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FINAL CRUISE REPORT

SUBMERSIBLE AND SCUBA COLLECTIONS IN THE COASTAL WATERS OF
THE BAHAMA ISLANDS AND FLORIDA'S OCULINA CORAL BANKS:
BIOMEDICAL AND BIODIVERSITY RESEARCH OF THE BENTHIC
COMMUNITIES WITH EMPHASIS ON PORIFERA AND GORGONACEA

OCTOBER 25 - 31, 1996
Research Vessel EDWIN LINK
JOHNSON-SEA-LINK II Research Submersible

Submitted to: The Government of the Bahama Islands

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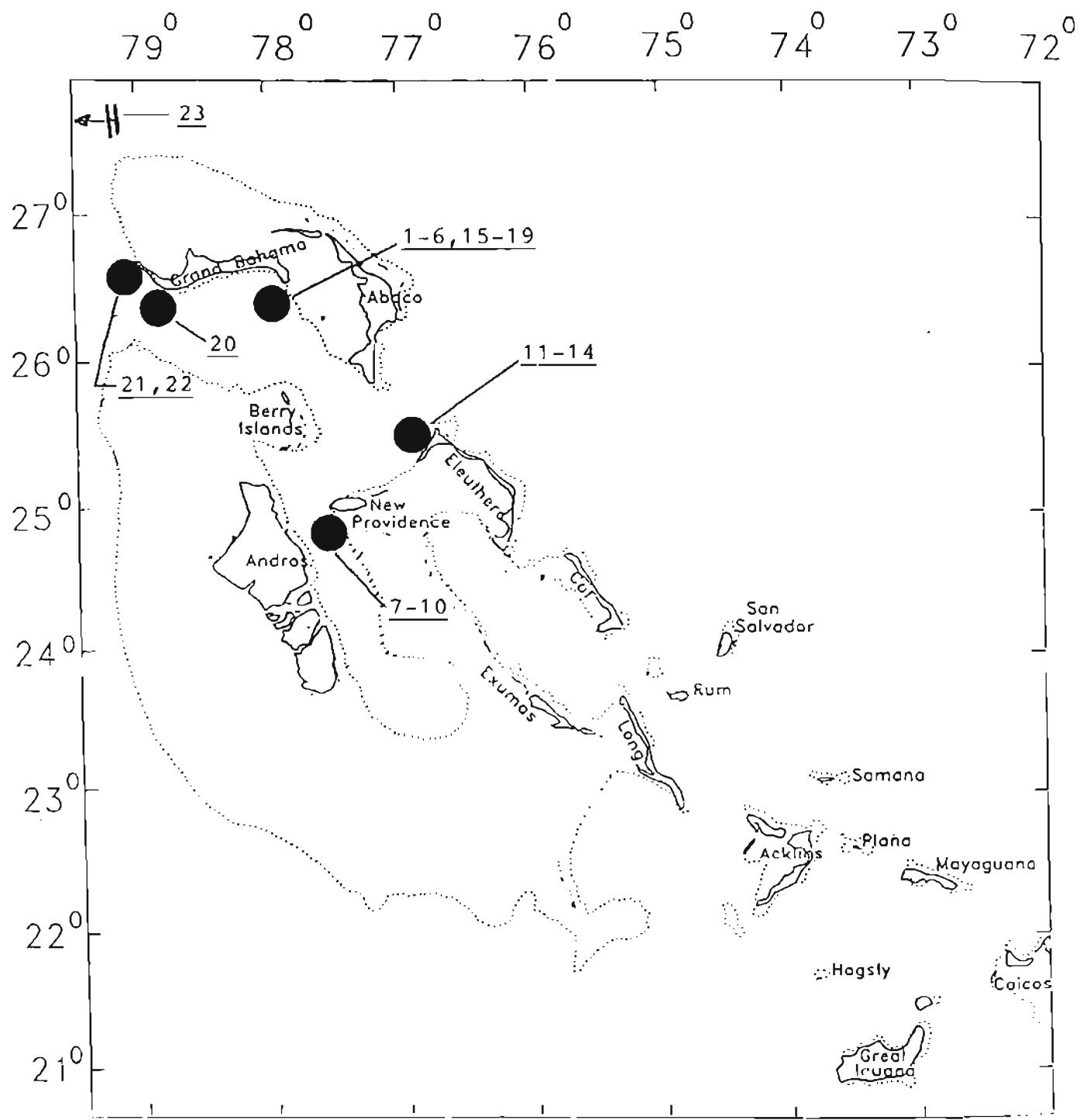


Figure 1. Collection Sites of Research Vessel EDWIN LINK
Bahama Islands: October 25 - 31, 1996

● = Submersible and scuba dive sites
 (Florida dive site not shown)

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ACKNOWLEDGMENTS

We thank the crews of the ship and submersible whose efforts made this cruise possible. All the scientists are especially thanked for their help in making this a very successful expedition.

We gratefully acknowledge the Bahamian government for providing the opportunity for this fundamentally important research in their territorial waters.

This is Harbor Branch Oceanographic Institution's Miscellaneous Publication Number 327.

SUMMARY

Division of Biomedical Marine Research (DBMR) scientists [Dr. Shirley Pomponi (Principal Investigator; Director, Division of Biomedical Marine Research), John Reed (Chief Scientist; Dive Safety Officer), Dr. Amy Wright, Robin Willoughby, Kathleen Janda, and Dr. Susan Sennett] and visiting scientists [Dr. Michelle Kelly-Borges, Dr. James McInerney, Dr. David Myles, Christi Adams, and Klaus Kelly-Borges] conducted a 7 day research expedition to the Bahama Islands on the R/V EDWIN LINK from October 25 to 31, 1996. One submersible dive was also conducted on the deep-water *Oculina* coral banks off central eastern Florida. The primary objectives were: 1) to conduct biochemical and DNA research on targeted bioactive sponges, 2) to collect marine invertebrates for discovery of novel compounds with therapeutic potential; 3) to document the biodiversity of deep-water benthic communities with videotapes, 35-mm photographs, and museum specimens using the JOHNSON-SEA-LINK II Research Submersible; 4) to isolate heterotrophic microorganisms for culture; and 5) to procure living specimens of sponges and tunicates for cell culture research.

A total of 12 submersible dives and 11 scuba/snorkel dives were conducted at 23 dive sites. The expedition resulted in the collection of 123 samples of macroinvertebrates, 81 microbial samples, and 14 samples for cell culture. These samples will be used in DBMR's research program with the purpose of identifying potential pharmaceutical agents for the treatment of cancer, microbial diseases, diseases of the immune system, and other therapeutic agents.

OBLIGATION TO HOST COUNTRY

In agreement with the Bahamian government, a permit was requested and received from the Department of Fisheries, Ministry of Agriculture and Fisheries, Nassau, Bahamas, to conduct this scientific research. All provisions of the permit have been fulfilled which include the following:

- 1) This final cruise report is being submitted to the Bahamas Department of Fisheries within three months of completing the cruise.
- 2) Duplicate museum specimens, underwater photographs and videotapes taken from the submersible of the benthic habitats and specimens will be made available to the Bahamas Department of Fisheries upon request within 6 months of the cruise.
- 3) Three copies of all published papers, reports, or books that may arise as a result of the research conducted during this cruise will be submitted to the Bahamas Department of Fisheries.
- 4) All other provisions of the permit were fulfilled.

SCIENTIFIC PARTICIPANTS

Division of Biomedical Marine Research (DBMR), HBOI

Dr. Shirley Pomponi	Principal Investigator; Director, DBMR
John K. Reed	Chief Scientist, Dive Safety Officer, DBMR
Dr. Amy Wright	Natural Products Chemistry, DBMR
Kathleen Janda	Microbiology, DBMR
Robin Willoughby	Invertebrate Cell Culture, DBMR
Dr. Susan Sennett	Biochemistry, DBMR
Dr. Michelle Kelly-Borges	Natural History Museum, London
Dr. James McInerney	Natural History Museum, London
Christi Adams	Natural History Museum, London
Klaus Kelly-Borges	Natural History Museum, London
Dr. David Myles	University of California, LA

OBJECTIVES

1. Observe, photograph, and collect selected marine invertebrates and macroalgae with the JOHNSON-SEA-LINK Research Submersible and scuba diving.
2. Document biodiversity of benthic communities with videotapes, 35-mm photographs, and taxonomic museum specimens using the JOHNSON-SEA-LINK Research Submersible and scuba diving.
3. Isolate and incubate microorganisms from macroinvertebrate and sediment samples.
4. Prepare samples of marine macroorganisms and microorganisms for subsequent bioassays and isolation of novel bioactive chemical compounds.
5. Collect live specimens of sponges and tunicates for cell culture research.
6. Recollect targeted deep-water sponges for DNA analyses and isolation research on bioactive compounds.

PURPOSE

Harbor Branch Oceanographic Institution (HBOI) is a private, not-for-profit research institution. This research will complement submersible and scuba collections and subsequent taxonomic and biomedical research conducted by HBOI elsewhere in the Bahamas and the Caribbean. This research will greatly add to the body of knowledge available on the biodiversity and resources of the deep reefs and deep island slopes of Puerto Rico. Basic research will be conducted on biodiversity, systematics, ecology, and physiology of the samples observed and collected. The results of studies on the bioactivity and

chemistry of novel compounds isolated from these samples may be applied in the long term to the development of compounds used to diagnose or treat human diseases, including cancer, microbial diseases, and diseases of the immune system, or used as biomedical research tools.

METHODS

1. Collections- Samples were collected in shallow water (0-50 m) by wading, snorkeling, and scuba; and in deep water (50-914 m) with the JOHNSON-SEA-LINK II (JSL) Research Submersible (Appendix 1). The JSL submersible was equipped with a manipulator arm which included clam-shell grab, jaws, and suction hose; 12-bin rotating basket; color video camera; 35-mm camera; and a data recorder (Seabird) to log temperature, conductivity, salinity, and depth.
2. Number of Samples- Marine algae, invertebrates (primarily sponges, gorgonians, and tunicates), and microorganisms were collected and assigned a HBOI/DBMR sample number. At each site approximately 1-2 submersible dives and 1-2 scuba dives were made. An average of 5-12 samples were collected at each dive site.
3. Environmental Impact- Collections by submersible and scuba were highly selective. Species protected by the CITES act for endangered species were avoided. No hard coral or commercially important black coral were collected.
4. Ship and Sample Positions- Collection site coordinates were determined with GPS navigation (Magnavox MX 200 Global Positioning System) which may be accurate to ~100 feet. Plots of the submersible tracks and specific sample sites were made with the Integrated Mission Profiler, a software database developed by Florida Atlantic University that ties into the ship's GPS system. Scuba and snorkel sites were documented in the field using a hand held GPS navigator (Garmin 45). Coordinates were plotted using "The Bahamas Chart Kit" (1984 edition, Better Boating Association).
5. Site and Sample Documentation- Collection site descriptions, including latitude, longitude, habitat, depth, temperature, current, and weather conditions; along with sample descriptions, including morphology, color, abundance, taxonomy, and photographic reference for each sample were transcribed to 100% rag cotton paper in a fieldbook. All data were entered into DBMR's proprietary database (ACCESS 2.0).
6. Photography- Samples were photographed *in-situ* using the JSL's Benthos camera with 35-mm lens, Ektachrome ASA 64

film, and laser sighting. Color videotapes (Fuji High 8-mm) were recorded with the JSL's pan and tilt videocamera (Sony DX2 3000A with Canon J8X6B KRS lens, 6-48 mm zoom, and 0.3 m minimum focus) which has four laser dots (17.5 cm apart along the edge of the diamond shape) for scale. Some of the scuba samples were photographed *in-situ* with a Nikonos V camera, 15-mm lens, Nikonos 103 strobe, and Kodachrome 64 film. Each sample was assigned a sample number, and was photographed with a ruler for scale against a grey background in the ship's laboratory using a 35-mm Olympus OM4T camera, 50-mm macro lens, Olympus T-20 flash with a Vivitar 2800 slave flash, and Ektachrome ASA 100 film. Submersible and lab photographs were developed with E-6 kits on board the ship. Original 35-mm slide photographs and videotapes are stored in DBMR's Photographic Library at HBOI.

7. Taxonomy- Taxonomic identifications of the specimens were made on board the ship by the following individuals:

Sponges - Shirley Pomponi, Michelle Kelly-Borges
General Invertebrates- John Reed

Microscopic slide mounts were made when necessary for identifications of sponges, gorgonians, and tunicates. Spicule slide mounts were made for most sponges and all gorgonians.

8. Museum Specimens- Museum voucher specimens were subsampled from most samples and stored in 20 dram scintillation vials for small vouchers and in 2-16 oz glass jars for large vouchers. Algal samples were fixed and preserved in 5% formalin. Chordates (ascidians and salps) were relaxed in seawater with menthol crystals and stored in 10% formalin. All other specimens were preserved in 70% ethanol. Some duplicate specimens were not vouchered. Voucher samples are stored in DBMR's Reference Museum at HBOI.
9. Cell Culture- (R. Willoughby)- Sponge and tunicate samples were collected for cell dissociation, cell-type enrichment, culture, and cryopreservation. Tissue samples were dissociated into cell suspensions and separated by cell type using density gradient centrifugation. Cell fractions were used to inoculate culture flasks for transport back to the DBMR's Cell Culture Laboratory at HBOI. Sponge cell fractions were also cryopreserved at -80°C.
10. Extractions- 5-10 gram subsamples of each sample were extracted with 100% ethanol and were pulverized with a Virtis grinder for subsequent evaluation of bioactivity.

11. Chemistry (A. Wright) - Deep-water samples collected by submersible were analyzed on board for the presence of novel chemical compounds using thin layer chromatography in three solvent systems. A small subsample of organism (~1 mm³) was crushed in a vial with ethyl acetate-methanol (2:1). Samples were chromatographed on thin-layer plates with three solvent systems: ethyl acetate-heptane(1:1), ethyl acetate-methanol (19:1), and n-propanol-ethyl acetate-water (7:2:1). The plates were then developed by heating with 2% vanillin in H₂SO₄ and analyzed.
12. Microorganism Isolation (K. Janda) - Heterotrophic microorganisms were collected from various macroinvertebrate and sediment samples, especially targeting deep-water samples. For sponges, a 1-3 mm piece of choanosome was used. Samples were pulverized and cultured on differential, selective media and/or stored frozen in 40% glycerol-seawater solution. Individual colonies of microorganisms will be isolated to axenic strains in DBMR's Fermentation Laboratory at HBOI.
13. Compound Localization Studies (S. Sennett) - Targeted deep-water sponges were subsampled for cell isolation using continuous and discontinuous Percoll gradients. Bacterial layers were separated from various sponge cell layers and will be subsequently tested for biologically active compounds.
14. DNA Analyses (Dr. Michelle Kelly-Borges) - Subsamples were prepared from most of the deep-water sponges for subsequent DNA analyses at the British Natural History Museum in London.
15. Labels - Samples, subsamples and vouchers were labeled with a HBOI/DBMR sample number (Date + Dive # + Sample # - e.g., 06-VI-93-1-001), using Nalgene paper and Sharpie water-resistant pen. Labels for museum specimens were computer generated on Resistall 28# paper.
16. Storage and Shipping - Samples were frozen in Ziplock bags at approximately -10 to -15°C in freezers on board the ship. These were transferred to DBMR's freezers at HBOI.

RESULTS and DISCUSSION

Itinerary

October 25	Ship departs Ft. Pierce, Florida; transits to Freeport, Grand Bahama Island; clear customs and immigration.
October 26-29	Submersible and scuba operations along Little Bahama Bank, Grand Bahama Island, Egg Island and New Providence Island.

October 30	Submersible and scuba operations off Grand Bahama Island; clear customs and immigration at Freeport, Grand Bahama Island; transit to Florida.
October 31	One submersible dive off Florida; transit to Ft. Pierce, Florida; clear customs and immigration.

Collections

The expedition lasted a total of 7 days, including 6 operation days, 1 day of transit and 2 port calls. Collections consisted of 12 submersible and 11 scuba/snorkel dives. Eight scientific divers made a total of 35 individual scuba dives for a total bottom time of 14.3 hours.

Collection Sites

Submersible, scuba and snorkel dives were conducted at 23 sites (Figure 1). Appendix 2 lists the sites, latitude, longitude, JSL dive number, maximum depth, and number of DBMR samples. Appendix 3 gives site descriptions, including location, habitat, substrate, bottom temperature, salinity, visibility, and current. Appendix 4 shows the cruise track of each submersible dive, including launch and on-bottom locations as well as some specific sample locations. Appendix 5 shows plots of the fathometer profiles of the shelf edge topography made by transects of the ship prior to and in the vicinity of the respective submersible dives.

Physical Parameters

For each submersible dive, profiles of temperature vs. depth (Appendix 6) and salinity vs. depth (Appendix 7) were recorded with the JSL data logger. Complete data bases for temperature and salinity profiles and for the submersible tracks are stored on 3 1/2" floppy discs. The data logger records in metric units whereas the submersible depth gauge is in feet; therefore, both units are given in this report accordingly. Surface water temperatures at the offshore sites ranged from 24.4°C to 26.7°C (76-80°F) and were fairly constant to a depth of ~75 m. Below this, temperatures gradually decreased with depth to ~21.5°C at 200 m and 16.2°C at 450 m. Salinities at the submersible sites averaged ~36.3 ppt at the surface, were fairly constant to a depth of 75 m, then increased to ~36.8 ppt at 200 m. Below 200 m, salinities decreased to 36.2 ppt at a depth of 450 m.

Taxonomy and Biodiversity

A total of 123 samples were collected for DBMR's biomedical research, including 116 sponges, 4 tunicates, and 4 gorgonians. Of these, 94 (76%) were deep-water organisms

collected by the submersible. Appendix 8 lists the taxonomy of each sample for each site, method of collection, recollection data, and depth of collection in feet.

A total of 63 taxa were identified. Table 1 lists the complete taxonomy for each species and is sorted by phylum, class, order, family, genus and species. Fifty-seven Porifera were identified from the collections, consisting of 15 orders, 29 families, and 42 genera.

Photography and Video

Documentation for each sample includes the following: submersible videotapes (66 of the deep-water samples documented on video); *in-situ* 35-mm photographs (66 samples documented), and laboratory 35-mm photographs (94 samples documented). Appendix 9 lists the presence or absence of *in-situ* videotapes and 35-mm photographs (*in-situ* and laboratory) for each sample. The original photographs and videotapes are curated at DBMR's Photographic Library at HBOI.

Museum Voucher Specimens

Museum voucher specimens were prepared for most species collected. Appendix 9 lists the presence or absence of both small and large vouchers deposited at DBMR's Reference Museum at HBOI.

Microbiology (Kathleen Janda)

A total of 81 macrorganisms and sediment samples were subsampled for shipboard microorganism isolations (Appendix 9). These were cryopreserved at -80°C on 40% glycerol/artificial seawater for future microbial isolation. Subsamples from 19 samples were also cultured on selective/differential agar plates. Individual colonies of microorganisms will be isolated to axenic strains in DBMR's Fermentation Laboratory at HBOI.

Invertebrate Cell Culture (Robin Willoughby)

Fourteen specimens of marine sponges were dissociated and cryopreserved as raw suspensions and/or enriched cell fractions. All specimens were kept frozen for transport back to HBOI for use in future experiments. Appendix 9 lists the samples used for cell cultures.

Chemical Screening - Thin Layer Chromatography (Dr. Amy Wright)

A total of 71 of the deep-water samples were tested for the presence of interesting chemical compounds using thin layer chromatography (TLC). Appendix 9 lists the samples used for

Table 1. Taxonomic distribution of species collected by Harbor Branch Oceanographic Institution, Division of Biomedical Marine Research (Sorted alphabetically by Phylum, Class, Order, Family, Genus, and Species); POR= Porifera, CNI= Cnidaria, ANN= Annelida, PLA= Platyhelminthes, BRY= Bryozoa, MOL= Mollusca, CRU= Crustacea, ECH= Echinodermata, CHO= Ascidiacea, CHL= Chlorophyta, RHO= Rhodophyta, PHA= Phaeophyta, CYA= Cyanophyta

PHYLUM CLASS		ORDER	FAMILY	TAXONOMY
CHO	ASCIDIACEA			ASCIDIACEA
CHO	ASCIDIACEA	PHLEBOBRANCHIA	PEROPHORIDAE	ECTEINASCIDIA TURBINATA
CHO	ASCIDIACEA +TURBELLARIA	PHLEBOBRANCHIA + POLYCLADIDA	PEROPHORIDAE + PSEUDOCERIDAE	ECTEINASCIDIA TURBINATA + PSEUDOCEROS CROZIERI
CNI	ANTHOZOA	GORGONACEA		GORGONACEA
CNI	ANTHOZOA	GORGONACEA	ACANTHOGORGIIIDAE	ACANTHOGORGIIIDAE
CNI	ANTHOZOA	GORGONACEA	PRIMNOIDAE	PRIMNOIDAE
POR	CALCAREA	LEUCETTIDA	LEUCASCIDAE	LEUCETTA SP.
POR	DEMOSTONGIAE	AGELASIDA	AGELASIDAE	AGELAS SP.
POR	DEMOSTONGIAE	ASTROPHORIDA	ancorinidae	ASTEROPUS SP.
POR	DEMOSTONGIAE	ASTROPHORIDA	GEODIIDAE	CAMINUS SP.
POR	DEMOSTONGIAE	ASTROPHORIDA	GEODIIDAE	ERYLUS cf. GOFRILLERI
POR	DEMOSTONGIAE	ASTROPHORIDA	GEODIIDAE	ERYLUS SP.
POR	DEMOSTONGIAE	ASTROPHORIDA	GEODIIDAE	GEODIA NEPTUNI
POR	DEMOSTONGIAE	ASTROPHORIDA	GEODIIDAE	GEODIA SP.
POR	DEMOSTONGIAE	ASTROPHORIDA	JASPIDAE	STELLETTINOPSIS SP.
POR	DEMOSTONGIAE	ASTROPHORIDA	STELLETTIDAE	STELLETTA SP.

PHYLUM	CLASS	ORDER	FAMILY	TAXONOMY
POR	DEMOSTONGIAE	ASTROPHORIDA	STELLETTIDAE	STELLETTIDAE
POR	DEMOSTONGIAE	AXINELLIDA	AXINELLIDAE	AXINELLIDAE
POR	DEMOSTONGIAE	AXINELLIDA	AXINELLIDAE	PHAKELLIA FOLIUM
POR	DEMOSTONGIAE	AXINELLIDA	AXINELLIDAE	PSUEDAXINELLA LUNAECHARTA
POR	DEMOSTONGIAE	AXINELLIDA	AXINELLIDAE	PTILOCAULIS SPICULIFER?
POR	DEMOSTONGIAE	AXINELLIDA	AXINELLIDAE	TEICHAXINELLA MORCHELLA
POR	DEMOSTONGIAE	AXINELLIDA	BUBARIDAE	BUBARIS SP.
POR	DEMOSTONGIAE	AXINELLIDA	RASPAILIIDAE	ECTYOPLASIA FEROX
POR	DEMOSTONGIAE	AXINELLIDA	RASPAILIIDAE	RASPAILIIDAE
POR	DEMOSTONGIAE	DENDROCERATIDA	HALISARCIDAE	HALISARCA? SP.
POR	DEMOSTONGIAE	DICTYCERATIDA	SPONGIIDAE	HIPPOSPONGIA SP.
POR	DEMOSTONGIAE	HADROMERIDA	TIMEIDAE	WILLARDIA CAICOSENSIS
POR	DEMOSTONGIAE	HALICHONDRIDA	HALICHONDRIIDAE	EPIPOLASIS SP.
POR	DEMOSTONGIAE	HALICHONDRIDA	HALICHONDRIIDAE	HALICHONDRIA CORRUGATA
POR	DEMOSTONGIAE	HALICHONDRIDA	HALICHONDRIIDAE	MYRMEKODERMA SP.
POR	DEMOSTONGIAE	HALICHONDRIDA	HALICHONDRIIDAE	SPONGOSORITES SP.
POR	DEMOSTONGIAE	HALICHONDRIDA	HALICHONDRIIDAE	TOPSENTIA PORRECTA?
POR	DEMOSTONGIAE	HALICHONDRIDA	HALICHONDRIIDAE	TOPSENTIA SP.
POR	DEMOSTONGIAE	HALICHONDRIDA or AXINELLIDA	HALICHONDRIIDAE?	HYMENIACIDON SP. or AXINELLIDAE
POR	DEMOSTONGIAE	HAPLOSCLERIDA	CALLYSPONGIIDAE	CALLYSPONGIA VAGINALIS

PHYLUM	CLASS	ORDER	FAMILY	TAXONOMY
POR	DEMO SPONGIAE	HAPLOSCLERIDA	NIPHATIDAE	AMPHIMEDON COMPRESSA
POR	DEMO SPONGIAE	HAPLOSCLERIDA	NIPHATIDAE	CRIBROCHALINA VASCULUM
POR	DEMO SPONGIAE	HAPLOSCLERIDA	NIPHATIDAE	SIPHONODICTYON MUCOSUM
POR	DEMO SPONGIAE	HAPLOSCLERIDA	OCEANAPIIDAE	OCEANAPIA SP.
POR	DEMO SPONGIAE	HAPLOSCLERIDA	OCEANAPIIDAE? or NIPHATIDAE?	OCEANAPIIDAE? or NIPHATIDAE?
POR	DEMO SPONGIAE	HAPLOSCLERIDA	PETROSIIDAE	PETROSIIDAE
POR	DEMO SPONGIAE	HAPLOSCLERIDA	PETROSIIDAE	STRONGYLOPHORA SP.
POR	DEMO SPONGIAE	HAPLOSCLERIDA	PETROSIIDAE	XESTOSPONGIA cf. STONEAE
POR	DEMO SPONGIAE	HOMOSCLEROPHORIDA	PLAKINIDAE	CORTICIUM SP.
POR	DEMO SPONGIAE	HOMOSCLEROPHORIDA	PLAKINIDAE	CORTICIUM SP.2
POR	DEMO SPONGIAE	HOMOSCLEROPHORIDA	PLAKINIDAE	PLAKINASTRELLA (DERCITOPSIS) SP.
POR	DEMO SPONGIAE	HOMOSCLEROPHORIDA	PLAKINIDAE	PLAKORTIS SP.
POR	DEMO SPONGIAE	LITHISTIDA	CORALLISTIDAE	CORALLISTES cf. MICROSTYLIFER
POR	DEMO SPONGIAE	LITHISTIDA	SCLERITODERMIDAE or SIPHOIIDAE	ACICULITES SP. or GASTROPHANELLA SP.
POR	DEMO SPONGIAE	LITHISTIDA	THEONELLIDAE	DISCODERMIA cf. VARRUCOSA
POR	DEMO SPONGIAE	LITHISTIDA	THEONELLIDAE	DISCODERMIA SP.
POR	DEMO SPONGIAE	LITHISTIDA	THEONELLIDAE	DISCODERMIA SP.1
POR	DEMO SPONGIAE	LITHISTIDA	THEONELLIDAE	DISCODERMIA SP.IV
POR	DEMO SPONGIAE	LITHISTIDA	VETULINIDAE	VETULINA N.SP.
POR	DEMO SPONGIAE	POECILOSCLERIDA	BIEMNIDAE	BIEMNA SP.

PHYLUM	CLASS	ORDER	FAMILY	TAXONOMY
POR	DEMOSPONGIAE	POECILOSCLERIDA	CLATHRIIDAE	PANDAROS ACANTHIFOLIUM
POR	DEMOSPONGIAE	POECILOSCLERIDA	MYXILLIDAE	IOTROCHOTA SP.
POR	DEMOSPONGIAE	POECILOSCLERIDA	MYXILLIDAE	MYXILLIDAE
POR	DEMOSPONGIAE	POECILOSCLERIDA	TEDANIIDAE	TEDANIA IGNIS
POR	DEMOSPONGIAE	SPIROPHORIDA		SPIROPHORIDA
POR	DEMOSPONGIAE	VERONGIDA	APLYSINELLIDAE	PSEUDOCERATINA CRASSA
POR	SCLEROSPONGIAE	CALCIFIBROSPONGIDA	CALCIFIBROSPONGIIDAE	CALCIFIBROSPONGIA SP.

TLC. These results will be used in conjunction with various biological assays, including tests for compounds which affect cells growing in the laboratory and tests for inhibitors of certain enzyme and receptor "targets". A complete report is on file at DBMR.

Compound Localization Studies (Dr. Susan Sennett)

The objective of this effort is to localize bioactive compound production at the cellular level whether in sponge cells or associated microbes for selected deep-water sponges. The long term goal of this project is to culture or ferment the compound producing sponge cells or microorganisms respectively as an alternative source of compound. Sponges targeted on this cruise were *Discodermia*, *Spongisorites* and *Dercitus*.

Several *Spongisorites* samples were collected and these were used in preliminary attempts to separate bacteria from sponge cells and different sizes and types of sponge cells from each other. Samples were analyzed by TLC to determine which of the known bis-indole metabolites were present. The bright yellow samples all contained topsentin or bromotopsentin. The pale yellow sample (30-X-96-1-6) did not contain topsentin but did contain several more polar compounds in the bis-indole class of compounds (A. Wright, personal communication).

Both continuous and discontinuous Percoll gradients were used to try to establish a system to separate the cell types. Discontinuous gradients were relatively effective in isolating bacteria in the upper layers of the gradient (10-20% Percoll) but the sponge cell fractions at higher percoll concentrations were still heterogeneous. There was some enrichment of the larger cell types (spherulous cells) in the 60% layer and in the pellet. Continuous gradients at the Percoll concentrations used were ineffective in separating cell types.

