The Underwater Catalog:
A Guide to Methods in Underwater Research

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1. Underwater Record Keeping: Computer; page 9
    a. WetPC: miniature PC with mask-mounted virtual display and novel, one-handed controller (Kord-pad)
    b. Mounted in waterproof housing on air tank
    c. www.aims.gov.au/pages/wetpc/wetpc.html

2. Underwater epoxy: Kop-Coat: page 13
    a. As no one bothers to answer e-mails sent to marine@kop-coat.com, send queries directly to West Marine at: cattech@westmarine.com
    b. West Marine usually has Kop-Coat in most of their branches...if one branch doesn't have it, they can locate a branch that does. Specific ordering info:
        i. Two-quart size (2 L): 201087; ca. $65
        ii. Two-gallon size (4 L): 230060; ca. $175
    c. West Marine will not ship Kop-Coat to a non-US address because it is "toxic". May or may not be allowed to transport in checked baggage

3. Attaching Gear to Hard Substrata: Drilling holes, page 18: ADD new section:
    Drop-In Anchors (for concrete, hard stone, and solid block)
    a. Insert anchor into predrilled hole (1_ inch) and tap flush with the surface
    b. Drive the setting tool (sold separately from anchors) into the anchor
    c. Anchor has terminal metal plug; when plug is set with setting tool, it expands the anchor sides in four directions to push against wall of hole
    d. Insert bolt or threaded rod into anchor (anchor is threaded)
    e. For a truly permanent anchor, insert epoxy into hole before inserting anchor
    f. A _ inch diameter hole requires an anchor with a _ inch outer diameter
    g. McMaster-Carr Supply Co. (http://www.mcmaster.com)
        i. Drop-In anchors (_ inch): 97083A310; ca. $0.32 each
        ii. Setting Tool (_ inch): 97077A120; ca. $3.25

4. Experimental Design: References; page 28
    a. Fletcher, D.J., and A.J. Underwood. 2002. How to cope with negative estimates of components of variance in ecological field studies. J. Exp. Mar. Biol. Ecol. 272:89-95.

5. Quantitative Sampling: General References; page 32
    a. Miller, A.W. and R.F. Ambrose. 2000. Sampling patchy distributions: comparison of sampling designs in rocky intertidal habitats. Mar. Ecol. Prog. Ser. 196:1-14. (cover data generated using high-density point-contact method: this area was sampled using different sampling units (single point, line transects, quadrats) distributed randomly or in a stratified design; results underscored difficulty of obtaining accurate cover estimates in rocky intertidal communities)
    b. Rumohr, H., I. Karakassis, and J.N. Jensen. 2001. Estimating species richness, abundance and diversity with 70 macrobenthic replicates in the Western Baltic Sea. Mar. Ecol. Prog. Ser. 214:103-110. (replicated [n=70] data set used to predict species richness and to assess accuracy of common estimates of abundance and diversity: >10 and >53 replicates required to include 66% and 95%, respectively, of species in 70 replicates; estimates of average abundance and Shannon-Wiener diversity index

using 5, 10, and 70 replicates resulted in < 40%, 30%, and 5% error, respectively; jackknife and S_4 predictors of species-richness were rather limited)

6. Census Methods for Fishes:
    a. General Comments; page 42
        i. Harvey, E., D. Fletcher, and M. Shortis. 2001. A comparison of the precision and accuracy of estimates of reef-fish lengths determined visually by divers with estimates produced by a stereo-video system. Fish. Bull. (US). 99:63-71
        ii. Harvey, E., D. Fletcher, and M. Shortis. 2001. Improving the statistical power of length estimates of reef fish: a comparison of estimates determined visually by divers with estimates produced by a stereo-video system. Fish. Bull. (US). 99:72-80.
    b. Visual Census: Stationary Observer: Baited traps (new category); page 46
        i. Stewart, B.D., and J.S. Beukers. Baited technique improves censuses of cryptic fish in complex habitats. Mar. Biol. Prog. Ser. 197:259-272. (compared baited technique with belt transect and tagging study; baited technique produced significantly higher density estimates for 3 of 4 most abundant cryptic species, but overestimated abundance of mobile species)
    c. General References; page 46
        i. Thompson, A.A., and B.D. Mapstone. 2002. Intra- versus inter-annual variation in counts of reef fishes and interpretations of long-term monitoring studies. Mar. Ecol. Prog. Ser. 232:247-257. (estimate temporal variation in abundances of tropical reef fishes attributable to sampling error at diurnal, daily, and monthly scales; also compares these estimate to inter-annual variation from processes such as mortality and recruitment)

