

# ***Distaplia alaskensis* sp.nov. (Ascidacea, Aplousobranchia) and other new ascidian records from south-central Alaska, with a redescription of *Ascidia columbiana* (Huntsman, 1912)**

Gretchen Lambert and Karen Sanamyan

**Abstract:** Alaskan ascidians are incompletely known and rarely sampled. The Smithsonian Environmental Research Center recently conducted an extensive survey of harbors and marinas for nonindigenous species at major marine traffic sites on the Kenai Peninsula and Prince William Sound in Alaska. Collections made during summer 1998 and 1999 included 12 species of ascidians, one of which is a new species of *Distaplia*, *D. alaskensis*. We consider it indigenous, though it could be cryptogenic because it was collected only from marina floats and no neighboring natural subtidal areas have ever been sampled. All the other species are natives except *Botrylloides violaceus*. This aggressive invader from Japan has recently spread rapidly along both coasts of the U.S.A. and Canada as well as in many other parts of the world, and is here reported from Alaska for the first time. *Ascidia columbiana* (Huntsman, 1912), synonymized in 1924 by Hartmeyer under *Ascidia callosa*, has now been shown to be a valid species, based on differences in morphology and reproductive mode; a redescription of *A. columbiana* is included here. Several species collected in 2000 at the Sitka Sea Farm mariculture facility near Sitka are also included. Because all these collections are from areas never before sampled for ascidians, all are new records for these species.

**Résumé :** Les ascidies d'Alaska sont mal connues et rarement échantillonnées. Le Centre de recherche en environnement du Smithsonian a procédé récemment à un inventaire important des ports commerciaux et ports de plaisance pour repérer les espèces non indigènes dans les principales voies de navigation de la péninsule de Kenai et du détroit du Prince-William en Alaska. Les récoltes des étés 1998 et 1999 contenaient 12 espèces d'ascidies, dont une nouvelle espèce de *Distaplia*, *D. alaskensis*. Nous croyons qu'il s'agit d'une espèce indigène, quoiqu'elle pourrait tout aussi bien être une espèce cryptogénique, puisqu'elle n'a été récoltée que dans des ports de plaisance et que les zones subtidales naturelles avoisinantes n'ont jamais été échantillonnées. Toutes les autres espèces sont indigènes, à l'exception de *Botrylloides violaceus*. Cet immigrant agressif du Japon a récemment envahi rapidement les côtes américaines et canadiennes et beaucoup d'autres endroits au monde et sa présence en Alaska est mentionnée ici pour la première fois. *Ascidia columbiana* (Huntsman, 1912), synonymisée par Hartmeyer en 1924 sous le nom d'*A. callosa*, s'est révélée une espèce valide, distincte par sa morphologie et son mode de reproduction, et on trouvera ici une nouvelle description d'*A. columbiana*. Plusieurs espèces récoltées en l'an 2000 à la mariculture Sitka Sea Farm, près de Sitka, sont également mentionnées. Comme toutes ces récoltes proviennent de régions jamais échantillonnées auparavant pour des ascidies, il s'agit de mentions encore inédites.

[Traduit par la Rédaction]

## **Introduction**

Approximately 70 species of ascidians are known to occur in Alaskan waters (Ritter 1899, 1901, 1913; Huntsman 1912a, 1912b, 1922; Ärbäck Christie-Linde 1922, 1928, 1934; Van Name 1945; Abbott 1966; Nishikawa 1991; O'Clair and

O'Clair 1998; Sanamyan 1993a, 1993b, 1996, 1998a, 1998b, 1999, 2000). The majority of the records were obtained from the Bering Sea, Arctic Ocean, and Aleutian Islands, primarily by dredging. There are a few published intertidal records but none made by SCUBA. The ascidian fauna of Alaska is still very incompletely sampled.

As part of a Smithsonian Environmental Research Center (SERC) study on nonindigenous species in Prince William Sound (PWS) and the Kenai Peninsula (Hinès and Ruiz 2000), a team of taxonomists surveyed numerous shallow-water localities at various times during the summers of 1998 and 1999. Fouling panels submerged for 3–4 months at each location over the summer months were also analyzed. A total of 12 species of ascidians were collected, including an undescribed *Distaplia* (Lambert 2000). While they contained few species compared with the total number known from Alaska, the collections include representatives of all 3 orders. This paper contains a summary of all the ascidians collected,

Received February 15, 2001. Accepted August 9, 2001.

Published on the NRC Research Press Web site at <http://cjz.nrc.ca> on November 14, 2001.

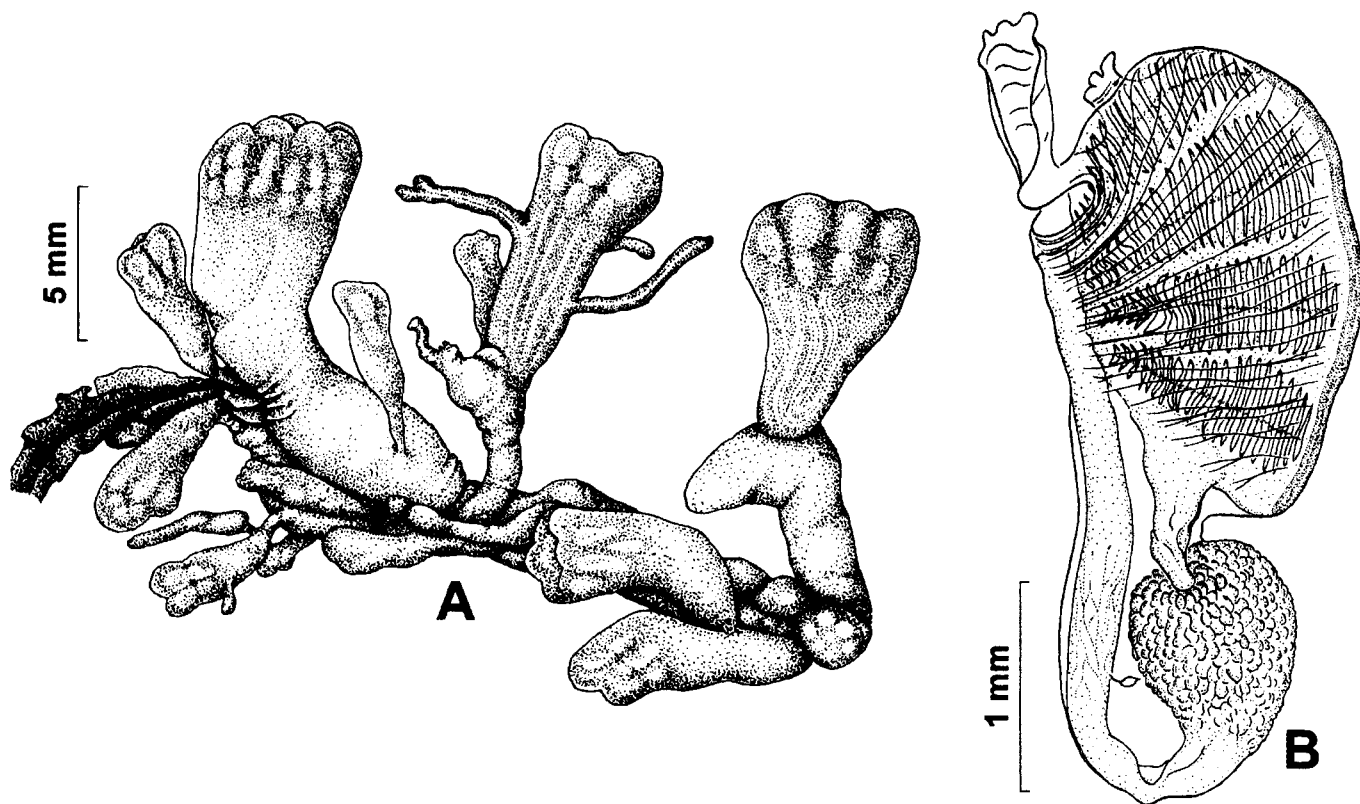
**G. Lambert.**<sup>1,2</sup> Friday Harbor Laboratories, 620 University Road, Friday Harbor, WA 98250, U.S.A.

**K. Sanamyan.** Kamchatka Institute of Ecology, 6 Partyzanskaya Street, Petropavlovsk-Kamchatsky, 683000, Russia.

<sup>1</sup>Corresponding author (e-mail: [glambert@fullerton.edu](mailto:glambert@fullerton.edu)).

<sup>2</sup>Present address: 12001 11th Avenue NW, Seattle, WA 98177, U.S.A.

**Fig. 1.** *Distaplia alaskensis* n.sp. (A) Young colony. (B) Immature zooid.



a description of *Distaplia alaskensis* sp.nov., and a redescription of *Ascidia columbiana* (Huntsman 1912), which was synonymized under *Ascidia callosa* in 1924 by Hartmeyer, but which has now been shown to be distinct from that species. Because all the collections are from areas never sampled for ascidians, all are new records for these species. The nonindigenous *Botrylloides violaceus* is reported from Alaska for the first time.

## Materials and methods

Ascidians were collected as part of several comprehensive rapid-assessment field surveys and analyses of a large number of fouling panels at numerous locations in PWS and at Homer and Seward on the Kenai Peninsula during the summers of 1998 and 1999. We personally examined all voucher specimens from both years, and one of us (G.L.) participated in the August 1999 surveys (Lambert 2000). Fouling panels were suspended in April–May of both years at depths of 1 and 3 m below mean low water at each site and retrieved between 7 and 17 September 1998 and 8 and 16 August 1999; all had a submergence time of about 4 months. A summary of the ascidian collection data for each site is given in the Appendix and by substrate in Table 2. A detailed description of the sites, methods, and results of the study is given in Hines and Ruiz (2000). Voucher samples were preserved in 10% seawater formalin buffered with sodium borate. All of the SERC voucher samples are stored at the SERC facility in Edgewater, Maryland. The type specimen of Huntsman's *A. columbiana* (described as *Ascidiopsis columbiana*) was borrowed from the Royal Ontario Museum in Toronto. The syntypes and other specimens of *Ascidia callosa* were borrowed from the Smithsonian Institution National Museum of Natural History, Washington, D.C. (NMNH); additional specimens from New Brunswick were loaned to us from the Atlantic Refer-

ence Centre, Huntsman Marine Science Centre, St. Andrews, N.B., Canada. The holotype and paratypes of *D. alaskensis* and vouchers of all other species except *B. violaceus* and *Pyura haustor* have been deposited at the California Academy of Sciences (CAS). Paratypes of *D. alaskensis* and vouchers of the other species have been deposited at SERC as described in the individual species accounts. Paratypes of *D. alaskensis* have also been deposited at NMNH. Paratypes of *D. alaskensis* and specimens of a few other species noted below have also been deposited at the Kamchatka Institute of Ecology, Petropavlovsk-Kamchatsky, Russia. Several species of ascidians from the Sitka Sea Farm mariculture facility were collected by M. Mottet in March and June 2000 and sent to G.L. for identification.

