

Apoptosis inducing factor (AIF), a phylogenetically conserved mitochondrial inter-membrane space flavoprotein has an essential role in caspase independent cell death. Nonetheless, AIF is also crucial for cell survival. This apparent functional duality highlights the notion that the apparatus involved in the suicide of a cell is closely linked to the basic cellular life-sustaining processes. AIF is essential for mitochondrial organization and energy metabolism. Although it executes critical cellular functions in a coordinated manner, the exact mechanism remains elusive.

D. discoideum is an excellent lower eukaryotic developmental model as it can switch between unicellular to multicellular phases. The present study aims to understand the role of AIF in cell survival, growth and development by its downregulation in *D. discoideum*. Constitutive *AIF* downregulated (dR) cells exhibited a slower growth rate and delay in developmental morphogenesis. Also, *AIF* dR cells manifested high intracellular ROS, oxidative DNA and protein damage and calcium levels with lower ATP content. Under oxidative stress, *AIF* dR cells showed early mitochondrial membrane depolarization followed by AIF translocation from mitochondria to nucleus and exhibited necrotic cell death as compared to paraptotic cell death of control cells (Fig. 8.1).

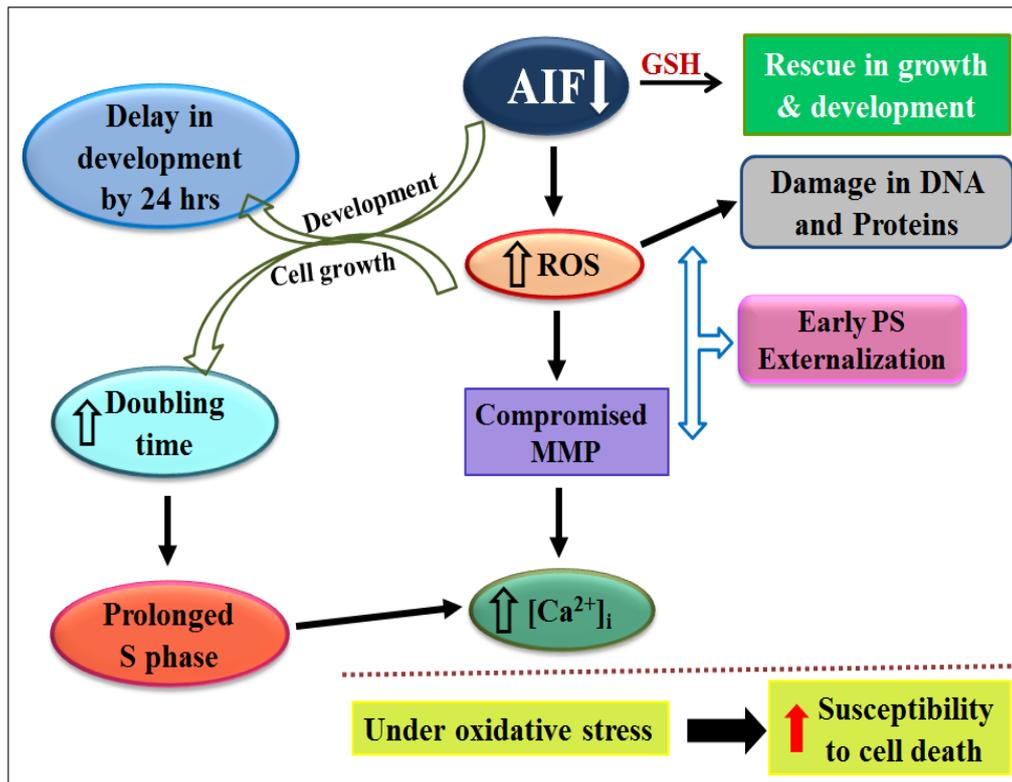


Fig. 8.1: Role of AIF in cell growth and development of *D. discoideum*: Reduced *AIF* levels result in a slower cell proliferation rate and delay in development. Under oxidative stress, reduced *AIF* increases the susceptibility to cell death.

