

Supplementary information

Manuscript: The relationship between oxidant levels and gut physiology in a litter-feeding termite

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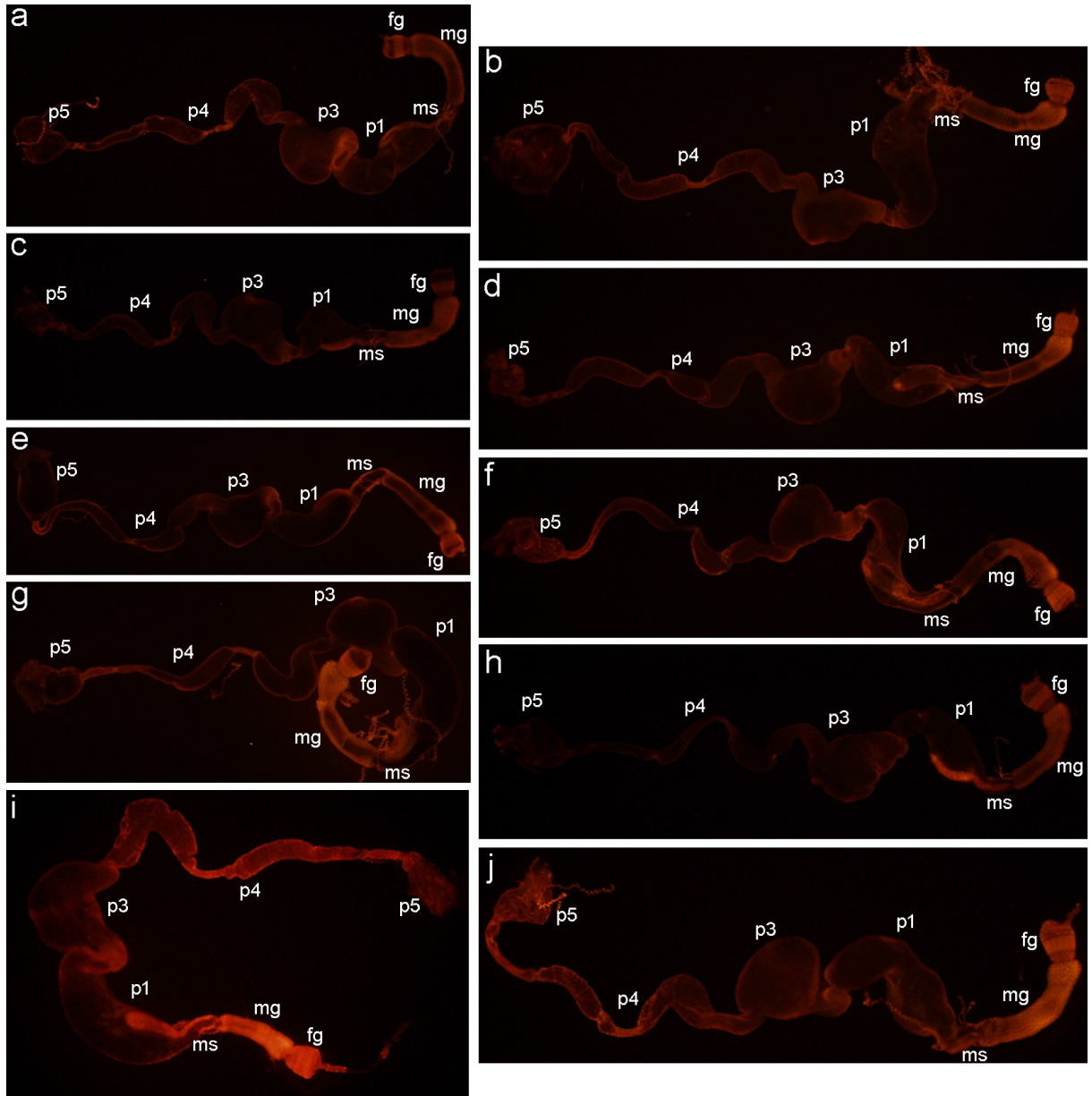


Fig. S1. Additional images to straight the profile of oxidant levels along *C. cumulans* alimentary canal. The guts were incubated with DHE and analyzed under a fluorescence stereomicroscope. Foregut (fg), midgut (mg), mixed segment (ms) and hindgut compartments – p1, p3, p4 and p5.

