Preliminary analyses of infection of western and southern sardine by a "tetracotyle" type digenean parasite from samples collected during the November pelagic biomass surveys and May pelagic recruit surveys, 2010-2014

C.D. van der Lingen^{1,2} and K. Mushanganyisi¹

¹ Resources Research, DAFF; ² MaRe, Department of Biological Sciences, UCT

Introduction

Data on infection of sardine by a digenean "tetracotyle" type parasite has supported the hypothesis of multiple sardine stocks off South Africa, and panel reports from both the 2013 and 2014 International Stock Assessment Workshops recommended that data on the presence of parasites by length should be included in sardine assessments (Smith *et al.*, 2013; Dunn *et al.*, 2014).

GLM analyses of infection data from a total of 4 327 sardine processed from commercial catch samples collected over the period 2011-2014 from both western and southern stocks showed that *Stock* was the most important contributor to explaining variance in prevalence of infection, followed by *Year* and then *Length* (van der Lingen *et al.*, 2015a). Samples of sardine have been collected for parasite analysis from pelagic recruit and spawner biomass surveys since 2010, and results from preliminary analyses of these data up to 2012 were presented by van der Lingen *et al.* (2015b), who reported strong gradients around the SA coast in mean parasite abundance for three size classes of fish. Samples collected during 2013 and 2014 have been processed and parasite data collected, and preliminary results are presented here.

Methods and materials

Totals of 2 851 sardine collected during pelagic biomass surveys and 1 937 sardine collected during pelagic recruit surveys conducted during the past five years (2010-2014) have been processed (Tables 1 and 2; Figure 1). Coverage has been unequal between surveys, years and between stocks, with more fish sampled from pelagic biomass surveys, and from the

western stock, particularly during the recruit surveys where coverage of south stock fish has been poorer. The catch location, caudal length (cm) and number of parasites found in each sardine's eyes are recorded, and two indices of infection per sample (i.e. fish from the same trawl) are calculated, namely infection prevalence (% of the sample infected), and mean parasite abundance (number of parasites.fish⁻¹).



Figure 1: Sample locations of sardine collected for "tetracotyle" type metacercariae parasite analysis during pelagic biomass (left) and recruit (right) surveys, 2010-2014.

Table 1: Number of fish (and number of samples) from Pelagic Biomass survey samplesexamined for "tetracotyle" type metacercarian parasites by putative stock and in total,2010-2014.

Year	Western	Southern	Total
2010	373 (16)	370 (22)	743 (38)
2011	542 (18)	431 (17)	973 (35)
2012	297 (8)	67 (1)	364 (9)
2013	175 (3)	103 (1)	278 (4)
2014	279 (12)	214 (9)	493 (21)
Total	1 666 (57)	1 185 (50)	2 851 (107)

Table 2: Number of fish (and number of samples) from Pelagic Recruit survey samplesexamined for "tetracotyle" type metacercarian parasites by putative stock and in total,2010-2014.

Year	Western	Southern	Total
2010	100 (2)	140 (4)	240 (6)
2011	571 (18)	0	571 (18)
2012	75(3)	55 (2)	130 (5)
2013	448 (5)	204 (3)	652 (8)
2014	225 (8)	119 (5)	344 (13)
Total	1 419 (36)	518 (14)	1 937 (50)

Spatial patterns in prevalence and mean parasite abundance are visualized by plotting these indices per sample for each year (and also as a composite of all years). Sardines are assigned to a 0.5cm caudal length class, and prevalence-by-length and mean abundance-by-length for the western (collected to the west of Cape Agulhas) and southern stock (collected to the east of Cape Agulhas) are derived for each year. Annual, stock-specific prevalence-at-length data from the biomass surveys are presently being incorporated in sardine assessment models in order to estimate movement between the two stocks (de Moor and Butterworth 2015).

Results: BIOMASS SURVEYS

Spatial patterns in the prevalence of infection of sardine during annual pelagic biomass surveys 2010 to 2014 are shown in Figure 2. In general, western sardine show moderate to high prevalence values and southern sardine show lower prevalence of infection, although moderate prevalence levels were observed in most southern samples in 2011 and high prevalence levels in some southern samples in 2014. The plot of the combined data shows that whereas most western samples have moderate to high prevalence levels, southern samples range from zero to high prevalence.

