µl**
Total volume	**20µl**

The reaction mix was incubated at the temperature and duration recommended by the manufacturer. The reaction was stopped by adding 3µl loading buffer (5% v/v glycerol, 0.1 M EDTA, 1% w/v SDS, 0.1% Bromophenl blue).The digested products accompanied with standard

marker (50 bp lader). in order to measure fragment size were than run on a 6% vertical polyacrylamide gel. The fragments were visualized by silver staining of the polyacrylamide gel.

Result:

The *Mnemiopsis leidyi* mtDNA 18s ribosomal RNA gene region was amplified using the PCR technique. Both digested and undigested PCR products showed that in *Mnemiopsis leidyi* mtDNA 18s rRNA gene region was approximately 800 pb in length (fig.1,2).

In the first step, the fragment containing 18s rRNA was separately digested by 8 restriction endonucleases *(Dra I, Bcl I, Alw 26I, Alu I, Msp I, Eco 47I, Rsa I and Hae III*). Table 2 shows the number and length fragment produced by enzymatic digestion of PCR product.

Table 2. The number and the length fragment which caused by digestive enzyme of PCR products in related to 18s rRNA in Caspian Sea cetenoihore.

No	Enzyme	Fragment number	Length of fragment
1	Alu I	4	170-256-207-192
2	*Dra I*	2	411-418
3	*Alw26 I*	2	802-30
4	*Bcl I*	2	106-723
5	*Eco 47I*	2	622-206
6	*Msp I*	3	542-229-57
7	*Hae III*	6	300-220-125-100-50
8	*Rsa I*	2	550-278

Six of eight restriction enzymes showed polymorphic patterns. These are *Alw 26I, Bcl I ,Msp I, Rsa I, Eco 47I and Hae III.* The electrophoresis patterns have been obtained by polyacrilamid gel. The size of fragments was the same for all samples by these enzymes. The restriction pattern for each enzyme was unique and similar among all of samples.

***Alu I* enzyme had 3 cut situation on PCR product. As result of enzymatic digestion function produce 4 fragments. This enzyme showed two different genotypes, A and B. The genotype A has four bands with a size about 250, 200, 190 and 170 pb and also genotype B had three bands with a size 400, 250 and 170 bP (fig. 3). Genotype B observed in 5 samples of Caspian Sea cetenophora(2 samples from Noryhern Caspian Sea and 3 samples from Southern Caspian Sea)**

Figure 4 shows two kinds of enzymatic digestion patterns of *Alu I* enzyme on polyacrilamid gel.

***Dra I* enzyme had got one cut situation on PCR product. As result of enzymatic digestion function produce 2 fragments. This enzyme showed two different genotypes, C and D. The genotype C had two bands with a size about 400 pb and also genotype D has two bands with a size 700, 130 bP (fig. 5). Genotype D observed in 6 samples of Caspian Sea cetenophora(2 samples from Noryhern Caspian Sea and 4 samples from Southern Caspian Sea).**

Figure 5 shows two kinds of enzymatic digestion patterns of *Dra I* enzyme on polyacrilamid gel.

***Alw 26 I* enzyme had one cut situation on PCR product. As a result of enzymatic digestion function**

produce 2 fragments(800 and 30 bp). This enzyme did not showe different genotype(fig.6).

***Bcl I* , *Eco 47I* , *Msp I* , *Hae III* and *Rsa I* similar bands in all samples. A morphometric banding pattern was observed in all samples by these enzymes.**

