PRELIMINARY POPULATION GENETIC STRUCTURE OF THE ENDANGERED DUSKY GROUPER, 
EPINEPHELUS MARGINATUS, IN MALTA AS REVEALED BY SIX MICROSATELLITE MARKERS.

Molly Buchholz 1* and Adriana Vella 1
1 Conservation Biology Research Group, University of Malta, Msida, MSD 2800, Malta - molly.sorensen@alumni.ubc.ca

Abstract
The objective of this study is to describe the population structure of the endangered marine fish, Epinephelus marginatus (Teleostei: Serranidae), in the Maltese archipelago for the purpose of localized conservation management planning. Preliminary genetic population structure of the Maltese dusky groupers was determined by six microsatellite markers. Initial results based on FST values comparing three spatial groups (Group A, North Malta; Group B, South Malta; and Group C, Hurd Bank) indicate group A and B are one continuous population inhabiting the Maltese archipelago (P(a-b) > 0.05) whereas group C, an area 23 km east of the main island, is identified as a sub-population to these groups (P(a-c) = 0; P(b-c) = 0). Results suggest that management should be designed to reflect the archipelago as a single population.

Keywords: Biogeography, Conservation, Genetics, Islands, Teleostei

Introduction
The dusky grouper, Epinephelus marginatus (Teleostei: Serranidae), is an endangered marine fish considered to be facing a very high risk of extinction in the wild. Due to concerns of population decline throughout their geographic range, the IUCN has currently listed them as endangered (EN) A2d [1]. Their native range includes the Mediterranean Sea, the eastern Atlantic Ocean along the west and south coasts of Africa around the cape to Mozambique as well as Brazil. Characterized by high site fidelity and protracted development to sexual maturity, this reef-associated protogynous hermaphrodite can usually be found in depths up to 50 meters off rocky coastal shores [2]. Localized population structure parameters include pelagic larval dispersal influenced by surface current direction during spawning season along with limited juvenile stage movement. The purpose of this research is to describe and define the status of the Maltese population of dusky groupers in order to best develop a comprehensive conservation management and monitoring strategy.

Materials and Methods
A total of 30 dusky grouper specimens with known catch locations were sampled from the Maltese archipelago between 2007-2009. Six fluorescently tagged microsatellite markers were used in this study to explicate the population structure: three primers developed for the red hind grouper [4]; Epinephelus guttatus (RH_CA_001, RH_CA_002 and RH_CA_008) and three developed for the gag grouper [5]; Mycteroperca microlepis (GA010, GA038 and GA045). Individuals were genotyped using an Applied Biosystems™ 3130xl capillary electrophoresis system. Tests of population differentiation were calculated using the software Arlequin [6].

Results
Three sample groups were identified based on log-likelihood assignment tests of individual genotypes to populations (Fig. 1). Within group analysis reveals observed allelic heterozygosity was greatest in group C (H_{a(a)} = 0.79, H_{b(b)} = 0.67 and H_{c(c)} = 0.81) while group A revealed the most unique haplotypes (n_{a(a)} = 31, n_{b(b)} = 12 and n_{c(c)} = 3). Rare breeding size males (T_{a(a)} > 85 cm) were present in all groups with a significantly higher incidence of large size individuals (T_{b(b)} ≥ 70 cm) observed in the west Gozo area of group B. Tests of population differentiation revealed little genetic variation between groups A and group B (F_{ST(a-b)} = 0.01, P > 0.05) while group C shows moderate differentiation from all other groups tested (F_{ST(a-c)} = 0.10, P = 0.0; F_{ST(b-c)} = 0.11, P = 0).

Discussion
Results indicate moderate genetic partitioning in a limited spatial range based on biogeographical isolation due to depth range and larval retention patterns. Based on preliminary FST values, group A and group B are considered to be a single congruent population inhabiting the Maltese archipelago whereas group C is identified as a sub-population to these groups. Due to long life history traits, population decline does not appear to have deleteriously effected within population allelic variation to date. West Gozo has been identified as a possible spawning site and area of conservation interest due to the relatively high concentration of large size individuals. Group A as a whole is also important since it contains a significant number of unique haplotypes. Average south eastern surface currents originating from the northern tip of Gozo during spawning season as a mechanism for pelagic larval dispersal is a probable mode for genetic homogenization within the Maltese population. We theorize Gozitan individuals may be an important source population to the rest of the archipelago. Therefore, we recommend management and pilot monitoring efforts initially focus on the northern (group A) and west (group B) Gozitan meta-populations.

References

Fig. 1. Map of collection locations for Maltese E. marginatus with 100 m bathymetry lines (NTS). Group A, North Malta (n = 13); Group B, South Malta (n = 11); and Group C, Hurd Bank (n = 6). Arrows indicate predominant surface current direction mean [3] for pelagic larval dispersal during the spawning months between June and August.