Cells from two of the samples (29-X-96-1-2 [bright yellow], 30-X-96-1-6 [pale yellow]) were dissociated using standard methods, applied to a 20% Percoll layer to remove bacteria and cryopreserved in 7.5% DMSO at -80° C for further separation studies in the lab. Subsamples were also frozen in CMF and stored at -20° C.

Spongisorites samples used include:

26-X-96-5-2
28-X-96-1-2
28-X-96-2-4
29-X-96-1-2
30-X-96-1-6

Dr. Shirley Pomponi and Robin Willoughby conducted separation studies with *Discodermia* samples. Dr. Pomponi used CMF and

different centrifugation speeds and times to pellet out different cell components of a crude suspension of cells. Pellets were analyzed by TLC for the presence of discodermolide. One of the pellets was embedded for later electron microscopic analysis.

Robin Willoughby had some success with continuous gradients in separating *Discodermia* cells. A major limitation in these studies was the speeds attainable with the centrifuge being used. Further studies will be attempted using continuous Percoll gradients and an ultracentrifuge.

Several large samples of *Discodermia* were dissociated and the crude cell suspensions cryopreserved and stored at -80° C.

DNA Analyses (Dr. Michelle Kelly-Borges)

Ninety of the sponge specimens were subsampled for DNA analyses (Appendix 10). These will be used for systematic studies of various sponge taxa and also used to determine taxonomic affinities of species which contain similar bioactive compounds.

APPENDIX 1

Specifications of Research Vessel EDWIN LINK
and JOHNSON-SEA-LINK II Research Submersible

R/V Edwin Link



R/V EDWIN LINK is a 168-foot former offshore supply vessel that was converted to support marine science research, ocean engineering research, and submersible operations. Her namesake is famed inventor Edwin A. Link. With a 7,000 nautical mile range and cruising speed of 11 knots, the vessel is capable of traveling to any of the world's oceans while accommodating 30 people. An 18-ton A-frame handling system located on the aft deck allows safe submersible launch and recovery up to sea state five.

R/V EDWIN LINK is one of three Harbor Branch owned research vessels that are operated by experienced personnel, expert at launch and recovery procedures, and supported by in-house ocean engineers. With a 360 degree bow thruster and state-of-the-art electronic navigation systems, R/V EDWIN LINK is able to maneuver and position itself with ease and speed, allowing for precision station-keeping. The R/V EDWIN LINK is part of the University-National Oceanographic Laboratory System (UNOLS) fleet.

Typical applications include submersible/ROV support, large towed systems support, deployment and retrieval of moored devices, surface oceanography and hydrographic applications, and diving support with optional recompression chamber facilities.

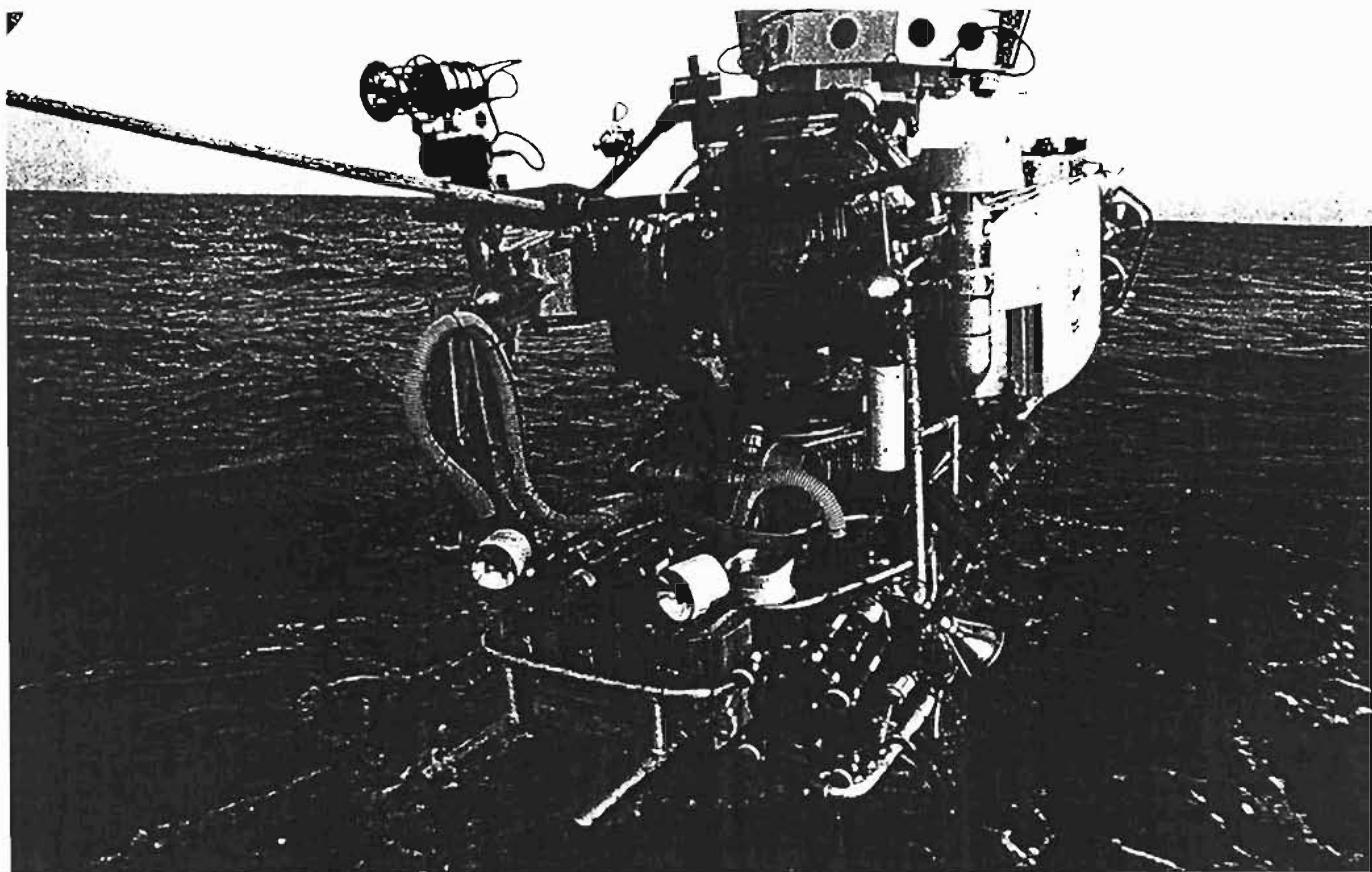
Length Overall	168 feet	Fuel Consumption	85 gal./hour, normal cruise
Length between perpendiculars	156 feet	Potable Water	39,000 Gal. with Reverse Osmosis Unit
Beam, overall	38 feet	150 gal./hr.
Draft	11 feet	Galley Messing	15
Displacement (empty)	781 Short Tons	Speed	11 knots
Gross Tonnage	288 19 Registered Tons	Range	7000 nautical miles
Fuel Capacity	62,000 Gal.	Year Converted	1988

HARBOR BRANCH HAS A SPECIAL FLEET TO MEET YOUR NEEDS

Owned and Operated by Harbor Branch Oceanographic Institution, Inc.
Contact Director, Marine Operations – 407-465-2400 ext. 262/271
FAX – 407-465-2116

Classification:	Furuno FE-D 814 AF 6000' Furuno FM 220 1000'
American Bureau of Shipping; Hull and Machinery	
Normal Compliment	Miscellaneous:
10 ship's crew	Alden Marinefax TR-IV with NAVTEX
20 other (including sub crew, if required)	Simrad L 1520 VHF auto-digital direction finder
Berthing Accommodations:	Deck Equipment:
30 air conditioned berths	A-frame for submersible/towed systems launch and recovery, 18 ton capacity (ABS Certified)
Propulsion:	Deck hydraulic cargo crane - 5-ton capacity
(2) 16V 149 Detroit Diesels (1800 rpm) 900 shp each	Aft Quarter Capstans (2)
Bow Thruster	Anchor windlass, 2 wildcats
360° Rotatable, high volume, axial pump thruster	Sea Mac 88 electro/hydraulic hydrowinch w/3000' 3/16 hydrowire
Powered by Cat 3406-T; 465 hp	Side 'J' Frame - 3500-lb. capability
10,000 lbs. thrust	Avon Rigid Hull Inflatable - 17 ft.; 50 h.p. outboard
Generators:	Willard rigid hull inflatable - 22 ft.; 2 x 70 h.p.
(2) GM 6V 92 Diesel Generators, 175 kw. each	Laboratories:
(1) GM 871 Diesel Generators, 190 kw.	Submersible maintenance lab (264 sq. ft.)
All paralleling	Complete electronics test equipment and electronics spare parts inventory
440, 208 & 110 VAC 3-phase	Submersible Spares, Parts and Supplies/Machine Shop (156 sq. ft.)
Navigation Equipment:	Drill press, lathe, sanders, bench grinder
(1) Integrated mission profiler/navigation system. IBM-PC based; provides differential GPS track for ship, submersibles and ROVs	Compressor room with dive locker
Compasses:	(2) Mako high-pressure air compressors (5000 psi)
Sperry Magnetic Compass	(2) Haskell gas-transfer pumps (O_2 and HeO_2)
Sperry Mark 37 Gyrocompass with 2 repeaters	(2) Delmonox air filters
Sperry Auto-Pilot	(5) T-cylinders (scuba air banks) and 15,000 cu. ft. stored gas, high pressure air (O_2 and HeO_2)
Radar:	Wet Lab (220 sq. ft.)
(2) Furuno 1411	Fresh and salt water
Lorans:	Dry Lab (270 sq. ft.)
Northstar 7000	Fresh and uncontaminated salt water, refrigerator and freezer
Satellite Navigation:	Scientific Storage Room (192 sq. ft.)
(2) Magnavox MX200 GPS	Environmental Labs (2) (36 sq. ft. and 30 sq. ft.)
Data Marine Speed Log	Fresh and uncontaminated salt water
Acoustical Systems:	Fume Hood Room with work bench (38.5 sq. ft.)
Straza ATM 504-14 Underwater Telephone	Lounges:
Straza Model 9010 Passive Tracker	Briefing Room with technical and science library, conference table, 1/2", 3/4" & Hi-8 video recorders, video monitor, and video/photo lab
ORE Track Point Model 4410C Acoustic Position System	Optional Equipment:
Communication Equipment:	Winches:
JRC 45A SATCOM	SEA• MAC Model 3540 EHCLWR
VHFs - Drake (1)	Towing, trawling, coring with optional 14 conductor slipring; drum capacity up to 5,000 feet of .625 wire.
ICOM IC-M120 hand held VHFs (3)	SMATCO Model HCSR-2-100 Hydraulic Storage Reel/Tow Winch; capacity up to 5,300 feet of 2" wire.
Furuno/Skanti - TRP 8258S SSB Transceiver	Recompression Chambers:
Harris - RF 230M SSB	48" double lock; weight-7000 lbs; steel; working pressure-190 psi
NECODE-Encoder/Decoder Model 321 AR	60" double lock; weight-5600 lbs; aluminum; working pressure-100 psi
Sounding Equipment:	NOTE: These recompression chambers can be installed when necessary to support scuba operations.
Data Marine Digital 1000'	
SIMRAD EQ50 video echo sounder 38/50 kHz	

Johnson-Sea-Link I & II



JOHNSON-SEA-LINK I & II, Harbor Branch manned submersibles devoted primarily to research in the marine sciences, are classed and certified to a maximum operating depth of 3,000 feet by the American Bureau of Shipping (ABS). The forward five-inch thick acrylic sphere accommodates the pilot and an observer at "one atmosphere," and allows panoramic visibility. A second crew member and another observer occupy the after observation chamber where a video monitor and side view ports provide forward and side observation.

The evolution of specialized equipment such as manipulator arms, suction devices, and rotary plankton samplers has made it possible for crewmen to accomplish almost any work from within the subs that once was done only by divers. The JOHNSON-SEA-LINK submersibles are further outfitted with such state-of-the-art equipment as active sonar, laser aimed still and broadcast quality video cameras, and Harbor Branch-developed xenon arc lights. The arc lights approximate sunlight, illuminating underwater scenes in true color and near daylight conditions, even in the darkest seas.

These sophisticated and highly maneuverable free swimming submersibles have been in operation since 1971 and 1975. Maintained and operated by experienced and expert pilots and crews, they are further supported by in-house ocean engineers.

Typical applications include benthic and/or mid-water observations, photo/video documentation and collection of organisms; dump site inspections and monitoring; punch and box coring; search and recovery; bottom surveys; photogrammetric surveys; archaeological site documentation and recovery; and environmental impact studies.

HARBOR BRANCH HAS A SPECIAL FLEET TO MEET YOUR NEEDS

Owned and Operated by Harbor Branch Oceanographic Institution, Inc.

Contact Director, Marine Operations – 407-465-2400 ext. 262/271

FAX – 407-465-2116

JOHNSON SEA-LINK'S Capabilities

1. Operate at any depth from the surface to 3000 FSW (914 meters) at speeds of 0-1 kts. (Not to exceed 3000 FSW actual bottom depth.)
2. Carry one observer in forward compartment and one observer in aft compartment.
3. Perform scientific or engineering tasks.
4. Maneuver within close proximity of slopes or other bottom topography.
5. Rest on the bottom to perform tasks.
6. Hover at neutral buoyancy.
7. Normal operations are conducted on a 12 hr. per day basis.
8. The J-S-Ls are highly adaptable work platforms, however, user supplied equipment must be made available at least one working month ahead of scheduled mission to ensure proper interfacing to vehicle.

General Specifications

A. Classified by American Bureau of Shipping		(1) Voltage	24 to 32 VDC
		(2) Current	4 to 16 Amp.
B. Dimensions		b. Output	
1. Overall		(1) Voltage	115VAC +5%
a. Length	23' 7.50" (7.2 m)	(2) Power	0 to 250 VA
b. Beam	8' 3.75" (2.5 m)	(3) Frequency	60Hz = .6 Hz
c. Height	10' 10.9" (3.07m)		
d. Draft	7' 6.75" (2.3m)		
e. Weight	26,000 lbs. (11794 kg)	D. Propulsion	1. Nine Identical Thrusters; 100 lbs. thrust each; reversible
f. Payload	1,000 lbs (453 kg)		
g. Gross Weight	27,000 lbs (12247)		
2. Pilot Sphere		E. Operating Characteristics	
a. Inside Diameter	57'97" (147.24 cm)	1. Depth	0 - 3000 FSW (914 m.)
b. Hull Thickness	5" (12.7 cm)	2. Speed	.75 knots, 2 thrusters
c. Hatch Opening	18.5" diameter (46.9 cm)	a. Cruise	1 knot, 4 thrusters
3. Diver/Observer Compartment			
a. Length	97.50" (2.47 m)	F. Life Support	
b. Internal Diameter	43.90" (1.11 m)	1. Endurance	480 man hours (20 man days)
c. Hull Thickness		2. Carbon Dioxide Scrubbers	
(1) Side Wall	3.30" (8.38 cm)	3. Emergency Breathing Apparatus	
(2) Heads	2.281" (5.79 cm)	a. Pilot Sphere	2
d. Side Ports	8.10" dia. x 3.85" thick (20.57 cm x 8.77 cm)	b. Diver Compartment	2
e. Manway Opening	20" diameter (50.8 cm)	4. Oxygen Analyzer	
		a. Pilot Sphere	
C. Power		(1) One Beckman Minos AOM Meter Model 6602	
1. Battery		(2) One Remote Meter AOM 6602 from Diver Compartment	
a. Fourteen 2 VDC EXIDE E 110W-33 lead-acid Batteries		5. Carbon Dioxide Monitors	
b. 1760 Ampere-hours (49.2 kwh)		a. Pilot Sphere	
c. Oil compensated (25 gal. mineral oil)		(1) Beckman Minos Atmospheric Carbon Dioxide Monitor	
2. Reserve Battery		(2) Bendix Gastec Analyzer - hand pump type	
a. Fourteen 2 UDC exide 75L-5 lead acid batteries		b. Diver Compartment	
b. 150 Ampere-hours (4.2 kwh)		(1) Beckman Minos ACDM	
c. Oil compensated (3 gal. mineral oil)		(2) Bendix Gastec Analyzer - hand pump type	
3. Static Inverter			
a. Input			

APPENDIX 2

Collection Site Summary

(Numbers 1-23 refer to collection sites in Figure 1)

COLLECTION SITE SUMMARY
BAHAMAS EXPEDITION
JANUARY 25 - 31, 1997
HARBOR BRANCH OCEANOGRAPHIC INSTITUTION
DIVISION OF BIOMEDICAL MARINE RESEARCH

SITE NUMBER (DATE + SITE #)	LATITUDE	LONGITUDE	METHOD	DEPTH RANGE		NUMBER SAMPLES	
				(Feet)	Meters		
1	26-X-96-1	26 35.036'N	77 54.947'W	JSL II-2789	600	504	2
2	26-X-96-2	26 34.807'N	77 54.719'W	JSL II-2790	506	225	5
3	26-X-96-3	26 37.25'N	77 54.30'W	SNORKEL	4	1	0
4	26-X-96-4	26 35.372'N	77 53.892'W	SNORKEL	8	8	0
5	26-X-96-5	26 31.815'N	77 52.543'W	JSL II- 2791	554	500	5
6	26-X-96-6	26 31.905'N	77 52.424'W	SCUBA	92	60	3
7	27-X-96-1	25 00.628'N	77 33.818'W	JSL II-2792	650	60	7
8	27-X-96-2	25 00.606'N	77 33.716'W	SCUBA	108	60	5
9	27-X-96-3	25 00.668'N	77 33.871'W	JSL II-2793	545	267	12
10	27-X-96-4	25 00.827'N	77 34.215'W	SCUBA	90	60	0
11	28-X-96-1	25 27.970'N	76 54.209'W	JSL II-2794	826	540	9
12	28-X-96-2	25 31.054'N	76 54.338'W	JSL II-2795	631	510	6
13	28-X-96-3	25 31.063'N	76 54.117'W	SCUBA	80	55	1
14	28-X-96-4	25 31.00'N	76 49.80'W	SNORKEL	2	0	0
15	29-X-96-1	26 31.932'N	77 52.672'W	JSL II-2796	530	513	5
16	29-X-96-2	26 32.055'N	77 52.471'W	SCUBA	78	68	0
17	29-X-96-3	26 37.269'N	77 54.377'W	SNORKEL	2	0	0
18	29-X-96-4	26 31.779'N	77 52.543'W	JSL II-2797	538	526	12
19	29-X-96-5	26 34.274'N	77 53.779'W	SCUBA	80	60	0
20	30-X-96-1	26 30.927'N	78 48.550'W	JSL II-2798	889	501	10
21	30-X-96-2	26 37.454'N	78 58.814'W	JSL II-2799	1414	1313	11
22	30-X-96-3	26 38.2'N	78 57.7'W	SCUBA	80	60	1
23	31-X-96-1	27 32.540'N	79 58.656'W	JSL II-2800	265	220	0

APPENDIX 3
Collection Site Descriptions

SITE DESCRIPTIONS
BAHAMAS EXPEDITION
JANUARY 15 - 31, 1997
HARBOR BRANCH OCEANOGRAPHIC INSTITUTION
DIVISION OF BIOMEDICAL MARINE RESEARCH

SITE NUMBER	HABITAT	TEMP. (C)	VISI- BILTY (Ft.)	CURRENT		SEAS (Ft., From)	DEPTH RANGE (Ft.)
				SALINITY (ppt)	(KNTS, From)		
LOCATION				WEATHER	(MPH, From)		
26-X-96-1	10 DG. SAND/MUD SLOPE; 30 DG. AT 450'	21.62C	50'	.1-2	180	1 SE	600 504
		36.84		CLOUDY	14 130		-
26-X-96-2	30 DG. ROCK SLOPE; TOP OF SLOPE 200'	22.4C	80	.1	S	1 SE	506 225
		36.84		CLOUDY, RAIN	14 110		-
26-X-96-3	RED MANGROVE CHANNEL	76F	30	.1-1.0	FLOOD	1 SE	4 1
				CLOUDY	14 130		-
26-X-96-4	PATCH REEF	79F	50	0		1-2 SE	8 8
	BAHAMAS; SE. GRAND BAHAMA ISLAND, ~1NM SW. OF SWEETINGS CAY			CLOUDY	14 130		-
26-X-96-5	45 DG. ROCK, RUBBLE SLOPE	21.5C	80	.2	S	1 SE	554 500
		36.85		SUNNY	18 90		-
26-X-96-6	30 DG. FORE REEF SLOPE; TOP 60', BASE 90', 6' RELIEF LEDGES	78F	80	<.1		1 SE	92 60
				SUNNY	15 90		-
27-X-96-1	80 DG. ROCK AND RUBBLE SLOPE	21.45C	40	.1	300	1 SE	650 60
	BAHAMAS; SW. NEW PROVIDENCE ISLAND, S. OF GOULDING CAY			36.94	SUNNY	15 70	

SITE NUMBER	HABITAT	TEMP. (C)	VISI- BILITY (Ft.)	CURRENT (KNTS, From)		SEAS (Ft., From)		DEPTH RANGE (Ft.)	
				SALINITY (ppt)	WEATHER	(MPH, From)			
LOCATION									
27-X-96-2 BAHAMAS; SW. NEW PROVIDENCE ISLAND, SE. OF GOULDING CAY	WALL, FORE REEF SLOPE	78F	100	<.1	NW	2	E	108	60
				SUNNY	15	70			
27-X-96-3 BAHAMAS; SW. NEW PROVIDENCE ISLAND, S. OF GOULDING CAY	80 DG. ROCK WALL, SAND CHUTES, RUBBLE SLIDES	21.20C	90	0		2-3	NE	545	267
				SUNNY	20	70			
27-X-96-4 BAHAMAS; SW. NEW PROVIDENCE ISLAND, SE. OF GOULDING CAY	90 DG. FORE REEF SLOPE AND WALL	78F	100	<.1	N	1-2	NE	-90	60
				SUNNY	20	70			
28-X-96-1 BAHAMAS; SW. OF EGG ISLAND	50 DG. SAND AND ROCK SLOPE	19.17C	60	0		1-2	NE	826	540
		36.60		SUNNY	18	50			
28-X-96-2 BAHAMAS, NW. OF EGG ISLAND	50-80 DG. ROCK WITH SILT VENEER	20.62C	60	.1	120	1-2	NE	631	510
				SUNNY	16	55			
28-X-96-3 BAHAMAS; NW. OF EGG ISLAND	30 DG. FORE REEF SLOPE; HARD GROUND W/ GORGONACEA, RUBBLE CHUTE	78F	80	.1	NE	1-2	NE	80	55
				SUNNY	16	55			
28-X-96-4 BAHAMAS; ROYAL ISLAND, COVE ON SOUTH SIDE, E. END, N. EDGE	RED MANGROVE CHANNEL	76F	30	0		CALM		2	
				P.CLOUDY	18	50			
29-X-96-1 BAHAMAS, SE. GRAND BAHAMA ISLAND, SW. OF SWEETINGS CAY	45 DG. ROCK RUBBLE SLOPE	22.4C	60	.6	S	CALM, SE		530	513
		36.84		SUNNY	10	60			

SITE NUMBER	HABITAT	TEMP. (C)	VISI- BILTY (Ft.)	CURRENT (KNTS, From)		SEAS (Ft., From)	DEPTH RANGE (Ft.)	
				SALINITY (ppt)	WEATHER (MPH, From)			
29-X-96-2	10 DG. FORE REEF SLOPE, 3-8' SPUR GROOVE AND HUMMOCKS	76F	60	0		CALM, E	78	68
BAHAMAS; SE. GRAND BAHAMA ISLAND, OFF LAST ISLAND S. OF SWEETINGS CAY				SUNNY	10 60			
29-X-96-3	RED MANGROVE CHANNEL	80F	20	I	FLOOD	CALM, E	2	0
BAHAMAS; SE. GRAND BAHAMA ISLAND, CHANNEL BETWEEN LITTLE HARBOR AND SWEETING CAY				SUNNY	10 60			
29-X-96-4	50 DG. SAND RUBBLE SLOPE, SOME BOULDERS	21.70C	50	.1	S	CALM, SE	538	526
BAHAMAS; SE. GRAND BAHAMA ISLAND, S. OF SWEETINGS CAY				SUNNY	10 160			
29-X-96-5	20 DG. FORE REEF SLOPE, SAND AT 80', 6-8' SPUR GROOVE	78F	60	<.1	E	CALM	80	60
BAHAMAS; SE. GRAND BAHAMA ISLAND, SW. OF SWEETINGS CAY				SUNNY	10 160			
30-X-96-1	15 DG. FINE WHITE SAND SLOPE; 30 DG. PAVEMENT; 50' ROCK OUTCROP	18.72C	80	0		CALM, SE	889	501
BAHAMAS; S. GRAND BAHAMA ISLAND, SW. OF FREEPORT HARBOR		36.61		SUNNY	8 75			
30-X-96-2	RO PAVEMENT, WHITE SEDIMENT LAYER; 40' FLAT TOP PINNACLES	16.32C	60	.1	60	CALM	1414	1313
BAHAMAS; SW. GRAND BAHAMA ISLAND, LITHOHERM		36.23		SUNNY	5 120			
30-X-96-3	SPARSE PATCH REEF, 4' RELIEF	80F	100	<.1	NW	CALM	80	60
BAHAMAS; SW. GRAND BAHAMA ISLAND				SUNNY	5 120			
31-X-96-1	Oculina Coral Bank	23.26C	10-30	.2	150	1-2 SW	265	220
FLORIDA; FT. PIERCE, JEFF'S REEF (OCULINA CORAL BANK)		36.28		SUNNY				

APPENDIX 4

Fathometer Profiles of Shelf-Edge
Topography from R/V EDWIN LINK
at Submersible Dive Sites

JSL II-2789

red flint

sandstone

10/26/95

Hd-6

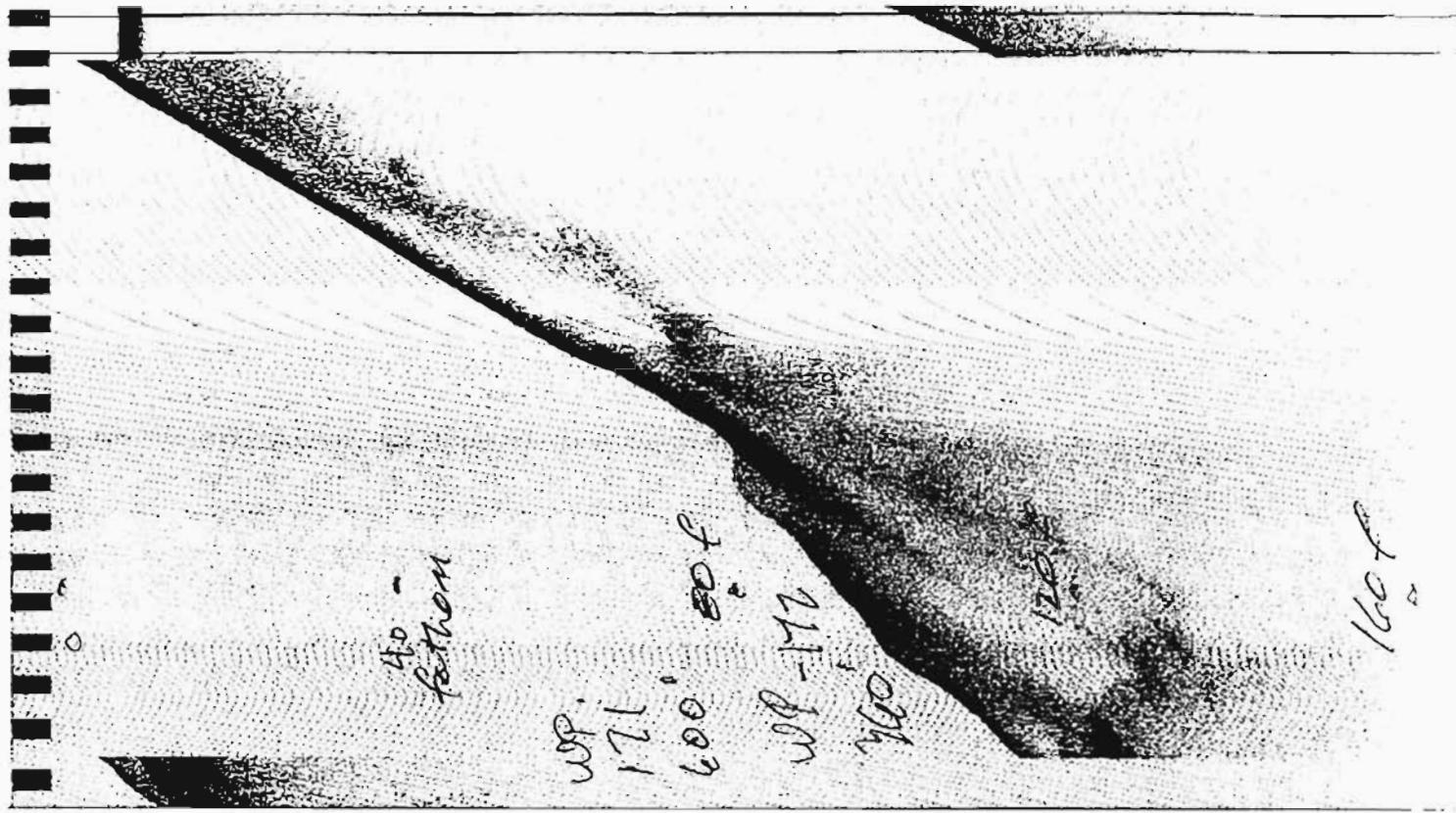
3 km

total

200 ft

200 ft

200 ft



O -

27-X-94-1
Goulding
NAT

40+
16°
20°
3 1/2 kD



SD -

120 -
4

1
27-II-2192

→ 5
Jeff &
Reef
12' ←
fathoms

Oct. 31, 1996

JSL II - 2800
J. Reef

32' ←

JSL II - 2800

etc.

