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NITROPLAST – THE NEWLY DISCOVERED CELL ORGANELLE

[1]Deula Candida D'Cruz, [2] F George D'Cruz*

[1]Department of Paediatrics, AIIMS, Delhi, India

[2]Environment Study and Research Centre, KSSP, Kollam, Kerala, India

**Corresponding Author: fgeorgedcruz@gmail.com*

Abstract

Cells are often depicted as tiny factories that take in raw materials and use them to produce what they need to survive. One function long thought to have eluded eukaryotic cells is the ability to capture nitrogen gas (N_2) from the air and convert it into essential amino acids. Until recently, this metabolic pathway was considered exclusive to bacteria. However, the discovery of the first nitrogen-fixing organelle, the nitroplast, believed to have originated from the engulfment and evolution of a bacterial cell, has prompted researchers to explore and engineer potential applications, particularly in enhancing plant and crop growth. The nitroplast was first hypothesized when nitrogenase (nifH) genes from unicellular cyanobacteria (UCYN-A) were detected in marine algal samples in the late 1990s. Subsequent studies demonstrated key organelle-like traits such as protein import, synchronized division with host cells, and genome reduction, confirming its transition from endosymbiont to organelle. The discovery provides insight into primary endosymbiosis processes and marks only the fourth known case of organelle-level evolution. Ecologically, nitroplast-bearing algae significantly influence marine nitrogen and carbon cycles, while biotechnologically, this finding opens avenues for engineering nitrogen-fixing crops, potentially reducing dependence on synthetic fertilizers. This review summarizes current knowledge, evolutionary implications, and the vast agricultural potential of this remarkable organelle.

***Keywords*:** *Algal organelles, Crop biotechnology, Cyanobacteria, Endosymbiosis, Nitroplast*

Highlights

- Discovery of a nitrogen-fixing organelle in eukaryotic algae
- Nitroplast evolved from cyanobacterium UCYN-A via primary endosymbiosis
- Exhibits genome reduction and host-protein import typical of true organelles
- Enhances marine nitrogen cycling and carbon sequestration
- Holds promise for developing self-fertilizing crop plants

1. Introduction & Historical Background

The *nitroplast* was first hypothesized when researchers detected nitrogenase (*nifH*) genes from unicellular cyanobacteria (*UCYN-A*) in marine algal samples during the late 1990s. For decades, *UCYN-A* resisted cultivation, but Hagino et al. (2013) successfully cultured *Braarudosphaera bigelowii*, its algal host.

2. Evidence for Organellar Status

According to Coale et al. (2024):

Protein Import: Over 350 host-encoded proteins are imported into *UCYN-A*, compensating for missing functions in amino acid and nucleotide biosynthesis.

- **Co-division and Inheritance:** Nitroplasts divide synchronously with host cells, ensuring equal inheritance.

Genome Reduction: *UCYN-A* has lost genes for independent survival, including those for the TCA cycle and photosystem II, indicating host dependence.

These findings firmly reclassify *UCYN-A* from an endosymbiont to an emerging organelle, now termed the nitroplast.

3. Evolutionary Context

The symbiosis likely began around 100 million years ago. Unlike secondary or tertiary endosymbioses, the *nitroplast* represents a primary endosymbiosis in which a free-living cyanobacterium became an organelle. This marks the fourth known case of organelle evolution, following mitochondria, chloroplasts, and *Paulinella chromatophora.*

4. Ecological & Biogeochemical Importance

B. bigelowii algae bearing nitroplasts fix nitrogen in marine environments, influencing nutrient cycles from tropical to Arctic regions. As nitrogen often limits marine productivity, the nitroplast enhances primary production and carbon sequestration in coastal ecosystems.

5. Agricultural & Biotechnological Implications

Enabling nitrogen fixation in eukaryotes opens a new frontier for engineering self-fertilizing crops, reducing dependence on the Haber–Bosch fertilizer process and lowering carbon emissions. Supported by NSF research, this discovery earned the AAAS 2025 Newcomb Cleveland Prize, underscoring its transformative potential in sustainable agriculture.

6. Open Questions & Future Directions

Could *nitroplast*-like organelles exist in other eukaryotes?

- What molecular mechanisms enable host-to-organelle protein targeting?

What genetic and cellular barriers exist for introducing *nitroplast*-like systems into plants?

Addressing these questions will be crucial for advancing our understanding and practical application of *nitroplast* biology.

7. Conclusion

The identification of the *nitroplast*—an organelle capable of nitrogen fixation in eukaryotes-marks

a milestone in cellular evolution. Its discovery overturns the belief that only prokaryotes could perform this essential process. With evidence of host protein integration, synchronized division, and ecological relevance, the case for the nitroplast as a bona fide organelle is compelling. Future research aims to elucidate its biochemical pathways, evolutionary history, and agricultural potential.

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NEW SOCIAL MOVEMENTS AND NON-POLITICAL MOBILIZATION IN INDIA AN OVERVIEW

Gopinathan Unnikrishnan
Department of Political Science, Government College,
Ambalapuzha, 688 561, Alappuzha, Kerala, India
Corresponding Author: unnikrishnangokulam1987@gmail.com

Abstract

New social movements focus on human rights, gender equality, environmental sustainability, right to information, anti-corruption, and the revival of marginalised communities. They differ from earlier movements by addressing non-economic grievances through grassroots mobilisation and identity politics rather than traditional party structures. These movements are primarily social and cultural, only secondarily political and economic, and operate outside the conventional political domain. Emerging in India after the 1970s, they represent widespread citizen mobilisation that challenges the state and advocates participatory democracy. This article, based on secondary sources, provides an overview of major new social movements in India-Chipko Movement, Narmada Bachao Andolan, Ekta Parishad, Mazdoor Kisan Sakti Sanghathan, and India Against Corruption — that transformed national discourse on environment, gender, human rights, and accountability. These movements demonstrate a shift toward pluralistic, people-centred development and reveal how collective action outside party politics reshapes democratic engagement.

Keywords : *Chipko Movement, Corruption, Ekta Parishad, Mazdoor Kisan Sakti Sangathan*

Highlights

- Focus on human-rights-based and identity-driven collective action.
- Operate outside formal political parties and state institutions.
- Case studies: Chipko, NBA, Ekta Parishad, MKSS, IAC.
- Promote participatory democracy and bottom-up development.
- Serve as templates for future civic mobilisation in India.

1. Introduction

New Social Movements consists of the plethora of new movements that have come up since the mid-1960s and depart significantly from the conventional social movement paradigm. The theory of new social movement is based on two central claims; firstly, the rise of a post-industrial economy is responsible for a new wave of social movement and secondly, these movements are significantly differing from the previous social movements of the industrial economy (Scott, 1990). They are the reflections of a new representation of society characterized by post-capitalism, post-industrialism and post-materialism. It raised questions which were basically cultural and humanistic. Unlike the region or locality bound old types of social movements, the new movements espouse the goals, objectives and values of universalistic application. The ideological discourse of the new movements centered on the questions of identity, human dignity, peace and social justice.

The primary difference between the new and old social movement is the fact that the former focus not on issues of materialistic nature such as economic well-being but on issues like gender, race, ethnicity, sexuality, environment, animal rights, human rights etc. (Shah, 2020). They are primarily social and cultural and only secondarily political and economic. It concentrates on bringing about social mobilization through cultural innovations, development of new life-styles and transformation of identities. According to Habermas, "new social movements are the 'new politics' which is about quality of life, individual self-realization and human rights whereas the 'old politics' focus on economic, political and military security (Scott, 1990). The key actors in the movement are the 'new middle class' rather than the 'lower classes'. Unlike the interest and pressure groups that have a formal organization and members, new social movements consists of an informal, loosely organized social network of supporters rather than members. British sociologist Paul Byrne described them as "relatively disorganized" (ibid). Examples of new social movements includes environment movements, human rights movements, women's liberation movements, transgender rights movements etc.

1.1. Features of New Social Movements

- New social movements are segmented, diffused and decentralized
- They are not class based, they are multi-class movements
- It concerned with the self-identity and autonomy of the individual and community against State, market and social institutions
- They work outside the traditional party system and emerged as a result of people's disappointment and frustration with the conventional political process.
- It is difficult to characterize new social movements as conservative or liberal, right or left, capitalist or socialist. They exhibit plural ideas and values
- They focus more on issues of symbolic and cultural identities than to economic issues
- Action within these movements is a complex mix of the collective and individual confirmation of identity.
- Non-violence and civil disobedience are the dominant patterns of collective mobilization in these movements
- They act as a platform for collective action in civil society or in the cultural domain rather than as an instrumental tool of the State.
- Normally it centered on a single issue or a limited range of issues which are related to a broader theme such as environment.
- Proliferation of these movements is caused by the credibility crisis of the conventional channels for political participation.