## Results

### Species descriptions

Order Aplousobranchia, family Holozoidae

#### *Distaplia alaskensis* new species

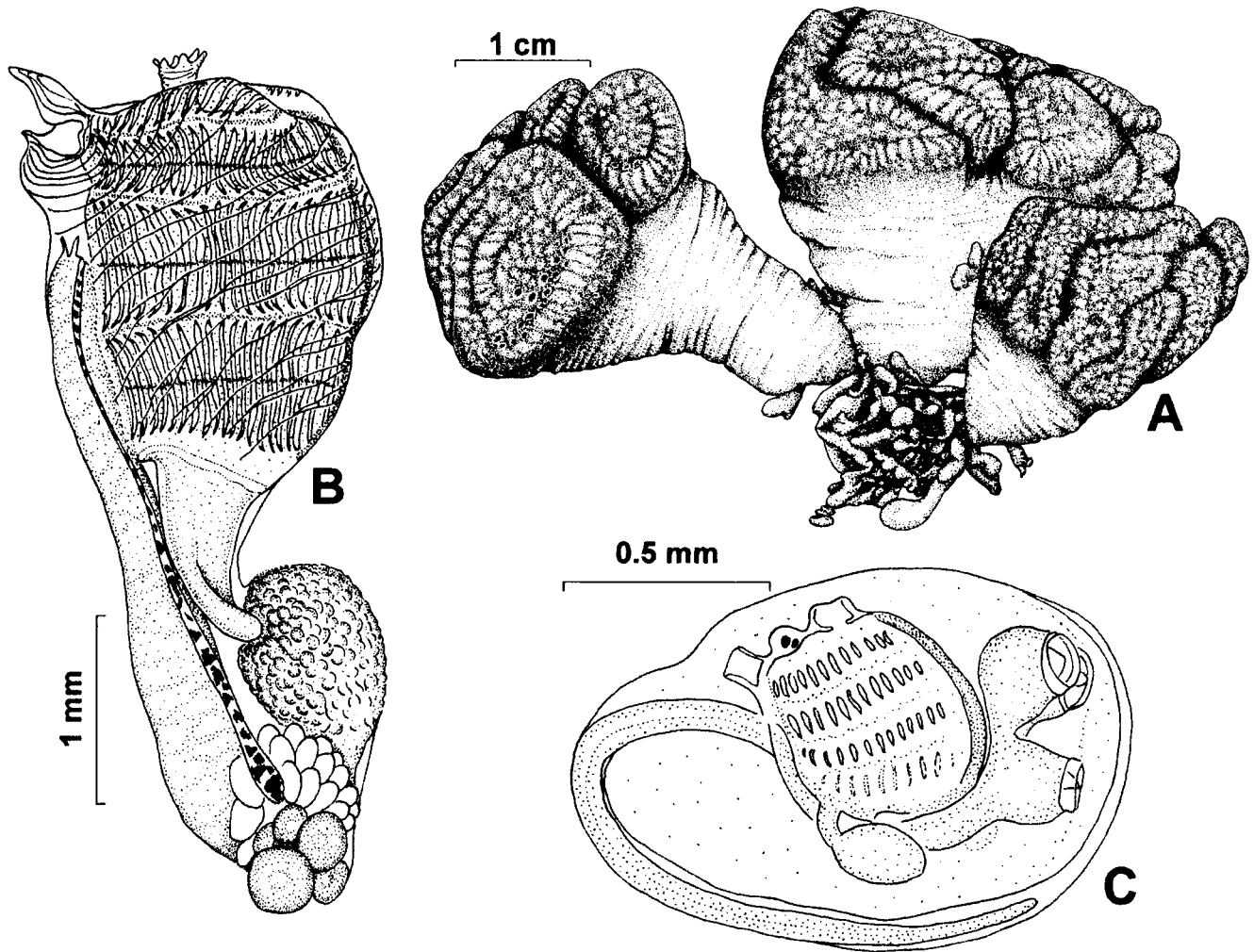
Figs. 1, 2

**TYPE MATERIAL:** *Holotype:* Consists of 3 large lobes, from suspended ropes at the small boat harbor, Homer, Kachemak Bay, Alaska; collected VIII-08-1999 by G. Lambert. CAS 152535.

*Paratypes:* Several colonies from the same sample as the holotype are deposited at the Kamchatka Institute of Ecology (KIE): KIE 1/1044. Additional paratypes are deposited at NMNH, U.S. National Museum (USNM) 1000000, CAS 152536, and SERC.

Several young colonies (or possibly fragments of one colony), small-boat harbor, Homer, Alaska, collected in October 1998 by G. Sonnevill: KIE 2/1045 and SERC. Additional

**Fig. 2.** *Distaplia alaskensis* n.sp. (A) Whole colony. (B) Mature zooid. (C) Unhatched tadpole larva.



immature sample collected V-11-1999, Homer Marina, by G. Sonnevil (SERC). Additional samples from Cordova Marina, Alaska, collected VIII-13-1999 are deposited at CAS 152537 and SERC.

#### Description

Colonies consist of several cone-shaped lobes gradually narrowing from the top to the base, with a small area of attachment arising from a tuft of short basal stolons (Figs. 1A, 2A). These lobes are elongate and flat-topped; the tops are usually subdivided by shallow furrows into oval or irregular areas each of which contains one or several systems of zooids whose limits are not always clear. Each system has a cloacal aperture at its center; there may be few or many zooids per system. The test is soft and translucent, especially on the heads. Mature colonies may be up to 5 cm in length but are always subdivided into numerous lobes 3 cm or less in diameter. Young colonies have smaller lobes arising from the basal stolons; the lobes may be several centimetres long but are only 5–8 mm in their greatest diameter, with one or rarely two or three systems of zooids. In both young and mature colonies each lobe is more or less distinctly divided into a cone-shaped stalk and a slightly expanded flat-topped head. The zooids (up to 4.5–5 mm long) are confined to the

heads (Figs. 1B, 2B). Many lobes have cylindrical and sometimes branched offshoots from their stalks or lateral surfaces of the heads. The offshoots contain vascular processes and sometimes also a few fully developed but small (2.3–2.8 mm in length) zooids at their tops. The test, especially in young colonies, is translucent and the zooids and brood sacs are clearly seen through it. In life, the colonies are tan or beige.

The thorax is somewhat longer and wider than the abdomen in fully relaxed specimens. In preserved specimens (Figs. 1B, 2B) it is usually somewhat contracted along the middorsal line and well expanded in other parts, causing it to be slightly curved with the dorsal side shorter than the ventral side. The branchial siphon has 6 lobes with well-developed circular muscles. In living specimens the atrial aperture is very large and expanded, exposing most of the first three rows of stigmata. In preserved zooids the atrial aperture is a transverse slit at the level of the first or first and second rows of stigmata. It bears a pronounced lip from its upper border. The free terminal border of the atrial lip has either 3 indistinct lobules or is almost smooth. The thorax bears 25–30 thin transverse muscles on each side, most of which extend from the endostyle to the dorsal line. Longitudinal muscles are lacking in the thorax wall, but there are

**Table 1.** Comparison of North Pacific *Distaplia* species.

	No. of stigmata per side	Stomach	No. and description of thoracic muscles per side	No. of larvae in brood pouch	Colony color	Colony shape
<i>D. alaidi</i>	24–28	Faint areolations or longitudinal plications	10, longitudinal	?	Transparent, colorless	Pedunculate
<i>D. alaskensis</i>	22–24	Large areolations	25–30, transverse	Up to 15	Tan	Conical
<i>D. dubia</i>	12–15	Longitudinal plications	15–20, longitudinal/oblique	?	White or pink	Flat
<i>D. miyose</i>	14	Longitudinal plications	20–30 longitudinal	?	Greenish	Flat
<i>D. occidentalis</i>	14–16	Irregular longitudinal plications	20–25, longitudinal and oblique	4–7	Purple, pink, tan	Mushroom
<i>D. rhavskii</i>	18–20	Areolations	20, transverse	2–3	Pink	Hemispherical
<i>D. systematica</i>	?	Smooth	20, transverse	?	Pink, brown/purple	Oval or round, flat
<i>D. unigermis</i>	12	?	30–40, longitudinal	1	?	Flat

**Table 2.** Substrates occupied by the ascidian species collected in this study.

Genus	Species	Solitary (S) or colonial (C)	Substrate* (this study only)
<i>Distaplia</i>	<i>alaskensis</i>	C	m, p, r
<i>Distaplia</i>	<i>occidentalis</i>	C	m, r
<i>Chelyosoma</i>	<i>productum</i>	S	i
<i>Corella</i>	<i>inflata</i>	S	m, o, p, r
<i>Corella</i>	<i>willmeriana</i>	S	p
<i>Ascidia</i>	<i>callosa</i>	S	m, p, r
<i>Ascidia</i>	<i>columbiana</i>	S	i, o, p
<i>Botrylloides</i>	<i>violaceus</i>	C	p
<i>Styela</i>	<i>truncata</i>	S	m, p, r
<i>Halocynthia</i>	<i>igaboja</i>	S	i
<i>Pyura</i>	<i>haustor</i>	S	i
<i>Molgula</i>	<i>retortiformis</i>	S	m, r

\*i, intertidal rocks; m, marina float; o, oyster bag; p, fouling panel; r, rope suspended from marina float.

two strong longitudinal muscles (not shown) along the middorsal line in the wall of the branchial sac. There are about 12–14 small oral tentacles in two size ranges. The usual four rows of long stigmata are crossed by well-developed parastigmatic vessels. The first three rows have 22–24 stigmata per side, the fourth row 21. Three long dorsal languets are displaced to the left side of the thoracic midline. The slightly curved oesophagus leads into the somewhat obliquely oriented stomach whose enlarged anterior end is slightly turned to the right side; a rectal valve is present. The stomach wall is very distinctly areolated and bright yellowish orange in life; this color fades somewhat in formalin. The large, regularly rounded areolations give the stomach a mulberry-like appearance. A gastric reservoir, more clearly visible in immature zooids (Fig. 1B), extends between stomach and intestine. The gut loop is almost vertical. Gonads occupy the right side of the gut loop (Fig. 2B) and consist of a rosette of 20–30 male follicles with a peripheral ovary containing a few relatively large ova. The sperm duct is very distended in some zooids.

The brood pouches are long and, unlike those of some other *Distaplia* species, always remain attached to the parent zooid throughout development; the parent does not die. Each brood pouch contains up to 15 developing larvae and embryos. The larval trunk is 1.1–1.3 mm long (Fig. 2C) with 12 or 13 stigmata per half row in the branchial sac.

#### Remarks

The species is characterized by a constellation of characters, including the conical shape, tan color, numerous brooded embryos, nearly transverse thoracic muscles, and especially the strongly areolated stomach (Table 1). It is most closely allied to *Distaplia rhavskii* Sanamyan, 1993 described from the Bering Sea and to *Distaplia occidentalis* Bancroft, 1899, which is common from south-central Alaska to California. *Distaplia rhavskii* is easily differentiated from *D. alaskensis*; the massive hemispherical colonies consist of numerous small lobes and the tunic is pinkish, the color remaining unchanged after several years in alcohol. The lobes in *D. alaskensis* have a very elongated conical

stalk and living colonies are a light mustard yellow or tan. *Distaplia occidentalis* colonies can vary a great deal in color: lavender, dark purple (the most common), yellow, orange, or tan. The lobes are broadly flattened anteriorly with a short narrowed stalk, and do not have the elongate conical shape of *D. alaskensis*.

The zooids of *D. rhavskii* and *D. alaskensis* are very similar, although those of *D. rhavskii* have slightly fewer stigmata per half row: 18–20. This difference is not significant and other features of the two species' zooids, including the shape and orientation of the stomach and the structure of its wall, and especially the number and orientation of the thoracic muscles, are almost identical. They also have a similar gastric reservoir, which was found in *D. rhavskii* after the holotype was reexamined. *Distaplia alaskensis* broods up to 15 embryos in its elongated brood pouch, while *D. rhavskii* broods only 2 or rarely 3 embryos at a time. *Distaplia alaskensis* and *D. occidentalis* are easily distinguished. In the former species the shape is a gradually narrowing cone, very elongated in a large colony, while in the latter the shape is more like a mushroom with a short stem and a broad hemispherical cap. Another difference is the stomach, with prominent areolations in *D. alaskensis* but with irregular longitudinal plications in *D. occidentalis*. There are also differences in the pattern of the musculature, number of stigmata per row, and number of embryos brooded (Table 1). In *D. alaskensis* the brood pouch remains connected to the living parent zooid, but in *D. occidentalis* the parent dies before the embryos mature and the brood pouch becomes isolated in the tunic. Ritter and Forsyth (1917) reported 12–14 stigmata per half row in *D. occidentalis* from California, Van Name (1945) gives the same numbers (but he apparently did not count them himself), and Michaelsen (1923) reported 10–14. We counted 13–16 and 14–16 stigmata per half row in two samples of *D. occidentalis* from Cordova, Alaska. *Distaplia alaskensis* has 21–23 stigmata per half row in small, young colonies with immature zooids and 21 in the most posterior row and 22–23 in the other rows in large colonies.