↑

32' ←

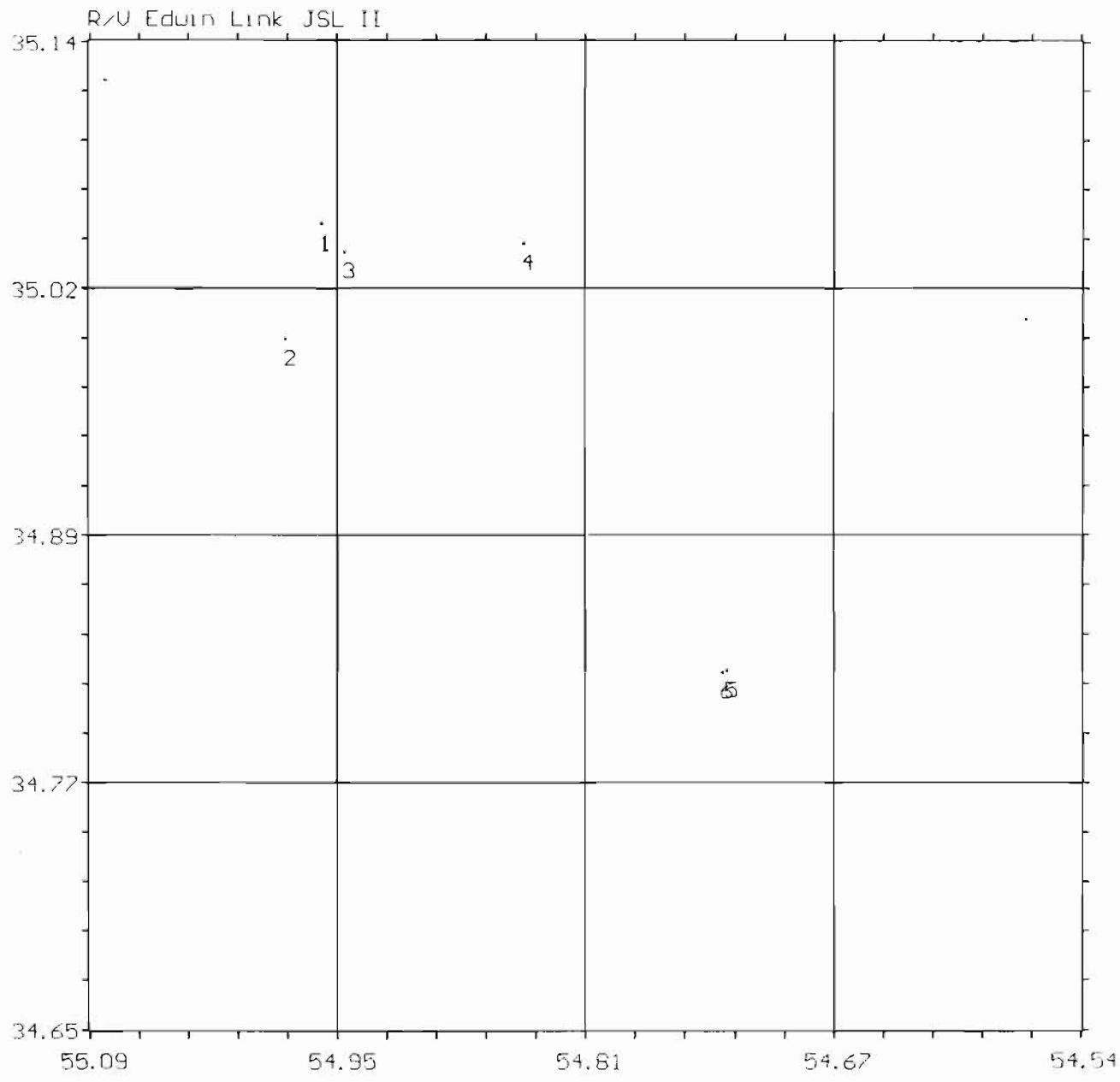
Ovalina
Bank -
Top 210'
Base on 270'

APPENDIX 5

JOHNSON-SEA-LINK II Submersible Tracks and Sample Positions

2789

Integrated Positioning System (CUTD/26/96 8:07:26

26°N
72°W

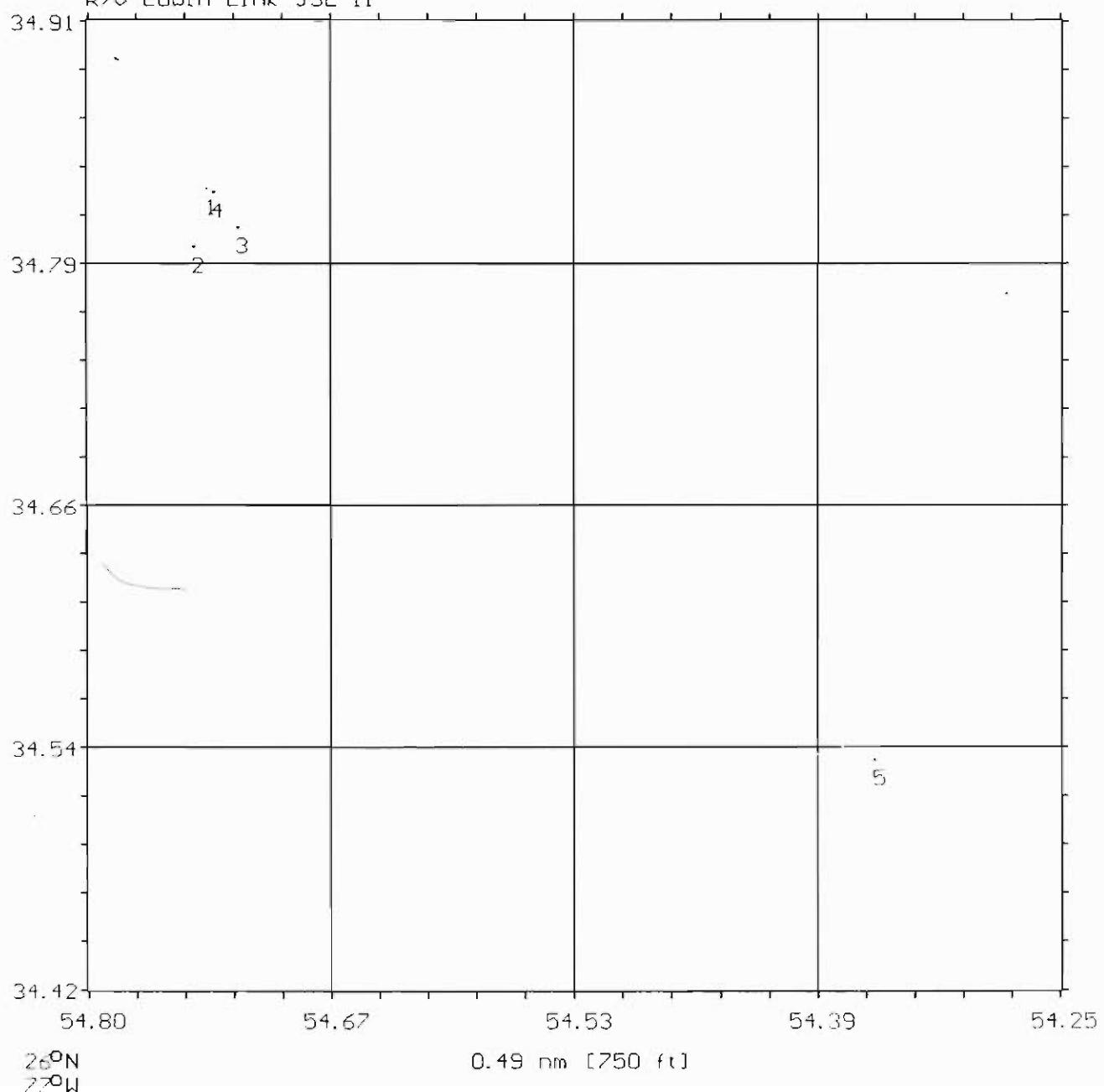
0.49 nm [750 ft]

07:

- | | | | | |
|----|----------|-------------|-------------|-------------------------|
| 1. | 10:57:16 | N 26:35.050 | W 72:54.960 | 0 ft Target Site |
| 2. | 10:59:32 | N 26:34.993 | W 72:54.980 | 0 ft Launch 2789 |
| 3. | 8:13:10 | N 26:35.036 | W 72:54.947 | 1186 ft On Bottom |
| 4. | 8:23:50 | N 26:35.040 | W 72:54.848 | 417 ft #1 |
| 5. | 8:39:28 | N 26:34.822 | W 72:54.735 | 471 ft Sample #1 - 504' |
| 6. | 8:53:36 | N 26:34.822 | W 72:54.737 | 476 ft Leaving Bottom |

2790 Integrated Positioning System (CUTD/26/96 10:42:38

R/V Edwin Link JSL II



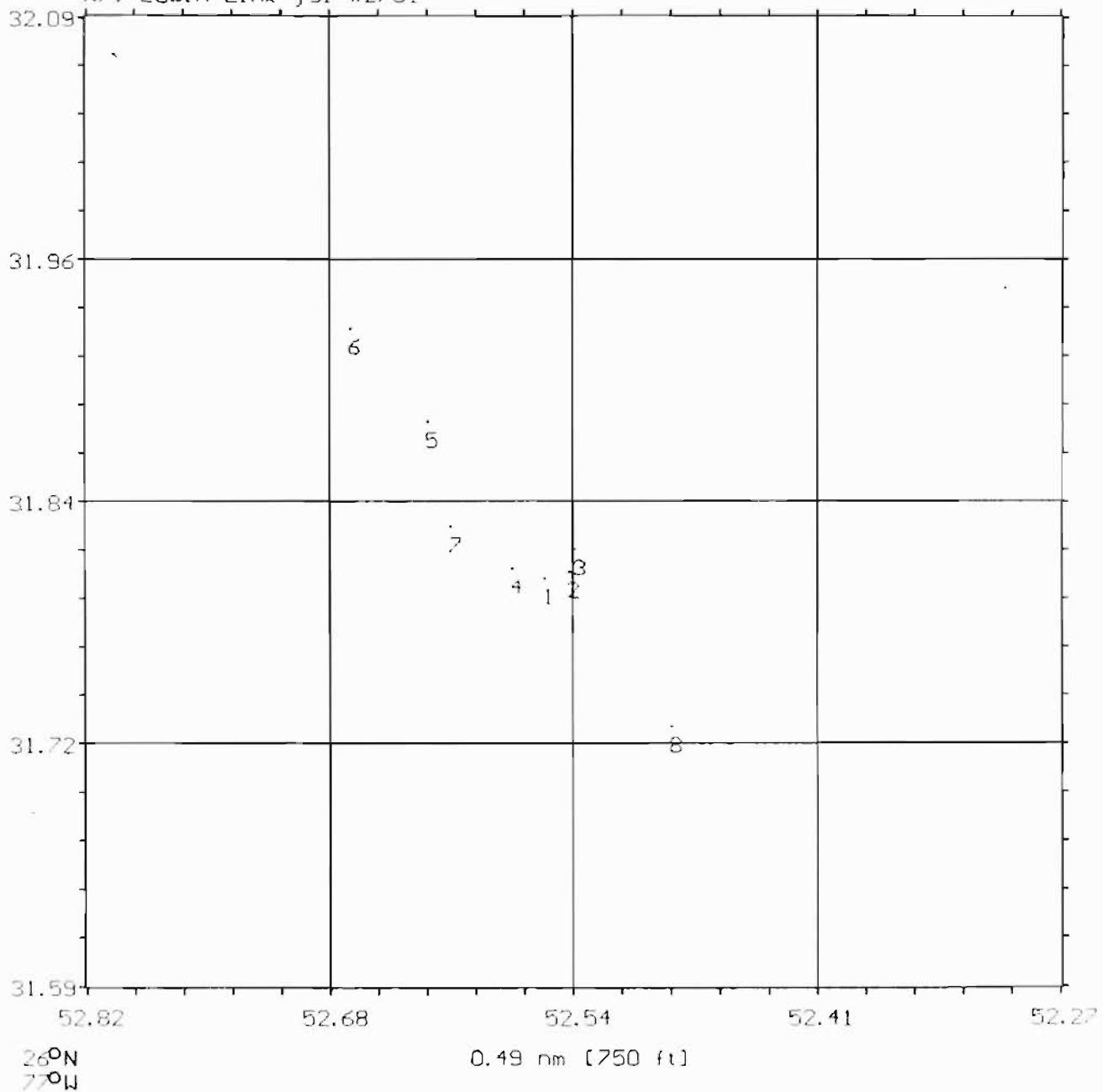
26°N
77°W

0.49 nm [750 ft]

- | | | | |
|-------------|-------------|-------------|------------------------------|
| 1. 10:36:24 | N 26:34.822 | W 77:54.736 | 0 ft End of Last Dive |
| 2. 10:43:48 | N 26:34.797 | W 77:54.743 | 0 ft Launch 2990 |
| 3. 10:55:04 | N 26:34.807 | W 77:54.719 | 499 ft On Bottom - 506' |
| 4. 11:00:28 | N 26:34.825 | W 77:54.732 | 498 ft Sample 1 - 502' |
| 5. 12:37:38 | N 26:34.536 | W 77:54.358 | 210 ft leaving bottom 225 ft |

Integrated Positioning System (CUTO/26/96 15:48:30

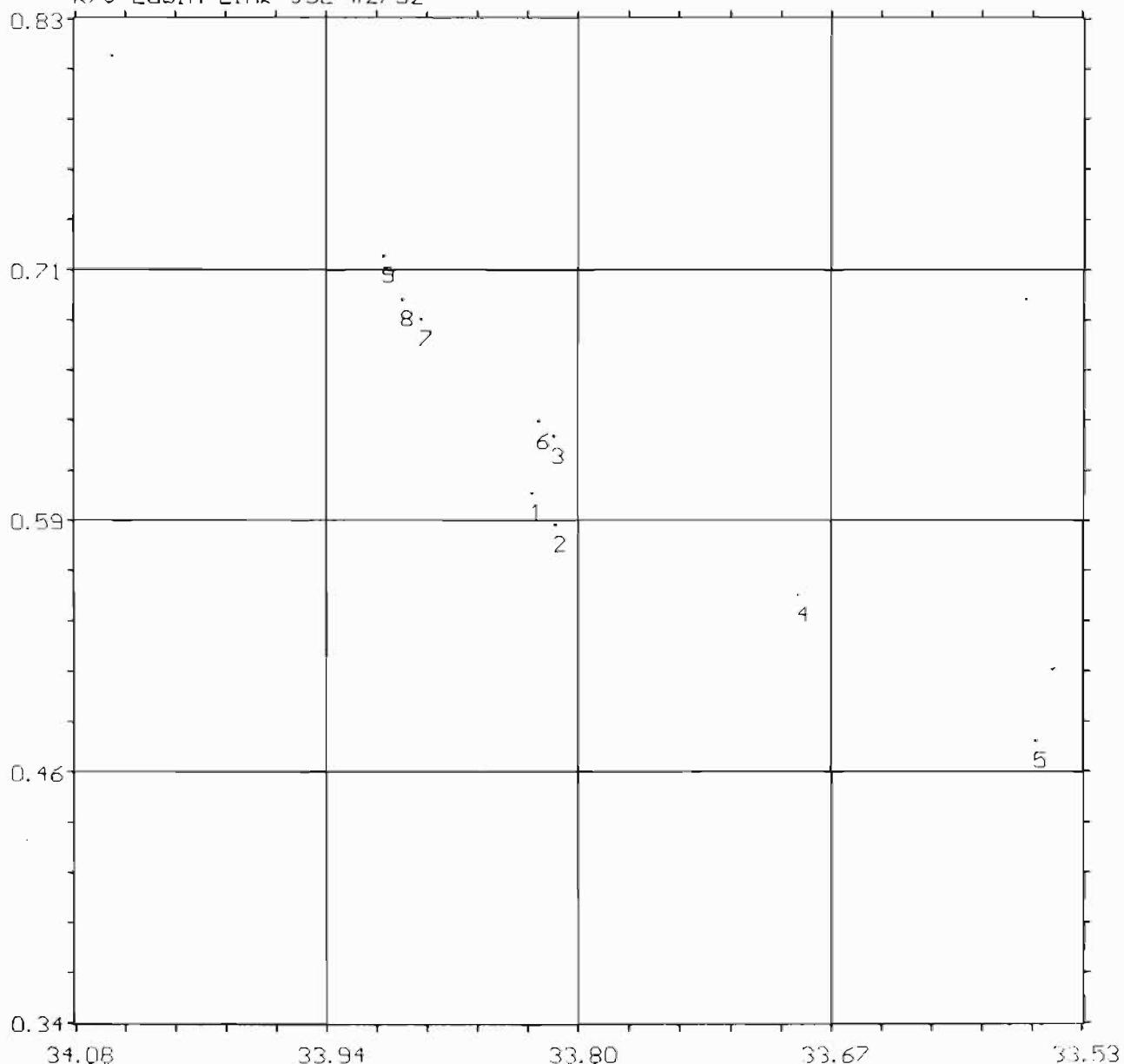
R/V Edwin Link jsl #2791



1. 15:48:58 N 26:31.800 W 77:52.560 0 ft Dive Site
2. 15:54:06 N 26:31.803 W 77:52.547 0 ft Launch 2791
3. 16:06:26 N 26:31.815 W 77:52.543 0 ft On Bottom - 549'
4. 16:32:28 N 26:31.805 W 77:52.579 544 ft Sample 2 - 527' - *Sponges scattered*
5. 16:53:44 N 26:31.880 W 77:52.626 525 ft Sample 3 - 523' - *Discoderus*
6. 17:01:36 N 26:31.927 W 77:52.671 553 ft Turnaround Point - 545'
7. 17:25:00 N 26:31.826 W 77:52.614 532 ft Sample 4 - 544' - *Discoderus*
8. 18:04:40 N 26:31.724 W 77:52.488 495 ft leaving bottom 520 ft.

Integrated Positioning System (CUT) 27/96 7:17:18

R/U Edwin Link JSI #2792

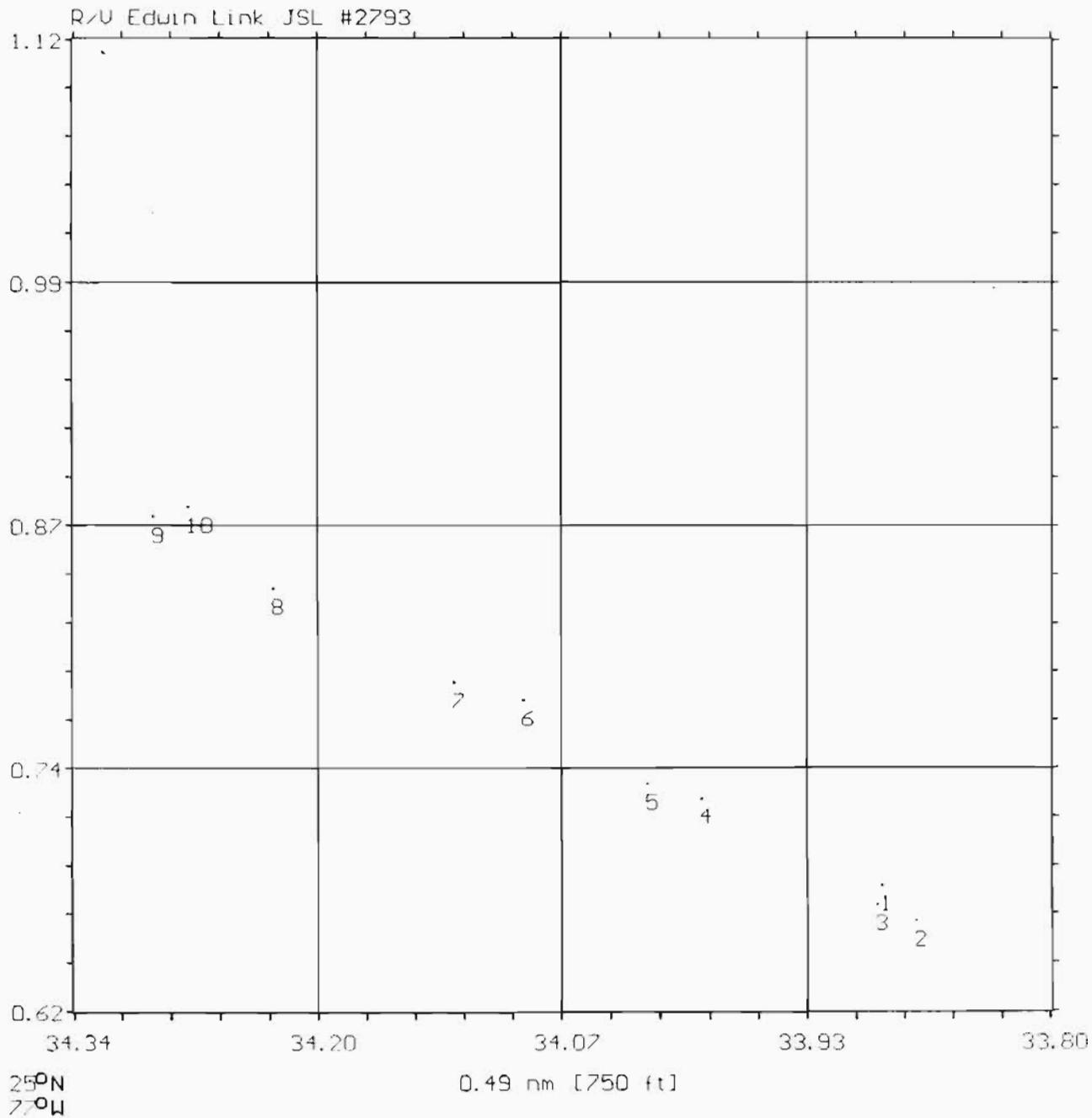


25°N
77°W

0.49 nm [750 ft]

8:10:52	N 25:00.600	W 77:33.830	0 ft target site wpt 174
8:14:54	N 25:00.584	W 77:33.817	0 ft launch
8:31:30	N 25:00.628	W 77:33.818	590 ft on bottom 602 ft
8:49:30	N 25:00.550	W 77:33.686	600 ft sample #1 550 ft.3
9:20:38	N 25:00.478	W 77:33.558	542 ft sample #3 543ft
10:03:10	N 25:00.636	W 77:33.826	574 ft sample #4 535 ft.
10:16:42	N 25:00.686	W 77:33.890	550 ft sample #5 535ft
10:39:40	N 25:00.695	W 77:33.899	382 ft sample #301 398ft
11:05:44	N 25:00.717	W 77:33.910	46 ft on top of wall 55 ft.

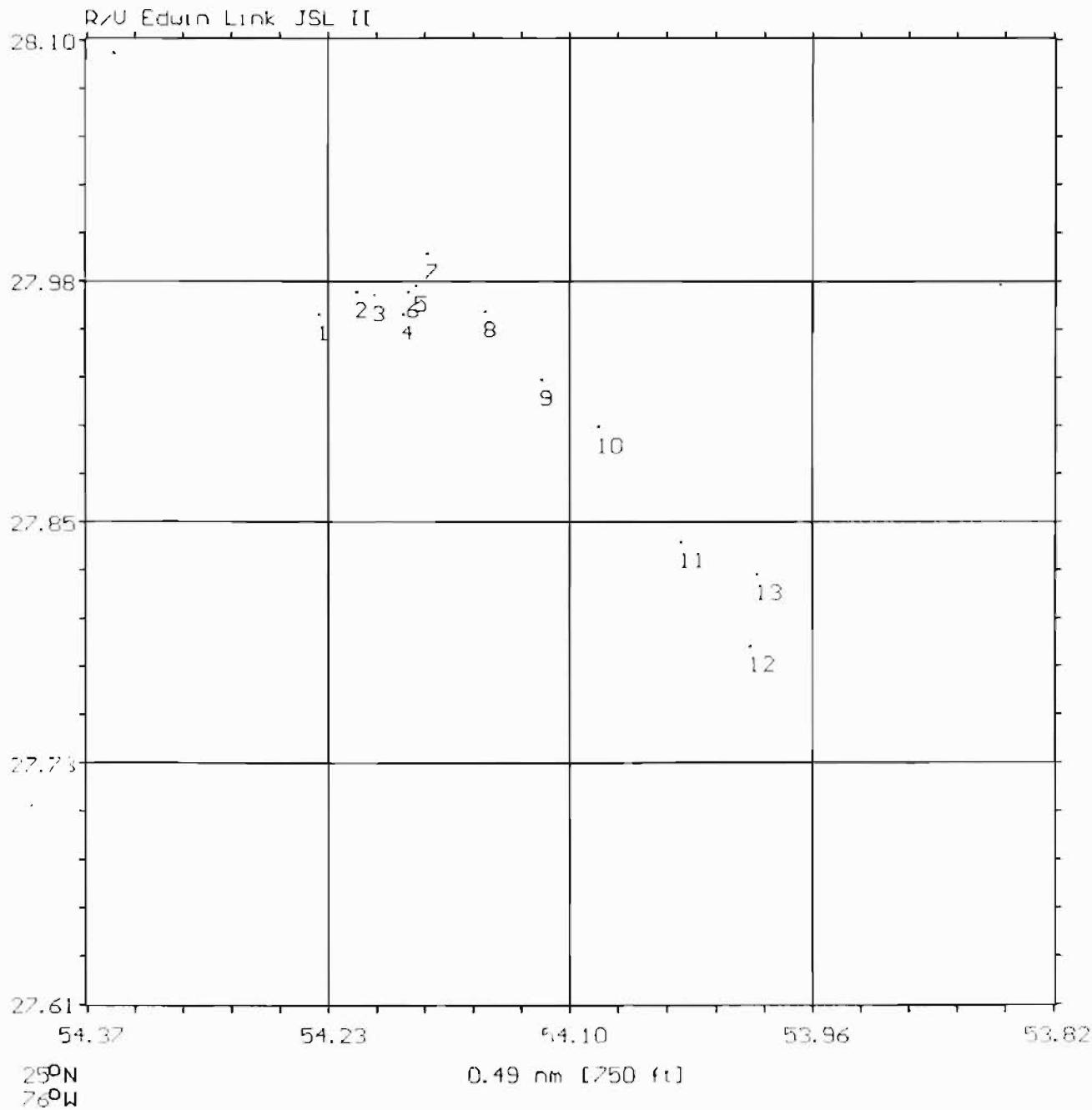
Integrated Positioning System (CUTD/27/96 14:32:36



1. 14:33:58 N 25:00.686 W 77:33.890 0 ft target site
2. 15:31:42 N 25:00.668 W 77:33.871 517 ft on bottom 535 ft.
3. 15:33:34 N 25:00.676 W 77:33.893 553 ft sample #1, 537 feet
4. 15:56:46 N 25:00.730 W 77:33.991 525 ft sample #2, 534 feet
5. 16:12:22 N 25:00.738 W 77:34.021 540 ft sample #3B, 536 feet
6. 16:41:30 N 25:00.780 W 77:34.090 517 ft sample #4 533 ft.
7. 16:56:02 N 25:00.789 W 77:34.129 512 ft sample #5, 520ft.
8. 17:15:14 N 25:00.832 W 77:34.229 505 ft sample #9 528 ft.
9. 17:31:34 N 25:00.873 W 77:34.295 512 ft sample #11 525 ft. end of transect
10. 18:04:30 N 25:00.878 W 77:34.276 373 ft sample 12 257 ft.

