NOAA Technical Memorandum NMFS-SEFSC-543

DEMOGRAPHIC MONITORING PROTOCOLS FOR THREATENED CARIBBEAN ACROPORA SPP. CORALS

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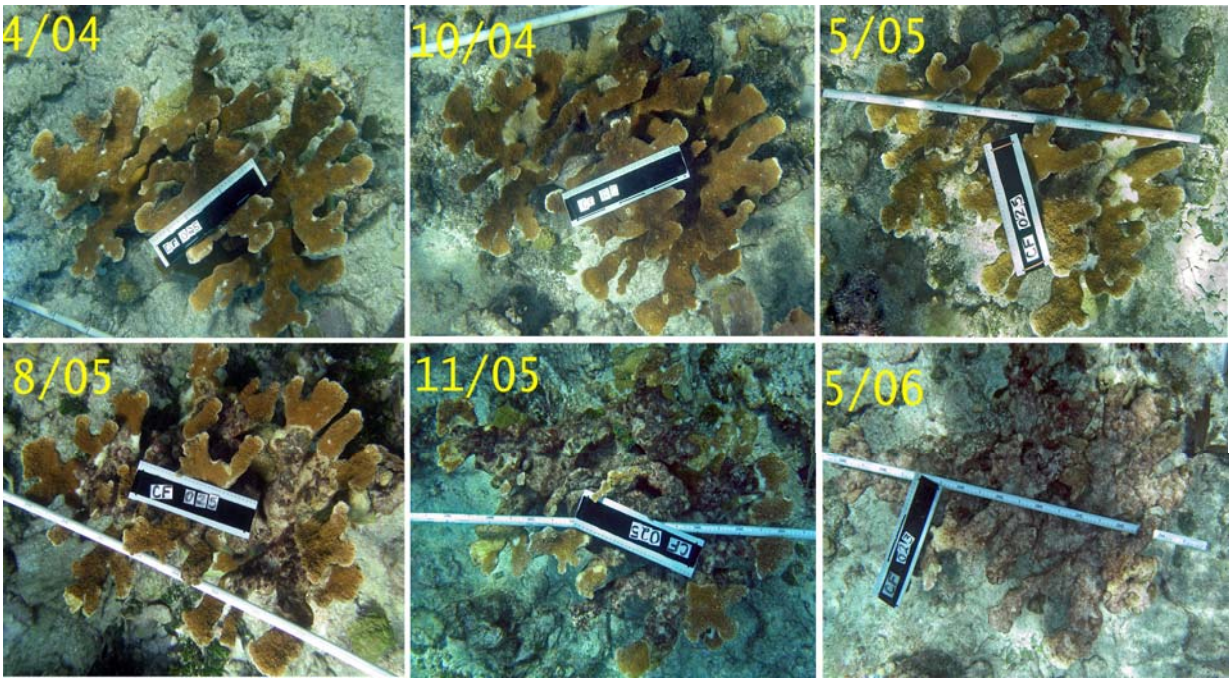

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National Marine Fisheries Service
Southeast Fisheries Science Center
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INTRODUCTION

PURPOSE

This manual is intended to serve as a basic but evolving platform to involve many partners in collecting comparable data sets focused on Atlantic *Acropora* spp. from a variety of locations throughout the Caribbean. This data will be utilized in the ongoing efforts and responsibilities of NOAA-Fisheries to assess species' status and promote recovery under the auspices of the US Endangered Species Act. The protocols described in this manual have been developed over a period of five years of focal monitoring of primarily *A. palmata* in the upper Florida Keys by our team. We have also applied these techniques in Tortola, BVI and Curacao. Obviously, every population and site is different and may require adaptation to the specific monitoring methods.

It should be noted that these techniques have been developed and applied primarily to *Acropora palmata* and the remainder of the document is phrased in terms of this species. However, similar types of data are needed for *Acropora cervicornis* and similar approaches should be appropriate, at least in theory. In our experience in the upper Florida Keys, *A. cervicornis* colonies are much more ephemeral (i.e. they suffer a high rate of complete mortality and complete colony dislocation) and occur at much lower density which makes following individual colonies in statistically robust numbers impractical. We hope to encourage partners to apply the protocols described herein to *A. cervicornis* (with appropriate adaptation) in their regions to begin to obtain similar data sets for this species.

ECOLOGICAL SIGNIFICANCE AND STATUS OF *ACROPORA PALMATA*

Acropora palmata is one of the dominant structural framework builders on Caribbean coral reefs (Goreau 1959; Shinn 1989). Its branched morphology and tendency to form dense, thicket-like, mono-specific stands naturally provide shelter to a variety of fish (Lirman 1999) and other reef organisms including the long-spined urchin, Diadema antillarum (Weil et al. 1984). Rapid skeletal growth and frequent fragmentation allowed it to dominate the reef crest throughout the Caribbean (Goreau 1959) in the past. The reef spurs found on high relief coral reefs in the Florida Keys and throughout the Caribbean were formed in large part by *A. palmata* (Shinn 1989). Because of its ability to create protective habitat, to foster reef

biodiversity, and to maintain the structural integrity of coral reefs, acroporid corals are considered ecologically irreplaceable(Bruckner 2002).

Since the 1970s acroporid species in the Caribbean have experienced extreme and accelerating declines estimated at 90 to 98 % throughout its range (Bruckner 2002). Much of the decline has been attributed to widespread disease impacts (Gladfelter 1982; Aronson, Precht 2001), but the overall loss is thought to have been exacerbated by physical breakage, more frequent and intense bleaching events as well as emerging diseases and anthropogenic effects (Porter et al. 2001).

A multitude of stressors potentially drive *A. palmata* decline including toxic substances, sedimentation and eutrophication, overfishing and related trophic shifts, hurricanes and climate change, disease, and possible synergistic effects. Presently, the ultimate cause(s) of acroporid decline and general reef demise are difficult to parse among these various sources because they usually occur together. Monitoring studies cannot provide robust linkages between these causes and their specific effects. Instead, more tractable proximal causes of decline, including quantifiable colony conditions such as disease and bleaching, fish and snail predation, sponge overgrowth and physical damage related to storms or direct human interaction will be investigated, as a crucial first step in identifying the relative contributions of specific stressors as ultimate causes of decline.

Given the extent and ongoing nature of *Acropora* spp. decline, NOAA-Fisheries responded to a petition by the Center for Biological Diversity and the evaluation of an expert Biological Review Team (Acropora Biological Review Team 2005) by listing both *Acropora palmata* and *A. cervicornis* as threatened under the US Endangered Species Act in May 2006 (NMFS 2006). The implications of this listing are still being worked out, but it invokes specific responsibilities for NOAA-Fisheries to continue to assess the status and orchestrate the recovery of these species throughout their geographic range.

PROJECT GOALS AND OBJECTIVES

There are many studies presently in place to assess the general status and trends of Caribbean coral reefs. *Acropora palmata* is often poorly represented in these studies since its natural distribution is along the reef crest while many studies focus survey efforts on fore reef areas. Furthermore, these studies typically survey randomly placed transects which are not well suited to *A. palmata*'s presently sparse and highly patchy distribution. As a result, very small numbers of *Acropora palmata* colonies end up in the being counted in general reef monitoring studies. While this accurately depicts the present densities of *Acropora*, it yields very little information on the condition and fate of these remaining colonies. Additionally, the lack of

consistency of survey methods greatly hinders the ability to compare the results of surveys between studies and, hence, regions.

The demographic (i.e. colony-based) monitoring approach described here aims to determine the relative importance of the many sources of mortality for populations of *A. palmata*. This method relies on tracking the performance of individual colonies (Fig. 1) to document the threats they face and their fate over time. By tracking the performance of randomly selected 'individual' colonies, the prevalence of a particular threat combined with the subsequent fate or lethality of affected colonies will show the ecological relevance of the various threats. For example, parrotfish bites may be extremely common among an acroporid population, but if effects on a colony are minor, they may be relatively 'unimportant' to the viability of the population. Management and conservation resources can be more effectively applied based on an understanding of the relative impacts of threats on populations. The tracking of individual colonies results in estimates of threat prevalence, threat impact (i.e. mortality), and change in colony size (growth and fragmentation).

A second aspect of this suggested monitoring approach aims to estimate recruitment, the establishment of new colonies in a population to (hopefully) offset the mortality from the treats observed. This is extremely difficult to do given the dynamic nature of fragments and the small size and likely cryptic habit of larval settlers. The best approach we have managed to come up with involves periodic careful, fine scale observations (i.e. mapping) of specific plots in order to detect the appearances of new colonies, including fragments that are attached and growing and small crusts which may be larval recruits.

The data generated by these survey techniques aim to address the following specific objectives:

1) Assess *A. palmata* density and condition in as many areas as possible throughout its geographic range.
2) Monitor sites for changes in abundance (recruitment or death), condition, and colony size.
3) Evaluate the relative ecological relevance and importance of threats to *A. palmata* including disease, predation, overgrowth, and physical breakage.

ANTICIPATED PRODUCTS

Survey data will yield basic population data including colony size, percent live tissue cover, recruitment, disease prevalence, predator infestation, sponge

overgrowth etc. Since site and plot selection may not be random (i.e., locations known to have *Acropora* spp. colonies), the described monitoring protocol is not intended to yield regional estimates of population abundance. However, changes in abundance due to recruitment and mortality can be rigorously evaluated. The random selection of colonies within the plot will yield information on the prevalence and variability of various threats among the general population. Ultimately, these estimates of demographic parameters such as growth, mortality, and recruitment will be useful in future efforts at population modeling for following and predicting population status and recovery in a rigorous quantitative way.

OVERVIEW

MAIN TASKS

1) Locate a site with *Acropora palmata*.
2) Select three 7m radius areas with moderately dense *A. palmata* as study plots. If the *A. palmata* patch is of sufficient extent, the location of the plots should be selected randomly. NOTE: Further discussion and alternative means of site selection and set-up are included below.
3) Install a stake or other marker at the center of each plot (preferably at random GPS point).
4) Randomly select and tag 12 *A. palmata* colonies using random list of polar coordinates (see 'Tagged Colony Selection' field sheet, Appendix A). DIVER B.
5) Survey tagged colonies within each plot using the 'Initial Colony Survey' field sheet (Appendix A). DIVER A.
6) Photograph each colony from several angles, always including top-down view, with scale to document colony condition and help identify the colony in future surveys. DIVER A (DIVER B can assist with locating colonies and placing scale and photoboard).
7) Careful examination of entire plot to record location of ALL *Acropora palmata* (selected and unselected, particularly small crusts and attached fragments) within each plot using the 'Annual Plot Map'

(Appendix A) field sheet to enable detection of new recruits in future surveys.

8) Take a GPS waypoint over the center stake at each plot.
9) Download and rename image files.
10) Enter field data into the database after each survey.
11) Conduct quarterly surveys to assess condition and photograph the tagged colonies (Steps 5-7 and 10 above).
12) Annually map all colonies in each study plot (Step 8 above).

Optional Tasks

1) Branch linear extension (growth) measurement.
2) Tissue sampling for genotype analyses.

MATERIALS

Suggested materials for conducting surveys (Appendix B):

1) Metal stakes (e.g. steel rebar ¼″ (1.2cm) diam, ~14″ (45cm) length)
2) Sledge hammer (2.5 lb (1.2 kg))
3) Aluminum numbered tags (1¼″ (3 cm) diam)
4) Cable ties (to fit through 7/64″ (0.28cm) hole in tag)
5) Nails, (approx 2 ¼" (5.7cm) length, 1/8″ (0.3 cm) diam shank, 3/8" (1 cm) diam head)
6) Transect line (10m length) with brass clip
7) Dive compass
8) GPS with waterproof case
9) PVC meter stick (marked 5cm)
10) Pencils (mechanical pencils work well)
11) Rubber bands/string for securing pencils & paper to clipboard
12) Aluminum clipboards
13) Dura Copy Underwater paper (http://www.riteintherain.com)
14) Photocopier or laser printer (for printing field datasheets)
15) Underwater digital camera (>5 Mpixels & wide angle is best)
16) 2 memory cards (bigger is better)
17) 2 camera batteries
18) Photo board (see Appendix C)
19) Field datasheet template files (worksheet name italicized; see Appendix A):
    a) Initial Survey.xls
        i) Initial Colony Survey Diver A
        ii) Tag Colony Selection Diver B
    b) Quarterly Survey.xls
        i) Quarterly Colony Cond Diver A
        ii) Quarterly Sn/ Fish Census Diver B
    c) Annual plot map.xls
        i) Annual Plot Map Diver A
        ii) Annual Plot Map Diver B
        iii) Map Diagram

PERSONNEL

This protocol is designed to be conducted by two SCUBA divers. Since *Acropora* typically occurs in very shallow habitats divers should be comfortable wearing SCUBA gear in shallow water and should be experienced in the challenges of

working in surge conditions. We suggest that in general monitoring sites with less than 2m between the TOP of the colonies and sea surface be avoided due to the likelihood of damage to the colonies. This general recommendation can be reconsidered based on the local conditions (e.g. very sheltered site or small widely spaced colonies) and the experience of the divers. Note that very shallow sites may seem amenable to survey on snorkel, however making comprehensive colony assessments and taking photographs is greatly compromised on snorkel. Additionally, some tasks such as hammering (tags and stake) are impractical to impossible on snorkel.

Field tasks are divided such that those assigned to Diver A are those involving relatively more training and/or subjective assessments that rely upon making consistent observations. Diver B is assigned more supporting tasks and collects quantitative data that is less dependent on consistent interpretation. The person who assumes the role of Diver A should therefore be the person who is most likely to be consistently available for follow-up surveys. Where possible the tasks are also distributed so that both divers will complete the survey in the same amount of time.

INITIAL SURVEY

SITE SELECTION AND SET-UP

Acropora palmata populations have variable geographic layouts in differing areas and different reef structures that may dictate different site selection and layout (e.g., fore-reef spur-and-groove typical of Florida Keys versus fringing *A. palmata* zone or patch reefs along St. John USVI). The extremely patchy distribution of surviving populations makes random selection of sites at the regional level impractical; any reef site in which *A. palmata* is found should be considered for observation. Ideally, each surveyed reef site should include at least three randomly-placed, 7m radius permanent survey plots (Fig. 2) to provide statistical replication for recruitment assessments. There are various situations that may dictate a less than-ideal set-up. In many areas, an *A. palmata* patch may be too small to accommodate multiple plots or to make random selection of plot location feasible. Any section of reef (7m diameter) with at least 10 *A. palmata* colonies is suitable (Fig. 2) but areas of live *A. palmata* 'thicket' should be avoided because the reliable designation of individual colonies is problematic (Fig. 1). In the Florida Keys, *A. palmata* is generally in remnant patches of limited extent so plots are often centered deliberately in these small patches on adjacent or nearby reef spurs (Fig. 2). The number of field sites selected should be based on the number that can be feasibly surveyed on a regular quarterly schedule. A single survey plot typically takes up to three hours to initially tag and assess, and may take up to two hours to fully map in (both tasks vary greatly depending on the number of colonies present). At least two people will be required to tag colonies, and survey/map each plot and assess colony condition.

The following list summarizes the properties of a 'suitable plot' location:

1) Water depth should be deep enough that divers can maneuver over colonies easily without touching them. Specifically, the top branches should be at least 6 feet (2m) below the sea surface. Note that in these areas even 'minor' tidal fluctuations can greatly affect the workability of a particular area. When choosing sites it is helpful to be aware of the current tidal state.

2) Avoid places where the colonies are growing in a thicket since 'demographic' monitoring is necessarily dependent on being able to distinguish individual colonies.

3) Counting the number of colonies that are in a 7m radius area should result in the same number (+/- 1) if counted repeatedly.

This becomes more difficult when colonies are larger, more closely spaced, or have poorly differentiated boundaries (discussed in detail below).

To re-iterate, the center of plot should be located randomly if feasible. Otherwise, after identifying a suitable plot location with an appropriate *A. palmata* density that is limited in extent, a metal stake should be centrally placed using a sledge hammer. Care should be taken to minimize disturbance by choosing areas for stake placement (and colony tag placement) that are not covered by coral or sponge and have adequate surrounding working space.

After placing the center stake, the number of colonies within 7 meters of the stake should be carefully counted by both divers. This count should include all colonies of any size or shape, except loose fragments (NOTE: loose fragments are not considered in the designation of colonies for ongoing observation, but they are counted in the plot-level assessments of recruitment). If there are only 10-12 colonies within the plot, all colonies can be automatically selected and tagged. However, this short-cut should be used only after observers have confirmed that only 10-12 colonies exist within the plot. Keep in mind that oftentimes additional small or remnant colonies are discovered during a careful search of the plot. For any plot with more than 12 colonies, the colonies selected for tagging must be chosen randomly (discussed below).

COLONY DETERMINATION & SELECTION

Acropora palmata, like other coral species, can often grow in unique orientations with variable morphologies. Colonies can fragment (after which the fragment can become its own colony), fuse with other colonies, and suffer partial mortality which divides a single colony into smaller isolated patches of tissue (Fig. 3). For an excellent discussion of these coral 'colony ambiguities' see, Hughes and Jackson (1980). For these surveys, the following rules should be applied to distinguishing single colonies:

1) Primarily, any continuous live tissue should be considered a single colony (Fig. 4).

2) If live tissue is not continuous, physically separated units of live tissue that are growing on the same underlying skeletal colony structure should be considered the same colony (Fig. 4).

3) If live tissue is not continuous and is located on underlying remnant/dead structure that is difficult to distinguish (i.e., one 'stem' or two?), patches of tissue within 1 meter of each other should be

considered a single colony. Patches more than 1 meter from surrounding tissue should be considered a separate colony (Fig. 4).

4) If the center (based on top view) of the colony is outside of 7m it should not be included.

The initial colony selection and tagging process involves two divers, referred to from here as 'Diver A' & 'Diver B'. At this stage, Diver B is responsible for selecting colonies and physically placing aluminum tags into the substrate and should carry the field sheet 'Tagged Colony Selection' (Fig. 5, Appendix A), which lists randomly generated coordinates (compass heading and distance from stake) to be used for selecting colonies. If only 10-12 *A. palmata* colonies are present in the plot, all should be tagged. If more than 12 colonies exist in the plot, 'Diver B' should randomly select colonies by following each 'random coordinate' listed on the field sheet ('Deg' = compass heading (0-355º), 'Dist' = distance from the central plot stake (1-7m)). If the center of a colony lies within 0.5 m of the first random coordinate, it should be selected and tagged. If there is no colony at the first random coordinate, 'Diver B' should proceed to the second random coordinate. Repeat this procedure until 12 colonies have been selected.

Each selected colony is tagged on adjacent substrate (Fig. 6). Different kinds of tags can be used; we use 3cm diameter, numbered, aluminum tags nailed into adjacent bare substrate, (or if necessary, by cable-tying it to an adjacent solid structure) because they are relatively inconspicuous (which is important to our permitting authorities and draws less attention of other visitors). More apparent tags, such as cattle ear tags can also be used. The tag should be placed close enough to the colony to clearly indicate to which colony it refers, but also far enough away to prevent future overgrowth by the colony. Less conspicuous tags, particularly, should be placed consistently on the side of the colony between the stake and the colony for ease of relocation. If this is not possible, the location can be noted on Diver B's field sheet. The tag numbers should be recorded along with the actual colony coordinates (heading and distance from stake) for each colony on Diver B's field sheet. Tags should not be attached directly to live tissue as they can be rapidly overgrown or irritate the surrounding tissue. Additionally, tags should not be nailed to an already dead portion of the colony because hammering can cause distal portions of the colony to break.

Once a colony is tagged by 'Diver B', 'Diver A' should assess of the condition of tagged colonies using the field sheet 'Initial Colony Condition' (Fig. 7, Appendix A). The maximum length, width and height of each colony should be recorded by 'Diver A' (described below), along with an estimate of % live tissue, and the presence of damselfish chimneys, bleaching, recent mortality, branch breakages,

predatory snails and damselfish. A detailed description of each of these assessment categories can be found in the section 'Assessment of Tagged Colonies' below.

After selecting and marking colonies, be sure to record a GPS waypoint for the center stake of each selected plot. It is useful to have an abbreviation for the site and number for the plot, for example the 2nd plot at 'French Reef' might be 'FR2,' and this should be recorded on the data sheet along with the date and any other notes that may assist subsequent surveys.

Assessment of Tagged Colonies

Size

On the field sheets, 'Initial Colony Survey' (Fig. 7) the observer will measure colony length ('L'), width ('W'), and height ('H') in centimeters. To ensure that measurements are straight, its best to use a solid measuring stick or measuring tape attached to a length of PVC pipe (or other 'measuring stick'). Length is a measure of the maximum colony diameter and width should be the dimension perpendicular to height (Fig. 8). Height should be measured as the maximum height of the colony from the base to top. These measurements may include dead portions of the colony, particularly at the base. Importantly, height measurements should be perpendicular to the substrate (pay close attention to the angle of the substrate; Fig. 8). It can be difficult to be consistent with the measurement of colony height since parts of a colony may differ dramatically in height. Keep in mind that this measurement is taken to estimate how much tissue is present (when used in conjunction with the % live estimate described below) so it is more useful to measure the height of most of the colony rather than measuring the colony height at a single taller branch. Even when the height is measured carefully, small differences in where it is measured can lead to misleading differences in measurements between surveys. Taking pictures from the sides of the colony will allow these measurements to be roughly verified by documenting whether major changes occurred in the colony height/structure since the previous survey.

Percent Live

The percentage of the measured colony that is covered with live tissue, '% live' should be estimated for each tagged colony. A colony with a score of 100% live (Fig. 9) would have live tissue covering the entire surface of the skeleton that was included when length, width, and height was measured (Fig. 9, Fig. 10). It is important to look at the undersides of branches and all sides of a colony when

making this estimate (Fig. 9, Fig. 10). Remember that bleached regions of the colony are still living and thus should be included in estimates of % live.

Colony Conditions

The following sections describe many conditions (most are detrimental conditions) that affect *A. palmata* colonies. Each colony is scored (presence/absence or a rank estimate of severity) for each condition. In the cases of bleaching and recent mortality conditions the conditions are further categorized by as to 'type' which is recorded on the data sheet by a specific code (explained further below). For the conditions that are scored with a qualitative rank, the rank should be scaled to the live area of the affected colony. For all ranked conditions the following scale applies:

0: Not present
1: Present, very sparse occurrence (≤5%)
2: Moderate colony coverage (10-25%)
3: High coverage, condition affecting 30-55% of the colony live area
4: Catastrophic extent, 60-85% of the colony's live area affected
5: Entire colony (≥90%) affected

Healthy/Unaffected

What does it look like?

A healthy colony is defined by tissue color and a lack of other detrimental conditions (recent mortality, bleaching, etc.). Healthy *Acropora* tissue (Fig. 11) varies in color from tan to a light orange to deep orange-brown. The intensity of the color may vary with depth and season, but a fair range of color can be considered 'within normal limits' (www.coralwatch.org uses a color chart to characterize coral health, healthy *Acropora palmata* typically corresponds to E5-E6 or D4-D6 on their chart). It is important to note that tissue around the growing edges of a colony is normally white (lacking zooxanthellae) but is not considered bleached or dead.

How is it scored?

Healthy colonies are designated on the field sheet only by the absence of all other tissue conditions. A colony may have less than 100% live tissue coverage but may still be considered 'healthy' (i.e. have no noted conditions) as long as all of the remaining tissue is normal (or within normal limits) and the dead portions are not recently dead (see below).

Recent Mortality: Rank

What does it look like?

When *Acropora* tissue dies it sloughs away leaving clean white skeleton exposed (Fig. 12). Freshly exposed skeleton is bright white with clearly visible, sharp corallite structure (Fig. 11b). Bare skeleton is gradually colonized by a thin turf of algae (approximately 1 week after tissue dies) and will eventually (6-8 weeks) appear similar to the surrounding substrate in terms of algal colonization. Over this time the corallite structure gradually erodes (Fig. 13).

Dead portions of a colony are distinguished as either 'recent mortality' or 'long dead'. Recent mortality includes areas of exposed skeleton that have little or no algal colonization and sharply defined corallite structure. These areas appear bright white or have a light brown/green tinge once filamentous algae are present. Based on past field observations, areas described here as 'recent mortality' have died within 4 to 6 weeks. Long dead areas have heavier algal colonization including fleshier types of algae and/or calcareous coralline algae (CCA). Additionally, the corallite structure gradually erodes so that long dead areas may appear completely smooth or only slightly bumpy. There is no definitive line where an area of dead skeleton is classified as recent or long dead (Fig. 12) and these processes vary with season and location.

How is it scored?

The extent of recent mortality should be qualitatively ranked on a scale from 0-5 (below) for each tagged colony regardless of the cause. The ranking is based on the amount (area) of the colony affected relative to the amount of living tissue on the colony (Figs 14-18). A 20cm lesion on a 30cm colony would get a much higher rank than the same size lesion on a 100cm colony.

0: Not present
1: Present, very sparse occurrence (≤5%)
2: Moderate colony coverage (10-25%)
3: High coverage, condition affecting 30-55% of the colony live area
4: Catastrophic extent, 60-85% of the colony's live area affected
5: Entire colony (≥90%) affected.

Recent Mortality: Type

The suspected cause(s) of the recent mortality ranked in the previous step is assigned a 'type' code(s) which is recorded on the field sheet (Fig. 7). The following

codes should be used to indicate the likely source(s) of the recent mortality that was ranked (each described further below):

WPx = white pox disease
WBD = white band disease
RTL = rapid tissue loss
FS = snail feeding scar
PFB = parrotfish bites
CB= ciliate band
CL= *Cliona* lesion
FP = fish poop
Unkn = unknown cause

In cases where there are multiple sources of recent mortality the rank should reflect the sum of all types of recent mortality. Where possible, each type should be recorded as primary, secondary etc. based on its relative impact to the live portion of the colony. For example a colony with large areas of RTL and a small feeding scar might be ranked a '3' and the 'RTL code would be recorded in the 1° column and the FS code in the 2° column.

For ranking purposes, all recent mortality types are grouped because the impact on the colony is the same regardless of cause. The causes that have been identified here are actually quite diverse including disease, predation, parasitism and incidental causes. The purpose of distinguishing these is to identify the influence that each has on the survivorship of *Acropora* colonies. The following section describes each type of recent mortality but it is important to emphasize here that many of the characteristic signs can be difficult to distinguish. This is particularly true of the three types that are considered to be diseases. For this reason some studies have grouped them as 'acroporid white disease'. While this is a conservative and valid approach we suggest attempting to distinguish the gross manifestations into more specific disease types since they can easily be grouped later. It is important to realize that rigorous 'diagnosis' requires identifying the pathogen which can NOT be done visually in the field. As a result distinguishing these three 'white diseases' is restricted to the pattern and rate of tissue loss (Fig. 19). Tissue loss can be band-like, patchy or irregular and sloughy. It is easiest to interpret disease signs when it is actively progressing (Fig. 19), but even if the progression has halted (recrudesced) and there is light algal colonization it is possible to roughly detect a pattern and the rate of progression.

White Band Disease (Type Code: WBD)

Early reports of White-band disease (WBD) described its appearance on *A. palmata* as "a sharp line of advance where the distally located zooxanthella-bearing

coral tissue is cleanly and completely removed from the skeleton, leaving a sharp white zone about 1 cm wide that grades proximally into algal successional stages" (e.g. Fig. 20; (Gladfelter 1982)). Reported rates of advance on *Acropora palmata* average 5.5 mm d^{-1} ((Gladfelter 1982)). In addition to the relatively uniform band-like pattern, it also progresses in a relatively uniform manner, often radiating up multiple branches at the same time (Fig. 20). Very close inspection of the tissue at the 'white band' often has a rough or ragged appearance as though the tissue is sloughing off of the skeleton at the 'active disease' front, but from more distant inspection it typically appears very uniform.

In Florida and parts of the Caribbean it is common to see WBD only on the undersides of branches, the progression of the disease often stops once it crosses to the upper surface of the branch ((Williams, Miller 2006); Fig. 21).

White Pox (Type Code:WPx)

White pox (WPx) produces scattered patches of recent mortality (often called 'lesions') on *A. palmata* colonies. The bacteria *Serratia marcescens* has been identified as the etiological agent of white pox (Patterson et al. 2002). 'Patchy necrosis' has also been used (Bruckner, Bruckner 1997; Rodriguez-Martinez et al. 2001; Weil et al. 2002) to describe lesions that are indistinguishable from those characterized as white pox. Here we will use the term white pox (WPx) to describe multifocal, irregular patches of recently exposed skeleton (Fig. 22). Patches (lesions) are typically 10-20cm across (pers. obs.) but the size can vary, and they can remove up to 12 cm^2 (mean 2.5 cm^2) of live tissue per day (Patterson et al. 2002). Unlike WBD, this disease does not progress at a steady rate, rather, lesions seem to form rapidly then stall (pers. obs). Once a lesion stalls and the skeleton is colonized by turf algae the tissue margin looses its ragged appearance and no longer appears to be 'actively' progressing. Stalled lesions may become active again but often they begin to heal (Fig. 23) (Lirman 2000). Lesions are also reported to coalesce into larger lesions (Rodriguez-Martinez et al. 2001).

Rapid Tissue Loss (Type Code: RTL)

Acropora colonies are also observed with large areas of recent mortality indicating that tissue loss occurred very rapidly, much faster and in a pattern not consistent with the accepted descriptions of either WBD or WPx (Fig. 24). It could be that our present descriptions of these diseases are not adequate to describe the full range of disease signs manifested, or it is possible that additional diseases affect *Acropora*. Since this can not be determined in the field, an additional condition named 'rapid tissue loss' (RTL) is used to describe colonies with large areas of recent mortality (Fig. 24, Fig. 25). These areas range from band-like to lesion-like and with a highly ragged tissue margin.

Corallivorous Snail Feeding Scars (Type Code: FS)

The short coral snail, *Coralliophila abbreviata* is a common predator on *Acropora* spp. and can consume more than 3 cm^2/day (Miller 2001). It has a white, ridged shell that is usually overgrown with algae (Fig. 26) making it difficult to recognize (see also 'Snail & Damselfish Census' section). It resides on *A. palmata* colonies for extended periods of time and is most often found in small groups (3 to 6 snails) adjacent to live tissue (Fig. 26) but may wander to other portions of the colony when 'resting' (not feeding; Fig. 26).

Feeding scars vary in size from a few square centimeters to 100 cm2 depending on the size and number of snails feeding in that area. The shape of a feeding scar is highly variable, but often has a scalloped margin and is almost always found along the tissue margin of a colony leading into a 'long dead' portion of the colony. Snails typically begin feeding at the base of a colony and leave behind a path (Fig. 27) of dead skeleton that can be obvious on larger colonies.

Feeding scars can be distinguished from disease associated recent mortality first and foremost by the presence of snails (see section 'multiple types' for exceptions). Additionally, while the overall shape of the scar may be irregular, the actual margin of live tissue is typically relatively clean compared to 'active disease' areas. Snails are only rarely observed feeding in an area surrounded by live tissue (Fig. 28), thus while it is conceivable that they could produce a WPx type lesion, it is unlikely that they would produce several lesions on one colony and then disappear from the colony.

However, feeding scars are often found on the undersides of branches and can appear similar to WBD (Fig. 28). In these cases, presence of the band of recent mortality on multiple branches is more consistent with WBD. If thorough searching of the colony does not reveal any snails and the recent mortality is otherwise consistent with WBD then it should be categorized as WBD.

Rapid tissue loss has been observed to coincide with snail feeding activity, when snails are present this can only be presumed if the area of recent mortality is significantly larger than can be explained by the number of snails present, or if the margin is highly irregular and 'ragged'. If the observer is in doubt and snails are present and no other disease lesions are present on the remainder of the colony, then the recent mortality should be presumed to be a feeding scar.

Parrotfish Bites (Type Code: PFB)

Parrotfish typically feed by scraping reef substrate for the endolithic algae and sponges. However scattered scrape marks (Fig. 29) are not uncommon on live *A. palmata*. They are distinctive from other types of recent mortality by the absence of corallites and chiseled appearance of the skeleton (Fig. 29). Stoplight parrotfish

(*Sparisoma viride*), are known to form loose feeding aggregations and repeatedly scrape a cluster of coral heads over days to weeks leaving scars up to 200cm^2 on *Montastraea annularis* (Frydl 1979). This 'focused biting' behavior (Humann, DeLoach 2002a; Bruckner et al. 2003) is poorly documented in *A. palmata* but clearly occurs sporadically (Fig. 30). This natural but destructive feeding habit removes live tissue and abrades the skeleton, occasionally affecting large continuous portions of a colony, typically along the entire margin of large branches.

Parrotfish bites should only be recorded if they affect the colony's live tissue, when they occur on long dead portions of the colony they do not need to be recorded. Additionally, a colony may bear marks that resemble parrotfish bites from a distance, however on closer inspection the corallites and polyps/tissue are intact, and the marks are simply bleached (Fig. 31). This is likely to be a stage of recovery from parrotfish bites (pers obs) but further documentation is needed. Regardless of the explanation, these bleached areas are not recent mortality and thus should not be included in the rank, instead they will be included as bleached tissue (discussed below).

Ciliate Band (Type Code: CB)

A dark greenish-black band associated with areas of recent mortality (Fig. 32) has recently been identified as infestation by the ciliate *Halofolliculina* sp. (Croquer et al. 2006). It is uncertain whether or not the ciliates are responsible for the recent mortality or if they are opportunistic colonizers of already compromised tissue. Regardless they are included here as a 'type' of recent mortality. Based on observations in Florida, when ciliates are present, they are generally the secondary type of recent mortality (Fig. 33) as they are often observed associated with other disease signs, feeding scars and *Cliona* lesions (discussed below). While it is altogether possible that they alone could cause tissue mortality, they should only be considered primary if they are present in all instances (i.e. lesions) of disease on that colony.

Cliona Lesions (Type Code: CL)

Cliona laticavicola is an orange excavating sponge found commonly in reef substrate. It does not form a thallus, but exists only inside substrates. It is visible only where its ostia burrow to the surface (Fig. 34) or in freshly fragmented substrates.

Cliona sp. can grow aggressively around the *A. palmata*'s live tissue margin and appear to drive tissue retreat. In cases where sponge is associated with a lesion (recent mortality), it should be included in the ranking and noted as type CL. *Cliona* present on the long dead portions of the colony or in healing lesions should not be assessed as recent mortality.

Fish Poop Lesions (Type Code: FP)

The flat palmate structure of *A. palmata* branches allows fish fecal material (Fig. 35; fish poop!) to settle and remain on the branches, particularly in calm weather. In many cases the material will smother the tissue underneath and result in a lesion ((Weil et al. 2002; Williams, Miller 2006)). *Acropora palmata* colonies are commonly seen with a few scattered small (<10cm) lesions. These can appear similar to WPx lesions and in fact it has been suggested that they are related (Bruckner 2002). Lesions caused by fish poop are often crescent shaped (Williams, Miller 2006) with small amounts of 'poop' remaining in the lesion. These types of lesions heal rapidly.

While fish poop is common on colonies, it does not always result in a lesion and should only be recorded as a 'type' of 'recent mortality' if it is responsible for a notable portion of the recent mortality ranking.

Unknown (Unkn)

If recent mortality is present and can not be identified as one of the described types, it should be recorded as unknown with notes as to likely types. In these cases taking several photographs (both wide angle and close up; see section 'Photographs' below) will allow it to be re-evaluated later. When categorized as 'unknown' the surveyor should note on the field sheet any additional information that may help evaluate it later. It is particularly important to note whether the tissue is bleached or if it is gone (i.e. recent mortality), while this can be difficult to determine in the field it is even harder to determine from a photograph (white areas do not photograph well). In many cases photographs taken at the next survey will show whether the area was dead or just bleached.

Bleached Tissue: Rank

What does it look like?

Acroporid tissue changes color from healthy to pale to completely translucent, bleached tissue when zooxanthellae are lost (Fig. 36). Totally bleached portions of colonies are bright white and from a distance look similar to very recent mortality. However, on close inspection polyps are visible and the sharp corallite structure of the skeleton is obscured by the translucent tissue. Bleaching may affect whole colonies in extreme situations, but more commonly, only part of the colony is bleached (Fig. 37). Furthermore, the affected tissue itself may be less than completely bleached resulting in pale tissue.

Tissue bleaching occurs for a variety of reasons including thermal stress, changes in light, tissue trauma or other acute stresses. Mass bleaching events are an

extreme example of acute bleaching, apart from these events, whole colonies are rarely fully bleached. Partially bleached colonies typically recover before the next survey (3 months). In contrast there are colonies with discrete patches of bleached tissue that persist for long periods (years). These patches are most often triangular or wedge shaped and found around the base of the colony (Fig. 38). Since there are no published descriptions of these patches we will refer to them as 'Wedgies'. Although they seem completely benign, their cause is uncertain and by noting their occurrence we can document any effects and perhaps eventually attribute causes.

How is it scored?

Bleaching is ranked on a scale from 0 to 5 (see scale in 'recent mortality' section above) on the field sheet. Pale tissue is treated here as a less severe form of bleaching. The ranking should reflect both the area bleached/pale tissue and the degree to which it is bleached relative to the amount of live tissue on the colony. As with the recent mortality rank, a small patch of bleached tissue would get a lower rank on a large colony than would the same sized patch on a smaller colony.

Bleached Tissue: Type

Codes should similarly be applied to describing the nature of colony bleaching.

Bl= bleached (intact, colorless live tissue)
W= "wedgies" (small, wedge-shaped areas of bleached live tissue)
R= rings (white ring-shaped areas of live tissue)

All bleaching should be recorded as the type code 'Bl' regardless of severity with two exceptions. Two specific patterns are coded separately due to their distinctive appearance. First, are the afore mentioned 'wedgies' They are typically small (~10cm max length) and found at the margins of a colony though they can occur anywhere on the colony. They are observed to persist for long periods (e.g., >2yrs of observation in the Florida Keys) and do not seem to change or adversely affect the colony. They are recorded as 'wedgies' (W) under bleaching so that an estimate of their prevalence and distribution can be documented. Second, rings of bleached tissue (Fig. 39) are sporadically observed on colonies. These are thought to result from yellowtail damselfish bites (Bruckner et al. 2003). The bleached tissue regains it's normal color within weeks, and frequently new rings will appear on the colony in that time.

Damselfish Impact

What does it look like?

Threespot damselfish (*Stegastes planifrons*; referred to from here simply as 'damselfish') can aggressively establish territories (Fig. 40) on large areas of *A. palmata* (Robertson et al. 1981). They are thought to farm algae 'lawns' (Fig. 41) within that defended territory. Impact on live tissue in a territory ranges from sparse bites that heal into chimney-like structures (Fig. 40) to lawns which result from dense clusters of bites that are rapidly colonized by algae leaving no live tissue on that portion of the colony. Bites initially appear as small (~1cm) spots of bleached or recently dead tissue usually interspersed with chimneys or other signs of damselfish activity. Damselfish nests (Fig. 42) also impact *A. palmata* colonies; they prefer to nest on the undersides of branches or sheltered portions of the colony. Eggs are deposited in the nests year round but seem to be more common in the summer. When eggs are present in the nest, they appear as dark purple-black masses, but when vacant, these areas of dead skeleton can resemble white-band diseased areas or healing lesions (Fig. 42). The same nest is used repeatedly which reduces the impact on live tissue, but when new nests are built live tissue is often cleared to create the nests.

How is it scored?

The amount of perceived impact (chimneys, bites, lawns & nests) to the colony should be ranked from 0 to 5 (Fig. 43 Fig. 44 Fig. 45) on the field sheet. Since these bites can include both bleached and dead areas that are too small to accurately classify and/or rank as either condition, they are included in the damselfish impact rank. Lawns are difficult to distinguish from other long dead portions of the colony, these should only be included in the rank if the area is surrounded with chimneys or other evident damselfish- inflicted damage.

Skeletal Anomalies

What does it look like?

Several types of skeletal/growth anomalies can affect *A. palmata* colonies. While the cause is unknown, tracking them over time will allow us to document their impact on overall colony performance.

Neoplasia (Np) (Fig. 46) are smooth bulbous growths on the colony surface with little or no coloration. Neoplastic regions typically occur along the new growth edge of *A. palmata* colonies. More technical description can be found in (Peters et al. 1986; Bruckner 2003; Gateno et al. 2003). Hyperplasia (Hp) (Fig. 46) is another condition commonly observed and appears as polyp calices that display accelerated and irregular growth. Affected polyps are large and pronounced and protrude above the colony surface (Bruckner 2003).

Colonies often have area around the margin where the polyps (corallites) are densely spaced and lacking zooxanthellae (Fig. 47). The cause of this pattern is unknown but often Yellowtail damselfish are closely associated so it is possible that it is territory or nesting related. For the purposes of this study, this phenomenon is called dense white polyps (DWP) and is broadly categorized as a skeletal anomaly. Additional cases of irregular corallite structure are also observed and simply classified as 'unknown'. Note that corallite structure can vary somewhat between colonies but this category is reserved for cases where the skeleton on a portion of a colony is notably different from the remainder of the colony.

How is it scored?

Skeletal anomalies are recorded only by the type of anomaly without regard to the severity of the anomaly. Skeletal anomaly types are recorded on the field sheet with the following codes:

Np = neoplasia
Hp = hyperplasia
DWP = dense white polyps
Unkn = unknown

Overgrowth

What does it look like?

Competition for space often occurs between sessile organisms, *Acropora* included (Fig. 48, Fig. 49). Its fast growth rate gives it an advantage in many situations, but the competitive interaction probably slows growth at best, but it often results in retreat of the tissue margin. This is especially true with *Cliona langae* (Fig. 48), an encrusting sponge (Weil et al. 2002; Bruckner et al. 2003) that is found in the Greater Antilles portion of the Caribbean. In Florida, overgrowth most often occurs with *Erythropodium caribaeorum* (Fig. 49), *Millepora complanata*, colonial tunicates and macrosponges. Regardless of the competing species, the interaction between *Acropora* and a competitor is typically marked by a margin of blanched tissue (pers. obs.).

How is it scored?

This sponge and other overgrowing organisms are not typically associated with recent mortality so it is not included as a 'type' of recent mortality. Instead it is classified as overgrowth and the type of organism should be recorded on the field sheet.

Fragmentation/ Physical Damage

The natural life history of acroporid corals relies strongly on fragmentation and the successful reattachment and growth of the fragments (Fong, Lirman 1995). This reproductive strategy has associated costs and benefits. It can foster rapid proliferation and spatial dominance; however it can also result in losses since loose fragments are more vulnerable to export or physical stress. Once broken, branches may nestle into surrounding reef structures and reattach, or they may be transported to sandy areas where they become abraded or smothered and can not attach. Although, asexual reproduction through fragmentation is a natural process, fragments are less likely to be retained in lower density stands (Fong, Lirman 1995; Miller et al. 2002), pers. obs.) typical of present populations.

What does it look like?

Branch breaks that occurred recently are fairly obvious, they are bright white, smooth and angular (Fig. 50). These areas are colonized by algae in the same way as recent mortality but should not be included as recent mortality since the tissue did not die. Older breaks can be difficult to recognize in the field, but can be detected in the photo series for each colony.