7. Quantitative Photography/Video: Quantitative Photography/Video:
    a. General Comments; page 49
        i. Freeware/shareware programs for performing measurements on digital images
            1. NIH-software Scion Image
                a. http://rsb.info.nih.gov/nih-image (website)
                b. http://rsb.info.nih.gov/ij/ (Mac, Win, Java)
            2. TNTlite
                a. www.microimages/com
                b. Versions for nearly all operating systems and free phone tech support
                c. Drawbacks:
                    i. Complex, will only use a small subset of tools
                    ii. Free version limits image size to 614 x 512 pixels
            3. Plugins for Photoshop: www.andromeda.com/info/measure.html
    b. Additional References; page 50
        i. Kollman, H., and M. Stachowitsch. 2001. Long-term changes in the benthos of the Northern Adriatic Sea: a phototransect approach. Mar. Ecol. 22:135-141. (annual 1 m^2 photoquads from 1984-1994)
    c. Behavior; page 52
        i. Heithaus, M.R., G.J. Marshall, B.M. Buhleier, and L.M. Dill. 2001. Employing Crittercam to study habitat use and behavior of large sharks. Mar. Ecol. Prog. Ser. 209:307-310. (attached video camera with integrated time-depth recorder to back of tiger sharks)

8. Methods for Collecting Organisms:
    a. Airlifts/Suction Devices: page 57
        i. Orth, R.J., and J. van Montirans. 1987. Utilization of a seagrass meadow and tidal marsh creak by blue crabs *Callinectes sapidus*. I. Seasonal and annual variations in abundance with emphasis on post-settlement juveniles. Mar. Ecol. Prog. Ser. 41:283-294.
        ii. Used a drop net and suction sampler, with pump and motor in an overhead boat

iii. 88% efficient and estimated densities were an order of magnitude greater than that obtained with conventional trawl gear

b. Fishes:
   i. Chemicals: Toxicants; page 63
      1. Ackerman, J.L. and D.R. Bellwood. 2000. Reef fish assemblages: a re-evaluation using enclosed rotenone stations. Mar. Ecol. Prog. Ser. 206:227-237. (small, enclosed, intensive rotenone stations provide reliable quantitative samples of small taxa)
   ii. Nets, new section; page 66
      1. BINCKE net
         a. Diver-held, closing hoop net
         b. Used to collect small fish and/or to retrieve SMURFs (see below)
         c. 3/16 inch mesh and dyed red (less perception by fish?) can collect 10 mm TL sailfish
         d. For details on construction, go to: http://www.biology.ucsc.edu/people/carr/, then click on People/Graduate Students/Former Students/Arnold Amman/Using SMURFs: A Primer
   iii. Traps:
      1. General Comments; page 66
         a. Robichaud, D., W. Hunte, and M.R. Chapman. 2000. Factors affecting the catchability of reef fishes in Antilean fish traps. Bull. Mar. Sci. 67:831-844. (more mobile species more likely to encounter and enter traps; traps are more attractive to fish in areas of low habitat complexity)
      2. Light Traps; page 68
         a. Meekan, M.G., P.J. Doherty, and L. White Jr. 2000. Recapture experiments show the low sampling efficiency of light traps. Bull. Mar. Sci. 67:875-885. (concluded that it is fruitless to convert light trap catches into densities, but light traps may provide useful measures of relative larval supply)
         b. Meekan, M.G., Wilson, S.G., Halford, A. and A. Retzel. 2001. A comparison of catches of fishes and invertebrates by two light trap designs, in tropical NW Australia. Mar. Biol. 139: 373-381.(compared large and small, drifting vs moored light traps; variation in efficiency of design appears to be small when compared to patchiness of zooplankton and fish assemblages)
         c. Fisher, R., and D.R. Bellwood. 2002. A light trap design for stratum-specific sampling of reef fish larvae. J. Exp. Mar. Biol. Ecol. 269: 27-37.(light traps radiated narrow beams of light so as to sample discreet depth strata)
      3. SMURFS (new section); page 68
         a. Standard Monitoring Units for Recruitment of Fishes
         b. A 1.3m long 0.3m dia meshed device designed to collect settling juvenile fishes up to 50 mm TL
         c. Rapidly detached and retrieved from spar buoys
         d. For detailed description of construction, deployment, and costs, go to: http://www.biology.ucsc.edu/people/carr/, then click on People/Graduate Students/Former Students/Arnold Amman/Using SMURFs: A Primer