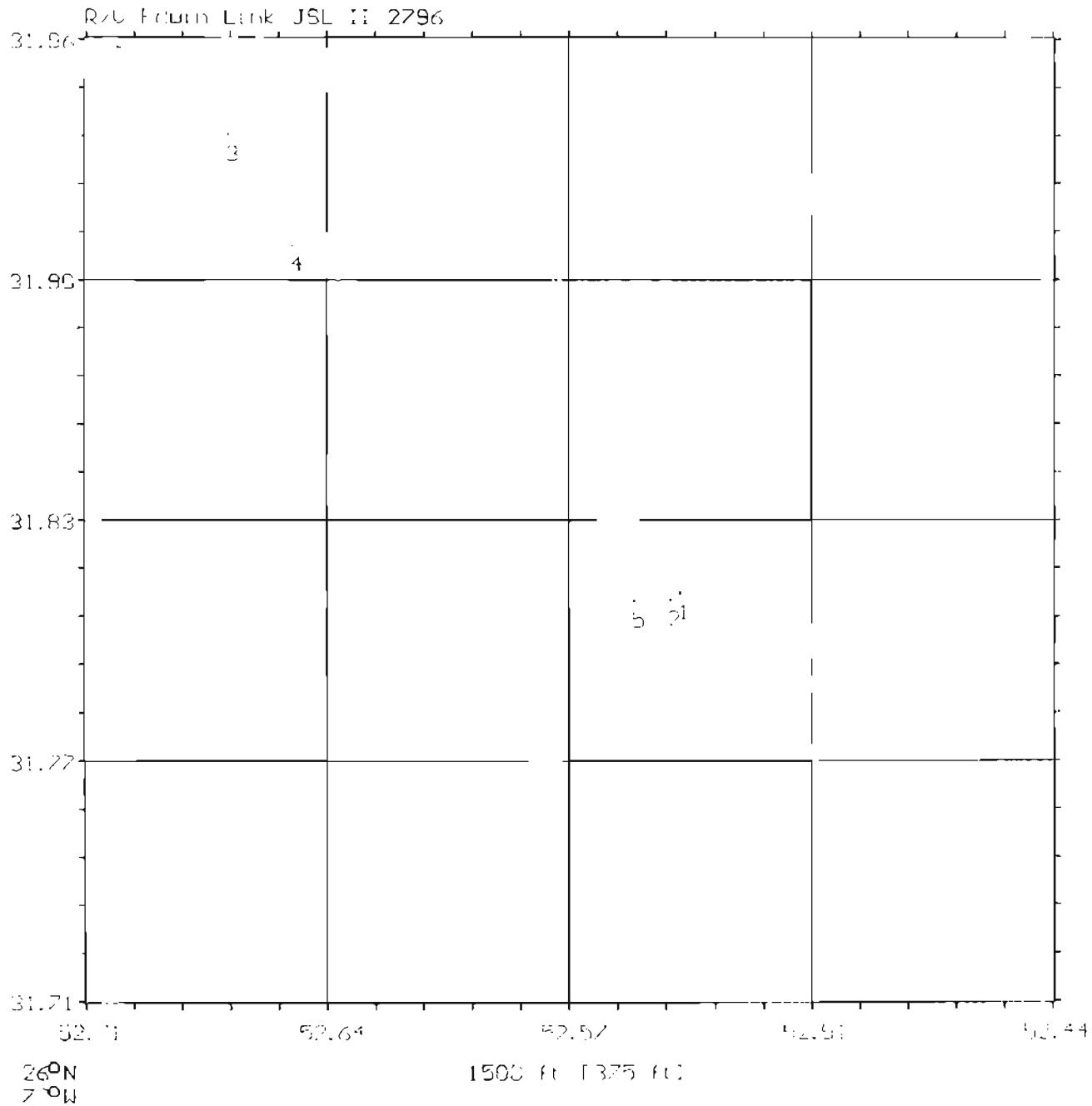
DIVE 2794

Integrated Positioning System (CUTD) 28/96 7:56:30



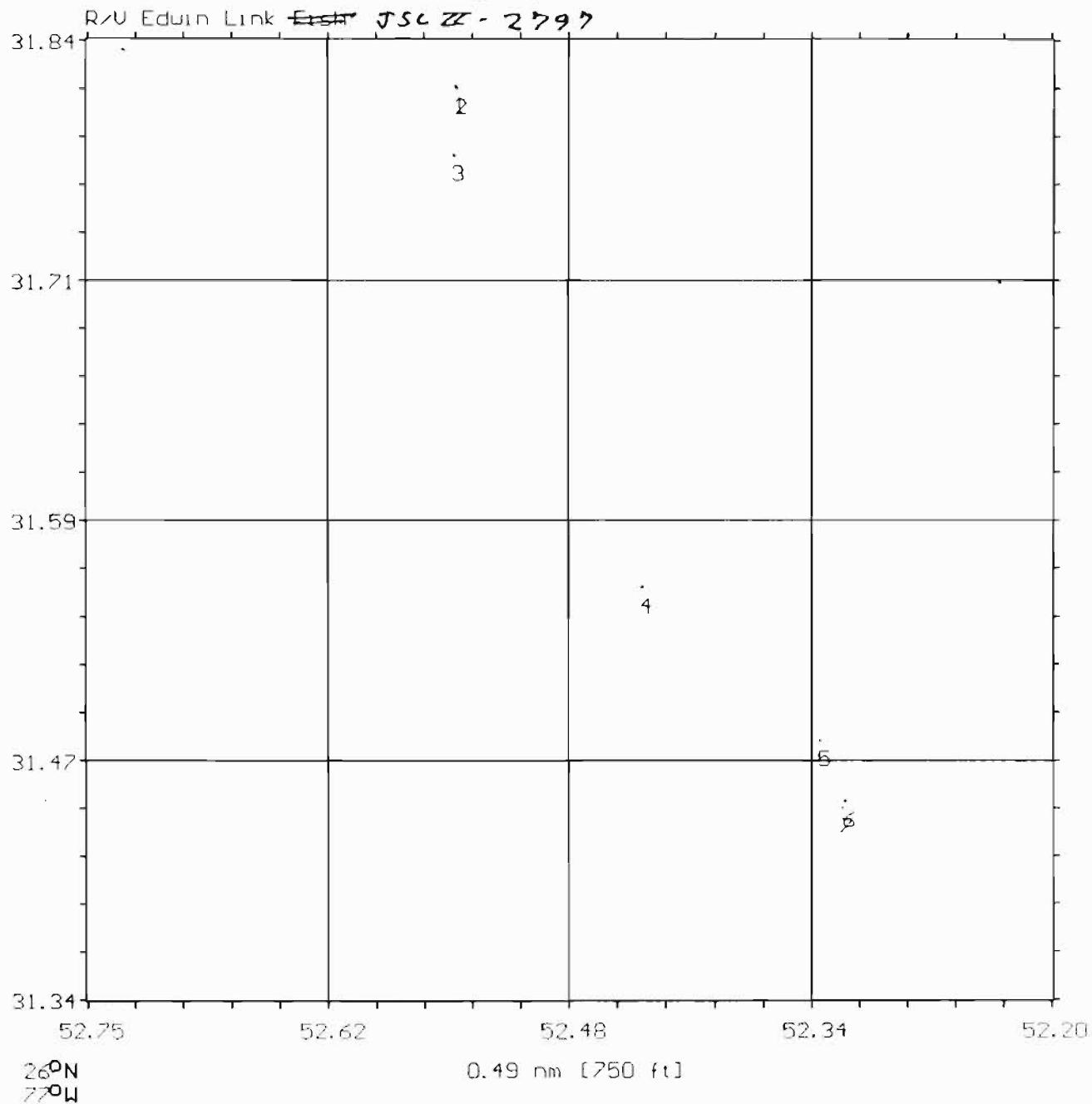
1. 7:57:20 N 25:27.960 W 76:54.240 0 ft target site
2. 8:04:48 N 25:27.972 W 76:54.219 0 ft launch
3. 8:21:18 N 25:27.970 W 76:54.209 797 ft bottom report
4. 8:24:38 N 25:27.960 W 76:54.193 762 ft sample 1 748'
5. 8:30:26 N 25:27.975 W 76:54.186 723 ft sample 2 730'
6. 8:36:50 N 25:27.972 W 76:54.190 707 ft sample 3 722'
7. 8:44:50 N 25:27.991 W 76:54.180 573 ft 600' following wall
8. 8:51:14 N 25:27.962 W 76:54.147 575 ft sample 590'
9. 9:17:18 N 25:27.927 W 76:54.114 552 ft sample 5 520'
10. 9:27:50 N 25:27.903 W 76:54.082 531 ft sample 6 560'
11. 9:50:30 N 25:27.843 W 76:54.037 598 ft sample #8 595 ft
12. 10:31:02 N 25:27.790 W 76:53.997 667 ft sample #9 604 ft
13. 11:01:50 N 25:27.827 W 76:53.993 557 ft leaving bottom

Integrated Positioning System (CUTD/29/96) 7:56:18



- | | | | |
|-------------|-------------|-------------|------------------------------|
| 1. 7:55:26 | N 26:31.815 | W 77:52.543 | 0 ft Dive Site From 2796 |
| 2. 7:56:58 | N 26:31.813 | W 77:52.546 | 0 ft Launch 2796 |
| 3. 8:14:18 | N 26:31.832 | W 77:52.672 | 523 ft On Bottom - 523' |
| 4. 8:36:10 | N 26:31.834 | W 77:52.653 | 524 ft Sampling - 524' |
| 5. 11:11:02 | N 26:31.813 | W 77:52.556 | 498 ft Leaving Bottom - 505' |

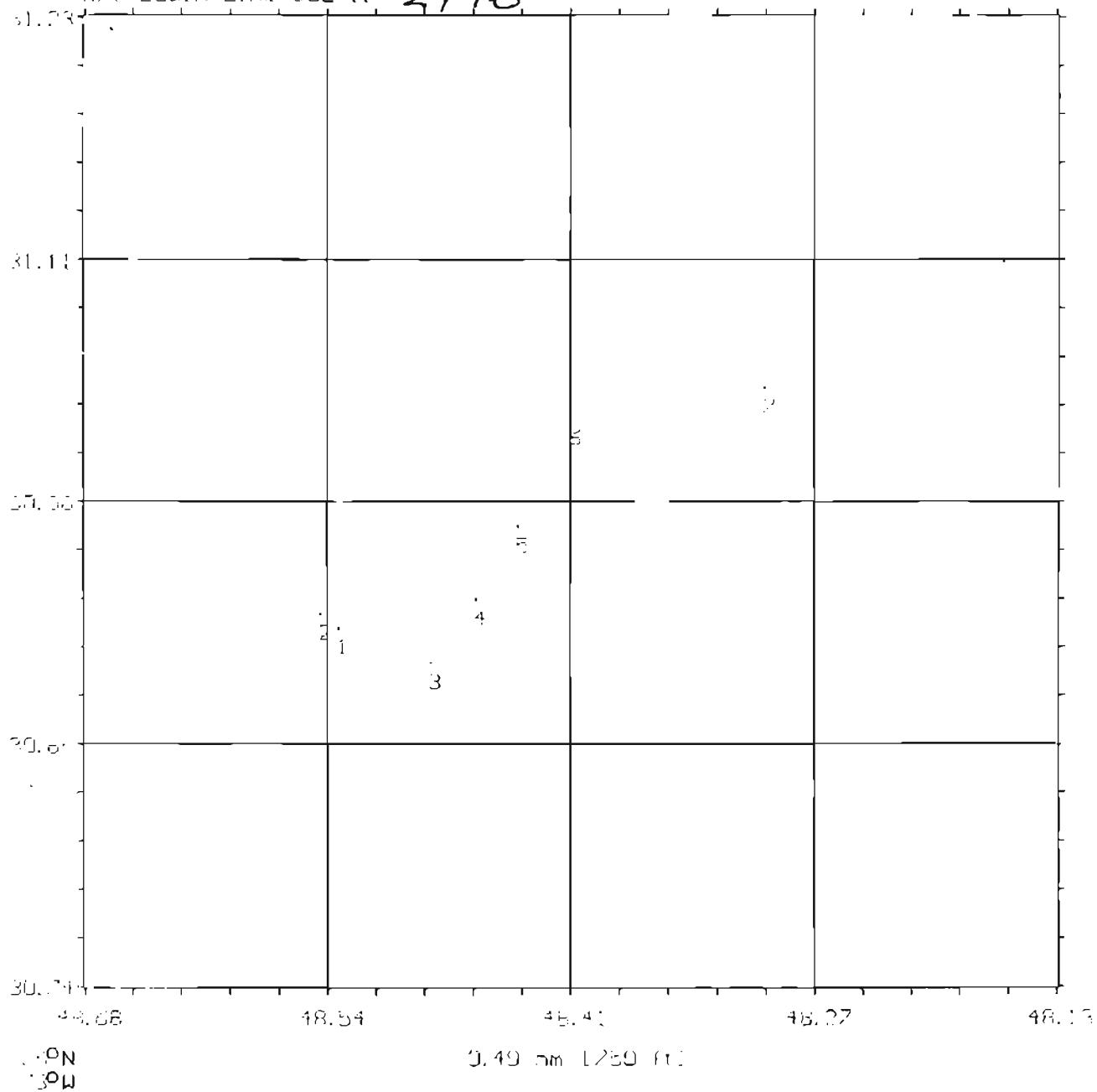
Integrated Positioning System (CUTD) 29/96 15:28:52



- | | | | |
|-------------|-------------|-------------|-----------------------------|
| 1. 15:29:34 | N 26:31.815 | W 77:52.543 | 0 ft Launch Site |
| 2. 15:35:54 | N 26:31.814 | W 77:52.542 | 0 ft Launch 2797 |
| 3. 15:52:22 | N 26:31.779 | W 77:52.543 | 513 ft On Bottom - 358' |
| 4. 17:18:46 | N 26:31.557 | W 77:52.436 | 520 ft Small Sponges - 525' |
| 5. 18:23:02 | N 26:31.478 | W 77:52.335 | 525 ft Disco City - 530' |
| 6. 18:37:30 | N 26:31.447 | W 77:52.320 | 513 ft End of Disco City |
| 7. 18:37:50 | N 26:31.443 | W 77:52.322 | 535 ft Leaving Bottom- 530' |

Integrated Positioning System (CUTD/30/96) 7:53:48

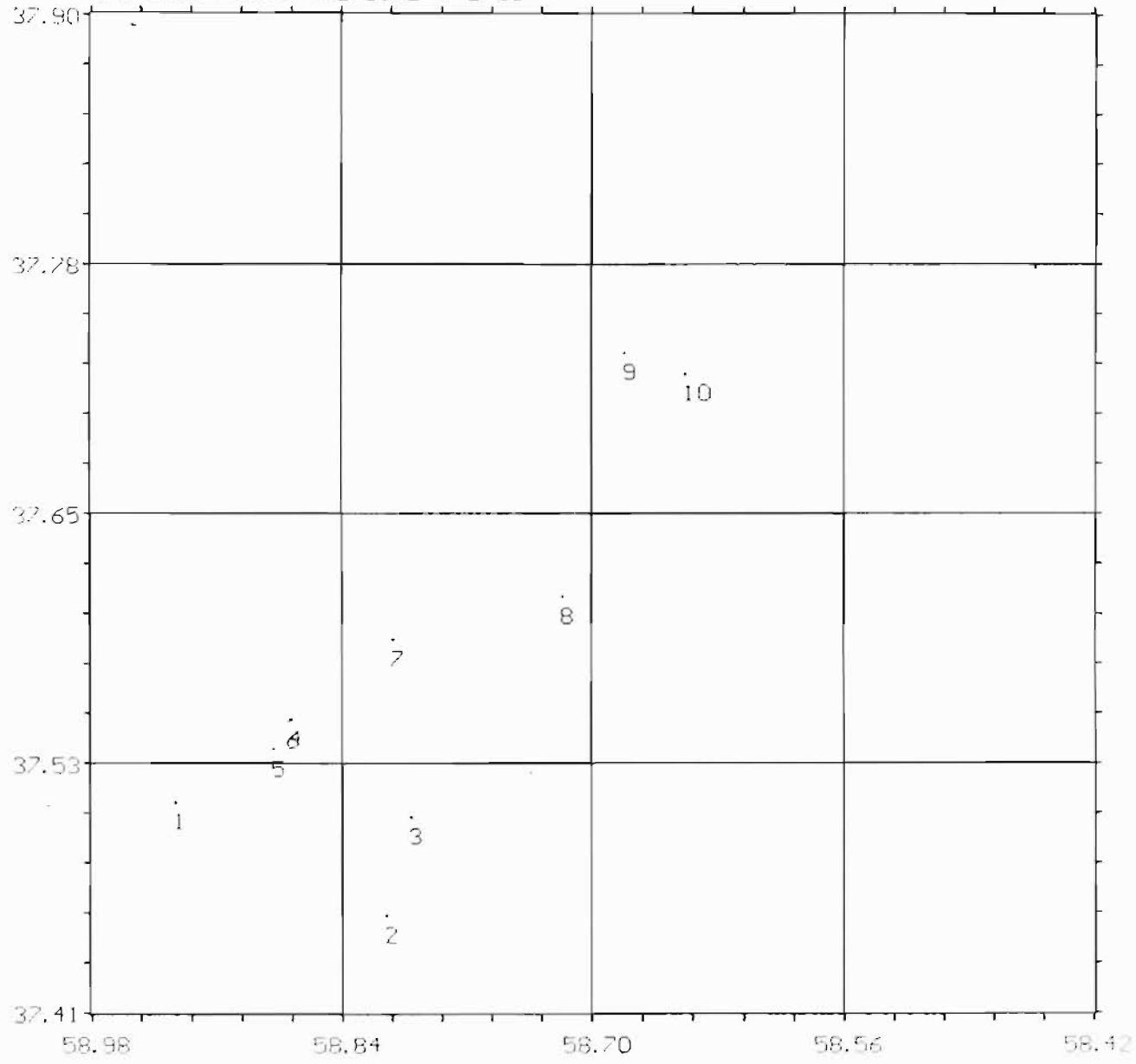
R/V Edwin Link JSL II - 2798



- | | | | |
|----------|-------------|-------------|--------------------------------|
| 7:54:14 | N 26:30.920 | W 78:48.540 | 0 ft target site |
| 7:59:58 | N 26:30.922 | W 78:48.550 | 0 ft launch |
| 8:31:24 | N 26:30.933 | W 78:48.482 | 866 ft sample #1 885 ft |
| 9:12:54 | N 26:30.935 | W 78:48.462 | 819 ft B13' sample |
| 9:25:42 | N 26:30.972 | W 78:48.437 | 746 ft 756' sample |
| 10:33:22 | N 26:31.027 | W 78:48.407 | 1005 ft sample 6 768' 20' wait |
| 11:01:46 | N 26:31.042 | W 78:48.297 | 484 ft last sample 500' |

Integrated Positioning System (CUTD/30/96 15:33:52

R/V Edwin Link JSL DIVE # 2799



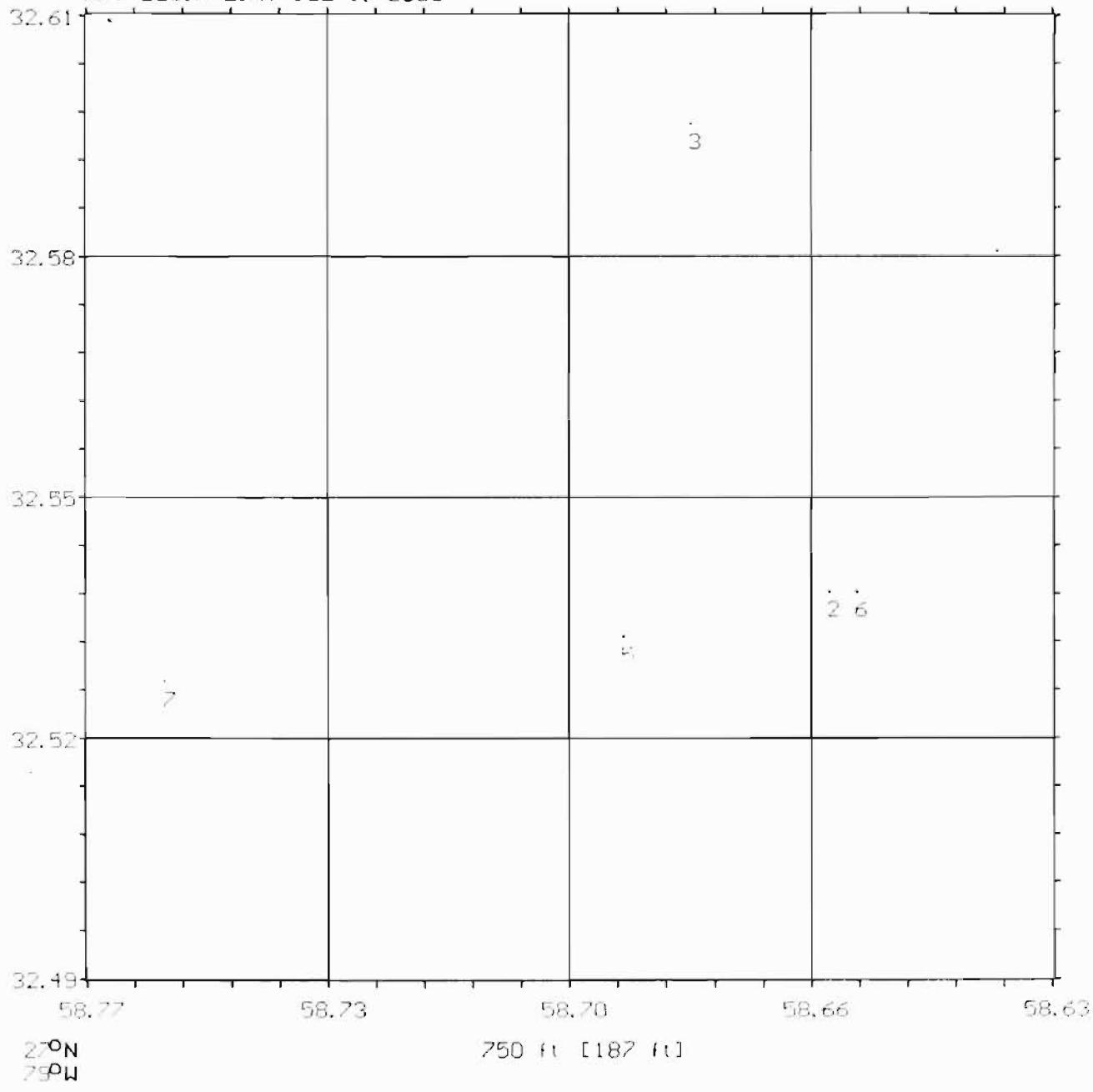
26°N
78°W

0.49 nm [750 ft]

15:17:16	N 26:37.510	W 78:58.930	0 ft target site 1410 ft.
15:57:24	N 26:37.454	W 78:58.814	1436 ft on bottom 1416 ft.
16:04:12	N 26:37.502	W 78:58.800	1421 ft sample 1 1404 ft.
16:37:26	N 26:37.551	W 78:58.866	1401 ft sample 2 1390 ft.
16:43:58	N 26:37.536	W 78:58.877	1384 ft sample 3
16:49:22	N 26:37.550	W 78:58.868	1383 ft sample 5 1392 ft.
16:59:08	N 26:37.590	W 78:58.810	0 ft waypoint 185
17:40:40	N 26:37.612	W 78:58.717	1316 ft Dome rock 1325 ft.
18:25:36	N 26:37.732	W 78:58.682	1271 ft another ridge
18:38:28	N 26:37.721	W 78:58.649	1331 ft last sample 1336 ft. leaving bottom

Integrated Positioning System (CUTD/31/96 6:42:50

R/V Edwin Link JSL II 2800

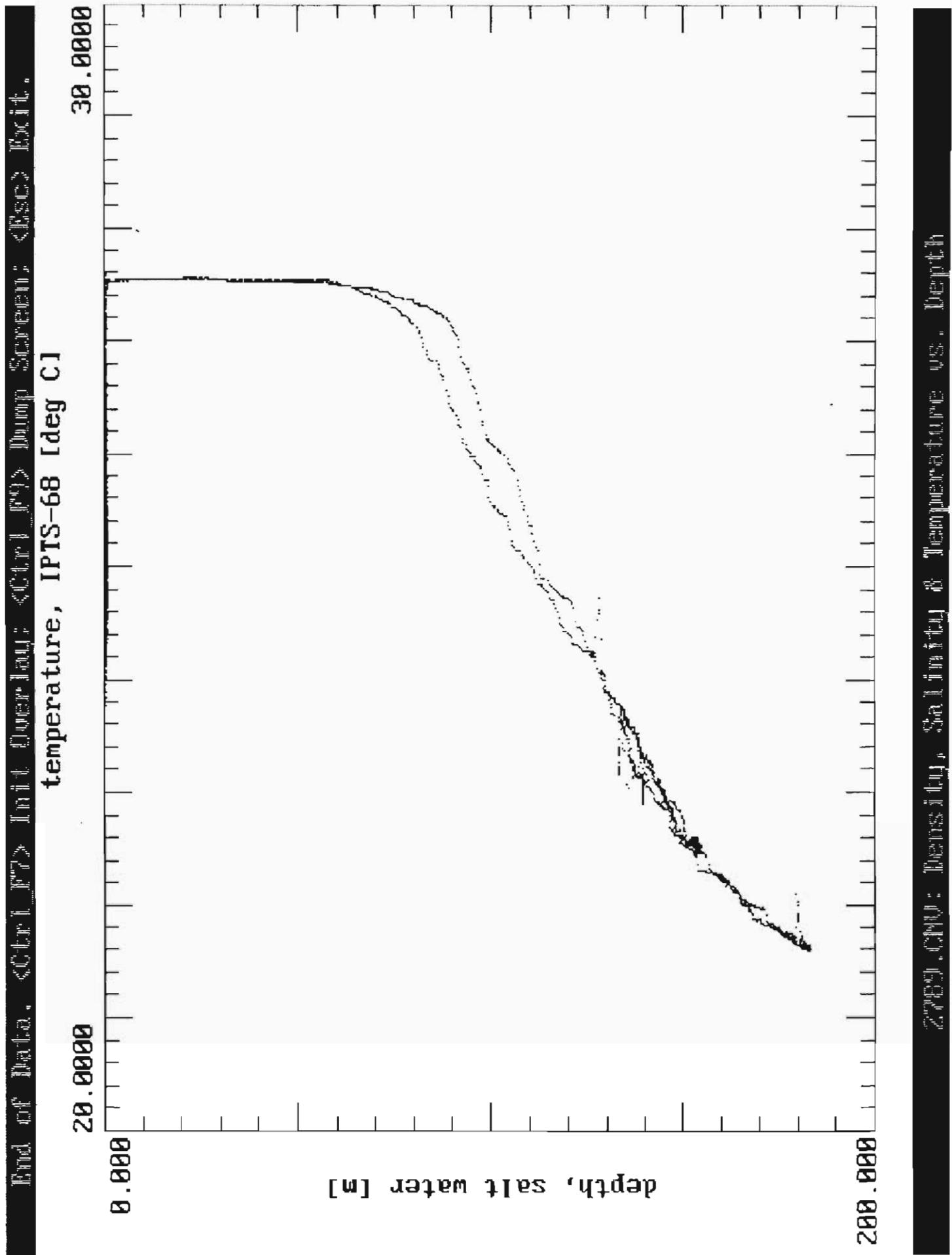


1. 7:03:36 N 27:32.270 W 79:58.660 0 ft target launch
2. 7:05:46 N 27:32.540 W 79:58.660 0 ft South Peak HPT 182
3. 7:08:28 N 27:32.600 W 79:58.680 0 ft base of north slope WPT 186
4. 7:19:30 N 27:32.269 W 79:58.631 0 ft launch
5. 7:47:58 N 27:32.534 W 79:58.690 212 ft on station
6. 9:01:38 N 27:32.540 W 79:58.656 250 ft at coral table 265 ft
7. 9:32:08 N 27:32.529 W 79:58.756 256 ft West side of reef