How is it scored?

Obvious broken branches on each colony should be counted and recorded on the field sheet according to the approximate diameter of the break (it is not important to take precise measurements of each break encountered).

sm = small <5cm
med = medium 5cm-10cm
lg = large 10cm-20cm
xlg = extra-large >20cm

Extreme disturbance events may 'fragment' entire colonies rather than individual branches. This would be scored as an extra-large break.

Snail and Damselfish Census

In addition to characterizing the *A. palmata* colony itself, we also document the associated predators. On each tagged colony, the number of corallivorous snails (*Coralliophila abbreviata* (Fig. 51) and *C. caribbaea*, Threespot damselfish (*Stegastes planifrons*) (Fig. 52), and Yellowtail damselfish (*Microspathodon chrysurus*) (Fig. 53) associated with each colony should be recorded on the field sheet.

Corallivorous Snails

Several types of snails are commonly found on *A. palmata* colonies, however only two species of *Coralliophila* spp. (Fig. 51) are significant predators of live tissue, and only these are to be counted in surveys. *Coralliophila abbreviata* (yellow operculum) is by far the most common species, but occasionally *Coralliophila caribbaea* (purple-brown operculum) is found feeding on *Acropora* colonies. They can be difficult to distinguish without removing them from the colony, so for the purposes of these surveys, both species can be counted and recorded under 'snails' on the datasheet.

Other snails (especially *Thais deltoidea;* Fig. 51) are commonly found on *Acropora* colonies. While the clean shells of these two are very different, both are very well camouflaged by heavy algal encrusted growth on their shells and may be difficult to distinguish while resting on the colony. When removed from the colony they are readily distinguished by the color of their operculum and body: *Coralliophila abbreviata* are bright yellow while *T. deltoidea* are deep purple. However, past observations have shown that attempting to replace snails that are removed from a colony for identification often fails. Frequently they are on undersides of branches and can't be replaced, and replacing them in other areas at the very least changes the placement of the feeding scars, but often the snail will wander off of the colony. All of these outcomes will affect the data being collected so should be avoided among tagged colonies. Thus, surveyors are encouraged to develop a search image by searching for snails on *Acropora* colonies outside of the study plot where they can be removed for closer inspection.

There are other visual ways to distinguish these snails that do not require disturbing them. First: Snails found associated with a feeding scar are most likely *Coralliophila* spp. *Thais deltoidea* does not feed on coral tissue, so is only coincidentally observed in the vicinity of a feeding scar. However, *Coralliophila abbreviata* is occasionally found with no apparent feeding scar, so the inverse is not necessarily true. Second: *Thais deltoidea* is typically observed alone (or occasionally feeding on another snail) while *C. abbreviata* is typically found in groups of 2 or more (most often 3 to 6).

Third: In spite of heavy algal growth it is often possible to see the rounded spires on *T. deltoidea*. *C. abbreviata* does not have this knobby appearance and it is occasionally possible to see the fine ridges through the algal growth.

Coralliophila shells that are heavily overgrown by algae can also appear similar to clumps of algae, tunicates or other non-descript algal encrusted lumps. Wiggling the lumps very gently can reveal if they feel fixed (i.e. skeletal substrate) or 'give' slightly (a snail shell wobbling on an attached foot), or have a more mushy texture such as a tunicate.

Based on thorough visual inspection and touch verification for suspicious algal covered lumps around the margins of live tissue, record the total number of corallivorous snails for each tagged colony.

Threespot Damselfish

Threespot damselfish (*Stegastes planifrons;* Fig. 52) are commonly associated with *Acropora spp.* and other corals. They aggressively defend a localized territory on live coral colonies and can inflict substantial damage to the live tissue in their territories. In addition to assessing the chimneys and other damage associated with their territory (described above), the number of adult ('Ad') and juvenile ('Juv') fish present on a colony are recorded during each survey. When colonies are closely spaced, a single fish may be defending a territory that spans across portions of 2 separate colonies. NOTE: In these cases the one fish should be counted twice (and note that it was the same fish), once for each colony since both colonies are being affected. Juveniles can be highly abundant but do not have the same impact as do the adults. These should be recorded in the separate column labeled 'juv'.

Other species of damselfish similar in appearance are often associated with *A. palmata*, and can even be more abundant than the threespot, but do not seem to result in colony damage so are not included in these surveys. In particular, Dusky, Beaugregory and Cocoa damselfishes can be common and similar in appearance to threespots (as adults). For specific distinguishing traits see one of the numerous field guides to reef fishes (Humann, DeLoach 2002b).

Yellowtail Damselfish

Yellowtail damselfish (*Microspathodon chrysurus;* Fig. 53) are also associated with *Acropora palmata* colonies and frequently overlap with the territory of a threespot, dusky or other damselfish. Adults typically roam over a larger area than the threespot (often multiple colonies) while juveniles remain in more confined areas. Their effects on the colony are both less dramatic and distinctive. Their bites may produce small (2cm) circular bleached patches ((Bruckner et al. 2003)) and they also occasionally nest around live *Acropora palmata* tissue. The number of adult and juvenile yellowtail roaming over a colony should be recorded at each survey. It can be difficult to determine the 'territory' of an adult yellowtail but if an individual appears closely linked to the colony it should be counted.

PHOTOGRAPHING COLONIES

Photographs are used to document what was observed during a survey so that future comparisons can be made. The data collected during the survey is needed for quantitative analyses, and could be adequate alone, however many of the observations in the field are qualitative and subjective so that taking photographs can allow data to be verified for consistency, particularly when comparing between observers.

At each survey, all tagged colonies should be digitally photographed from several angles with scale and colony tag number included. Diver A should take the photographs since it is important to document the conditions noted in the colony condition assessment. At minimum, photographs should be taken from each of these angles:

1) Planar (top down) view of the whole colony with scale (meter stick) and tag number (on 'photoboard' see below and Appendix C).
2) Side view of the whole colony from at least 2 sides, but preferably more for larger colonies).
3) Close-up view of any special conditions affecting the colony (disease lesions, predation feeding scars, etc).

Tips:

1) See the suggested 'photoboard' (Appendix C) for a method of including the colony tag number in the photograph which is stable (even in moderate surge). This board can also serve as scale for smaller (<50cm) colonies.
2) The scale/photoboard should be placed on top of the colony and level for the planar view photograph. It will only work as accurate scale if it is parallel to the camera lens and colony surface. If placed and removed gently it will not damage the coral, however it is preferable to place it on already dead areas, if present.
3) In cases where the colony is large and/or too shallow for the planar view to fit in one frame, simply take several overlapping photos from far away as possible. In these cases it is still useful to have an additional view with the entire colony in one frame though it will be taken from an oblique angle.
4) If the scale or photoboard obscures important colony details, a second picture can be taken without them.

5) The photoboard and scale only need to be in the first few whole colony photos so long as all photos of that colony are taken consecutively and the board is placed in the first photo of the next colony.

6) We have found it to be more efficient to photograph all colonies after the colony assessment data are collected.

7) If Diver B has completed all tasks, he/she can assist Diver A by locating and the next colony, placing the scale(s) and preparing the photoboard with the proper colony designation while Diver A completes the close-up condition photos for the previous one. This allows the photographer to concentrate on the colony at hand and saves time.

8) If many cases of the same type of condition (e.g., many disease lesions) are present on one colony it is not necessary to photograph each one, select a few representative shots.

A typical plot with 12 colonies will have approximately 6 photos per colony meaning 72 files per plot, so it is crucial to keep the files organized. At the end of the day, the pictures should be downloaded and renamed using a conventional naming scheme. Our surveys use the following mask: SI###-1a where SI is a 2 letter site abbreviation and ### is the three digit tag number followed by a dash then the survey number (1 in the above example) and a different serial letter assigned to each image. This task is a lot easier using a 'batch rename' tool available in many image processing software programs or as standalone shareware programs available online. The date and time and other file information will be retained as part of the file information so it is not necessary to include it in the file name.

MAPPING *ACROPORA PALMATA* COLONIES

Use the field sheets 'Annual Plot Map' to record the coordinates (distance from stake and compass bearing) of all *A. palmata* colonies within each plot, including tagged and untagged colonies, very small colonies and attached fragments. Loose fragments that are not mapped should be tallied as they are encountered in the mapping process. Use the rules described above to distinguish the individual colonies to be mapped. Plot mapping will require at least two divers, and will likely require at least 1 long dive per plot.

Prior to mapping, tag numbers and coordinate information ('distance from stake', 'compass heading from stake') for tagged colonies should be recorded on the field sheets 'Annual Plot Map', then confirmed during the mapping process.

During the mapping dive, Diver B should remain over the central stake, and hold the measuring tape in place while Diver A extends the measuring tape to the center of each colony (Fig. 54). 'Diver B should read the compass heading from the central stake to the colony and signal it to Diver A. IMPORTANT: Note that the compass should be held at least 50cm from the stake to avoid magnetic distortion from the stake. Diver A should assign sequential numbers to each untagged colony, and record the distance from the central stake to the center of each colony, the size (length, width, and height), % live tissue and colony type for each colony on the field sheet 'Annual Plot Map'. This process should be repeated for all *A. palmata* colonies within a plot, including very small colonies and attached fragments. As the diver-team comes across a tagged colony, the original coordinates (determined during the tagging process) can be quickly confirmed.

It is worth a specific reminder that dive compasses are designed to be read through the side window, not from the top. Many experienced divers tend to forget or (worse) be inconsistent in reading their compass from the top. MAKE SURE YOU ARE CONSISTENT both in reading your compass through the side window and in the direction the heading of each colony is recorded (i.e. FROM stake TO colony). By the same token, if on subsequent annual mapping surveys you have trouble locating many or all of the previously mapped colonies, it might be worth checking the opposite heading.

While assigning 'colony type' for each colony on the data sheet (Diver A), avoid touching the colonies or fragments as the re-attachment process can be easily interrupted. Try to assess the nature of each fragment visually only!

On the data sheet, 'type' should be filled in as follows:

- Branched colony (BC) - 'normal' looking colony with branches, may have some partial mortality.
- Remnant colony (RC) - live tissue mostly encrusting, no/few branches .
- Attached fragment (AF) - live fragment (usually a branch) with some signs of attachment to the reef. If attachment of a fragment can not be determined visually, map it and note the uncertainty. It is important not to touch fragments since they may be in the early stages of forming and attachment.
- Stable Fragment (SF) - Occasionally large (>75cm) portions of colonies are broken off and found 'loose' in the plot. While they may not be attached to the substrate they can be considered 'stable' due to their weight, structure and/or location. These 'fragments' should be

counted among the colonies initially chosen for tagging and mapped along with other attached colonies.

- Loose Fragment (LF) - live fragment (usually a branch), loose in rubble pile or on reef, obvious fresh white break. These should be tallied during mapping but do not need to be mapped or measured. Note that occasionally loose fragments land on living tissue, these should not be considered separate fragments, but rather part of the colony on which they have landed.

POST SURVEY TASKS

1) Download and rename photo files as described above under 'Photographing Colonies'; organize files by colony (i.e., one folder per colony).
2) It is worthwhile to compare the previous survey photo of each colony along with the most recent to verify that the colony was correctly identified and that the colony condition was accurately recorded.
3) Make a contact sheet with one picture of each colony to assist locating colonies on the next survey. Many image processing software programs will do this automatically!.

QUARTERLY SURVEYS

Subsequent surveys should ideally occur quarterly (approximately every three months) or sooner in the case of a known disturbance event (e.g. mass bleaching or hurricane). Tasks 5 and 6 are repeated during quarterly surveys. However both Diver A and B will collect data since Diver B is not occupied with colony selection. Diver A will use the 'Quarterly colony condition assessment' field sheet (Fig. 55) to record colony condition observations. Diver B will locate the tags and clear them so that the number can be read. Then Diver B will count the snails and damselfish using the 'Quarterly snail and fish census' field sheet (Fig. 55). If an additional diver is available to participate, it would be desirable for that person to perform branch extension measurements/photos (see optional tasks).

The small aluminum tags can be difficult to locate, even after only 3 months. Diver B should be prepared to scrape turf algae or chip CCA from the tags so that they can be read. In case some tags can not be found, contact sheets made prior the survey can greatly improve the chances of relocating a colony. Furthermore, in

cases of ambiguous colony boundaries, these photos can improve the reliability of colony measurements.

OPTIONAL TASKS

LINEAR BRANCH EXTENSION

Overall colony dimensions and % live tissue give one measure of how much colonies (or total live area) are changing over time (growing or shrinking), but these measures are imprecise. We would also like to measure the rate of branch extension as a different, more precise measure of colony growth. This is also a measurement that has been used in previous published studies of *Acropora palmata* ecology and physiology and so can be compared to historical (e.g. pre-die-off conditions).

Materials & Methods

1) Small, thin, beaded cable ties, preferably at least two colors (e.g., http://www.coleparmer.com/catalog/product_view.asp?sku=0683050)
2) Small, cm-scale ruler
3) Digital underwater camera

We have been marking ~ 4 branches per colony and will use these measurements to calculate the average linear extension for the colony. A cable-tie loop is placed tightly around (preferably) each of these four branches to form a benchmark. The small (~5 in), flexible beaded cable ties are best for this application as they are less obtrusive and the beaded flexible shaft of the tie will nestle and "grip" amongst the individual corallites on the branch whereas heftier cable ties would crush more corallites in order to get a firm fit. More than one cable tie can be linked together if needed. It is important that the cable tie be tight and not move since its location will be used to gage branch length. Place the cable tie fairly close to the branch tip (1-2 cm) to improve the resolution of the length measurements. Note that this cable tie will likely be completely overgrown within ~6 months so sequential cable ties will need to be used to monitor branch growth over time.

We have devised a designation system with the branch nearest the plot stake being designated "A", and then working clockwise around the colony designating branches "B", "C", and "D". A top down photo is taken of the whole colony to facilitate locating the designated branches on future surveys. The small cable ties are visible in a full colony photo at the initial placement, but may be overgrown by

coral tissue on subsequent surveys, so the full colony photo can be used to focus ones' search for the small, loose tail of the cable tie.

At the initial marking and at subsequent surveys, the maximum distance from the cable tie to the branch tip should be recorded. Depending on your capacity (field vs. lab time), this can be done in the field (most effectively using a transparent ruler), or from measuring close-up digital photos of each branch tip, including an opaque ruler next to the branch in the photo. Linear extension (growth) is calculated as the difference in length between subsequent surveys.

GENOTYPE SAMPLING

Acropora spp. are highly clonal organisms. That is, many colonies originate from fragments of mother colonies (i.e. branch that breaks off and re-attaches to the reef to form a new colony) rather than from sexual larvae. Hence, many colonies within a site may be genetically identical. We hope to learn two things from genotyping the monitored colonies; 1) the genotype of a colony may have a strong influence on colony characteristics and performance (e.g. growth rate, disease susceptibility) that we are monitoring and we want to understand this relationship better, 2) we want to know relatively how many genotypes are present in each plot as this gives an indication of the relative success of larval recruitment in that population.

Note: The tissue sampling described here requires extensive permitting, special shipping considerations, and specialized laboratory analysis. Please contact SEFSC for advice and coordination prior to undertaking genotype tissue sampling.

Materials & Methods

1) Snipping tool (e.g. garden pruners or wire cutters and/or small chisel (1/4 inch) and hammer
2) Small plastic bags with labels (e.g. 60ml size Whirl-pak)
3) Cryo-vials 2 ml for sample storage
4) Ethanol (non-denatured > 95%) for sample preservation
5) Rite-in-the-Rain paper (http://www.riteintherain.com/ and search for 'grid sheets') for interior labels in EtOH. (Note that plastic types waterproof paper such as Dura-Copy will dissolve in ethanol!)

The goal is to obtain a very small amount of live *Acropora spp.* tissue with the minimum harm/disturbance to the colony. As little as 0.5 cm^2 of tissue is adequate to extract enough DNA for the analysis. We have found the most precise way to obtain such small samples of *A. palmata* is using a flat-mouthed snipper (as you

would use for snipping off nail heads) to snip a tiny bit of branch tip or the smallest size of wood-chisel (1/4 inch) to chip a small piece of tissue/skeleton from the encrusting base of a branchless colony. Snipping branch tips is probably more feasible for *A. cervicornis* since they are less likely to have an encrusting base. Place this small sample in a plastic bag in the field with a label with the colony number. You can either pre-label the bags, or include a small label inside the bag. On the surface or upon return to land, the sample should be transferred from the bag into a small cryo-vial (or larger sample jar if needed). Forceps can be helpful in this step and the sample can be further fragmented if needed to fit into the small vials. Construct a small label on the Rite-in-the-Rain paper with pencil to fit inside the vial and include the colony number, site/plot number, and date. It is also good to label the outside of the vial with the same information but this is not sufficient as the ethanol can dissolve permanent marker. Inside labels are NECESSARY.

If possible, it is good to keep the samples in a freezer (-80°C freezer is ideal) until shipment and analysis can be arranged. Again, prior arrangements need to be made for shipping and analysis of these samples.

REFERENCES

Acropora Biological Review Team (2005) Atlantic *Acropora* Status Review Document. Report to National Marine Fisheries Service, Southeast Regional Office.

Aronson RB, Precht WF (2001) White-band disease and the changing face of Caribbean coral reefs. Hydrobiologia 460:25-38

Bruckner AW (2003) Field Guide to Western Atlantic Coral Diseases and Other Causes of Coral Mortality. In: E. Corcoran EG (ed). UNEP-WCMC, Marine and Coastal Programme, Cambridge, UK

Bruckner AW (2002) Proceedings of the Caribbean *Acropora* Workshop potential application of the U.S. Endangered Species Act as a conservation strategy: April 16-18, 2002, Miami, Florida NOAA Technical Memorandum NMFS-OPR-24, Silver Spring, MD, 184 pp.

Bruckner AW, Bruckner RJ (1997) Outbreak of coral disease in Puerto Rico. Coral Reefs 16:260-260

Bruckner AW, Corcoran E, Green E, O'Caroll J (2003) Field guide to western Atlantic coral diseases and other causes of coral mortality. UNEP World Conservation Monitoring Centre; Marine and Coastal Programme, Cambridge, UK

Croquer A, Bastidas C, Lipscomp D, Rodriguez-Martinez RE, Jordan-Dahlgren E, Guzman HM (2006) First report of folliculinid cilliates affecting Caribbean scleractinian corals. Coral Reefs 25:187-191

Fong P, Lirman D (1995) Hurricanes cause population expansion of the branching coral *Acropora palmata* (Scleractinia): Wound healing and growth patterns of asexual recruits. Marine Ecology 16:317-335

Frydl P, Phillips RC, McRoy CP (1979) The effect of parrotfish (Scaridae) on coral in Barbados, W.I. Int. Rev. Gesamt. Hydrobiol. 64(6):737-748

Gateno D, Leon A, Barki Y, Cortes J, Rinkevich B (2003) Skeletal tumor formations in the massive coral *Pavona clavus*. Marine Ecology-Progress Series 258:97-108

Gladfelter WB (1982) White-Band Disease in *Acropora palmata* : Implications for the Structure and Growth of Shallow Reefs. Bulletin of Marine Science 32:639-643

Goreau TF (1959) The ecology of Jamaican coral reefs: I. species composition and zonation. Ecology 40:67-90

Humann P, DeLoach N (2002a) Reef Coral Identification : Florida, Caribbean, Bahamas : including marine plants. New World Publications, Jacksonville, Fla

Humann P, DeLoach N (2002b) Reef Fish Identification : Florida, Caribbean, Bahamas. New World Publications, Jacksonville, Fla

Lirman D (2000) Lesion regeneration in the branching coral *Acropora palmata*: effects of colonization, colony size, lesion size, and lesion shape. Marine Ecology-Progress Series 197:209-215

Lirman D (1999) Reef fish communities associated with *Acropora palmata*: Relationships to benthic attributes. Bulletin of Marine Science 65:235-252

Miller MW (2001) Corallivorous snail removal: evaluation of impact on *Acropora palmata*. Coral Reefs 19:293-295

Miller MW, Baums IB, Williams DE, Szmant AM (2002) Status of Candidate coral, *Acropora palmata*, and its snail predator in the upper Florida Keys National Marine Sanctuary: 1998-2001. NOAA Technical Memorandum, NMFS-SEFSC-479, Miami, FL, 26 pp. 26 pp

NMFS (2006) Endangered and Threatened Species: Final Listing Determinations for Elkhorn Coral and Staghorn Coral. Federal Register 71:26852-26861

Patterson KL, Porter JW, Ritchie KE, Polson SW, Mueller E, Peters EC, Santavy DL, Smiths GW (2002) The etiology of white pox, a lethal disease of the Caribbean elkhorn coral, *Acropora palmata*. Proceedings Of The National Academy Of Sciences USA 99:8725-8730

Peters EC, Halas JC, McCarty HB (1986) Calicoblastic neoplasms in *Acropora palmata* , with a review of reports on anomalies of growth and form in corals. Journal of the National Cancer Institute 76:895-912

Porter JW, Dustan P, Jaap WC, Patterson KL, Kosmynin V, Meier OW, Patterson ME, Parsons M (2001) Patterns of spread of coral disease in the Florida Keys. Hydrobiologia 460:1-24

Robertson DR, Hoffman SG, Sheldon JM (1981) Availability of Space for the Territorial Caribbean Damselfish *Eupomacentrus planifrons*. Ecology 62:1162-1169

Rodriguez-Martinez RE, Banaszak AT, Jordan-Dahlgren E (2001) Necrotic patches affect *Acropora palmata* (Scleractinia : acroporidae) in the Mexican Caribbean. Diseases Of Aquatic Organisms 47:229-234

Shinn EA (1989) Reefs of Florida and the Dry Tortugas : Miami to Key West, Florida, July 2-7, 1989. American Geophysical Union:45 pp.

Weil E, Hernández-Delgado EA, Bruckner AW (2002) Distribution and Status of Acroporid Coral (Scleractinia) Populations in Puerto Rico. In: Bruckner AW (ed) Proceedings of the Caribbean *Acropora* Workshop potential application of the U.S. Endangered Species Act as a conservation strategy: April 16-18, 2002, Miami, Florida. NOAA Technical Memorandum NMFS-OPR-24, Silver Spring, MD, pp 67-94

Weil E, Losada F, Bone D (1984) Spatial variations in density and size of the echinoid *Diadema antillarum* Philippi on some Venezuelan coral reefs. Bijdragen tot de Dierkunde 54:73-82

Williams DE, Miller MW (2006) Importance of disease & predation to the growth & survivorship of juvenile *Acropora palmata* & *Acropora cervicornis*: A demographic approach. Proc 10th Int Coral Reef Symp:1096-1104

GLOSSARY

active (tissue loss) - describes disease lesions that are believed to be actively progressing based on exposed skeleton with no algal colonization and frequently irregular jagged lesion margins.

attachment point - tissue & skeletal growth formed when a fragment fuses with the substrate. Fragments attaching to the substrate frequently produce several attachment points.

base - portion of the colony encrusting the substrate

bleached - Coral tissue that is lacking zooxanthellae- Tissue appears translucent but is intact.

calcareous coralline algae (CCA) - light pink to burgundy encrusting algae with a calcified thallus also known as 'pink pavement'.

chimney - Growths of *Acropora* spp. tissue resulting from threespot damselfish bites. Repeated bites during tissue healing result in upward growth of tissue and skeleton which are frequently toped with filamentous algae.

colony - Operationally defined as an area of continuous tissue OR disconnected patches of tissue found on the same underlying skeletal structure (presumed to be formerly continuous).

corallite - cup shaped calcium carbonate structure secreted by and containing a single polyp

filamentous algae -– Fast-growing algal forms that quickly colonize 'new' reef surfaces such as newly dead coral skeleton. The intensity of filamentous algal growth can be used to gauge the approximate age or rate of progression of an area of recent mortality

fragment - broken portions of standing colonies found loose on the surrounding substrate. Once the fragment attaches to the substrate it can be considered (here) an 'asexual recruit'.

genet - the genetic individual resulting from a single planula (zygote). For clonal organisms such as *Acropora* a genet often includes numerous ramets (asexually produced individuals). Determining if colonies belong to the same genet requires genotyping in the lab and can only be guessed based on observations in the field.

healing – re-growth of healthy tissue over a former lesion or scar.

isolate - physiologically continuous patch of tissue regardless of size or shape.

lesion - Area of recently exposed skeleton that is presumed to result from disease

margin - boundary between live tissue and adjacent substrate (including *Acropora* skeleton/dead areas)

polyp - single unit of the coral colony contained within a corallite.

ramet - products of asexual reproduction by a given ramet genet.

recent (tissue loss) - Exposed Acropora skeleton with little or no colonization by filamentous algae and relatively sharp, observable corallite structure.

recruit - smaller colonies (<20cm) are frequently described in the literature as 'recruits' since they are presumed to have appeared recently. In this study a recruit is a colony that as appears in the study plot only after the initial survey. It includes both fragments that have re-attached to the substrate (asexual recruits) and new colonies that appear from settled planulae (sexual recruits).

remnant - describes patch(es) of tissue (isolates) that result from a larger colony that lost substantial tissue leaving only 1 or a few disjointed patches of tissue. Typically describes patches that have grown new branches since they were isolated.

snails - In the context of this protocol, the term 'snails' refers only to those found to prey on *Acropora*, chiefly *Coralliophila abbreviata* but also *Coralliophila caribbaea* where present.

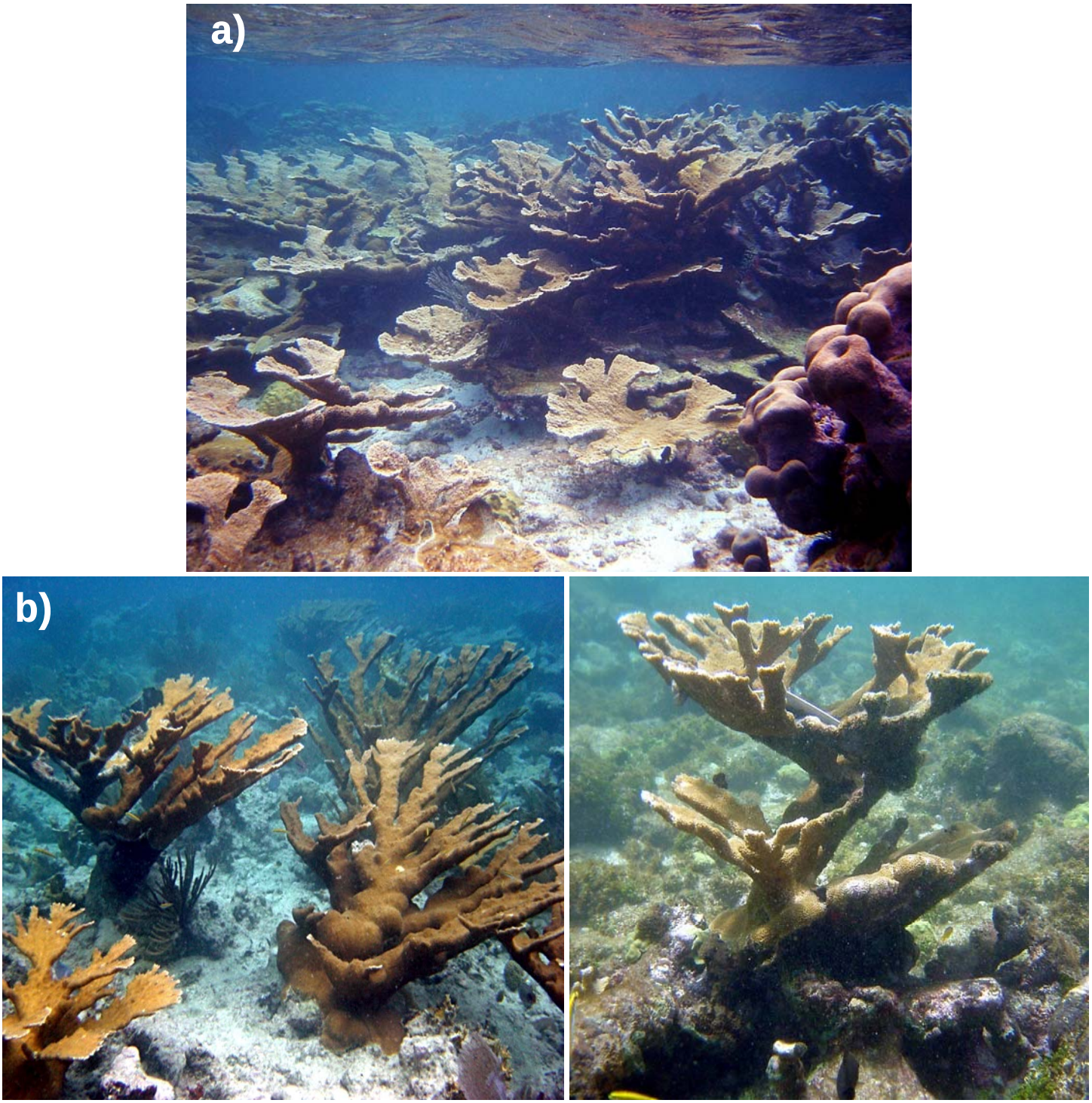


Figure 1. a) While dense monospecific patches or "thickets" are characteristic of *Acropora palmata,* these complex structures are not conducive to the 'demographic' monitoring methods described here. 'Demographic' monitoring tracks the performance of individual colonies (b-c) over time. If colonies are densely spaced in a small area it becomes difficult to discern individuals from their neighboring colonies. There are distinct boundaries separating individual 'colonies' (b-c).

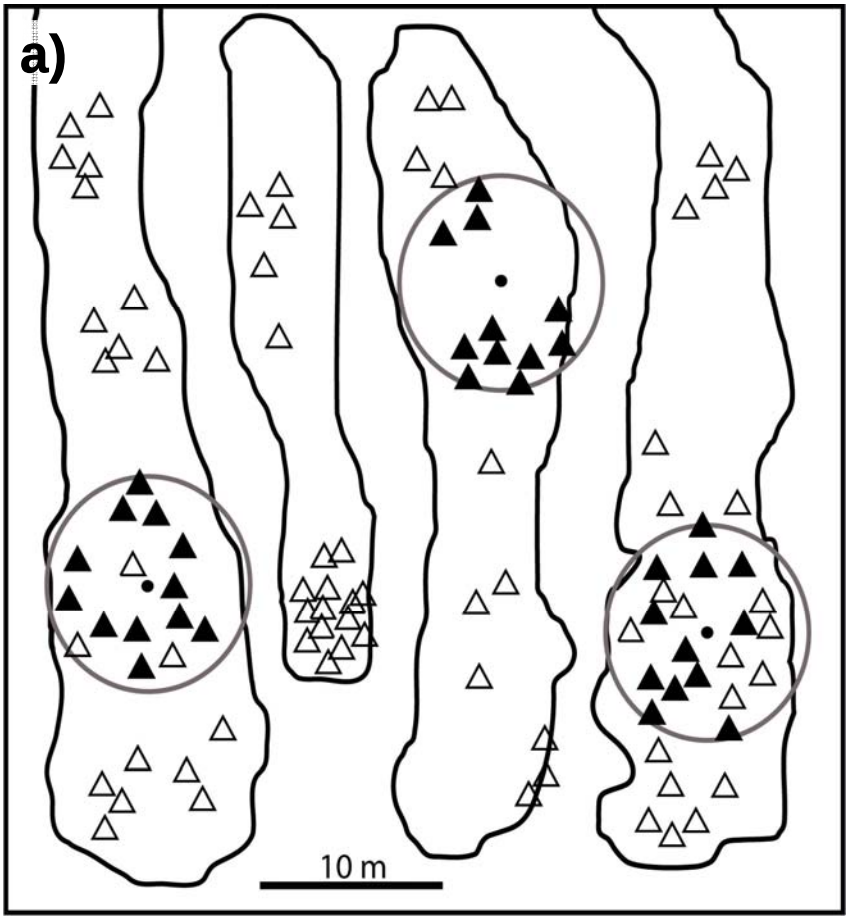
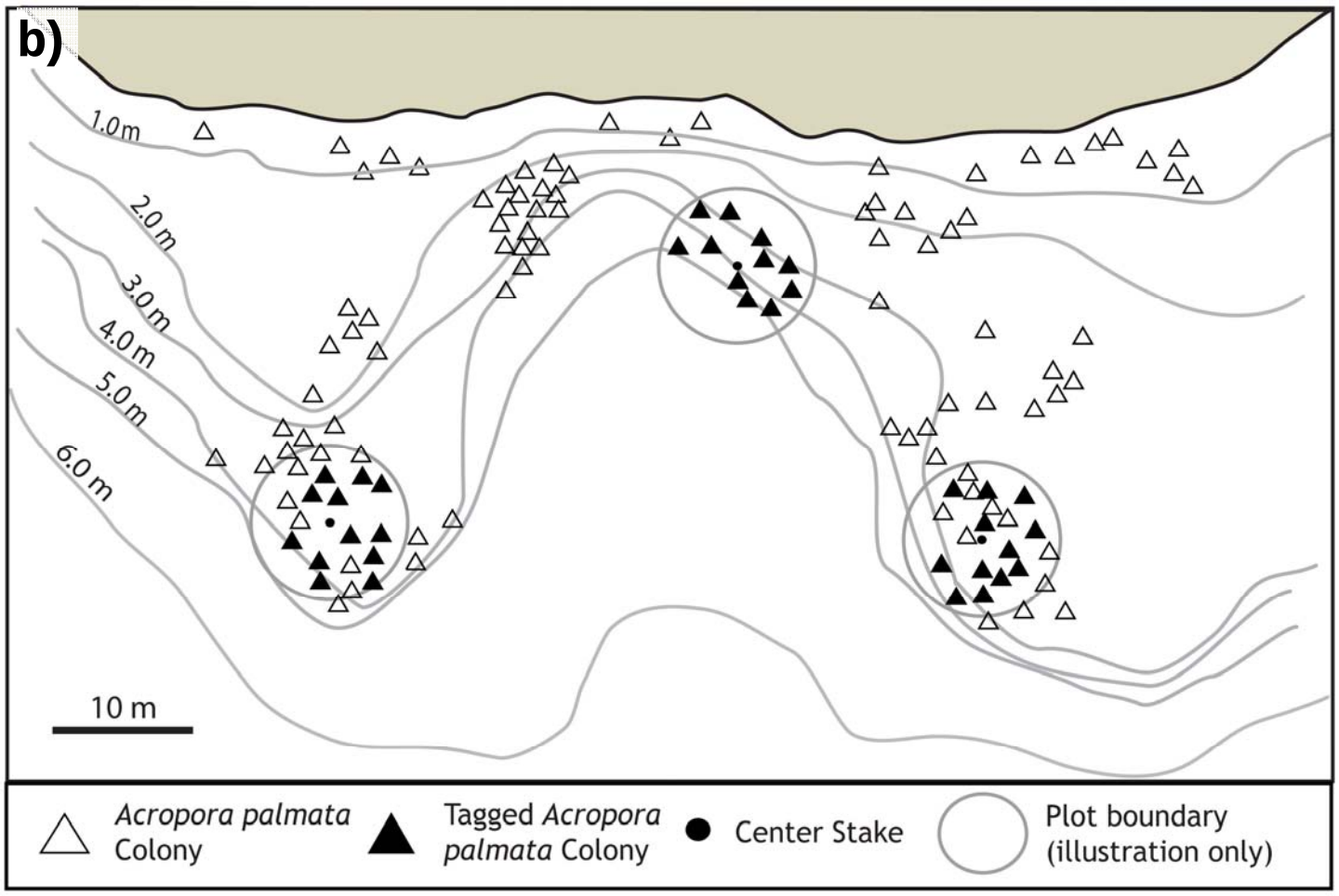


Figure 2. Schematic illustration of plot placement. a) an example based on a spur and groove type reef structure and b) from a fringing type reef. Acropora palmata colonies are represented by (Δ) and (▲). Shaded triangles represent those that would be selected for tagging using randomly generated compass bearings and distances from the central plot stake (●). There are a total 35 colonies selected within the three plots. Tagged colonies in the plots on the far left and far right in both frames were selected randomly. All colonies in the center plot of both frames were tagged because there are less than 12 colonies within the plot area.



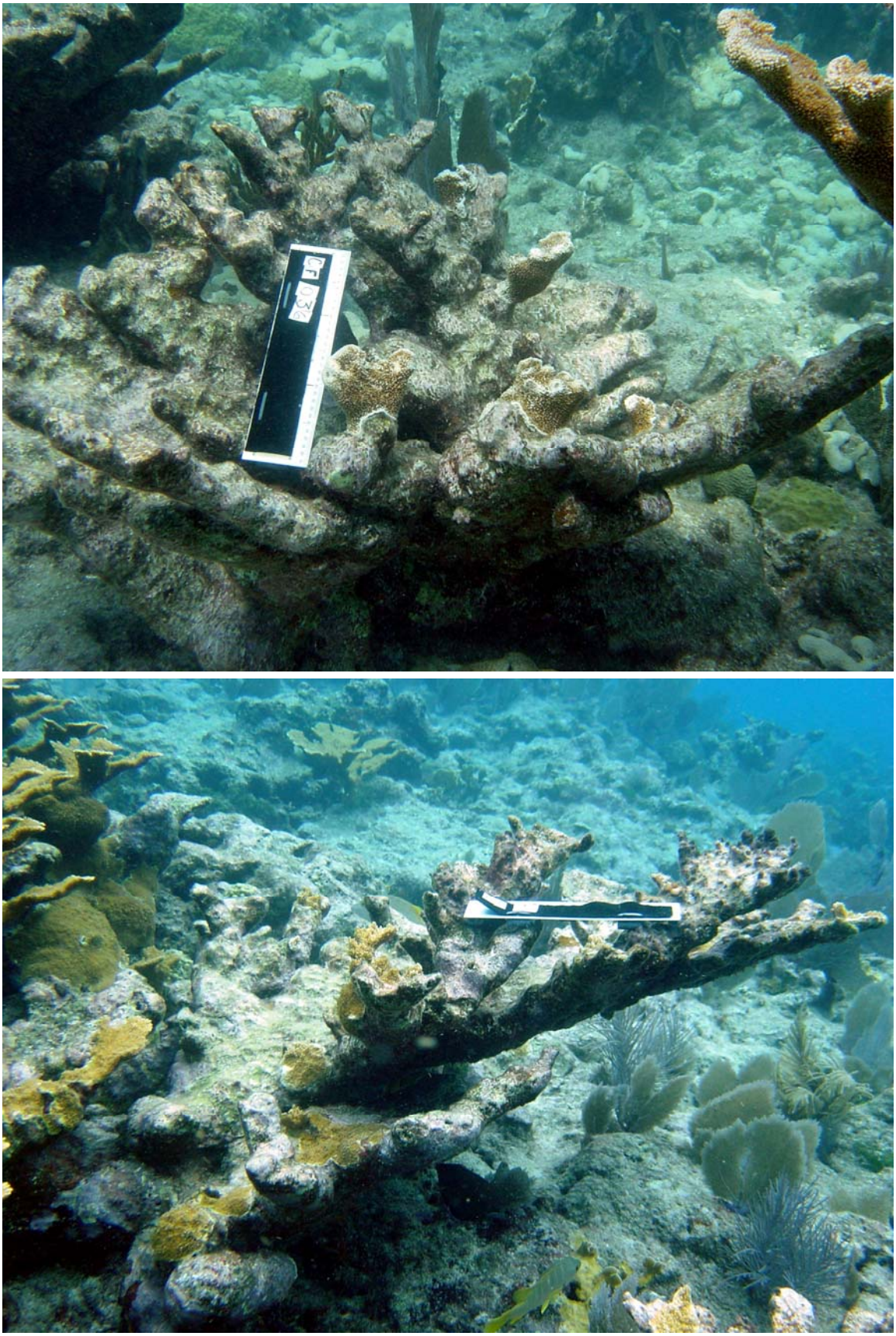


Figure 3. Remnant patches of live tissue on large colony skeletons represent challenges in designating what is an individual colony for monitoring. This designation is crucial to tracking changes in the colonies and recruitment so it is important to be consistent over time. Guidelines are presented in the text to help the surveyor make consistent decisions on the boundaries of a colony.

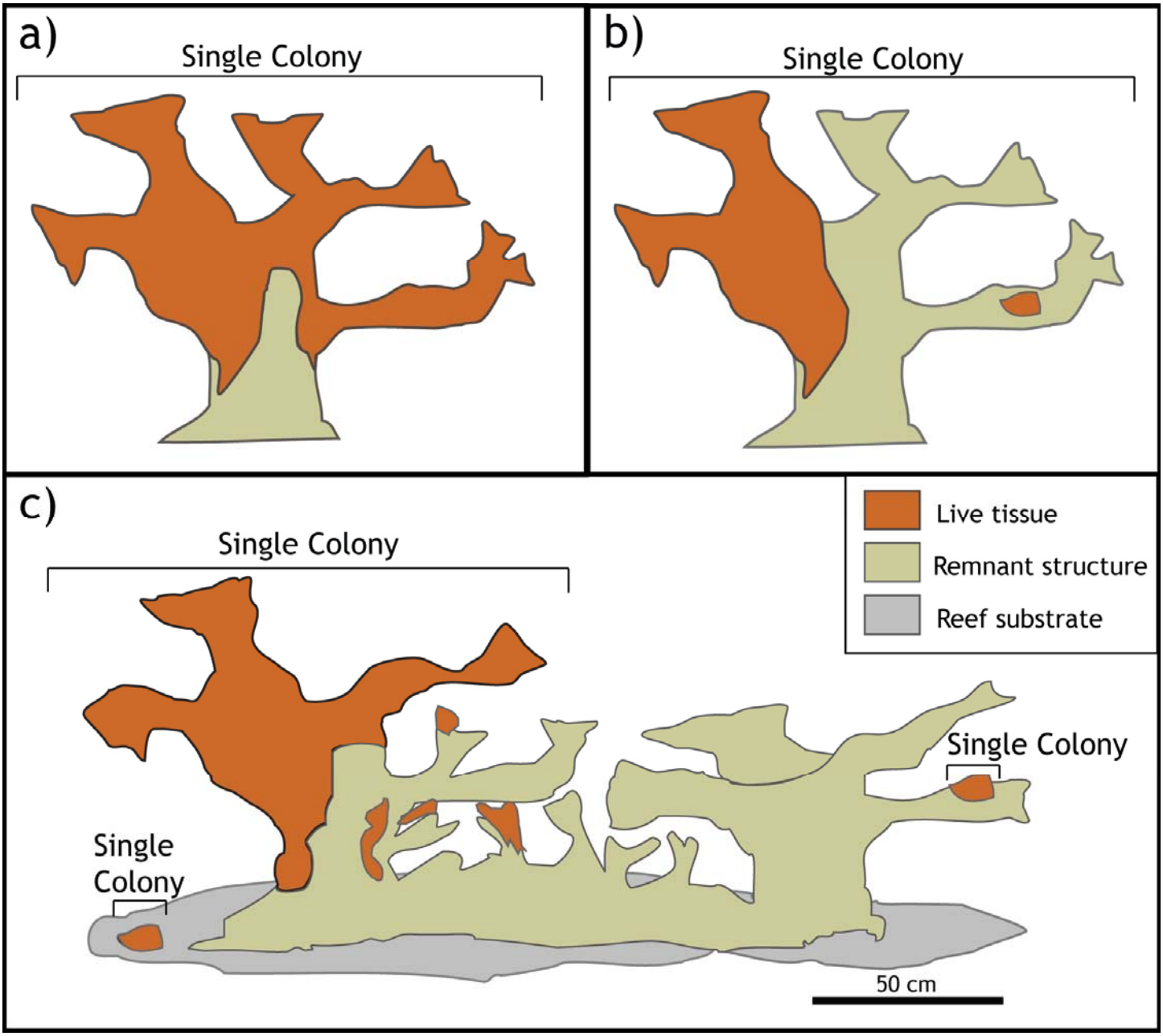


Figure 4. Illustration of three rules used to distinguish colonies. In (a) live tissue is clearly continuous. In (b) live tissue is not physically connected, but the two patches of live tissue are on the same remnant structure, so the two patches should be considered the same colony. In (c), the three regions of live tissue should be considered separate colonies. Some ambiguity exists in distinguishing whether the remnant/dead structures of the remaining tissue patches are related, and because the tissue patch on the right is more than 1 meter from the larger tissue patch on the left, these two patches should be considered separate colonies. The small patch of live tissue on the lower left is within one meter of the larger live patch on the left, but is clearly not on the same remnant/dead structure, so should therefore be considered a separate colony.

