

## PREFACE

Since the dawn of immunology research and in spite of the dominant spotlight on mammalian and in particular human immunology, it may appear startling that invertebrates—and among others earthworms have continued to be an important model to solve several mysteries. In the sixties when transplantation experiments paved the path of understanding the recognition of self/non-self, it has also set the stone rolling for next decades to intensify research on earthworm immune mechanisms that evolved to prevent the invasions of pathogens. The skin constituted of epidermis and cuticle drained with muco-polysaccharides, casing the earthworm's body, is considered to be the first nonspecific antimicrobial barrier. The epidermis is outlined by a mono-layer epithelium of supporting cells, basal cells and secretor cells. The basal cells play a central role in wound healing and graft rejection, often wielding phagocytic activity. These phagocytes are coelomocytes which are freely flowing cells in the coelomic cavity. Recently, these coelomocytes have received particular attention in order to study immunity processes. The communication with the external environment is maintained by every segment of the coelomic cavity and so the skin fails to fully prevent the microbes from entering the coelomic cavity. As a result, the coelomic cavity is not aseptic and always contains microorganisms from the outer environment. Yet, there are well-organized mechanisms that keep the growth of microorganisms under control.

Moreover, earthworms play a key role in soil biology by providing ideal conditions for the growth of microorganisms. Biomass of soil, in many ecosystems, constituted of invertebrate components is largely dominated by earthworms. In the present study, the interrelationship between habitat (processed cow dung) and the earthworm (*Eisenia fetida*) has been studied in the laboratory mimicking the *in-situ* conditions. *In situ* environmental conditions versus those of the animal gut present distinct habitats and challenges for microbes due to differences in water activity regimens, pH, oxygen levels, etc. Cow-dung was used as a model system to study survivability of the randomly ingested microbes (bacteria, Achaea, fungi, and protozoa) during passage through the gut of the dung feeder earthworm *Eisenia fetida*. The cow-dung contains high titres of cultivable *Bacillus* spp. ( $> 10^{13}$  g<sup>-1</sup> processed

cow dung) throughout the period of processing until it gets suitable for feeding the earthworms. Microbes of aerated cow-dung, after being devoured by the earthworm, passes through an itinerant anoxygenic micro-zone through suffering assaults or being selectively favoured by the gut *in-situ* factors. Since several *Bacillus* species are predominant with titres  $> 10^{11} \text{ g}^{-1}$  gut content, it was hypothesized that the journey of *Bacillus* population from cow dung to earthworm gut and back to the environment has an important regulatory effect on the overall dynamics of *Bacillus* population. The thesis has elucidated in details the swings in population dynamics of the subset of ingested dung-microbes under unique micro-conditions of the earthworm gut. Knowing fully well the limitation of culture-dependent studies, whole metagenome sequence of *E. fetida* gut content was analysed to understand the spectra of microbial diversity prevailing within earthworm system. Attempting to answer another fundamental question-‘How do these worms manage to regulate the bacterial population under control?’ was also one of the other major objectives set in this thesis. Side-by-side, wound healing including regeneration (after amputation) with concurrent monitoring of innate immune status and indigenous micro-flora of the coelomic fluid have been studied. Finally differential gene expression in coelomocytes under bacterial challenge was analysed via genome-wide transcriptomics.

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