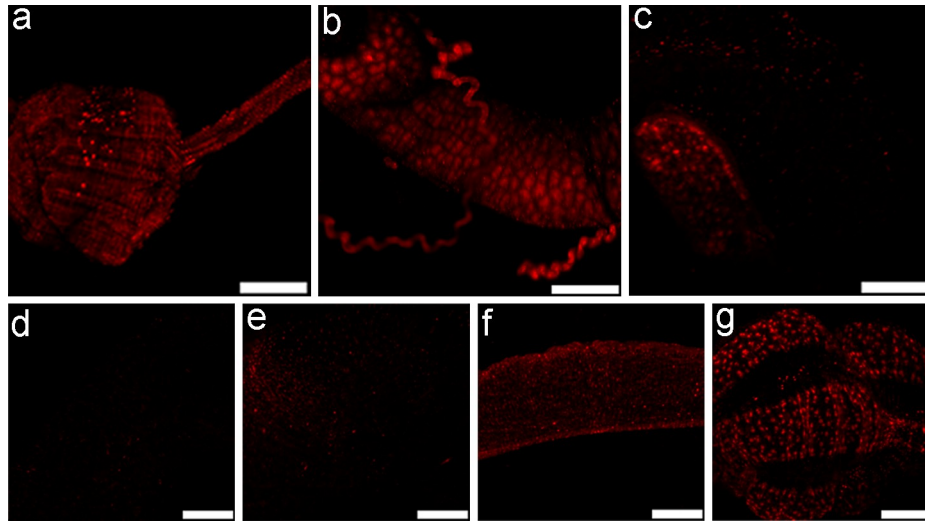


Fig. S2. Further details of oxidant levels along *C. cumulans* alimentary canal. Guts labelled with DHE and analyzed under a confocal microscope employing the “z-stack” function and overlap of 350 μm. **a** Foregut, **b** midgut, **c** mixed segment and hindgut **d** p1, **e** p3, **f** p4 and **g** p5 compartments. Bars = 100 μm.

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ZnSOD      1  -----MPVKAVCVLNGE
CcSOD      1  RRVAITWGGTRSCWSSLEVQAGLVV*FTG*FTK*SLFQGTERKRPLIMPLKAVCVLKGE
CgSOD      1  -----MPIKAVCVLNGE
CfSOD      1  -----MPIKAVCVLNGE

ZnSOD      13  VVKGTLFFDQVNPDSVPRVTGEVTGLSQGLHGFHIEHFGDNTNGCIISAGAHFNPHQKDHG
CcSOD      58  VVKGTVFFDQDDPESEVVKVTGEVTGLSKGLHGFHIEHFGDNTNGCTSAGAHFNPYQKDHG
CgSOD      13  VVKGTLFFDQENPDSAVKVTGEVTGLSKGLHGFHIEHFGDNTNGCTSAGAHFNPYQKDHG
CfSOD      13  VVKGTLFFEQENPDSAVKVTGEVTGLSKGLHGFHIEHFGDNTNGCTSAGAHFNPYQKDHG

ZnSOD      73  SPTEIRHVGDLGNI EAGSNGVAKVDITDKLISLSGAQNIIGRTL VVHADPDDL GKGGE
CcSOD     118  GPDDQVRHVGDLGNVEADSNVAKVDIADKLISLTGAHNIIGRTVVVHADPDDL GKGGE
CgSOD      73  GPDDEV RHVGDLGNI EADSSGVAKVDIADKLISLTGAHNIIGRTL VVHADPDDL GKGGE
CfSOD      73  GPDDEV RHVGDLGNI EADSSGVAKVDIADKVISLTGAHNIIGRTL VVHADPDDL GKGGE

ZnSOD     133  LSKTTGNAGARSACGVVGI AKI-----
CcSOD     178  LSKTTGNAGARSACGVVGI AKI*CMLHVKVFLVVC*QRPGLACSWKFK*QCRLPSIK*V
CgSOD     133  LSKTTGNAGARSACGVVGI AKI-----
CfSOD     133  LSKTTGNAGARSACGVVGI AKI-----