Diver B Tagged Colony Selection

Site_____ Date_____

Stake Depth: _____

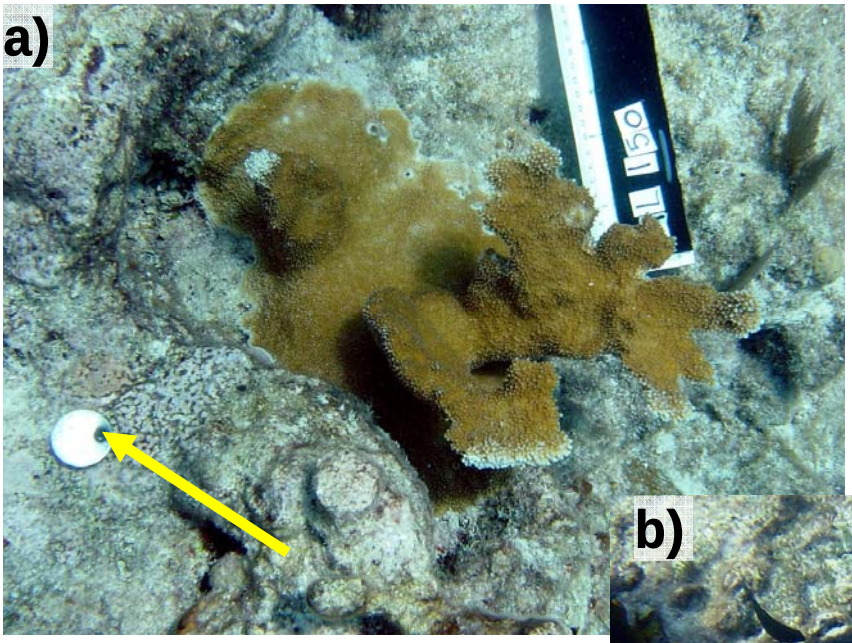
Stake Waypoint#

Plot	Colony	Tag #	Deg	Dist	Notes (tag loc., depth, etc)
1	1				
1	2				
1	3				
1	4				
1	5				
1	6				

Random Coordinates:

Plot 1		Plot 2	
Deg	**Dist**	**Deg**	**Dist**
270	6.5	190	7.0
145	4.0	320	3.0
125	4.5	260	2.5
95	0.5	65	2.5
315	0.5	115	3.0
285	7.0	175	3.5
210	1.5	10	5.0
50	6.0	275	3.5

Figure 5. Field sheet used by Diver B to select the colonies to be tagged. On the right are sets of randomly generated coordinates that are followed by Diver B. If the center of a colony is found within 0.5m of this point it should be tagged. Then the actual distance and degrees of the center of the tagged colony should be recorded with the tag number on the left side of the field sheet.



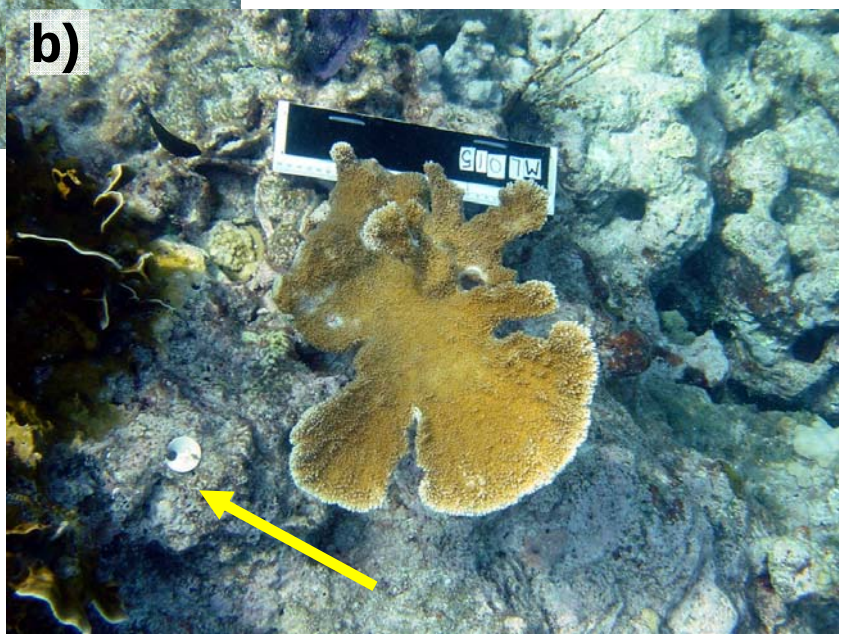
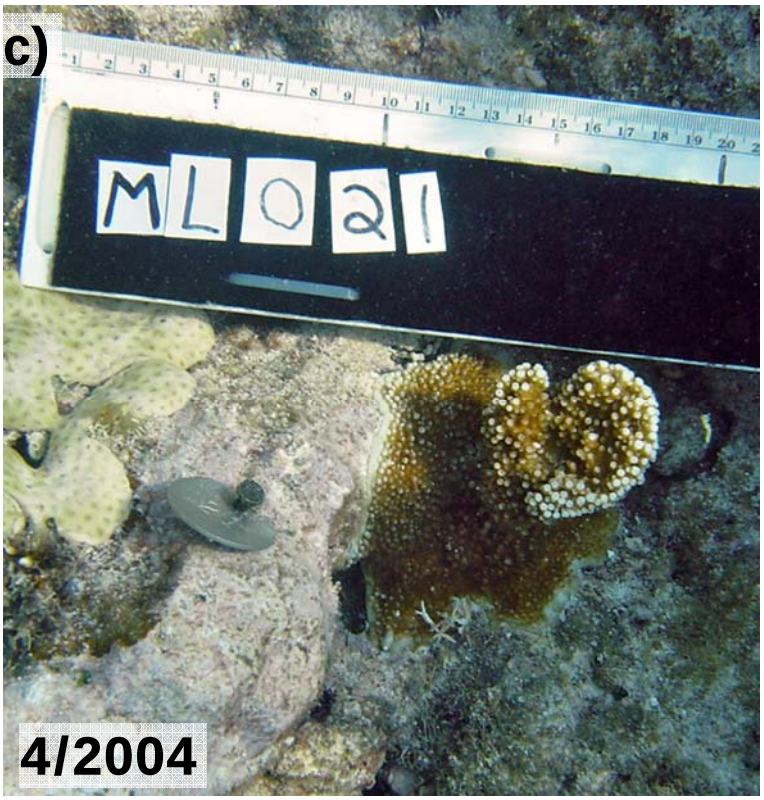
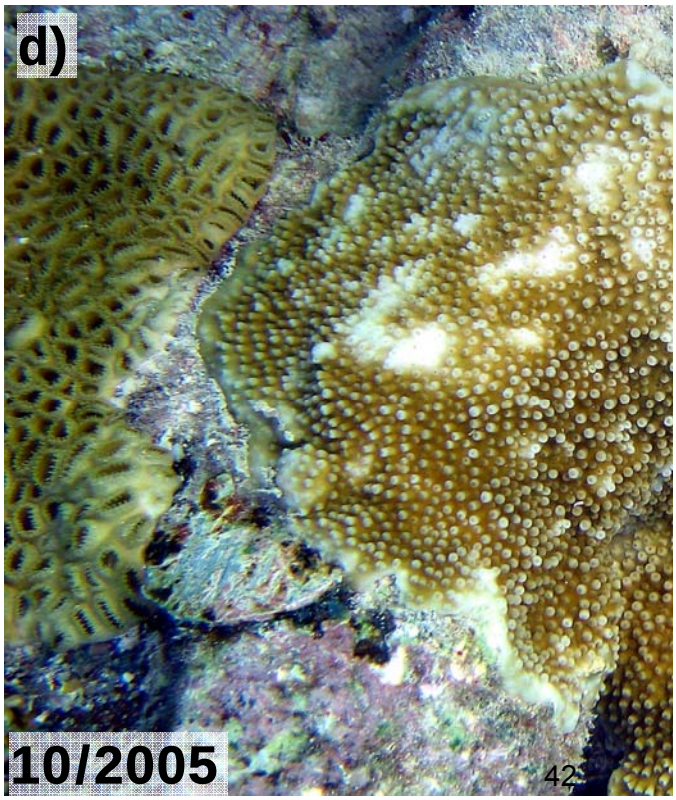


Figure 6. Tags should be nailed to an adjacent area of bare substrate, close enough to clearly identify the tagged colony, but far enough to allow for the tissue around the base to grow. Arrow indicates tag. Tags shown in frames a and b are appropriately spaced. The tag in frame c is too close to the base of the colony, 18 months later the tag is being overgrown by the colony and adjacent *Palythoa caribaeorum* .





a)

Diver A: Initial Colony Survey

Site Horseshoe Reef Plot HS #1 Date 4/20/2006

X	Tag #	L	W	H	% Live	Recent mort*				Bleaching**			Damselfish Impact (Rank)	Skel anoms***	Overgrowth	# Snails	# 3-Spot damsels		# Yellowtail damsels		# Branch Breaks				Notes
						Rank	Types			Rank	Types														
							1°	2°	3°		1°	2°					Ad	Juv	Ad	Juv	sm	med	lg	xlg	
	056	12	9	11	100	0	-	-	-	-	-	-	1	-	-	1	0	0	0	0	0	0	0	0	
	057	175	130	65	70	2	FS	RTL	-	-	-	-	3	-	-	8	2	1	1	0	0	1	0	0	FS contiguous with RTL
	058	70	45	60	20	-	-	-	-	2	R	W	0	DWP	-	0	0	0	1	1	0	0	0	0	
	059	85	60	50	80	2	PFB	-	-	-	-	-	0	-	-	-	-	-	-	-	2	-	-	-	breaks are near intense PFB

Figure 7. Field Sheet used by Diver A to record the initial survey of the colonies to be tagged. The fields are explained in detail in the text. a) Fields have been filled in for 4 colonies at a hypothetical site called Horseshoe Reef. b) note that the codes for several of the fields are listed at the bottom of the field sheet.

b)

Additional Notes:

* **Recent mortality types:** WPx= white pox, WBD= white band, RTL= rapid tissue loss, FS=snail feeding scar, CB=cilliate band, PFB= parrrotfish bites, CL=Cliona lesion, FP=fish poop, Unkn= unknown ** **Bleaching types:** Bl= bleached, R= rings, W= wedgies ***Skeletal anomolies: Hp=hyperplasia, Np=neoplasia, DWP=dense white polyps, Unkn=Unknown

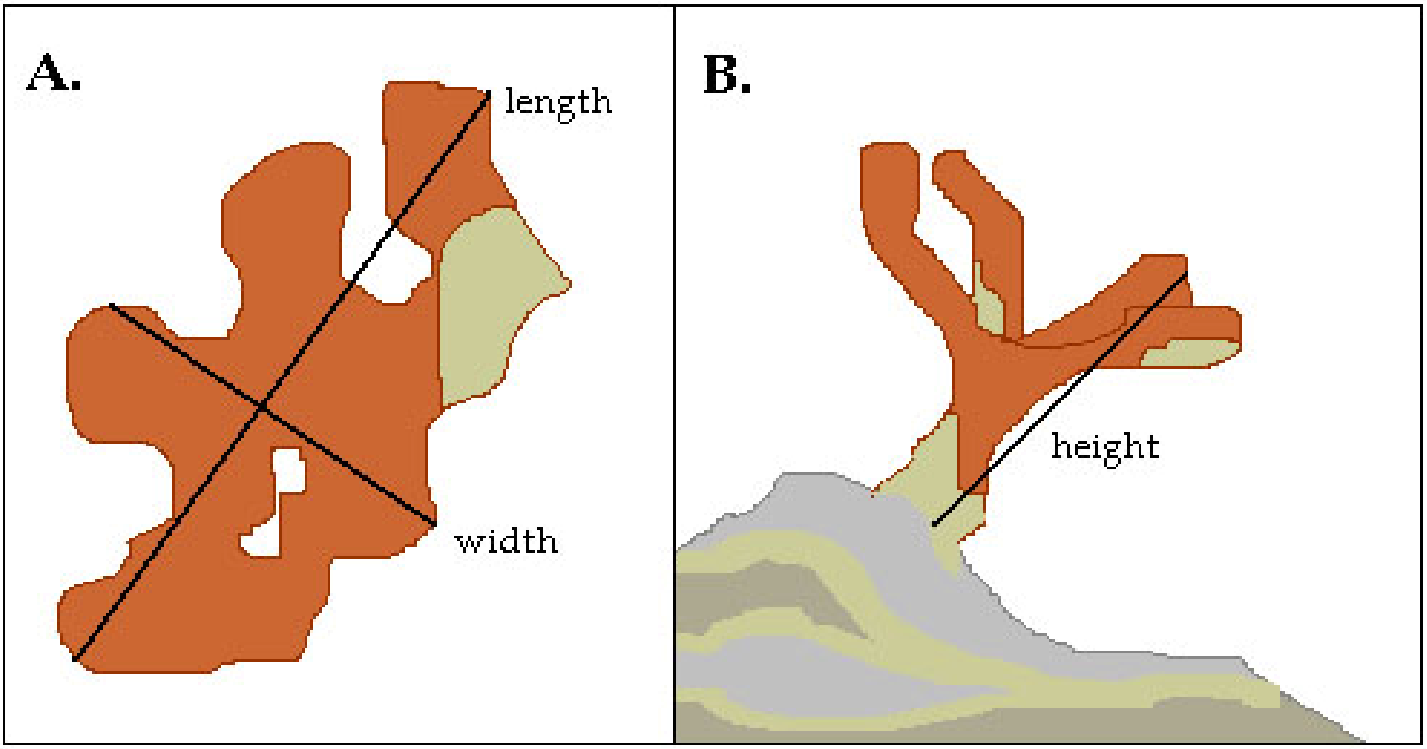


Figure 8. Orientation of colony measurements of A) the maximum length and width, and B) height (perpendicular to the substrate) of a tagged colony.

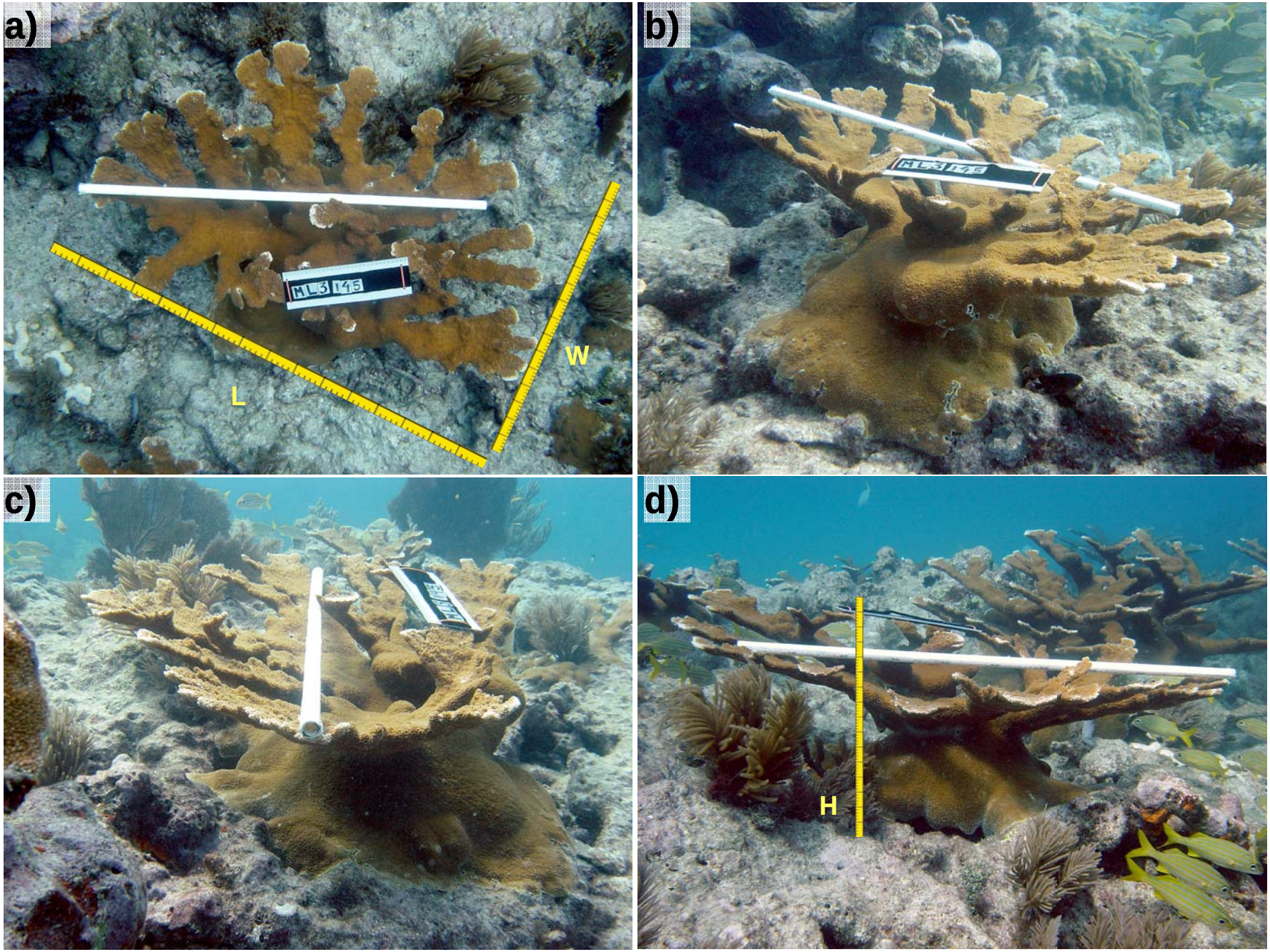


Figure 9. Photos taken of a colony that was assessed as 100% live. All 4 sides are shown to emphasize that all portions of the colony that were included in the LxWxH measurement must be considered when estimating the % live. Frames a and d show the orientation of the length (L), width (W) and Height (H) measurements.

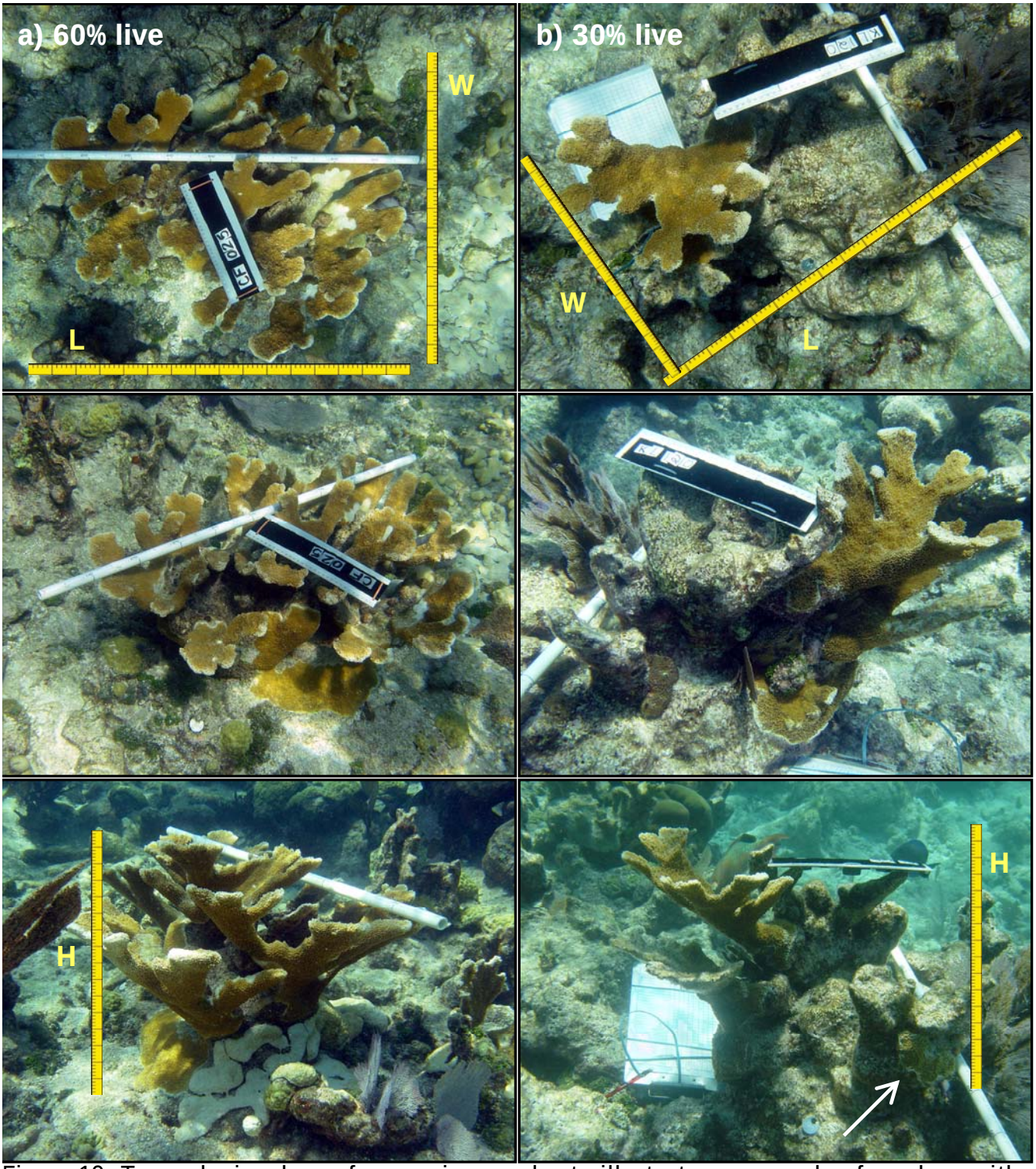
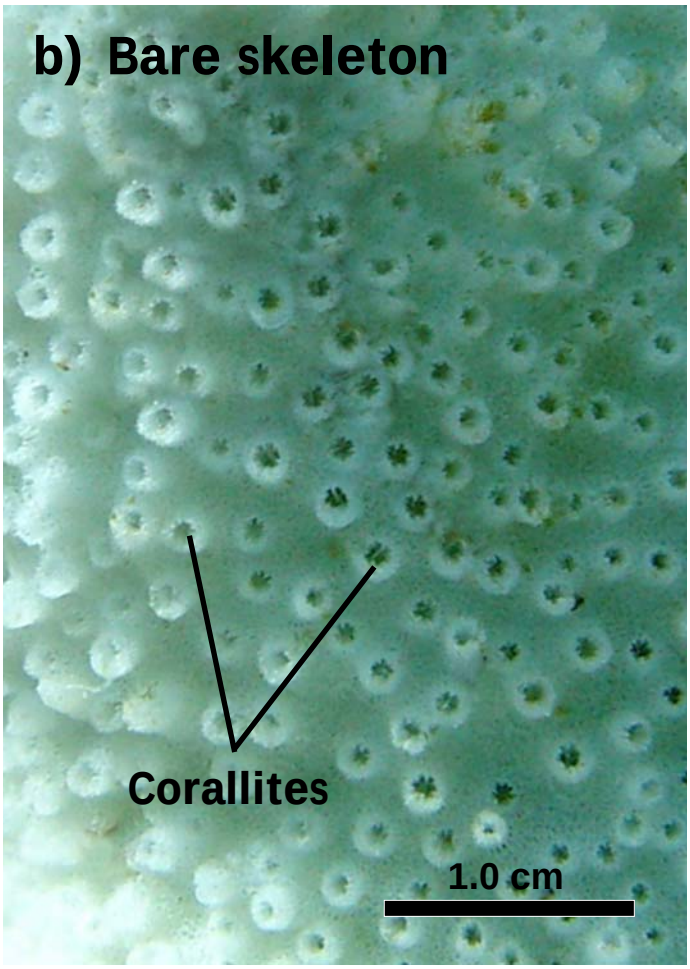
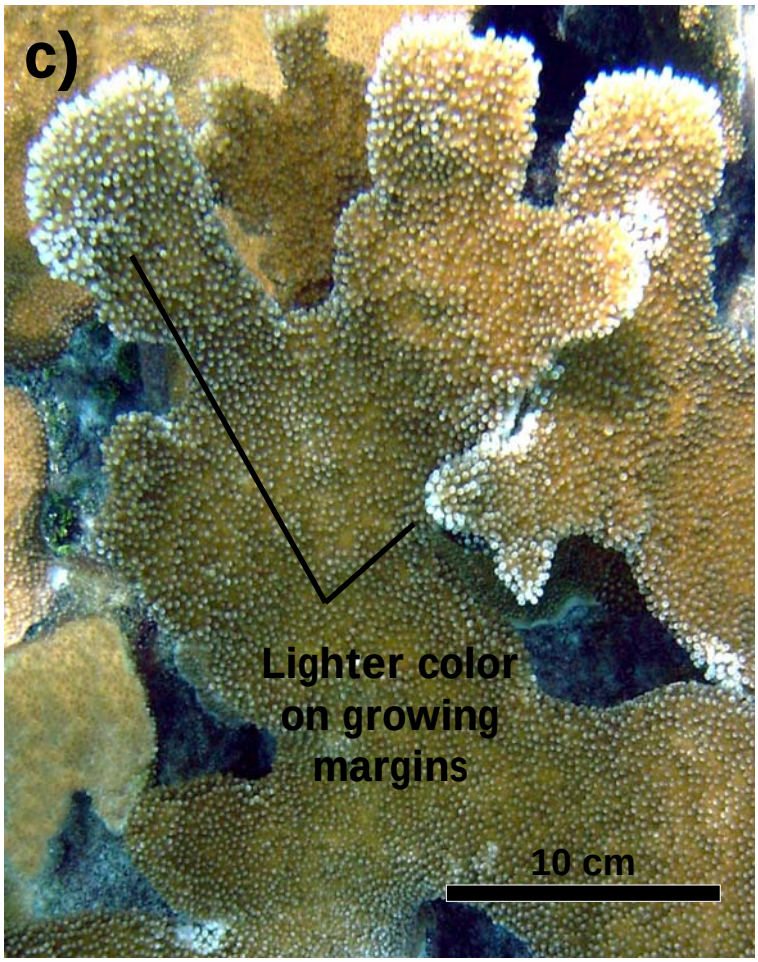
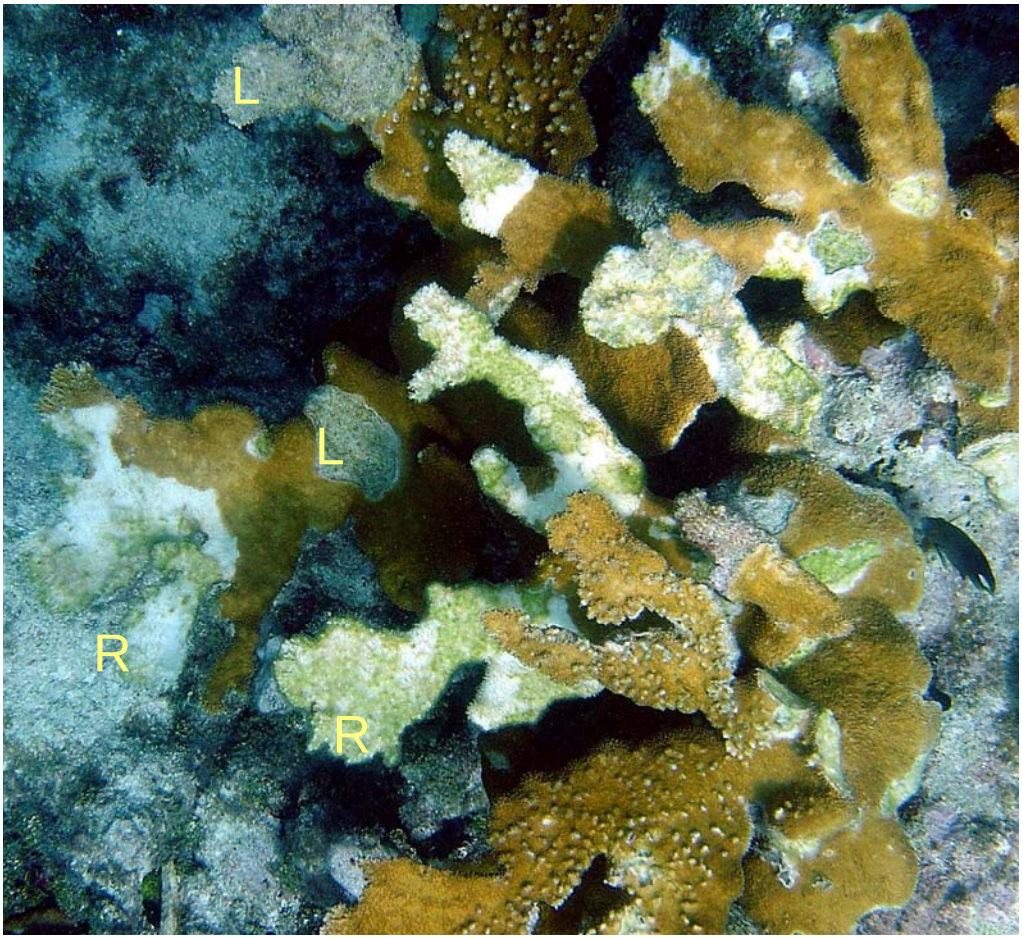


Figure 10. Two colonies shown from various angles to illustrate an example of a colony with an estimated a) 60% coverage of live tissue and b) 30% cover of live tissue. The yellow rulers are superimposed to demonstrate how the dimensions (length (L), Width (W) and height (H) were measured when estimating the %live for that colony. The meter stick (white pole) is used to take measurements in the field and to provide scale in plan view (first frame) images. Note that in 10-b the plan view photo is not directly over the colony making it hard to indicate the length and width of this colony. Furthermore, because two thirds of the colony is dead it is difficult to differentiate the colony margins. The meter stick was placed at the base of the colony to highlight the colony boundary. Note also that Height is measured from the lowest live point (at arrow in bottom frame) rather than extending to the seafloor. Both ways are valid as long as the % live takes this into account.

Figure 11. Healthy *Acropora* colonies are golden to orange brown in color (a). The tissue itself is translucent but harbors zooxanthellae which give it color. The calcium carbonate skeleton is bright white beneath the tissue (b). Colonies are comprised of many individual polyps which each secrete a cup shaped corallite. Polyps are withdrawn into the corallites during the day but often the tentacles protrude slightly above the corallite and can be seen on close inspection. Colony margins, especially branch tips are normally lighter in color where new tissue has grown but has not yet acquired zooxanthellae (c).







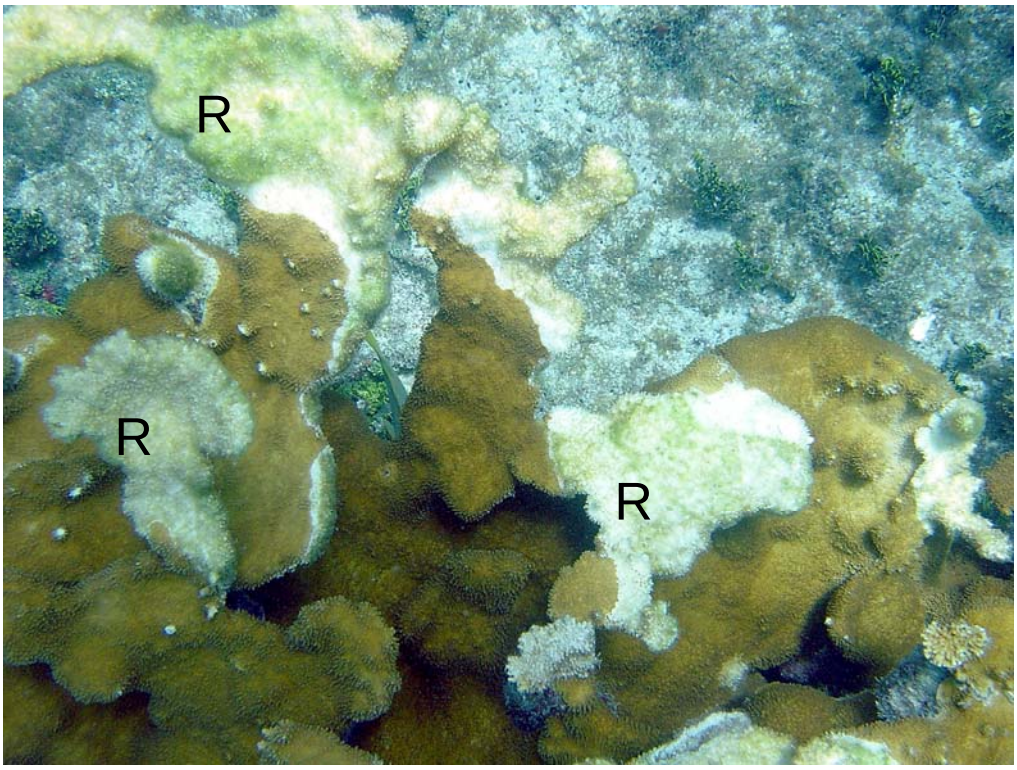


Figure 12. When tissue is lost the bright white coral skeleton is exposed and gradually undergoes succession. In the first few weeks it is possible to roughly distinguish 'recently dead' areas from 'long dead' areas of a colony based on the relative amounts of algal colonization and skeletal erosion. 'Recently dead' (R) areas have light or no algal colonization and the corallite structures are still sharply defined. 'Long dead' (L) areas have eroded corallites and heavier colonization by filamentous algae and/or fleshy or calcareous algae. In general the longer the skeleton has been exposed the darker it appears but it is helpful to look at the corallites (or lack of) to help distinguish recent and long dead.

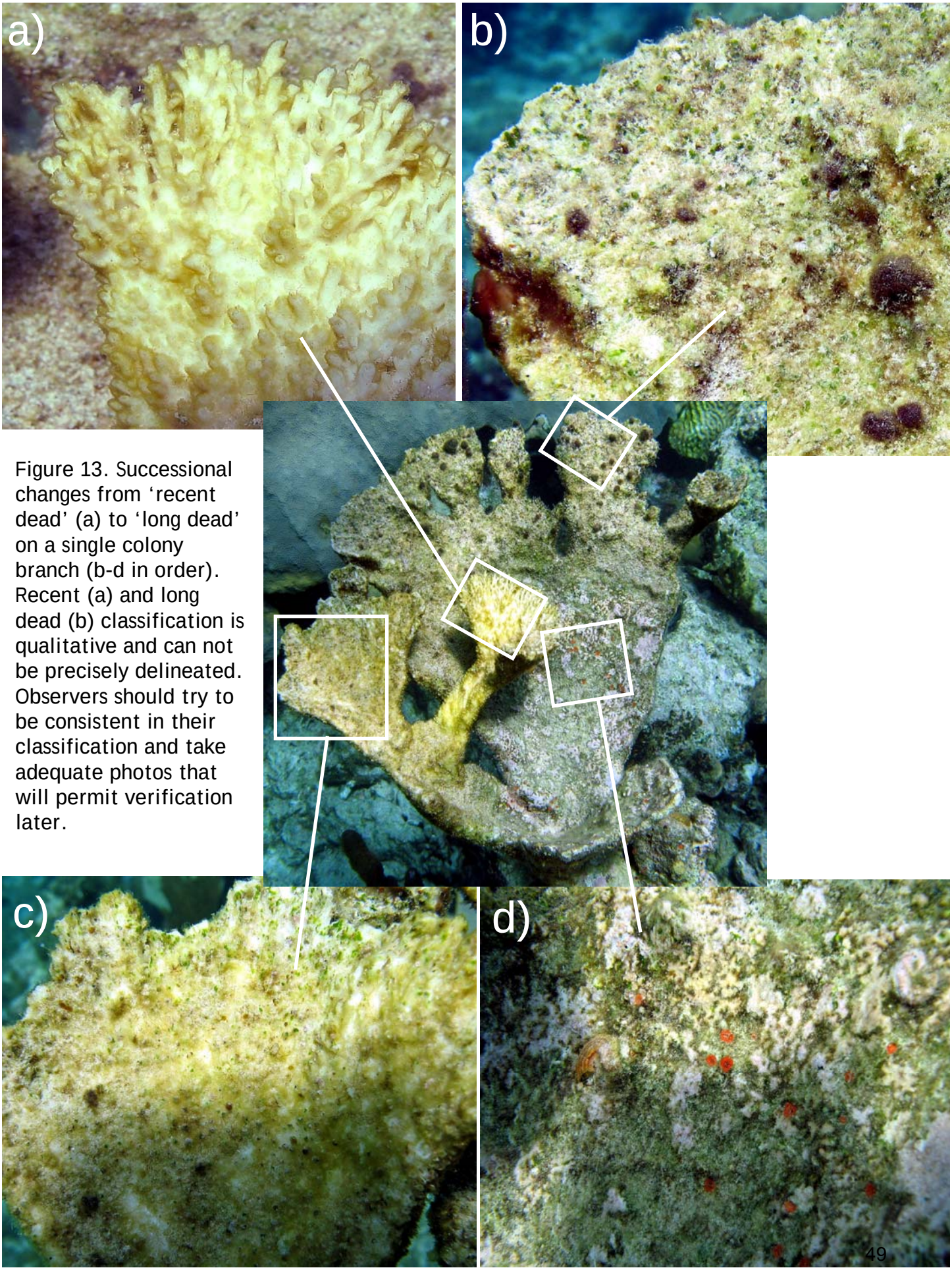


Figure 13. Successional changes from 'recent dead' (a) to 'long dead' on a single colony branch (b-d in order). Recent (a) and long dead (b) classification is qualitative and can not be precisely delineated. Observers should try to be consistent in their classification and take adequate photos that will permit verification later.

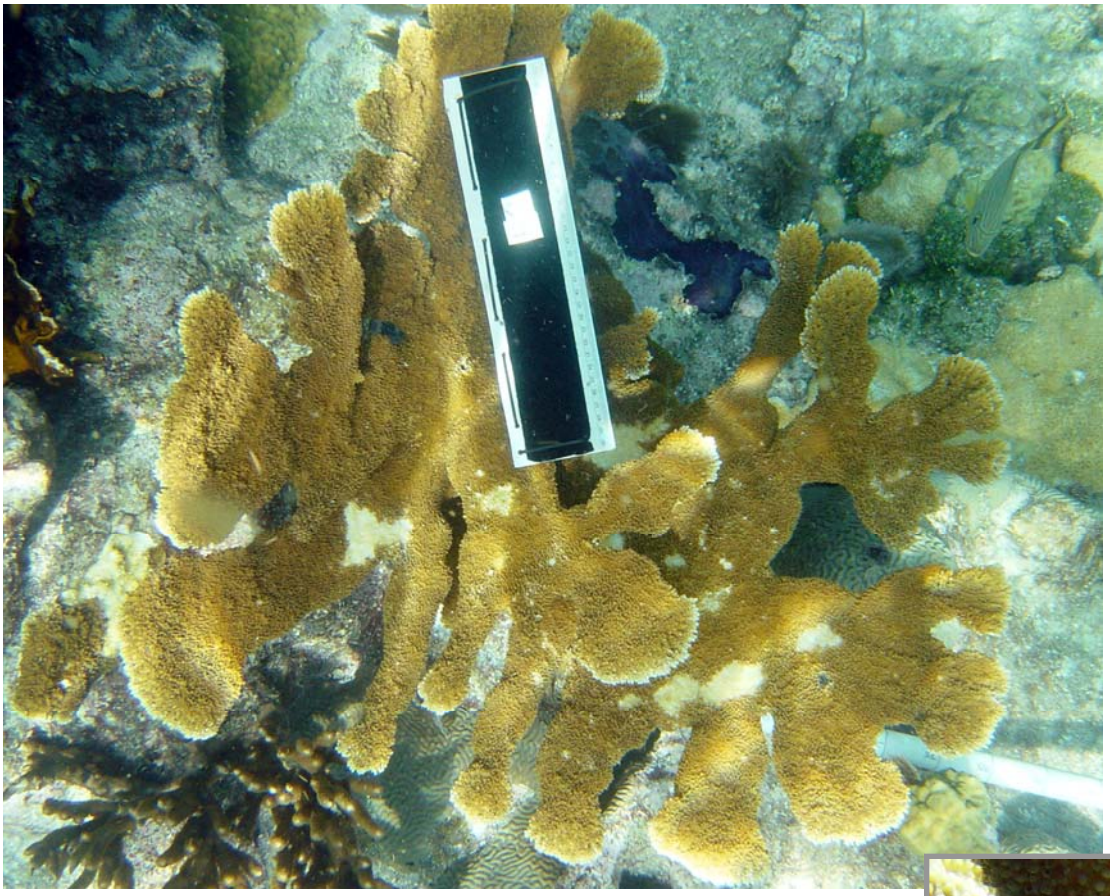
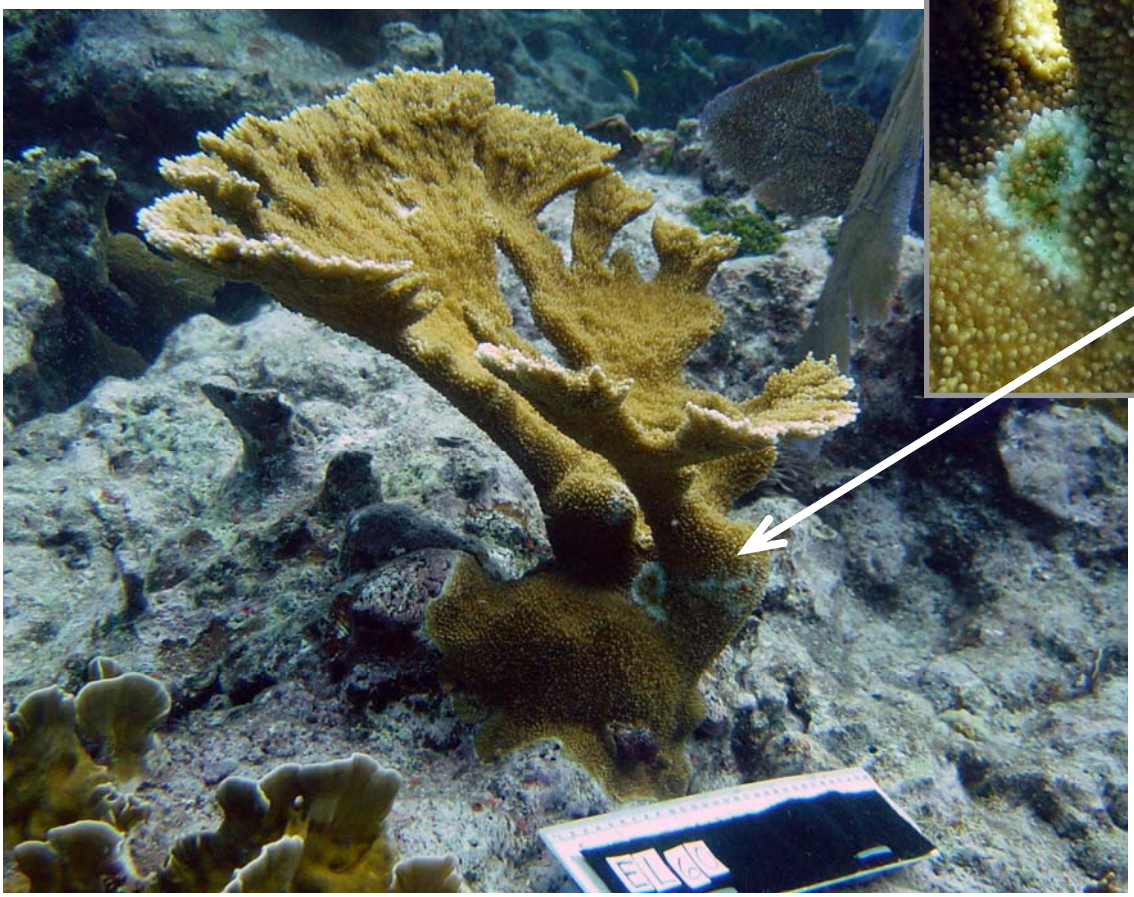


Figure 14. Two examples of colonies with a recent mortality rank= 1. Relative to the size of these colonies the recent mortality is “present but very sparse” (see text).