9. Tagging Methods:
   a. Algae, Types of Tags, Mass tagging of propagules; page 73
      i. Reproductive thalli
         1. Stain with 0.05% Toluidine Blue for 1-2 hrs
         2. Return thalli to field; after 24 hrs sample surrounding area with small airlift for stained propagules

3. Kendrick, G.A. and D.I. Walker. 1991. Dispersal distances for propagules of *Sargassum spinuligerum* (Sargassaceae, Phaeophyta) measured directly by vital staining and venturi suction sampling. Mar. Ecol. Prog. Ser. 79:133-138

ii. Fertilized gametes
1. Tag with calcofluor white (Sigma, St. Louis, MO, USA), a fluorescent stain that binds to cell walls
2. Unfertilized eggs, which do not have cell walls, are not stained
3. Suspensions of sperm and eggs were prepared and filtered through 200 µL mesh to remove debris
4. Staining occurred for ca. 1 hr in a final calcofluor solution of 0.13 $g \cdot L^{-1}$
5. Gametes were released on a rising tide and detected by sampling the water column and assaying caged ceramic tiles attached to the shore
6. Dudgeon, S., J.E. Kübler, W.A. Wright, R.L. Vadas, and P.S. Petraitis. 2001. Natural variability in zygote dispersal of *Ascophyllum nodosum* at small spatial scales. Funct. Ecol. 15:595-604.

b. Shelled Molluscs:
i. Bee Tags; page 76
1. The Bee Works, 5 Edith Dr., RR #2, Orillia, ON, Canada, L3V 6H2; (705) 326-7171, (705) 325 3461 (fax); email: admin@beeworks.com; sales, sales@beeworks.com; www.beeworks.com

ii. Mass tagging of Cohorts; page 77
1. Moran, A.L. 2000. Calcein as a marker in experimental studies of newly-hatched gastropods. Mar. Biol. 137:893-898

iii. Internal Tags (new category)
1. Glazer, R.A., K.J. McCarthy, R.L. Jones, and L. Anderson. 1997. The use of underwater metal detectors to locate outplants of the mobile marine gastropod, *Strombus gigas* L. Proc., Gulf. Carib. Fish. Inst. 49. (used external aluminum tags)

c. Sea Urchins:
i. Test penetration; page 77
1. Hagen, N.T. 1996. Tagging sea urchins: a new technique for individual identification. Aquaculture 139:271-284 (reviews external tags and tagging methods)
2. Duggan, R.E., and R.J. Miller. 2001. External and internal tags for the green sea urchin. J. Exp. Mar. Biol. Ecol. 258:115-122 (used anchor tags, metal and nylon screws; anchor tags caused 50% mortality in 1 month, nylon screws show no affect on behavior with 55-75% retention after 3 months)