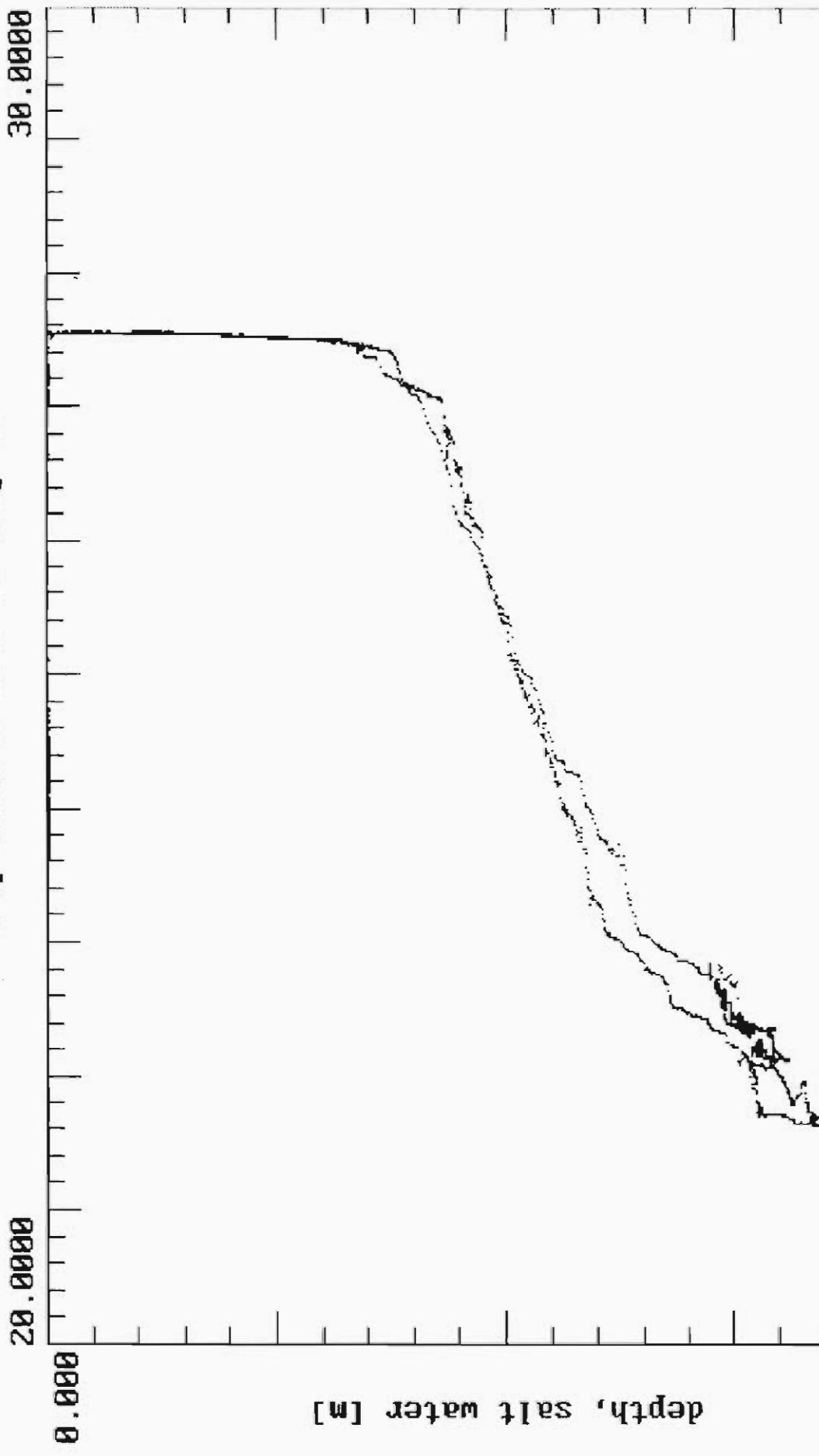
APPENDIX 6

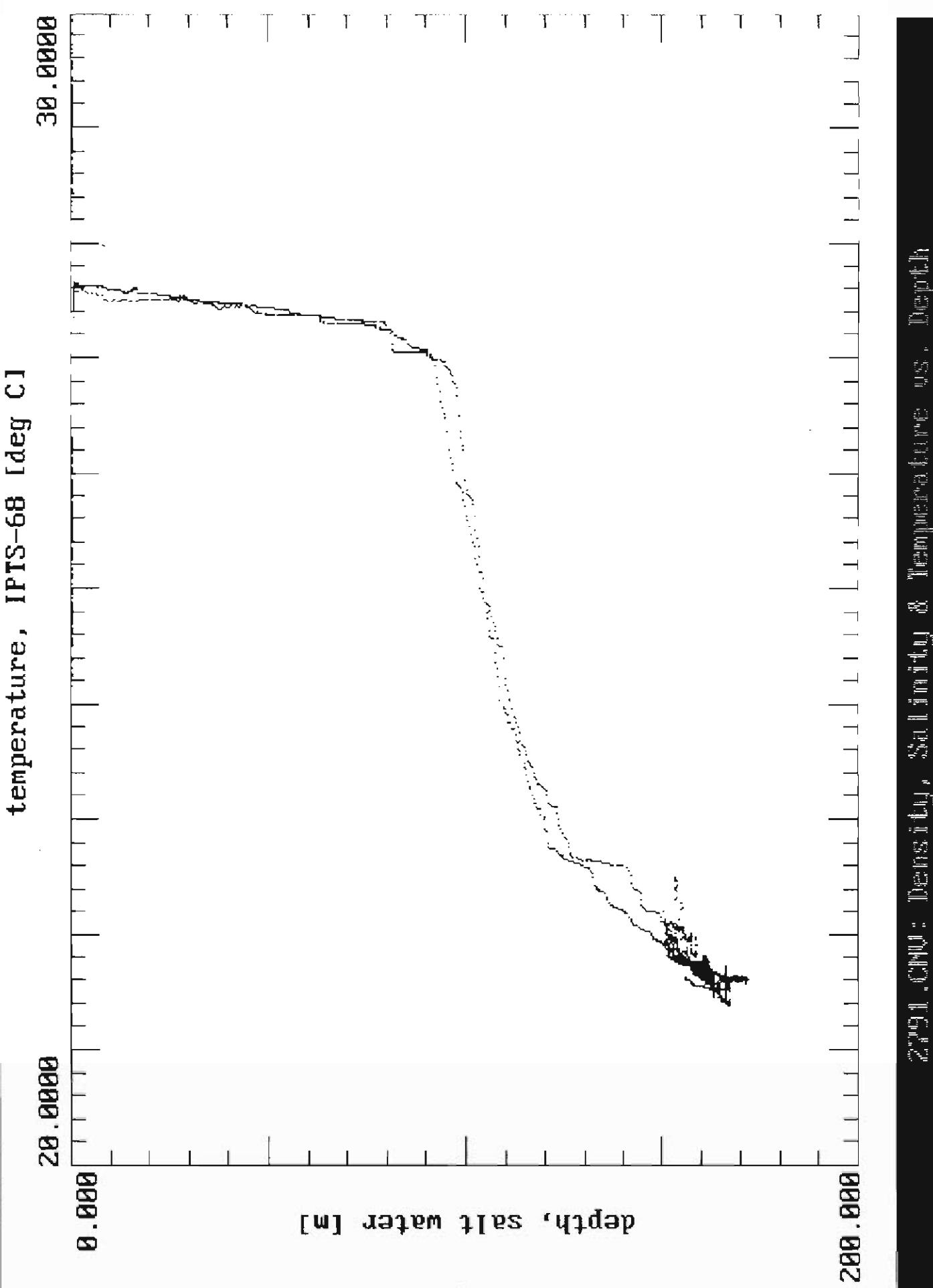
JOHNSON-SEA-LINK II Temperature-Depth Profiles

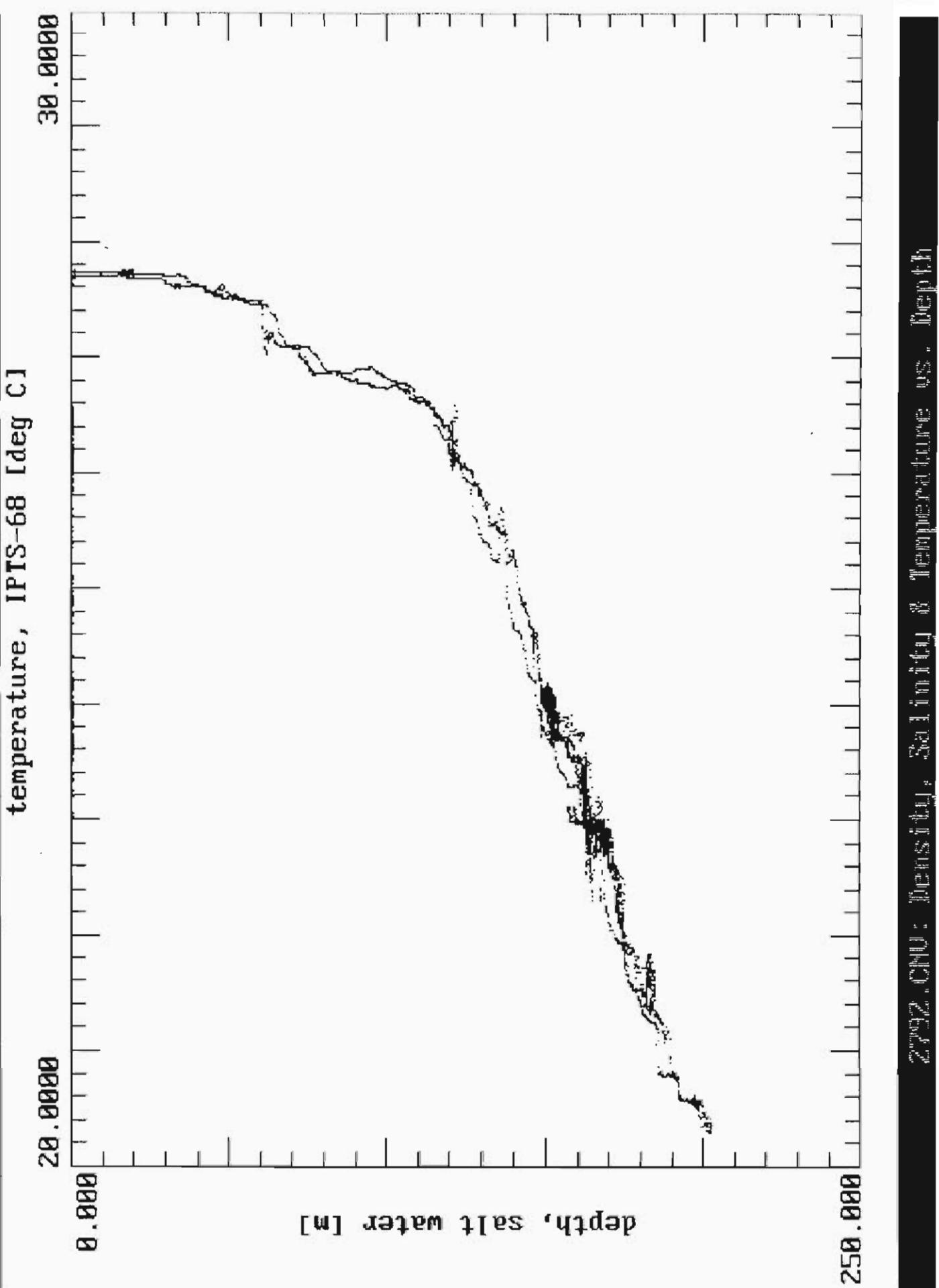
(Number at bottom of figure refers to the JSL Dive Number, e.g.,
2800 = JSL II-2800)



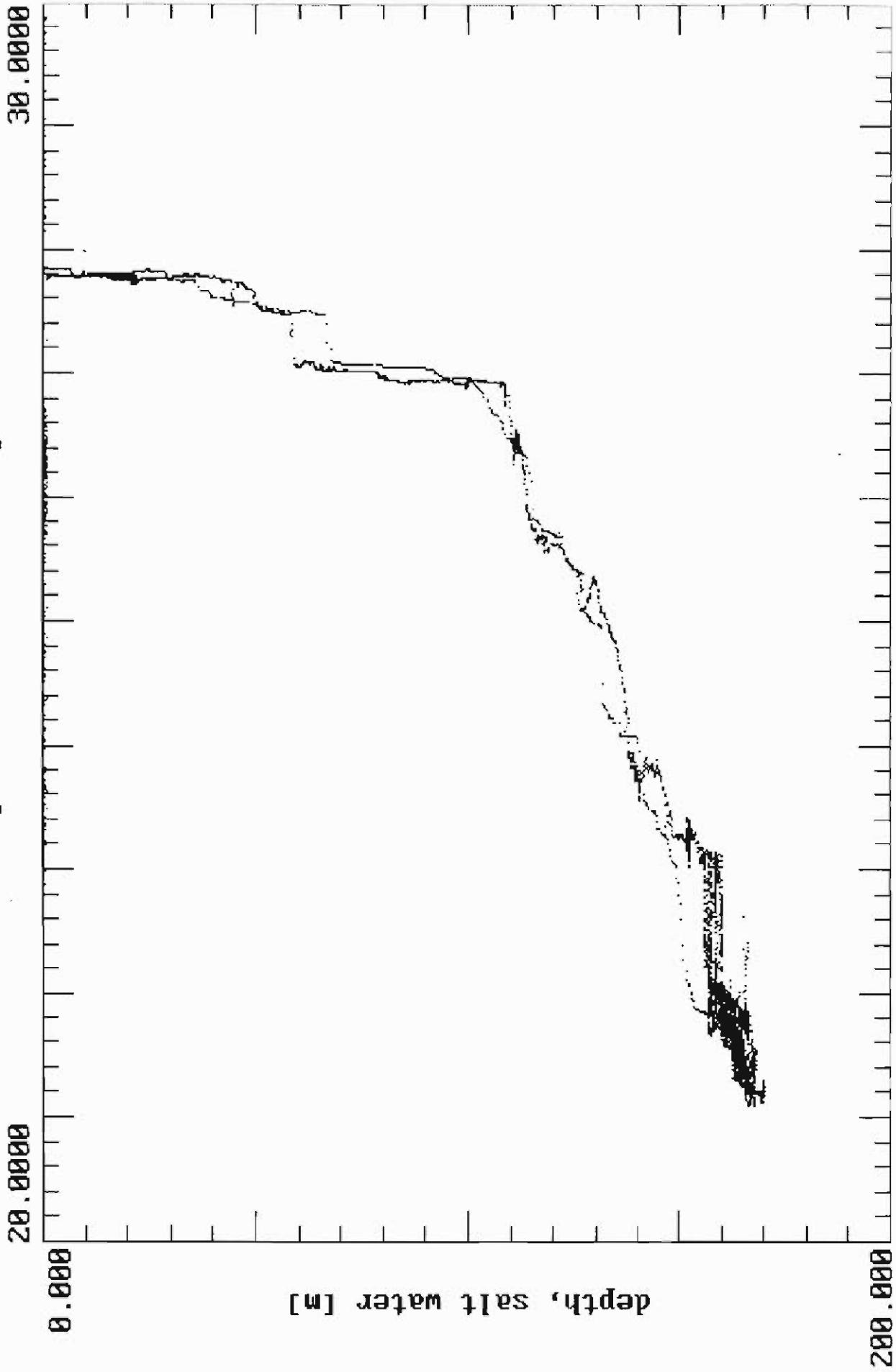
temperature, IPTS-68 [deg C]

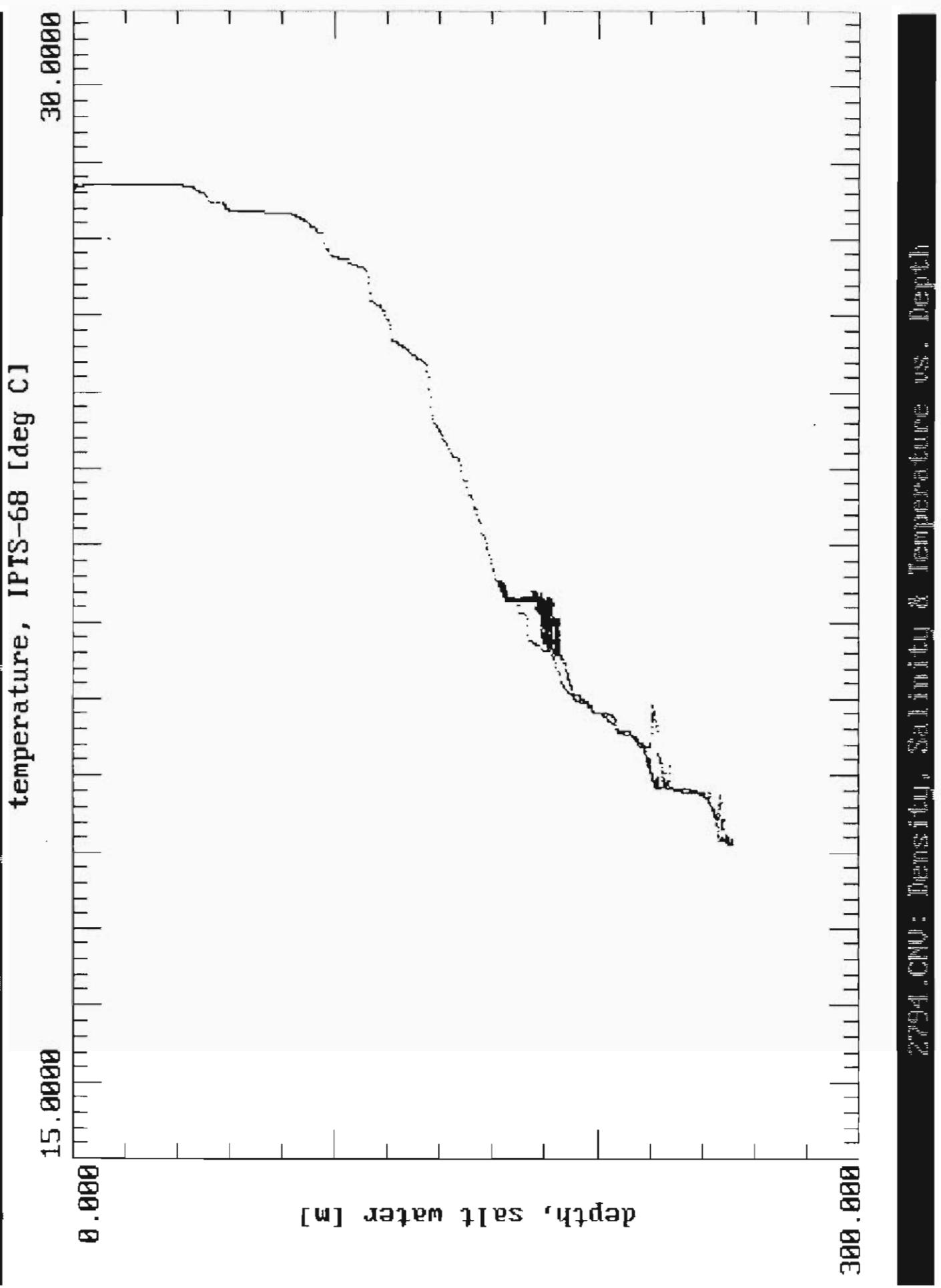


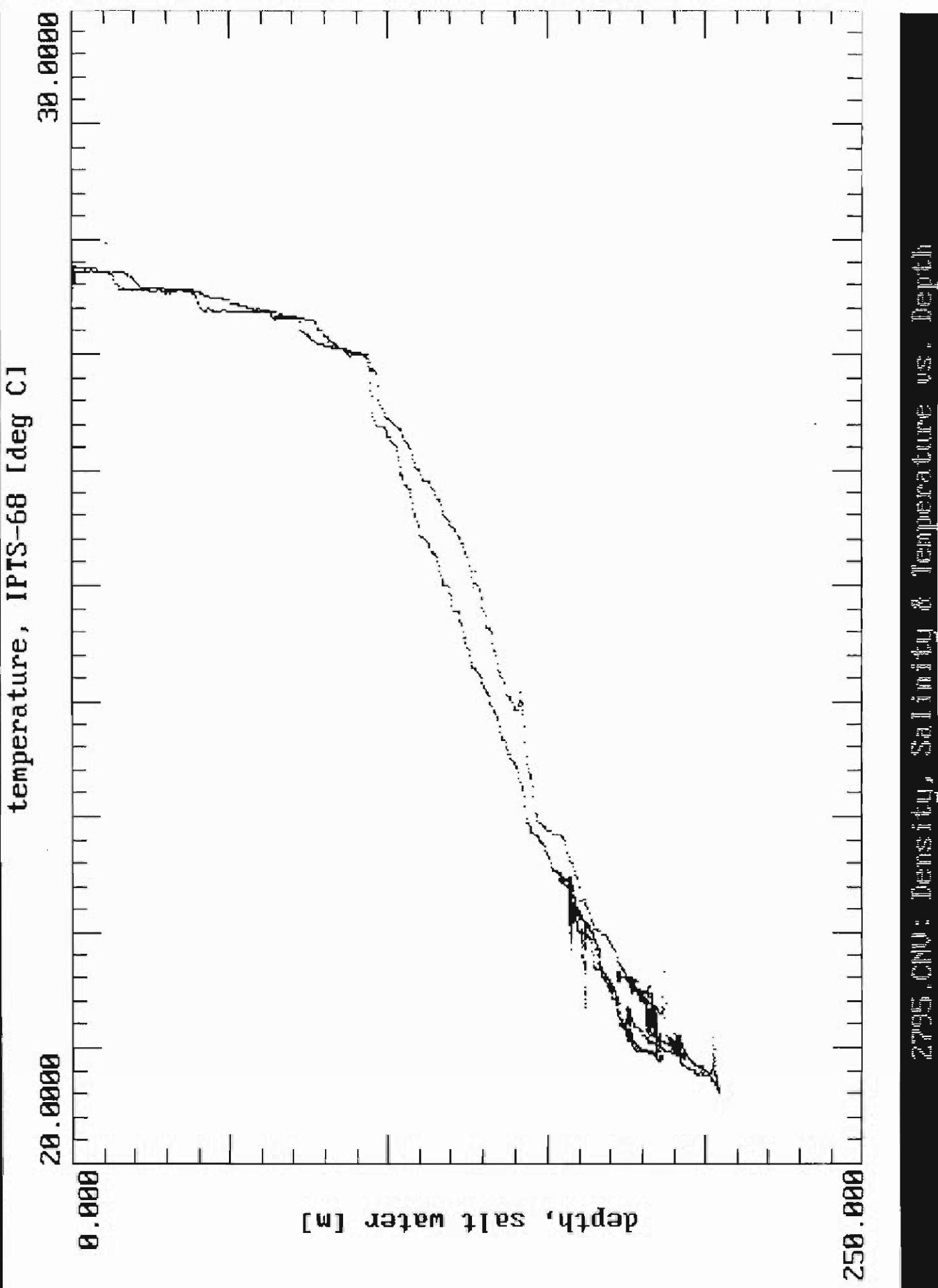




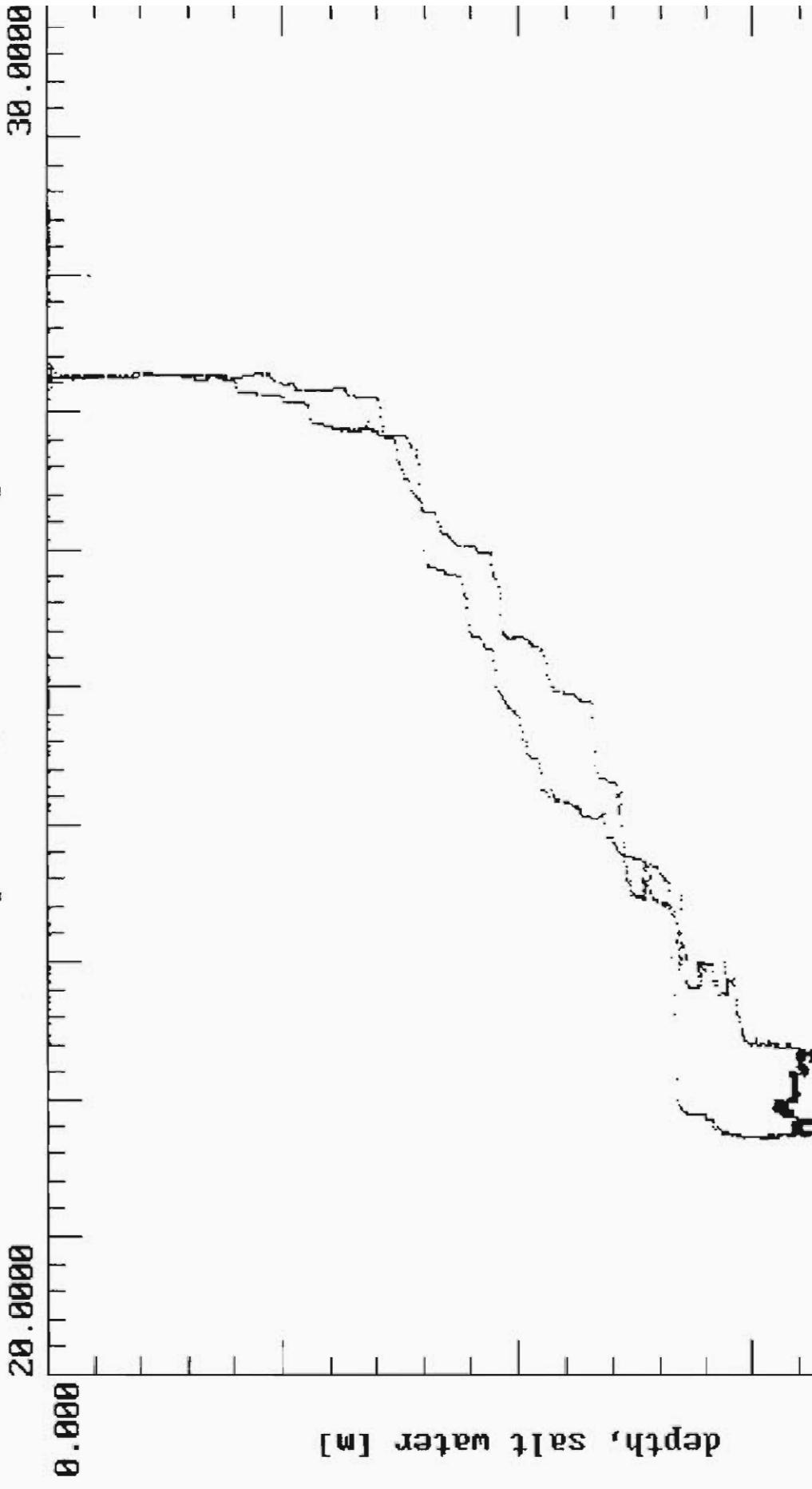
temperature, IPTS-68 [deg C]







temperature, IPTS-68 [deg C]



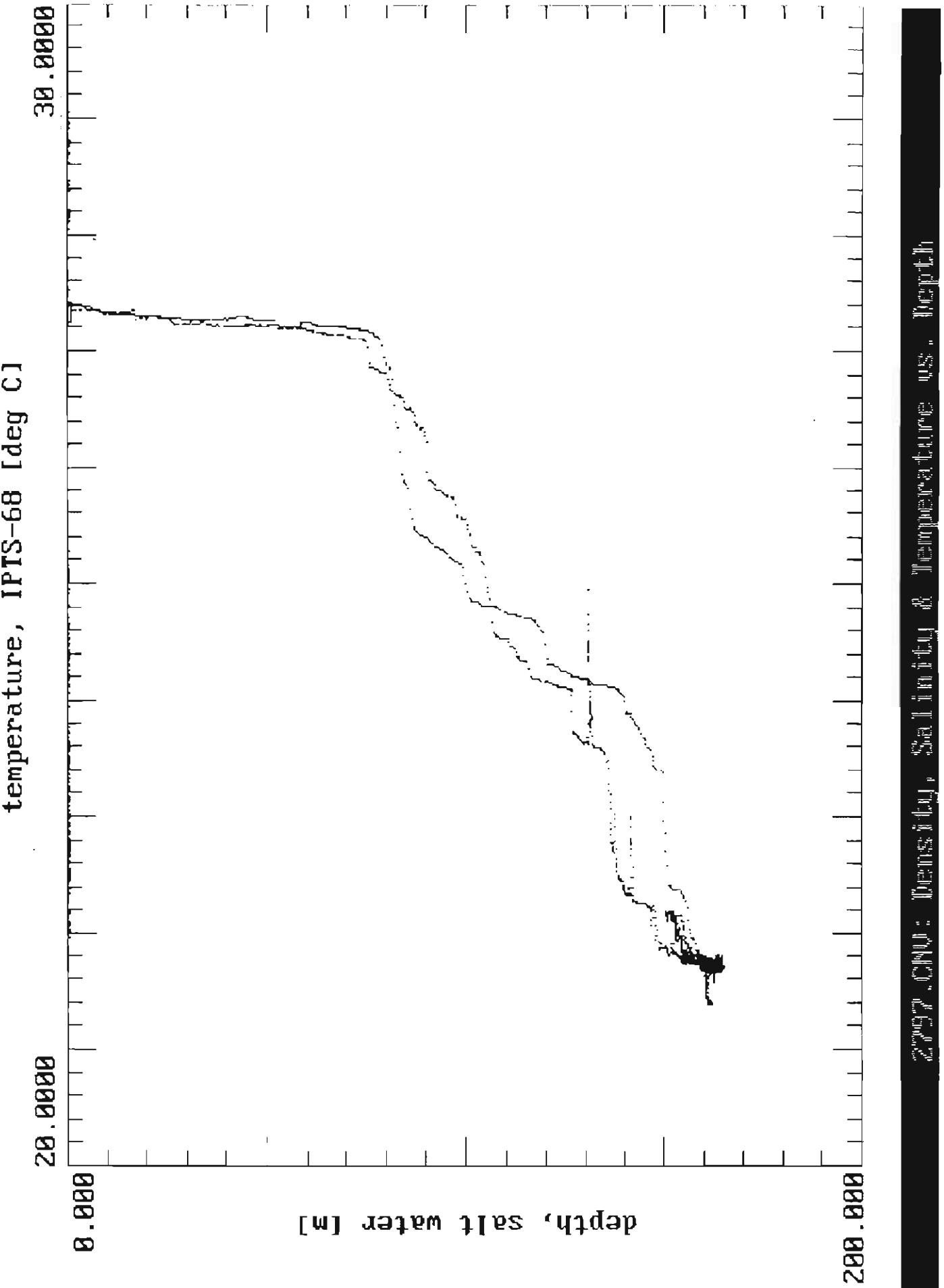
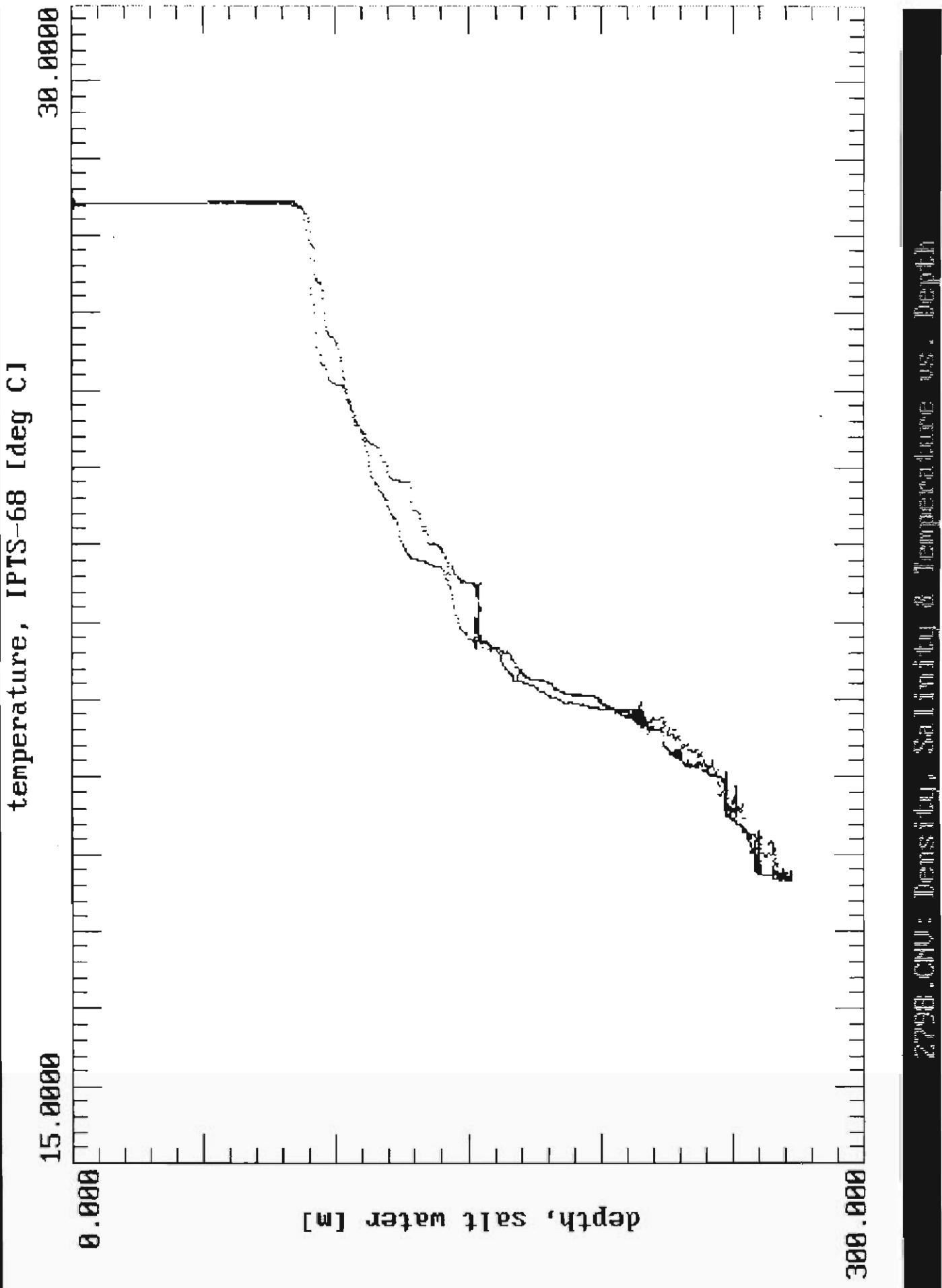
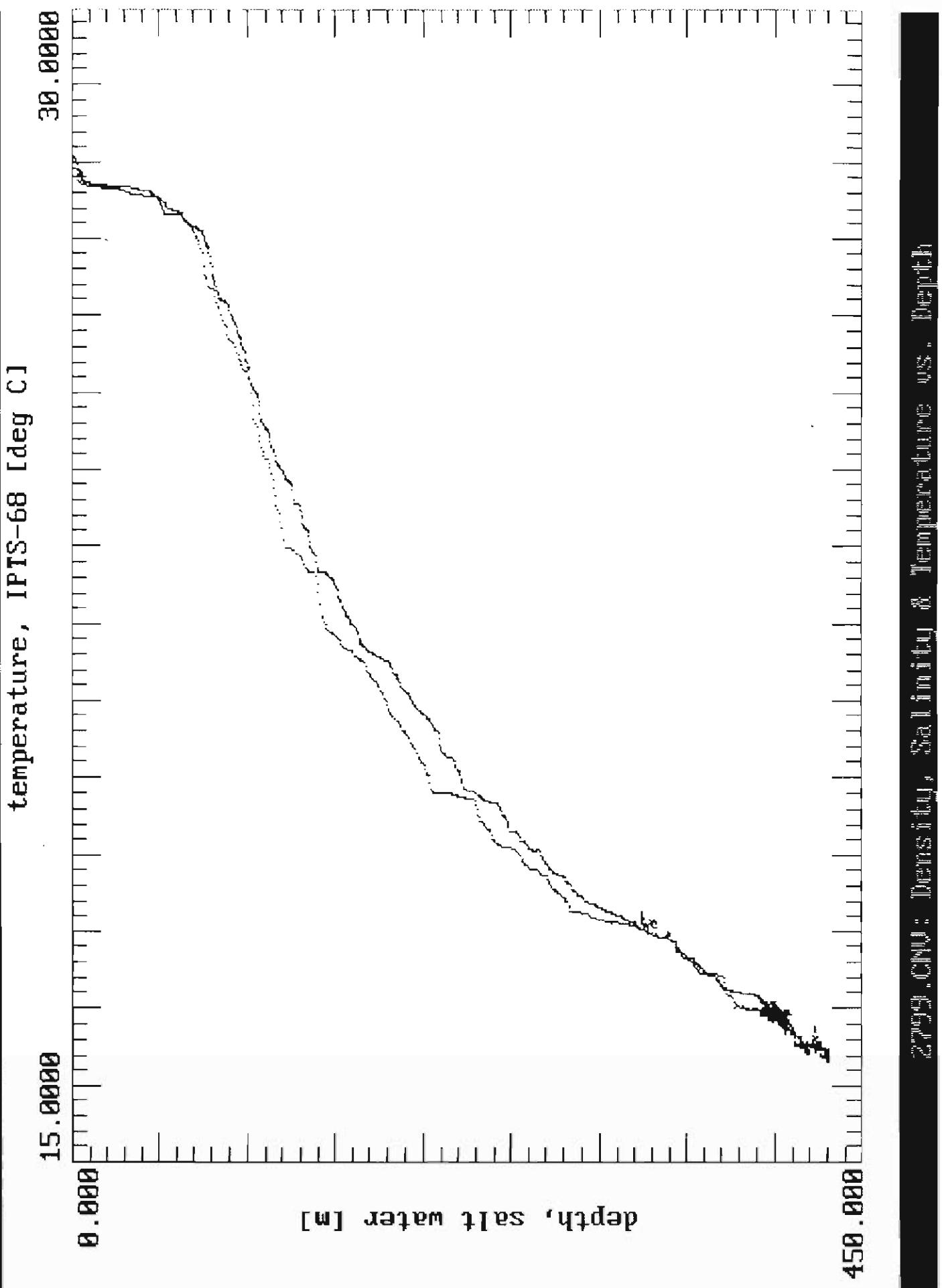