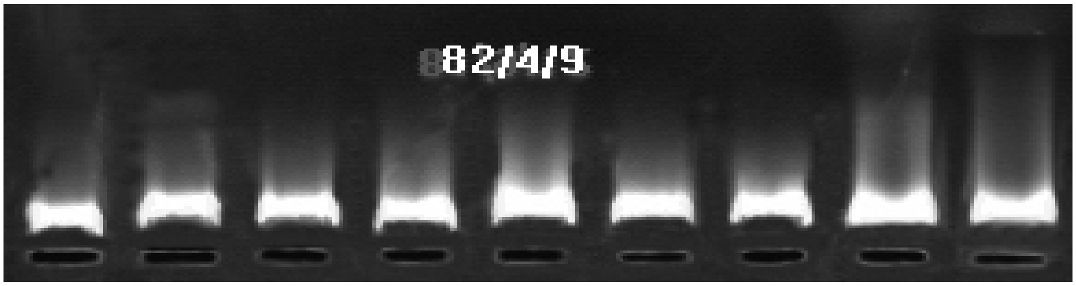

Figure 1 – DNA extraction of *Mnemiopsis leidyi* on Agarose gel

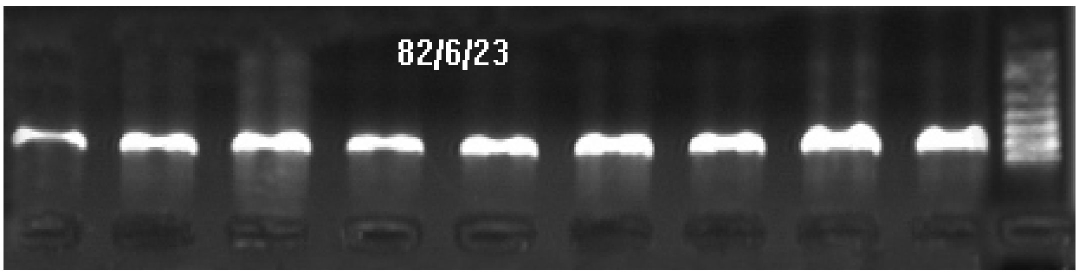

Figure 2 – PCR product of *Mnemiopsis leidyi* 18s rRNA gene on Agarose gel

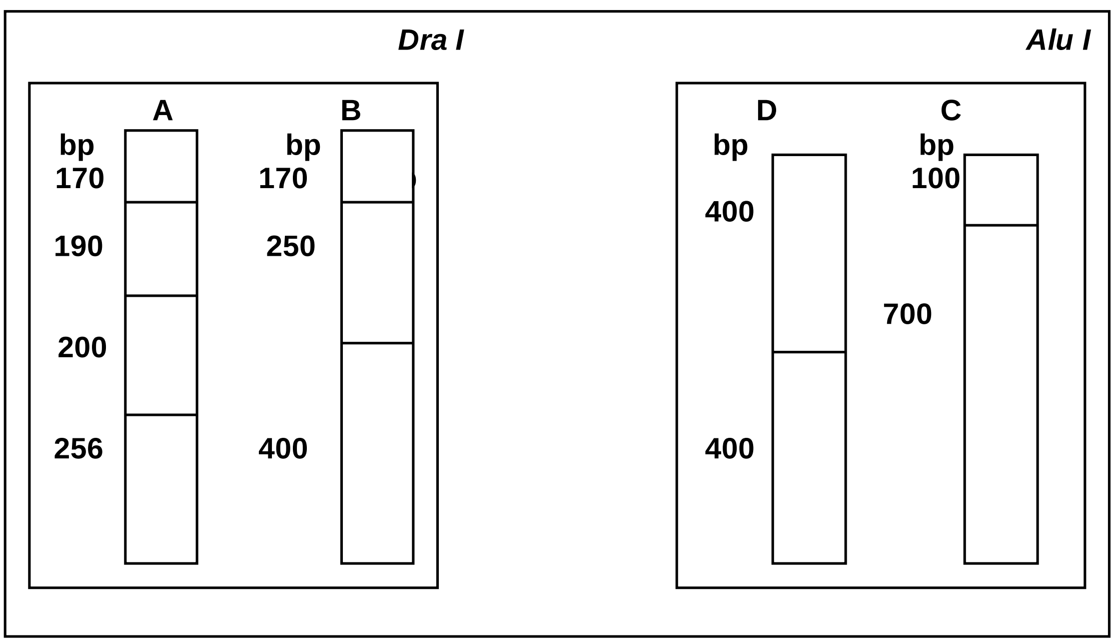

Figure 3 – Different genotypes showed with *Dra I* & *Alu I* enzymes

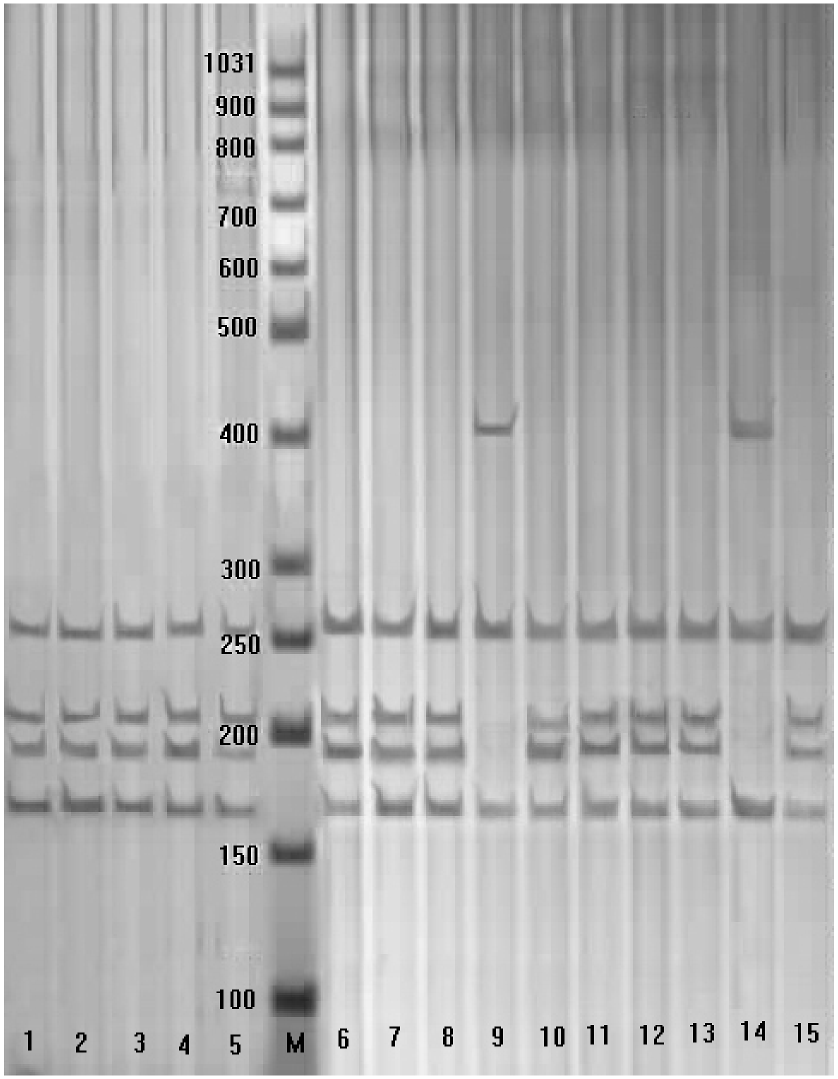