*Distaplia alaskensis* differs from all other *Distaplia* species described from the North Pacific Ocean by its unique combination of characters. *Distaplia systematica* Tokioka, 1958 is the only other species with transverse thoracic muscles known from the North Pacific, but the zooids have a smooth stomach. In *Distaplia alaidi* Sanamyan, 1993, known only from two cormidia, the colony consists of heads arising from long cylindrical stalks, the zooids have a large atrial opening, and there are 24–28 stigmata per half row. Some colonies of *Distaplia dubia* (Oka, 1927) may resemble young colonies of *D. alaskensis* (see plate 11, Fig. 2, in Tokioka 1953) but the stomach has faint longitudinal plications rather than being areolated, and has 15–20 longitudinal muscles on each side. *Distaplia miyose* Tokioka, 1962 has 14 stigmata per half row and 20–30 thoracic longitudinal muscles. *Distaplia unigermis* Ivanova-Kazas, 1965 always has flat colonies and 30–40 longitudinal muscles (if correctly counted), 12 stigmata per half row, and always just a single embryo in the brood pouch.

This new species has thus far been found only on man-made structures at Homer and Cordova marinas; the other sites sampled had much lower surface salinities or were in

more exposed areas. At this time we have insufficient data to consider it either native or cryptogenic. It has never been reported from anywhere else; thus the species name *alaskensis* defines the type locality. The existing Alaskan records, made many decades ago, are from intertidal collections or dredging; since there have never been any ascidian collections made in Alaska by SCUBA, the subtidal sheltered rocky crevices favored by *Distaplia* spp. have never been sampled. *Distaplia occidentalis*, native to the Pacific coast of the U.S.A. and Canada, occurs abundantly on artificial structures as well as subtidally. Ritter (1901) in his description of *D. occidentalis* complained that "the species is so exceedingly variable" that he may have collected more than one species but decided to lump them together. We were not able to locate his type specimen or paratypes of *D. occidentalis*.

DISTRIBUTION: Alaska, PWS: Homer Marina, Kenai Peninsula; Cordova Marina.

*Distaplia occidentalis* Bancroft, 1899

Fig. 3

*Distaplia occidentalis* Bancroft, 1899, pp. 59–112; Ritter and Forsyth 1917, pp. 462–464; Van Name 1945, p. 149 (synonymy and description).

MATERIAL EXAMINED: Numerous colonies collected VIII-13-1999 from the floats at Cordova Marina, PWS. Several color morphs present, including purple and orange-tan. Up to six brooded larvae present in isolated brood pouches in the tunic. CAS 152538 (purple), CAS 152539 (tan), and USNM 1000001.

#### Remarks

This species was actually named and described by W.E. Ritter, but Bancroft's paper (1899) on oogenesis, using Ritter's unpublished notes, appeared before Ritter's description (Ritter 1901), and thus the first publication on it belongs to Bancroft. See above and Table 1 for morphological characters distinguishing this species from *D. alaskensis*.

The species described by Sanamyan (1993a) as *Distaplia* sp. aff. *clavata* has more prominent areolation of the stomach and the brood pouch remains attached to the living parent zooid, which continues to function normally.

DISTRIBUTION: In Alaska: PWS: Cordova Marina; Kodiak I. (Ritter 1901); Chichagof I., Bertha Bay rocky intertidal (O'Clair and O'Clair 1998). Outside Alaska: British Columbia to southern California, low intertidal to 15 m depth (Ritter and Forsyth 1917; Van Name 1945; Lambert et al. 1987).

Order Phlebobranchia, family Corellidae

*Chelyosoma productum* Stimpson, 1864

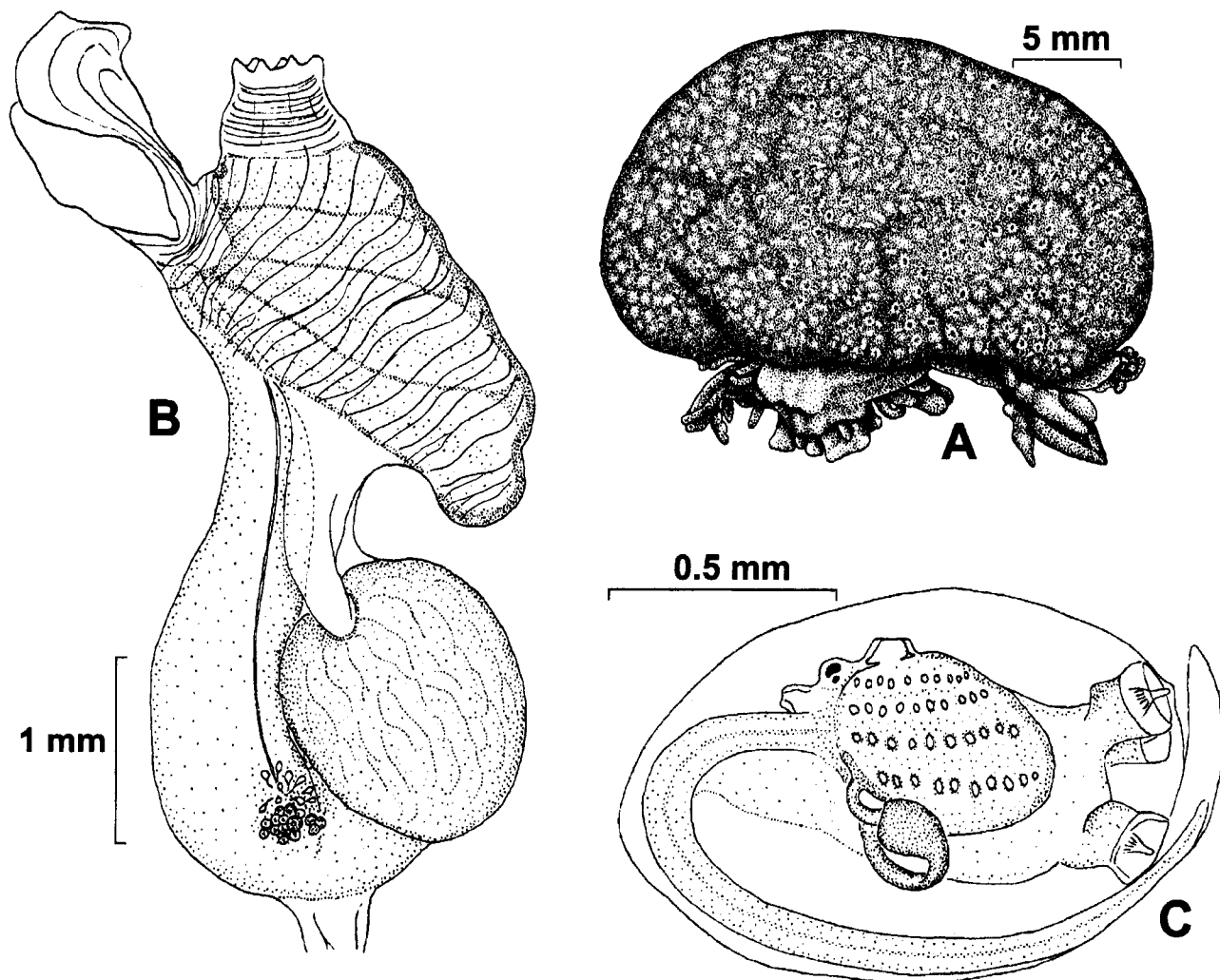
*Chelyosoma productum* Stimpson, 1864, p. 161; Van Name, 1945, p. 207 (synonymy and description); Lambert et al. 1987.

MATERIAL EXAMINED: PWS: Tatitlek, 12 specimens on intertidal rocks at low tide, VIII-12-1999. CAS 152540.

#### Remarks

A widely distributed and common ascidian, distinguished

**Fig. 3.** *Distaplia occidentalis*. (A) Colony. (B) Individual zooid. (C) Unhatched tadpole larva.



by its spiral stigmata and the pattern of plates on the oral disk.

**DISTRIBUTION:** In Alaska: PWS: Tatitlek; Sitka Sound: Passage I. (O'Clair and O'Clair 1998). Outside Alaska: British Columbia to southern California, intertidal to 50 m depth (Huntsman 1912a, 1912b; Van Name 1945; Lambert et al. 1987).

*Corella inflata* Huntsman, 1912

*Corella inflata* Huntsman, 1912a, pp. 114, 123; 1912b, p. 121. Van Name, 1945, p. 213–214 (part, under *C. willmeriana*); Lambert et al. 1981 (description and photographs).

*Corella willmeriana*: Van Name, 1945, p. 213–214 (part); Lambert, 1968; Lambert and Lambert, 1978.

**MATERIAL EXAMINED:** PWS: Tatitlek, VIII-11-1999, numerous specimens from both the fouling panels and their frames at about 3 m depth, with brooded larvae, also 1 small specimen on an oyster cage across the bay from Tatitlek; Cordova Marina floats, VIII-13-1999, numerous, with brooded larvae; Valdez, on fouling panel at Alyeska terminal Berth 5, IX-08-1998; Chenega small boat harbor floats IX-1998. Cordova specimens deposited at CAS 152541, the rest at SERC.

Sitka: 11 specimens with brooded larvae from Sitka Sea Farm net, collected III-24-2000 by M. Mottet.

#### Remarks

This species was synonymized under *C. willmeriana* by Van Name (1945) but determined to be a valid species by Lambert et al. (1981). It is distinguished from the free-spawning *C. willmeriana* by numerous differences in morphology, including the greatly expanded atrial chamber in which the larvae are brooded and released when competent to settle, some hours after hatching (Lambert et al. 1981, 1995).