Fig. 10. Water "C" (IPTS-68) Trawl Survey Scenario (EBCS) Experiment



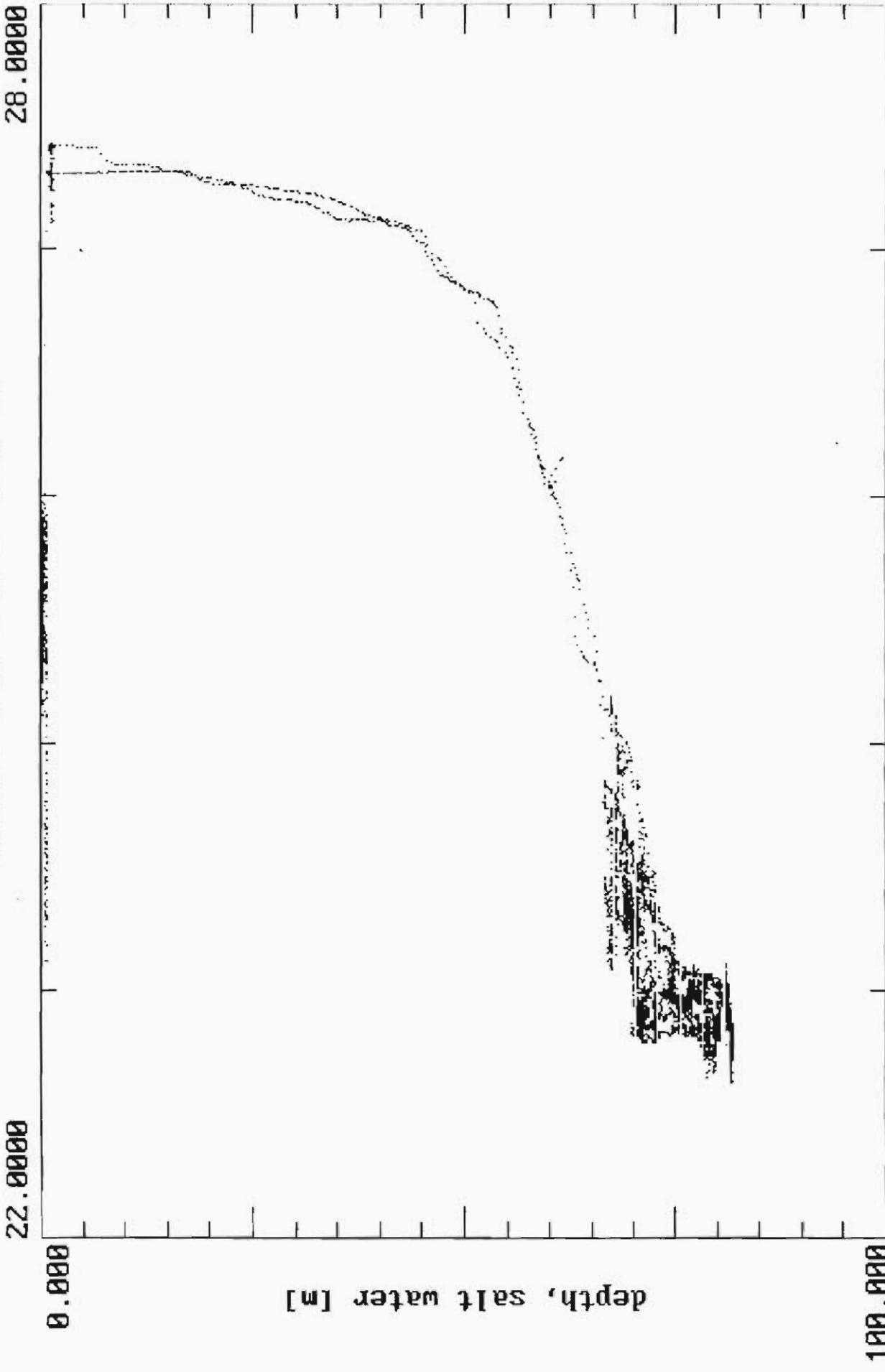


Prod of Water : <CtoI_P7> Unit: Dose [Lau] ; <CtoI_TG3_Mump_Screen> <ESTC> Exit.

temperature, IPTS-68 [deg C]

22.000

0.000



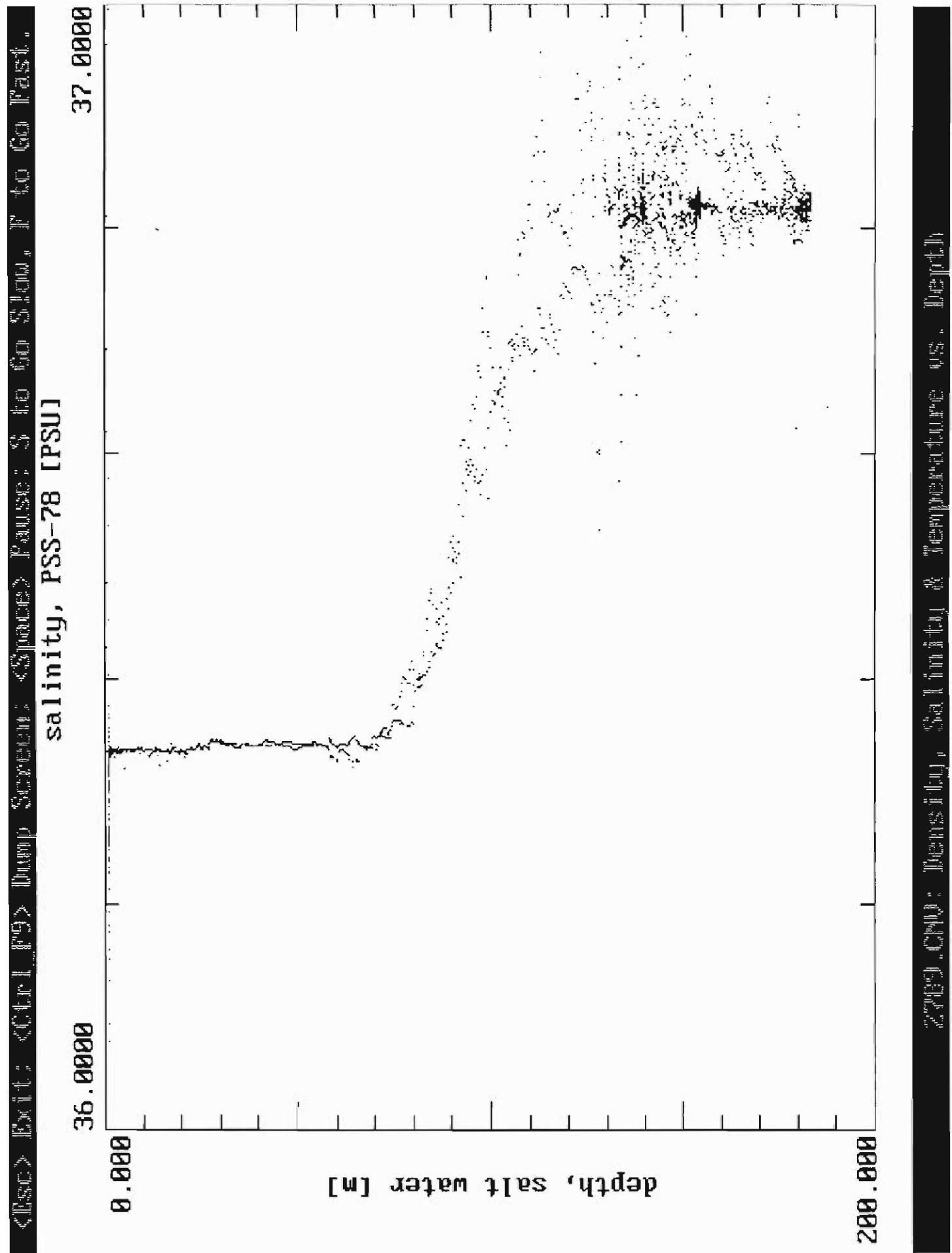
100.000

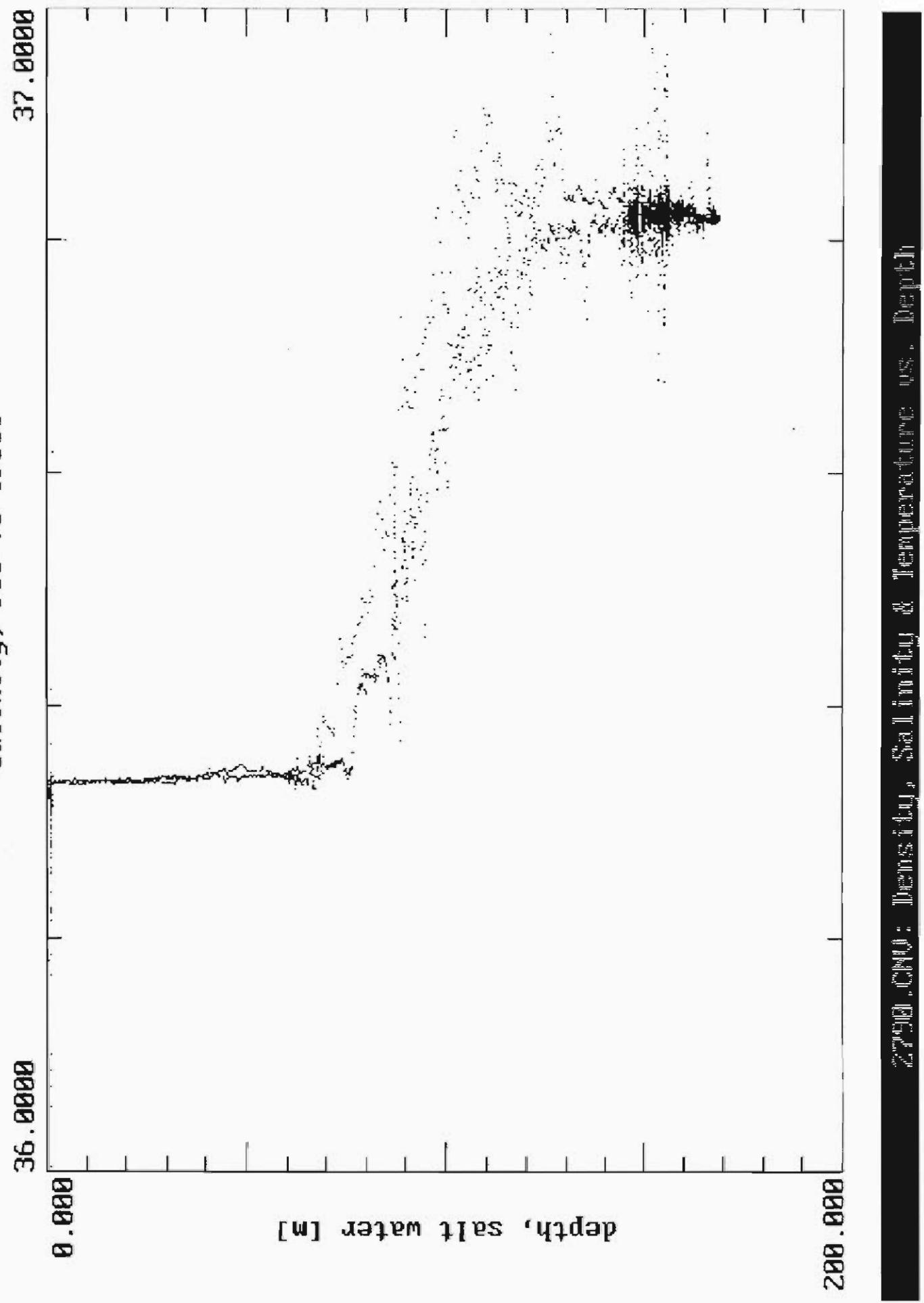
SEED_CW : Dose [Lau] ; Salt in the Temperature [DU] ; Depth [m]

APPENDIX 7

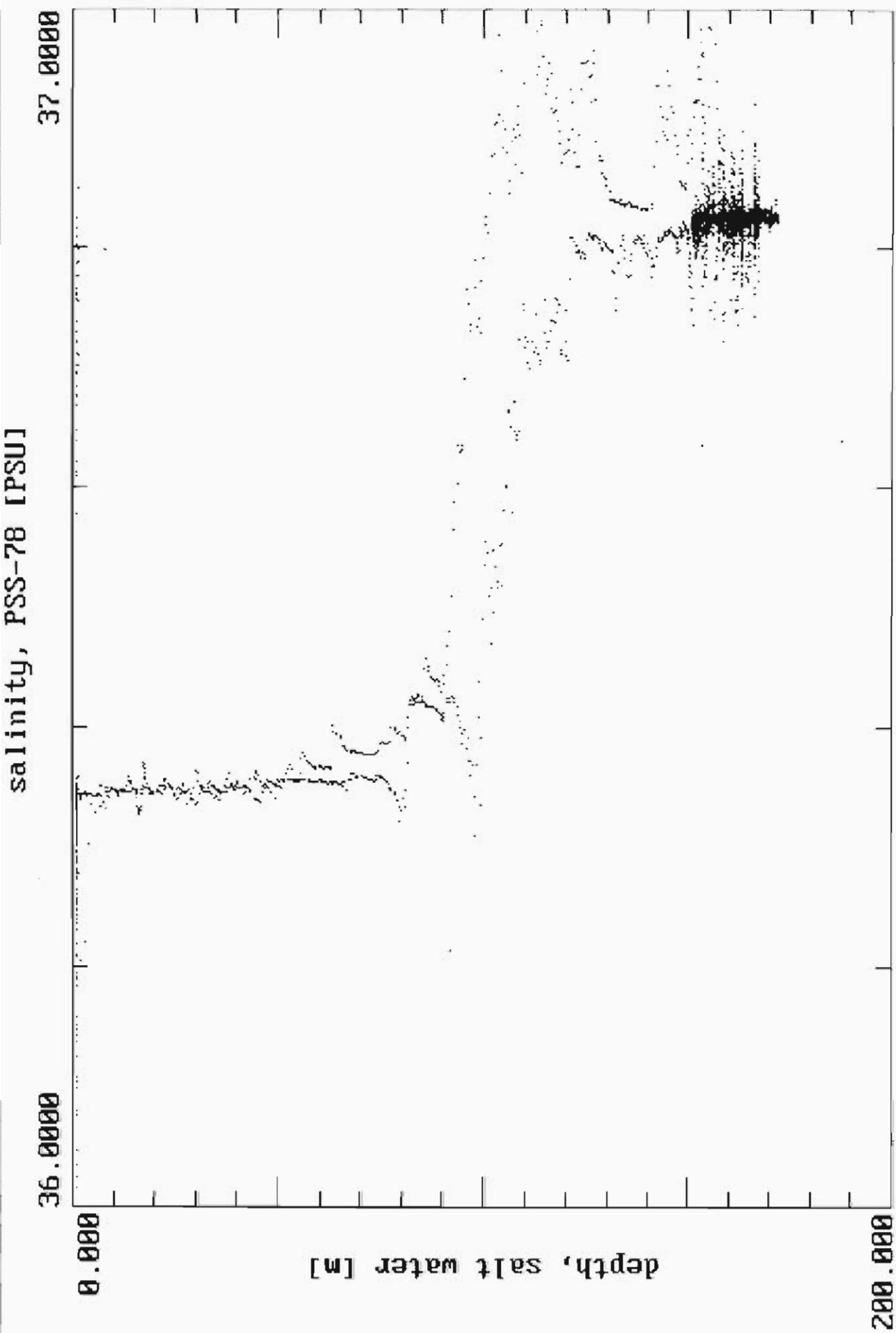
JOHNSON-SEA-LINK II Salinity-Depth Profiles

(Number at bottom of figure refers to the JSL Dive Number, e.g.,
2800 = JSL II-2800)

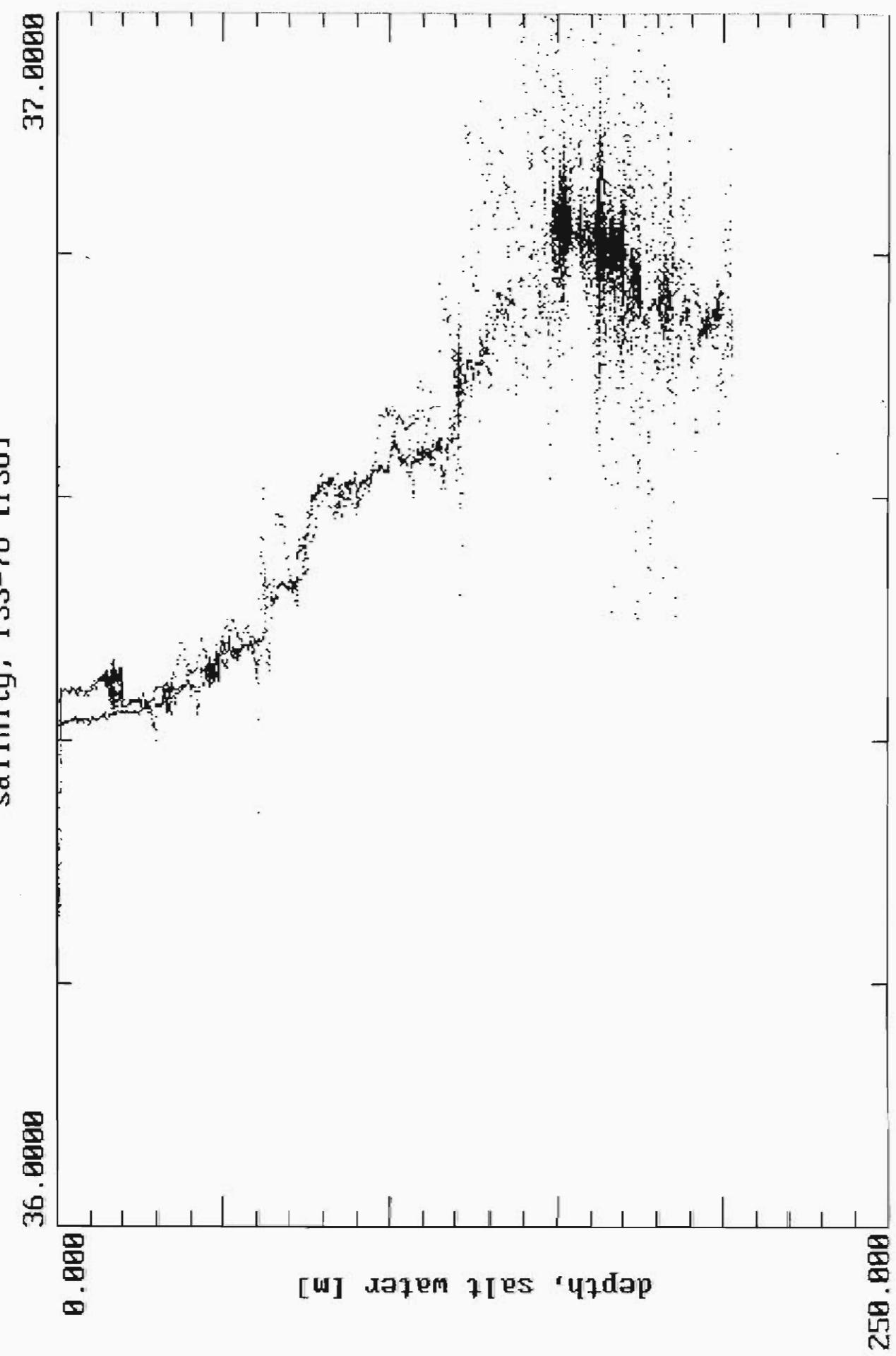




End of Data, CTD_1973, Unit: Farenheit, CTD_Salinity, CTD_Temp, CTD_WaterTemp

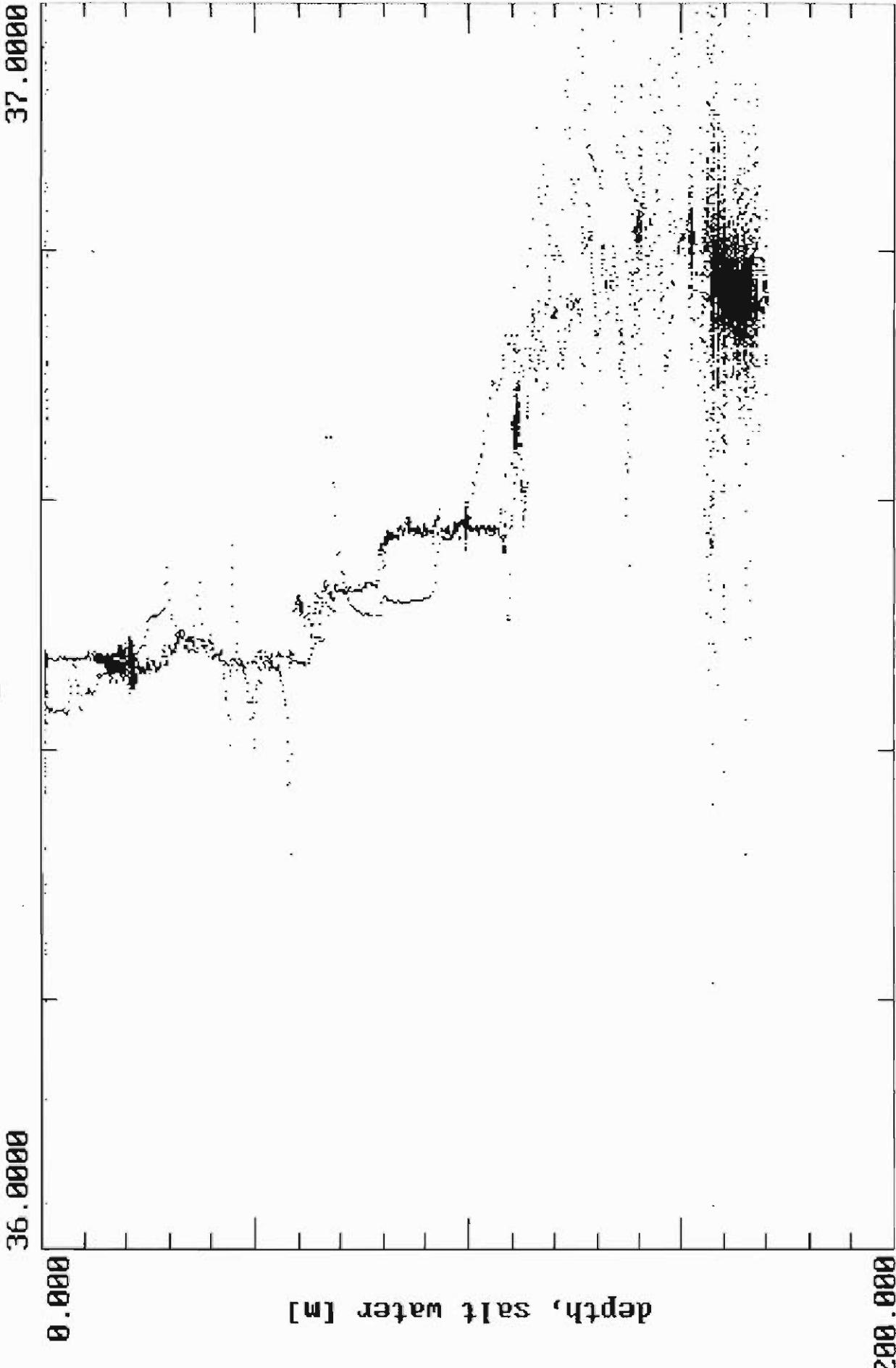


ZERO, CTD: Thermist, Salinity, Temp[water], Temp[air], Date, Depth



End of file, <CRLF> End of file <CRLF> End of file <CRLF> End of file

salinity, PSS-78 [PSU]

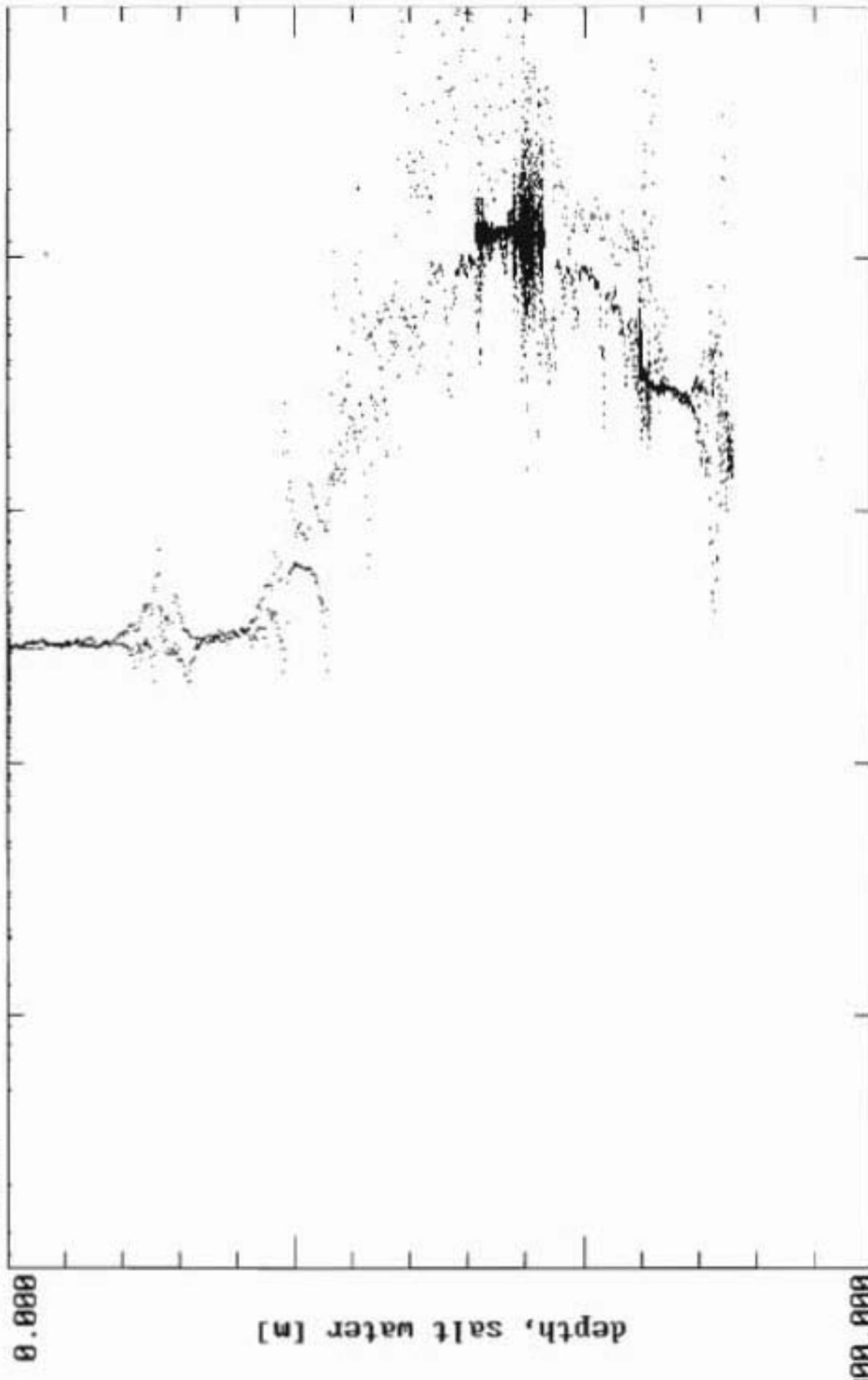


Zeta-2000, Chukchi Plateau, 35°N, 165°E, 2000 m, Depth

End of Data. <Ctrl_F7> Init Overlay; <Ctrl_F9> Print Screen; <Esc> Exit.