ii. Internal; page 78
1. Hagen, N.T. 1996. Tagging sea urchins: a new technique for individual identification. Aquaculture 139:271-284 (implanted PIT tags, which were activated and recorded by an electronic reader; See: “Fishes: Internal Tags: Types: PIT tags; page 87)
2. Duggan, R.E., and R.J. Miller. 2001. External and internal tags for the green sea urchin. J. Exp. Mar. Biol. Ecol. 258:115-122 (used aluminum welding rod and poultry tags inserted into coelom, detected by an underwater metal detector; all tags were retained for the 4 month study)

d. Crustaceans; page 80
i. Kneib, R.R., and M.C. Huggler. 2001. Tag placement, mark retention, survival and growth of juvenile white shrimp (*Litopenaeus setiferus*) Pérez Farfante, 1969) injected with coded wire tags. J. Exp. Mar. Biol. Ecol. 266: 109-120. Best retention (95-100%) was achieved in abdominal musculature, which also yielded the greatest survival rates (81-100%)

e. Fishes: Telemetry; page 92

i. Klimley, A.P., B.J. LeBoeuf, K.M Cantara, J.E. Richert, S.F. Davis, and S. Van Sommeran. 2000. Radio-acoustic positioning as a tool for studying site-specific behavior of the white shark and other large marine species. Mar. Biol. 138:429-446

10. Methods to Measure Growth:
    a. Algae: Staining techniques:
        i. Calcofluor White M2R; page 93
            1. Cole, K. 1964. Induced fluorescence in gametophytes of some Laminariales. Can J. Bot. 42:1173-1183.
            2. Hsiao, S.I.C., and L.D. Druehl. 1973. Environmental control of gametogenesis in *Laminaria saccharina*. IV. *In situ* development of gametophytes and young sporophytes. J. Phycol. 9:160-164.
            3. Nakazawa, S.K., K. Takamura, and M. Abe. 1969. Rhizooid differentiation in *Fucus* eggs labeled with Calcofluor White and birefringence of cell wall. Bot. Mag. Toko 82:41-44
            4. Serrão, E.A., L. Kautsky, and S.H. Brawley. 1996. Distributional success of the marine seaweed *Fucus vesiculosus* L. in the brackish Baltic Sea correlates with osmotic capabilities of Baltic gametes. Oecologia 107:1-12.
        ii. Alizarin Red ; page 94
            1. Multer H.G. 1988. Growth rate, ultrastructure and sediment contribution of *Halimeda incrassata* and *Halimeda monile*, Nonsuch and Falmouth Bays, Antigua, WI. Coral Reefs 6:179-186
            2. Payri C.E. 1988. *Halimeda* contribution to organic and inorganic production in a Tahitian reef system. Coral Reefs 6:251-262.
        iii. FungiFluor (new category); page 94
            1. Deployed glass sides in field on which kelp zoospores settled
            2. Immersed slides in 20% solution of FungiFluor (0.01% Cellufluor) for 24 hrs (non-lethal biostain binding non-specifically to beta-linked polysaccharides and fluoresces at 400-440 nm)
            3. After staining, slides transferred to individual culture dishes for growth
            4. Edwards, M.S. 1999. Using *in situ* substratum sterilization and fluorescence microscopy in studies of microscopic stages of marine macroalgae. Hydrobiologia 398/399:253-259.
    b. Corals: Alizarin Red; page 94
        i. Dodge R.E. 1984 Coral calcification rates by the buoyant weight technique: effects of Alizarin staining. J. Exp. Mar. Biol. Ecol. 75:217-232
    c. Molluscs, Brachiopods, Sponges; page 95
        i. Moran, A.L. 2000. Calcein as a marker in experimental studies of newly-hatched gastropods. Mar. Biol. 137:893-898
    d. Fishes; page 96
        i. Benîot H.P. and P. Pepin. 1999. Individual variability in growth rate and the timing of metamorphosis in yellowtail flounder *Pleuronectes ferrugineus*. Marine Ecology Progress Series 184:231-244.