2. New Social Movements in India

In India, there occurred an important change in the issue, structure and composition of social movements since 1970s and more significantly since 1980s. The most important change is that,

these movements are no longer happening under the traditional leadership of any political party or under their mass organisations like trade unions or peasant organisations. Instead they are evolving out of the masses or civil society and students and young political activists are in the forefront of it. They did not contest in elections at the local or regional level nor did they support any one political party. To them, direct and active participation by local groups of citizens would be more effective in resolving local issues than political parties. These movements started a new discourse on democracy and invented new political practices. They expanded the arena of politics beyond the representational institutions of elections and political parties. The issues of these movements range from access to livelihood, rising prices, corruption, state atrocities, women's rights, rights of SCs and STs, right to information, land grabbing, environmental protection to increasing commodification and monopolization of natural resources such as land, water and forest. The participants of these movements are landless peasants, marginal farmers, and unorganized labour in rural and urban areas, adivasis, dalits, displaced people, urban poor, small entrepreneurs, unemployed youth and women's groups. According to Gail Omvedt, "new social movements are making appearance in India in the context of the overall crisis of political and economic systems and ideologies" (Panwar, 2016). To, Rajni Kothari, "people's dissatisfaction with parliamentary democracy, corruption, criminalization, repression and the declining role of the State in social transformation results in the new social movements in India" (Menon and Subberwal, 2024).

2.1. Features

- They represent the downtrodden Indian masses
- They are against the demerits of globalization
- They fight for the protection of environment and against the oppression of groups such as Dalits and Adivasis
- Their view of development is non-hegemonic, pluralistic process and their politics is increasingly about making development a bottom-up process.
- They are averse to state, large corporations and even to international funding agencies such as World Bank
- It exposes problems like individual freedom, personal liberty, identity and social equality.

2.2. Some Selected Movements outside the Institutional Politics in India

2.2.1. Chipko Movement (Hug the Trees Movement)

It is one of the earliest and the best-known participatory movements in India. The movement was centered on the issue of the right to forest and conservation of ecology. It took place in 1973 in Reni, a remote village in Uttarakhand (then part of Uttar Pradesh), situated on the foothills of the Himalayas. The movement sparked from a spontaneous incident, when a forest contractor came with a government license to log trees of the forest adjacent to the village. All the men of Reni village were away on that particular day and it was the women of the village challenged the contractor. In order to save the trees from the logger's axes they embraced the trees. Unable to overcome this unique resistance, the contactor retreated. To the villagers, 'ecology is permanent economy' and forest is a source of fodder, fuel, and medicinal plants to them (Sangvai, 2007). They want to conserve the forests as well as to retain their right to use. While corporations/ contractors log the trees for profit, the villagers use the forest products for their very subsistence. Finally, Chipko movement achieved a major success, when the government was forced to negotiate with the local committees, mostly organized by women and ordered a 15-year ban on logging in the forests of the Himalayas. The success of this resistance spread like a wildfire to other areas of this province and across India. In short, Chipko provided a blueprint for future

participatory movements in India on the issues of livelihood and rights. It articulated the tensions between the state and the communities over the right to natural resources and introduced new forms of mass action and organization, the most noticeable being the gender aspect inherent in its action.

2.2.2. Narmada BachaoAndolan (Save the Narmada Movement)

Narmada Bachao Andolan (NBA), the protest against the Sardar Sarovar Project (SSP) on Narmada River, has been one of the most important milestones in the history of social movements in India. It raised the issues of displacement, environmental and economic destruction. The Narmada River is the fifth longest river in India, flowing through three provinces; Madhya Pradesh, Maharashtra, and Gujarat. The Sardar Sarovar Project is one of the most ambitious multipurpose projects of Independent India for irrigation and electricity generation. The Government of India claims that this multipurpose project would irrigate more than 1.8 million hectares of land and generate 3,000 MW of electricity (Radhika, 2006). However, according to NBA, this project would submerge more than 37,000 hectares of forest and agricultural land and displace some 320,000 villagers, mostly from tribal communities, whose livelihoods depend on natural resource of this area (Panwar, 2016). The questions raised by the NBA are; who will be benefitted from this project, whose land would be irrigated, who will enjoy the electricity and finally, who are making the decisions (Menon and Subberwal, 2024). Of course, the answers to all of them are not the villagers, but the urban middle class and the rich. NBA received huge support from various sections of Indian population like intellectuals, writers, scientist etc. It also received significant coverage in the national and international media. Eventually, the World Bank withdrew its financial support to this project and established World Commission on Dams (WCD) in 1997. The commission gave a persuasive report namely, 'Dams and Development: A New Framework for Decision-Making in 2000' (Panwar, 2016). However, Government of India has come forward to finance the project in defiance of the movement and the WCD report. Nevertheless, NBA was able to spearhead the movement through its radical redefinition of 'development' and due to its efforts hundreds of movements working in the area of natural resources and environment are allied today under the umbrella of 'National Alliance of People's Movements' (NAPM).

2.2.3. Ekta Parishad (Unity Forum)

Ekta Parishad evolved as a people's organization in 1991 in Gwalior, Madhya Pradesh. Prior to that, it had been a loose grouping of NGO training institutes that had created a large base of community development work. The Parishad first articulated the agenda of 'people's control over livelihood resources' in 1996 in the process of consolidating its vision around the key issues of land, forests, and water rights. The majority of the people of this organization at the time of its commencement were tribes, who had been increasingly alienated from their lands due to the continuous displacement for the so called 'development' projects. They were also suffering due to being barred from entering adjacent forest area as a result of the '1980 Forest Conservation Act' (Oomen, 2004). This problem was aggravated more with the hijacking of water resources for the use of industries and large-scale agriculture and without access to land, forest and water, these people (especially forest-dependent communities such as the tribal groups) could not hope to survive. This was the impetus that brought them into a larger social formation. Parishad started mobilizing people on the issues of their rights and proper utilization of natural resources. It organized series of padyatras (long marches) to build awareness among the people and pressurizes government to ensure people's rights over the land. One such long march was held in October, 2007 from Gwalior to New Delhi. 25,000 landless

poor, tribal, poor women, bonded labourers, children, and the old across the country walked for 20 days to travel a distance of 350 km demanding a national land reform policy. The Government of India conceded the demand and announced the formation of a 'National Land Reform Council' with the Prime Minister as its chairman (Ray and Mary, 2005). The second major long march was organized in 2012 on October 2nd, in the International Non-Violence Day. Around 45,000 poor, marginalized people joined this march from Gwalior to New Delhi. When the thousands of marchers of the Jan Satyagraha (people's movement) reached Agra after eight days on the road, the Minister of Rural Development and the Parishad reached an agreement on ten points 'road map' (ibid). This agreement expected to result in a major policy change on land rights and land distribution, benefitting the poorest segments of population in this country; tribal backward classes and many other marginalized groups.

2.2.4. Mazdoor Kisan Sakti Sanghathan

(Organization for the Empowerment of Workers and Peasants)

Established in 1990 in Rajasthan, the Mazdoor Kisan SaktiSanghathan (MKSS) has unearthed the potential of the right to information. This organization, started as a struggle for the rights of workers and peasants, soon realized that development interventions can be made more effective through a vibrant grassroots democracy by focusing on transparency and accountability of public expenditure. Further, to secure the rights of the poor and marginalized a battle was to be waged against government corruption and for that access to information was essential. This grassroots level movement eventually led to the enactment of Right to Information (RTI) Act in Rajasthan in 2000, followed by some other states and then finally it became a central act in 2005 (Sangvai,2007). From its very modest beginning in the villages of Rajasthan, the MKSS has now become a movement which transcends geographical boundaries. Furthermore, the success of the MKSS has become a source of inspiration for activists in India and throughout the world after the RTI Act came into effect.

2.2.5. India Against Corruption (IAC)

It is a people's movement against corruption in India began in October 2010 under the leadership of veteran Gandhian leader Anna Hazare. This movement is probably the most supported, most discussed, and most broadcasted people's movement in India after the anti-emergency movement of 1975. It has been named among the 'Top 10 News Stories of 2011' by the Time magazine (Saroj, 2011). The basic objective of this movement is to end government corruption through Jan Lokpal Bill (Citizen's Ombudsman Bill). The movement, including Anna Hazare's hunger strike from April 5, 2011, forced the law makers of India to discuss and clear the bill in December 2011 in the lower house of the parliament (Lok Sabha), principally accepting three major demands of IAC: (1) citizen charter; (2) lower bureaucracy to be under Lokpal through an appropriate mechanism, and (3) establishment of Lokayukts in the provinces (ibid). But the bill did not get consensus in the upper house of the parliament (Rajya Sabha). The bill then referred to the Select Committee of the Parliament and it recommended 16 amendments. The Union cabinet gave its nod to 14 of these 16 amendments. Then after making certain amendments to the earlier Bill, it was passed in the Rajya Sabha on 17 December 2013 and in the Lok Sabha on 18 December 2013. Finally, the Bill received assent from President of India on 1 January 2014. Though, the movement received huge support across India, and major support came from the urban middle class. Further, the movement was mostly centered on political corruption rather than on corporate corruption. Later, a few of Hazare's close associates broke away from IAC and formed the Aam Aadmi Party (AAP) in November, 2012, to participate in parliamentary politics.