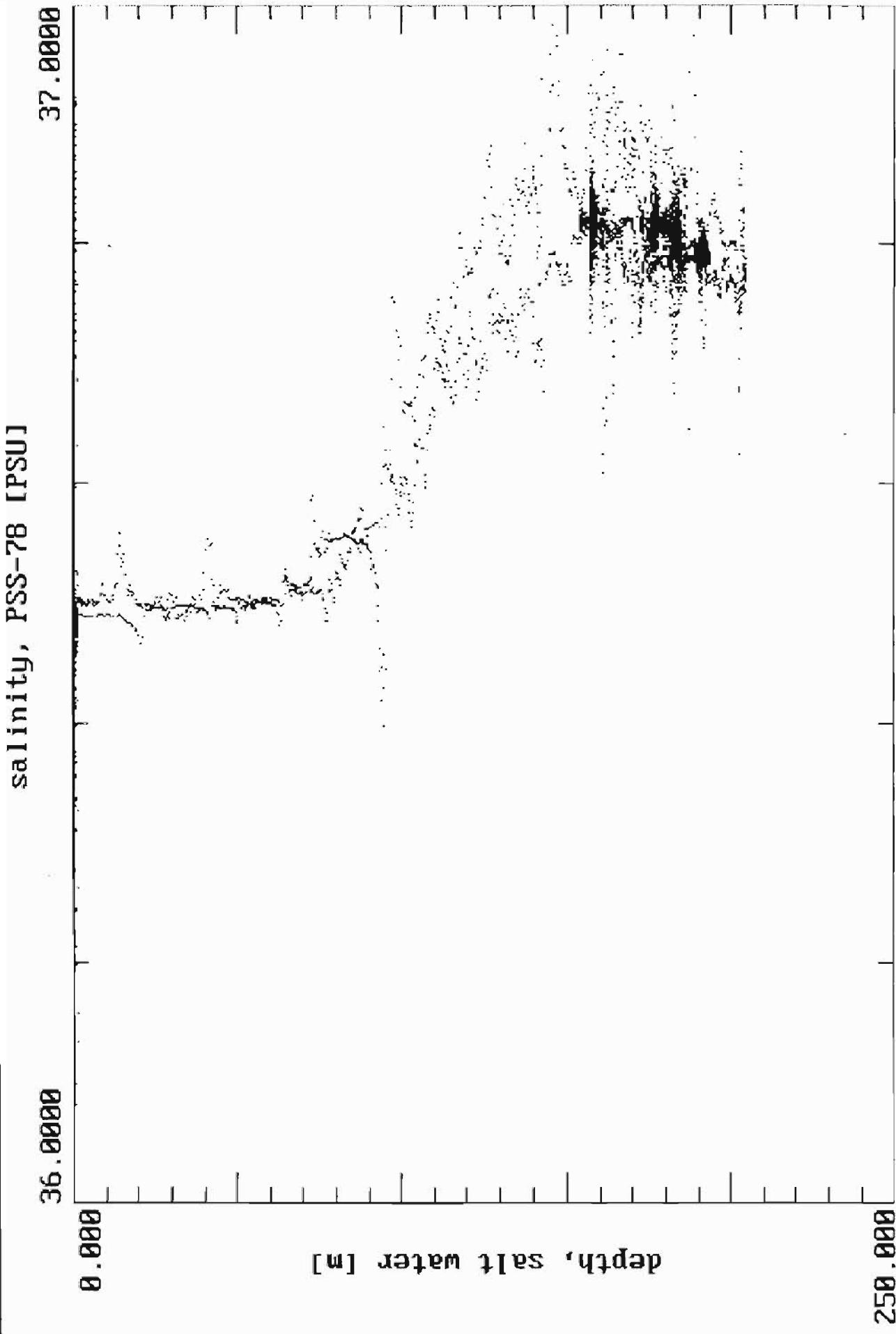
salinity, PSS-78 [PSU]

37.0000
36.0000
0.0000

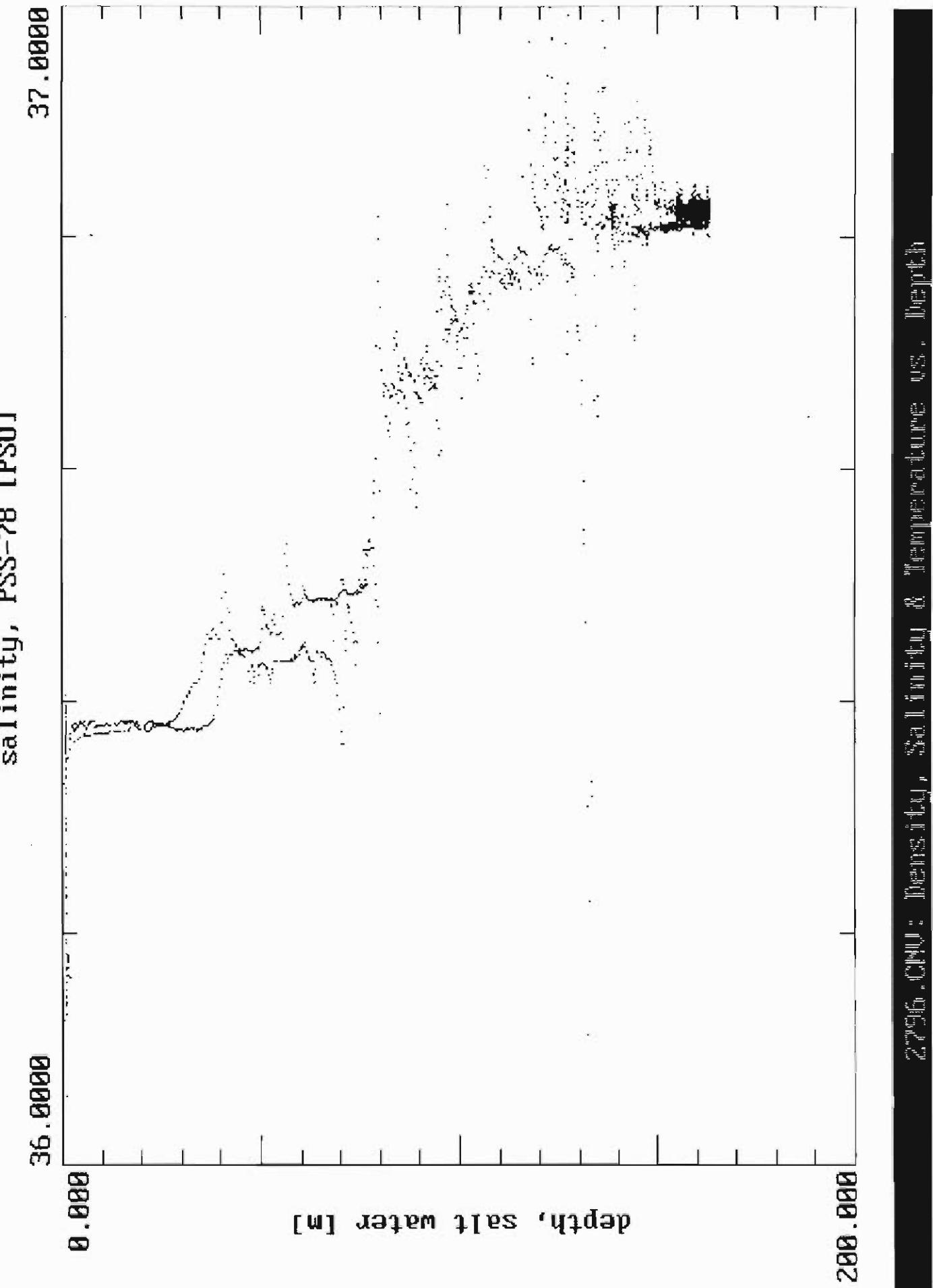


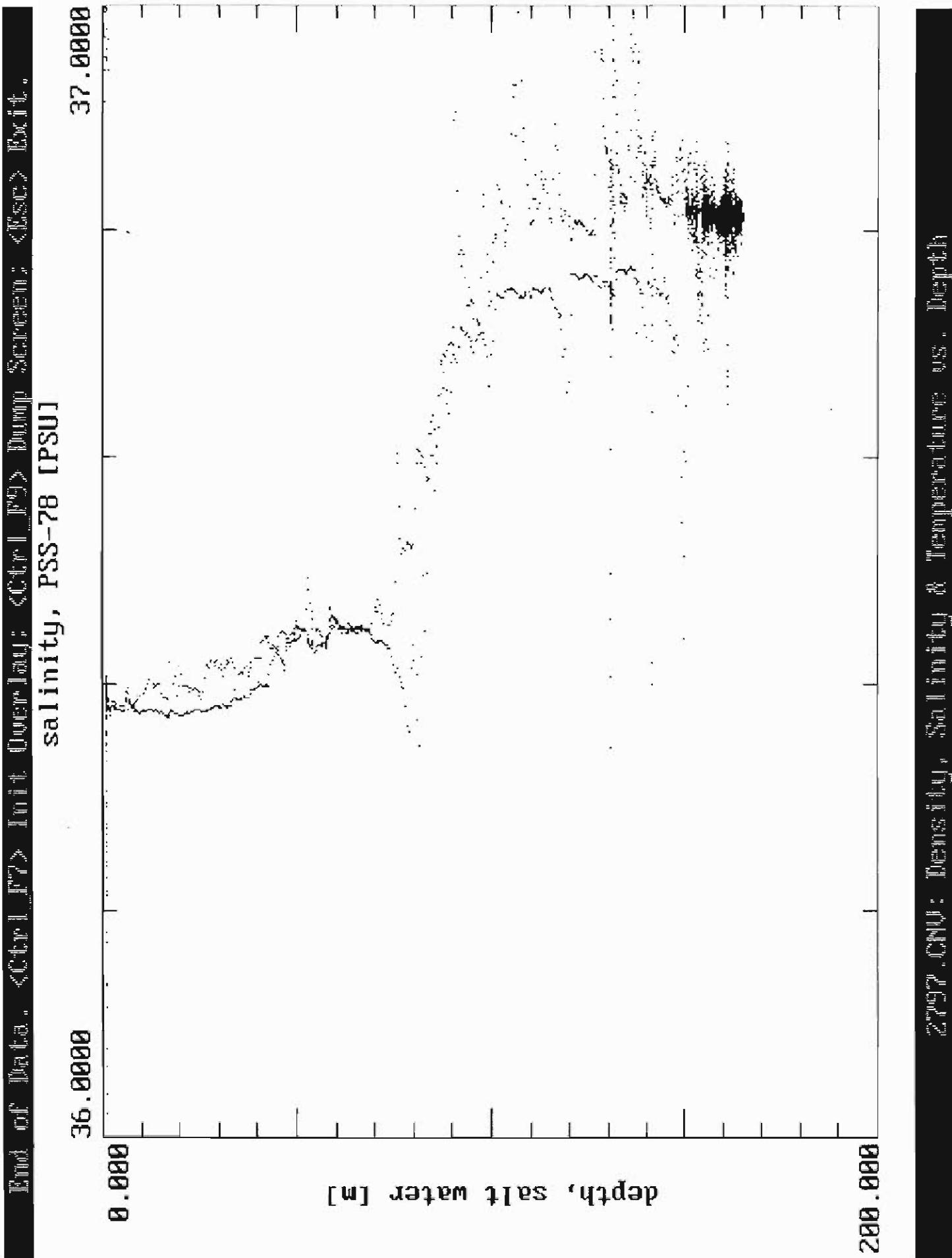
Z794.CNU: Density, Salinity & Temperature vs. Depth

File of Data : <CCW> TPS_19930520_002 Temperature <Egg> Buoy

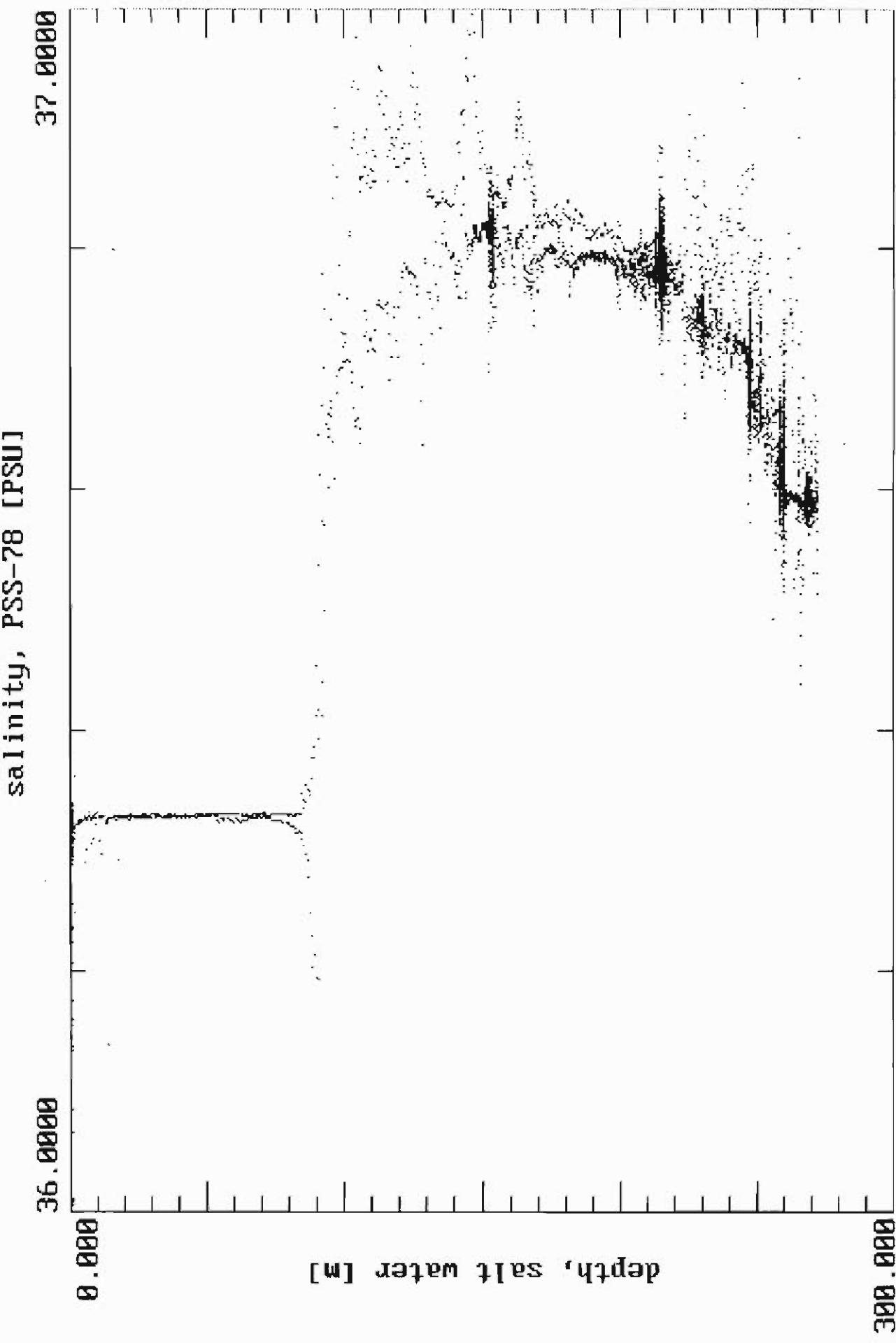


250.000 260.000 270.000 280.000 290.000 300.000 310.000 320.000 330.000 340.000 350.000 36.000 37.000

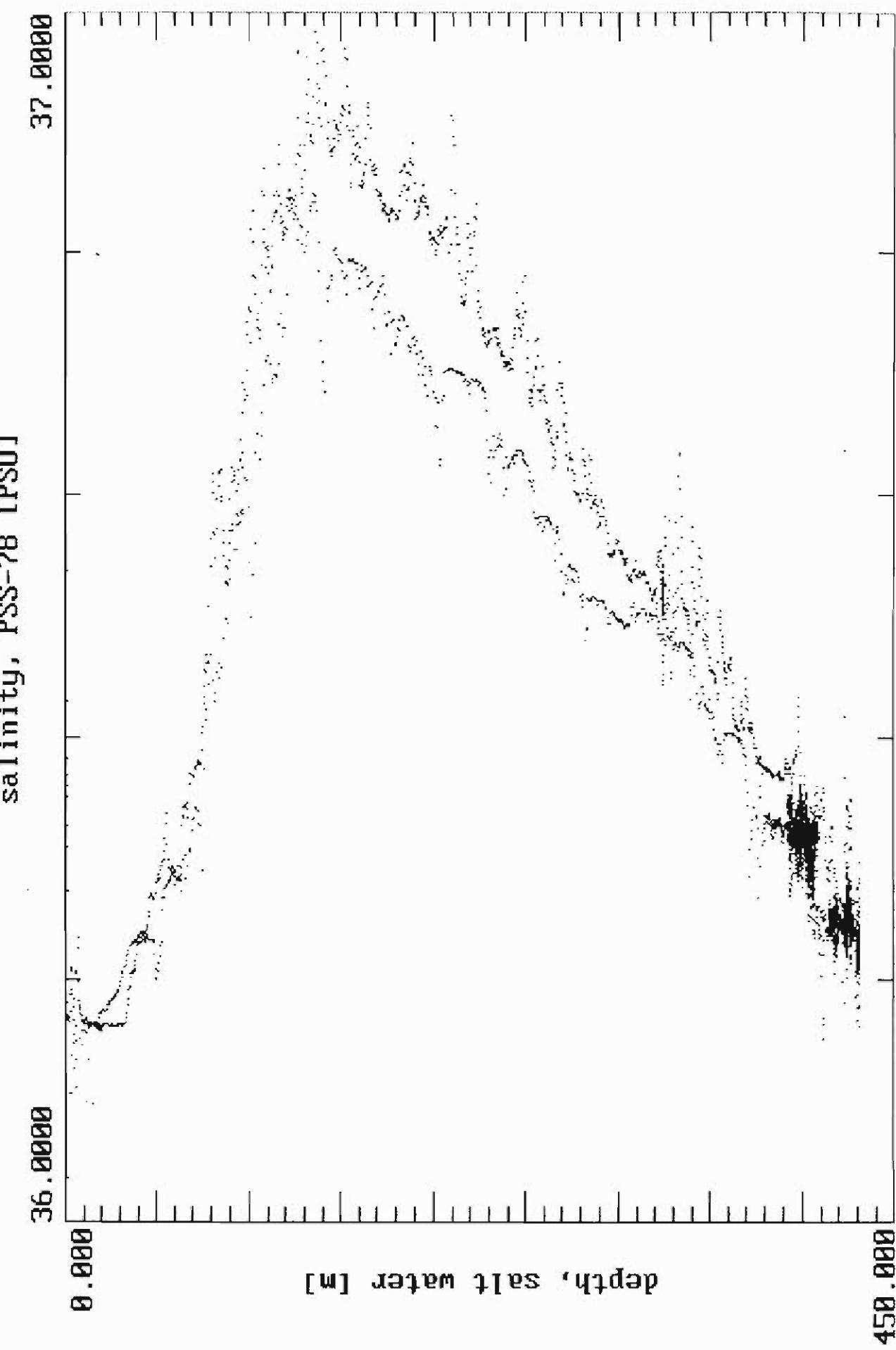


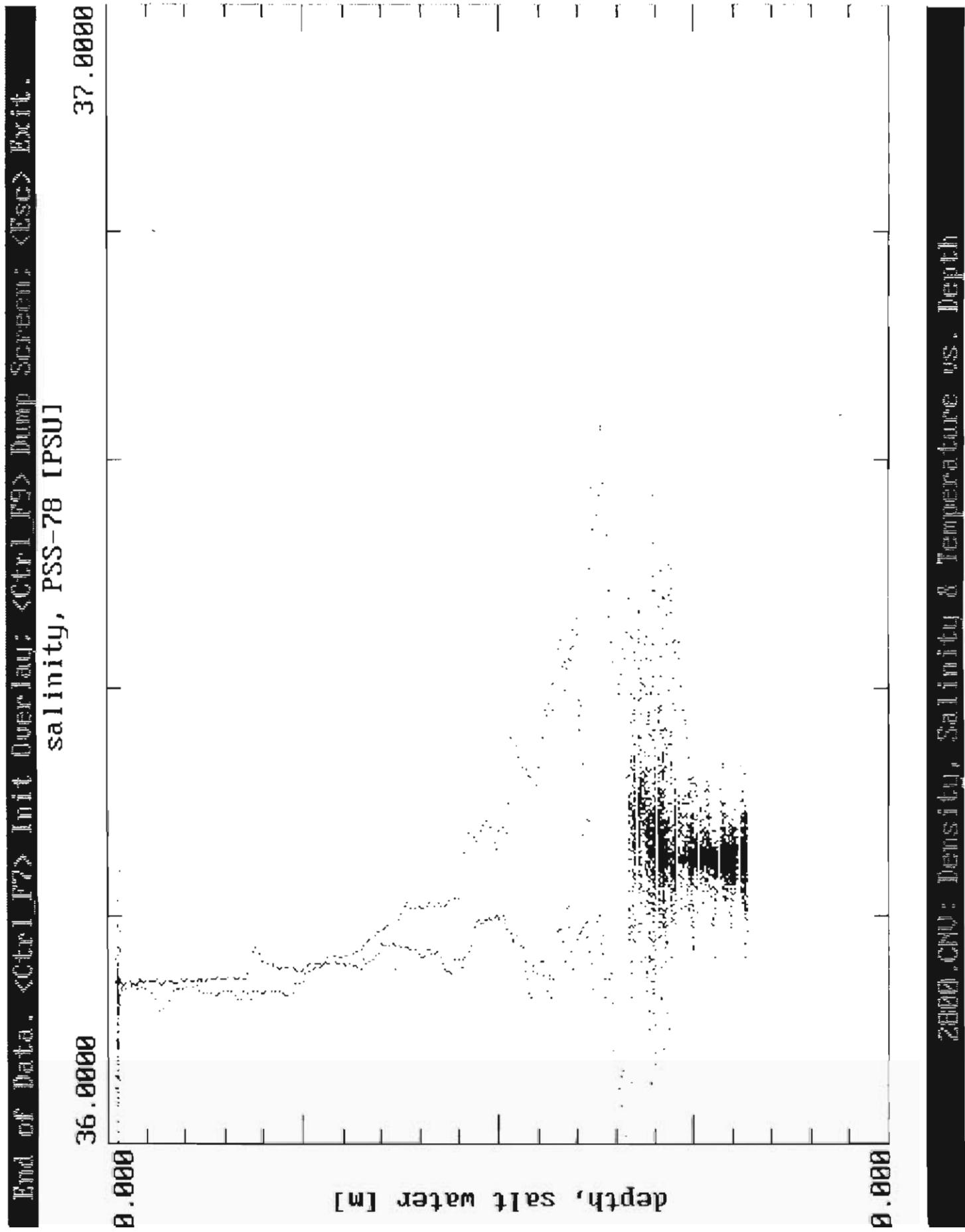


End off Data, <Ctrl+P>, Init, Open File: <Ctrl+T>, Dump Structure, <ESC>, Exit.