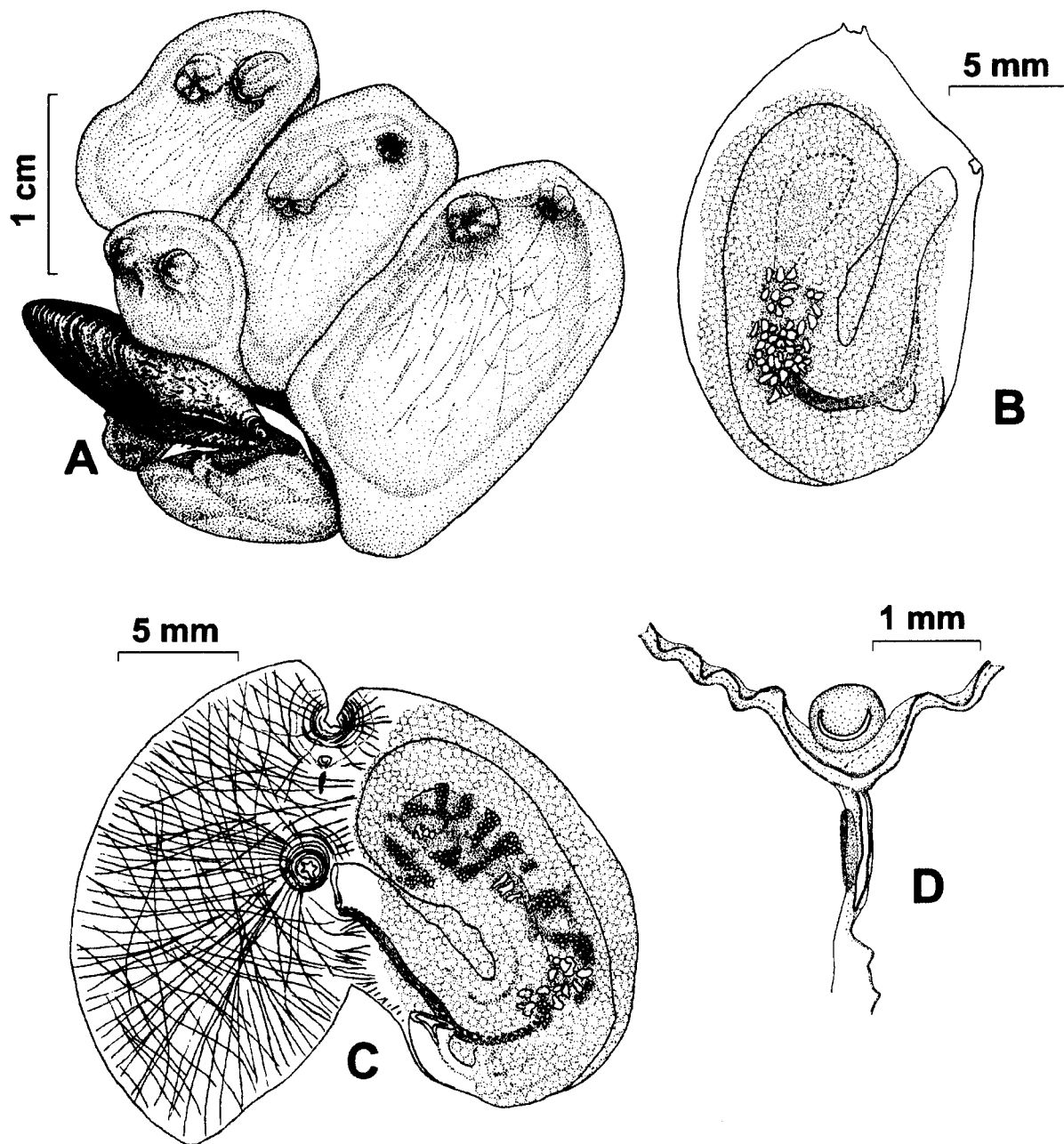
**DISTRIBUTION:** In Alaska: PWS: Tatitlek, Cordova, Valdez, Chenega; Sitka (present study). Outside Alaska: northeast Pacific Ocean: British Columbia to Washington State, shallow subtidal (Huntsman 1912a, 1912b; Van Name 1945; Lambert et al. 1987).

*Corella willmeriana* Herdman, 1898

*Corella willmeriana* Herdman, 1898, p. 252; Van Name, 1945, p. 213 (synonymy, part); Lambert et al. 1981.

**MATERIAL EXAMINED:** PWS: Cordova, VIII-13-1999, several

**Fig. 4.** *Ascidia callosa*. (A) Several individuals in a clump. (B) Left side of a specimen removed from the tunic. (C) Dissected specimen, branchial sac removed. (D) Dorsal tubercle and ganglion.



on fouling panels; Valdez, VIII-14-1999, 6.5 m depth. Deposited at SERC.

#### Remarks

See above for morphological characters distinguishing this species from *Corella inflata*. During the 60 years that *C. inflata* was synonymized under *C. willmeriana*, the two species were lumped together in studies, such as Lambert (1968) and Lambert and Lambert (1978), that were actually carried out with *C. inflata*.

**DISTRIBUTION:** In Alaska: PWS: Cordova Marina, on floats; Valdez, on fouling panels (present study). Outside Alaska: British Columbia to Washington State, 0–50 m depth

(Huntsman 1912a, 1912b; Van Name 1945; Lambert et al. 1981, 1987).

#### Family Ascidiidae

*Ascidia callosa* Stimpson, 1852

Fig. 4

*Ascidia callosa* Stimpson, 1852, p. 228; Huus, 1930; Ärnäck Christie-Linde, 1934, p. 53; Van Name, 1945, p. 178 (part; not *Ascidiopsis columbiana* Huntsman 1912a, 1912b); Millar, 1966, p. 55.

*Ascidia adhaerens* Ritter, 1901, p. 227.

**MATERIAL EXAMINED:** USNM: 5 syntypes collected by

W. Stimpson from Grand Manan, New Brunswick, and Eastport, Maine, in 1850; one previously dissected. All specimens were very disintegrated. No brooded embryos present. USNM 781 collected on gravel by W.H. Dall, VII-12-1880, U.S. Coast Survey, from Kodiak I., Chiniak Bay, Alaska, 16–25 fathoms, 4 specimens, identified by W.R. Ritter as *A. adhaerens*, the largest already dissected and now disintegrated. Many brooded embryos (short-tailed tadpoles) present in peribranchial cavity. USNM 2581, collected by W.H. Dall 1872 (his No. 944), U.S. Coast Survey, Shumagin Is.: New Harbor, Unga I., identified by W.R. Ritter as *A. adhaerens*; many brooded tadpoles present.

Huntsman Marine Science Centre, St. Andrews, New Brunswick, Canada: ARC 9954019, Letang River, N.B., VII-19-1948, labelled *Ascidia* sp., 3 specimens collected by the RV *Clupea harengus*; ARC 9954020, Letang River, N.B., December 1970, collected by J. Caddy, labelled *Ascidia* sp., 2 specimens, one with many brooded tadpoles in atrial chamber; ARC 9954021, Letite Harbour, N.B., VII-17-1972, collected by H. Gow, labelled *Ascidia* sp., 2 specimens; ARC 9159127, Deer I., N.B., wharf piling at Richardson, 10 m depth, collected by C. Hadfield, July 1990, labelled *Ascidia callosa*, 2 specimens, one dissected and contains brooded embryos in atrial chamber.

Homer, Alaska, numerous specimens collected from floats and ropes at the marina, VIII-08-1999 (CAS 152542), growing in clumps with smaller individuals attached to larger ones as shown in Fig. 4A and also Ritter's Plate 27, Fig. 1 (1901). Brooded embryos present in the peribranchial cavity of a few individuals (CAS 152543). Largest specimen 5.5 cm in length. Also Homer, Alaska, IX-03-1998, SERC OH-02-P2, 1 specimen; Chenega, PWS, IX-07-1998, 1 specimen (SERC). Also common at Cordova Marina, PWS, VIII-13-1999, some with brooded larvae (CAS 152544, USNM 1000002).

### Description

The tunic is soft, easily torn, and about 1–2 mm thick. It is not papillate around the siphons, but very small papillae may be present on the sides of the animal. The tunic margins are rounded, in contrast to the Alaskan specimens of *A. columbiana* in which the tunic edges spread out in a thin sheet that adheres to the substrate. Both siphons are 6-lobed, pinkish orange; sometimes the entire mantle is bright orange. There is an even meshwork of muscles on the right side (Fig. 4C), with a fringe of short muscles on the left side (not shown) but none covering the gut. The oral siphon is at the extreme anterior end of body, the atrial siphon close to it but slightly posterior. There are about 20 oral tentacles in a specimen 2.9 cm in length and 14 plus 5 very tiny ones in a 3 cm long specimen. The dorsal tubercle is a simple C shape opening anteriorly, the ganglion is elongate and located just posterior to the dorsal tubercle. The preoral region is either smooth or has extremely minute papillations. There are fewer than 20 longitudinal vessels per side, very irregular: 17 on the right and 14 on the left in a 3 cm long specimen. In many individuals some of the longitudinal and horizontal vessels are incomplete. Intermediate papillae are present, but only at those points where the stigmata are dividing and secondary horizontal vessels are forming. Branchial meshes are

elongate, with more than 10 stigmata per mesh. The oviduct and sperm duct follow the posterior curve of the intestine (Figs. 4B, 4C), unlike *A. columbiana*. The gut, intestine, and gonads are somewhat obscured by a thick covering of renal vesicles (dotted circles in Figs. 4B and 4C).

### Remarks

Ritter (1901) did not observe any brooded larvae in his numerous specimens from Alaska collected in 1899. We have not been able to locate these samples. However, we did observe many brooded larvae in Alaska specimens collected by Dall in 1872 and 1880 and identified by Ritter. *Ascidia callosa* from the North Atlantic Ocean, on both the North American and European sides, brood their larvae (Huus 1930; Ärnback Christie-Linde 1934).

DISTRIBUTION: In Alaska: Popof I. (Shumagin Is.), Yakutat Bay, Glacier Bay (Ritter 1901); also Kodiak I. (USNM 781, collected by W.H. Dall, VII-12-1880) and Shumagin Is. (USNM 2581, collected by W.H. Dall, 1872) identified by W.E. Ritter; PWS: Homer, Cordova, Chenega (present study). Outside Alaska: northeast Pacific coast, unclear because of the lumping together of *A. callosa* and *A. columbiana* for many decades. Probably extending south from Alaska to northern British Columbia. Common in northern Europe (Huus 1930; Ärnback Christie-Linde 1934), the northwest Atlantic (Van Name 1945), and the northwest Pacific (Sanamyan 1997, 1998b).

*Ascidia columbiana* (Huntsman, 1912)

Fig. 5

*Ascidopsis columbiana* Huntsman, 1912a, pp. 113 ff., 1912b, p. 110; Ärnback Christie-Linde, 1934, p. 53; Carson, 1980, p. 5.

*Ascidia callosa* Van Name, 1945, p. 178 (part); Lambert, 1970.

?*Ascidella griffini* Herdman, 1898, p. 256.