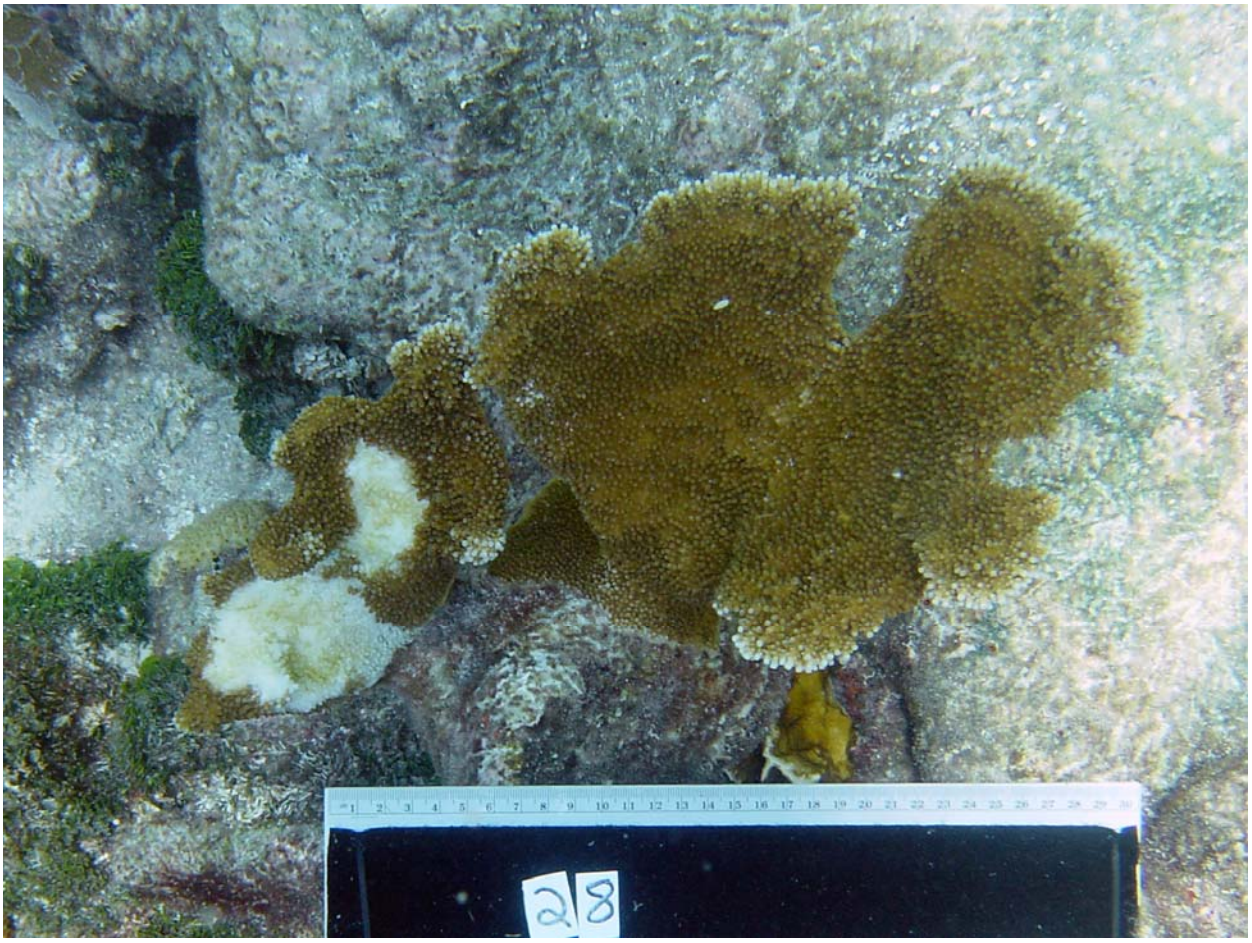
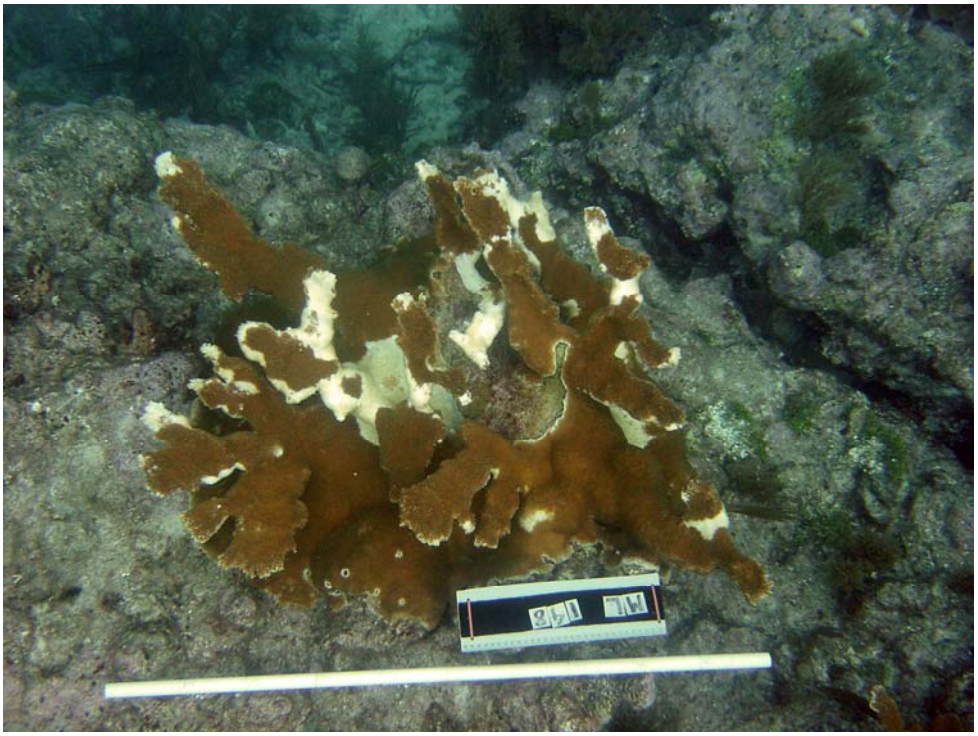
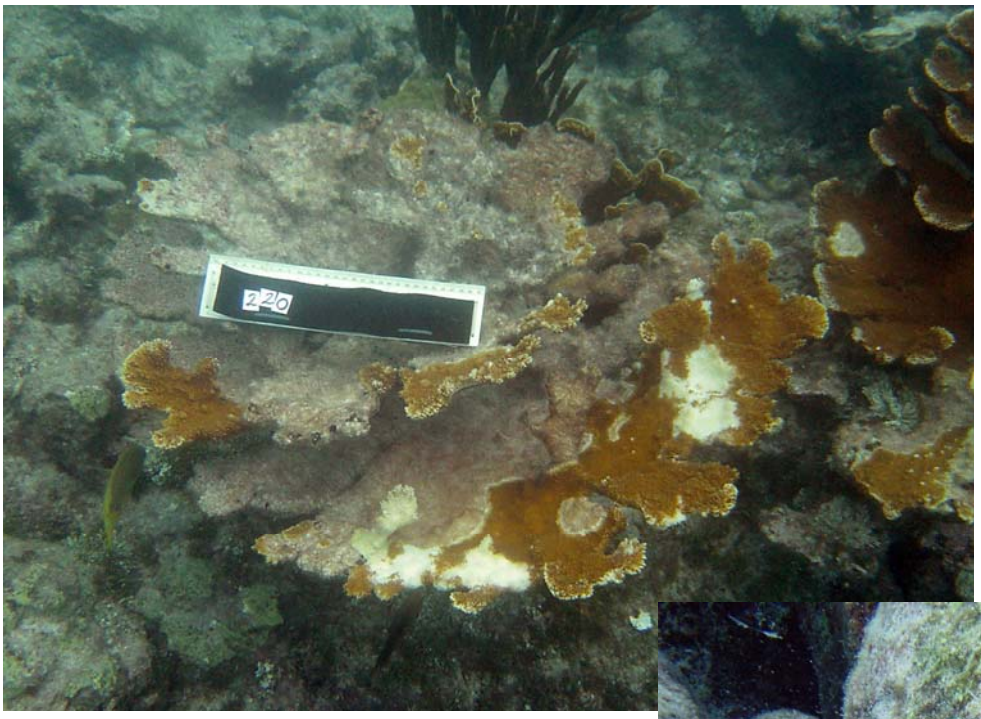


Figure 15. Two examples of colonies with a recent mortality rank= 2. Relative to the size of these colonies the recent mortality is “moderate(10-25%)” (see text).





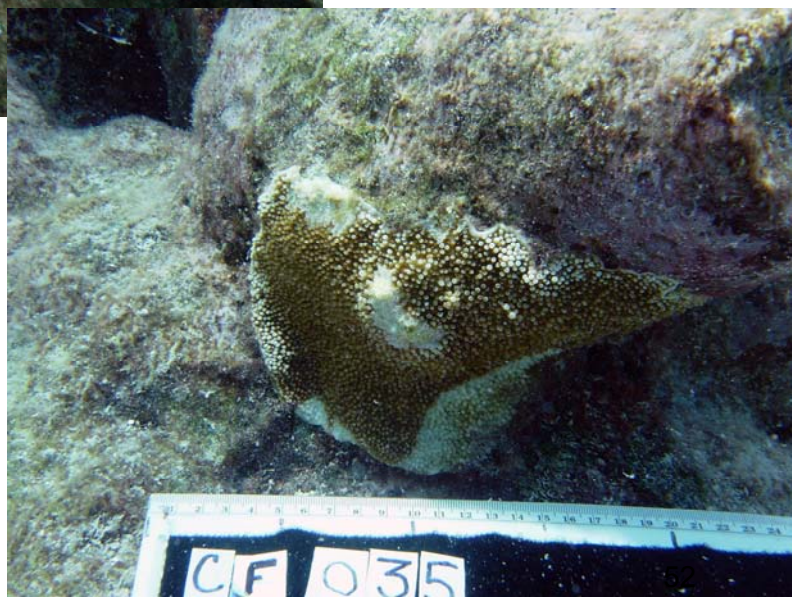


Figure 16. Examples of colonies with a recent mortality rank= 3. Relative to the size of these colonies the recent mortality is “High, affecting 30-55% of the colony live area ” (see text).

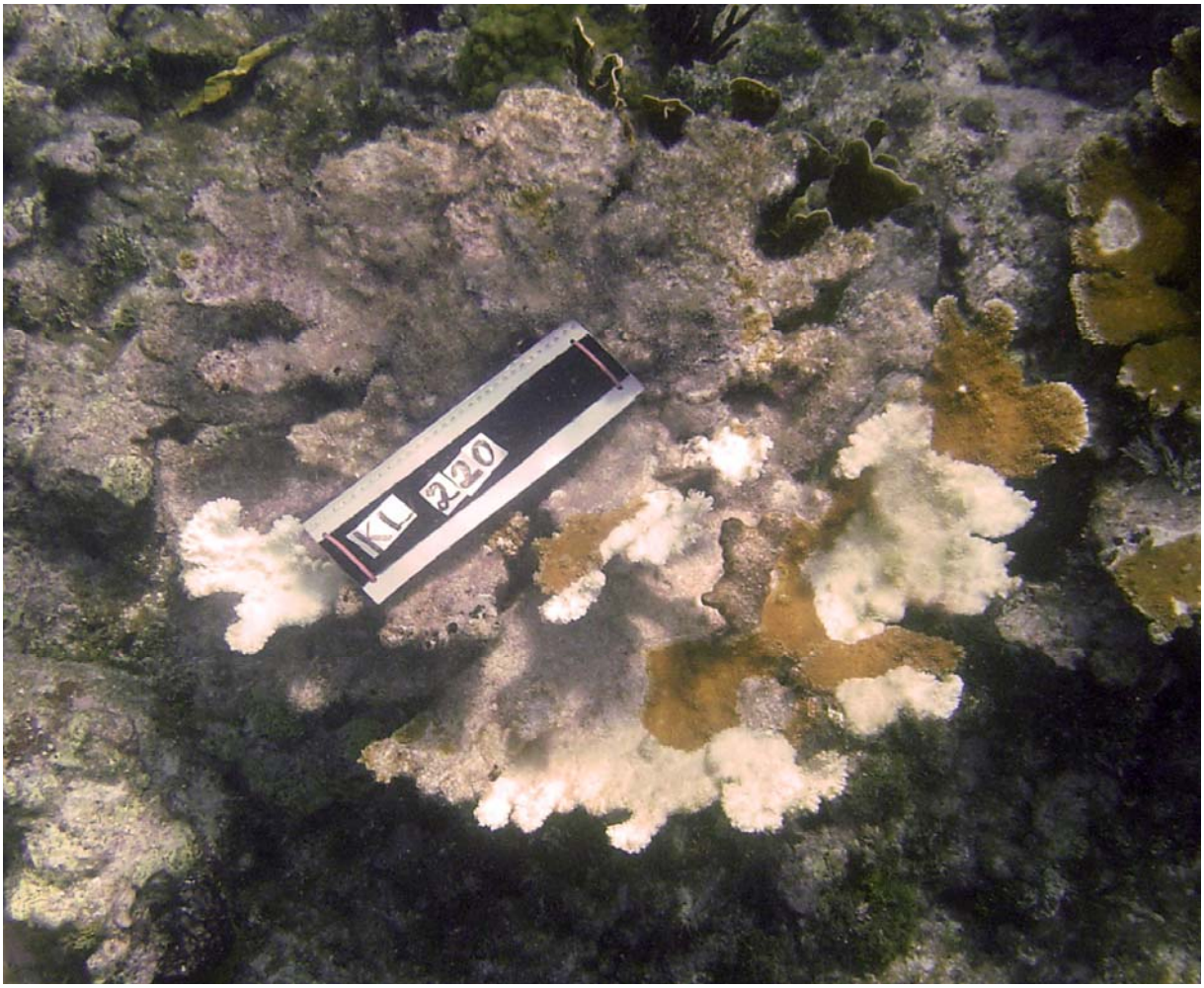
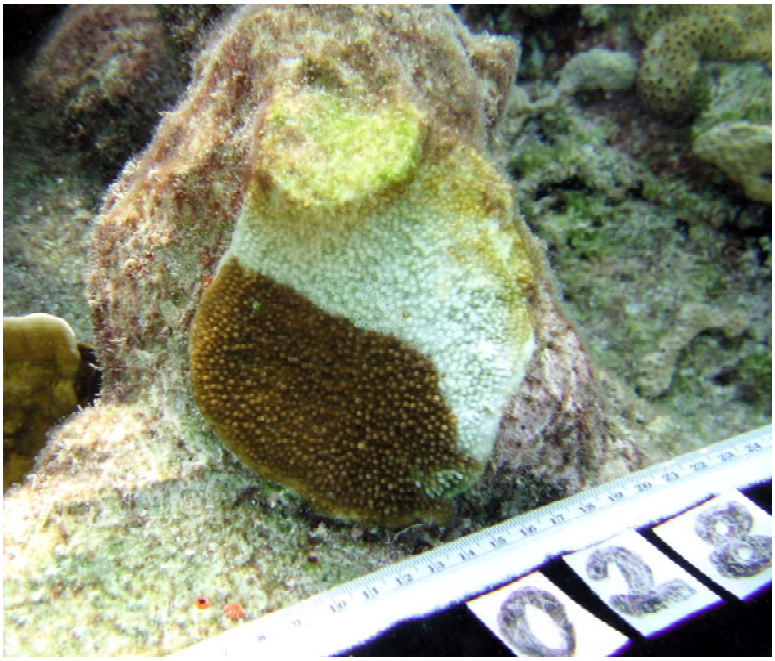
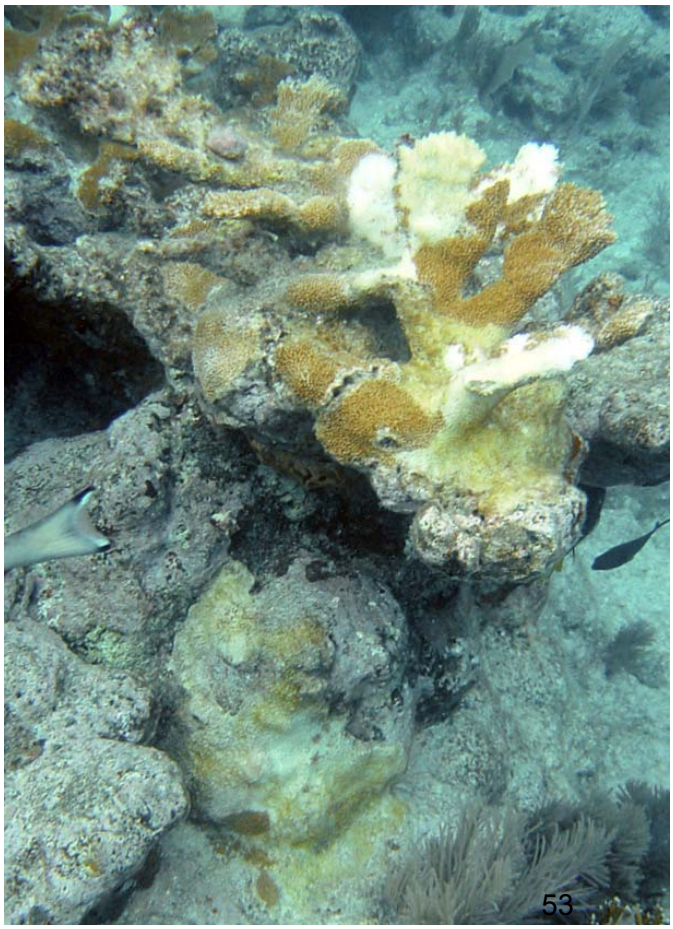
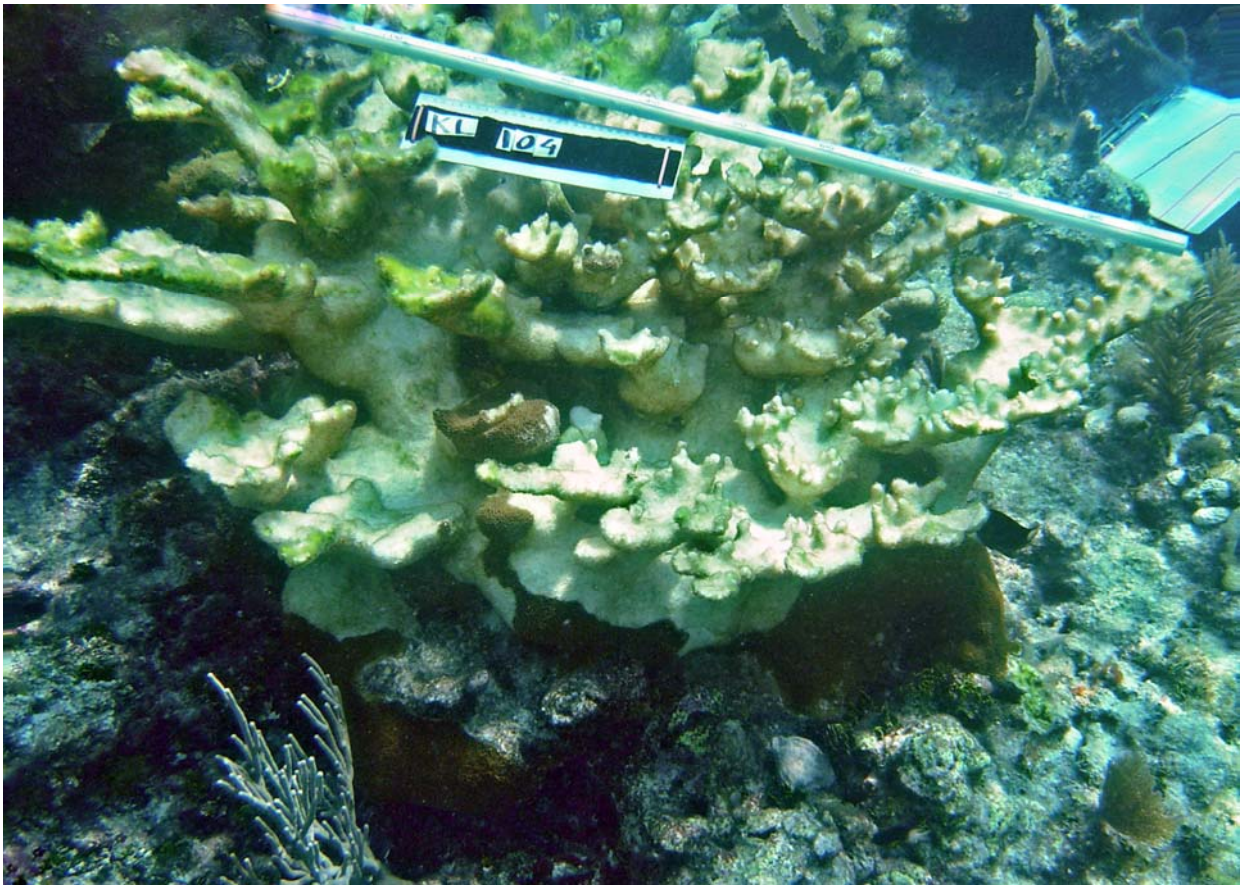


Figure 17. Examples of colonies with a recent mortality rank= 4. Relative to the size of these colonies the recent mortality is “Catastrophic affecting, 60-85% of the colony’s live area” (see text).







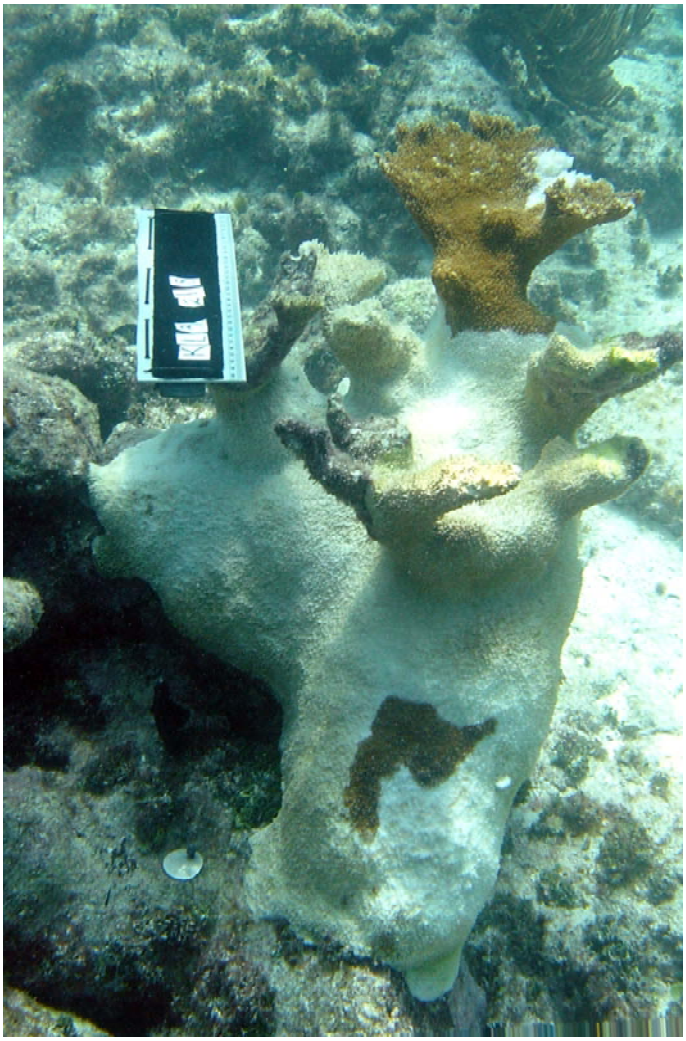
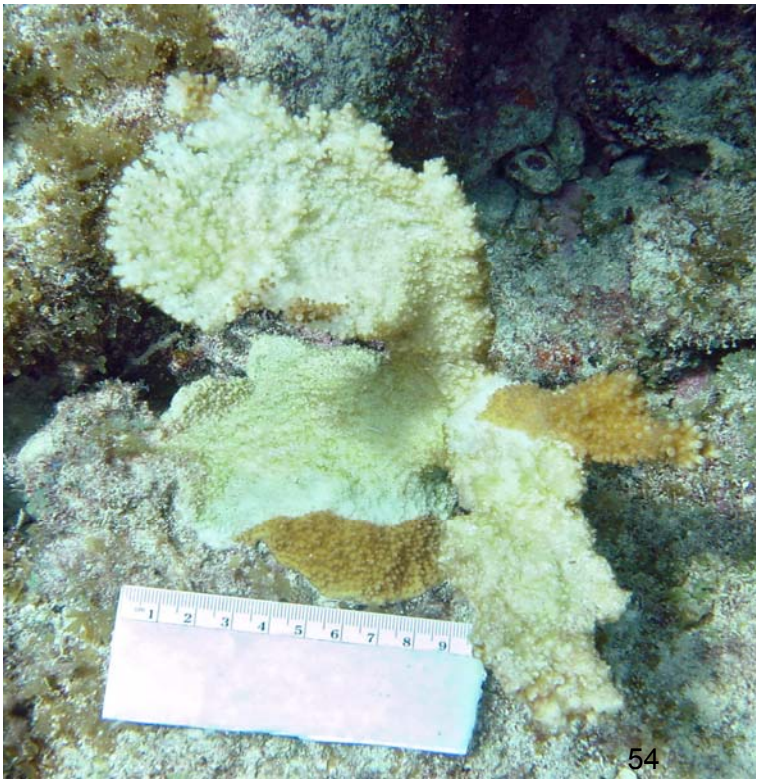


Figure 18. Examples of colonies with a recent mortality rank= 5. Relative to the size of these colonies the recent mortality affects almost the “entire colony >/= 85%” of the colony’s live area” (see text).Fortunately this degree of recent mortality is rarely observed.



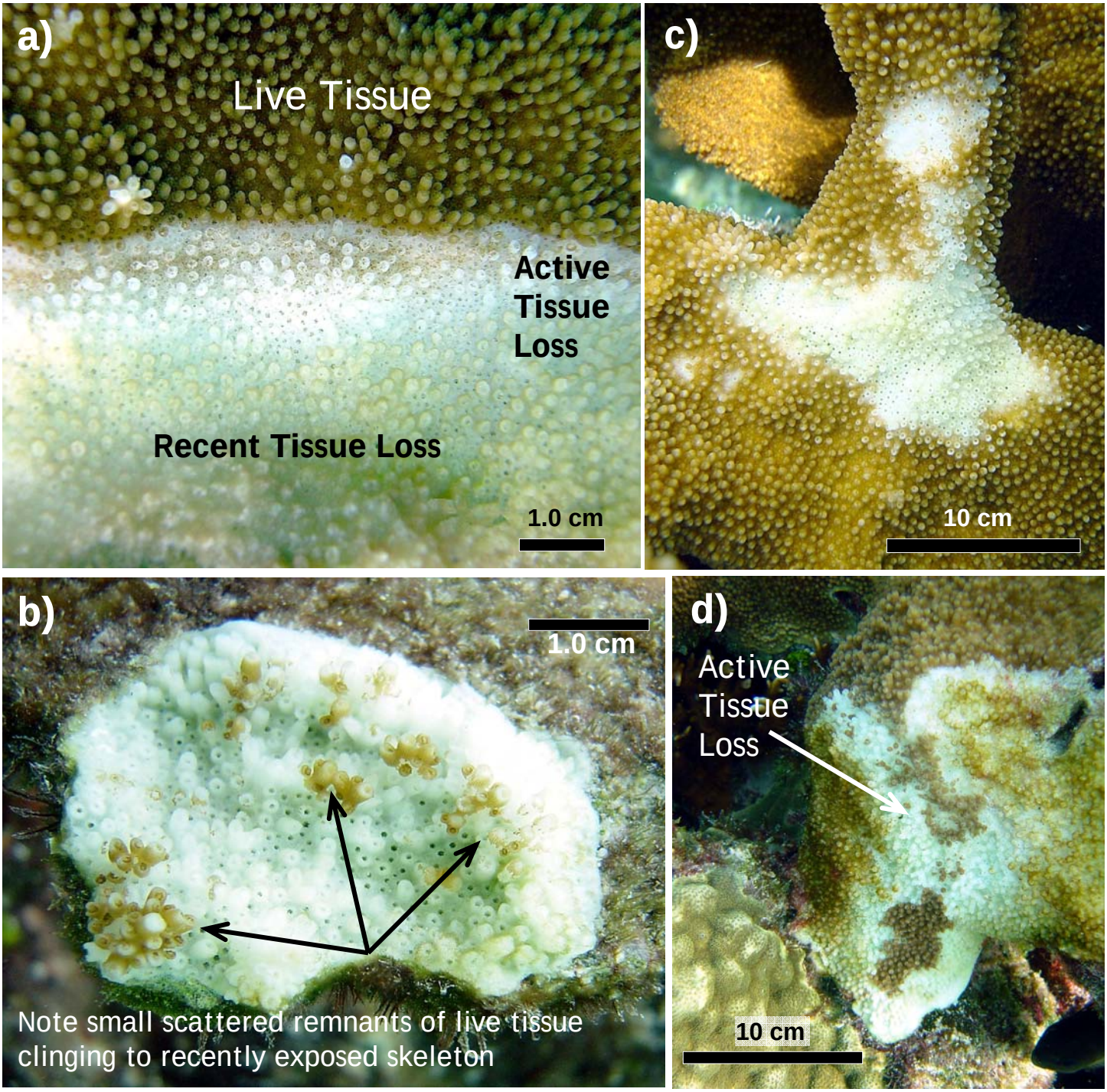
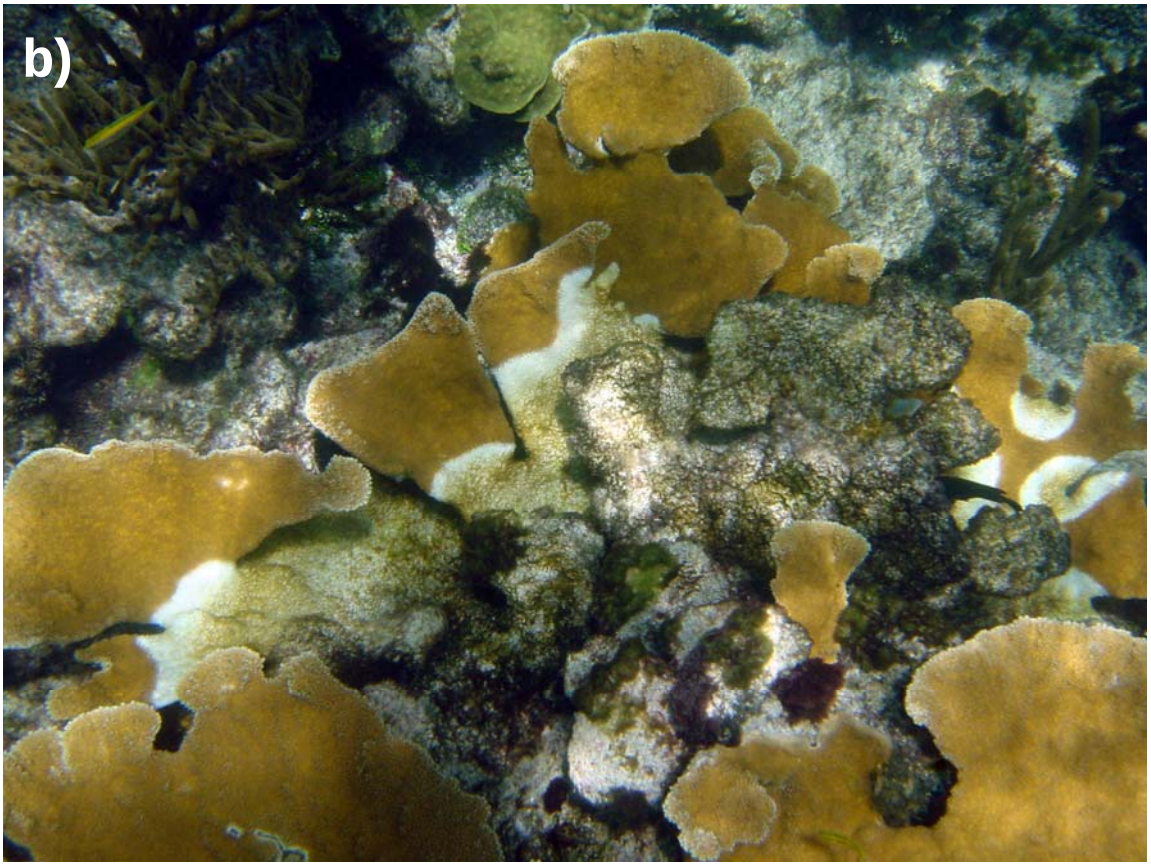


Figure 19. Exposed skeleton that is bright white with NO algal colonization frequently has a jagged margin (a) indicating that tissue loss is actively progressing. The irregular margin results from small pieces of tissue that are still clinging to the skeleton (only seen on close inspection) (b). For the purposes of the colony condition assessment described in this protocol 'active' tissue loss is not distinguished from 'recent' tissue loss, but recognizing it can help recognize the pattern and rate of tissue loss which aids in assigning it to the appropriate type. a) band-like pattern of tissue loss is relatively uniform; c) patchy tissue loss is exposed skeleton typically surrounded by live tissue.

Figure 20. White band disease has a distinctive band of recent mortality. The bright white band of exposed skeleton grades into increasingly algal colonized skeleton indicating that the band is progressing gradually along the branch. Typically the band progresses from the base towards the tips (a) and is often progressing evenly up adjacent branches (a-b)



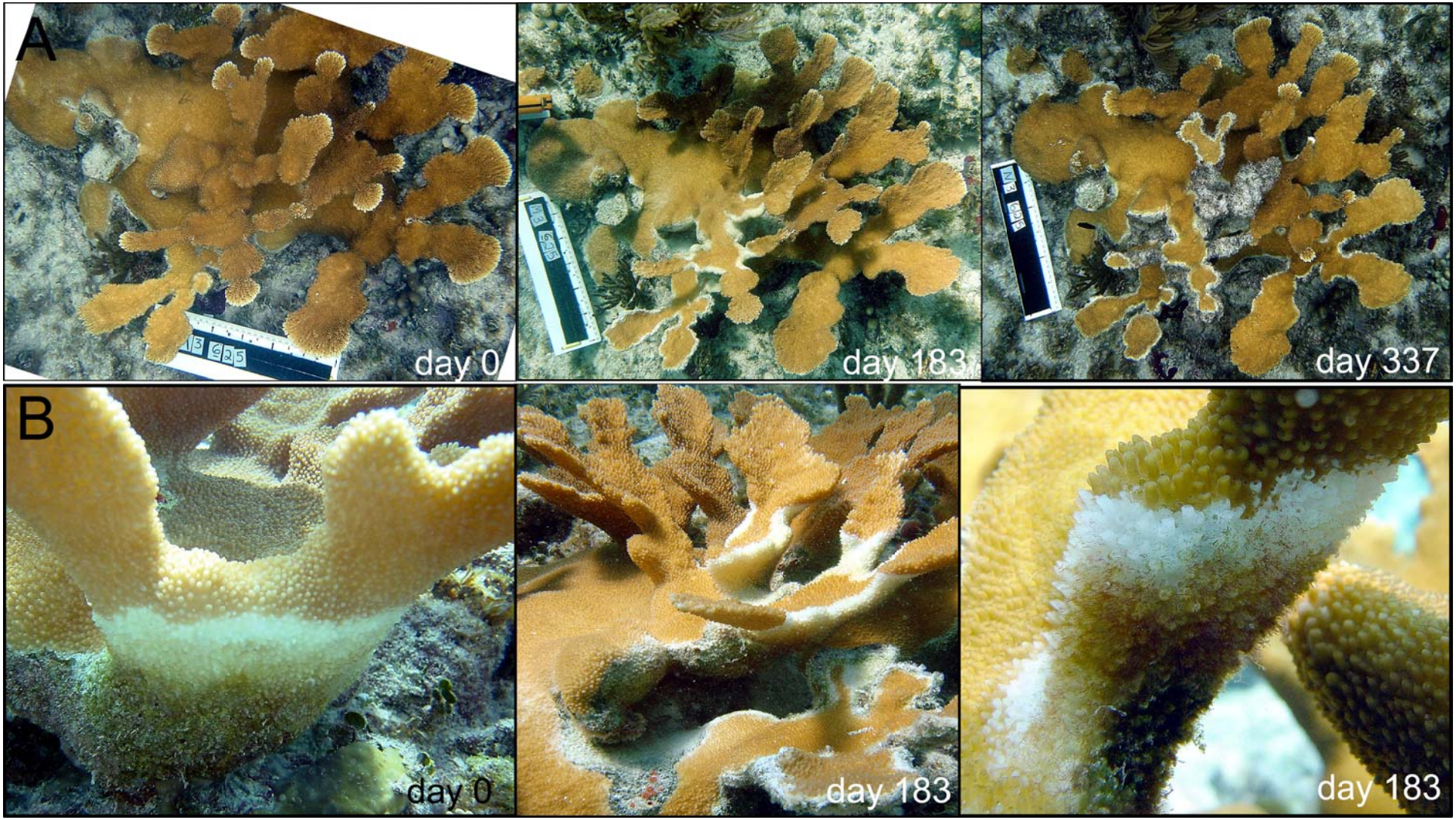


Figure 21. In the Florida Keys, WBD is often observed to advance from the base of the colony along the under side of branches. The disease front often stalls as it reaches the branch margins, leaving branches with a 'white outline' when viewed from above (Reproduced from Williams & Miller 2006). A) and B) show time series of two separate colonies.