ZnSOD      -----
CcSOD     234  DSISAS*GTVS*CMI*IFIPYLCNKESHII*EKKKX
CgSOD      -----
CfSOD      -----

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Fig. S3. Multiple amino acid sequence alignment of *C. cumulans* SOD with sequences from other insects. The protein was denominated Cc (*C. cumulans*) SOD and compared to *Zootermopsis nevadensis* (ZnSOD – KDR12362), *Coptotermes gestroi* (CgSOD – ALM23457) and *Coptotermes formosanus* (CfSOD – AGM32998). Identical amino acids (black), amino acids from the same classification group according to the side chain (grey) and codon coding for a termination amino acid (*). The highest score blast X matches to *C. cumulans* SOD was 8e-98 for SOD1 (*C. gestroi*). Sequence ID: ALM23457.1.

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RpGPX 1 -----MRYYLLIVSL
TdGPX 1 -----
CcGPX 1 RVPITAGGRREV*GSVGRVHIDW*GCCRQVFVSI*TAIAEERHNFTSVKEFAYKVKSEGL
ZnGPX 1 -----MLVPGI

RpGPX 11 ILLATSIIILH-YQLTGFYSTEHTMAESEKDASSVYDFTVKDIAGNDISLEKYRGHVLIIV
TdGPX 1 -----RTMATSEKDATSVYDFTVKDISGNDVSLEKYRGHVLIIV
CcGPX 58 QLFCAAFCALRLLSTRAPVMAATPEAWKNASSIYEFTVKDINGQDVSLEKYRGHVAVIV
ZnGPX 7 QLFGTALCALRLLSTRTPVMASTPEAWKNASSIYDFTVKDINGQDVSLEKYKGHAAIIV

RpGPX 70 NVASQCGLTSTNYKELVELDEKYRETKGLRILAFPCNQFGSQEPGSREDIVCFMROKNAS
TdGPX 40 NVASQCGLTSTNYKELVELDEKYRDTKGLRILAFPCNQFGGQEPGSREDIVCFMQKNAT
CcGPX 118 NVASKCGEFTPTNYKELAEIHDQYAESKGLKILAFPCNQFNGQEPGTPEEIVCFAKSKNAK
ZnGPX 67 NVASKCGLTATNYKELAEIHDNYADSKGLRILAFPCNQFNSQEPGNSEEIVCFAKSKNAK

RpGPX 130 FEMFDKIEVNGSNNAHPLWKYLKSKQGGTLVDNLIKWNFTKFIIDKNGQPVERHGPMTNPSK
TdGPX 100 FEMFDKIEVNGNNAHPLWKYLKSKQGGTLVDNLIKWNFTKFIIDKNGQPVERHGPMTNPSK
CcGPX 178 FDMFEKIDVNGKNAHPLWKYLEYKQGGTLGDFIKWNFTKFIIDKNGQPVERHGPNVDPSK
ZnGPX 127 FDLFEKIDVNGNNSHPLWKYLKSKQGGTLGDFIKWNFTKFIIDKNGQPVERHGPNVDPK

RpGPX 190 ILSSELEKYW-----
TdGPX 160 LVSCLEKYW-----
CcGPX 238 LVSSFEKYW*CP*RYTX
ZnGPX 187 LVSNLEKYW-----

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Fig. S4. Multiple amino acid sequence alignment of *C. cumulans* GPX with sequences from other insects. The protein was denominated Cc (*C. cumulans*) GPX and compared to *Rhodnius prolixus* (RpGPX – JAA75589), *Triatoma dimidiata* (TdGPX – JAP03111) and *Zootermopsis nevadensis* (ZnGPX – KDR22003). Identical amino acids (black), amino acids from the same classification group according to the side chain (grey) and codon coding for a termination amino acid (*). The highest score blast X matches to *C. cumulans* GPX was 4e-124 for phospholipid hydroperoxide GPX isoform X1 (*Z. nevadensis*). Sequence ID: XP_021914630.1.