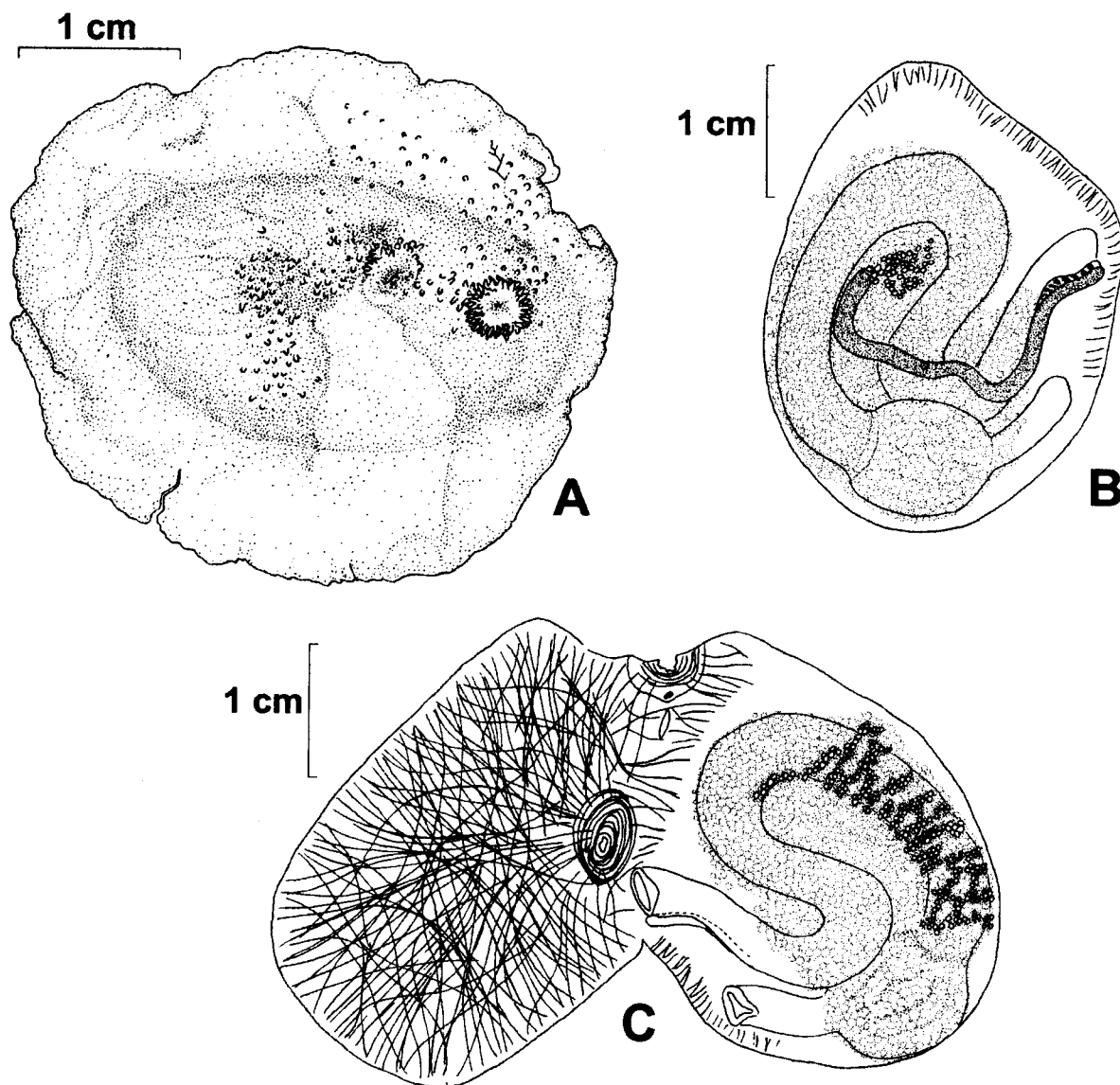
MATERIAL EXAMINED: PWS: Fairmont Bay oyster farm, VIII-10-1999, numerous specimens on oyster cages, the largest 4 cm in length (CAS 152545); Valdez VIII-14-1999, fouling plate at 6 m depth, 1 small specimen; Tatitlek, VIII-12-1999, a few small specimens on fouling panels (SERC) and their concrete block anchor (CAS 152546), and two intertidally on rocks, the largest 4.2 cm in length (CAS 152547). Sitka: 3 specimens from Sitka Sea Farm net collected III-24-2000 by M. Mottet. British Columbia: Jesse Is. in Departure Bay, Huntsman's type specimen collected at extreme low tide on rock, summer 1909 (Royal Ontario Museum); San Juan I., Washington, XII-1999: several specimens from marina floats.

### Description

Reexamination of Huntsman's type specimen: The tunic is thin, especially along the edges, where it spreads out on the substrate. Both siphons are on the upper surface. The branchial siphon is near but not at the extreme anterior edge, the atrial siphon is slightly behind and to the left. Numerous papillae are present on the tunic around the siphons. The dorsal tubercle is U-shaped. There are 43 oral tentacles, mostly of two alternating sizes. Intermediate papillae are present on parts of the branchial sac but only where new horizontal vessels are forming; there are at least 14 stigmata



**Fig. 5.** *Ascidia columbiana*. (A) Entire animal, top view. (B) Left side of a specimen removed from the tunic. (C) Dissected specimen, branchial sac removed.



per mesh. The dorsal lamina has ribs, extended as short teeth. As mentioned and figured by Huntsman (1912*b*), the oviduct and sperm duct cross the descending intestine anterior to the stomach; they do not follow the posterior border of the intestine as is the usual case in *Ascidia*. The stomach, intestine, and gonads are somewhat obscured by a thick layer of renal vesicles (dotted circles in Figs. 5B and 5C). Specimens from San Juan Island agree in all characters with Huntsman's type specimen, except that the tunic is much thicker, up to 5 mm thick in some regions in large specimens 4.5–5 cm in length, and often rounded rather than spread out thinly at the edges.

From Huntsman (1912*b*, pp. 110–113): "Attached by the entire left side to lower surfaces of stone. Upper (right) surface sloping very gradually to margin of attached surface and rough from the presence of wrinkles and short papillae with vascular cores. The latter are sometimes hardly discernible except near the apertures, where they are always distinct and large. The colour is brownish or reddish or in specimens

little exposed to light, transparent or greyish. Apertures, oral near the middle of the anterior end of part of test containing body, atrial from 1/3 to 1/2 length of body from oral along left margin of upper surface, i.e., along dorsal edge; oral 6 to 8-lobed, usually 7-lobed, atrial 6-lobed. Lobes very indistinct or indiscernible. The papillae may be arranged in radial rows corresponding to the lobes. Test 1/4–1 mm thick, vascular throughout. Musculature forms a close network on right side, absent on left: the fibres pass in every direction, but on the siphons they are arranged in the usual circular and radial layers, and near the margin they run at right angles to the margin and thin out as they reach the left side. Pharynx does not extend posterior to the intestinal loop. Tentacles 25–35, various sizes. Prebranchial zone smooth. Dorsal tubercle from round to transversely elongated, its aperture from crescent to horseshoe-shaped, with the horns turned in or even bent outwards again. It is partly hidden behind the projecting thin anterior lip of peripharyngeal groove. Ganglion, narrow, of medium length, placed just

behind tubercle. Gland, below ganglion, of about the same width or slightly more and somewhat shorter. Dorsal lamina strongly ribbed on both sides anteriorly, but only on left side posteriorly; margin toothed corresponding to ribs, teeth especially distinct at widest part of lamina, just in front of the oesophagus. Longitudinal bars 20–21 on the left side, and 22–26 on the right. Papillae at junctions with transverse vessels and also between junctions, but not throughout. They precede the division of stigmata and the appearance of new transverse vessels. At least 4 sizes of transverse vessels. The smallest often cross stigmata, preceding division of the latter. One to two plications of wall between successive longitudinal bars. Meshes elongated transversely, containing 8–20 stigmata each. Stomach with diameter much greater than that of intestine, its wall with 12–20 shallow longitudinal folds. Intestinal loop fairly close, bent in the usual manner with the concavity dorsal. Typhlosole on the right side becoming anterior in the rectum. Anus behind anterior end of the loop, or about the same level; two-lipped, margin thick and somewhat recurved. Ovary chiefly on right side of first part of the intestine, in the form of numerous branches. Testis in the form of small lobes on left side of intestinal loop and on both sides of stomach. Oviduct appears on the left side near the middle of the intestinal loop, passes across left side of second bend of intestine and then runs along the postero-dorsal border of rectum, ending just behind the anus.”

The Alaska specimens agree with Huntsman’s description in all respects. There is an even meshwork of muscles on the right side (Fig. 5C) that extends very briefly onto the left side (Fig. 5B); Huntsman’s interpretation of this was that muscles were absent on the left.

#### Remarks

*Ascidia columbiana* was synonymized under *A. callosa* by Hartmeyer (1924), and this determination was followed by subsequent workers (see Van Name 1945). Ärnback Christie-Linde (1934) maintained them as two distinct species, with some doubt. The 2 specimens from Kamchatka that she examined have the papillated tunic and other characters of *A. columbiana*, though she does not mention whether the sperm duct crosses the intestine anterior to the stomach. Herdman’s *Ascidiella griffini* from Puget Sound also has a papillated tunic and a papillated prebranchial region, but he does not include a description of the gonoducts. Published descriptions of *A. callosa* do not state whether the prebranchial area is smooth or papillate, but Huntsman (1912a, 1912b) states that it is smooth in both *A. columbiana* and *A. adhaerens*. Freshly collected specimens from the San Juan Islands, Washington, have a thicker, more rounded tunic than Huntsman’s type specimen.

*Ascidia columbiana* is distinguished from *A. callosa* by the presence of prominent tunic papillae always in a dense circle around the siphons (Fig. 5A) and sometimes also covering most of the tunic, by the gonoducts crossing the intestine along the anterior edge of the stomach (Fig. 5B), and by the absence of larval brooding in the peribranchial cavity. The free-spawning habit of this species (under the name *A. callosa*) was remarked upon by Lambert (1970, p. 22), who observed spawning in the laboratory and quoted R.L. Fernald (personal communication), who observed free spawning in the field.

DISTRIBUTION: In Alaska: PWS, Sitka (present study). Outside Alaska: British Columbia (Huntsman 1912a) to Washington State (Lambert et al. 1987 as *A. callosa*); Kamchatka, 20 m depth, V-12-1921, VI-04-1922 (Ärnback Christie-Linde 1934).

Order Stolidobranchia, family Styelidae

*Botrylloides violaceus* Oka, 1927

*Botrylloides violaceus* Oka, 1927, pp. 608–609; Saito et al. 1981, pp. 357–368.

MATERIAL EXAMINED: PWS: Tatitlek, VIII-11-1999, numerous very small zooids on fouling panels. The youngest appeared to be only hours post metamorphosis and the largest colony contained only two zooids. Deposited at SERC. Sitka: several large colonies from Sitka Sea Farm oyster net, collected by M. Mottet, III-24-2000 (CAS 152548) and VI-12-2000, the latter with mature larvae (CAS 152549).

#### Description

See Saito et al. (1981) for a detailed redescription with drawings and photographs. Its most distinguishing character is the large larva with 24–32 lateral ampullae.

#### Remarks

*Botrylloides violaceus* is not native to Alaska or to any part of the northeast Pacific. It is considered to be a Japanese species. It appeared on the U.S. Pacific coast more than 20 years ago; Fay and Vallee (1979) recorded an unidentified *Botrylloides* species containing giant larvae with 32 ampullae from subtidal areas around several of the Channel Islands. Since that time it has spread and become extremely abundant from southern California to British Columbia. In southern California it was confused with *Botrylloides diegensis* until recently (Lambert and Lambert 1998), but it is distinguished from this species by the giant larvae with 24–32 ampullae. This is the first report of its presence in Alaska. Although large colonies were not observed at Tatitlek, the fouling panels contained numerous newly settled zooids most of which appeared healthy. Thus, somewhere close to the panels there had to be mature colonies that were supplying the short-lived nonfeeding tadpoles. *Botrylloides violaceus* incubates its embryos; the tadpoles that are released are huge and complex, and usually swim for just a very brief period, perhaps only a few minutes, before settling. The unusual tadpole morphology allows for easy recognition of this invasive species (Saito et al. 1981). *Botrylloides violaceus* is very abundant at Sitka Sea Farm near Sitka (M. Mottet, personal communication); an examination by one of us (G.L.) of several large colonies confirmed the identification. It is not known when the species first appeared in the Sitka area. Because the Tatitlek specimens were newly settled zooids near an oyster farm, it is possible that the species may have been introduced through transport of oyster shells, bags, and (or) spat.

Ritter (1901) described *Botryllus magnus* from Kodiak Island at 60 m depth and Popof Island on rocks at low tide on the Alaska peninsula. It has several characters that are similar to those of *B. violaceus*: a uniform “dusky purple” color, large zooids at least 3.5 mm long, and usually 11–12 rows of stigmata. However, it has “distinct, circular, regular”

systems, which he has clearly illustrated in his Plate XXX, Fig. 34; in *B. violaceus* the systems are meandering, difficult to determine, and never circular. *Botryllus magnus* was recorded recently by Sanamyan (2000) from Medny Island (Commander Islands). Although this specimen lacks larvae and gonads are indiscernible, it is easily distinguished from *B. violaceus* by its complete second row of stigmata and clearly circular systems.