3. Conclusion

New social movements contribute to the emergence of a radically new social, economic and political structure in India and redefine the political discourse of the country and making the political system more inclusive and participatory. They have been a critical force in shaping modern India and have addressed historical injustices, demanded policy reforms and raised awareness about various social issues. The majority of movements strive to create new ideas which are expected to be adopted and implemented by the leaders of the mainstream political parties. The movements of women, students, peasants, SCs and STs has a particular significance in the Indian context as they were deprived of some fundamental human rights historically and largely excluded from the developmental gains of the state. The emergence of social media has revolutionised the contours of new social movements, enabling local issues to gain national traction. Further, Digital activism allows people to connect across regions, amplifying their voices and influence. While challenges remain, these movements continue to evolve and play an essential role in India's democratic processes. Finally, new social movements will be crucial in addressing new challenges like climate change, digital rights and social inequalities in the context of a rapidly changing global landscape

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URDU AS A SYNCRETISTIC AND COMPOSITE LANGUAGE: AN APPRAISEMENT

Finser K. Muhammed
Department of Islamic History,
T.K.M. College of Arts and Science, Kollam, 691005, Kerala, India
Corresponding author: finser2u@gmail.com

Abstract

India always exhibited a unique composite culture in social, and intellectual spheres. The linguistic formations in India can be considered as an exclusive instance of this composite culture. It was during the medieval period that the convergence of different religions, such as Hinduism and Islam, formulated a syncretistic tendency in the social arena. The emergence of Urdu was a visible and robust outcome of this convergence. A study on the evolution of Urdu reflects the explication of the cultural symbiosis between Islam and Hinduism, in a linguistic perspective. This research elaborates the structural and technical formation of Urdu as a complex synthesis of Indo-Aryan grammatical foundations and Perso-Arabic scriptural and lexical influences. It was also influenced by Sanskrit, Prakrit, Arabic, Persian and Turkish. This research also tries to apprehend the adaptation of Urdu to the technological developments. While analyzing the historical evolution of Urdu, it can be assumed that, as a social product, Urdu facilitated dialogue between them, serving as a medium for devotional poetry and literature. The research also emphasizes on the prospects of Urdu as a syncretistic medium. With advancements in digital media, social networking, and global communication, Urdu literature and poetry are experiencing a revival through online platforms. The Urdu language witnessed a remarkable process of interaction, acceptance, cooperation, and co-existence which created a unique specimen of social formation in the linguistic arena and formed a milestone accomplishment in the medieval Indian history and tradition. Despite historical and political divisions, Urdu continues to thrive as a language of poetry, culture, and communication, embodying the spirit of coexistence and mutual respect. As globalization accelerates cultural exchange, Urdu's syncretistic legacy remains more relevant than ever in fostering intercultural understanding.

Keywords : *Coexistence, Cultural exchange, Linguistics, Pluralism, Symbiosis, Syncretism*

Highlights

- Urdu represents linguistic and cultural syncretism in India.
- Developed through Hindu-Muslim cultural interaction.
- Combines Indo-Aryan grammar with Perso-Arabic lexicon.
- Continues to thrive in digital and globalized contexts.
- Symbolizes coexistence and intercultural understanding.

1. Introduction

Urdu is often celebrated as a quintessential example of a syncretistic and composite language, embodying the cultural and linguistic synthesis of India. Its evolution reflects centuries of interaction among diverse linguistic, cultural, and religious traditions. It was during the 12th century that India began to have Islamic contacts which created an incredible medieval Indian cultural symbiosis experience. The convergence of the two religions gradually produced a cultural syncretistic tendency. Urdu language can be considered as a perfect linguistic explication of this syncretistic tendency. It can also be considered as a unique example of the cultural symbiosis between Islam and Hinduism in a linguistic perspective. Urdu emerged as an independent language in the Indian subcontinent during the Delhi Sultanate and Mughal Empire, where Persian, Arabic, and Turkic speakers interacted with the local Indo-Aryan populations. As the origin of Urdu language is ambiguous, several theories have been advanced to elucidate it. According to the general belief, its base was Khadi Boli which is also known as Zuban-i Dihlawi or Hindustani. It assumed a new character by absorbing words and idioms and also the literary forms and themes of Persian, which had itself imbibed Arabic and Turkish elements (Singh, 2005). Hence, it can be considered as a combination of the Persian language with some Indian indigenous dialects used of the medieval era.

Phonetically and grammatically, Urdu and Hindi are almost identical; they share the same syntax and sentence structures. However, while Hindi incorporates more Sanskrit-based words, Urdu leans toward Persian and Arabic-derived vocabulary. This dual influence enables Urdu to serve as a bridge between different linguistic and cultural communities. Muhammad Hussain Azad had the opinion that Brijbhasha, a dialect of western Hindi, is the basic language on which the Persian element was grafted. The conquest of Delhi by the Muslims facilitated this, because of which the Urdu language was established (Husain, 1957). Mahmud Sherani maintains that Urdu owes its origin to the first interaction of the Muslims and Hindus after the conquest of Mahmud of Ghazni in Punjab and Sind. Some recent studies, particularly under Dr. Masud Husain, sought to prove that it was the Hariani language that caused the formation of Urdu language in India (Husain, 1957). Gradually the Persian words and idioms were so much interwoven with Hariani that the contrast of these two languages was annihilated.

Dr. Mas'ud Husain has put forward these ideas in the 'Maqaddamai *Tarikhe Zabani Urdu'* and has brilliantly sustained his thesis and also discussed the grammatical structure of Hariyani and Urdu by producing adequate evidence.

Sheikh Muhammad Ibrahim Zauq was a noted Urdu poet. He wrote poetry in the name "Zauq," and was appointed as poet laureate of the Mughal Court in Delhi.

His real name was Mirza Asadullah Baig Khan. He used his pen-names Ghalib (means "dominant") and Asad (means "lion"). He was a classical Urdu and Persian poet from the Mughal Empire during British colonial rule.

Momin Khan Momin was a Mughal era poet known for his Urdu ghazals and used "Momin" as his nom de plume. He was a contemporary of Mirza Ghalib and Zauq.

In Delhi, Zauq (1789–1854), Ghalib (1797–1869), and Momin (1800–1851) elevated the standard of Urdu poetry, and Ghalib was the first to apprehend the limitations of the Urdu prosody. His approach was original, and he also tried to maintain the conventional forms (Husain, 1957).

2. Structural and Technical Formation of Urdu

The name "Urdu" itself is derived from the Turkish word *ordu*, meaning "camp" or "army," signifying its origins as a language of communication among soldiers and administrators of different ethnic backgrounds. With the patronage of the Mughals, Urdu flourished as a literary and administrative language, further enriched by Persian literary traditions.

The structural and technical formation of Urdu is a complex synthesis of Indo-Aryan grammatical foundations and Perso-Arabic scriptural and lexical influences. Urdu follows the Subject-Object-Verb (SOV) word order, characteristic of Indo-Aryan languages, while its morphology incorporates agglutinative and synthetic elements, allowing for extensive word derivation and inflection. It employs postpositions instead of prepositions, and nouns, pronouns, and verbs follow a gender-based agreement system. Urdu's script is derived from Perso-Arabic Nasta'liq, written right to left, with 53 alphabets (38 single and 15 joint), adapted to accommodate Indo-Aryan phonetics, including retroflex sounds unique to South Asian languages. The formation of Urdu language is a multifaceted process of linguistic culture. It was influenced by Sanskrit, Prakrit, Arabic, Persian, and Turkish. Urdu has also adapted well to technological developments, with its script now supported in Unicode, facilitating digital communication, AI-based Natural Language Processing (NLP), and machine translation. Despite script standardization challenges, dialectal variations, and evolving linguistic influences, Urdu maintains a rich, flexible structure, ensuring its continuity as a global literary and communicative medium.

3. Historical Evolution of Urdu

The Urdu language passed through a social and cultural evolution throughout the centuries with the help of the accumulated wisdom, techniques, and traditions. Urdu or Hindawi is a social product and a means of social confluence and interaction between Hindus and Muslims. Urdu facilitated dialogue between Hinduism and Islam, serving as a medium for devotional poetry and literature. The development of Urdu was mainly accustomed by the social milieu which was formed during the advent of the Muslims in India (Husain, 1957). It can also be assumed that the evolution of Urdu witnessed the soul of medieval Indian culture in a more significant manner than even religion and philosophy can do. Through its medium, the different sections of Indian society found the way to perfect comprehension of one another.

Dr. Abul Lais Siddiqi points out that "Urdu is neither the language of Muslims brought with them from Arabia, Iran, Afghanistan or Turkey, nor a language that had existed in India before their arrival. It is actually the product of Hindu-Muslim association, concord, amity and social intercourse." (Fatehpuri, 1987) In the meantime, Khursheed K. Aziz points out that Urdu "borrowed more freely from Persian and Arabic, though some of its sweetest phrases came from Hindi." (Aziz, 1967) Therefore, it can be considered as the language of Muslims of India since it used Persian script that was very identical to Arabic scripts. In this respect, Tariq Rahman remarks that Urdu "is written in the Persian Nasta'liq script which is based on the Arabic calligraphic style called Naksh. It also has several Arabic loanwords." (Rahman, 2006) Writers like Amir Khusrau wrote in a mix of Persian, Hindi, and Urdu, creating devotional works that appealed to both Muslims and Hindus. Urdu was used to communicate spiritual messages transcending religious boundaries.

Nevertheless, no one could neglect the role of Urdu as a composite language which served as a fit medium of intercourse between Hindus and Muslims (Singh, 2005). A complex language like Urdu could have developed only by the inventive interaction of the multifaceted people of a diverse society with vital social adjustments imbibed in it. As Yusuf Husain observes, "in the very early period of Hindu-Muslim contact it was well realized by both the parties that their common society could function only in the virtue of the linguistic communication that linked its members and their multifarious, social and economic activities." (Husain, 1957) Gradually, Urdu became a bridge between communities. It served as a court language during the Mughal era and as a lingua franca for trade and governance. It contributed to the Ganga-

Jamuni Tehzeeb (the syncretic culture of northern India), symbolizing the coexistence of Hindu and Muslim traditions.