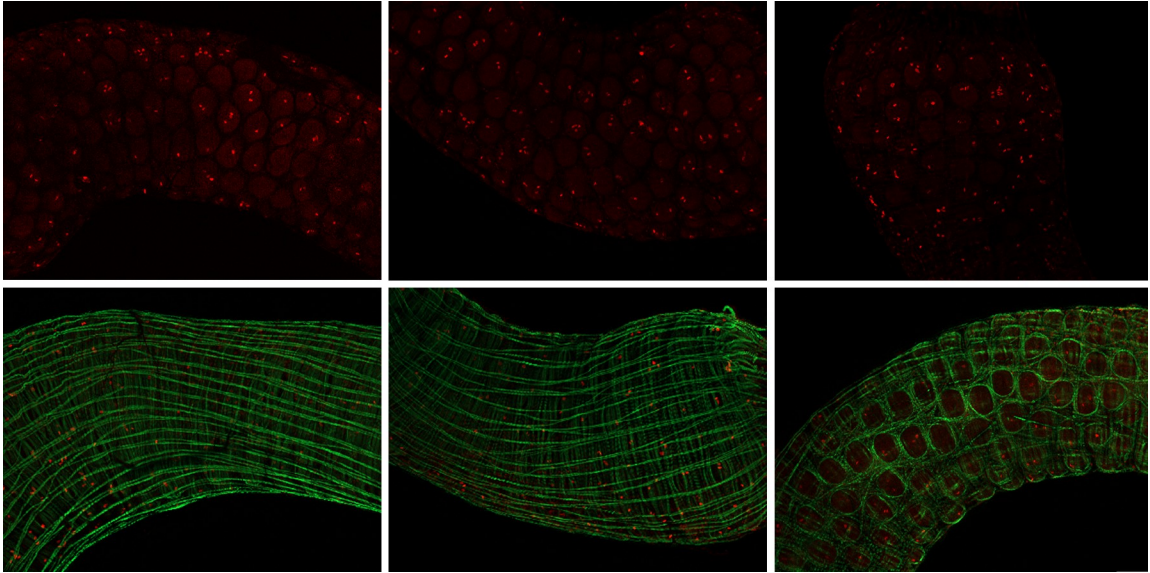


Fig. S5. Additional images to straight the profile of midgut cell renewal. The mitotic cells were revealed by anti-pHH3 antibody (red). The co-marking of Alexa Fluor 488 phalloidin (green) highlights muscle fibers. Analysis under a fluorescence stereomicroscope employing the “z-stack” function and overlap of 350 μm .

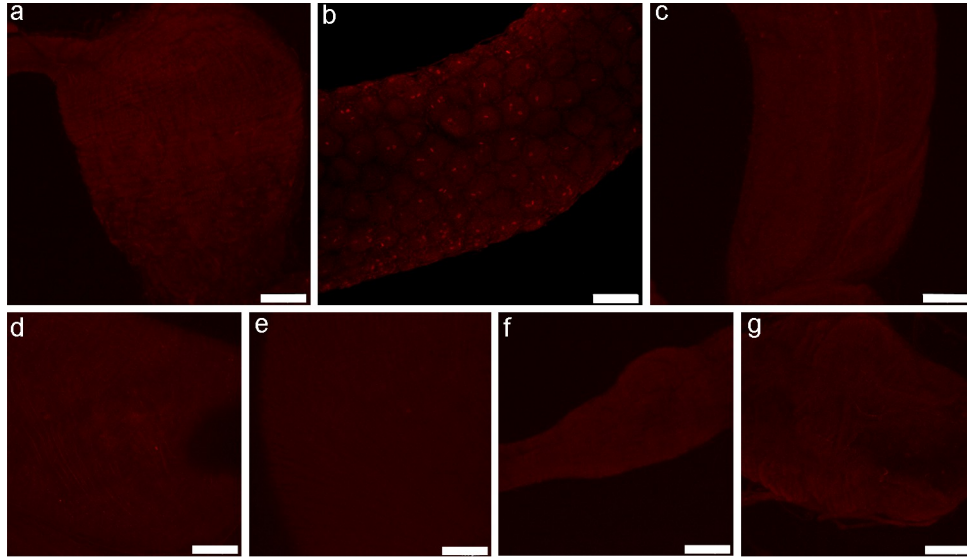


Fig. S6. Further details of mitotic profile of *C. cumulans* alimentary canal. Mitotic cells revealed by anti-pHH3 antibody (red) and analyzed under a confocal microscope employing the “z-stack” function and overlap of 350 µm. **a** Foregut, **b** midgut, **c** mixed segment and hindgut **d** p1, **e** p3, **f** p4 and **g** p5 compartments. Bars = 100 µm.