Figure 4 – Restriction digest patterns observed with Alu I enzyme. Lines 1-5 Black Sea samples, lines 6-10 Nourth of Caspian Sea samples, Lines 10-15 South of Caspian Sea samples, line M marker.

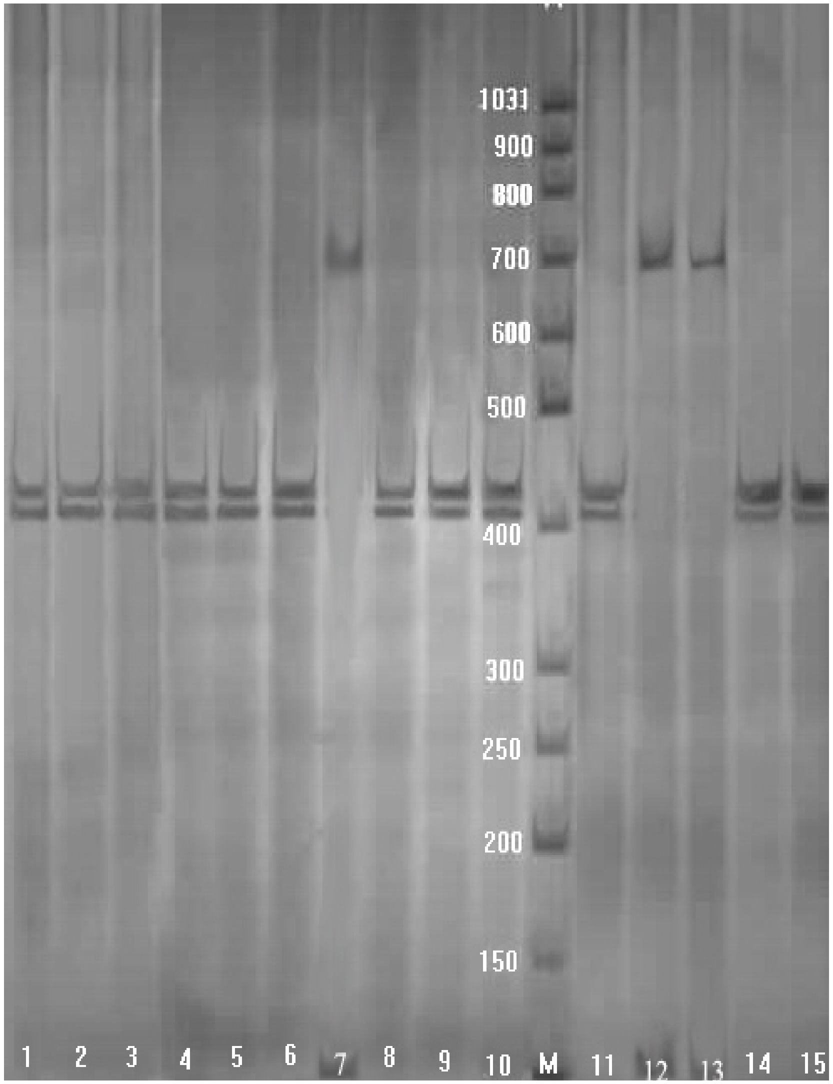

Figure 5 - Restriction digest patterns observed with Dra I enzyme. Lines 1-5 Black Sea samples, lines 6-10 Nourth of Caspian Sea samples, Lines 11-15 south of Caspian Sea samples, line M marker.