Van Name (1945) agreed with Hartmeyer (1923) that Ritter's *B. magnus* was probably a synonym of the Atlantic *Botrylloides aureum* Sars, 1851; nevertheless, he listed it as a valid species. Neither description includes any reference to the morphology of the tadpole. The second row of stigmata is complete in Van Name's illustration (Fig. 136) of *B. aureum*. *Botrylloides aureum* is described as "reddish purple to purplish black" with 16 branchial tentacles, 11–13 rows of stigmata, and the embryos developing singly in brood pouches on each side of the adult, which remains alive and functional. *Botrylloides violaceus* usually has 8 branchial tentacles and 11 rows of stigmata, never 13, and "the [incubatory] pouch including an embryo remains in the test after the degeneration of the parent zooid till the tadpole hatches out" (Saito et al. 1981). Thus, while it is possible that *B. magnus* could be *B. aureum*, neither fits the description of *B. violaceus* and we consider it a recent invader. Identification of difficult groups such as the botryllids, and their possible point of origin, would be greatly aided by molecular techniques such as the use of 18S ribosomal DNA sequences (C.S. Cohen et al. 1998; Huber et al. 2000; Swalla et al. 2000); we recommend that future collections include a small sample prepared appropriately. New techniques that can utilize formalin-fixed material for polymerase chain reaction (PCR) and DNA analysis (Yue and Orban 2001) may soon allow even museum specimens to be analyzed.

**DISTRIBUTION:** In Alaska: PWS, Tatitlek; Sitka (present study). Outside Alaska: British Columbia to Ensenada, Baja California (A. Cohen et al. 1998; unpublished observations); north-eastern U.S.A. (unpublished observations); Japan (Oka 1927; Tokioka 1953; Saito et al. 1981; Nishikawa 1991); Kurile Is., Sea of Japan (Sanamyan 2000); Vladivostok (K. Sanamyan, unpublished observations); Mediterranean Sea: Venice lagoon (Manni et al. 1995).

### *Styela truncata* Ritter, 1901

*Styela truncata* Ritter, 1901, p. 241; Van Name, 1945, pp. 314–316 (synonymy); Sanamyan, 2000, p. 71.

**MATERIAL EXAMINED:** PWS: Cordova Marina, VIII-13-1999, numerous individuals on long ropes suspended from floats, as well as on the floats (CAS 152550), and eight on fouling panels suspended from floats at the marina (SERC). Homer Marina, Kachemak Bay, Cook Inlet, VIII-08-1999: one small individual on G float (CAS 152551) and one individual on fouling panel, IX-03-1998, HO-04-P1 (SERC). Sitka: 5 specimens from Sitka Sea Farm net, collected III-24-2000 by M. Mottet.

### Description

The specimens are irregularly conical in shape, wide at the base and narrow at the anterior end, where the two siphons are close together. The largest individuals are 2 cm

tall × 3 cm across the base, 1.5 cm tall × 2 cm across, and 2.2 cm tall × 1.8 cm across. The siphons are orange-red, with 4 fleshy lobes covered with minute siphonal spines. Posterior to the siphons the tunic is pinkish tan fading to tan posteriorly. The anterior tunic is lumpy, especially around the siphons, while the posterior tunic is either smooth or irregularly grooved anteroposteriorly, depending on how contracted the individuals are.

The internal anatomy is as described by Ritter (1901) and Van Name (1945). The branchial sac, with four well-developed folds on each side, is characterized by the presence of only one longitudinal vessel between each fold. (*Styela gibbsii*, the only other species with which it might be confused, always has more than one vessel between each fold and usually four to six.) There are two large sinuous ovaries on each side, with about 8–12 large branched testes attached to the mid and posterior regions of the ovaries. Brooded larvae were present in the peribranchial chamber of a few individuals. Ritter (1901) states that though the gonads of his specimens appeared ripe, he did not observe any brooded larvae.

**DISTRIBUTION:** In Alaska: PWS, Cordova Marina floats, abundant (present study); Khantaak I., Yakutat Bay, VI-21-1899, about 50 specimens on rocks at low tide (Ritter 1901); Sitka (present study). Outside Alaska: British Columbia to southern California (Huntsman 1912b; for summary see Van Name 1945); Medny I. (Commander Is.) (Sanamyan 2000).

### Family Pyuridae

#### *Halocynthia igaboja* Oka, 1906

*Halocynthia igaboja* Oka, 1906, p. 45; Van Name, 1945, pp. 362–363 (synonymy).

*Halocynthia okai* Ritter, 1907, p. 11.

**MATERIAL EXAMINED:** PWS: Tatitlek, VIII-12-1999, low intertidal on rock, 1 very small specimen 7 mm in diameter (CAS 152552).

### Description

The specimen is immature but with dense soft spines covering the entire body and with short, inconspicuous reddish siphons. See Van Name (1945) for a complete description.

### Remarks

The taxonomy of this species is problematical. Several authors have attempted to determine whether there are several closely related species or whether all the various forms should be lumped together under one name, in which case it would be *Halocynthia hilgendorfi* (Traustedt, 1885). Most likely, however, *H. hilgendorfi* is distinct from *H. igaboja* because the long tunic spines of the former are confined to the siphons (for a detailed discussion see Van Name 1945). *Halocynthia igaboja* is large, oval or globular in shape, up to 100 mm in length, and densely covered with soft tunic spines up to 12 mm long with short, recurved side branches. The siphons are anterior, very short and often inconspicuous, and usually reddish. Nishikawa (1991) synonymized *H. igaboja* under *Halocynthia hispida* in opposition to Kott (1985) but now agrees that *H. hispida* should be reserved for the Australian form (T. Nishikawa, personal communication). Oka (1906) described a number of new *Halocynthia* species from north-

ern Japan, but those having page priority over *H. igaboja* have a different spine pattern from *H. igaboja*. Ritter (1907) described *H. okai*, which agrees in all morphological characters with *H. igaboja*. Thus, either all of these species are variants of *H. hilgendorfi* or there are several sibling species (T. Nishikawa, personal communication). Until the problem is resolved we wish to retain the long-established name *H. igaboja*.

**DISTRIBUTION:** In Alaska: PWS, Tatitlek (present study); Bertha Bay, Chichagof I. (O'Clair and O'Clair 1998: 337); near Kodiak I., 57 m depth (Sanamyan 1996 as *H. hispida*). Outside Alaska: numerous locations from British Columbia (Huntsman 1912a, 1912b) to southern California, low intertidal to 270 m depth (Van Name 1945); Japan, Otaru rocks, Hokkaido (Oka 1906).

*Pyura haustor* (Stimpson, 1864)

*Cynthia haustor* Stimpson, 1864, p. 159; Van Name, 1945, pp. 338–340 (synonymy and description); Lambert et al. 1987, p. 472.

**MATERIAL EXAMINED:** PWS: Tatitlek, VIII-12-1999, 4 specimens seen in deep rock crevices at low tide, 1 incomplete specimen collected (SERC).

### Description

All features as in Van Name (1945) and Lambert et al. (1987). This species is distinguished by its dark reddish brown, thick, tough, ridged tunic and long divergent red-tipped siphons located at the anterior end of the body. It has 6 well-developed branchial folds on each side. It is very common all along the northeast Pacific coast.

### Remarks

Van Name (1945) mentions that "eggs and larvae may often be found in the peribranchial cavity" without giving any further details regarding where these animals were collected. The *P. haustor* of Puget Sound and the San Juan Islands are exclusively free-spawners; no brooded larvae have ever been found (C. Lambert, personal communication).

**DISTRIBUTION:** Depth: intertidal to 450 m. In Alaska: PWS, Tatitlek (present study); Sitka Sound and other areas of southeast Alaska (O'Clair and O'Clair 1998: 338); Shumagin Is. (Van Name 1945), near Sanak I., dredged at 75 and 114 m depth (Sanamyan 1996). Outside Alaska: British Columbia to southern California, low intertidal to 194 m depth, common (Ritter and Forsyth 1917; Van Name 1945; Lambert et al. 1987).

### Family Molgulidae

*Molgula retortiformis* Verrill, 1871

*Molgula retortiformis* Verrill, 1871, p. 56; Van Name, 1945, pp. 422–424 (synonymy and Figs. 308–310).

**MATERIAL EXAMINED:** Homer Marina, Kachemak Bay, Cook Inlet, VIII-08-1999, 10 specimens: 2 large (58 mm tall × 45 mm across at widest region and 52 × 52 mm) and 8 small specimens, on a rope or on the side of an adjacent floating dock about 0.5–1 m below the water surface (CAS 152553). Also 2 small specimens from fouling panel E1 (SERC).

### Description

The description agrees in every respect with that of Van Name (1945). The tunic is soft, translucent, and 3–7 mm thick, thickest at the posterior end where they were attached. The surface is covered with numerous very small papillae that appear to be the terminal bulbs of the tunic vascular system, which is very highly developed. The body is globular, though somewhat taller than wide.

The branchial siphon has 6 lobes; the siphonal lining is grooved between the 6 thickened areas. Each lobe tip terminates in a short papilla that projects outward at the opening. The atrial opening has 4 lobes. The dorsal tubercle is large and elevated, fleshy, opening posteriorly, and turned to the right in one of the large individuals but not in the other; the horns are inrolled, each making a complete turn as in Van Name (1945, Fig. 309). There are 7 large branchial folds per side, 13 robust longitudinal vessels per fold, and no vessels between the folds. The stigmata are very irregular, mostly short and straight. The stomach is large, dark reddish brown, and lobulated; the kidney, on the right side, is large, sausage-shaped, and clear.

The ovary is well developed on each side except in one large individual in which the left testis (overlying the stomach) is present but the left ovary is missing. No incubated embryos are present in the peribranchial cavity; this species is known to be a free spawner with direct-developing embryos (Van Name 1945; Bates 1995). Diagnostic of this species, the gonads are separate: the left ovary is located above the stomach, the left testis overlying and below the stomach. On the right side the ovary is above the kidney, the testis below it (see Fig. 308 in Van Name 1945).

### Remarks

This molgulid is distinguished by the arrangement of the gonads and by its large size; specimens up to 10 cm long were collected by Ritter (1901). It is not a brooder; the freely spawned eggs quickly become sticky and attach to substrates near the adult (including the adult tunic), and the embryos undergo direct development without a swimming tadpole stage (Van Name 1945; Bates 1995). The larval ocellus is absent and the ampullae develop prior to hatching. Thus, it is not surprising that this species occurs in clumps and often large dense populations, since larval dispersal is usually minimal.

**DISTRIBUTION:** Depth: low intertidal to 200 m depth, though usually 80 m or less; prefers hard substrates but has been dredged occasionally from sand and gravel. In Alaska: Homer Marina, Kenai Peninsula (present study); Sitka, 30 m depth, VI-15-1899 (Ritter 1901); southeast Bering Sea (Ritter 1913); Cape Thompson area, Chukchi Sea (Abbott 1966). Outside Alaska: northwest Pacific Ocean: Kurile Is. and Kamchatka (Sanamyan 1993b), Commander Is. (Ritter 1913; Sanamyan 1997), White Sea, Siberian Arctic; North Atlantic: east coast of Canada south to Massachusetts, Iceland, Greenland, Hudson Bay, Norway (Van Name 1945; Bates 1995).

### Discussion

Homer, Tatitlek, and Cordova, the three locations with the highest surface salinity, had the greatest number of species and individuals on the substrates sampled (Appendix). No

ascidians were recorded from the floating docks at Seward, Whittier, or Valdez, where the surface salinity was only 10–12 parts per thousand, although fouling panels at Valdez submerged at about 6.5 m depth did have some ascidians. Most ascidians require a salinity of at least 25–27 parts per thousand and prefer 30 or higher (Sims 1984; Marin et al. 1987; Vázquez and Young 1996, 2000).