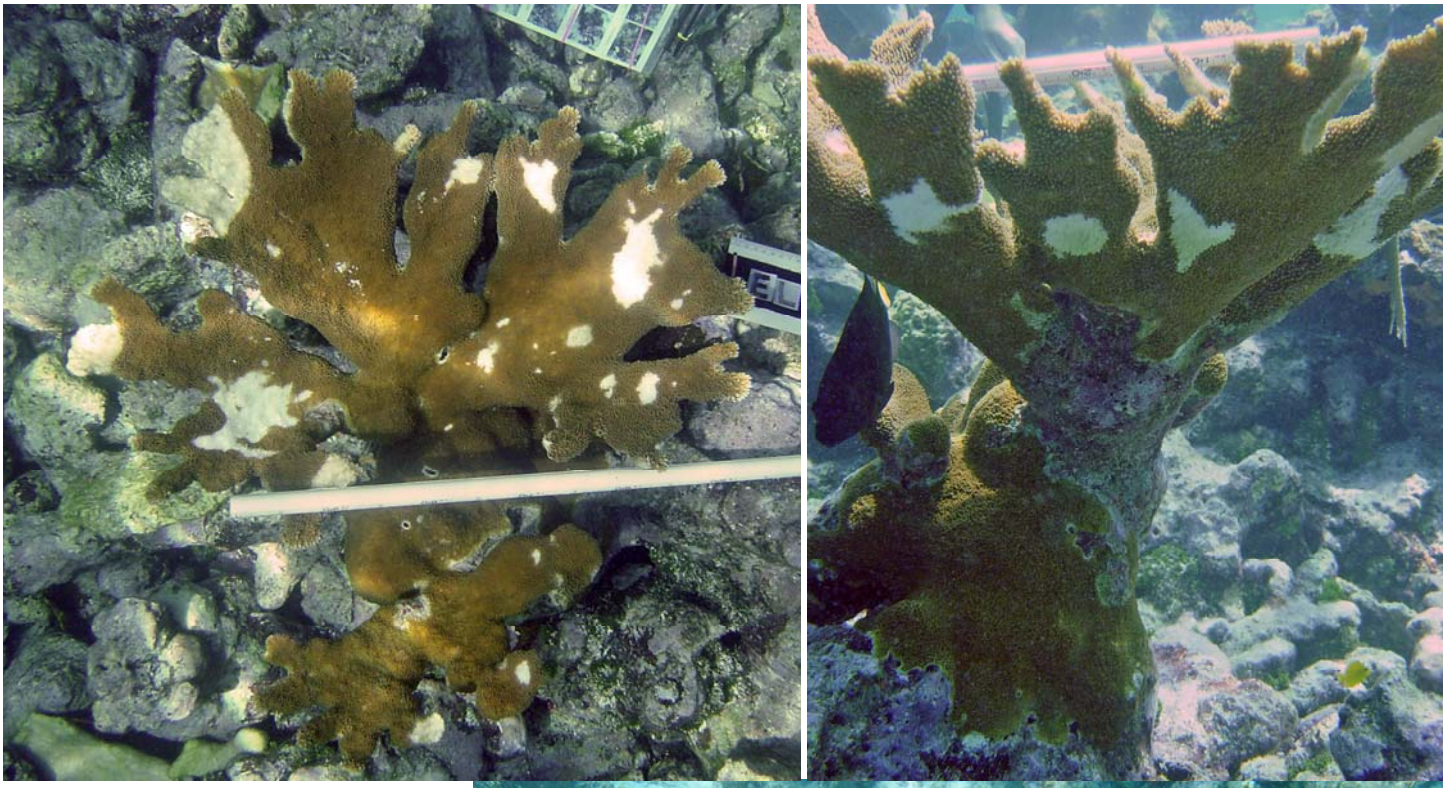
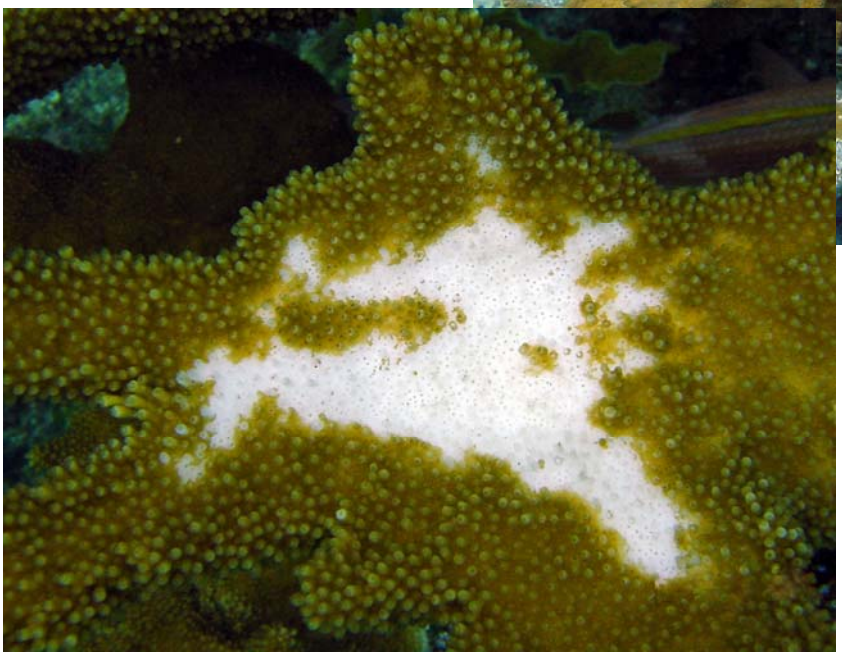


Figure 22. White pox type recent mortality is characterized by multifocal and irregular patches of exposed skeleton or 'lesions' with rates of tissue loss of 12 cm^2 per day. Lesions' can be found on any part of the colony



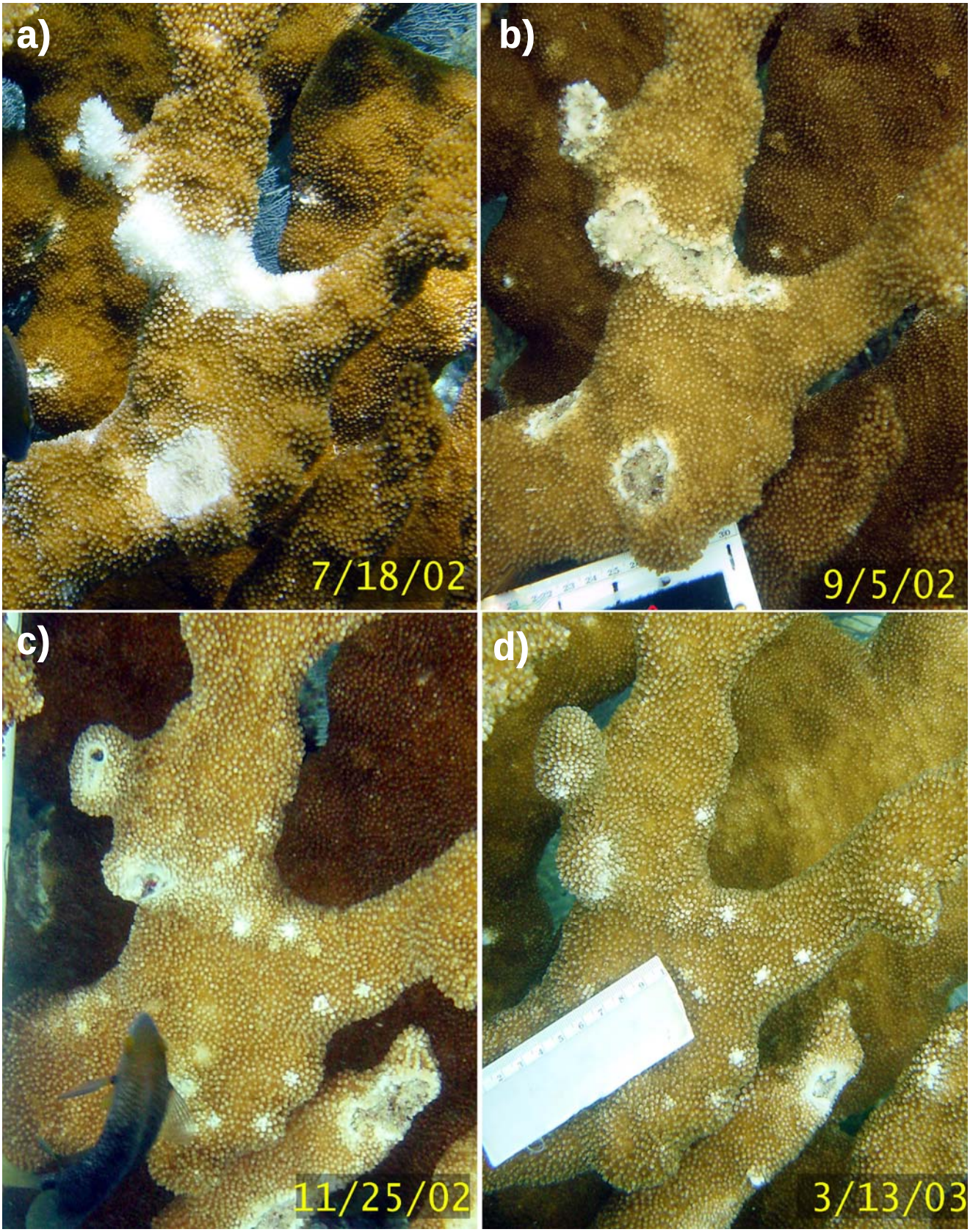
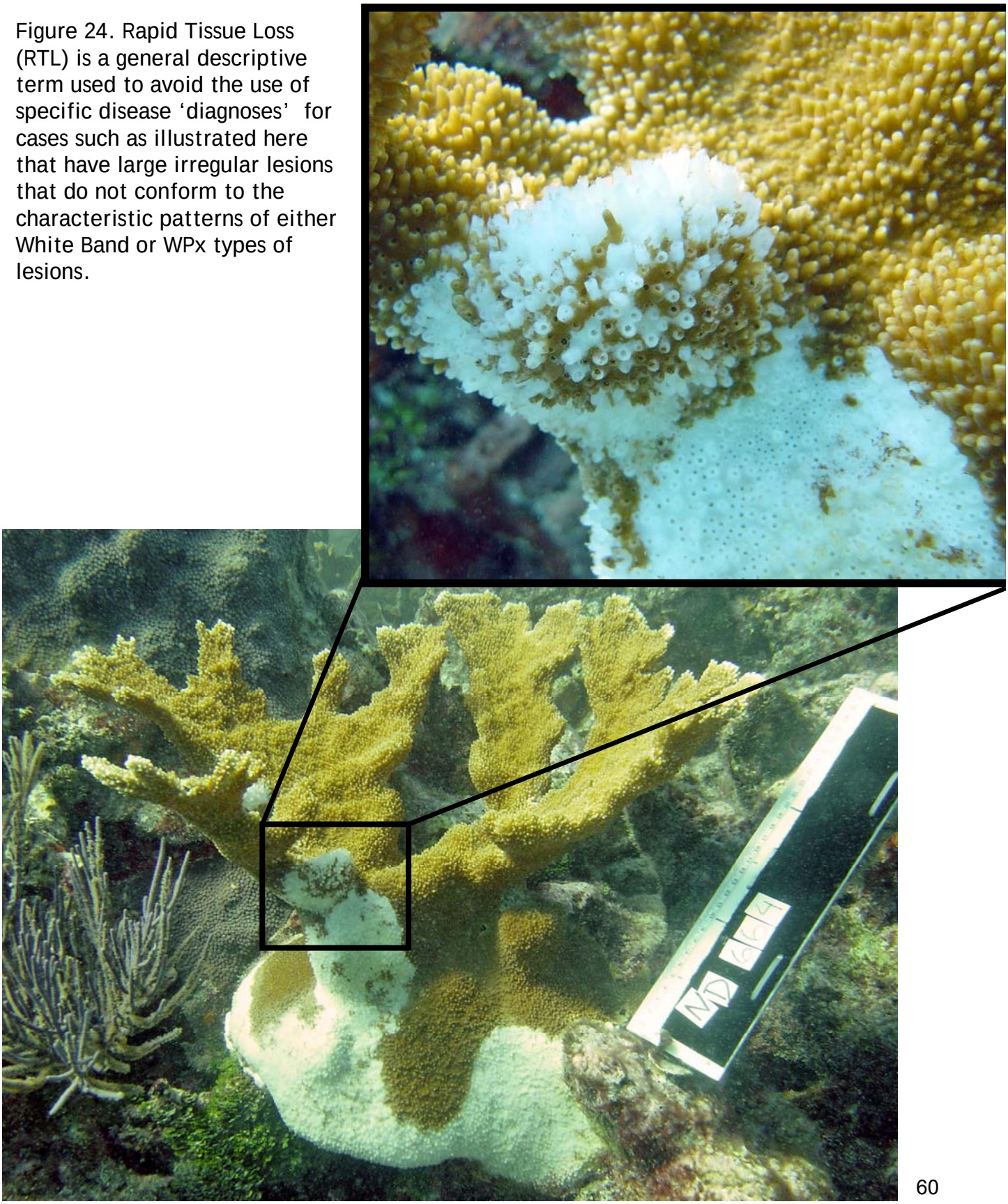


Figure 23. a) recent mortality classified as white Pox (WPx) lesions. b) lesions are healing, note the white lip around the advancing tissue which is typical of new tissue growth. In frames b-d the original lesions are observed to heal completely. c) two additional lesions in the bottom right have appeared since the last photo but are also in the process of healing which continues in d; small white spots termed damselfish chimneys also appear, the threespot damselfish is also visible in frame c.

Figure 24. Rapid Tissue Loss (RTL) is a general descriptive term used to avoid the use of specific disease 'diagnoses' for cases such as illustrated here that have large irregular lesions that do not conform to the characteristic patterns of either White Band or WPx types of lesions.



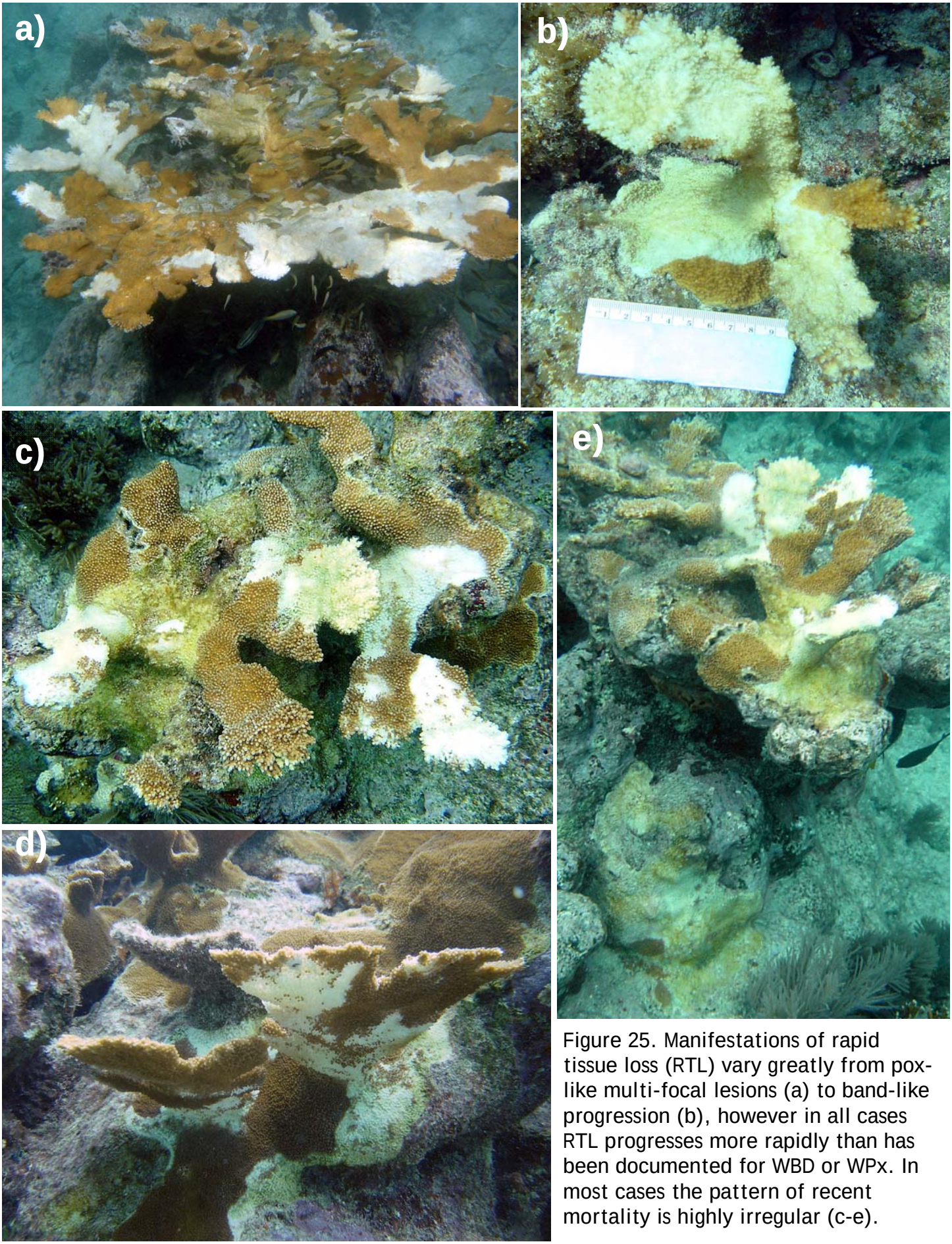


Figure 25. Manifestations of rapid tissue loss (RTL) vary greatly from pox-like multi-focal lesions (a) to band-like progression (b), however in all cases RTL progresses more rapidly than has been documented for WBD or WPx. In most cases the pattern of recent mortality is highly irregular (c-e).

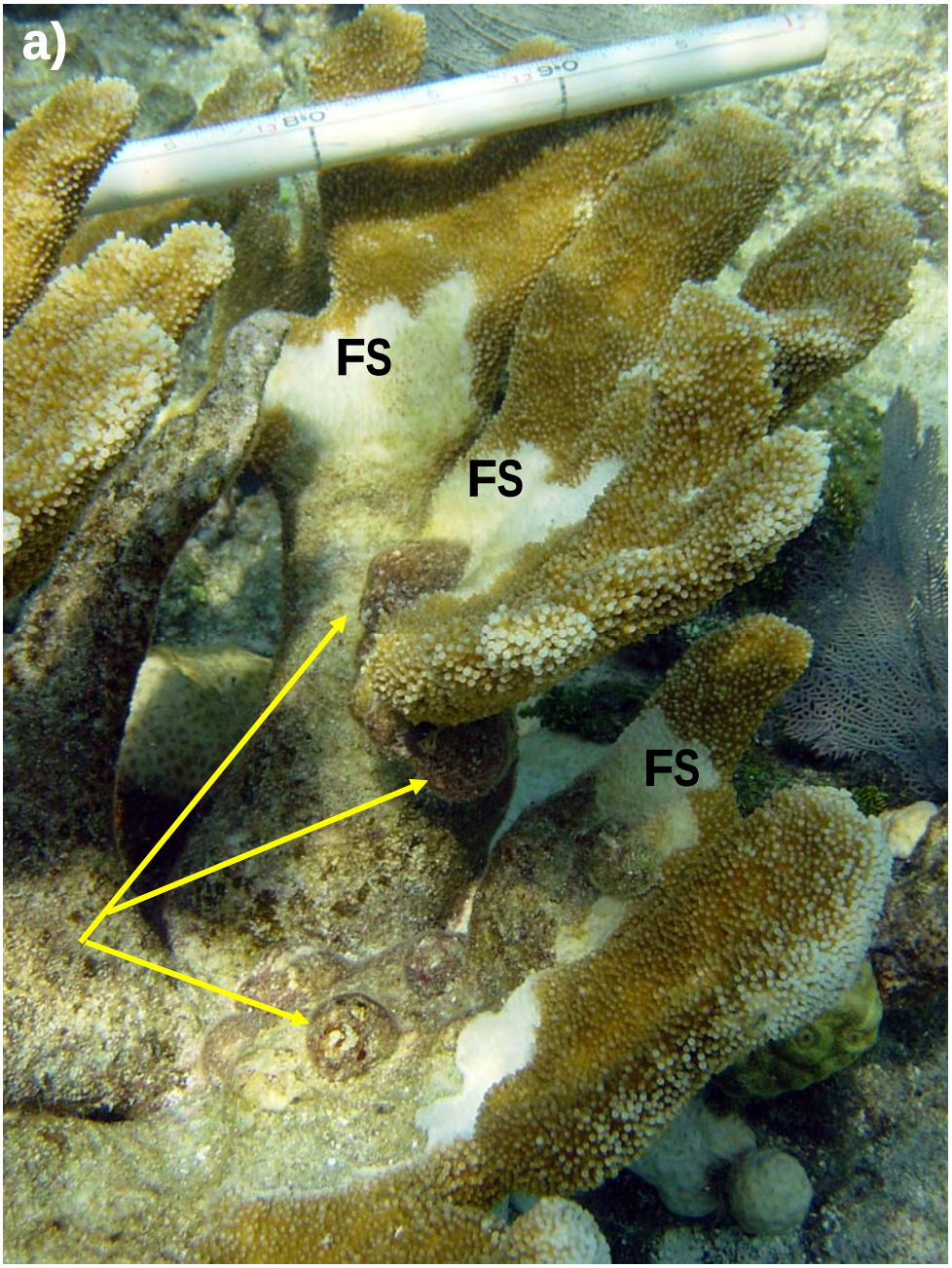
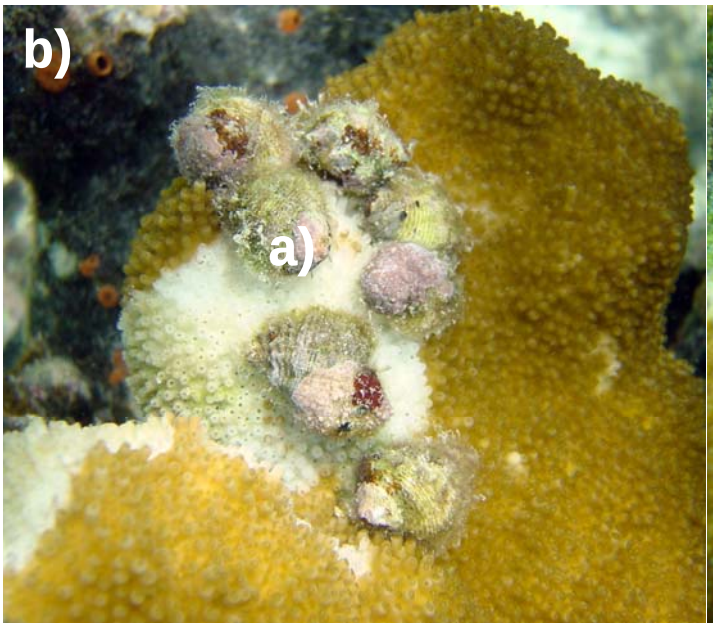


Figure 26. *Coralliophila abbreviata* feeds on *Acropora* spp. (and other coral) tissue leaving an area of bare skeleton (recent mortality) called a feeding scar (FS). They are often found feeding in a group (typically 3-6 snails) but algal colonization on their shell makes the highly cryptic, resting on dead substrate at the base of colonies or undersides of branches when not actively feeding. They usually remain on a single colony for extended periods of time (months or longer). While they are often found feeding (b) but when not feeding they migrate away from the live tissue and are found 'resting' (c) and outside of the feeding scar. Note the scalloped pattern at the margin of the feeding scar and live tissue is apparent in frames a and c.



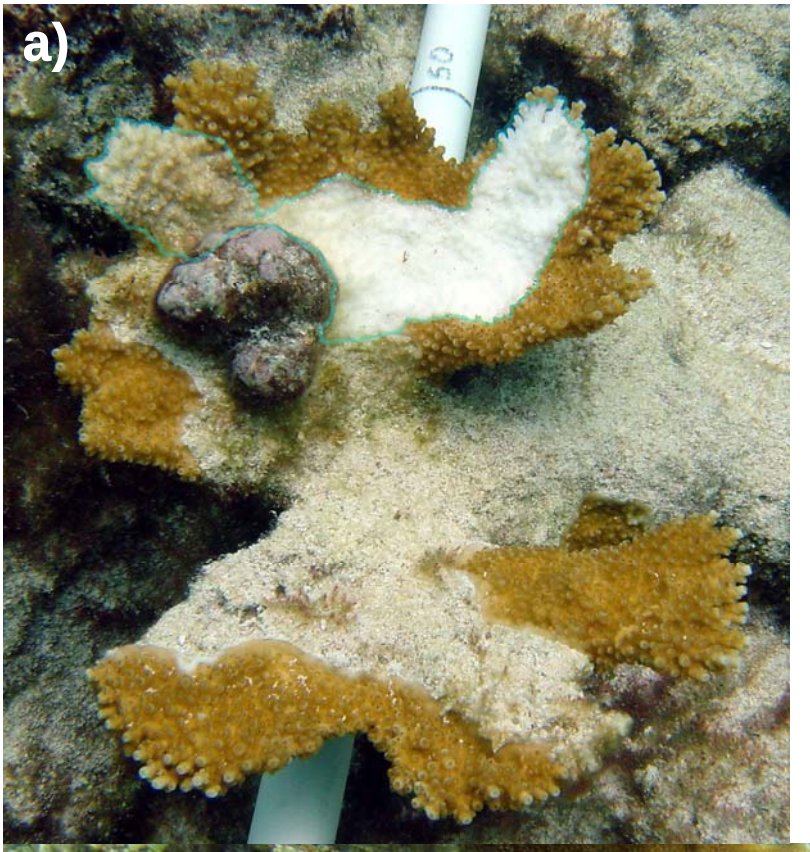
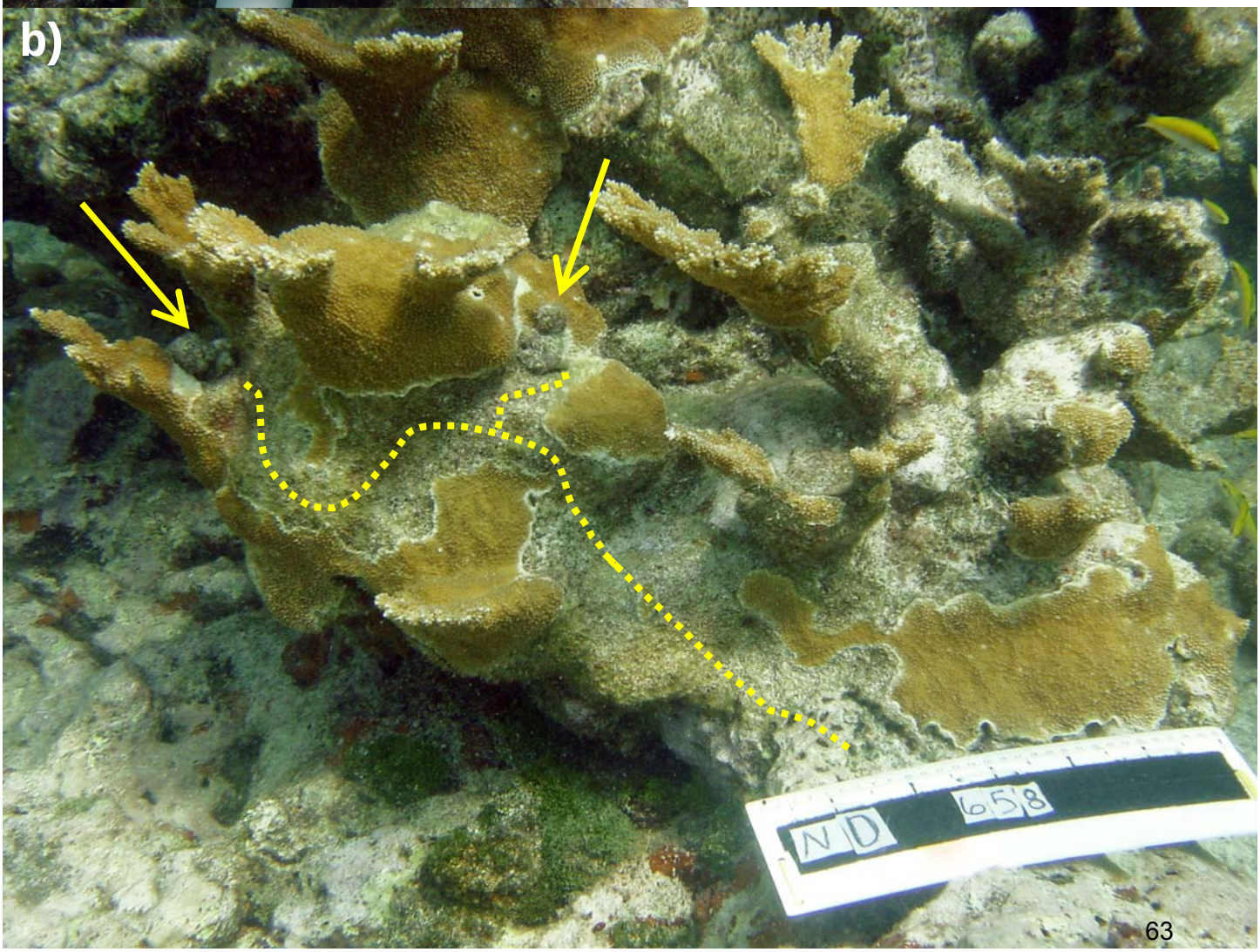


Figure 27. *Coralliophila abbreviata* predation typically leaves a path or trail of denuded skeleton behind. a) shows a trail of recent mortality cased by snail feeding (i.e. Feeding scar (FS)), and older dead areas on the branch which may be areas fed on by snails in the past. These snails appear to have encountered the branch tip and turned back a few times in the area shown in this frame. However, only the bright white area (outlined in blue) would be considered recent mortality and assigned FS as type, the rest is speculation! b) extensive 'snail trails' can be seen on larger colonies, the dotted yellow line shows a probable trail created by snails (at arrows) feeding over the past 6-8 months.



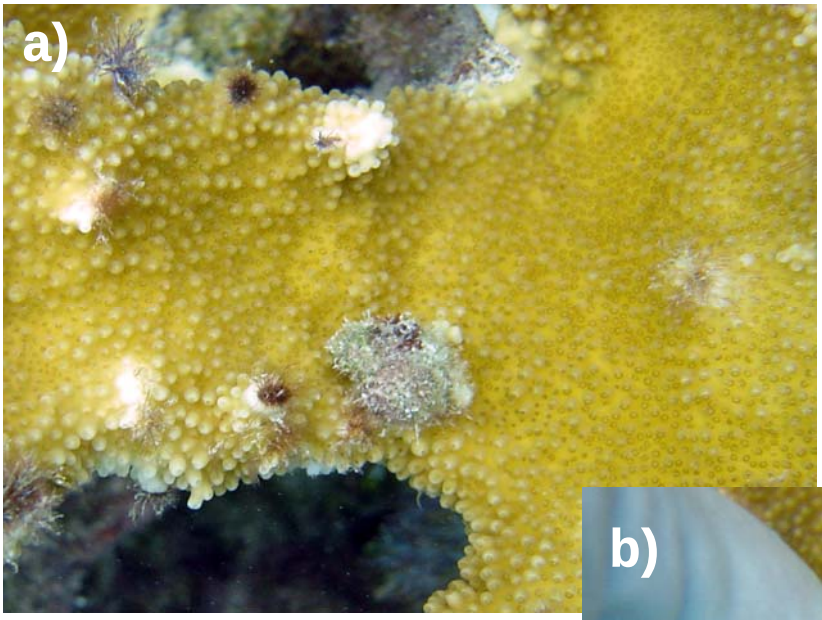
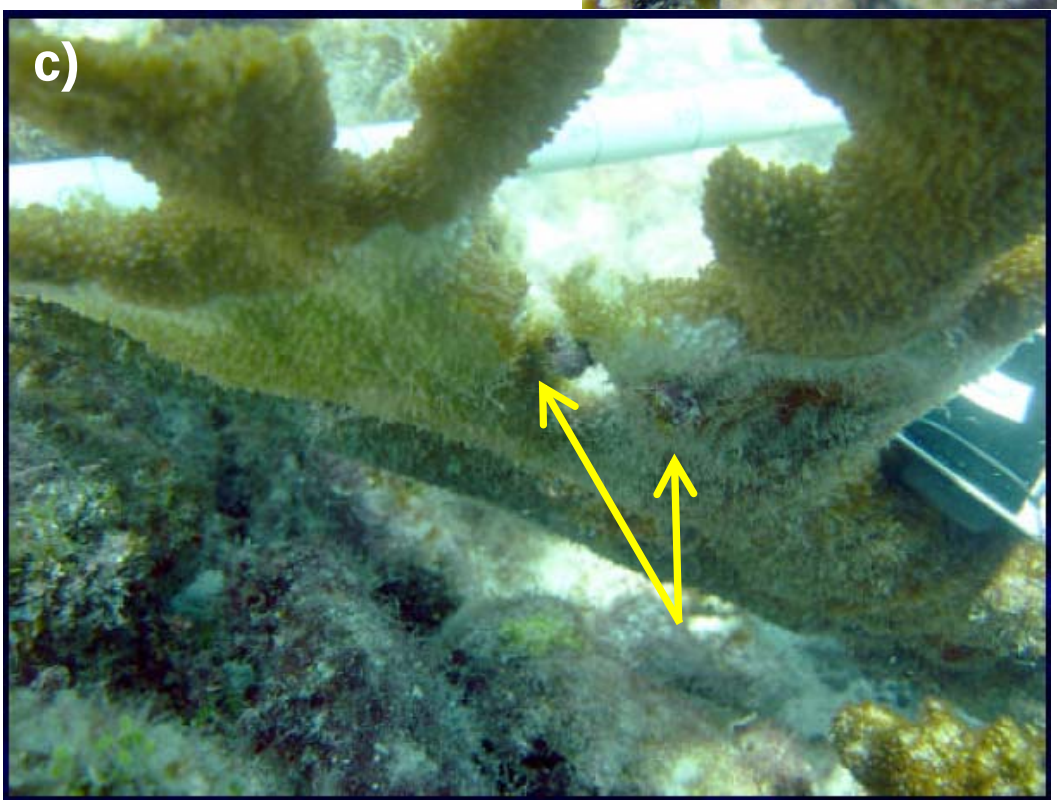


Figure 28. *Coralliophila abbreviata* are only rarely found feeding in an area entirely surrounded by live tissue (a-b). In general, snails should be excluded as the cause of lesions completely surrounded by live tissue (e.g. white pox) unless the snail is observed. c) snails (at arrow) commonly feed on the underside of a branch resulting in a band-like FS that might easily be mistaken for WBD. (see text for further details)



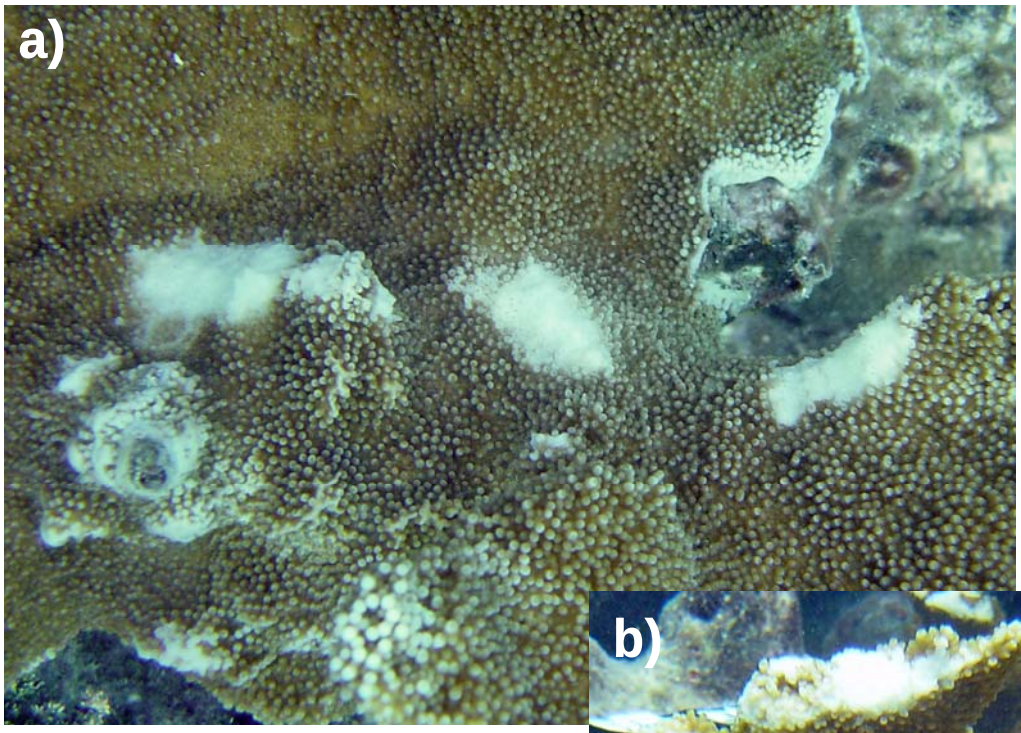
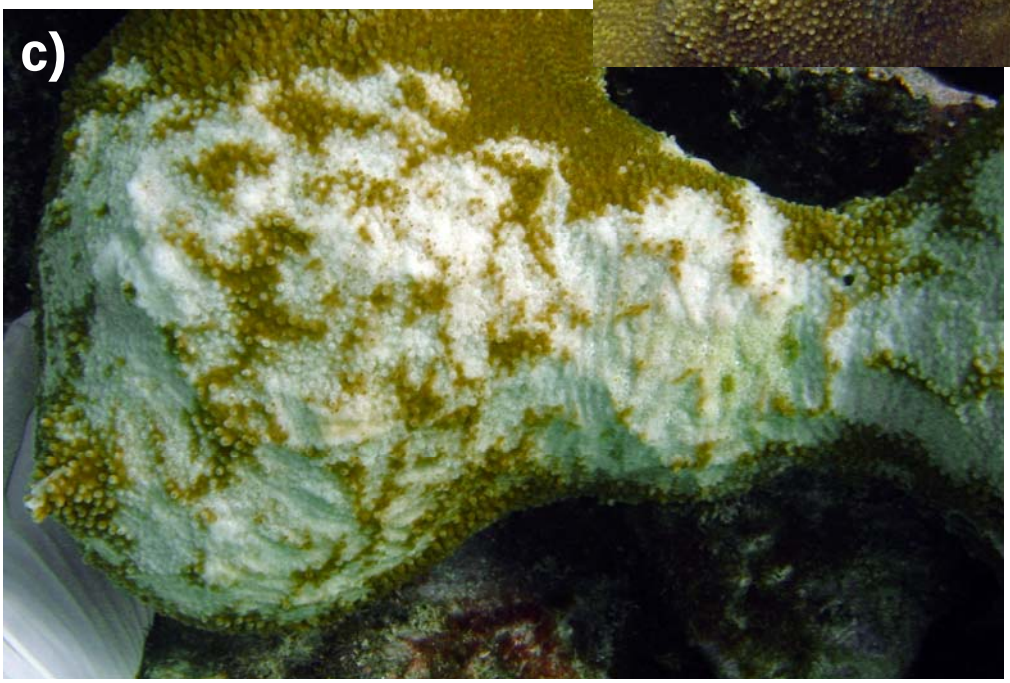


Figure 29. Parrotfish typically feed by scraping reef substrate for the endolithic algae and sponges. However scattered bite marks (a) are not uncommon on live *A. palmata*. Scrapes are more common on raised parts of the colony and branch tips (b). Occasionally the biting becomes more intense and concentrated in a behavior known as 'focused biting' (c).



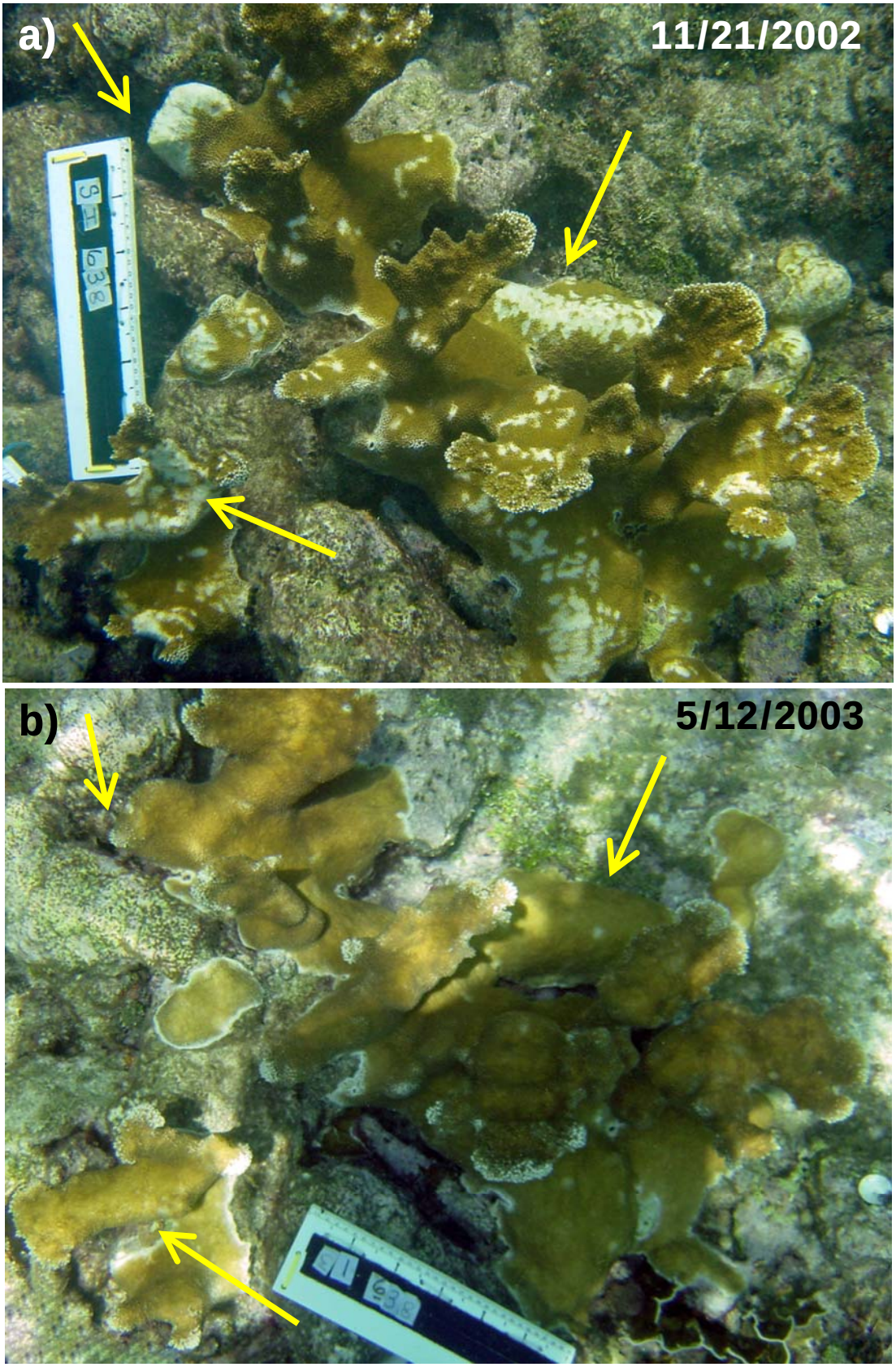


Figure 30. a) Focused biting by Parrotfish (*Sparisoma viride*) can result in substantial areas of tissue loss and skeletal damage. These bites are usually focused on ridges, sides of branches or other protruding portions of a colony (at arrows). b) However colonies are able to heal rapidly, the pictured colony healed completely in less than 6 months (in fact a new proto branch has formed at the upper left arrow).

Figure 31. Colonies are occasionally found with the characteristic appearance of focused biting (b), however on closer inspection the white marks have intact corallites and bleached polyps (a). Since the tissue is intact, it should not be considered as recent mortality, instead it will be recorded as a type of bleaching.