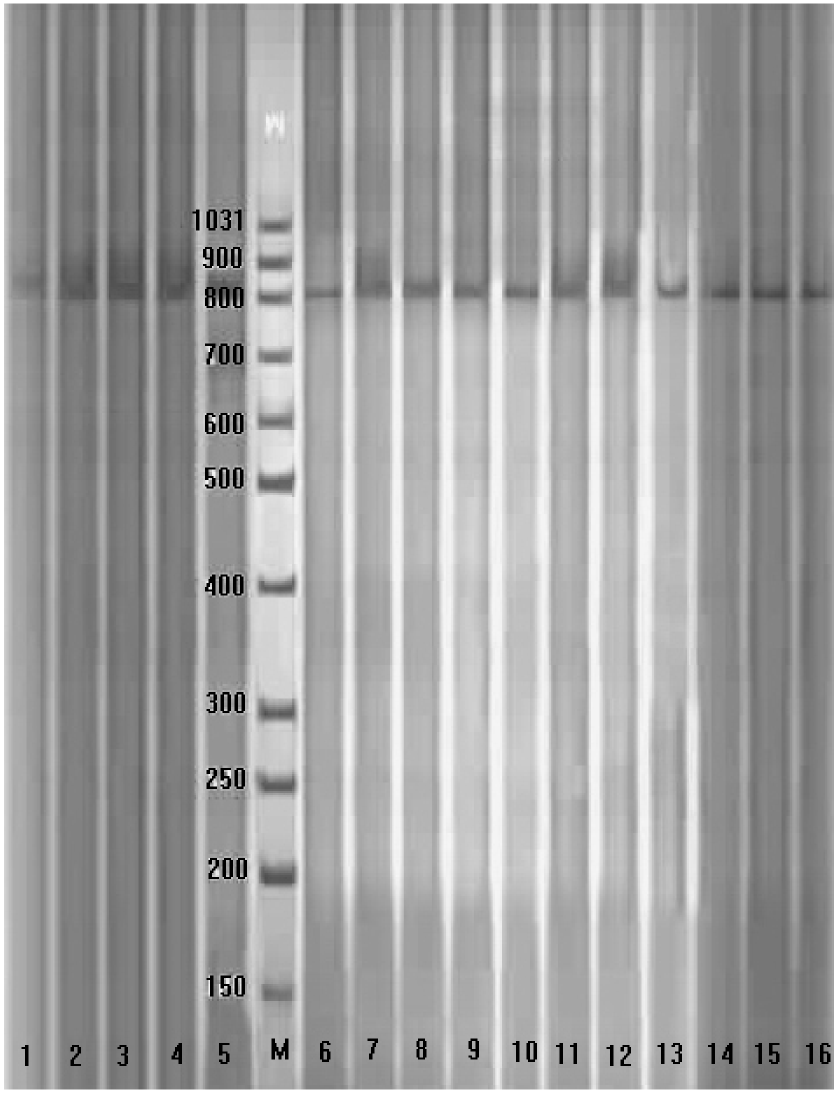

Figure 6 – Restriction digest patterns observed with Alw 26 I enzyme. Lines 1-5 south of Caspian Sea samples, lines 6-10 Nourth of Caspian Sea samples, Lines 11-15 Black sea samples, line M marker.