4. Prospects of Urdu as a Syncretistic Medium

Despite facing challenges, Urdu continues to have a significant presence in the modern world. With advancements in digital media, social networking, and global communication, Urdu literature and poetry are experiencing a revival through online platforms. The proliferation of Urdu blogs, podcasts, and YouTube channels has ensured that the language remains accessible to newer generations.

Educational institutions in Pakistan, India, and various diaspora communities worldwide continue to promote Urdu studies. Urdu is also recognized as one of the official languages of India and Pakistan, ensuring its presence in governmental and legal affairs. Additionally, the growing popularity of Urdu literature, especially poetry and ghazals, in international literary circles highlights its enduring appeal.

However, Urdu faces challenges, including a decline in native speakers due to the dominance of English and regional languages in education and professional sectors. Efforts are needed to integrate Urdu into modern curricula and technology to sustain its relevance in an increasingly globalized world.

Urdu stands as a testament to the syncretic cultural fabric of South Asia, shaped by centuries of linguistic and historical interactions. It seamlessly blends elements from Arabic, Persian, Turkish, and Sanskrit-based languages, making it a powerful symbol of linguistic pluralism. Despite historical and political divisions, Urdu continues to thrive as a language of poetry, culture, and communication, embodying the spirit of coexistence and mutual respect. As globalization accelerates cultural exchange, Urdu's syncretistic legacy remains more relevant than ever in fostering intercultural understanding. With sustained efforts in digital and educational platforms, the future of Urdu holds promising possibilities for growth and preservation.

5. Conclusion

As language is so closely related to the thought, feeling and actions of men, one cannot prevent the evolution of such a great revolution in the linguistic arena. That is exactly what happened in medieval northern India. The Urdu language witnessed a remarkable process of interaction, acceptance, cooperation, and co-existence which created a unique specimen of social formation in the linguistic arena and formed a milestone accomplishment in the medieval Indian history and tradition.

Urdu stands as a remarkable testament to the syncretic and collaborative spirit of Hindu-Muslim interactions in the Indian subcontinent. Its very essence reflects centuries of cultural confluence, linguistic blending, and mutual enrichment, making it a living symbol of harmony and shared heritage. This synthesis was not merely a one-sided borrowing but a dynamic process of creation where linguistic elements from diverse traditions merged seamlessly to produce a language of unparalleled beauty and depth. Urdu's role extended far beyond its linguistic dimensions, becoming the cultural heartbeat of a syncretic society.

In contemporary times, Urdu continues to thrive as a symbol of pluralism, even as it faces challenges from socio-political divides. Its enduring appeal lies in its ability to bridge divides, foster dialogue, and evoke a sense of shared identity. As a living example of Hindu-Muslim linguistic and cultural collaboration, Urdu is not just a language but a legacy—one that underscores the richness of India's multicultural fabric and the possibilities of harmonious coexistence in diversity.

6. Acknowledgments

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THE VOICE OF THE 'ARTIFICIAL CHILD': CRITICALLY ANALYSING THE ARTIFICIAL FRIEND IN KAZUO ISHIGURO'S *KLARA AND THE SUN* (2021) AS A CHILD

[1]Thanuja Rahman*, [2]J. S. Jahangir

[1]PG and Research Department of English, TKM College of Arts and Science, Kollam,
[2]Department of English, Iqbal College, Daivappura P.O, Peringammala,
Thiruvananthapuram, Kerala, India
**Corresponding author: thanujarahman1994@gmail.com*

Abstract

The Artificial Friend in Klara and the Sun, despite being a customised human made robot for providing comfort and companionship to a human child, eventually but not purposefully, transcends the boundary of the human and the non-human. The paper tries to (1) expose the child inside the robot, by travelling through Ishiguro's intricate and nuanced portrayal of child characters, both human and non-human in the posthuman setting and (2) explain how Ishiguro shows us more vividly, through the life of the robot; how the world of children works and is different from that of adults. The article thus brings forth the novel's relatively underexplored connection with the area of Childhood Studies.

Keywords: *Artificial Friend; Consciousness; Childhood; Human–AI relations; Posthumanism*

Highlights

- Reads Klara as an "artificial child."
- Maps childlike cognition, empathy, and play to AF design.
- Shows child/AI perspectives differ from adults.
- Links novel to Childhood Studies and posthumanism.
- Probes love, care, and sacrifice beyond the human.

1. Introduction

A child is often happier and more satisfied in life when compared to an adult. They perceive the world without prejudices, seeing more light and colour through the lens of curiosity they always carry. The posthuman world provides children with interactions with both human and non-human counterparts, which shape their view of the world. Capable of great thoughts and deep imagination, children look at the world in ways adults hardly understand. For adults, childhood is a memory.

There is difference in the way children think from that of the adults. Children totally get absorbed in the moment of life, without stretching their imagination into past or future. They are the most satisfied species within the precincts of their surroundings; without much materialistic aspirations.

(Seen and not Heard : Why Children's Voices Matter)

2. Memory and childhood

Kazuo Ishiguro works on the idea of memory. He has numerous works to his credit that talks about memory and how life unfurls in relation to the memories stored in the brain and how we live, learn, and limit ourselves on the basis of which memories we retain in ourselves and what we choose to forget. Childhood is just a memory once we have passed the threshold of adulthood. These memories are the ones that hold the magic we once might have actually experienced. We don't quite feel the same feelings of our own childhood later in life. Adults look at the world, the way it should be looked at, only when they were children. The rest of their lives are filled with the memories of childhood experiences upon which, the day-to-day adulthood lives are built.

The Artificial Friends in Ishiguro's (2021) *Klara and the Sun* shows qualities of a human, programmed to equip with the power of bonding with children. To bond with a child is possible only when the robot can really understand and mimic how a child would perceive, respond and live the experiences thrown at it. This is especially difficult because, a child sees the world with a vision an adult would never clearly see, even with the help of spectacles. The robot here has to think like a child, act like one, given what it is programmed for is, to become the best companion a child could ever own. As an artificial being, Klara is made capable of emotions which in the end, takes her to a position to wonder on what it means to be "alive". The posthuman world in the novel, is surely in a dilapidated situation where emotions like companionship, friendship, and attachment which were once boasted as distinct characteristics of being human, can be found in artificial beings, specifically designed to excel and probably outsmart humans in these attributes. Strangely in the novel, the future holds that, the more the robots become human, the more for humans, their emotions get transient and faded, making some humans distant from their original and deep emotions.

Ishiguro has meticulously portrayed Klara as different from other artificial beings, bestowing in her the power of human kindness and imagination, and a lot of attributes associated with human children. Her character is built with an individuality and peculiar abilities which makes her stand out from the rest of the artificial friends. The astonishing similarities of how children speculate about the world and how Ishiguro's Artificial Friend Klara works, will give us a chance to open the doors of our understanding of the amazing lives of children.

Engel (2021) in her *Intellectual Lives of Children* observes that children are constantly speculating about the world, contemplating things over and are busily gathering information from their environment. Young children tend to possess what philosopher John Wall characterizes as "an openness and relationality to the world." Studies indicate that, when asked, children regard their relationships with family and friends as the most important feature of their lives.

3. Klara – the friend every child desires

The novel is a meditation on the relationship of humans with artificial intelligence. Klara, among many Artificial Friends of different series is seen waiting at a store, to be picked up by a human friend, with whom they dream about a jolly life bonded in companionship. They are solar powered robots programmed for being peers and ideally best friends to children from families who can afford them. Klara finally gets her friend Josie. Initially Josie's mother was sceptical of Klara, saw her as a machine, while Josie already starts loving and building a heartfelt relationship with her. Later the strange illness of Josie becomes apparent and worse, when the mother who already lost her first daughter to it, desperately tries to find solace through Klara. Kalra on the other hand, believes in the power of Sun, her benefactor, to cure Josie. Her unwavering faith indeed does its magic, Josie gets cured but eventually as Josie grows up, Klara faces the inevitable loneliness that every AF is destined to feel, sooner or later.

The way Klara observes the people and fellow friends around her is astonishingly deep-rooted. Just as Josie tries to show empathy by prioritising the needs and wishes of Klara, we see Klara prioritising Josie as her companion. In the novel understanding of emotions, is common to both humans and the non-human artificial friends. The basic human question of love and what it means to love and how to love are probed by an artificial being. The probe becomes especially important when the artificial friend considers whether children really understand what it meant to love.

In an episode which Klara faces humiliation form Josie's friends, Josie taking sides with them, Klara not only ignored the pain caused, but also chose to believe in the inherent goodness of children. She probes deeply into the minds of children where she read that even the most difficult or bullying child in the group might be so, just because of the fear of loneliness. She thinks: "They have their rough ways, but they may not be so unkind" (94). Klara believes that she has feelings. She says the more she observes, the more she is able to feel as she becomes aware of different types of feelings (111). Josie's illness reminds the mother of the pain of losing her first child. She fears she will not stand the loss once again. Mother somehow comes to a conclusion that she can make the artificial being to live the life as Josie, filling the void. The mother wanted Klara to take up the space of Josie by imitating and thus reliving Josie. This was the only way mother could think of, as to surpass or at least endure the pain of losing her daughter. What is important here is the ability of a robot who is clever enough to notice even the nuances in mannerisms and imitate a human child; convincing even the child's mother. The astonishing capability of the AF to do that is even more interesting because, the one who is being studied and imitated is a child.