Three of the 12 species are colonial, the other 9 are solitary forms. Although the number of species collected was not large, all 3 orders of Ascidiacea are represented and include a total of 6 families. All of the colonials and 3 of the solitaries brood their embryos and release metamorphosis-competent swimming tadpoles; most are known to have an extremely short planktonic period, often only a few minutes (Lambert 1968; Lambert et al. 1995; G. and C. Lambert, unpublished observations). Five of these did harbor mature embryos: *D. alaskensis* n.sp., *D. occidentalis*, *C. inflata*, *A. callosa*, and *Styela truncata*. Individuals of the sixth brooding species, *B. violaceus* from Tatitlek, were all newly settled juveniles, but the parent colonies from which they were released, though not personally observed, were obviously reproductive during August 1999. Colonies of *B. violaceus* collected in June 2000 from Sitka Sea Farm contained numerous brooded larvae. It is not known how long this species has been present in Alaska.

*Molgula retortiformis*, though not a brooder, has direct development of the anural larva, thus bypassing the planktonic phase altogether (Bates 1995). The eggs, which become sticky within a few minutes of being spawned, attach to and undergo development on nearby hard surfaces, including the adult tunic. Bates (1995) and others (Huber et al. 2000) have studied the evolution of direct development in ascidians. Rather than supporting Berrill's substrate hypothesis (1931), in which he considered that species living in a homogeneous environment do not need larval structures for habitat selection, perhaps this represents adaptation to life in rather extreme environments, analogous to the brooding behavior described above, that results in the successful establishment of large clumps of breeding adults by preventing the sweeping away and loss of larvae from suitable habitats.

It is surprising that the new *Distaplia* species is very abundant at both Homer and Cordova marinas, yet has gone unrecognized for nearly a century. This species may have been much rarer during Ritter's and Huntsman's time. Its preferred habitat is apparently sheltered surfaces, shallow but never exposed at low tide and situated away from very much light. It is thus "pre-adapted" for the huge surface area and specialized environment provided by marina floats and the many submerged ropes up to 3 m in length that are often suspended from floats. In this respect it is comparable to the native species *D. occidentalis*, which is extremely abundant on floats at many marinas (Lambert et al. 1987; A. Cohen et al. 1998). Before the building of large marinas, this particular type of habitat was not abundant. Environmental conditions on marina floats are somewhat unstable, as the floats are subjected to changing seasonal conditions and anthropogenic activities. Many native ascidians, as well as invaders, are well adapted to take advantage of this instability. *Distaplia* spp., like most aplousobranchs, are very fast growing; they reach sexual maturity in a few weeks, reproduce, and then die back to a dedifferentiated basal portion

than can survive until environmental conditions are again suitable for rapid growth. It is possible that if Ritter (1901) or Huntsman (1912a, 1912b) collected it, they may have considered it merely a variant of *D. occidentalis*. Ritter (1901) stated in his description of *D. occidentalis* that "the species is so exceedingly variable and the quantity of material of the Alaska form now available for examination so small that I do not feel justified in regarding the differences as of more than individual value." Natural subtidal areas adjacent to Homer and Cordova were not sampled in this study and have never been sampled to our knowledge, thus it is not possible to say whether *D. alaskensis* is a member of these communities. Until more information is available on the subtidal ascidians of the Kenai Peninsula and PWS we cannot be sure of the origin of *D. alaskensis*. It is possible, but we feel doubtful, that it is an invader; it has never been collected from the Canadian Pacific coast or during extensive sampling around Japanese and Kamchatka waters. Thus, we hesitate to designate it native, cryptogenic, or introduced at this time.

Two species of the genus *Ascidia* were collected. One corresponds to *A. adhaerens* Ritter, 1901, the other to *A. columbiana* (Huntsman, 1912). Hartmeyer (1924) synonymized these two northeast Pacific species under *A. callosa*, and Van Name (1945) followed Hartmeyer's scheme. *Ascidia callosa* was described in 1852 by Stimpson from Maine and New Brunswick specimens. Reexamination of Stimpson's syntypes of *A. callosa* and Dall's specimens of *A. adhaerens* (identified by Ritter), borrowed from the Smithsonian Institution National Museum of Natural History, has confirmed that they are morphologically identical. Both Atlantic and Pacific forms are brooders. Thus, *A. adhaerens* remains a synonym under *A. callosa*. *Ascidia columbiana*, on the other hand, was confirmed to be distinct from *A. callosa* and is here reinstated as a valid species. The employment of molecular techniques such as 18S rDNA markers (Wada et al. 1992; C.S. Cohen et al. 1998; Swalla et al. 2000) might be of great help in resolving taxonomic difficulties such as distinguishing these *Ascidia* species. New techniques, described by Yue and Orban (2001), are becoming available to analyze the DNA of formalin-fixed museum specimens, not previously considered utilizable.

*Molgula retortiformis* and *A. callosa* are the only species collected on this expedition that are considered native to both the North Pacific and North Atlantic (Van Name 1945). *Distaplia occidentalis*, *D. alaskensis*, *C. productum*, *C. inflata* and *C. willmeriana*, *P. haustor*, and *S. truncata* have been recorded only from the northeast Pacific. *Halocynthia igaboja* is known from both the northwest and northeast Pacific. *Botrylloides violaceus* is a recent invader of the northeast and northwest Pacific, North Atlantic, and Mediterranean Sea.

## Acknowledgments

We thank the many members of the SERC team who participated in the Alaska expedition, particularly the principal investigators, Drs. A.H. Hines and G.M. Ruiz, without whose help this collection would not have been possible. Financial support for the SERC study was provided by the Regional Citizens' Advisory Council of Prince William Sound, the

U.S. Fish and Wildlife Service, and a National Sea Grant from the National Oceanic and Atmospheric Administration. We are indebted to C. Lambert for his willing and able assistance with collecting, microscopy, and critical reading of the manuscript, to G. Sonnevill for collecting additional specimens of *D. alaskensis*, and to M. Mottet for providing the specimens from Sitka Sea Farm. L. Cole facilitated the loan of specimens from the Smithsonian Institution National Museum of Natural History in Washington, D.C., G. Pohle the specimens from the Atlantic Reference Centre, Huntsman Marine Science Centre, St. Andrews, New Brunswick, and M. Zubowski the type specimen of *A. columbiana* from the Royal Ontario Museum in Toronto. We also thank N. Sanamyan for her excellent drawings of some specimens. K. Sanamyan gratefully acknowledges the Russian Foundation of Basic Researches for personal grant No. 00-04-48727. G.M. Ruiz and an anonymous reviewer are warmly thanked for their numerous improvements to the manuscript.

## References

- Abbott, D.P. 1966. The ascidians. In *Environment of the Cape Thompson Region, Alaska*. Chap. 30. Edited by N.J. Wilimovsky and J.N. Wolfe. U.S. Atomic Energy Commission, Washington, D.C. pp. 839–841.
- Ärnäck Christie-Linde, A. 1922. Tunicata 1. Styelidae and Polyzoidae. In *Northern and Arctic invertebrates in the collection of the Swedish State Museum*. K. Sven. Vetenskapsakad. Handl. **63**(2): 1–62.
- Ärnäck Christie-Linde, A. 1928. Tunicata 3. Molgulidae and Pyuridae. In *Northern and Arctic invertebrates in the collection of the Swedish State Museum*. K. Sven. Vetenskapsakad. Handl. **49**(9): 1–99.
- Ärnäck Christie-Linde, A. 1934. Tunicata 4. Cionidae, Ascidiidae, Agnesiidae, Rhodosomatidae. In *Northern and Arctic invertebrates in the collection of the Swedish State Museum*. K. Sven. Vetenskapsakad. Handl. **13**(3): 1–91.
- Bancroft, F.W. 1899. Orogenesis in *Distaplia occidentalis* Ritter (M.S.), with remarks on other species. Bull. Mus. Comp. Zool. **35**: 59–112.
- Bates, W.R. 1995. Direct development in the ascidian *Molgula retortiformis* Verrill, 1871. Biol. Bull. (Woods Hole, Mass.), **188**: 16–22.
- Berrill, N.J. 1931. Studies in tunicate development. Part II. Abbreviation of development in the Molgulidae. Philos. Trans. R. Soc. Lond. B Biol. Sci. **219**: 281–346.
- Carson, R. 1980. The Huntsman collection (Urochordata: Ascidiacea) at the Royal Ontario Museum, with an annotated catalogue of Huntsman's types from British Columbia. R. Ont. Mus. Life Sci. Misc. Publ. pp. 1–18.
- Cohen, A., Mills, C., Berry, H., Wonham, M., Bingham, B., et al. 1998. Report of the Puget Sound Expedition, Sept. 8–16, 1998: a rapid assessment survey of non-indigenous species in the shallow waters of Puget Sound. Washington State Department of Natural Resources, Olympia.
- Cohen, C.S., Saito, Y., and Weissman, I.L. 1998. Evolution of allorecognition in botryllid ascidians inferred from a molecular phylogeny. Evolution, **52**: 746–756.
- Fay, R.C., and Vallee, J.A. 1979. A survey of the littoral and sublittoral ascidians of southern California, including the Channel Islands. Bull. S.C. Acad. Sci. **78**: 122–135.
- Hartmeyer, R. 1923. Ascidiacea. In *Danish Ingolf Expedition*. Vol. 2. Part 6. Zoological Museum, Copenhagen.
- Hartmeyer, R. 1924. Ascidiacea. In *Danish Ingolf Expedition*. Vol. 2. Part 7. Zoological Museum, Copenhagen.
- Herdman, W.A. 1898. Description of some simple ascidians collected in Puget Sound, Pacific coast. Trans. Liverpool. Biol. Soc. **12**: 248–267.
- Hines, A.H., and Ruiz, G.M. 2000. Biological invasions of cold-water coastal ecosystems: ballast-mediated introductions in Port Valdez/Prince William Sound, Alaska. Final project report, 15 March 2000, for the Smithsonian Environmental Research Center, Edgewater, Md.
- Huber, J.L., da Silva, K.B., Bates, W.R., and Swalla, B.J. 2000. The evolution of anural larvae in molgulid ascidians. Semin. Cell Dev. Biol. **11**: 419–426.
- Huntsman, A.G. 1912a. Ascidians from the coasts of Canada. Trans. R. Can. Inst. **9**: 111–148.
- Huntsman, A.G. 1912b. Holosomatous ascidians from the coast of western Canada. Contrib. Can. Biol. 1906–1910. pp. 103–185.
- Huntsman, A.G. 1922. Arctic ascidians. In *Report of the Canadian Arctic Expedition 1913–1918*, Vol. 6. 3B–14B. F.A. Acland, Ottawa, Ont.
- Huus, J. 1930. Ascidiologische Notizen. I. Zur Verbreitung, Morphologie und Biologie von *Ascidia callosa* Stps. Bergens Mus. Arb. 1930, Nat. rekke no. 5. pp. 1–11.
- Kott, P. 1985. The Australian Ascidiacea. Mem. Queensl. Mus. **23**: 1–440.
- Lambert, C.C. 1970. Genetic transcription during the development and metamorphosis of the tunicate *Ascidia callosa*. Ph.D. thesis, University of Washington, Seattle.
- Lambert, C.C., and Lambert, G. 1978. Tunicate eggs utilize ammonium ions for flotation. Science (Washington, D.C.), **200**: 64–65.
- Lambert, C.C., and Lambert, G. 1998. Non-indigenous ascidians in southern California harbors and marinas. Mar. Biol. (Berl.), **130**: 675–688.
- Lambert, C.C., Lambert, G., and Kozloff, E.N. 1987. Phylum Urochordata. In *Marine invertebrates of the Pacific Northwest*. Chap. 23. Edited by E.N. Kozloff. University of Washington Press, Seattle. pp. 467–479.
- Lambert, C.C., Lambert, I.M., and Lambert, G. 1995. Brooding strategies in solitary ascidians: *Corella* species from north and south temperate waters. Can. J. Zool. **73**: 1666–1671.
- Lambert, G. 1968. The general ecology and growth of a solitary ascidian, *Corella willmeriana*. Biol. Bull. (Woods Hole, Mass.), **135**: 296–307. [The correct name is now *C. inflata*.]
- Lambert, G. 2000. Focal taxonomic collections: ascidians. In *Biological invasions of cold-water coastal ecosystems: ballast-mediated introductions in Port Valdez/Prince William Sound, Alaska*. Edited by A.H. Hines and G.M. Ruiz. Final project report, 15 March 2000, for the Smithsonian Environmental Research Center, Edgewater, Md. pp. 9C10 1–7.
- Lambert, G., Lambert, C.C., and Abbott, D.P. 1981. *Corella* species in the American Pacific Northwest: distinction of *C. inflata* Huntsman, 1912 from *C. willmeriana* Herdman, 1898 (Ascidiacea, Phlebobranchia). Can. J. Zool. **59**: 1493–1504.
- Manni, L., Zaniolo, G., and Burighel, P. 1995. Oogenesis and oocyte envelope differentiation in the viviparous ascidian *Botrylloides violaceus*. Invertebr. Reprod. Dev. **27**: 167–180.
- Marin, M.G., Bressan, M., Beghi, L., and Brunetti, R. 1987. Thermal tolerance of *Ciona intestinalis* (L., 1767) at different developmental stages. Cah. Biol. Mar. **28**: 47–57.
- Michaelsen, W. 1923. Neue und altbekannte Asciden aus dem Reichsmuseum zu Stockholm. Mitt. Zool. Mus. Hamb. **40**: 1–60.
- Millar, R.H. 1966. Tunicata Ascidiacea. Mar. Invertebr. Scand. **1**: 1–123.