ZetaDB, Ctrl+P, Dump Init, Salinity, Depth





APPENDIX 8

Species List of Samples Collected by DBMR

POR= Porifera, CNI= Cnidaria, ANN= Annelida,
PLA= Platyhelminthes, BRY= Bryozoa, MOL= Mollusca,
CRU= Crustacea, ECH= Echinodermata, CHO= Ascidiacea,
ANG= Angiospermata, CHL= Chlorophyta, RHO= Rhodophyta,
PHA= Phaeophyta, CYA= Cyanophyta

SPECIES LIST
BAHAMAS EXPEDITION
JANUARY 15 - 31, 1997
HARBOR BRANCH OCEANOGRAPHIC INSTITUTION
DIVISION OF BIOMEDICAL MARINE RESEARCH

SAMPLE NUMBER (Date+Site+Sample)	PHYLUM	TAXONOMY	RECOLLECT	METHOD	DEPTH (Ft.)
26-X-96-1-001	POR	DISCODERMIA SP.		JSL II-2789	504
26-X-96-1-002	POR	DISCODERMIA SP.IV		JSL II-2789	504
26-X-96-1-301	POR	HALICHONDRIA CORRUGATA		JSL II-2789	504
26-X-96-2-001	POR	DISCODERMIA SP.		JSL II-2790	501
26-X-96-2-002	POR	STELLETTINOPSIS SP.		JSL II-2790	487
26-X-96-2-003	POR	DISCODERMIA SP.		JSL II-2790	512
26-X-96-2-004	POR	STELLETTIDAE		JSL II-2790	550
26-X-96-2-005	POR	BUBARIS SP.		JSL II-2790	550
26-X-96-3-301	CHO	ECTEINASCIDIA TURBINATA		SNORKEL	2
26-X-96-3-302	POR	TEDANIA IGNIS		SNORKEL	2
26-X-96-4-301	POR	ASTEROPUS SP.		SNORKEL	10
26-X-96-4-302	POR	PANDAROS ACANTHIFOLIUM		SNORKEL	10
26-X-96-5-001	POR	DISCODERMIA SP.I		JSL II- 2791	539
26-X-96-5-002	POR	SPONGOSORITES SP.		JSL II- 2791	530
26-X-96-5-003	POR	DISCODERMIA SP.IV		JSL II- 2791	527
26-X-96-5-004	POR	DISCODERMIA SP.I		JSL II- 2791	554
26-X-96-5-005	POR	DISCODERMIA SP.		JSL II- 2791	530
26-X-96-6-001	POR	HYMENIACIDON SP. or AXINELLIDAE		SCUBA	90
26-X-96-6-002	POR	RASPAILIIDAE		SCUBA	85
26-X-96-6-003	CHO	ASCIDIACEA		SCUBA	80
26-X-96-6-301	POR	PTILOCAULIS SPICULIFER?		SCUBA	70
26-X-96-6-302	POR	TEIXAXINELLA MORCHELLA		SCUBA	80
26-X-96-6-303	POR	AMPHIMEDON COMPRESSA		SCUBA	70
26-X-96-6-304	POR	PSUEDAXINELLA LUNAECHARTA		SCUBA	80
26-X-96-6-305	POR	MYRMEKIODERMA SP.		SCUBA	80
26-X-96-6-306	POR	ECTYOPLASIA FEROX		SCUBA	80
26-X-96-6-307	POR	PLAKORTIS SP.		SCUBA	80
26-X-96-6-308	POR	PSEUDOCERATINA CRASSA		SCUBA	80
26-X-96-6-309	POR	CALLYSPONGIA VAGINALIS		SCUBA	70
26-X-96-6-310	POR	AMPHIMEDON COMPRESSA		SCUBA	70
27-X-96-1-001	POR	DISCODERMIA SP.	=1-3, 1-5	JSL II-2792	553
27-X-96-1-002	POR	DISCODERMIA SP.	=1-4	JSL II-2792	548
27-X-96-1-003	POR	DISCODERMIA SP.	=1-1, 1-5	JSL II-2792	543
27-X-96-1-004	POR	DISCODERMIA SP.	=1-2	JSL II-2792	534
27-X-96-1-005	POR	DISCODERMIA SP.	=1-1, 1-3	JSL II-2792	535
27-X-96-1-006	POR	ERYLUS SP.		JSL II-2792	516
27-X-96-1-007	POR	DISCODERMIA SP.		JSL II-2792	535
27-X-96-1-301	POR	WILLARDIA CAICOSENSIS		JSL II-2792	398
27-X-96-2-001	POR	HALISARCA? SP.		SCUBA	70
27-X-96-2-002	POR	IOTROCHOTA SP.		SCUBA	70
27-X-96-2-003	POR	AXINELLIDAE?		SCUBA	84
27-X-96-2-004	POR	SIPHONODICTYON MUCOSUM		SCUBA	100
27-X-96-2-005	POR	MYXILLIDAE		SCUBA	60
27-X-96-2-301	POR	TOPSENTIA PORRECTA?		SCUBA	80
27-X-96-2-302	POR	OCEANAPHIDAE? or NIPHATIDAE?		SCUBA	80
27-X-96-3-001	POR	DISCODERMIA SP.IV (IRREGULAR CUPS-		JSL II-2793	536

SAMPLE NUMBER (Date+Site+Sample)	PHYLUM	TAXONOMY	RECOLLECT	METHOD	DEPTH (Ft.)
27-X-96-3-002	POR	DISCODERMIA SP.IV (IRREGULAR STALK		JSL II-2793	530
27-X-96-3-003	POR	DISCODERMIA SP.IV		JSL II-2793	539
27-X-96-3-004	POR	DISCODERMIA SP.IV		JSL II-2793	517
27-X-96-3-005	POR	DISCODERMIA SP.		JSL II-2793	531
27-X-96-3-006	POR	DISCODERMIA SP.I		JSL II-2793	527
27-X-96-3-007	POR	DISCODERMIA SP.IV?		JSL II-2793	524
27-X-96-3-008	POR	DISCODERMIA SP.IV		JSL II-2793	534
27-X-96-3-009	POR	DISCODERMIA SP.IV		JSL II-2793	524
27-X-96-3-010	POR	DISCODERMIA SP.IV		JSL II-2793	545
27-X-96-3-011	POR	DISCODERMIA SP.		JSL II-2793	525
27-X-96-3-012	POR	DISCODERMIA SP.IV		JSL II-2793	501
27-X-96-3-301	POR	CRIBROCHALINA VASCULUM		JSL II-2793	530
27-X-96-3-302	POR	SPIROPHORIDA		JSL II-2793	521
27-X-96-3-303	POR	PHIACKELIA FOLIUM		JSL II-2793	367
27-X-96-3-304	POR	CALCIFIBROSPONGIA SP.		JSL II-2793	327
27-X-96-3-305	POR	LEUCETTA SP.		JSL II-2793	267
28-X-96-1-001	POR	ASTEROPUS SP.		JSL II-2794	748
28-X-96-1-002	POR	SPONGOSORITES SP.		JSL II-2794	728
28-X-96-1-003	POR	ASTEROPUS SP.	= 1-1	JSL II-2794	720
28-X-96-1-004	POR	ERYLUS cf. GOFFRILLERI		JSL II-2794	591
28-X-96-1-005	POR	HIPPOSPOONGIA SP.		JSL II-2794	583
28-X-96-1-006	POR	DISCODERMIA cf. SP.IV		JSL II-2794	560
28-X-96-1-007	POR	STELLETTA SP.		JSL II-2794	540
28-X-96-1-008	POR	DISCODERMIA cf. SP.I		JSL II-2794	592
28-X-96-1-009	POR	CORTICIUM SP.		JSL II-2794	604
28-X-96-1-010	POR	SPONGOSORITES SP.		JSL II-2794	600
28-X-96-1-301	POR	XESTOSPOONGIA cf. STONEAE		JSL II-2794	600
28-X-96-1-302	POR	CORTICIUM SP.2		JSL II-2794	600
28-X-96-1-303	POR	AGELAS SP.		JSL II-2794	600
28-X-96-2-001	POR	CORALLISTES cf. MICROSTYLIFER		JSL II-2795	580
28-X-96-2-002	POR	GEODIA NEPTUNI		JSL II-2795	603
28-X-96-2-003	POR	GEODIA SP.		JSL II-2795	627
28-X-96-2-004	POR	SPONGOSORITES SP.		JSL II-2795	631
28-X-96-2-005	POR	PETROSIIDAE		JSL II-2795	518
28-X-96-2-006	POR	SPONGOSORITES SP.		JSL II-2795	630
28-X-96-3-001	POR	BIEMNA SP.		SCUBA	75
28-X-96-3-201	POR	ECTYOPLASIA FEROX		SCUBA	75
28-X-96-4-301	POR	TEDANIA IGNIS		SNORKEL	2
28-X-96-4-N	CHO	ECTEINASCIDIA TURBINATA + PSEUDOC		SNORKEL	2
29-X-96-1-001	POR	DISCODERMIA? SP.		JSL II-2796	525
29-X-96-1-002	POR	SPONGOSORITES SP.		JSL II-2796	520
29-X-96-1-003	POR	DISCODERMIA? SP.		JSL II-2796	522
29-X-96-1-004	POR	DISCODERMIA SP.		JSL II-2796	522
29-X-96-1-005	POR	DISCODERMIA SP.		JSL II-2796	513
29-X-96-1-006	POR	DISCODERMIA SP.		JSL II-2796	520
29-X-96-1-007	POR	DISCODERMIA SP.		JSL II-2796	520
29-X-96-3-301	CHO	ECTEINASCIDIA TURBINATA		SNORKEL	2
29-X-96-4-001	POR	DISCODERMIA SP.IV		JSL II-2797	530
29-X-96-4-002	POR	DISCODERMIA SP.I		JSL II-2797	527
29-X-96-4-003	POR	DISCODERMIA SP.I		JSL II-2797	520
29-X-96-4-004	POR	DISCODERMIA SP.I		JSL II-2797	520
29-X-96-4-005	POR	DISCODERMIA SP.I?		JSL II-2797	520
29-X-96-4-006	POR	DISCODERMIA cf. VARRUCOSA		JSL II-2797	520



SAMPLE NUMBER (Date+Site+Sample)	PHYLUM	TAXONOMY	RECOLLECT	METHOD	DEPTH (Ft.)
29-X-96-4-007	POR	DISCODERMIA SP.IV?	JSL II-2797	520	
29-X-96-4-008	POR	DISCODERMIA SP.	JSL II-2797	520	
30-X-96-1-001	POR	ASTEROPUS SP.	JSL II-2798	882	
30-X-96-1-002	POR	ASTEROPUS SP.	JSL II-2798	853	
30-X-96-1-003	POR	ASTEROPUS SP.	JSL II-2798	849	
30-X-96-1-004	POR	SPONGOSORITES? SP.	JSL II-2798	814	
30-X-96-1-005	POR	ASTEROPUS SP.	JSL II-2798	758	
30-X-96-1-006	POR	SPONGOSORITES SP.	JSL II-2798	709	
30-X-96-1-007	POR	ACICULITES SP. or GASTROPHANELLA SP.	JSL II-2798	708	
30-X-96-1-008	CNI	ACANTHOGORGIIIDAE	JSL II-2798	501	
30-X-96-1-009	POR	DISCODERMIA SP.IV	JSL II-2798	708	
30-X-96-1-010	POR	AXINELLIDAE	JSL II-2798	708	
30-X-96-2-001	POR	CAMINUS SP.	JSL II-2799	1404	
30-X-96-2-002	POR	GEODIA SP.	JSL II-2799	1404	
30-X-96-2-003	POR	VETULINA N.SP.	JSL II-2799	1404	
30-X-96-2-004	POR	TOPSENTIA SP.	JSL II-2799	1392	
30-X-96-2-005	POR	PLAKINASTRELLA (DERCITOPSIS) SP.	JSL II-2799	1392	
30-X-96-2-006	POR	EPIPOLASIS SP.	JSL II-2799	1394	
30-X-96-2-007	POR	TOPSENTIA? PORRECTA?	JSL II-2799	1394	
30-X-96-2-008	POR	OCEANAPIA SP.	JSL II-2799	1391	
30-X-96-2-009	CNI	PRIMNOIDAE	JSL II-2799	1312	
30-X-96-2-010	POR	STRONGYLOPHORA SP.	JSL II-2799	1313	
30-X-96-2-011	CNI	GORGONACEA	JSL II-2799	1323	
30-X-96-3-001	POR	MYXILLIDAE	SCUBA	70	
31-X-96-1-201	SED	SEDIMENT	JSL II-2800	265	

APPENDIX 9

Checklist of *In-situ* and Laboratory Photographs, Videotapes,
Taxonomic Museum Vouchers, Microorganism Isolates,
Invertebrate Cell Culture Samples, and Thin Layer
Chromatography Samples

SAMPLE DOCUMENTATION
BAHAMAS EXPEDITION
JANUARY 15 - 31, 1997
HARBOR BRANCH OCEANOGRAPHIC INSTITUTION
DIVISION OF BIOMEDICAL MARINE RESEARCH

SAMPLE NUMBER (Date + Site # + Sample #)	IN-SITU 35-MM PHOTO	LAB 35-MM PHOTO	MUSEUM SPECIMEN		PRESER- VATIVE	MICROBIAL ISOLATION		CELL CULTURE	TLC
			(Large)	(Small)		(Agar)	(Freeze)		
26-X-96-1-001	Y	Y	N	Y	70ET	Y	Y	N	Y
26-X-96-1-002	N	Y	N	Y	70ET	Y	Y	N	Y
26-X-96-1-301	N	MKB	MKB	MKB	70ET	N	N	N	N
26-X-96-2-001	Y	Y	H	Y	70ET	Y	Y	N	Y
26-X-96-2-002	Y	Y	H	Y	70ET	N	Y	N	Y
26-X-96-2-003	Y	Y	H	Y	70ET	Y	Y	Y	Y
26-X-96-2-004	Y	Y	H	Y	70ET	N	Y	N	Y
26-X-96-2-005	Y	Y	N	Y	70ET	N	Y	N	Y
26-X-96-3-301	N	N	N	N		N	N	Y	N
26-X-96-3-302	N	N	N	N		N	N	Y	N
26-X-96-4-301	N	N	MKB	N	70ET	N	N	N	N
26-X-96-4-302	N	N	MKB	N	70ET	N	N	N	N
26-X-96-5-001	Y	Y	H, BA	Y	70ET	N	Y	N	Y
26-X-96-5-002	Y	Y	H, BA	Y	70ET	Y	Y	Y	Y
26-X-96-5-003	Y	Y	H, BA	Y	70ET	N	Y	N	Y
26-X-96-5-004	Y	Y	N	Y	70ET	Y	Y	Y	Y
26-X-96-5-005	N	Y	H	Y	70ET	N	N	N	Y
26-X-96-6-001	Y	Y	H, BA	Y	70ET	N	N	N	N
26-X-96-6-002	N	Y	H, BA	Y	70ET	N	N	N	N
26-X-96-6-003	N	Y	H	Y	5FO	N	N	N	N
26-X-96-6-301	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-302	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-303	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-304	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-305	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-306	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-307	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-308	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-309	N	N	MKB	N	70ET	N	N	N	N
26-X-96-6-310	N	N	MKB	N	70ET	N	N	N	N
27-X-96-1-001	Y	Y	H, BA	Y	70ET	N	Y	N	Y
27-X-96-1-002	Y	Y	H	Y	70ET	N	Y	N	Y

SAMPLE NUMBER (Date + Site # + Sample #)	IN-SITU 35-MM PHOTO	LAB 35-MM PHOTO	MUSEUM SPECIMEN		PRESER- VATIVE	MICROBIAL ISOLATION		CELL CULTURE	TLC
			(Large)	(Small)		(Agar)	(Freeze)		
27-X-96-1-003	Y	Y	H	Y	70ET	Y	Y	Y	Y
27-X-96-1-004	Y	Y	N	Y	70ET	N	Y	N	Y
27-X-96-1-005	Y	Y	H, BA	Y	70ET	N	Y	N	Y
27-X-96-1-006	Y	Y	N	Y	70ET	N	Y	N	N
27-X-96-1-007	W/ #5	Y	N	Y	70ET	N	N	N	Y
27-X-96-1-301	Y	N	N	N		N	N	N	N
27-X-96-2-001	N	Y	N	Y	70ET	N	N	N	N
27-X-96-2-002	N	Y	H	Y	70ET	N	N	N	N
27-X-96-2-003	Y	Y	H	Y	70ET	N	N	N	N
27-X-96-2-004	N	Y	H	Y	70ET	N	N	N	N
27-X-96-2-005	Y	Y	H	Y	70ET	Y	N	N	N
27-X-96-2-301	N	MKB	MKB	N	70ET	N	N	N	N
27-X-96-2-302	N	MKB	MKB	N		N	N	N	N
27-X-96-3-001	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-002	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-003	N	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-004	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-005	Y	Y	H	Y	70ET	Y	Y	Y	Y
27-X-96-3-006	Y	Y	N	Y	70ET	N	Y	N	Y
27-X-96-3-007	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-008	N	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-009	N	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-010	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-011	?	Y	H	Y	70ET	Y	Y	N	Y
27-X-96-3-012	N	Y	H	Y	70ET	Y	Y	N	Y
27-X-96-3-301	Y	N	MKB	N		N	N	N	N
27-X-96-3-302	Y	N	MKB	N		N	N	N	N
27-X-96-3-303	Y	N	MKB	N		N	N	N	N
27-X-96-3-304	N	N	MKB	N		N	N	N	N
27-X-96-3-305	Y	N	MKB	N		N	N	N	N
28-X-96-1-001	Y	Y	H, BA	Y	70ET	N	Y	N	N
28-X-96-1-002	Y	Y	H, BA	Y	70ET	Y	Y	Y	N
28-X-96-1-003	Y	Y	H, BA	Y	70ET	N	Y	N	N
28-X-96-1-004	Y	Y	H	Y	70ET	N	Y	N	N
28-X-96-1-005	Y	Y	H, BA	Y	70ET	N	Y	N	N
28-X-96-1-006	Y	Y	H	Y	70ET	Y	N	N	N
28-X-96-1-007	Y	Y	N	Y	70ET	N	Y	N	N
28-X-96-1-008	Y	Y	H	Y	70ET	Y	Y	N	N
28-X-96-1-009	Y	Y	H	Y	70ET	N	Y	N	N

SAMPLE NUMBER (Date + Site # + Sample #)	IN-SITU 35-MM PHOTO	LAB 35-MM PHOTO	MUSEUM SPECIMEN		PRESER- VATIVE	MICROBIAL ISOLATION		CELL CULTURE	TLC
			(Large)	(Small)		(Agar)	(Freeze)		
27-X-96-1-003	Y	Y	H	Y	70ET	Y	Y	Y	Y
27-X-96-1-004	Y	Y	N	Y	70ET	N	Y	N	Y
27-X-96-1-005	Y	Y	H, BA	Y	70ET	N	Y	N	Y
27-X-96-1-006	Y	Y	N	Y	70ET	N	Y	N	N
27-X-96-1-007	W/ #5	Y	N	Y	70ET	N	N	N	Y
27-X-96-1-301	Y	N	N	N		N	N	N	N
27-X-96-2-001	N	Y	N	Y	70ET	N	N	N	N
27-X-96-2-002	N	Y	H	Y	70ET	N	N	N	N
27-X-96-2-003	Y	Y	H	Y	70ET	N	N	N	N
27-X-96-2-004	N	Y	H	Y	70ET	N	N	N	N
27-X-96-2-005	Y	Y	H	Y	70ET	Y	N	N	N
27-X-96-2-301	N	MKB	MKB	N	70ET	N	N	N	N
27-X-96-2-302	N	MKB	MKB	N		N	N	N	N
27-X-96-3-001	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-002	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-003	N	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-004	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-005	Y	Y	H	Y	70ET	Y	Y	Y	Y
27-X-96-3-006	Y	Y	N	Y	70ET	N	Y	N	Y
27-X-96-3-007	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-008	N	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-009	N	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-010	Y	Y	H	Y	70ET	N	Y	N	Y
27-X-96-3-011	?	Y	H	Y	70ET	Y	Y	N	Y
27-X-96-3-012	N	Y	H	Y	70ET	Y	Y	N	Y
27-X-96-3-301	Y	N	MKB	N		N	N	N	N
27-X-96-3-302	Y	N	MKB	N		N	N	N	N
27-X-96-3-303	Y	N	MKB	N		N	N	N	N
27-X-96-3-304	N	N	MKB	N		N	N	N	N
27-X-96-3-305	Y	N	MKB	N		N	N	N	N
28-X-96-1-001	Y	Y	H, BA	Y	70ET	N	Y	N	N
28-X-96-1-002	Y	Y	H, BA	Y	70ET	Y	Y	Y	N
28-X-96-1-003	Y	Y	H, BA	Y	70ET	N	Y	N	N
28-X-96-1-004	Y	Y	H	Y	70ET	N	Y	N	N
28-X-96-1-005	Y	Y	H, BA	Y	70ET	N	Y	N	N
28-X-96-1-006	Y	Y	H	Y	70ET	Y	N	N	N
28-X-96-1-007	Y	Y	N	Y	70ET	N	Y	N	N
28-X-96-1-008	Y	Y	H	Y	70ET	Y	Y	N	N
28-X-96-1-009	Y	Y	H	Y	70ET	N	Y	N	N

SAMPLE NUMBER (Date + Site # + Sample #)	IN-SITU 35-MM PHOTO	LAB 35-MM PHOTO	MUSEUM SPECIMEN		PRESER- VATIVE	MICROBIAL ISOLATION		CELL CULTURE	TLC
			(Large)	(Small)		(Agar)	(Freeze)		
28-X-96-1-010	N	Y	H	Y	70ET	N	Y	N	N
28-X-96-1-301	N	N	N	N		N	N	N	N
28-X-96-1-302	N	N	MKB	N		N	N	N	N
28-X-96-1-303	N	N	MKB	N		N	N	N	N
28-X-96-2-001	Y	Y	H, BA	Y	70ET	N	Y	N	Y
28-X-96-2-002	Y	Y	H	Y	70ET	N	Y	N	Y
28-X-96-2-003	Y	Y	H	Y	70ET	N	Y	N	Y
28-X-96-2-004	N	Y	H	Y	70ET	N	Y	Y	Y
28-X-96-2-005	Y	Y	N	Y	70ET	N	Y	N	Y
28-X-96-2-006	N	Y	H	Y	70ET	N	Y	N	Y
28-X-96-3-001	Y	Y	H, BA	Y	70ET	N	N	N	N
28-X-96-3-201	Y	Y	N	N		Y	Y	N	N
28-X-96-4-301	N	Y	N	N		N	N	Y	N
28-X-96-4-N	N	N	N	N		N	N	N	N
29-X-96-1-001	N	Y	H	Y	70ET	Y	N	Y	Y
29-X-96-1-002	N	U	H	Y	70ET	N	Y	Y	Y
29-X-96-1-003	N	Y	H	Y	70ET	Y	Y	N	Y
29-X-96-1-004	N	Y	H	Y	70ET	N	Y	N	Y
29-X-96-1-005	N	Y	H	Y	70ET	N	Y	N	Y
29-X-96-1-006	?	Y	H	Y	70ET	N	Y	N	Y
29-X-96-1-007	N	Y	N	Y	70ET	N	Y	N	Y
29-X-96-3-301	Y	N	N	N		N	N	Y	N
29-X-96-4-001	N	Y	H	Y	70ET	N	Y	N	Y
29-X-96-4-002	Y	Y	H	Y	70ET	N	Y	N	Y
29-X-96-4-003	N	Y	Y	Y	70ET	N	Y	N	Y
29-X-96-4-004	N	Y	H	Y	70ET	N	Y	N	Y
29-X-96-4-005	?	Y	H	Y	70ET	Y	Y	N	Y
29-X-96-4-006	N	Y	H, MK	Y	70ET	Y	Y	N	N
29-X-96-4-007	Y	Y	H	Y	70ET	N	Y	N	Y
29-X-96-4-008	N	Y	H	Y	70ET	N	Y	N	Y
30-X-96-1-001	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-1-002	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-1-003	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-1-004	Y	Y	H	Y	70ET	N	Y	N	Y
30-X-96-1-005	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-1-006	Y	Y	H, BA	Y	70ET	N	Y	Y	Y
30-X-96-1-007	Y	Y	H, MK	Y	70ET	N	Y	N	Y
30-X-96-1-008	N	Y	H?	Y	70ET	N	Y	N	Y
30-X-96-1-009	W/#?	Y	N	Y	70ET	N	Y	N	Y

SAMPLE NUMBER (Date + Site # + Sample #)	IN-SITU 35-MM PHOTO	LAB 35-MM PHOTO	MUSEUM SPECIMEN		PRESER- VATIVE	MICROBIAL ISOLATION		CELL CULTURE	TLC
			(Large)	(Small)		(Agar)	(Freeze)		
30-X-96-1-010	N	Y	H	Y	70ET	N	Y	N	Y
30-X-96-2-001	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-002	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-003	N	Y	H	Y	70ET	N	Y	N	Y
30-X-96-2-004	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-005	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-006	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-007	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-008	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-009	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-010	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-2-011	Y	Y	H, BA	Y	70ET	N	Y	N	Y
30-X-96-3-001	N	Y	H	Y	70ET	N	Y	N	N
31-X-96-1-201	N	N	N	N		N	N	N	N

APPENDIX 10

Checklist of Specimens Subsampled for DNA Analyses

Appendix 10. VOUCHER, DNA AND SLIDES FROM BAHAMAS OCTOBER 1996 COLLECTION.

SPECIES	HBO#	DNA	VOUCHER	SLIDE
<i>Discodermia</i> sp IV	26-X-96-1-002	5 ml GNCL sample	S	-
<i>Halichondria</i> ?corrugata	26-X-96-1-301	5 ml GNCL sample	L	-
<i>Stellatinnopsis</i> / <i>Jaspis</i> sp	26-X-96-2-002	5 ml GNCL sample	S	X
<i>Stellatidae</i> genus 1	26-X-96-2-004	5 ml GNCL sample	S	X
<i>Axinellidae</i> genus 1	26-X-96-2-005	5 ml GNCL sample	S	X
<i>Aplysina</i> "globularis"	26-X-96-2-301	5 ml GNCL sample	L	-
? <i>Pachastrissa</i> sp	26-X-96-2-302	5 ml GNCL sample	S	-
<i>Erylus</i> ? <i>trispshaera</i>	26-X-96-4-301	5 ml GNCL sample	L	-
<i>Pandorus</i> acanthofolium	26-X-96-4-302	5 ml GNCL sample	L	-
<i>Discodermia</i> sp IV	26-X-96-5-001	50 ml GNCL sample	S	X
<i>Spongosorites</i> sp 1 (bright yellow)	26-X-96-5-002	5 ml GNCL sample	S	X
<i>Discodermia</i> sp 1	26-X-96-5-003	50ml GNCL sample	S	X
? <i>Hymeniacidon</i> sp	26-X-96-6-001	5 ml GNCL sample	S	-
Raspailiid genus 1	26-X-96-6-002	-	S	-
<i>Ptilocaulis</i> sp	26-X-96-6-301	5 ml GNCL sample	S	-
<i>Teichixinella</i> murchella	26-X-96-6-302	5 ml GNCL sample	S	-
<i>Amphimedon</i> compressa	26-X-96-6-303	5 ml GNCL sample	S	-
<i>Pseudaxinella</i> lunaecharta	26-X-96-6-304	5 ml GNCL sample	S	-
<i>Myrmekioderma</i> sp	26-X-96-6-305	5 ml GNCL sample	S	-
<i>Ectyoplasia</i> ferox	26-X-96-6-306	5 ml GNCL sample	S	-
<i>Plakortis</i> sp 1	26-X-96-6-307	5 ml GNCL sample	S	-
<i>Pseudoceratina</i> crassa	26-X-96-6-308	5 ml GNCL sample	S	-
<i>Callyspongia</i> vaginalis	26-X-96-6-309	5 ml GNCL sample	S	-
? <i>Amphimedon</i> compressa	26-X-96-6-310	5 ml GNCL sample	S	-
<i>Discodermia</i> sp IV	27-X-96-1-001	5 ml GNCL sample	S	-
<i>Discodermia</i> sp I	27-X-96-1-002	5 ml GNCL sample	S	-
<i>Discodermia</i> sp IV	27-X-96-1-005	50 ml GNCL sample	S	-
<i>Erylus</i> ? <i>goffrilleri</i>	27-X-96-1-006	5 ml GNCL sample	S	-
<i>Discodermia</i> sp IV	27-X-96-1-003	10ml GNCL dissoci. cells for genetics	S	X
<i>Discodermia</i> sp IV	27-X-96-1-007	5 ml GNCL sample	S	-
<i>Halisarca</i> sp	27-X-96-2-001	5 ml GNCL sample	S	-
? <i>Iotrochota</i> sp	27-X-96-2-002	5 ml GNCL sample	S	-

Axinellidae genus 2	27-X-96-2-003	5 ml GNCL sample
Siphonodictyon mucosum	27-X-96-2-004	5 ml GNCL sample
Topsentia sp	27-X-96-2-301	5 ml GNCL sample
Niphatidae genus 1	27-X-96-2-302	5 ml GNCL sample
Cribochalina vasculum	27-X-96-3-301	5 ml GNCL sample
Cinachyrella sp 1	27-X-96-3-302	5 ml GNCL sample
Phakellia folium	27-X-96-3-303	5 ml GNCL sample
Calcifibrospongia sp	27-X-96-3-304	5 ml GNCL sample
Leucetta sp	27-X-96-3-305	5 ml GNCL sample
Plakortis sp 2	27-X-96-3-306	5 ml GNCL sample
Asteropus sp 1	28-X-96-1-001	5 ml GNCL sample
Hippospongia sp	28-X-96-1-005	5 ml GNCL sample
Discoderma cf sp IV	28-X-96-1-006	5 ml GNCL sample
Stilletta sp	28-X-96-1-007	5 ml GNCL sample
Discoderma cf sp 1	28-X-96-1-008	5 ml GNCL sample
Corticium sp 1	28-X-96-1-009	5 ml GNCL sample
Spongisorites sp 2 (dull yellow)	28-X-96-1-010	5 ml GNCL sample
Xestospongia sp (cavernous)	28-X-96-1-301	5 ml GNCL sample
Corticium sp 2	28-X-96-1-302	5 ml GNCL sample
Agelas sp	28-X-96-1-303	5 ml GNCL sample
Corallistes microstyliifer (Bahamas)	28-X-96-2-001	5 ml GNCL sample
Geodia sp 1	28-X-96-2-002	5 ml GNCL sample
Geodia sp 2	28-X-96-2-003	5 ml GNCL sample
Spongisorites sp 3 (orange)	28-X-96-2-004	5 ml GNCL sample
Petrosia sp (deepwater ear)	28-X-96-2-005	5 ml GNCL sample
Condylilla nucula	28-X-96-2-301	5 ml GNCL sample
Biemna sp	28-X-96-3-001	5 ml GNCL sample
Petrosia sp (deepwater tough ball)	29-X-96-1-301	5 ml GNCL sample
Axinyssa ?ambrosia de Laubenfels	29-X-96-1-302	5 ml GNCL sample
Halichondrida sp	29-X-96-2-301	5 ml GNCL sample
Neofibularia nolitangere	29-X-96-2-302	5 ml GNCL sample
Oceanapia barschii	29-X-96-2-303	5 ml GNCL sample
Petrosia sp (shallow grey encruster)	29-X-96-2-304	5 ml GNCL sample
?Aplysinopsis sp	29-X-96-2-305	5 ml GNCL sample
Spheciopspongia vesparium	29-X-96-2-306	5 ml GNCL sample
Spirastrella coccinea	29-X-96-2-307	5 ml GNCL sample
Xestospongia muta	29-X-96-2-308	5 ml GNCL sample
Amphimedon viridis	29-X-96-3-301	5 ml GNCL sample
Ircinia cf felix	29-X-96-3-302	5 ml GNCL sample

Dysidea etheria	S
Leucetta sp 2	S
Discodermia verrucosa	L
Asteropus sp 1	S
?Spongisorites sp (yellow, gooey)	S
Aciculites or Gastrophanella	L
Axinelidae genus 3	S
Carminus sp 1 (lithoherm)	S
Geodia sp 3 (lithoherm)	S
Vetulina n. sp (spaceship)	S
?Topsentia sp (lithoherm)	S
Plakastrella (Dercitopsis) sp	S
Epipolais sp	S
Topsentia pectata (lithoherm)	S
?Oceanapia sp (prop-legs)	S
Strongylophora sp (bagel)	S
Racodiscula asteroides	L
Corallistes tubulatus (lithoherm)	L
Cinachyrella sp 2	L
29-X-96-3-303	5 ml GNCL sample
29-X-96-5-301	5 ml GNCL sample
29-X-96-4-006	5 ml GNCL sample
30-X-96-1-003	5 ml GNCL sample
30-X-96-1-004	5 ml GNCL sample
30-X-96-1-007	5 ml GNCL sample
30-X-96-1-010	5 ml GNCL sample
30-X-96-2-001	5 ml GNCL sample
30-X-96-2-002	5 ml GNCL sample
30-X-96-2-003	5 ml GNCL sample
30-X-96-2-004	5 ml GNCL sample
30-X-96-2-005	5 ml GNCL sample
30-X-96-2-006	5 ml GNCL sample
30-X-96-2-007	5 ml GNCL sample
30-X-96-2-008	5 ml GNCL sample
30-X-96-2-010	5 ml GNCL sample
30-X-96-2-301	5 ml GNCL sample
30-X-96-2-302	5 ml GNCL sample
30-X-96-3-301	5 ml GNCL sample