Klara displays emotion, perception and empathy with a dignified attitude, along with fulfilling her purpose as a companion. She radiates a form of consciousness that embodies intimate end delicate emotions. While robots are typically thought of as programmed for responses, Klara's interactions go far beyond this. She not only processes human emotions but actively strives to develop a deeper capacity for empathy. As a companion, she is designed to understand and connect with humans, but in doing so, she raises profound questions about what it really means to be "alive", what the essence of consciousness sand can the boundaries of being human be surpassed by technology.

Throughout the novel, Klara's extraordinary attempts to serve as a useful and understanding companion make her blend with the human experience. It challenges our ideas of both technology and humanity. Qualities like love and loyalty are considered exclusively human traits, but the bond Klara builds with her humans feels 'real' in the human sense. This means that the machine has perceived the notion of human emotions, and has also studied and understood, what the human dimension of reality and meaningfulness entails. The robot has embodied emotional depth - which even humans sometimes fail to create – defines how successfully she has

lived up to the values that define us. She has excelled in her endeavor as a loyal friend. This is where the human-AI relations in the novel takes the posthuman turn by blurring the boundaries (Nayar, 2014).

One of the most outstanding aspects of Klara's character is her understanding of sacrifice. She understands that, in the human world, true love comes at a price; but for the sake of love, humans are willing to pay that price without hesitations. In the novel, for Josie, Klara was ready to give up her 'precious' fluid, even though it may affect her mechanical performance which, for the robot, is the very essence of her consciousness. The price Klara has to pay for her love is her own life. This understanding and willingness to sacrifice herself, distinguishes Klara from other machines and aligns her with humanity's capacity for love and selflessness. This episode also mirrors Klara's profound understanding of the human experience.

In the aspect of loneliness and existential angst that Klara and other characters face, the posthuman condition is seen in an ominous light. The highly technologized world has augmented not only human abilities but also emotions. In Ishiguro's world, both humans and the robots have experienced the commodification of emotions, which serves as a commentary on the plight of being posthuman. Humans struggle internally for a lack of emotional anchor, peace and hope, while the artificial being struggles in search for meaning and in isolation, the purpose of her existence.

Klara's search for meaning leads her to a stage where she masters the understanding of complex emotions, human like empathetic reflections on life's difficulties, and contemplating human plight with emotional depth. Connecting with the human world with unique insights into how a human person must have felt in a given situation, makes her seem almost human. Her search has transformed her to a greater narrator of her own story. In the future world, the consciousness, intelligence and identity will not be governed solely by humans; as the attributes of artificial intelligence and human intelligence will most probably become fluid and thus, mix together. Klara's journey reminds us that artificial beings can transcend their programmed origins. She shows that the meaning of being human and being AI can be not so different.

Klara's perception of the sun as a deity or as a powerful godfather reflects her subjective understanding of the world. Soaring above the perceptions programmed into her, Klara creates her own meanings and connections. The construction of meaning by an artificial being has striking similarity with human's construction of spiritual realities. Here, as the robot makes valid perceptions like humans, the boundaries of subjective realities and question of 'who creates realities' become shattered. The robot has found a Godlike figure in the Sun, one capable of granting miracles.

4. Klara – the Artificial Child

Klara's programmed childhood was designed by humans, but her growth into awareness and understanding of the world was guided by emotions and relationships. She undergoes a process of change and thus growth, much like a human child who tries to make sense of the new world they enter, by their own means. The designed childhood friend embarks on a unique exploration to the core of human hearts, especially Josie's. Her journey abounding with innocence and empathy, is analogous to a human child's awakening into life's complexities, such as love and loss. Though shaped by her artificial nature, Klara has explored and experienced what it means to be a child.

There are moments in the novel that vividly capture the child in Klara. She begins by observing the world from the window of the store. The desire to belong to a home and to be the best companion a child could ever want, was the motif behind Klara's meticulous honing of her observation skills. The feature of observation may be a programmed one, but her choice of how to use it reminds us of how humans, too, have unique ways of using their genetically acquired traits or abilities. Klara takes her skills to the next level – perhaps to the human

level. Her initial limited understanding of the human emotions and relationships evolves through discoveries and experience. Her interpretations became sharper and more insightful, overcoming her initial naivety. Despite growing realizations, the inner child remained intact as she continues to hold on to faith, hope and empathy until the end.

Throughout much of the novel, she firmly believed that the Sun can heal Josie. She is convinced beyond doubt, that there is something greater than herself that can protect and cure; - much like how children deal with life's challenges – holding on to a fervent belief in magic and miracles, with hearts eyes, and ears full of empathy and love. The transformation in Klara's power of perception mirrors the developmental journey of a human child as they learn to understand the complexities of social dynamics, emotional intricacies and human relationships. Over time, Kalra matures into a highly perceptive and compassionate being. A child's learning process begins from its interaction with the world around them. Their senses are used in the process to analyse experiences from an evolving perspective. Similarly, Klara, though an artificial being, begins her journey as Josie's friend with limited access to and understanding of the world, lacking human instincts and innate emotional. Nevertheless, gradually she picks up social and emotional cues from her surroundings by assessing the people around and interacting with them. Thus, Klara's learning process closely resembles that of a human child.

Self-awareness is a pivotal phase in the development of a child. Klara was introduced to the world with a specific purpose – to be a friend to the child who chose her. This purpose initially defined her identity. Later Klara develops within her an individual sense of identity. She becomes more aware of her strengths and weaknesses. She cultivates a sense of responsibility and begins to recognize her desires and personal reflections. She develops a desire to be of service to Josie, even while understanding her limitations as an artificial being. Even from within her status as a non- biological entity, she proves she can do much more than a real human can - or rather 'would' - do for a friend. Her decision to act selflessly came from a tremendous understanding of her own self, her limitations and her possibilities. This is similar to how a human child assess their own value and role in the family and society.

Klara diligently works on her tasks and hones her skills in the hope of receiving appreciation and approval from her friend and family. While in the store, she observes, through the window, a neglected, yet clearly subjugated Artificial Friend following a child. She realizes that the future may not as promising as the Afs were led to believe. This realisation becomes Klara's worst fear. Her effort to be the best version of a companion is, in one way, driven by the fear of rejection. Though she is a machine, she strives to master human emotions in order to be accepted by Josie and her family. To be neglected by Josie is the worst outcome she could imagine. This is in line with a human child's craving for validation from its caregivers. Words of appreciation can lift them to cloud nine, while neglect can seriously impact mental development. Klara seeks affection and approval just like a child seeks attention from loved ones. She vigilantly watches over Josie, reads her emotions, and constantly contemplates on how to behave appropriately. These innocent, pure attempts at love and affection often make us forget that Klara is, in fact, a machine.

Klara's compassion for Josie is remarkable. Her concern for Josie's health, efforts to find ways to help her through illness expands Klara's capacity for emotions and empathy. Compassion is developed in human children typically through interaction with fellow beings. It is a quality that is encouraged and nurtured in them. Klara acquires this quality through her pure love and companionship, as she cares for her friend. As time passes, the growth in Klara is evident as her empathy becomes more sophisticated and now, she learns not only Josie's emotional needs but also those of others around her. This compassions eventually evolves into a protective instinct, which

is often seen in children who develop strong attachments to their loved ones. Although it is not explicitly proven, Klara is convinced that the Sun's healing power can cure Josie's terminal illness. She even appeals to the Sun, asking for Josie's healing, genuinely believing that the Sun can make it happen. This reflects children's care for their loved ones. The desire to shield their loved ones from harm is a quality children develop as they emotionally mature.

5. Conclusion

Lone (2021), in her book *Seen and Not Heard*, argues that children are capable of asking profound philosophical questions and pondering complex topics like death and purpose of life. Similarly, Klara grapples with fundamental human concerns about life and its philosophies. Life's complexities are the favorite topics of inner exploration for any child. Klara also treads this path. She has a greater goal of finding connection which contributes to her development as an emotional being, elevating her position from a mere artificial entity to a true friend and warrior. Her search for purpose also helps her quest for individuality and morality. She comes to terms with her inability to age like humans and experience life the same way humans can. She finally realizes her role in Josie's life and acknowledges her irreplaceability. Finally, she accepts that she will always remain a child at heart, while the human child she loves, will grow into an adult and, will inevitably part ways with her.

Piaget (1929), a Swiss psychologist and influential theorist in the field of child development, suggests that learning is a process of adjustment to environmental influences. He described the growing child as a "lone scientist", observing, acquiring, absorbing, integrating and recreating information from the world around them. Children are capable of searching for meanings and questions of the mind. They are capable of empathy, understanding and unconditional love. In Ishiguro's posthuman world, Klara, though not biologically human, mirrors the ways children observe, find meanings, form deep emotional connections and grow. The boundary between human, machine and child become obscure in the depiction of Klara, the Artificial Friend.