- Nishikawa, T. 1991. The ascidians of the Japan Sea. II. Publ. Seto Mar. Biol. Lab. **35**: 25–170.
- O'Clair, R.M., and O'Clair, C.E. 1998. Southeast Alaska's rocky shores. Plant Press, Auke Bay, Alaska.
- Oka, A. 1906. Notizen über japanischen Ascidien I. Annot. Zool. Jpn. **6**: 37–52.
- Oka, A. 1927. Zur Kenntnis der japanischen Botryllidae (Vorläufige Mitteilung). Proc. Imp. Acad. Jpn. **3**: 607–609.
- Ritter, W.E. 1899. A contribution to the knowledge of the tunicates of the Pribilof Islands. In The fur seals and fur-seal islands of the North Pacific Ocean. Part III. Government Printing Office, Washington, D.C. pp. 511–537.
- Ritter, W.E. 1901. Papers from the Harriman Alaska Expedition. XXIII. The ascidians. Proc. Wash. Acad. Sci. **3**: 225–266.
- Ritter, W.E. 1907. The ascidians collected by the United States Fisheries Bureau steamer *Albatross* on the coast of California during the summer of 1904. Univ. Calif. Publ. Zool. **4**: 1–52.
- Ritter, W.E. 1913. The simple ascidians from the northeastern Pacific in the collection of the United States National Museum. Proc. U.S. Natl. Mus. **45**: 427–505.
- Ritter, W.E., and Forsyth, R.A. 1917. Ascidians of the littoral zone of southern California. Univ. Calif. Publ. Zool. **16**: 439–512.
- Saito, Y., Mukai, H., and Watanabe, H. 1981. Studies on Japanese compound styelid ascidians II. A new species of the genus *Botrylloides* and redescription of *B. violaceus* Oka. Publ. Seto Mar. Biol. Lab. **26**: 357–368.
- Sanamyan, K. 1993a. Ascidians from the north-western Pacific region. 1. Polycitoridae. *Ophelia*, **37**: 163–173.
- Sanamyan, K. 1993b. Ascidians from the north-western Pacific region. 2. Molgulidae. *Ophelia*, **38**: 127–135.
- Sanamyan, K. 1996. Ascidians from the north-western Pacific region. 3. Pyuridae. *Ophelia*, **45**: 199–210.
- Sanamyan, K. 1997. Ascidians of the Commander Islands. [In Russian with English summary.] In Benthic flora and fauna of the shelf zone of the Commander Islands. Edited by A. Rzhavsky. Dalnauka, Vladivostok, Russia. pp. 254–261.
- Sanamyan, K. 1998a. Ascidians from the north-western Pacific region. 4. Polyclinidae and Placentelidae. *Ophelia*, **48**: 103–135.
- Sanamyan, K. 1998b. Ascidians from the north-western Pacific region. 5. Phlebobranchia. *Ophelia*, **49**: 97–116.
- Sanamyan, K. 1999. Ascidians from the north-western Pacific region. 6. Didemnidae. *Ophelia*, **51**: 143–161.
- Sanamyan, K. 2000. Ascidians from the north-western Pacific region. 7. Styelidae. *Ophelia*, **53**: 67–78.
- Sims, L.L. 1984. Osmoregulatory capabilities of three macro-sympatric stolidobranch ascidians, *Styela clava* Herdman, *S. plicata* (Lesueur), and *S. montereyensis* (Dall). J. Exp. Mar. Biol. Ecol. **82**: 117–129.
- Stimpson, W. 1852. Several new ascidians from the coast of the United States. Proc. Boston Soc. Nat. Hist. **4**: 228–232.
- Stimpson, W. 1864. Descriptions of new species of marine Invertebrata from Puget Sound, collected by the naturalists of the North-west Boundary Commission. Proc. Acad. Nat. Sci. **16**: 153–161.
- Swalla, B.J., Cameron, C.B., Corley, L.S., and Garey, J.R. 2000. Urochordates are monophyletic within the deuterostomes. Syst. Biol. **49**: 52–64.
- Tokioka, T. 1953. Ascidians of Sagami Bay. Iwanami Shoten, Tokyo.
- Tokioka, T. 1958. Contribution to Japanese ascidian fauna. 12. Sporadic memoranda. Publ. Seto Mar. Biol. Lab. **6**: 313–326.
- Tokioka, T. 1962. Contribution to Japanese ascidian fauna. 19. Additions to Japanese ascidian fauna, with notes on two already known species. Publ. Seto Mar. Lab. **10**: 259–282.
- Van Name, W.G. 1945. The North and South American ascidians. Bull. Am. Mus. Nat. Hist. **84**: 1–476.
- Vázquez, E., and Young, C.M. 1996. Responses of compound ascidian larvae to haloclines. Mar. Ecol. Prog. Ser. **133**: 179–190.
- Vázquez, E., and Young, C.M. 2000. Effects of low salinity on metamorphosis in estuarine colonial ascidians. Invertebr. Biol. **119**: 433–444.
- Verrill, A.E. 1871. Descriptions of some imperfectly known and new ascidians from New England. Am. J. Sci. Ser. 3, **1**: 54–58, 93–100, 211–212, 288–294, 443–446.
- Wada, H., Makabe, K.W., Nakauchi, M., and Satoh, N. 1992. Phylogenetic relationships between solitary and colonial ascidians, as inferred from the sequence of the central region of their respective 18S rDNAs. Biol. Bull. (Woods Hole, Mass.), **183**: 448–455.
- Yue, G.H., and Orban, L. 2001. Rapid isolation of DNA from fresh and preserved fish scales for PCR. Mar. Biotechnol. **3**: 199–204.

## Appendix.

Collection records, 1998–1999.

### Kenai Peninsula

VIII-08-1999, Homer Marina, Kachemak Bay, Cook Inlet (salinity 27‰ at 0.3 m depth, 30‰ at 2 m depth; 10°C).

Most of the following records are from suspended ropes.

*Ascidia callosa*: common, especially about halfway between the inner and outer ends of the marina. Some contain brooded larvae in the atrial chamber. Two small specimens were collected from fouling panels.

*Distaplia alaskensis*: very abundant at most locations in the marina, especially on ropes 1–2 m deep. A few small colonies were collected from one of the fouling panels.

*Molgula retortiformis*: 2 large and 8 small specimens, on a rope and on lower side of float. Two small specimens on fouling plate E1.

*Styela truncata*: 1 small individual.

VIII-09-1999, Seward Marina (salinity 11‰ at 0.3 m depth; 11.5°C).

No ascidians either at the marina or Lowell Point rocky intertidal (11‰, 11.5°C).

### Prince William Sound

VIII-10-1999, Whittier Marina. No salinity or temperature readings were taken. Salinity very low; no ascidians on floats.

VIII-10-1999, Fairmont Bay oyster farm (salinity 25‰ at 0.3 m depth; 14°C).

*Ascidia columbiana*: numerous small specimens on concrete blocks of the fouling panels and one large one on bottom of submerged 4 ft long oyster bag.

VIII-11-1999, Valdez Marina. No salinity or temperature readings were taken; no ascidians were observed on the marina floats or suspended ropes.

VIII-11-1999–VIII-12-1999, Tatitlek (salinity 27‰ at 0.3 m depth; 17°C, surface).

(i) SERC fouling panels and frames, 3 m depth:

*Ascidia columbiana*: a few small specimens on the concrete blocks for the fouling panels and one on panel PL3-1.

*Corella inflata*: common on both the panels and frames at about 3 m depth, with brooded larvae in the atrial chamber.

*Botrylloides violaceus*: numerous very small zooids on fouling panels including PL3-1.

(ii) Intertidal rocks, low tide, morning.

*Ascidia columbiana*: 1 large specimen collected by J. Goddard.

*Chelyosoma productum*: about 12 specimens seen, 2 collected.

*Halocynthia igaboja*: 1 very small immature specimen collected by J. Goddard.

*Pyura haustor*: 4 specimens seen in deep rock crevices, 1 incomplete specimen collected.

(iii) Oyster cages from oyster-culture facility across the bay: 1 small *C. inflata*.

VIII-13-1999, Cordova Marina (salinity 27‰ at 0.3 m depth, 13°C, surface, low tide).

*Distaplia alaskensis*: abundant colonies with many mature larvae.

*Distaplia occidentalis*: common; several color morphs, including purple and orange-tan, large heads, with mature larvae.

*Ascidia callosa*: numerous specimens on floats and suspended ropes. Some contain brooded larvae in atrial chamber.

*Corella inflata*: large, common, full of brooded larvae in atrial chamber.

*Styela truncata*: numerous specimens on floats and ropes. Some contain brooded larvae in atrial chamber.

SERC fouling panels

*Distaplia alaskensis*: numerous small colonies.

*Ascidia callosa*: 12 small specimens.

*Corella inflata*: 1 small specimen.

*Corella willmeriana*: several specimens.

*Styela truncata*: 8 specimens.

VIII-14-1999, SERC Valdez fouling panels, 6.5 m depth.

*Ascidia columbiana*: 1 small specimen.

*Corella willmeriana*: 3 individuals, 1 large and 2 small.

#### Additional SERC ascidian vouchers

Homer Marina, Kachemak Bay, Cook Inlet, IX-03-1998, 1 *A. callosa* and 1 *S. truncata* on fouling panels.

Homer Marina, X-1998, *D. alaskensis* collected by G. Sonnevil from floats. Colonies without gonads or larvae.

Homer Marina, V-11-1999, *D. alaskensis* collected by G. Sonnevil from floats. Colonies small, immature; no gonads or larvae.

Port San Juan, Chenega small-boat harbor, IX-07-1998, 1 *A. callosa* and 2 *C. inflata*.

Port San Juan, Chenega small-boat harbor, no date (but probably IX-07-1998), 7 *C. inflata*, sample MI-05-1-?

Valdez, IX-08-1998, 2 *C. inflata* at Alyeska berth 5.