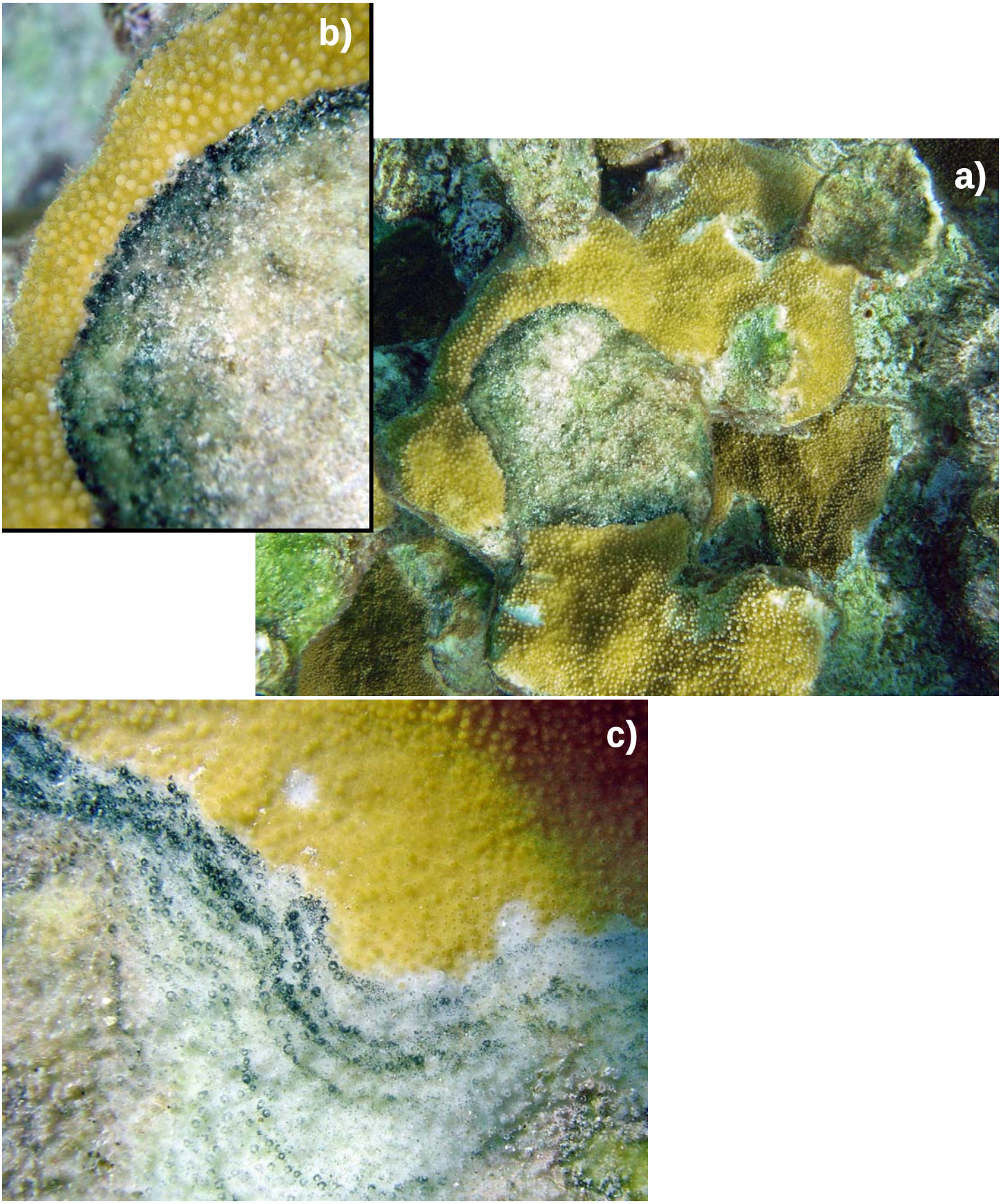
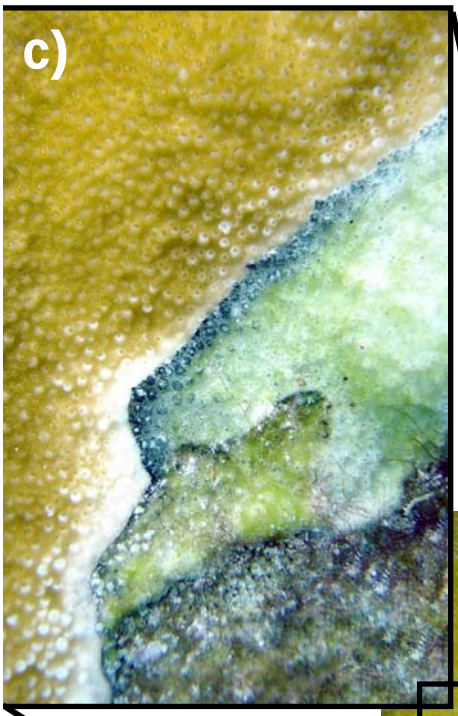
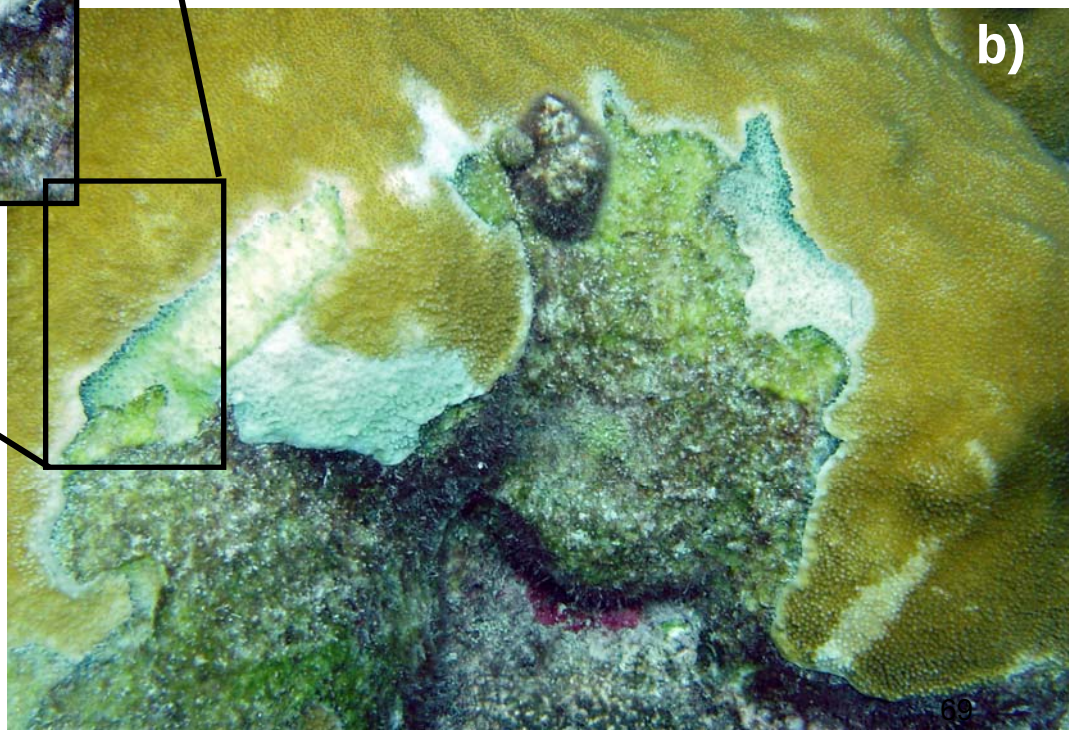
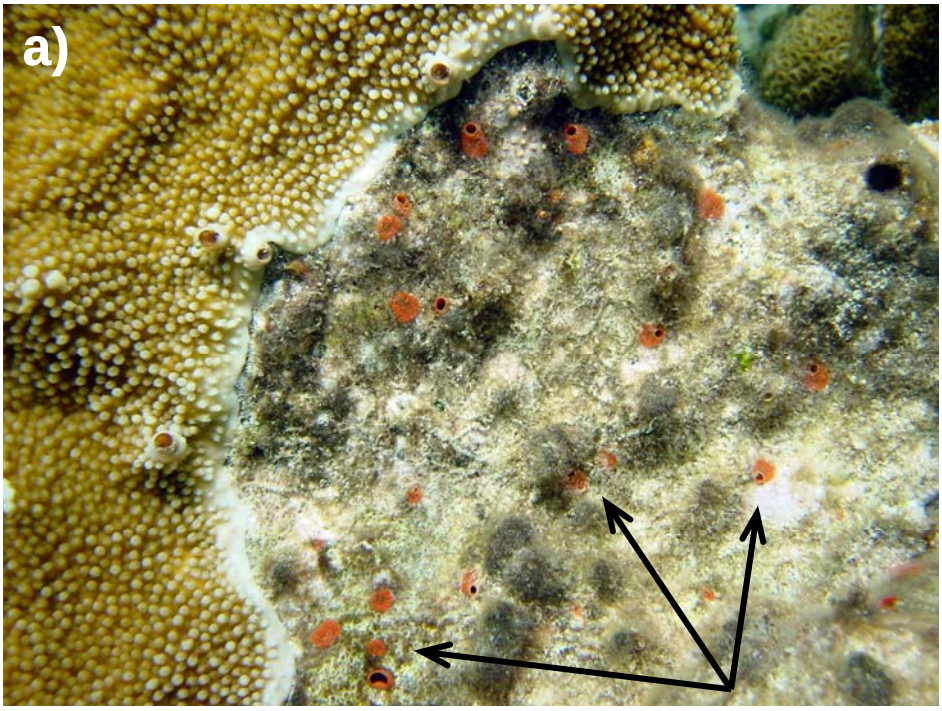


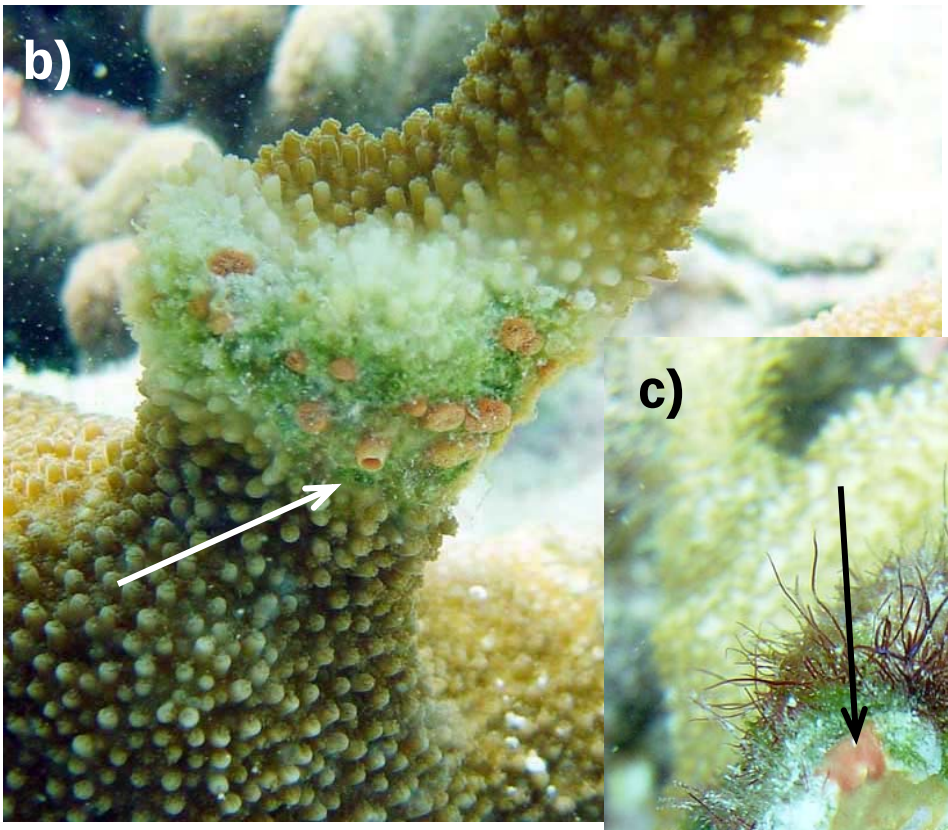
Figure 32. a) A dark greenish black band of ciliates is sometimes seen associate with recent mortality. (b) The ciliates vary in density, forming a thick fuzz resembling black band disease in some cases c) The band appears more sparse in areas of faster tissue retreat.

Figure 33. Cilliate bands are very conspicuous but may co-occur with less distinctive types of recent mortality. A) when a cilliate band occurs with a white band of recent mortality it is impossible to know if the cilliates are the sole cause of the mortality or if white band disease is the primary cause and cilliates are a secondary 'cause'. b) Snail feeding scar is the primary type of recent mortality and c) cilliates are secondary.









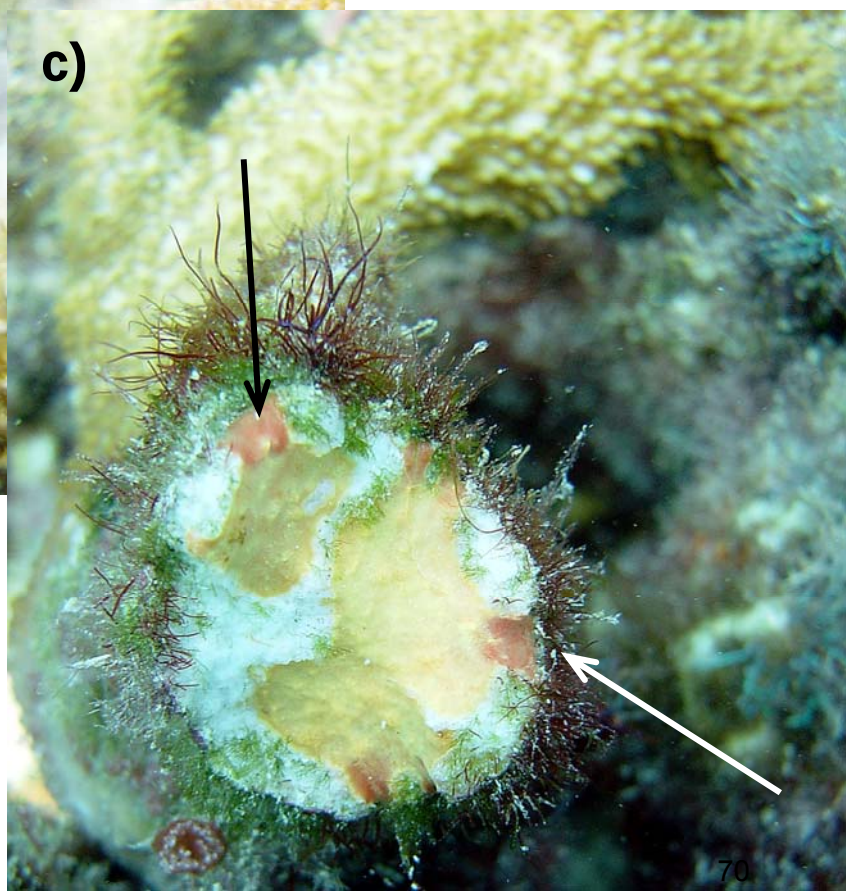


Figure 34. a) Cliona sp.? Is very common on reef substrates including long dead *A. palmata* skeleton and is visible only where its ostia protrude (at arrows). b) Ostia can also protrude through live tissue resulting in a lesion. c) this fresh branch break shows a cross section through an excavated branch. The ostia can be seen (at arrows) along with the main body of the sponge.

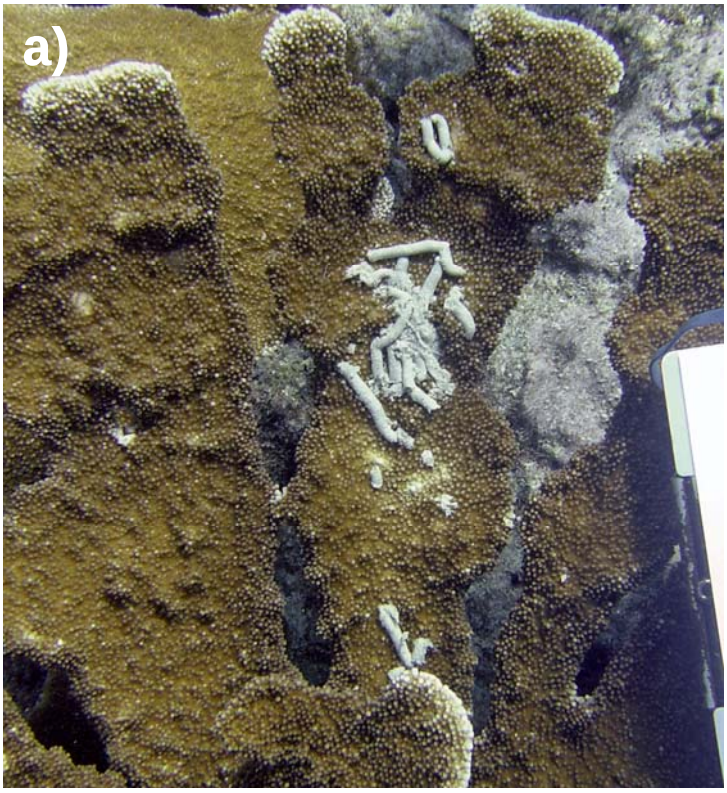
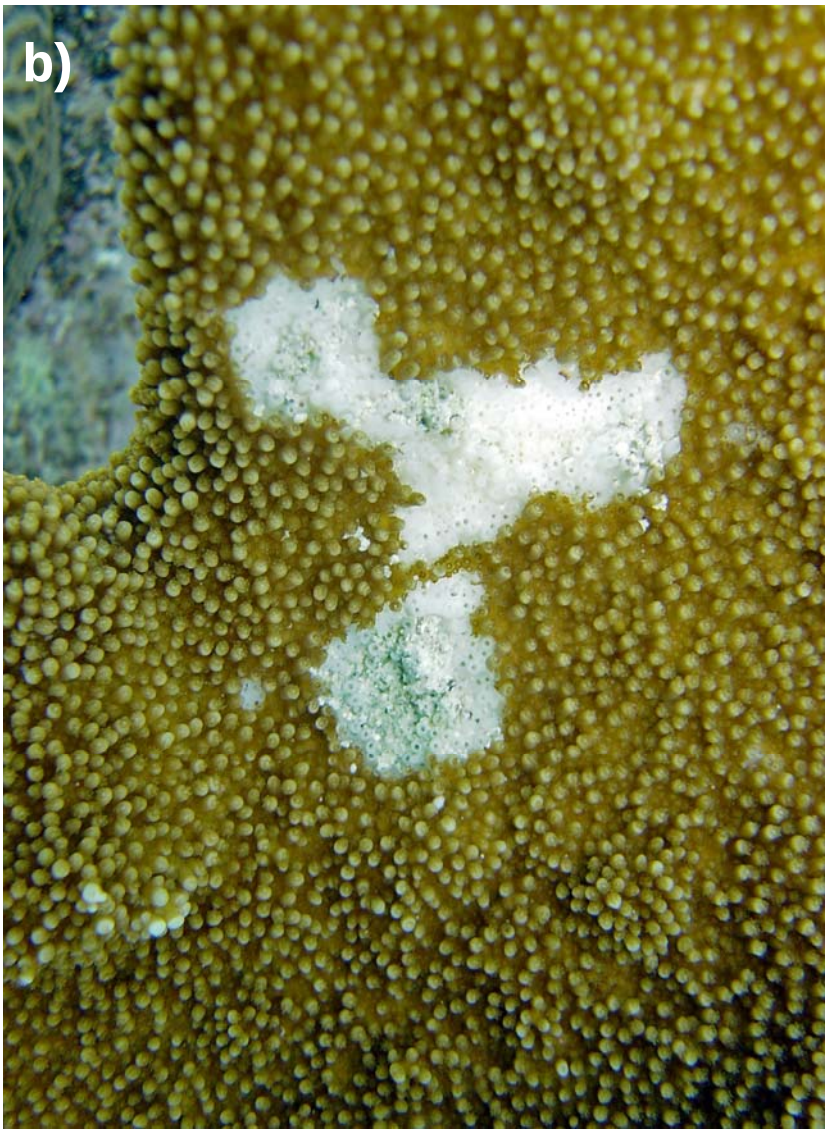
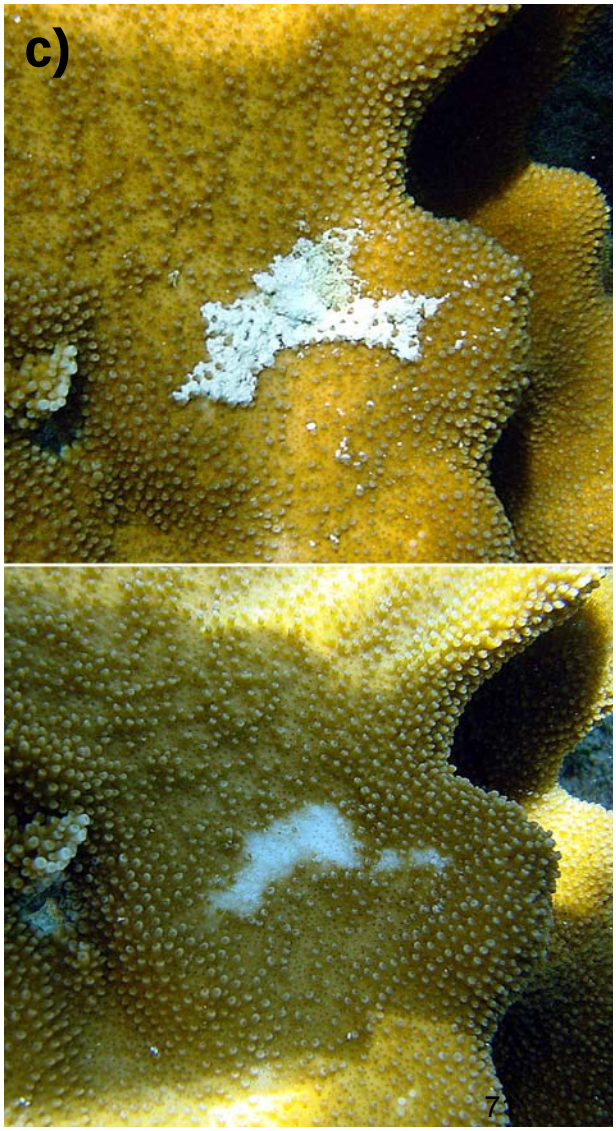


Figure 35. a) When fish fecal material (Fish Poop) lands on *A. palmata*'s flat branches and remains there, the tissue underneath can get smothered resulting in b) scattered irregular lesions that can resemble a white pox lesions. c) Fish poop lesions are often crescent shaped and may have traces of fish poop among the denuded corallites.





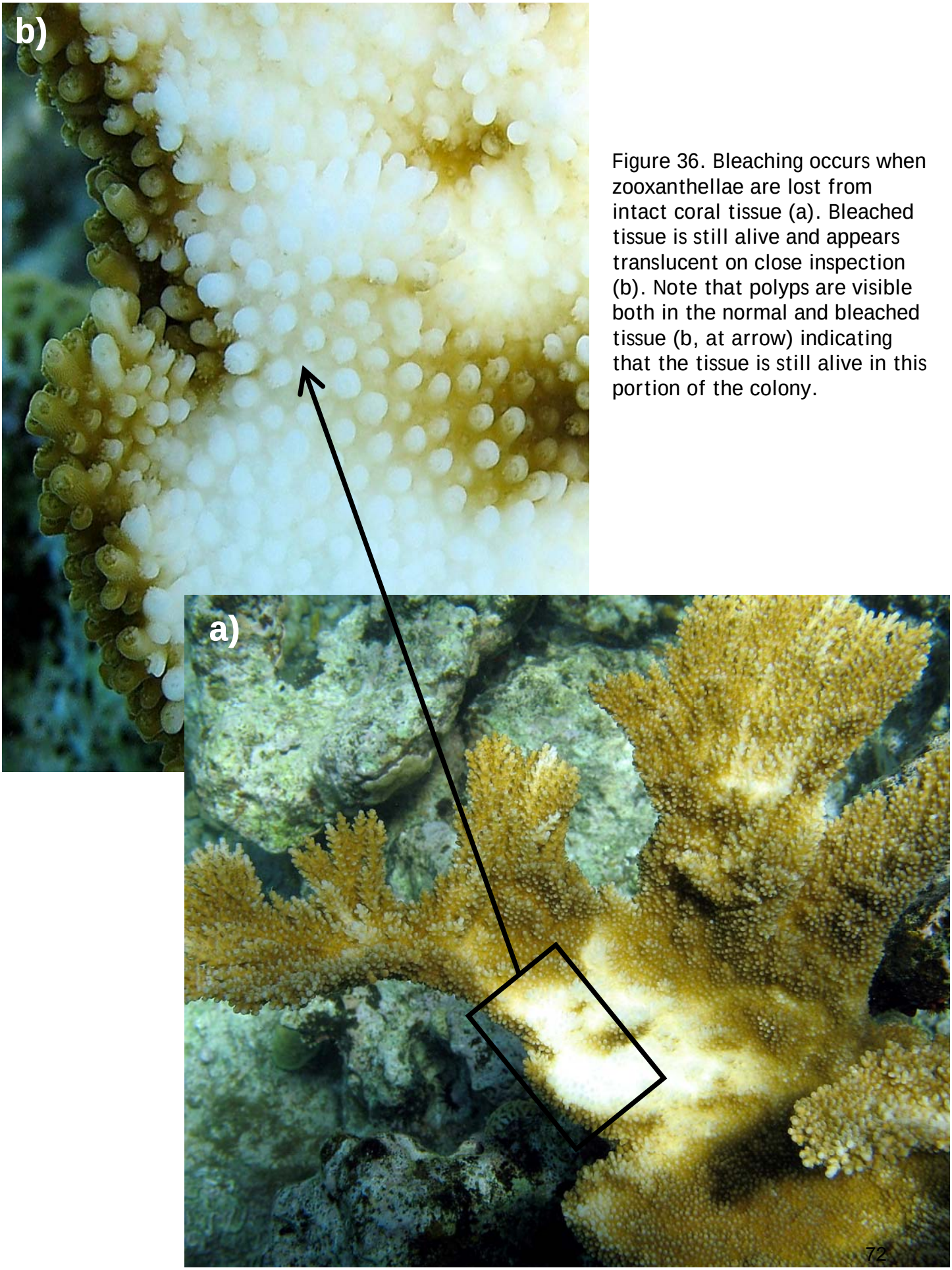


Figure 36. Bleaching occurs when zooxanthellae are lost from intact coral tissue (a). Bleached tissue is still alive and appears translucent on close inspection (b). Note that polyps are visible both in the normal and bleached tissue (b, at arrow) indicating that the tissue is still alive in this portion of the colony.

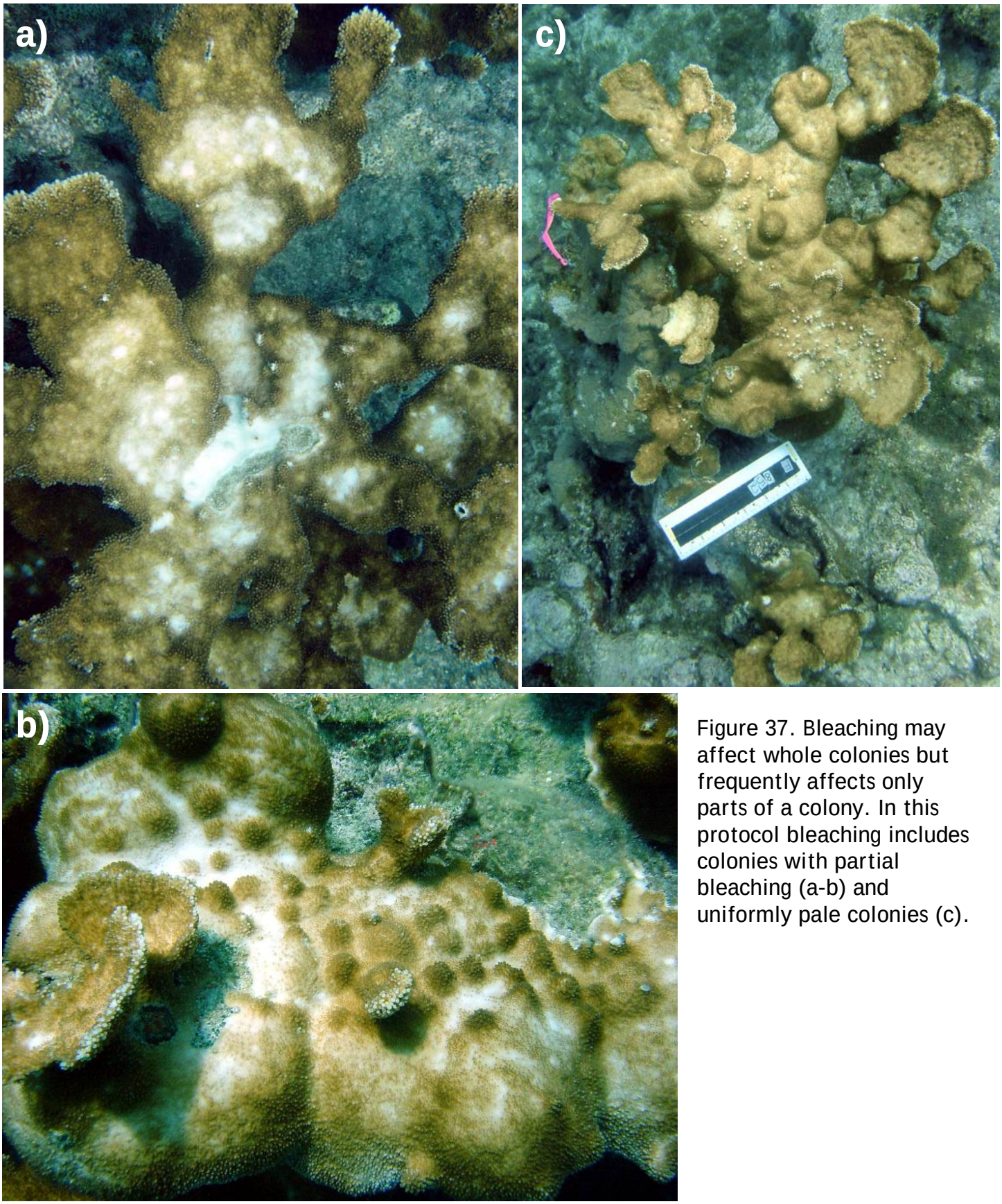
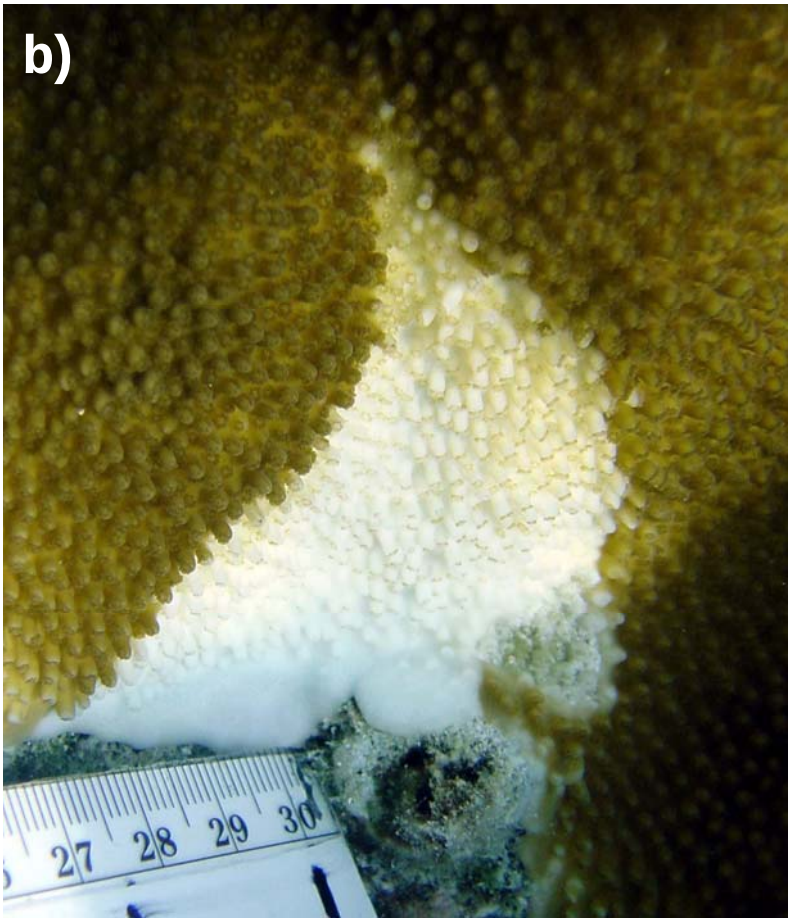


Figure 37. Bleaching may affect whole colonies but frequently affects only parts of a colony. In this protocol bleaching includes colonies with partial bleaching (a-b) and uniformly pale colonies (c).

Figure 38. Wedge-shaped area of bleached tissue (known informally as 'Wedgies') are usually found around the base of a colony leading to the tissue margin (a-b), but they can occasionally be found in in the interior of the colony surrounded by live tissue (c). These marks persist nearly unchanged for at least several years.



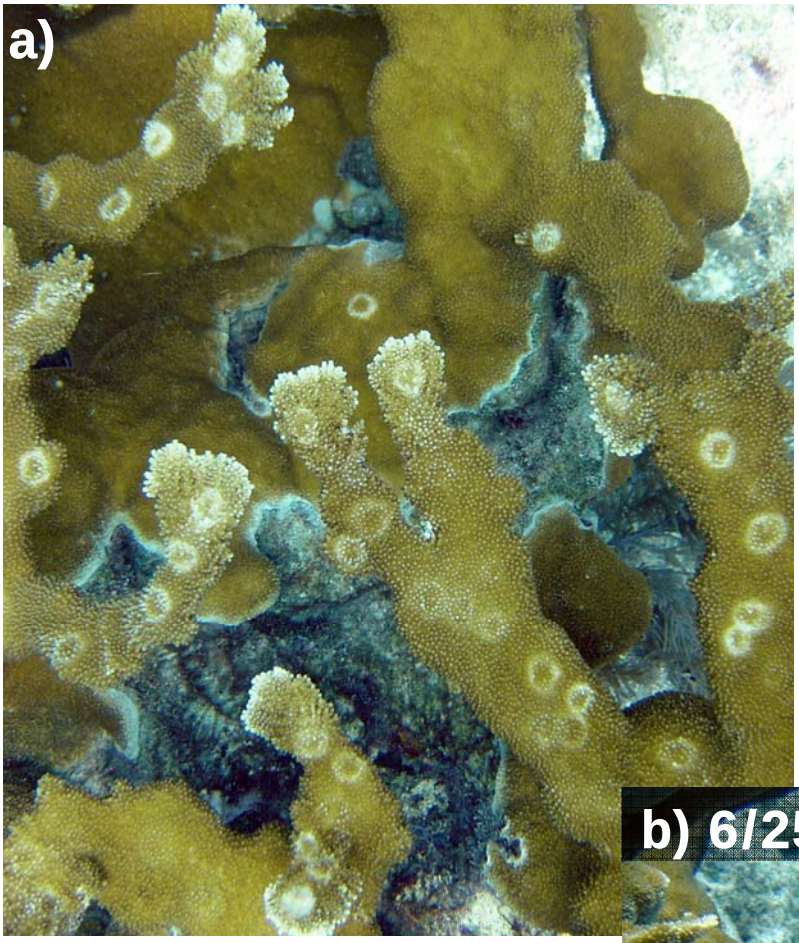
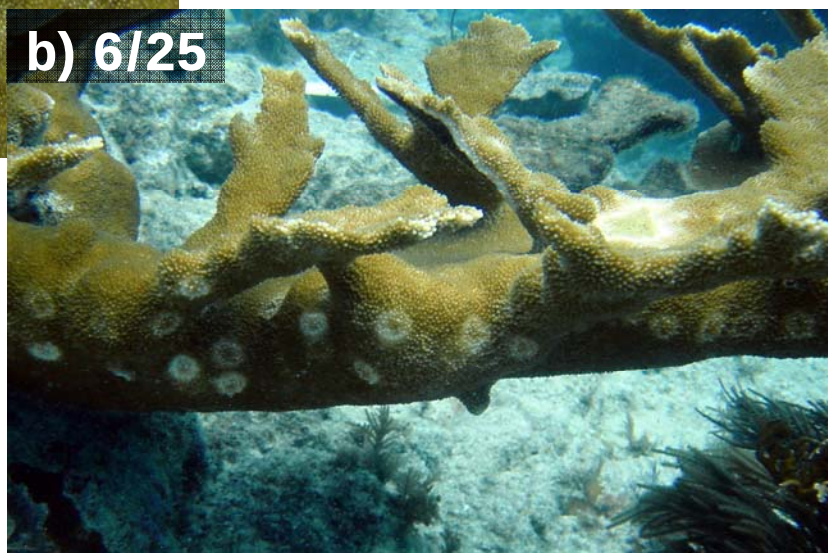
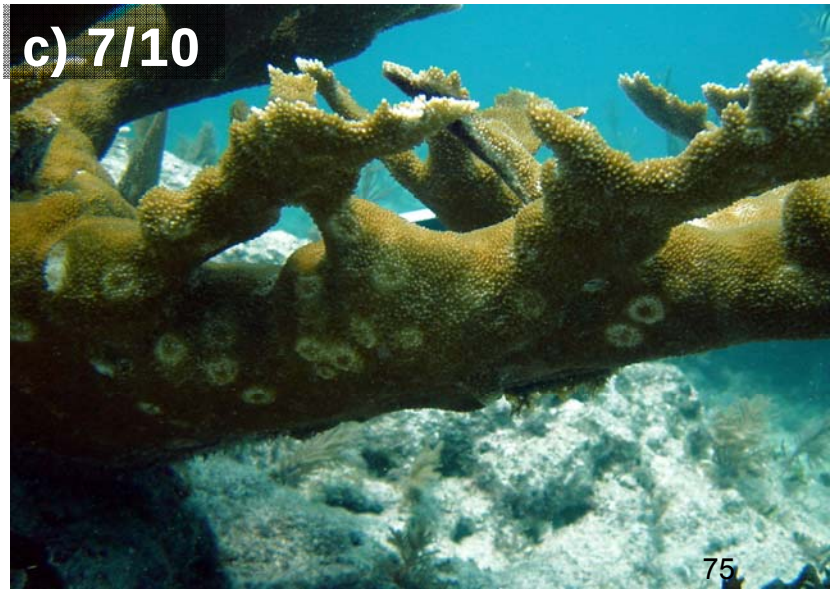
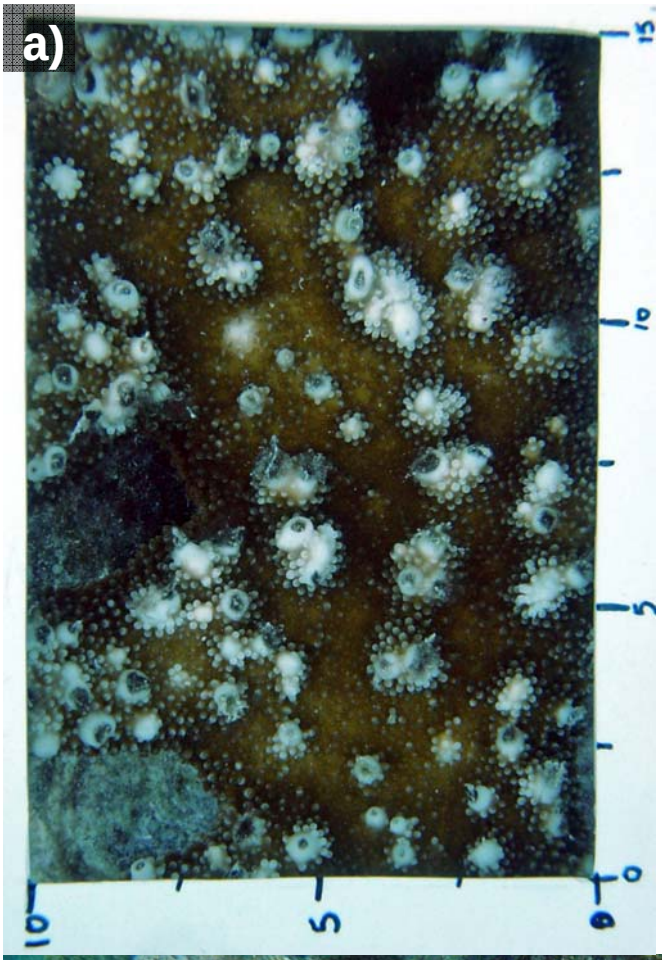


Figure 39. Ring-shaped bleached areas are thought to be caused by yellowtail damselfish bites. These marks routinely appear and heal on a colony b) After 2 weeks some of the rings present in frame b are still present while others have disappeared and new rings have appeared.







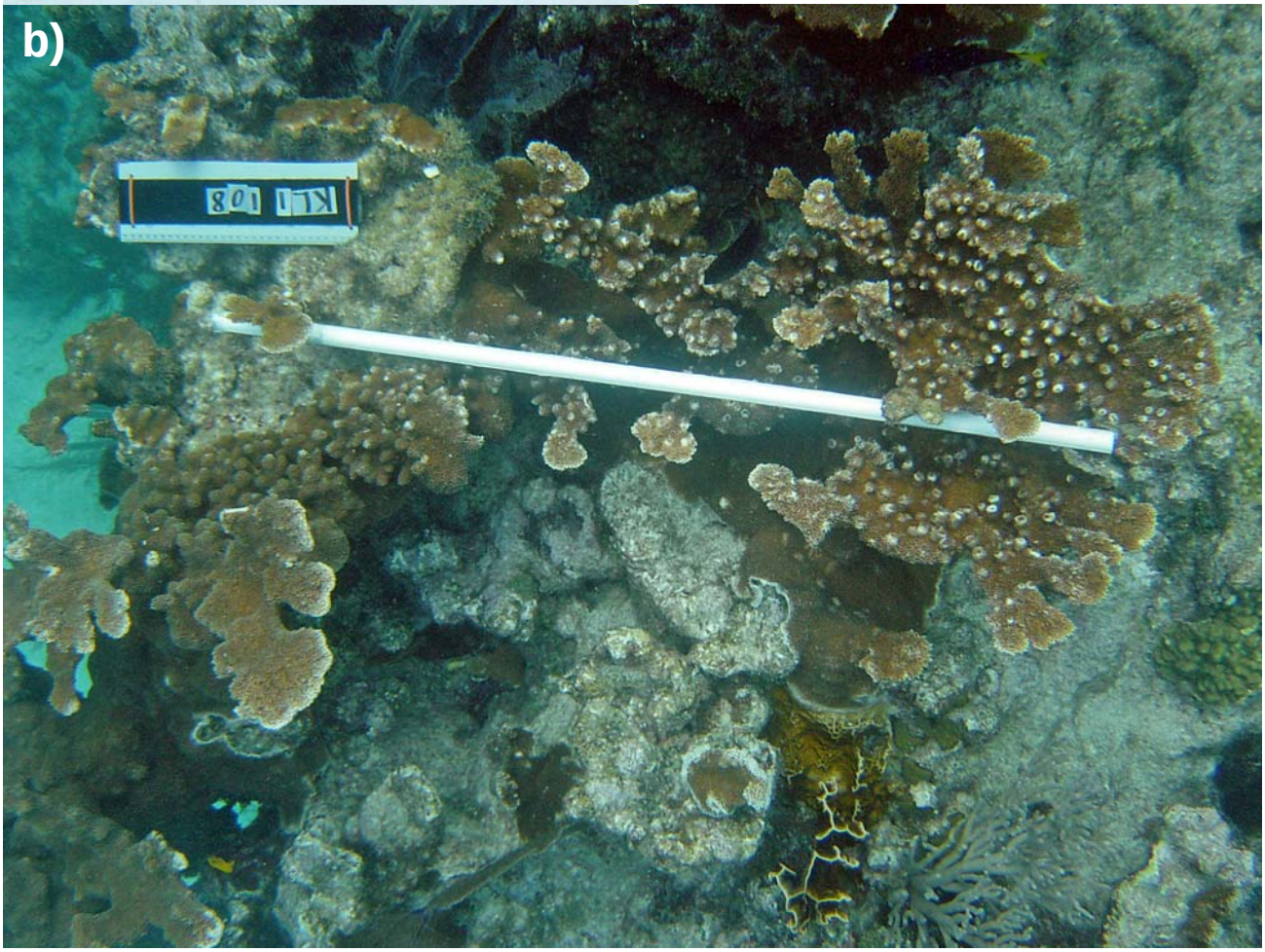


Figure 40. Threespot Damselfish maintain territories on *A. palmata* (b). They repeatedly bite the tissue, clearing space for algal growth. As tissue begins to heal over the bite, it is repeatedly interrupted by more bites. The result is a 'chimney' (a) which is a small tube like protuberance, often open at the top and capped with algae. The tissue at the top of the chimney is often white like other areas of new *Acropora* growth. The freshly bitten areas are also white but are flush with the surrounding tissue.