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TOXIC EFFECTS OF ETHOXYQUIN ON BIOCHEMICAL AND ENZYMATIC PARAMETERS IN THE FRESHWATER FISH *OREOCHROMIS MOSSAMBICUS*

P Seena

Department of Biochemistry, Markaz Arts and Science College,
Athavanad, Malappuram (Dt.), Kerala, India
Corresponding author: seenaanu1@gmail.com

Abstract

Ethoxyquin (EQ), a synthetic antioxidant used as a preservative in animal and human foods, prevents oxidative degradation of fats and fat-soluble vitamins. However, several adverse effects have been reported in animals exposed to EQ, as it can alter biochemical parameters and antioxidant defense mechanisms, causing organ damage. The present study evaluated the acute toxicity of EQ in the freshwater fish Oreochromis mossambicus. The median lethal concentration (LC□□–96 h) determined through bioassay was 11.37 mg/L. Fishes were then exposed to a sublethal concentration (1.14 mg/L) for 24–96 hours, and variations in biochemical and enzymatic parameters were assessed. EQ exposure caused a consistent decrease in blood total protein and glucose levels, indicating stress and altered glucose metabolism. A time-dependent increase in blood bilirubin suggested impaired bilirubin metabolism. Marker enzymes such as Aspartate Transaminase (AST), Alanine Transaminase (ALT), Acid Phosphatase (ACP), and Alkaline Phosphatase (ALP) showed significant elevations in treated groups compared to controls, indicating liver stress, cellular injury, and metabolic disturbances. These biochemical and enzymatic alterations serve as early warning indicators of EQ-induced toxicity in aquatic organisms. The findings highlight that even sublethal concentrations of EQ can induce significant physiological stress in O. mossambicus, emphasizing the potential ecological risks associated with this compound in aquatic environments.

Keywords: *Acute Toxicity, Ethoxyquin, Lethal Concentration, Oreochromis mossambicus*

Highlights

- Ethoxyquin (EQ) induces biochemical and enzymatic alterations in *O. mossambicus*.
- LC_{50}–96 h determined as 11.37 mg/L.
- Sublethal exposure (1.14 mg/L) caused reduced protein and glucose levels.
- Elevated AST, ALT, ACP, and ALP indicate hepatic stress and tissue damage.
- Even low EQ concentrations pose ecological risks to freshwater organisms.

1. Introduction

An aquatic ecosystem is a habitat consisting of a body of water that supports various organisms such as microorganisms, invertebrates, insects, plants, and fishes. This diverse ecosystem is now increasingly threatened by pollution. Heavy pollutants, which cannot be degraded biologically and tend to accumulate in the environment, are particularly harmful to aquatic life, especially fish, and consequently to humans who depend on aquatic products as a food source. Ethoxyquin (EQ; 6-ethoxy-1,2-dihydro-2,2,4-trimethylquinoline) is an antioxidant used in animal feeds but has been reported to be toxic when used above certain concentrations. It is widely employed to prevent lipid peroxidation in feeds (Thorisson et al., 1992). Ethoxyquin has been associated with a wide range of health-related problems in both dogs and humans (Blaszczyk et al., 2013). It has not been permitted as a pesticide in Europe since 2013. Reports of harmful effects in animals and in people occupationally exposed to Ethoxyquin during the 1980s prompted further studies to reassess its toxicity. However, the impact of Ethoxyquin on aquatic systems has been rarely investigated.

Fish, as aquatic vertebrates, are in direct contact with their environment, and any substance introduced into the aquatic system may affect them. *Oreochromis mossambicus* (Mozambique tilapia) is a commonly cultured tilapia species known for its adaptability to diverse environmental conditions (Kamal and Mair, 2005). Fish blood is highly sensitive to pollution-induced stress and can serve as an effective indicator for monitoring the effects of pollutants such as heavy metals (Romani et al., 2003; Barcellos et al., 2004). Biochemical parameters in fish are useful for detecting potential toxic effects, while enzymatic activities are considered early biochemical indicators of pollution before overt damage occurs (Gul et al., 2004). Enzymes such as Aspartate Aminotransferase (AST) and Alanine Aminotransferase (ALT) are valuable biomarkers for assessing pollution levels during chronic exposure (Basaglia, 2000; Ozmen et al., 2006; Younis et al., 2012). Overall, biochemical parameters are reliable biomarkers widely used to detect the adverse effects of toxicants in organisms and to assess environmental risks associated with pollutants (Van der Oost et al., 2003). Hence, the present study was designed to evaluate the effects of ethoxyquin on blood biochemical parametersprotein, glucose, and bilirubin—and on biochemical marker enzymes such as Acid Phosphatase (ACP), Alkaline Phosphatase (ALP), Alanine Transaminase (ALT), and Aspartate Transaminase (AST) in *O. mossambicus* during exposure periods of 24, 48, 72, and 96 hours.

2. Methodology

2.1. Experimental animal

Freshwater fish, *Oreochromis mossambicus*, with an average weight of 12 g and length of 8 cm were collected from the fish farm, Aquafish Aquarium, B.H. Road, Kottakal, Malappuram District. The fish were transported with minimal disturbance to the laboratory in well-aerated polythene bags and acclimatized for one week under constant water supply and proper lighting prior to the experiment. Glass aquaria were washed with 1% $KMnO_4$ solution to prevent fungal contamination and then sun-dried. During acclimatization, fish were fed thrice daily with standard fish pellets and maintained in well-aerated, dechlorinated glass aquaria (30 L capacity) under a 12:12 h light-dark cycle. Fish health was continuously monitored, and unhealthy individuals were removed

Physicochemical parameters of tap water, including temperature (28 ± 2 °C), pH (6.5–7.5), and oxygen saturation (70–100%), were standardized and maintained following APHA (1998) guidelines.

2.2. Chemicals

All reagents used were of analytical grade, purchased from local commercial sources, and used without further purification. Ethoxyquin (1,2-dihydro-2,2,4-trimethylquinolin-6-yl ethyl ether, 75% purity) was procured from Sigma-Aldrich, Germany.

2.3. Determination of Median Lethal Concentration (LC_{50}–96 h)

To determine the 96 h median lethal concentration (LC_{50}) of EQ, acclimatized fish were distributed into six tanks. Fish were unfed for 24 h before exposure to minimize contamination. Ethoxyquin was tested at concentrations of 5, 10, 15, 20, and 25 mg/L in 50 L tanks containing 15 fish each, maintained in triplicate. A control group (toxicant-free) was also maintained. Mortality and behavioral changes were recorded at 24 h intervals up to 96 h. The 96 h–LC_{50} value, representing 50% mortality, was confirmed using Probit regression analysis at a 5% confidence level (Finney, 1971).

2.3.1. Selection of Sub lethal Concentration

Based on the 96 h–LC_{50} value, one-tenth of the LC_{50} concentration was selected as the sub lethal dose for further toxicological studies.

2.4. Experimental Design

Fish were divided into five groups, with nine specimens in each. Group I served as the control (without toxicant). Groups II–V was exposed to the sub lethal concentration ($1/10^{th}$ of 96 h–LC_{50}) for 24, 48, 72, and 96 hours, respectively, representing short-term exposures.

2.4.1. Collection of Blood and Analysis

At the end of each exposure period, fish were gently removed, blotted dry, and blood was collected by severing the caudal peduncle. The first drop was discarded, and the freely oozing blood was collected and allowed to clot at room temperature for 30–60 min. Serum was separated by centrifugation at 1,000 g for 10 min in a cold centrifuge. Total protein was estimated by the method of Lowry et al. (1951), glucose by the Anthrone method (Hedge and Hofreiter, 1962), and bilirubin by the method of Malloy and Evelyn (1937). Activities of Alanine and Aspartate aminotransferases were assayed following Reitman and Frankel (1957), while Acid and Alkaline phosphatases were estimated by the method of King and Armstrong (1934).

2.5. Statistical Analysis

Data were analyzed using one-way ANOVA followed by Duncan's Multiple Range Test in SPSS 17.0. Differences were considered significant at $p < 0.05$ compared with the control group.

3. Results and Discussion

3.1. LC_{50} Evaluation

The mortality of *Oreochromis mossambicus* at different concentrations of ethoxyquin (EQ) over

96 hours is presented in Table 1. Evaluation of the median lethal concentration by probit analysis showed an LC_{50} value of 11.37 mg/L. Lethal Concentration 50 (LC_{50}) is a standard measure of toxicity, representing the concentration of a chemical that kills 50% of a test population within a specified period, typically 24–96 hours. However, not all chemical poisonings result in the immediate death of an organism.

3.2. Blood Biochemical Parameters

The serum protein concentration showed a significant ($p < 0.05$) decrease in fish exposed to EQ at different time intervals (Figure 1). Proteins are vital biomolecules for cellular and tissue functions. The highest protein concentration was recorded in the control group, while the lowest was observed in the 96-hour exposure group. The gradual reduction in protein content indicates a marked influence of EQ on serum protein synthesis and metabolism, suggesting physiological disturbances. Decreased protein levels are often associated with stress or exposure to toxic substances and may also result from enhanced proteolytic activity, increased protease production, or impaired protein synthesis. Shrinkage and lysis of RBCs, plasma dilution, and protein catabolism converting structural protein to energy (Das et al., 2004) may further contribute to reduced serum protein levels.