A similar pattern to that seen in prevalence of infection is observed in the spatial distribution of mean parasite abundance (Figure 3); this is not surprising since mean parasite abundance is correlated with prevalence. The highest mean parasite abundances were observed in western fish in 2013 and in western and southern fish in 2014.

Length frequency distributions, prevalence-at-length and mean parasite abundance-atlength plots for western and southern fish for each year are shown in Figures 4 to 6. Between-stock differences in prevalence- and mean parasite abundance-at-length were apparent in most years, although the degree of difference varied between years; 2010 and 2014 appeared to show the strongest difference whereas differences were smaller for 2011-2013. Composite length frequency distributions, prevalence-at-length and mean parasite abundance-at-length plots for western and southern fish derived by combining the data from all five years into a single dataset are shown in Figure 6. Sardine become infected at a size of 10.5cm CL off both coasts, and prevalence-at-length shows a steady increase from around 15% at 11 cm CL to around 60% at 21 cm CL for western fish. Prevalence of infection in small (<12.5 cm) southern sardine is similar to levels shown by western fish of the same size (although sample sizes for small southern fish are themselves small), but prevalence then declines and remains at around 10% or less until about 17 cm CL, after which it increases rapidly and is higher than for western fish for sizes of 19.5 cm and above (although this may be an artefact of the number of fish examined as sample sizes for these larger fish are again small).



Figure 2: Sample locations (crosses) and prevalence of infection (% circles; size proportional to prevalence) of sardine by year sampled during annual pelagic spawner biomass surveys 2010-2014; and a composite plot for the combined data (bottom right; note symbols are differently scaled for this last plot).



Figure 3: Sample locations (crosses) and average parasite abundance (parasites.fish⁻¹; squares; size proportional to abundance) of sardine sampled during pelagic spawner biomass surveys 2010-2014; and a composite plot for the combined data (bottom right; note symbols are differently scaled for this last plot).



Figure 4: Length frequency distributions (n; upper), prevalence of infection by caudal length class (centre) and mean parasite abundance by caudal length class (lower; standard error bars are shown) for western (west of 20°E) and southern (east of °E) sardine sampled during the 2010 and 2011 Pelagic Biomass Surveys.



Figure 5: Length frequency distributions (n; upper), prevalence of infection by caudal length class (centre) and mean parasite abundance by caudal length class (lower; standard error bars are shown) for western (west of 20°E) and southern (east of °E) sardine sampled during the 2012 and 2013 Pelagic Biomass Surveys.



Figure 6: Length frequency distributions (n; upper), prevalence of infection by caudal length class (centre) and mean parasite abundance by caudal length class (lower; standard error bars are shown) for western (west of 20°E) and southern (east of °E) sardine sampled during the 2014, and for the composite 2010 to 2014 Pelagic Biomass Surveys (note that standard error bars for the composite abundance plot are derived using the numbers of fish samples per length class).

The composite plot of mean parasite abundance-at-length shows low values for both western and southern fish up to around 15cm CL, above which it increases steadily for sardine from both stocks up to around 20 cm CL (Figure 6). Mean parasite abundance is higher for western compared to southern fish from 15 to 20 cm CL, and the standard errors do not overlap for most observations within this size range, although this is difficult to see in Figure 6 (see zoom in Figure 7). Above 20 cm CL, the standard errors are substantially larger and mean parasite abundance does not appear to be different for fish of this size from the two stocks.

Parasite abundance data are highly skewed, with the vast majority (75% for western and 84% for southern) of fish being uninfected and the numbers of infected fish decreasing with

increasing parasite number (Figure 7). Because of this skewedness the data should be transformed, and a log transformation (log[# parasites + 1]) has been applied. Such a transformation will reduce the impact of individual fish with high parasite loads (i.e. >10 parasites) on the calculation of mean abundance. Abundance-at-length for untransformed and transformed data are shown in Figure 7, for both the full data range (means) and a zoom to focus on fish between 10 and 20 cm CL.