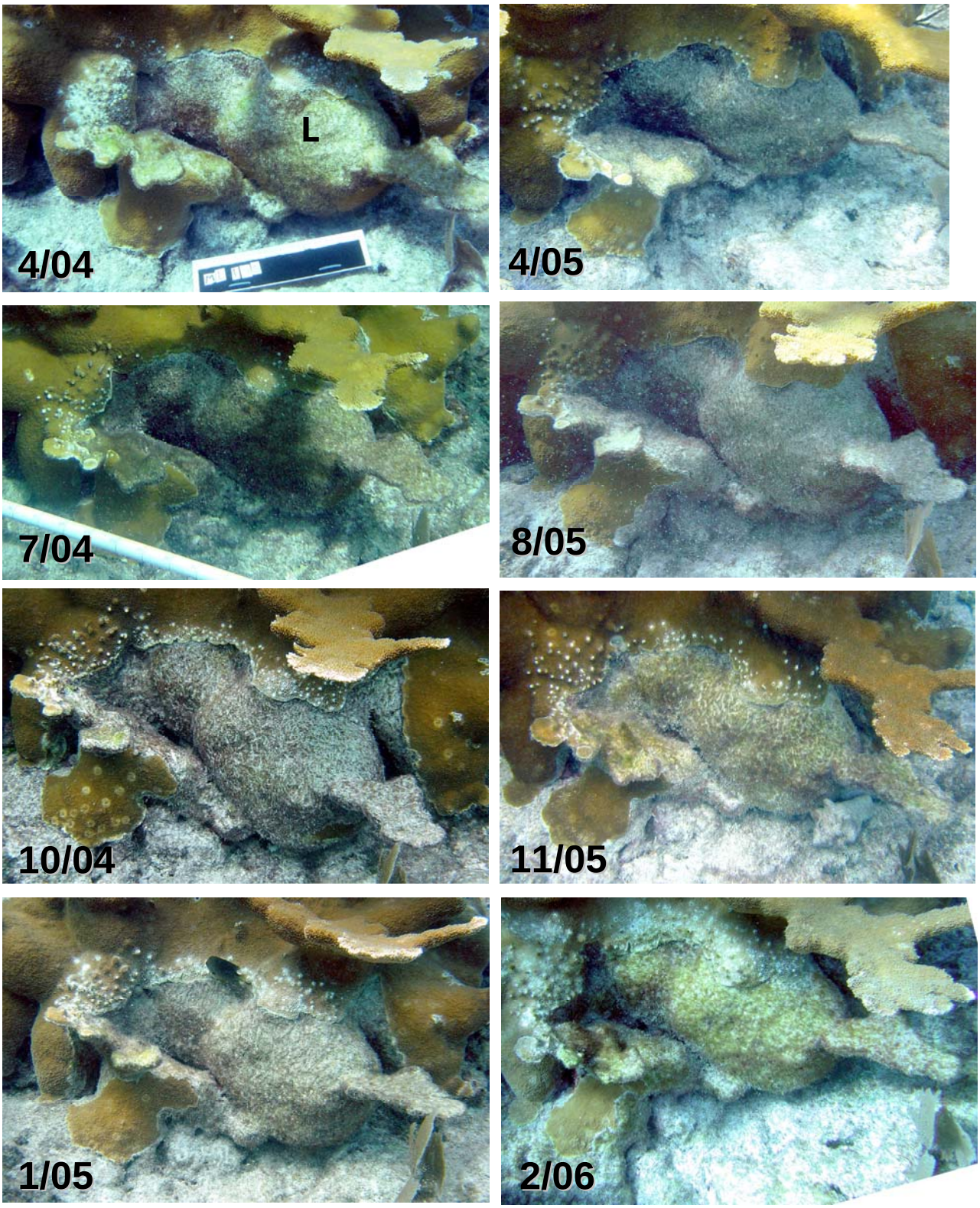


Figure 41. Time series of a threespot damselfish territory over a two year period. A few chimneys are present around the margins, and though they change in number at each survey, the overall impact on the *Acropora* colony remains fairly constant. The chimneys are surrounding the damselfish's 'lawn' (L) which is an algal turf covered part of the colony. While we can't assume that the fish was the original cause of the tissue loss on this colony, it is preventing regrowth of tissue in that area thus the whole area, lawn + chimneys should be included when ranking the impact on this colony. Note that this territory is located at the base of a colony which is somewhat unusual).

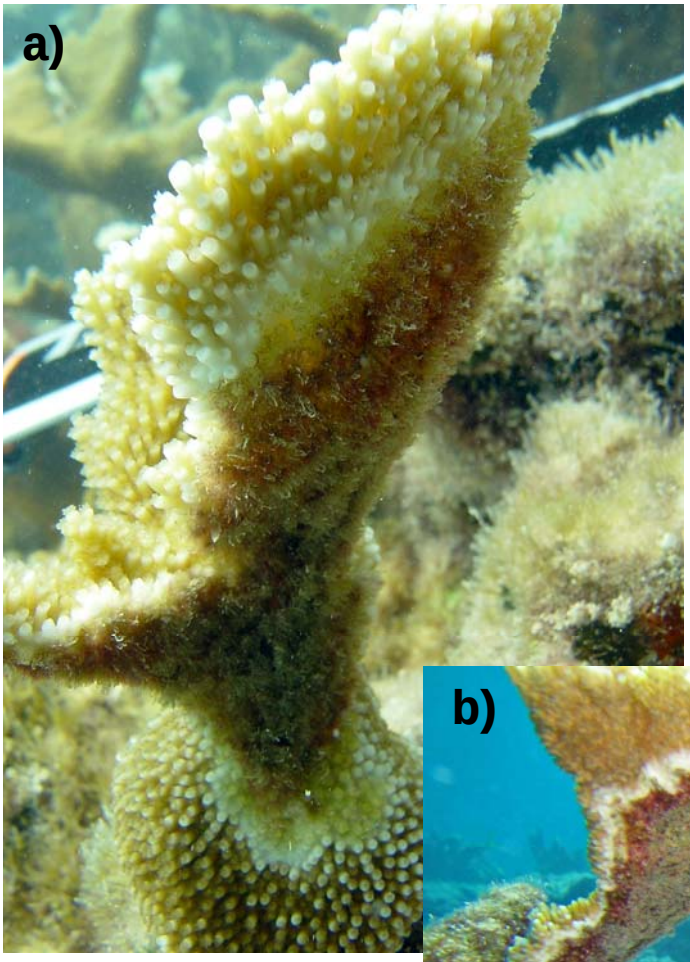
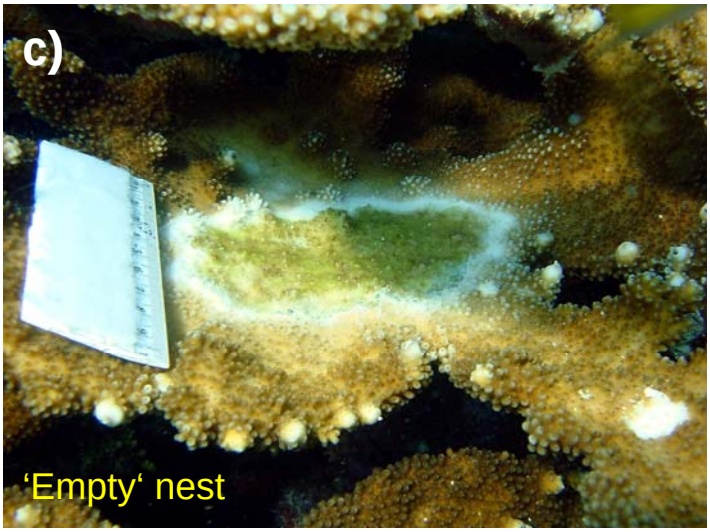
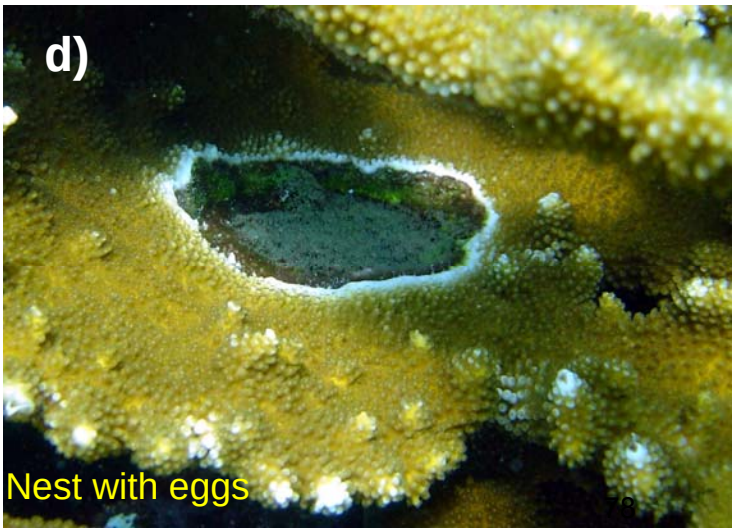


Figure 42. Male threespot damselfish guard eggs laid within their territory. Nests are typically on the underside of a branch and surrounded by live *Acropora* tissue (a-b). A given nest seems to be used repeatedly, frame c and d were taken more than a year apart. When eggs are present the nest will have an grey iridescent cast, often over a very fine deep red filamentous algae turf. The area occupied by a nest should be included in the damselfish impact rank, though it is usually minor compared to the chimney and lawn impacts.





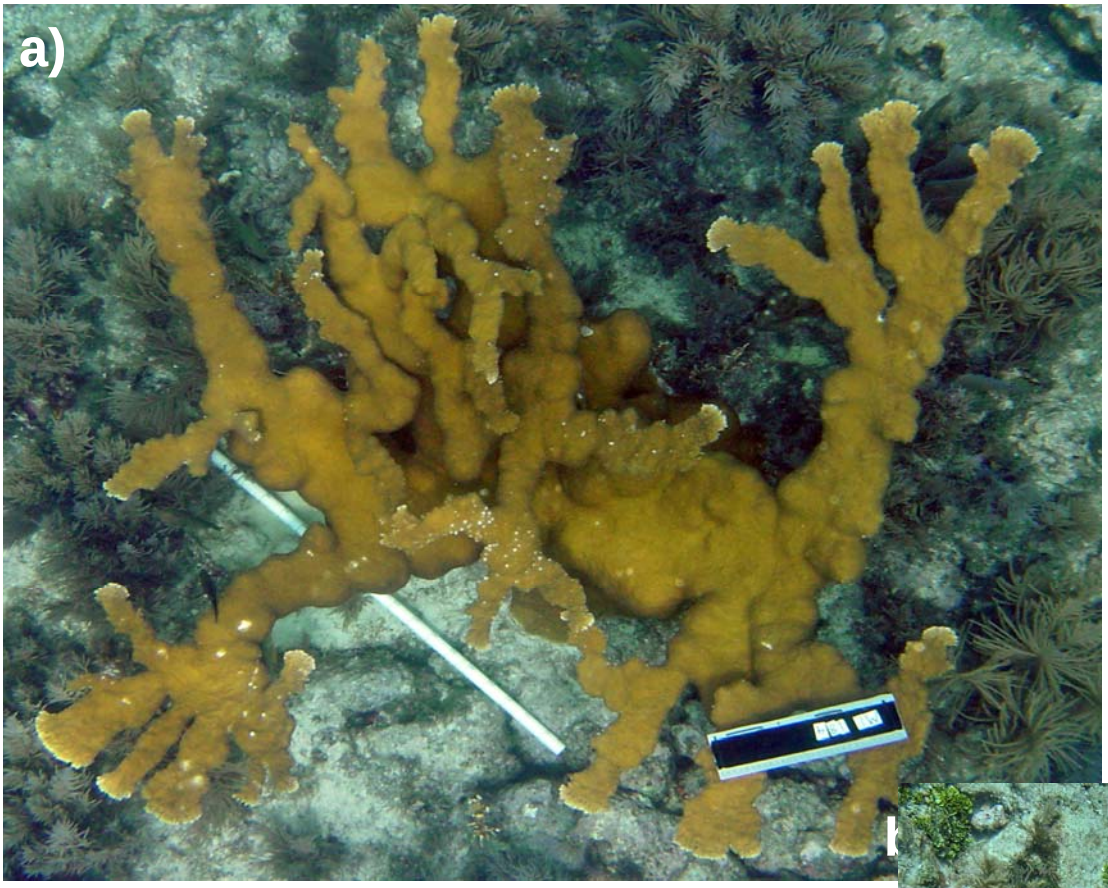
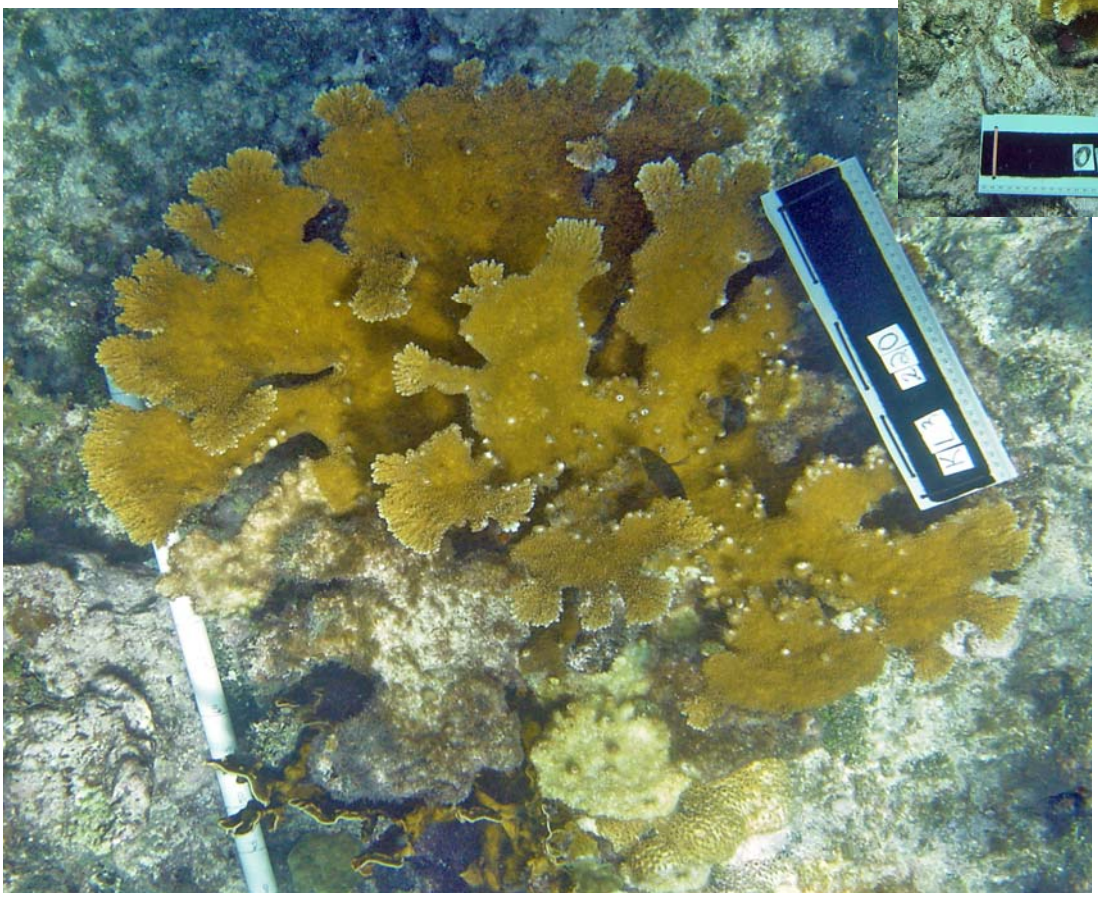


Figure 43. Examples of damselfish impact with rank=1 (a-b) and rank=2 (c).



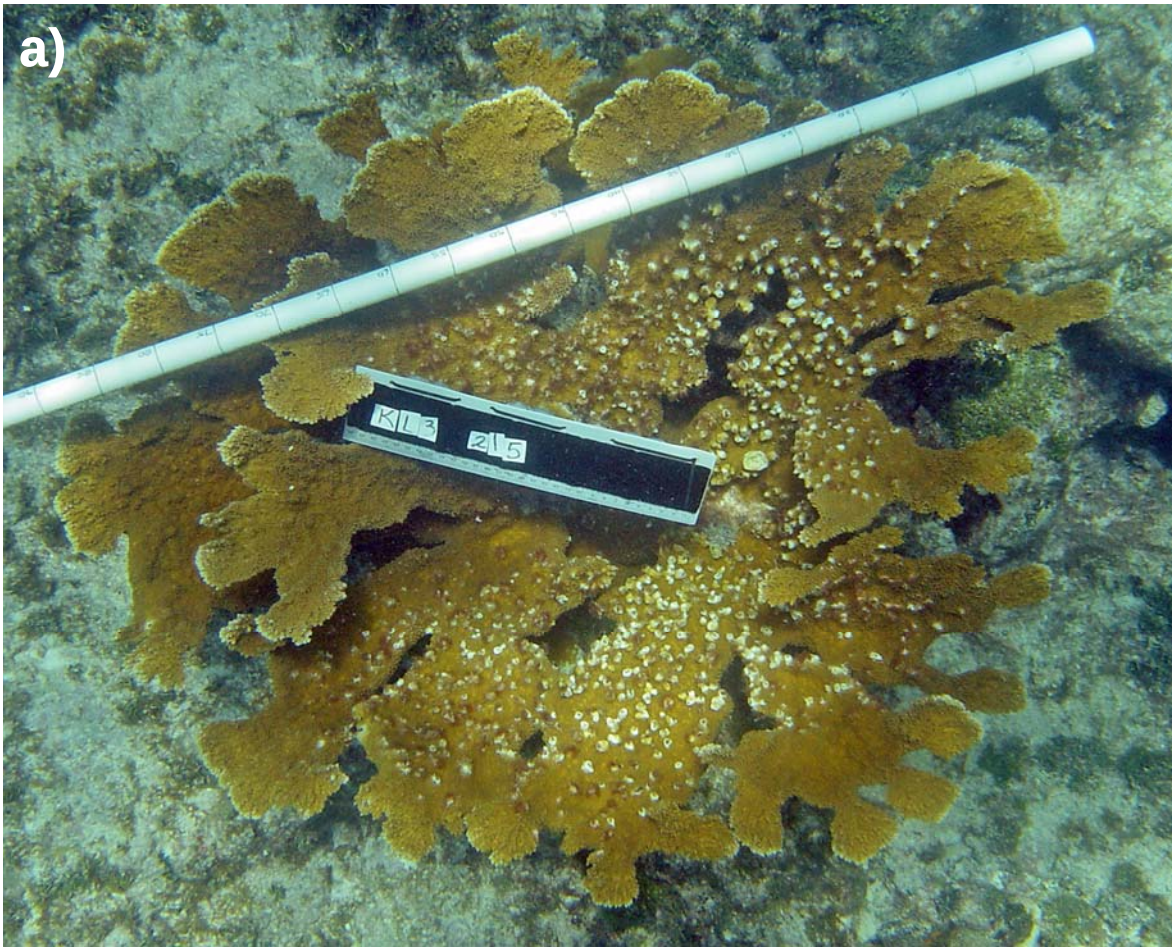


Figure 44. Examples of damselfish impact ranks 3 (a-b) and 4 (c).

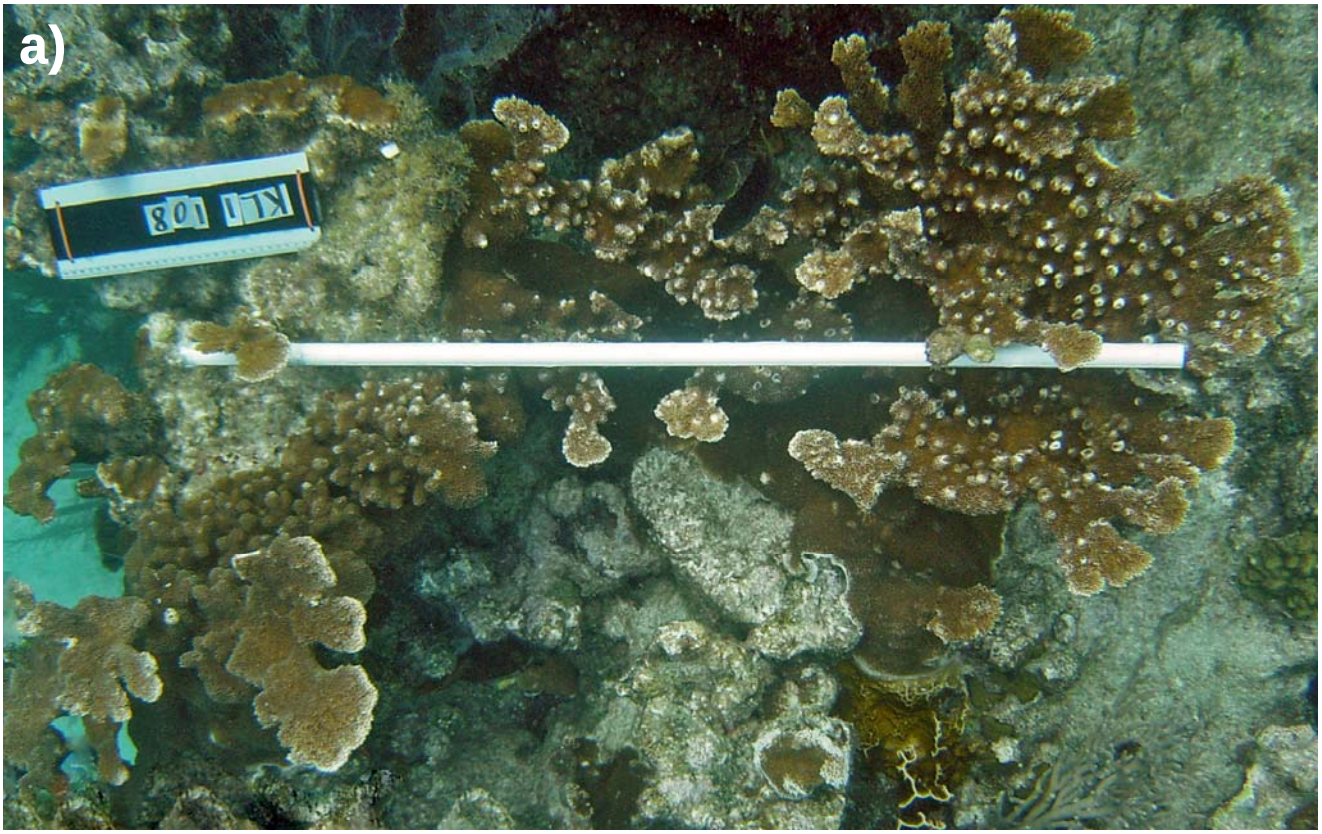
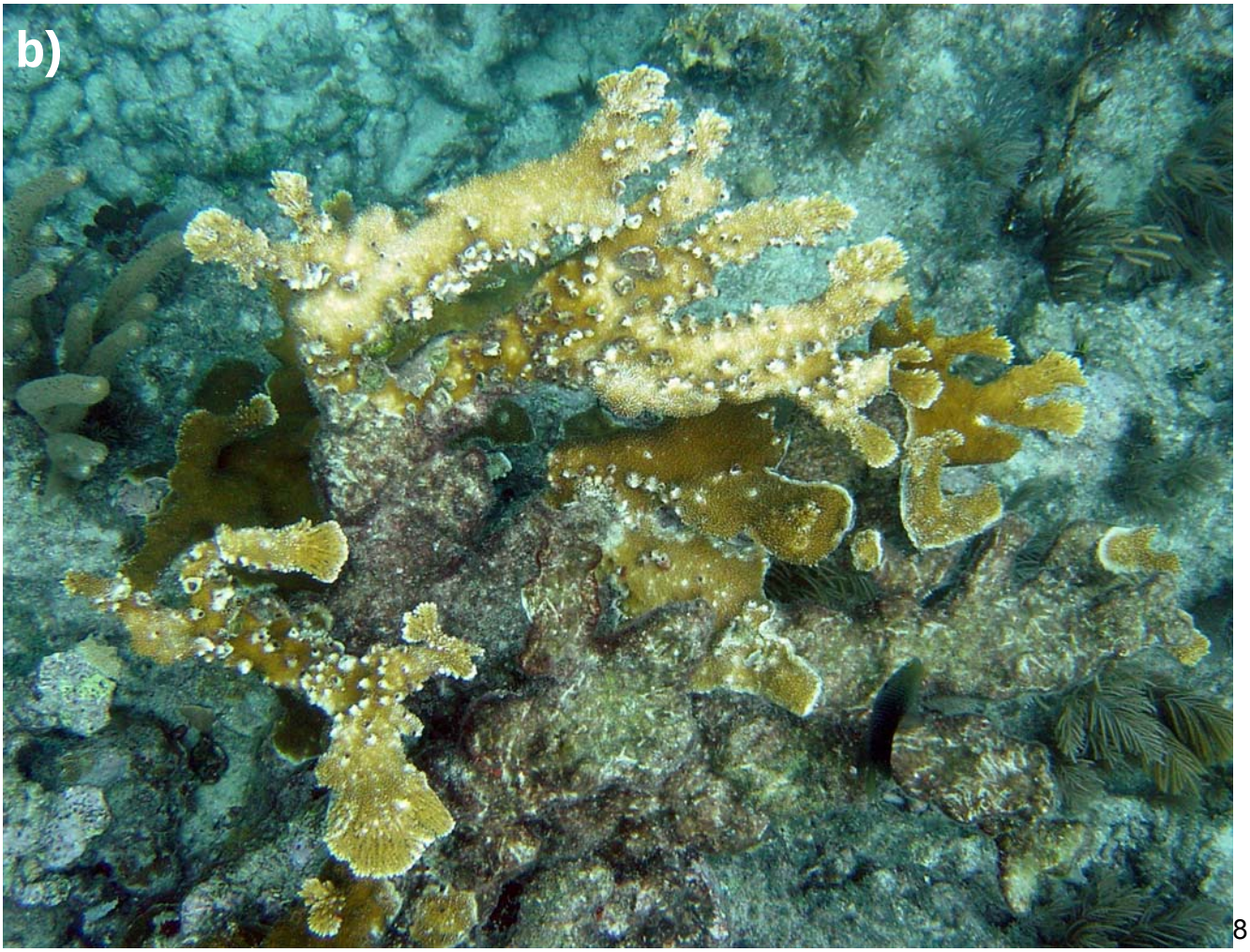


Figure 45. Examples of damselfish impact rank 5.



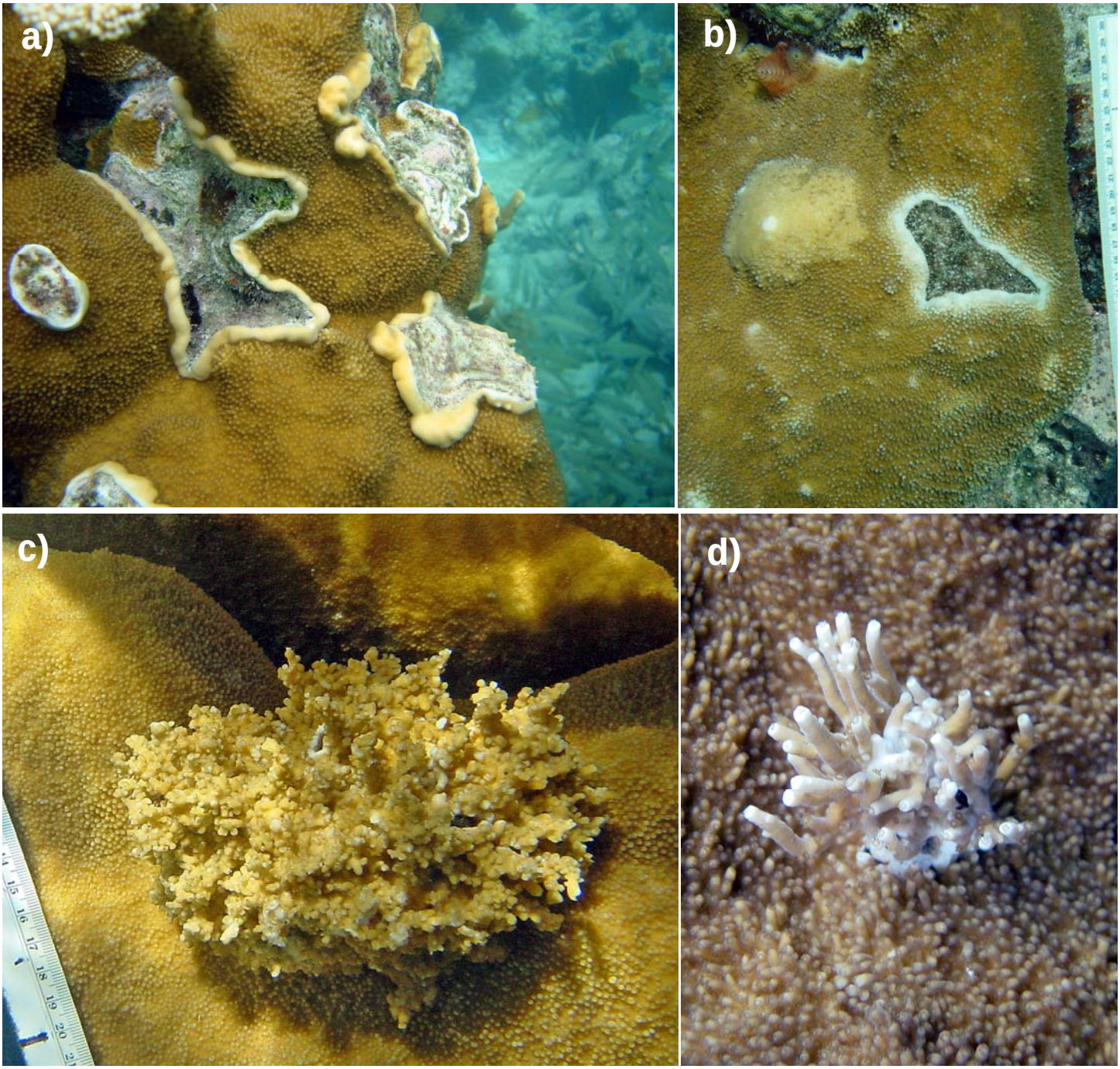


Figure 46. Irregular growth that is relatively smooth and depleted in zooxanthellae is termed neoplastic growth (a-b) and recorded as 'NP' in the skeletal anomaly column of the field sheet. Irregular growth that has zooxanthellae and defined, albeit abnormal, corallites is termed hyperplastic growth and recorded as 'HP' (c-e).

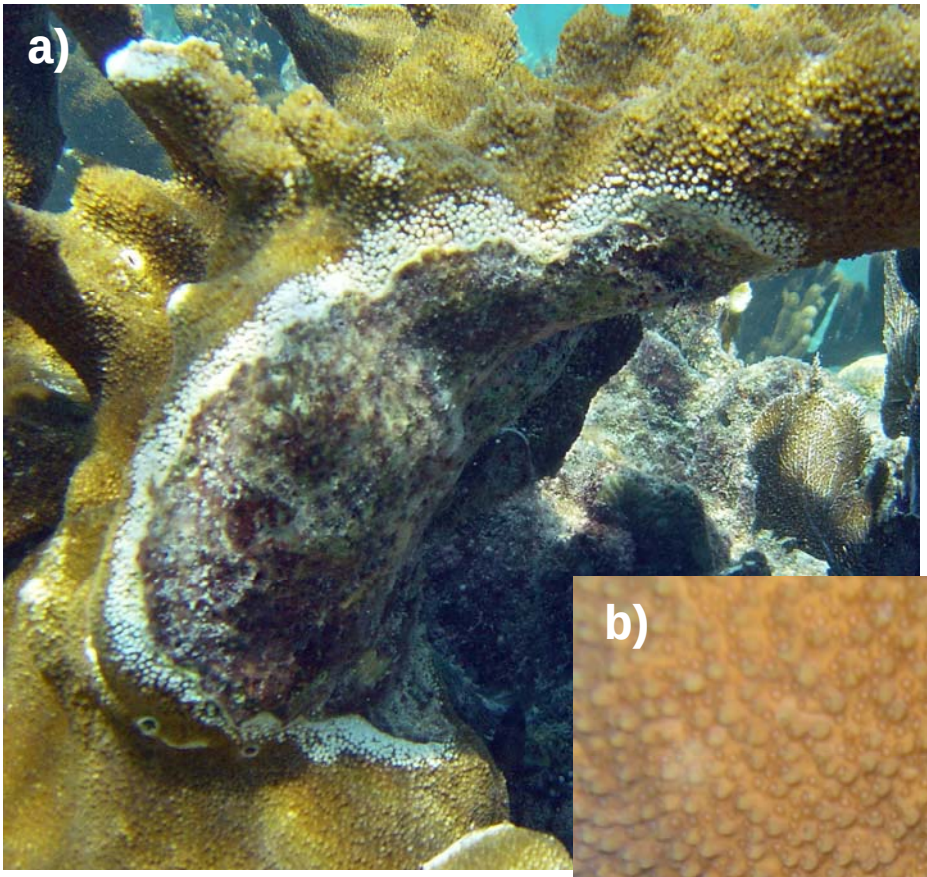
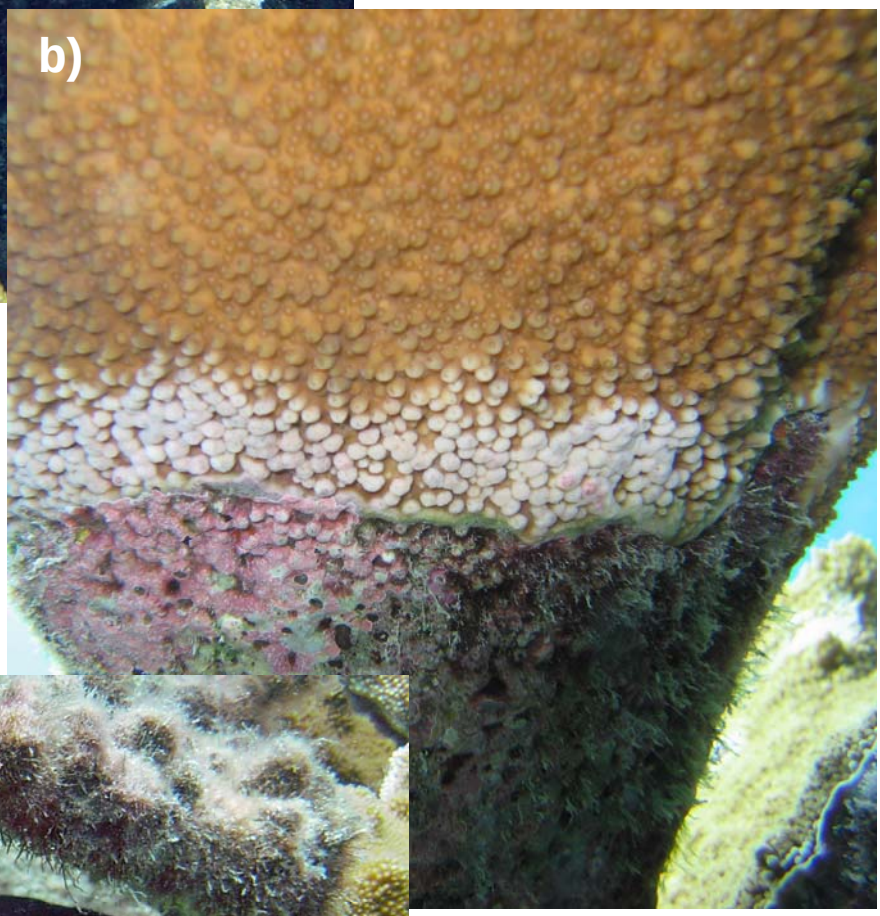


Figure 47. Colonies are occasionally observed with a band of white, densely packed of corallites. There is no recent mortality associated with the bands however it gradually retreats across the colony. These bands are closely associated with yellowtail damselfish (c), and may be the result of repeated nesting activity.



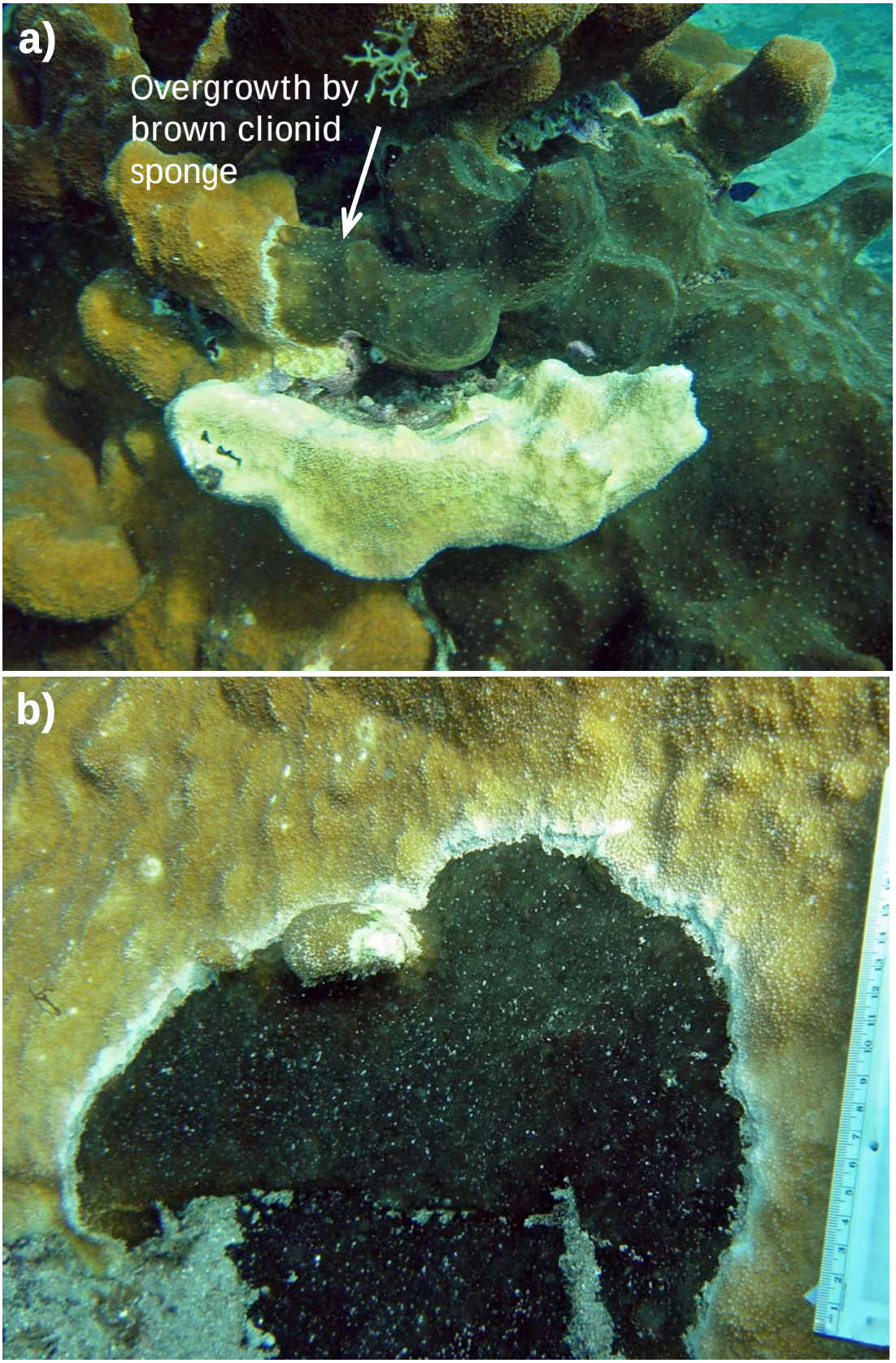


Figure 48. *Cliona langae* is a very thin encrusting sponge that aggressively competes with or invades coral tissue and can readily overgrow it (a-b). It has a velvet feel and is chocolate to olive brown in color. Note that the white tissue at the margin is not recent mortality. It is more accurately characterized as a tissue rejection response.

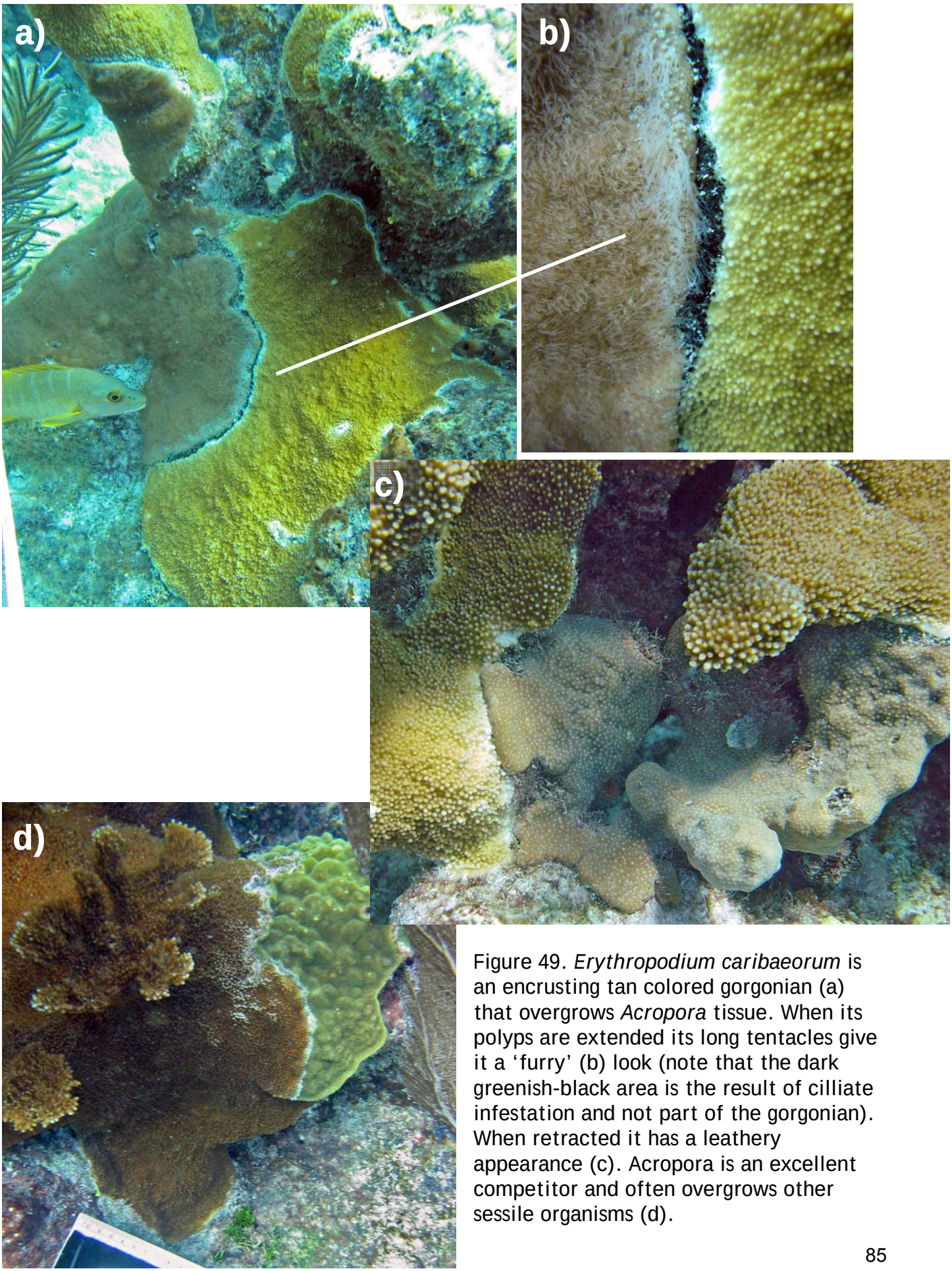


Figure 49. *Erythropodium caribaeorum* is an encrusting tan colored gorgonian (a) that overgrows *Acropora* tissue. When its polyps are extended its long tentacles give it a 'furry' (b) look (note that the dark greenish-black area is the result of cilliate infestation and not part of the gorgonian). When retracted it has a leathery appearance (c). Acropora is an excellent competitor and often overgrows other sessile organisms (d).