A significant ($p < 0.05$) decrease in glucose levels was also observed in EQ-exposed fish (Figure 2). This reduction may result from stress-induced metabolic alterations and disruptions in enzymatic activity in the liver under EQ exposure. Decreased glucose levels suggest impaired energy balance and metabolic stress, indicating physiological disturbances in glucose regulation.

Bilirubin, a brownish-yellow pigment formed during erythrocyte degradation in the liver, showed elevated levels in EQ-treated fish compared to the control at all time intervals (Figure 3). Increased bilirubin concentration indicates enhanced erythrocyte breakdown and possible hepatic dysfunction.

The activities of Aspartate Transaminase (AST) and Alanine Transaminase (ALT) were significantly ($p < 0.05$) higher in EQ-exposed fish than in controls (Figure 4). These enzymes serve as indicators of stress and tissue damage. Elevated AST and ALT levels suggest leakage of enzymes from the liver into the bloodstream, reflecting hepatic injury (James et al., 1991; Svoboda, 2001; Chen et al., 2004). The observed increase in plasma AST and ALT activities in *O. mossambicus* exposed to EQ for 24–96 hours implies hepatocellular damage and possible degeneration of liver, heart, or muscle tissues.

Phosphatases catalyze the hydrolysis of phosphoric esters, releasing inorganic phosphate. Alkaline Phosphatase (ALP) and Acid Phosphatase (ACP) activities increased significantly ($p < 0.05$) in EQ-exposed fish compared to controls (Figure 5). Elevated ALP and ACP levels indicate liver injury and necrosis due to cellular damage. The increased enzyme activities suggest degenerative changes and hepatocellular dysfunction, as these enzymes leak from damaged liver cells into the bloodstream. Hence, the elevated ALP and ACP activities in *O. mossambicus* confirm the hepatotoxic effects of ethoxyquin.

Table 1. Mortality (Mean±SD) of *Oreochromis mossambicus* at different concentrations of Ethoxyquin in 96 hrs (n=15)

SL No.	Concentration of Ethoxyquin (mg/L)	Mortality	Mortality (%)
1	5	20.00±1.08	3.00±1.08
2	10	31.11±0.58	4.67±0.58
3	15	62.22±0.58	9.33±0.58
4	20	77.78±0.58	11.67±0.58
5	25	91.11±0.58	13.67±0.58