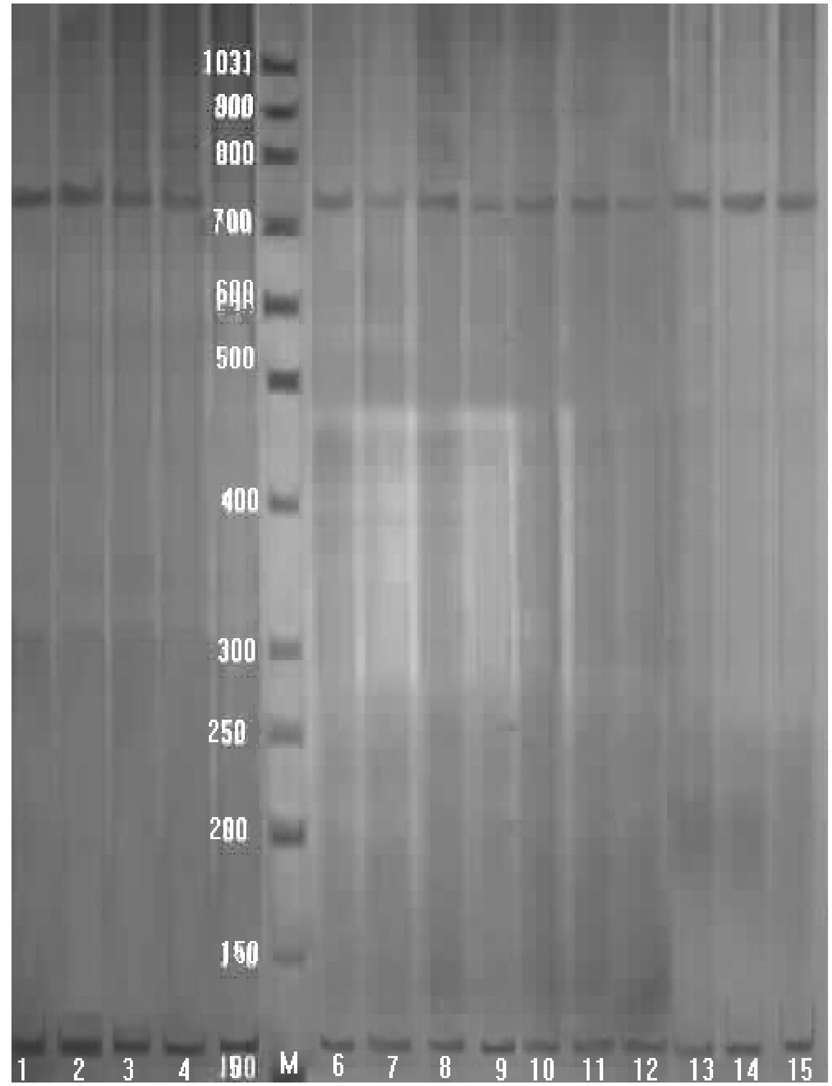

Figure 7 – Restriction digest patterns observed with Bcl I enzyme. Lines 1-5 south of Caspian Sea samples, lines 6-10 Nourth of Caspian Sea samples, Lines 11-15 Black sea samples, line M marker.

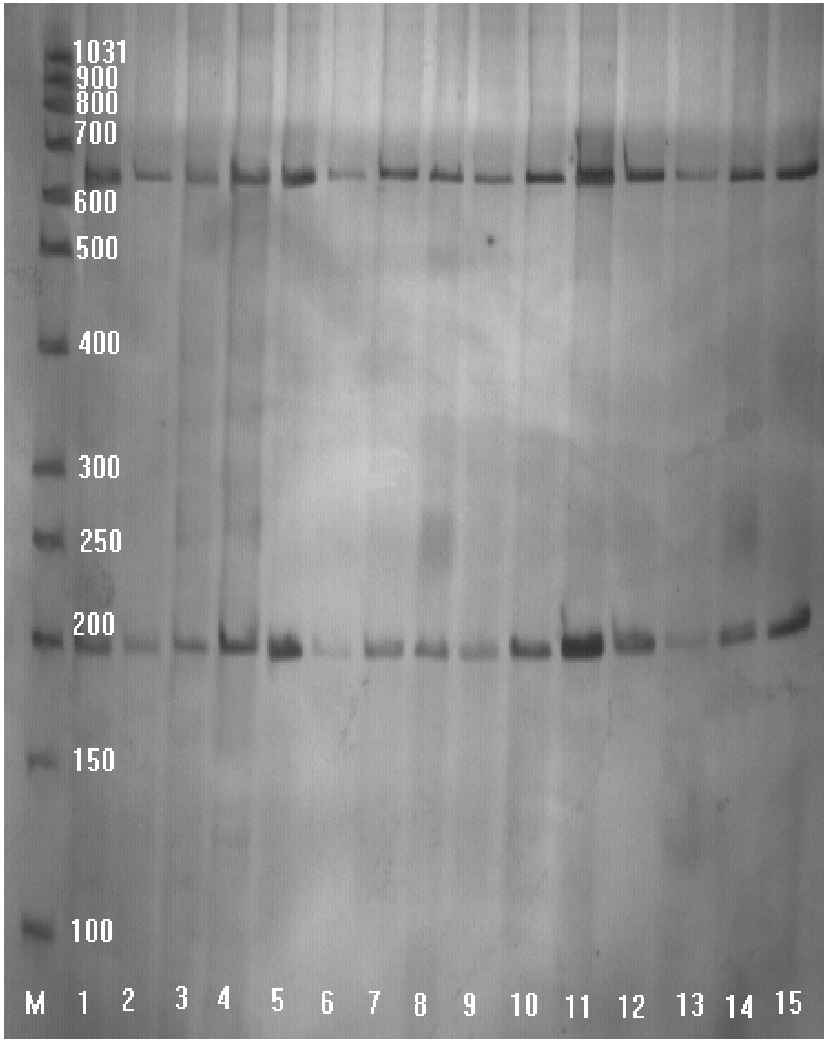

Figure 8 – Restriction digest patterns observed with Eco47 I enzyme. Lines 1-5 south of Caspian Sea samples, lines 6-10 Nourth of Caspian Sea samples, Lines 11-15 Black sea samples, line M marker.