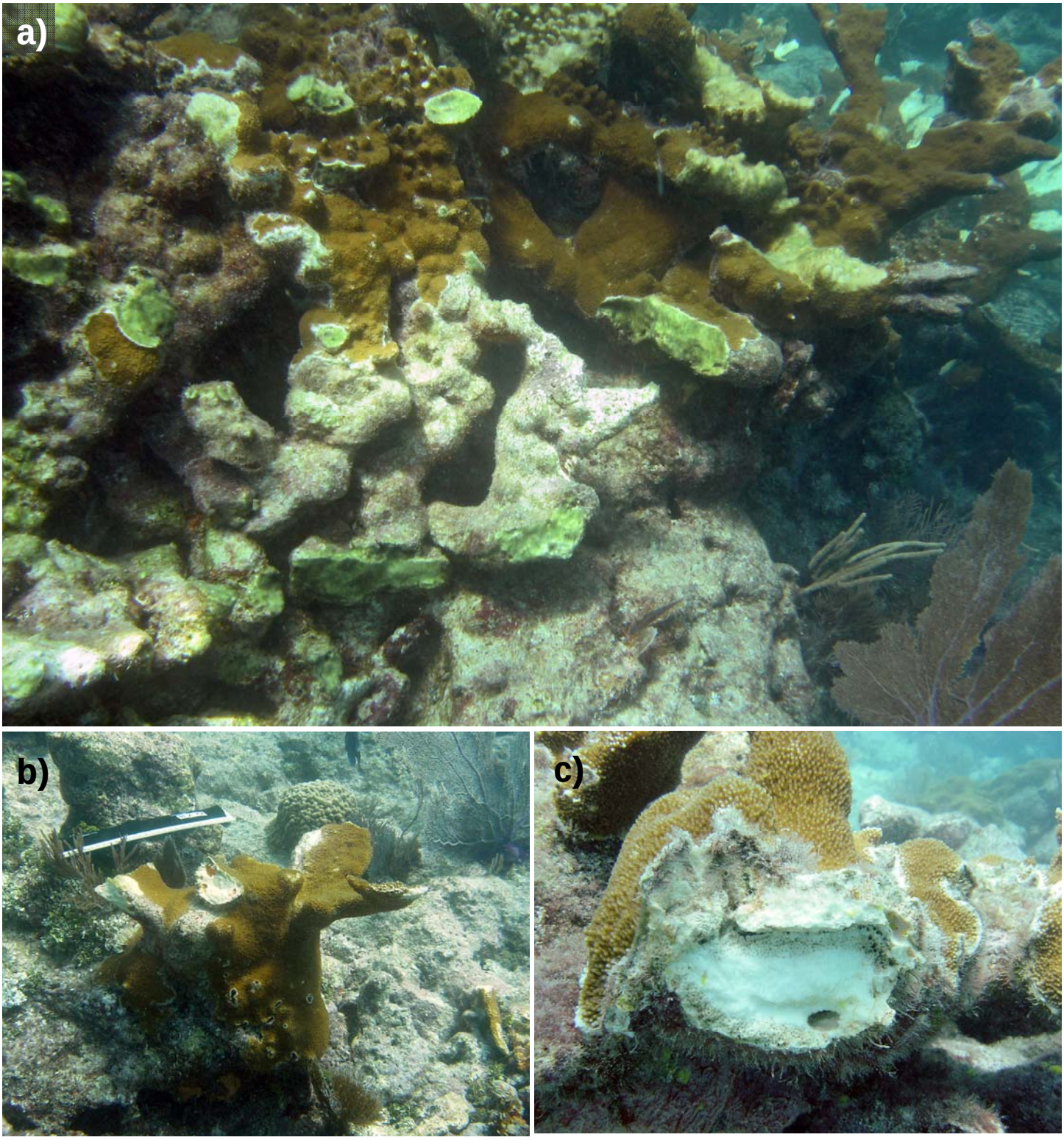


Figure 50. Fragmentation is an important aspect of *Acropora* life history, allowing rapid clonal reproduction. Fragmentation is scored as the number and ranked size (diameter) of branches that have broken off, as determined by examination of the broken scars on the focal colony. In the top picture, the scars have a greenish cast due to colonization of algal turf following an acute disturbance event.

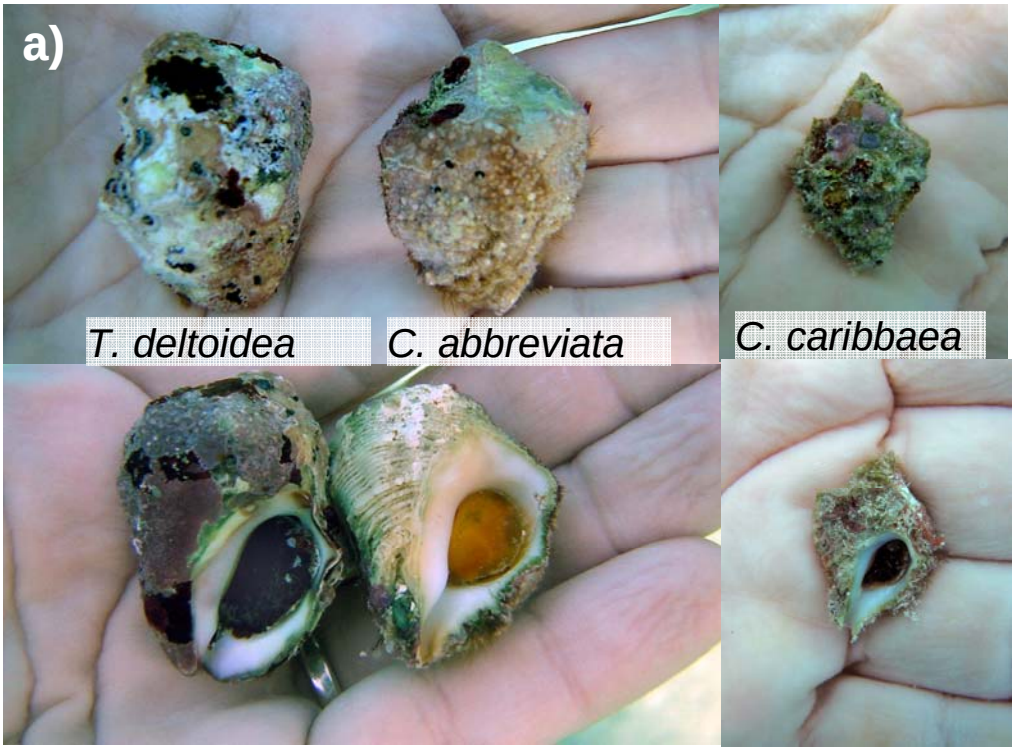


Figure 51. *Coralliophila abbreviata* and *C caribbaea* are corallivores commonly found feeding or resting on *Acropora* colonies. It is important to distinguish them from other common snails (e.g. *T. deltoidea)* that do not feed on coral so that an accurate census of these predators can be done. *C. abbreviata* is more common on Acropora than *C. caribbaea*. These three species can be distinguished based on some physical traits and habits.

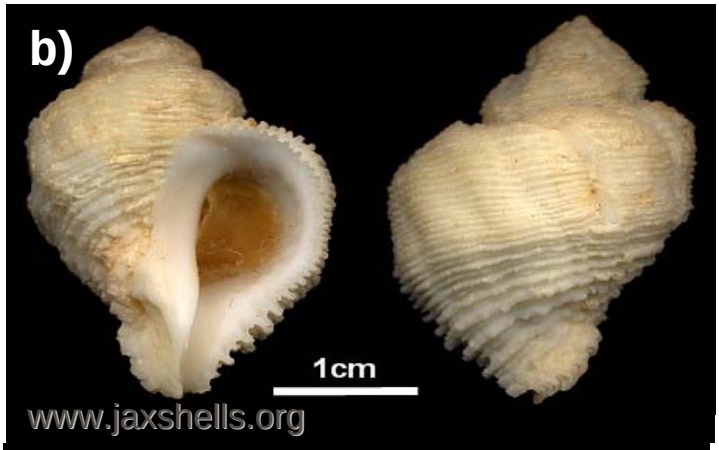


Figure 51b. *Coralliophila abbreviata*: Feeds on coral tissue typically found in a small group associated with live tissue or a feeding scar. Shell has fine ridges that are sometimes visible through the encrusting algae. Bright yellow body and gold operculum. *C. caribbaea* (shown in frame a) is smaller at full size and has a white or tan body with a burgundy operculum. They are also found in a small group and near a feeding scar.

Figure 51c. *Thais deltoidea*: Feeds on other snails and is commonly found on reef substrate including the dead portions of *Acropora* colonies. Only coincidentally associated with live coral tissue. Dull knobs on the outer whorl that are subtly visible through the encrusting algae. Burgundy body and very dark purple/brown operculum.

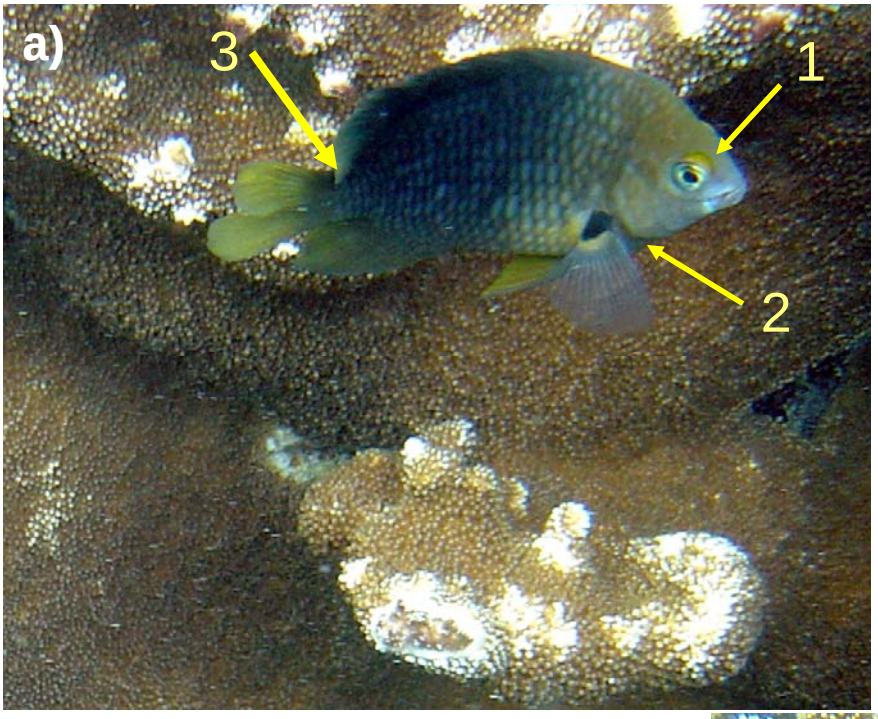
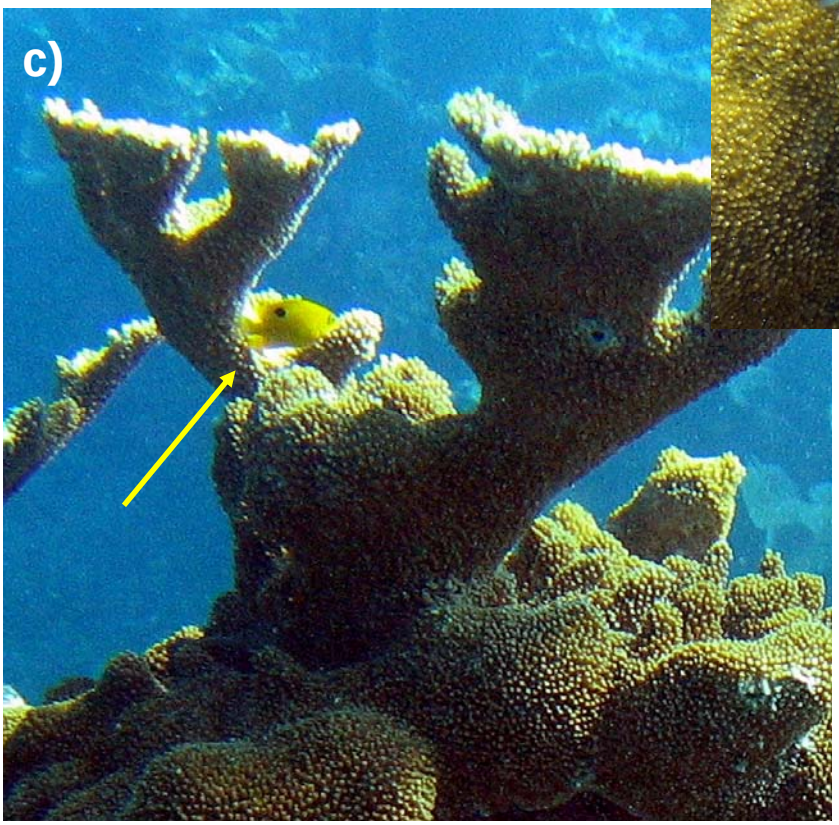


Figure 52. Threespot damselfish (*Stegastes planifrons*) adults are similar in appearance to Dusky, Beaugregory & Cocoa damselfishes. Their body color varies from light to dark yellowish brown. Their most reliable distinguishing features are the spots & behavior: a) They have three spots: 1. bright yellow eyelid; 2. black & yellow patch at base of pectoral fin; 3. black patch at base of caudal fin. At least one of these will be visible (note that the cocoa has a similar spot on the base of the caudal fin). c) They are very territorial and often will aggressively approach observers, particularly around egg masses. Other species of damselfish tend to be less aggressive but this varies within a species. c) Juveniles are bright yellow with 2 black spots & not aggressive.



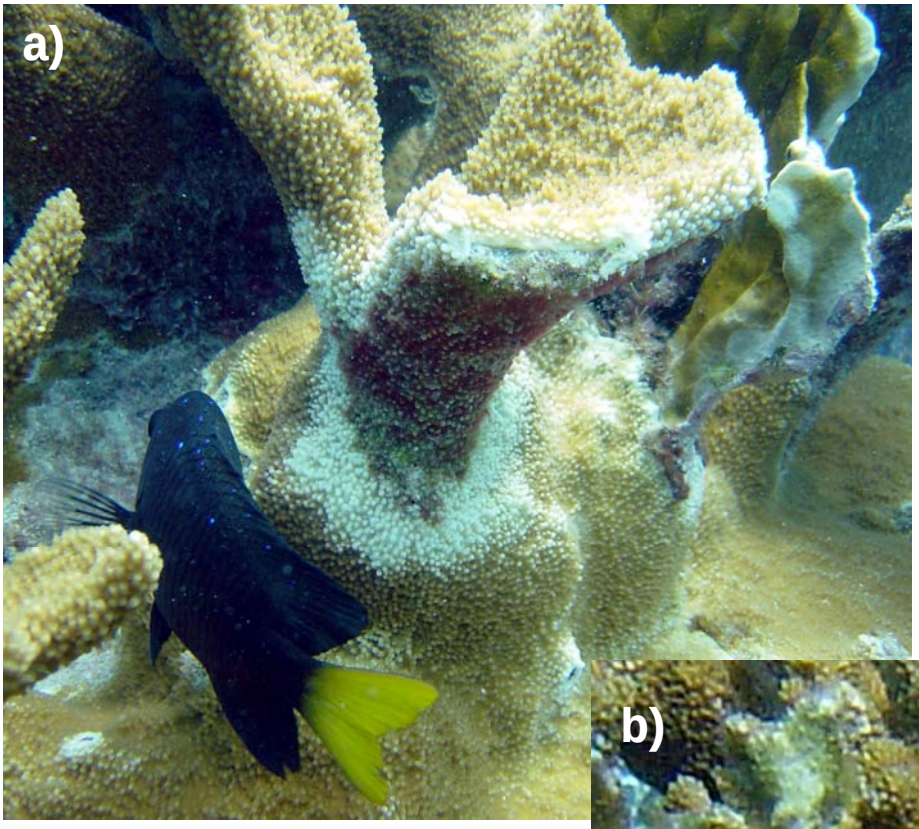
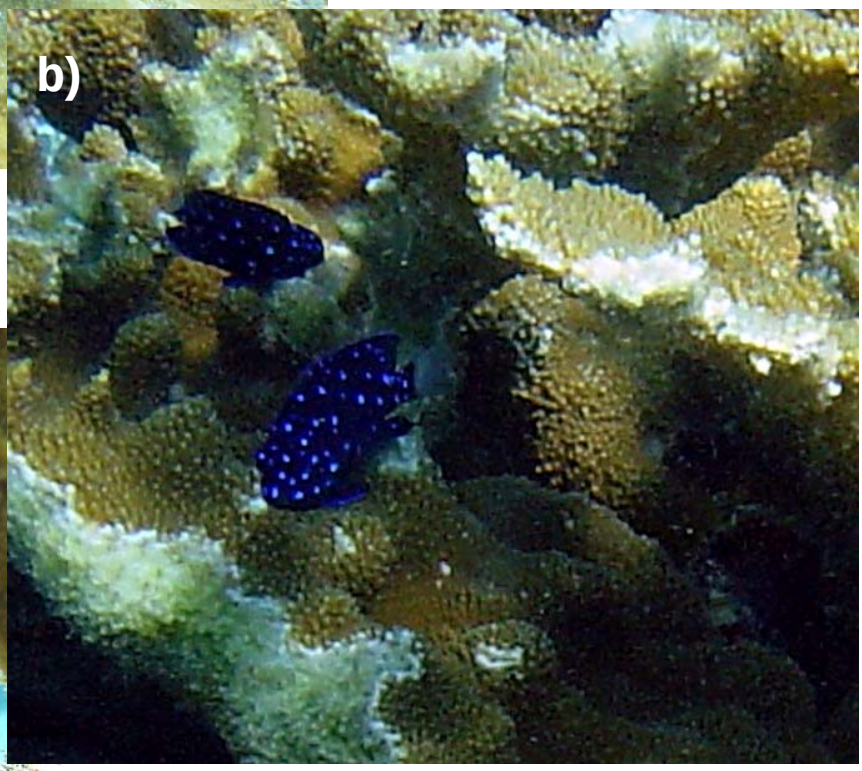
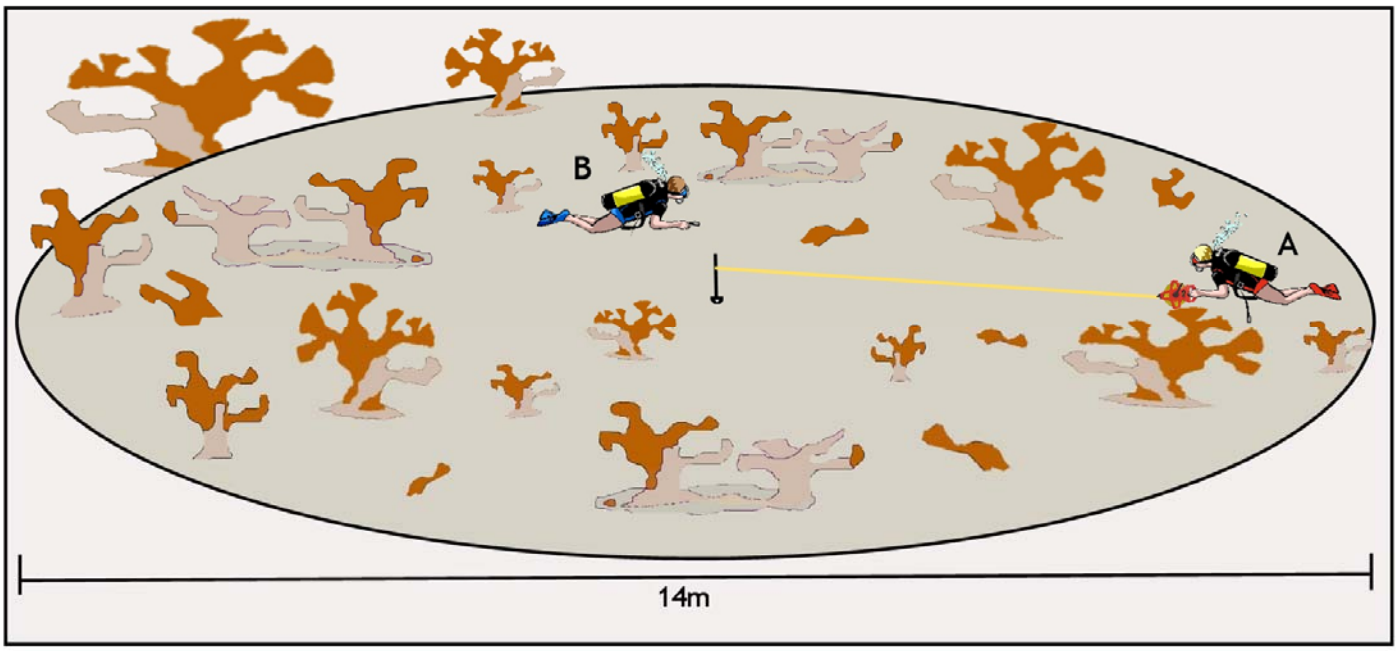


Figure 53. Yellowtail Damselfish (*Microspathodon chrysurus*) are easily distinguished by their yellow tail as adults (a) and vivid blue spots as juveniles (b). They are often found sheltered among the branches (c). While they are closely associated with *A. palmata*, they often 'patrol' a territory that may include more than one colony. Frame a also shows a nest, note that it is surrounded by a white band (see also Figure 47) that should not be confused with white band disease (see Figure 20).



a)



b)

Annual Plot Map (DiverA: Distance from stake, colony description)

Plot_____ Date_____

Untagged Col #	Deg	Dist	L	W	H	% Live	Colony Type**	Untagged Col #	Deg	Dist	L	W	H	% Live	Colony Type**
X1								X17							
X2								X18							
X3								X19							
X4								X20							
X5								X21							

Confirm previously measured coordinates:

Tag #	Deg	Dist

c)

Annual Plot Map (Diver B: compass heading)

Plot_____ Date_____

Untagged Col #	Deg	Untagged Col #	Deg
X1		X16	
X2		X17	
X3		X18	
X4		X19	
X5		X20	
X6		X21	

Confirm previously measured coordinates:

Tag #	Deg

Plot_____ Date_____

Untagged Col #	Deg	Untagged Col #	Deg
X1		X16	
X2		X17	
X3		X18	
X4		X19	
X5		X20	
X6		X21	

Confirm previously measured coordinates:

Tag #	Deg

Figure 54. a) Schematic illustration of the plot mapping procedure. b) Diver A records colony information and c) Diver B reads the bearing to each colony designated by Diver A.

a)

Diver A Quarterly colony condition assessment

Site_____ Plot _____ Date_____

X	Tag #	L	W	H	% Live	Recent mort*				Bleaching**			Damselfish Impact (Rank)	Skel anoms***	Notes
						Rank	Types			Rank	Types				
							1°	2°	3°		1°	2°			

b)

Diver B Quarterly snail and fish survey

Site_____ Plot_____ Date_____

X	Tag #	Degree	Distance	# Snails	# 3-Spot damsels		# Ylwtail damsels		# Branch Breaks				Notes
					Ad	Juv	Ad	Juv	sm	med	lg	xlg	

Figure 55. Data collection is divided between a) diver A and b) Diver B for the quarterly surveys.

APPENDICES

Diver A: Initial Colony Survey

Site_____ Plot _____ Date_____

X	Tag #	L	W	H	% Live	Recent mort*				Bleaching**			Damselfish Impact (Rank)	Skel anoms***	Overgrowth	# Snails	# 3-Spot damsels		# Yellowtail damsels		# Branch Breaks				Notes
						Rank	Types			Rank	Types														
							1°	2°	3°		1°	2°					Ad	Juv	Ad	Juv	sm	med	lg	xlg	

Additional Notes:

* **Recent mortality types:** WPx= white pox, WBD= white-band, RTL= rapid tissue loss, FS=snail feeding scar, CB=cilliate band, PFB= parrrotfish bites, CL=Cliona lesion, FP=fish poop, Unkn= unknown ** **Bleaching types:** Bl= bleached, R= rings, W= wedgies ***Skeletal anomolies: Hp=hyperplasia, Np=neoplasia, DWP=dense white polyps, Unkn=Unknown

Diver B Tagged Colony Selection

Site_____ Date_____

Stake Depth: _____

Stake Waypoint#

Plot	Colony	Tag #	Deg	Dist	Notes (tag loc., depth, etc)
1	1				
1	2				
1	3				
1	4				
1	5				
1	6				
1	7				
1	8				
1	9				
1	10				
1	11				
1	12				

Site_____ Date_____

Stake Depth: _____

Stake Waypoint#

Plot	Colony	Tag #	Deg	Dist	Notes (tag loc., depth, etc)
2	1				
2	2				
2	3				
2	4				
2	5				
2	6				
2	7				
2	8				
2	9				
2	10				
2	11				
2	12				

Notes:

Random Coordinates:

Plot 1		Plot 2	
Deg	Dist	Deg	Dist
75	3.0	300	4.5
135	4.5	35	3.0
260	5.0	105	3.5
135	1.5	215	0.5
110	3.5	240	4.0
190	0.5	30	4.5
275	1.5	10	0.5
175	4.0	85	2.0
140	2.5	125	7.0
200	6.5	225	1.5
225	5.0	310	2.5
225	7.0	30	1.0
270	4.5	280	3.5
40	5.5	125	6.5
280	1.5	245	6.5
345	1.0	150	5.0
60	3.0	15	5.0
30	1.5	20	5.0
175	4.0	260	1.5
115	3.5	290	2.0
35	2.0	20	0.5
235	6.0	20	6.5
85	5.0	300	3.0
40	2.0	215	0.5
110	2.0	80	0.5
190	6.0	305	3.5
0	1.5	230	6.5
150	2.5	20	4.0
225	5.5	65	7.0
110	7.0	300	0.5
230	7.0	340	2.5
55	4.5	50	5.5
205	1.5	30	7.0
85	2.5	215	3.5
185	2.5	275	2.5
335	5.0	195	1.5
295	2.0	20	2.0
285	1.0	105	6.5
165	2.0	350	7.0
195	7.0	50	6.5
155	4.0	80	4.0
205	4.5	5	1.0
150	3.5	155	3.0
320	6.0	170	4.5
325	4.5	295	1.0
295	1.5	135	3.0
220	5.5	55	4.0

Diver A Quarterly Colony Condition Assessment

Site_____ Plot _____ Date_____

X	Tag #	L	W	H	% Live	Recent mort*				Bleaching**			Damselfish Impact (Rank)	Skel anoms***	Notes
						Rank	Types			Rank	Types				
							1°	2°	3°		1°	2°			

Site_____ Plot _____ Date_____

X	Tag #	L	W	H	% Live	Recent mort*				Bleaching**			Damselfish Impact (Rank)	Skel anoms***	Notes
						Rank	Types			Rank	Types				
							1°	2°	3°		1°	2°			

* **Recent mortality types:** WPx= white pox, WBD= white-band, RTL= rapid tissue loss, FS=snail feeding scar, CB=cilliate band, PFB= parrotfish bites, CL=Cliona lesion, FP=fish poop, Unkn= unknown ** **Bleaching types:** Bl= bleached, R= rings, W= wedgies ***Skeletal anomolies: Hp=hyperplasia, Np=neo DWP=dense white polyps, Unkn=Unknown

Diver B Quarterly Snail and Fish Census

Site_____ Plot_____ Date_____

X	Tag #	Degree	Distance	# Snails	# 3-Spot damsels		# Ylwtail damsels		# Branch Breaks				Notes
					Ad	Juv	Ad	Juv	sm	med	lg	xlg	

Site_____ Plot_____ Date_____

X	Tag #	Degree	Distance	# Snails	# 3-Spot damsels		# Ylwtail damsels		# Branch Breaks				Notes
					Ad	Juv	Ad	Juv	sm	med	lg	xlg	

Annual Plot Map (Diver A: Distance from stake, colony description)

Plot_____ Date_____

Untagged Col #	Deg	Dist	L	W	H	% Live	Colony Type**	Untagged Col #	Deg	Dist	L	W	H	% Live	Colony Type**
X1								X17							
X2								X18							
X3								X19							
X4								X20							
X5								X21							
X6								X22							
X7								X23							
X8								X24							
X9								X25							
X10								X26							
X11								X27							
X12								X28							
X13								X29							
X14								X30							
X15								X31							
X16								X32							

Confirm previously measured coordinates:

Tag #	Deg	Dist

LF** Tally:

A-5

Plot_____ Date_____

Untagged Col #	Deg	Dist	L	W	H	% Live	Colony Type**	Untagged Col #	Deg	Dist	L	W	H	% Live	Colony Type**
X1								X17							
X2								X18							
X3								X19							
X4								X20							
X5								X21							
X6								X22							
X7								X23							
X8								X24							
X9								X25							
X10								X26							
X11								X27							
X12								X28							
X13								X29							
X14								X30							
X15								X31							
X16								X32							

Confirm previously measured coordinates:

Tag #	Deg	Dist

LF** Tally:

* Transcribe from Diver 2's data sheet after the dive

**BC= Branched Colony, RC=Remnant Crust, AF= Attached Fragment, SF=Stable Fragment LF=Loose Fragment (Count only)

Annual Plot Map (Diver B: Compass Heading)

Plot_____ Date_____ Confirm previously measured coordinates:

Untagged Col #	Deg	Untagged Col #	Deg	Tag #	Deg
X1		X16			
X2		X17			
X3		X18			
X4		X19			
X5		X20			
X6		X21			
X7		X22			
X8		X23			
X9		X24			
X10		X25			
X11		X26			
X12		X27			
X13		X28			
X14		X29			
X15		X30			

Plot_____ Date_____ Confirm previously measured coordinates:

Untagged Col #	Deg	Untagged Col #	Deg	Tag #	Deg
X1		X16			
X2		X17			
X3		X18			
X4		X19			
X5		X20			
X6		X21			
X7		X22			
X8		X23			
X9		X24			
X10		X25			
X11		X26			
X12		X27			
X13		X28			
X14		X29			
X15		X30			

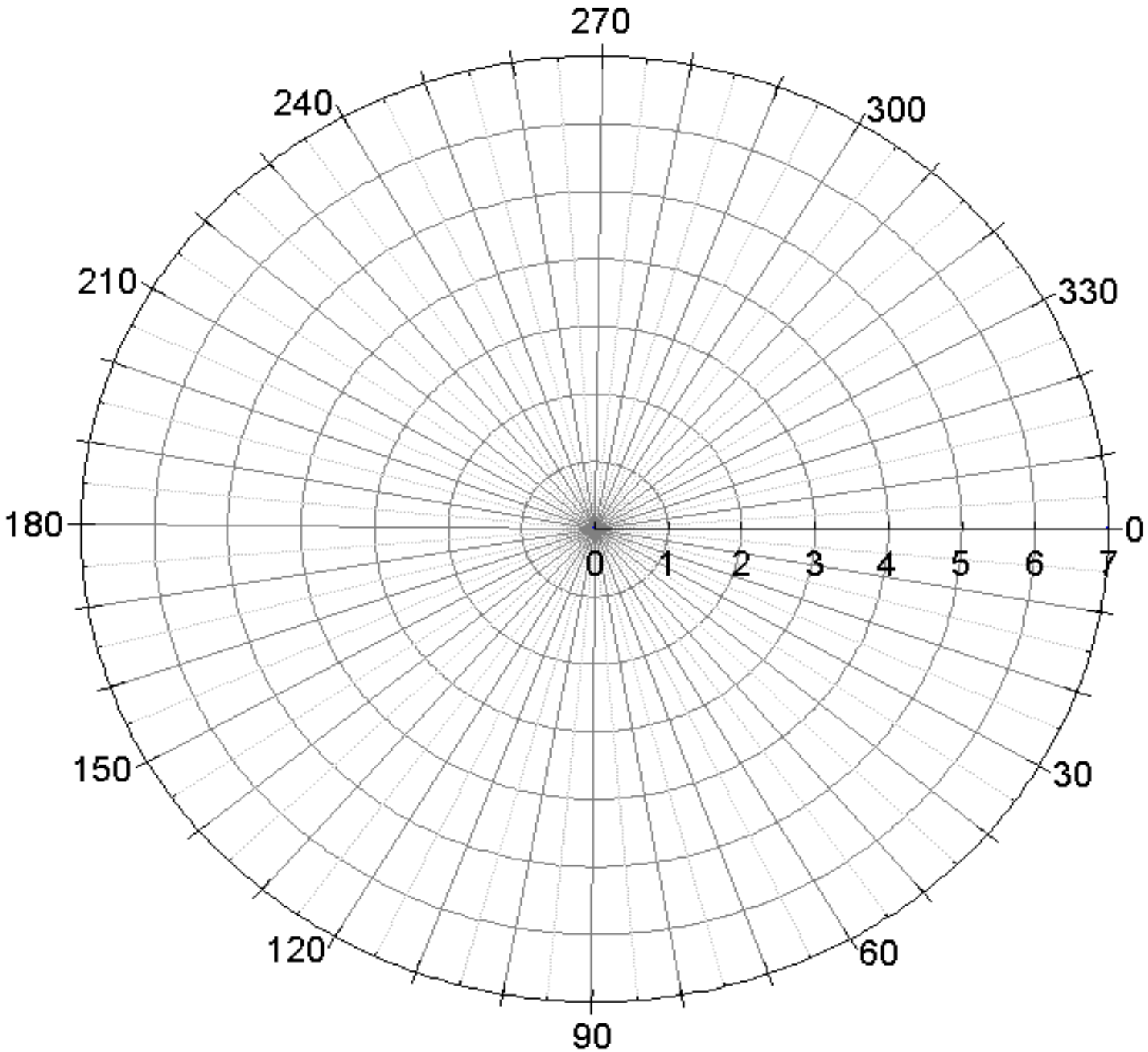
Plot_____ Date_____ Confirm previously measured coordinates:

Untagged Col #	Deg	Untagged Col #	Deg	Tag #	Deg
X1		X16			
X2		X17			
X3		X18			
X4		X19			
X5		X20			
X6		X21			
X7		X22			
X8		X23			
X9		X24			
X10		X25			
X11		X26			
X12		X27			
X13		X28			
X14		X29			
X15		X30			

Plot_____ Date_____ Confirm previously measured coordinates:

Untagged Col #	Deg	Untagged Col #	Deg	Tag #	Deg
X1		X16			
X2		X17			
X3		X18			
X4		X19			
X5		X20			
X6		X21			
X7		X22			
X8		X23			
X9		X24			
X10		X25			
X11		X26			
X12		X27			
X13		X28			
X14		X29			
X15		X30			

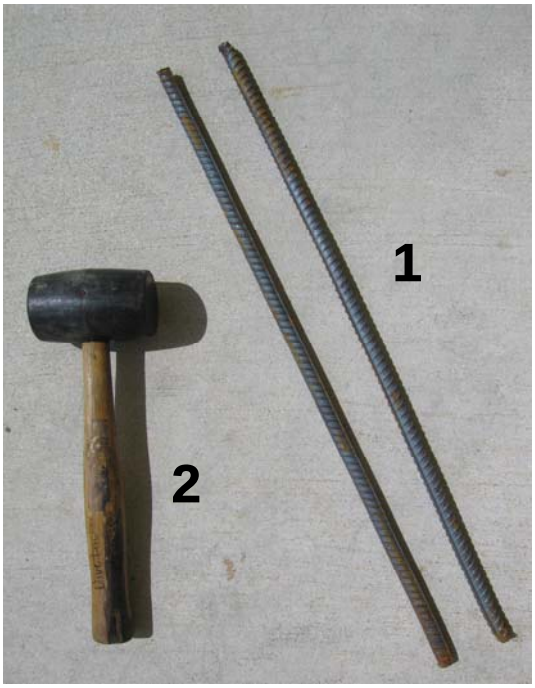
Annual Plot Map **Plot** _____ **Date** _____

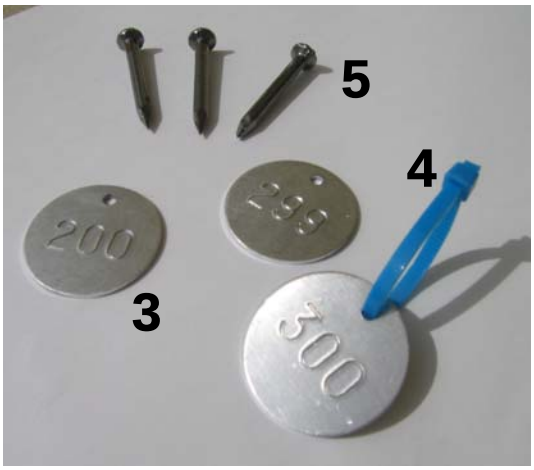


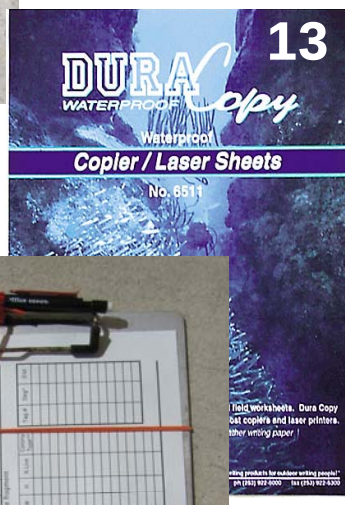
Notes _____

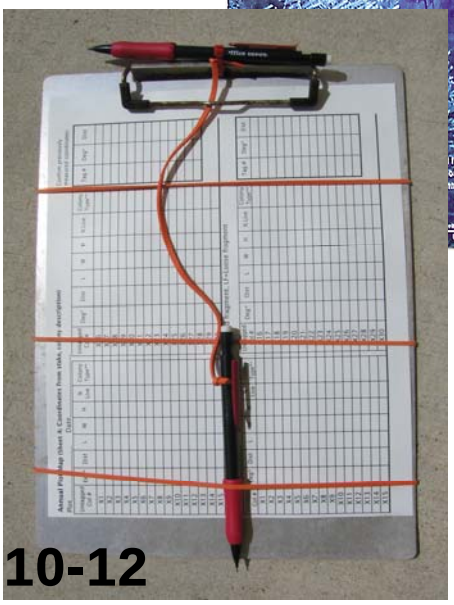
A-7

Loose Fragment Tally:









Key

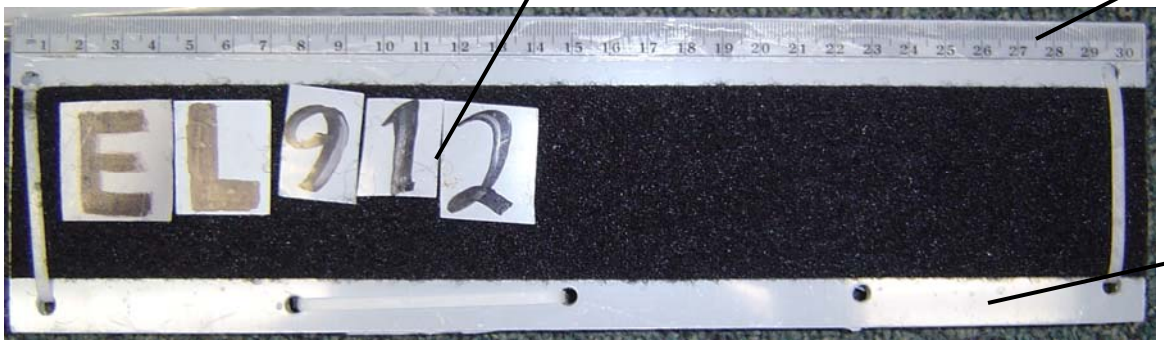
1. Metal stakes
2. Sledge hammer
3. Aluminum numbered tags
4. Cable ties
5. Nails
6. Transect line (10m length)
7. Dive compass
8. GPS with waterproof case
9. PVC meter stick (marked 5cm)
10. Pencils
11. Rubber bands/string
12. Aluminum clipboards
13. Dura Copy underwater paper

Photoboard: for scale and labeling in field photos

Front

Bold, individual letters & numbers on front indicate colony or sample number in photo

Metric ruler is readable in close up photos. The board itself provides 30cm scale in wide shots



3 1/4”

12”

PVC Board- Grey (not white) color reduces glare in photos

Back

Small distributed fishing weights eliminate buoyancy and provide balance & stability

Letters & numbers not being used are stored on the back of the board

Materials:

1) Metric Ruler (plastic)
2) PVC sheet (0.3mm thick)
3) PVC board (2.5mm thick)
4) Adhesive Velcro (Heavy Duty)
   (two 12″x 2″ strips)
5) Fishing Weights (6 x 1.5oz)
6) Super Glue
7) Box cutter
8) Scissors
9) Cable ties
10) Sharpie (Wide/Chisel Tip)
11) Drill (not shown)

Instructions:

A. Cut 2.5mm thick PVC to 12″ x 3 ¼″ using box cutter (score deeply then break)
B. Glue metric ruler flush with the top edge of the board.
C. Affix one strip of Velcro to board below the ruler
D. Affix second strip of Velcro to the back side of the board (centered)
   IMPORTANT: Both sides of the board must have the <u>same</u> side (hook OR loop) of velcro, the mate to both strips will be used later.
E. Drill holes 2 on each end and 2 along the bottom edge. Each pair should be ~2 ½″ apart.
F. Attach fishing weights to the board by lacing the cable ties through the holes.
G. Cut remaining two strips of Velcro in half lengthwise (1″ x 12″ strips)
H. Affix Velcro strip just below (<1/8″) the top margin of the 0.3mm thick PVC sheet, cut the sheet just below the Velcro. Repeat this with the remaining 3 strips of Velcro.
I. Use the Sharpie to write the appropriate letters and numbers and cut to the strips into individual letters.
J. Stick the letters not being used on the Velcro on the back side of the board.