The present study also underscores the ability of mitochondria-localized AIF to influence the redox metabolism, thereby maintaining respiratory capacity, assembly of Electron Transport Chain (ETC), mitochondrial DNA content, fusion-fission processes and morphology in *D. discoideum*. It acts as a survival protein possibly through its oxido-reductase property. Interestingly, constitutive *AIF* dR cells showed amelioration in cell growth upon the antioxidant treatment, providing a piece of evidence for the cellular protective function of AIF as an ROS regulator. Moreover, overexpression of *AIF* also affects mitochondrial homeostasis, altering mitochondrial fission-fusion profile, increasing ROS levels and reducing mtDNA pool (Fig. 8.2).

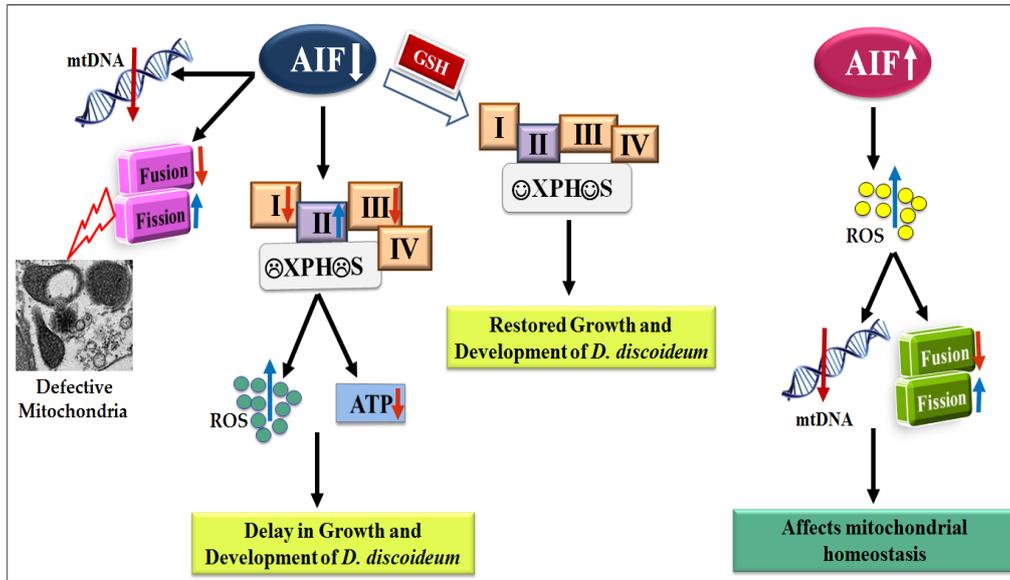


Fig. 8.2: Role of AIF in mitochondrial functions and morphology of *D. discoideum*: Downregulation of *AIF* compromises OXPHOS and thereby increased ROS levels and decreased ATP production ultimately affecting cellular growth and development which was restored by GSH treatment. *AIF* deficiency also affects mitochondrial fusion-fission events causing defective mitochondrial structure. Overexpression of *AIF* also affects mitochondrial homeostasis, altering mitochondrial fission-fusion profile, increasing ROS levels and reducing mtDNA pool.

Thus, the findings of the present study provide an exemplar where AIF is implicated in cellular growth and development by regulating ROS levels and maintaining mitochondrial integrity and functioning. One of the intriguing facets of the most common complex I mitochondriopathies is hypomorphic and/ deletion *AIF* mutation which causes >30% of mitochondrial deficiencies. The *AIF* deficient models could be instrumental for therapeutic approaches in complex I and *AIF* deficiencies thus making it possible to identify target metabolic and stress-response pathways. In addition to this, various underlying mechanisms of AIF's mitochondrial activities would contribute to other diseases such as obesity, diabetes, etc. Thus, it is useful to find out AIF inhibitors that may act as cyto-protective or cyto-toxic agents against AIF

related diseases or mitochondrial diseases. Moreover, it would indeed be worthwhile to identify mitochondrial interacting targets of AIF to clarify whether it is an assembly factor of ETC or takes part in other mitochondrial activities in the inter-membrane space. This finding encompasses our understanding of molecular mechanisms underlying AIF related mitochondrial respiratory chain and redox balance diseases that may contribute to the development of new therapeutic targets.

In conclusion, our findings enlighten the physiological role of AIF in mitochondrial homeostasis of both the vegetative and developmental stages of *D. discoideum*, controlling mitochondrial energy production by stabilizing respiratory chain complex assembly, maintaining mtDNA pool, mitochondrial structure and morphology via fusion-fission process, while its NADH activity functions as a redox channel.