11. Manipulation of Substrata; page 104
    a. Sterilization of Substrata:
        i. Edwards, M.S. 1999. Using *in situ* substratum sterilization and fluorescence microscopy in studies of microscopic stages of marine macroalgae. Hydrobiologia 398/399:253-259. (bleach was injected into subtidal, waterproof tents to remove all microscopic algal spores)
    b. Settling Plates; Other designs for settling plates:
        i. Plastic “biospheres”, scrubber pads, nylon bristle brushes, etc.: see “Collecting Invertebrate Larvae-Settlement Collectors”, page 60.
        ii. Artificial substrata made of scrubbers tend to under-sample amphipods and be dominated by suspension-feeding polychaetes

1. Smith, S.D.A., and M.J.Rule. 2002. Artificial substrata in a shallow sublittoral habitat: do they adequately represent natural habitats or the local species pool? J. Exp. Mar. Biol. Ecol. 277: 25-41.

12. Manipulation of Organisms:
    a. Attachment of Sessile Organisms; page 105
        i. Rinkevich, B. 2000. Steps towards the evaluation of coral reef restoration by using small branch fragments. Mar. Biol. 136:807-812. (restoration protocols may be applied either by sacrificing whole large colonies via pruning high numbers of small fragments or, by pruning only a few small branches from each one of many genets)
    b. Tethering/references; page 105
        i. Aronson, R.B., K.L. Heck Jr, and J.F. Valentine. 2001. Measuring predation with tethering experiments. Mar. Ecol. Prog. Ser. 214:311-312. (critiques Kneib and Scheele, 2000)
        ii. Ball, B., A. Linnane, B. Munday, R. Browne, and J.P. Mercer. 2001. The effect of cover on *in situ* predation in early benthic phase European lobster *Homarus gammarus*. J. Mar. Biol. Ass. U.K. 81:639-642.
        iii. Kneib, R.T., and C.E.H. Scheele. 2000. Does tethering of mobile prey measure relative predation potential: An empirical test using mummichogs and grass shrimp. Mar. Ecol. Prog. Ser. 198:181-190.
        iv. Smith, L.D. 1995. Effects of limb automony and tethering on juvenile blue crab survival from cannibalism. Mar. Ecol. Prog. Ser. 116:65-74.
        v. Wahle, R.A., and R.S. Steneck. 1992. Habitat restrictions in early benthic life: experiments on habitat selection *in situ* predation with the American lobster. J. Exp. Mar. Biol. Ecol. 157: 91-114.
    c. Artificial Substrates: Mimicking Biological Species: Artificial Seagrass; page 109
        i. Lee, S.Y., C.W. Fong, and R.S.S. Wu. 2001. The effects of seagrass (*Zostera japonica*) canopy structure on associated fauna: a study using artificial seagrass units and sampling of natural beds. J. Exp. Mar. Biol. Ecol. 259:23-50.

13. Manipulation of Abiotic Factors in the Field: Nutrients; page 111
    a. References:
        i. Miller, M.W., M.E. Hay, S.L Miller, D. Malone, E. Sotka, and A. Szmant. 1999. Effects of nutrients versus herbivores on reef algae: a new method for manipulation nutrients on coral reefs. Limnol. Oceanogr. 44:1847-1861. (fertilizer spikes sealed in cinder blocks; nutrients diffused through a hole drilled in block and around/through a slab of coral rock)
        ii. Worm, B., T.B.H. Reusch, and H.K. Lotze. 2000. *In situ* nutrient enrichment: methods for marine benthic ecology. Internat. Rev. Hydrobiol. 85: 359-375. (reviewed 18 published enrichment methods, recommend coated fertilizer pellets because they provide gradual nutrient release, realistic nutrient gradients and even application)

14. Submersible Pumps
    a. References:
        i. Mundy, C., R. Babcock, I. Ashworth, and J. Small. 1994. A portable, discrete-sampling submersible plankton pup and its use in sampling starfish eggs. Biol. Bull. 186:168-171.

15. Special Applications: Diving in Polluted Water; page 122
    a. Barsky, S.M. Diving in High-Risk Environments, 3rd Ed. Hammerhead Press (info@hammerheadpress.com)