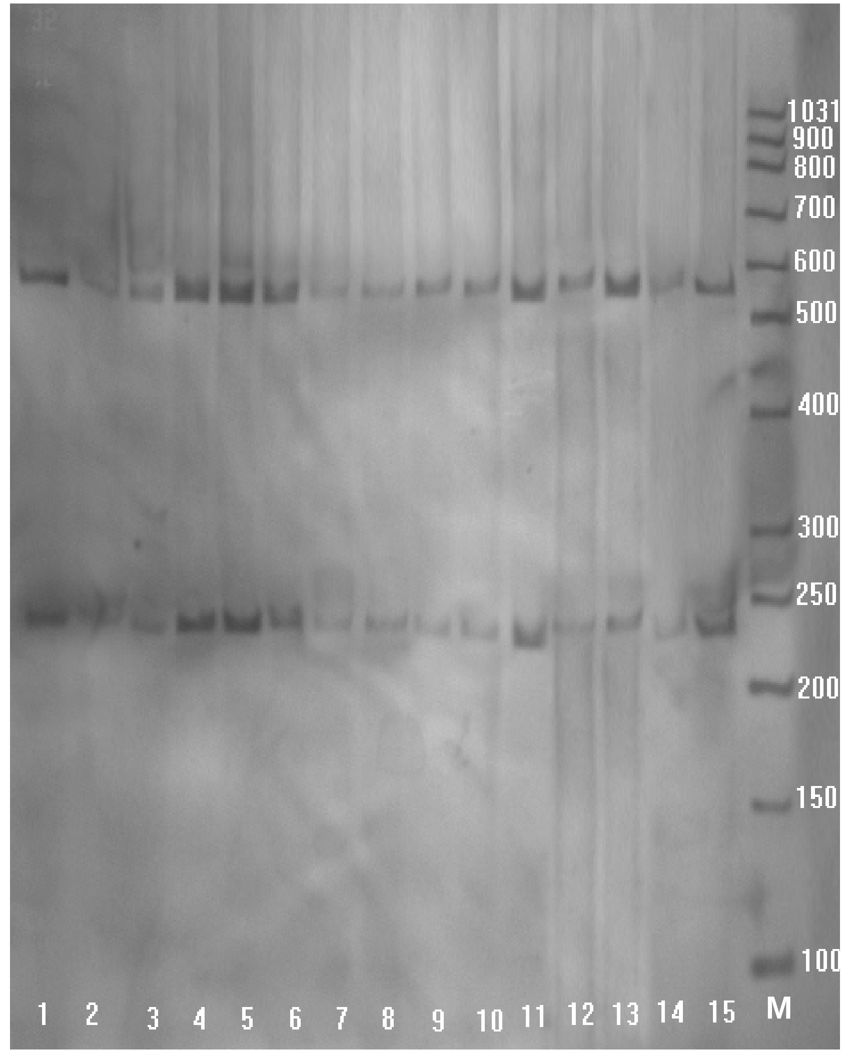

Figure 9 – Restriction digest patterns observed with Msp I enzyme. Lines 1-5 south of Caspian Sea samples, lines 6-10 Nourth of Caspian Sea samples, Lines 11-15 Black sea samples, line M marker.