Figure 7: Frequency distribution of the number of parasites per fish (top plot); mean untransformed parasite abundance by caudal length class (middle plots; standard error bars are shown); and mean transformed (log[# parasites + 1]) parasite abundance by caudal length class (lower plots; standard error bars are shown) for western and southern sardine for the composite 2010 to 2014 Pelagic Biomass Surveys (note that standard error bars for the composite abundance plot are derived using the numbers of fish samples per length class, and that the right hand plots in the middle and lower panels are zooms of the left hand plots).

Results: RECRUIT SURVEYS

Year-specific analyses of spatial variability in prevalence and mean parasite abundance, and of prevalence-at-length and mean abundance-at-length for sardine collected during recruit surveys have not yet been conducted, but composite length frequency distributions, prevalence-at-length and mean parasite abundance-at-length plots for western and southern fish derived by combining the data from all five years into a single dataset are shown in Figure 8.



Figure 8: Length frequency distributions (n; upper), prevalence of infection by caudal length class (centre) and mean parasite abundance by caudal length class (lower; standard error bars are shown) for western (west of 20°E) and southern (east of °E) sardine for the composite 2010 to 2014 Pelagic Recruit Surveys (note that standard error bars for the composite abundance plot are derived using the numbers of fish samples per length class).

Very few small (<10 cm CL) sardine were collected from the southern stock during these surveys but both stocks have reasonable coverage overall for fish of 10-20 cm CL. Recruit samples show that western sardine first become infected at around 8 cm CL, and that prevalence increases from around 20% at 10 cm CL to around 50% for fish of 14 to 18 cm CL, before declining sharply in larger fish. Southern fish show a lower prevalence of infection than western fish up to around 18 cm CL, after which the values are similar and declining. Mean parasite abundance is higher in western than southern fish over the size range 13-16 cm CL, but larger southern fish appear to have higher mean parasite abundances.

Discussion

Data on prevalence of infection and parasite abundance for sardine collected during research surveys over the period 2010-2014 are either available (biomass surveys) or will soon be available (recruit surveys) for incorporation in stock assessment models, and may prove useful in estimating movement of sardine between the two stocks. Although statistical analyses have yet to be performed on the parasite data from fish collected during DAFF research surveys, the pattern of a higher prevalence of infection and higher parasite abundances in western fish compared to southern fish seen in commercial samples appears to hold for both the biomass and recruit survey samples. Pelagic biomass surveys are conducted in spring, and results in Weston *et al.* (2015) indicate that spring and summer are when differences in prevalence of infection and parasite abundance between the western and southern stocks are at their least, whereas these differences are at their highest in autumn and winter (when the recruit survey is conducted). That inter-stock differences in prevalence and mean parasite abundance appear to be larger for fish of 13-17 cm CL than for fish >17 cm suggests that movement from the west to the south also occurs in these larger fish.

References

- de Moor CL, Butterworth DS. 2015. Some initial results using the revised two mixing-stock hypothesis for South African sardine. *FISHERIES/2015/NOV/SWG-PEL/43*, 17pp
- Dunn, A., Link, J.S., Punt, A.E., Stefansson, G., and Waples, R.S. 2014. International Review
 Panel Report for the 2014 International Fisheries Stock Assessment Workshop, 1-5
 December 2014, UCT. MARAM/IWS/DEC14/General/4, 19pp
- Smith, A.D.M., Cox, S., Parma, A. and A.E. Punt. 2013. International Review Panel Report for the 2013 International Fisheries Stock Assessment Workshop, 2-6 December 2013, UCT. MARAM IWS/DEC13/General/4, 22pp.

- van der Lingen CD, Reed CC, Hendricks M, Nunkoo I, Winker H. 2015. Update on the processing for "tetracotyle" type metacercariae of sardine from commercial catch samples, 2011-2014, and preliminary analyses thereof. *FISHERIES/2015/FEB/SWG-PEL/02, 7pp.*
- van der Lingen CD, Weston LF, Ssempa NN, Reed CC. 2015. Incorporating parasite data in population structure studies of South African sardine *Sardinops sagax*. *Parasitology* 142: 156-167.
- Weston LF, Reed CC, Hendricks M, Winker H, van der Lingen CD. 2015. Stock discrimination of South African sardine (*Sardinops sagax*) using a digenean parasite biological tag. *Fisheries Research* 164: 120-129.