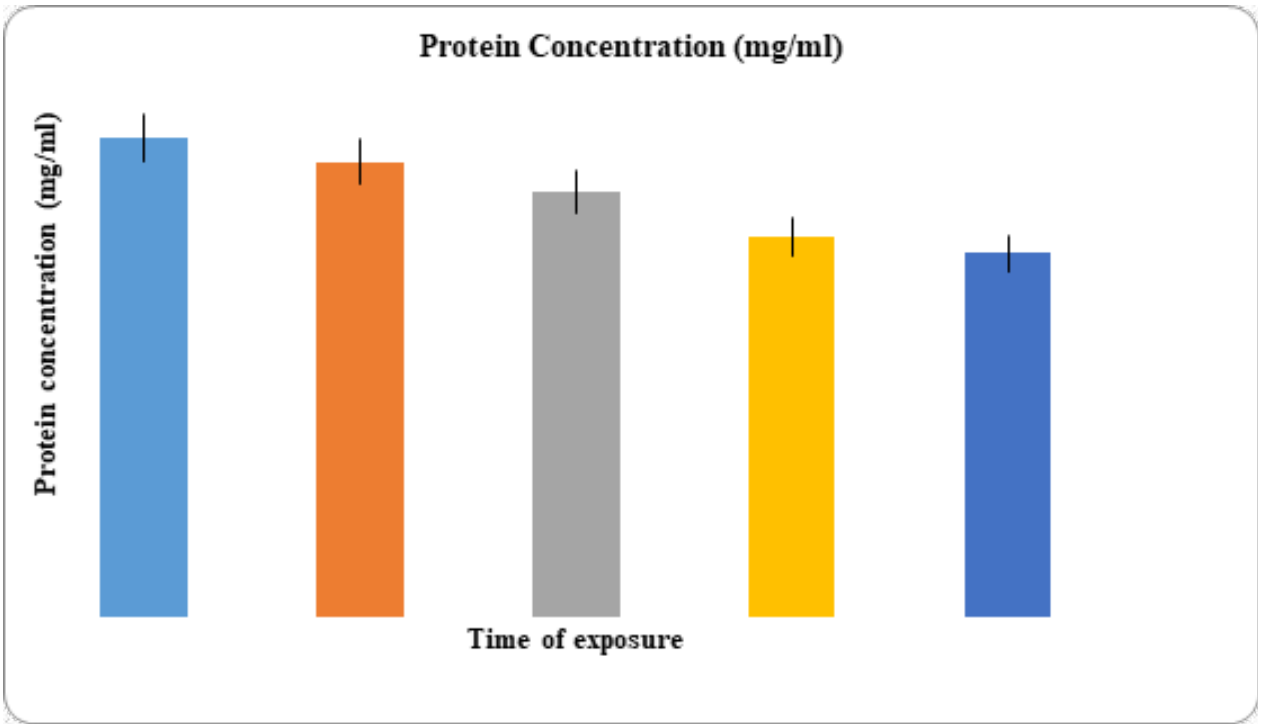

Figure 1. Effect of EQ on concentration of serum protein

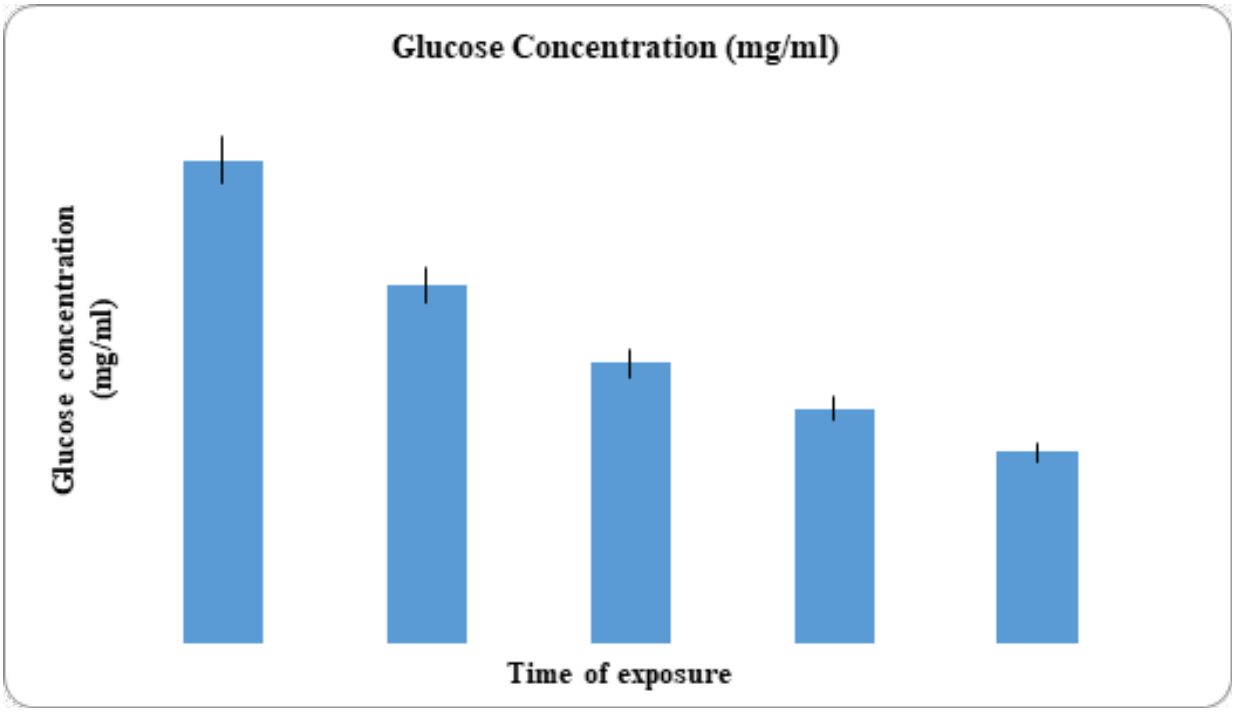

Figure 2. Effect of EQ on concentration of serum glucose

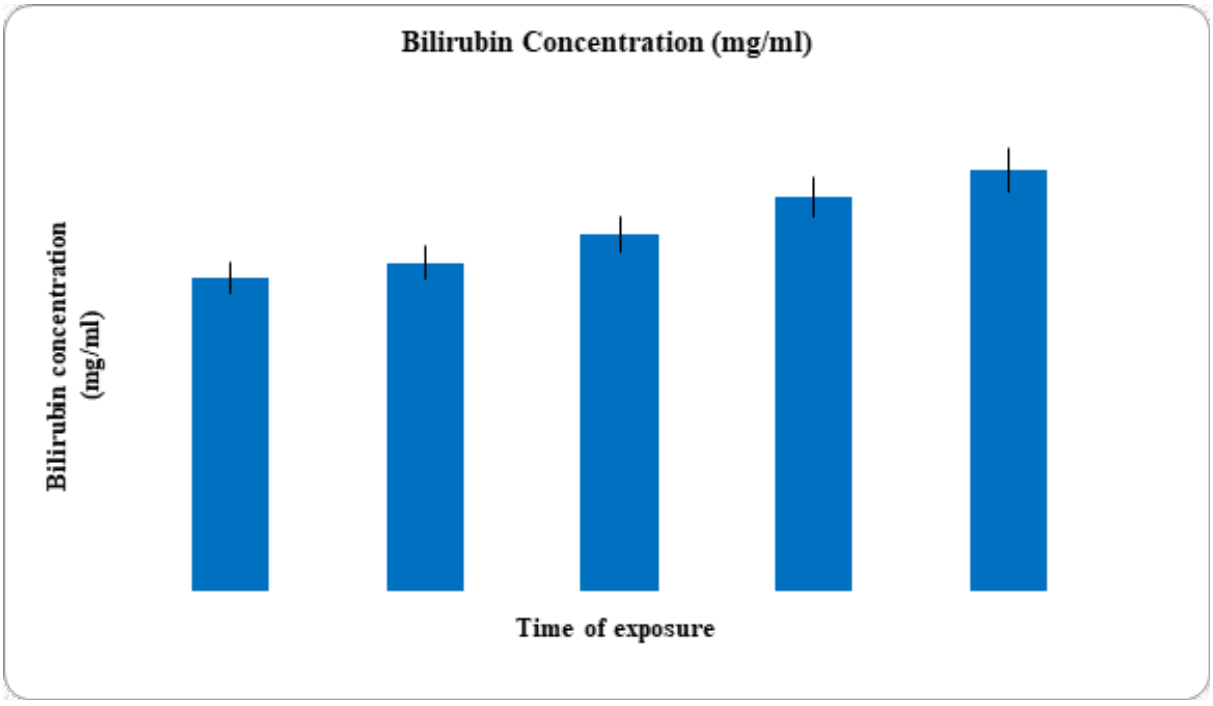

Figure 3. Assessment of Serum Bilirubin on EQ Exposure

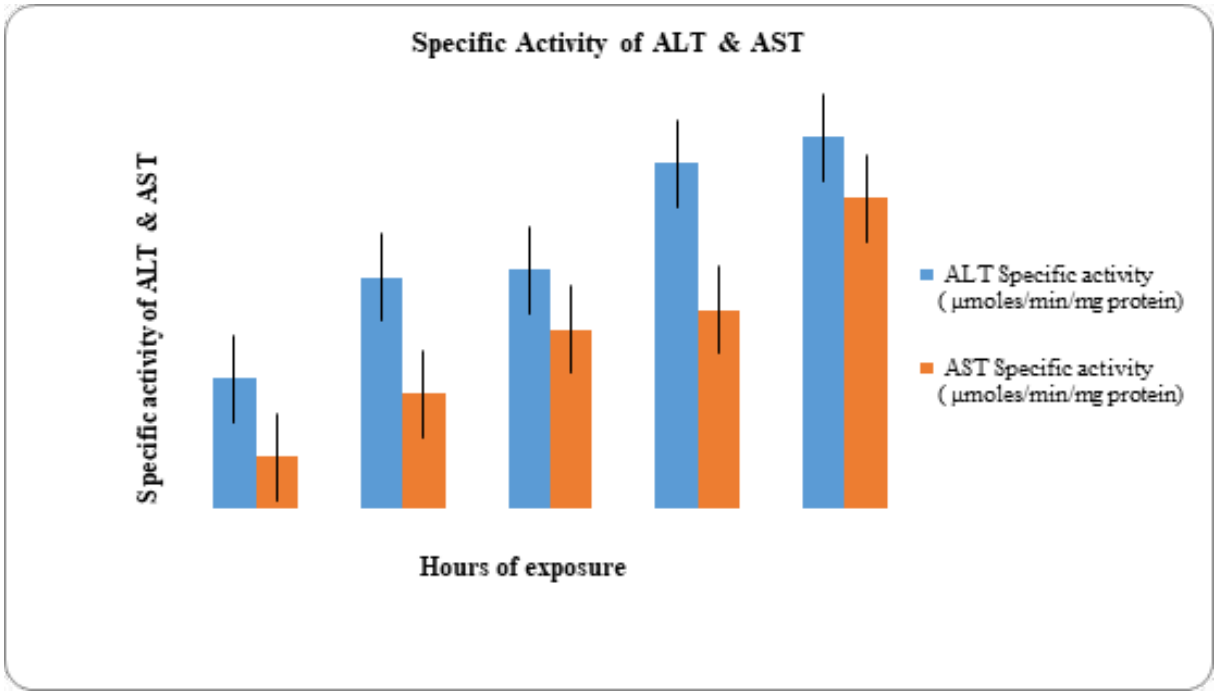

Figure 4. Effect of EQ on concentration of serum ALT and AST Activity

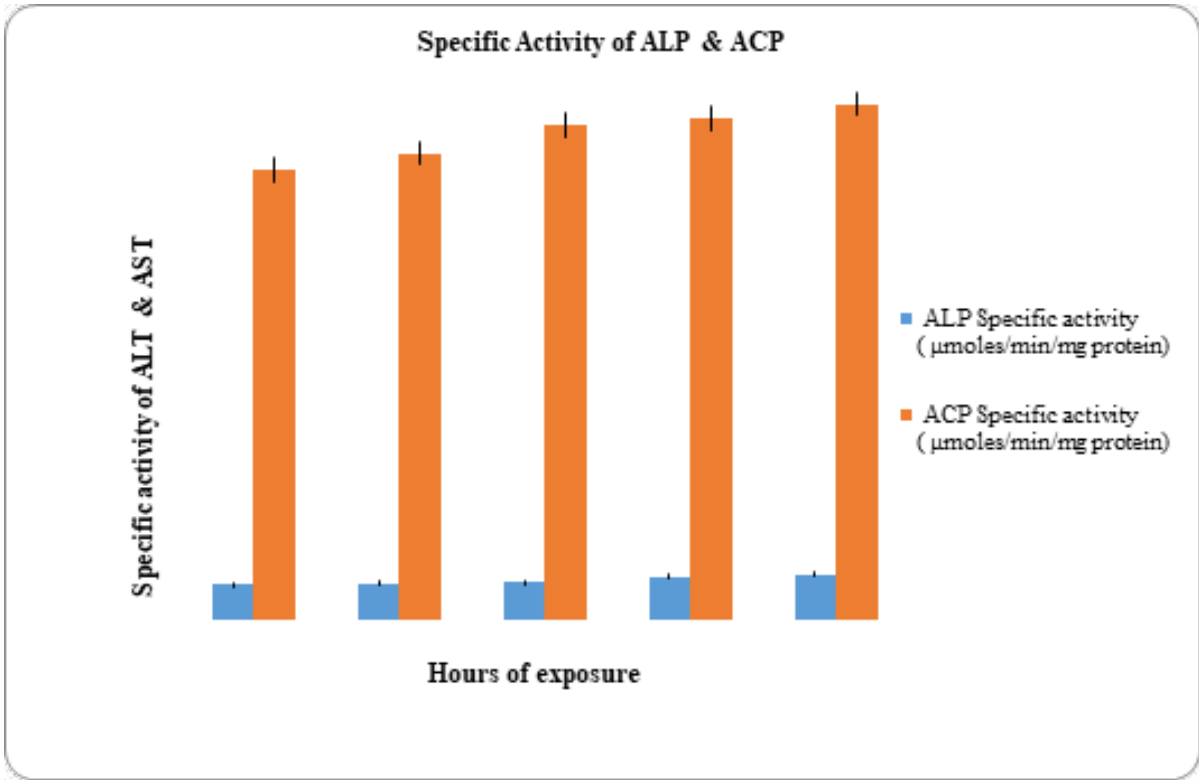

Figure 5. Effect of EQ on concentration of serum ALP and ACP Activity

4. Conclusion

In the present study, the toxic effect of Ethoxyquin (EQ) on *Oreochromis mossambicus* was determined by assessing LC_{50} values calculated at different exposure periods. The fishes showed mortality even at low concentrations, and as the exposure duration decreased, mortality occurred at higher concentrations. The effects of EQ on biochemical variables such as protein, glucose, and bilirubin were also analyzed. Tilapia exposed to EQ exhibited a consistent decrease in blood total protein levels with increasing exposure duration. The experimental fish likely metabolized more proteins in their bloodstream to meet energy demands, resulting in reduced serum protein levels. Additionally, enhanced proteolytic activity, increased protease enzyme production, or decreased protein synthesis under stress may contribute to this reduction. EQ exposure also caused a marked decline in glucose levels across different time intervals, indicating stress-induced hypoglycemia due to hepatic enzyme activation. A significant rise in bilirubin levels was observed, suggesting liver injury and enzyme impairment, as bilirubin levels in fish vary with hepatic health. The study further evaluated the impact of EQ on biochemical markers such as AST, ALT, ACP, and ALP, all of which showed elevated concentrations in treated groups compared to controls. These detoxification enzymes and oxidative stress biomarkers serve as key indicators for identifying polluted sites. Such biomarkers can aid environmental managers in assessing fish exposure to contaminants and evaluating detoxification responses. Overall, the results indicate that both acute and sub lethal EQ exposure induce significant alterations in biochemical and enzymatic parameters of *O. mossambicus*, providing early warning signals of toxicity and ecological risk in aquatic environments.

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