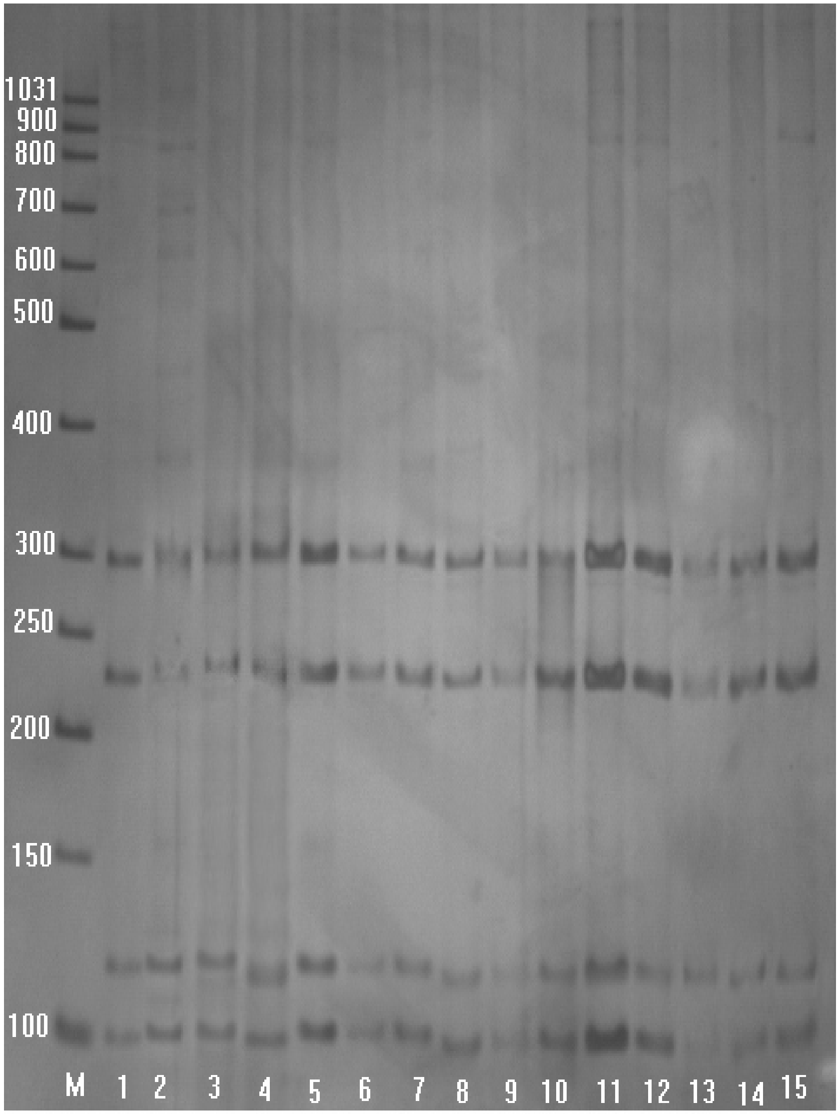

Figure 10 – Restriction digest patterns observed with Hae III enzyme. Lines 1-5 south of Caspian Sea samples, lines 6-10 Nourth of Caspian Sea samples, Lines 11-15 Black sea samples, line M marker.

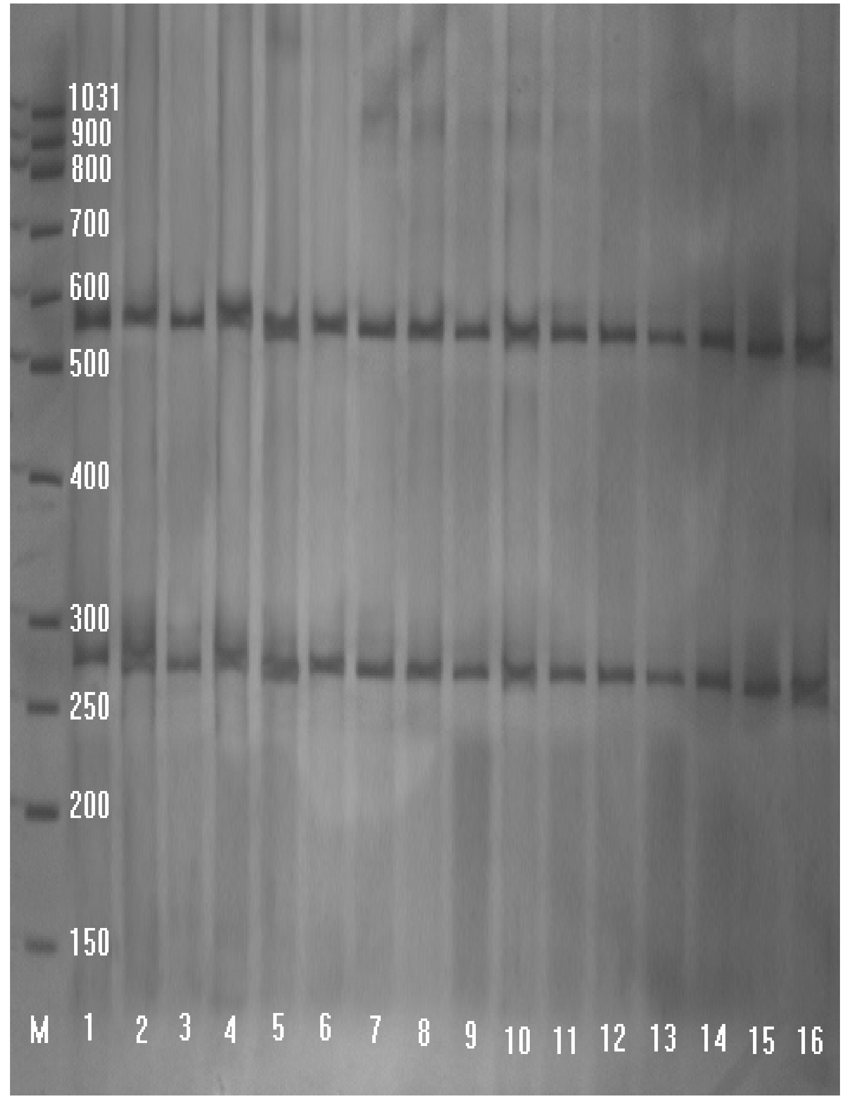

Figure 11 – Restriction digest patterns observed with Rsa I enzyme. Lines 1-5 south of Caspian Sea samples, lines 6-10 Nourth of Caspian Sea samples, Lines 11-16 Black sea
1 **samples, line M marker.**

Reference

1-Berrebi, P., 1996. Speciation of the genus Barbus in the north Mediterranean basin: recent advances from biochemical genetics. Biological conservation. Vol. 72, pp. 237-249.

2-Cronin, M. A.; Hilis, S., Born, E. W. and potton, C., 1994. mtDNA variation in atlantic and pacific waleruses. Gen. 3. 2001. 72, pp. 1035-1043

3-Hynes, R. A.; Ferguson, A. and Mccann, M. A., 1996. Variation in mtDNA and post – glacial colonization of north western Europe by brown trout. Journal of fish Biology. Vol. 48, pp. 54-61.

4-Miller, S. A.; Dykes, D.D. and Polesky, H.F., 1988. A simple salting out procedure for entreating DNA from human nuckated cells. Vol. 16, 1215 p.

Pourkazemi, M., 1996. Molecular and biochemical genetic analysis of sturgeon stock from the south Caspian Sea. School of Biological Science, University of Wales. 258 p.

5-Miccea, P., Steven, H. D., Mitchell, L., 2001. A molecular phylogenetic farme work for the phylum ctenophora using 18s rRNA Geneso Molecular phylagenetics and Evaluation. Vol. 21, No. 2, pp. 218-230

6-Mills, C., 2001. Phyllum ctenophora list of all valid species, science 238 : 933-937.

7- Panaud, O., Chen, X. and McCouch, S.R., 1996. Development of microsatellite markers and characterization of single sequence length polymorphism (SSLP) in rice Gen Genet 252: 597-607

8-Rezvani Gilkolaei, S., 1997. Molecular population genetic studies of sturgeon species in the south Caspian Sea. School of Biological Sciences, University of wales. 196 p.

9-Rezvani Gilkolaei, S., 2000. Study of mtDNA varation of Russian sturgeon population from the south Caspian Sea using RFLP analysis of PCR amplified ND5/6 gene regions. Iranian Journal of Fisheries Sciences, Vol. 2, No.1, pp. 13-36.

10-Rob, Desalle and Birestin, V. J., 1996. PCR identification of black cariar. Nature. 381 (16 May) : 197-198.

11-Williams, J.G.K., Kubelic, AR, Livak, K.J., Rafalski, J.A. and Tingey, S.V., 1990. DNA polymorphisms amplified by arbitray primers are useful as genetic markers. Nucleic Acids Res 18: 6531-6535.

Recommendations

Of The First Regional Technical Meeting on possible introduction of Beroe ovata into The Caspian Sea (22-23 Feb, 2004 Tehran – I.R. Iran)

Based on decisions made in the 20th meeting of Aquatic Bioresources Commission of Caspian Sea (Astrakhan, Russia 2003) and support of Caspian Environment Programme (CEP) in order to mitigate *Mnemiopsis leidyi*, First Regional Technical Meeting on Possible Introduction of Beroe ovata into the Caspian Sea is held in Tehran, 22-23rd of Feb. 2004. Representatives of Caspian littoral states, Azerbaijan Republic, Islamic Republic of Iran, Kazakhstan Republic, Russian Federation and Turkmenistan along with international scientists from Belgium, Ukraine, Germany, Turkey and CEP representative were present.

In this technical and professional meeting, 21 research papers in the field of Caspian, Black and Azov seas ecology, biodiversity changes in environmental community structure after invasion of *M .leidyi*, were discussed.

In addition results of experiments on *B. ovata* that is the best species in order to biologic control, and the consequences of its introduction in the Caspian ecosystem were considered.

Participants have agreed in following points:

1. Invasion of *M. leidyi* from black sea to the Caspian sea has caused ecologic and biologic modifications in one hand and kilka fishes stocks has decreased considerably in the

other hand. These changes caused socio-economic disasters.

2. Researches carried out in the field of possibility of introduction of B. ovata into the Caspian Sea have responded to all questions asked in this purpose. Experiments show the possibility of *B. ovata* reproduction in the Caspian Sea ecosystem. It feed only from *M. leidyi*. It is not harmful for the other species of Caspian environment such as zooplankton and phytoplankton. Its introduction would be beneficial to this ecosystem. In addition, its introduction does not cause any microbial and parasitic infestation to the species of the Caspian Sea.

3. Regarding the results of realized researches, about artificial introduction of B. ovata to the Caspian Sea, and considering heavy losses on fish stocks, participants of the technical meeting recommended that B. ovata must be introduced to the Caspian Sea.
4. Action plans for introduction of B. ovata for biological control of M. leidyi in to the Caspian sea:

- Technical agreement reached on 22 - 23 Feb 2004
- IFRO to prepare and share the presentations minutes of the meetings (Feb 2004)
- ToR for setting up release program (by 10th March 2004) by Dr Negarestan
- Preparation and design of the releasing program (June-July 2004)..
- National agreements reached by the July 2004
- Full regional agreements reached Oct-Nov 2004 (at the CEP Steering Committee or Aquatic Bioresources commision). Attempts will be made for an earlier consensus through communication perhaps by Aug 2004.
- Release of Beroe together with sampling of DNA library 2004-2005
- Start (or continue) of monitoring on the release follow up
- Complementary activities (modeling etc..)

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