



Original Research Paper

FIELD EVALUATION OF YEAST-FERMENTATION CO₂ AS AN ATTRACTANT IN MOSQUITO TRAPS IN GWALIOR, INDIA

Raju Kushwah^b, Laxmi Gound^b, O. P. Agrawal^b, Sadhana Shrivastava^a and Divya Gupta^{c,a}

^aReproductive Biology and Toxicology Lab, School of Studies in Zoology, Jiwaji University, Gwalior- 474011

^bEntomology Research Lab, School of Studies in Zoology, Jiwaji University, Gwalior- 474011

^cAmity Institute of Biotechnology, Amity University Madhya Pradesh

Corresponding author: Dr. Divya Gupta / Divya121gupta@gmail.com

ABSTRACT

Mosquito-borne diseases continue to pose a significant global health challenge, highlighting the need for sustainable and cost-effective control strategies. Conventional methods relying on chemical insecticides often result in environmental hazards and the development of resistance in mosquito populations. This study evaluates the efficacy of a yeast-sugar fermentation system as a natural carbon dioxide (CO₂) source to enhance mosquito trap performance. Field experiments were conducted in Gwalior, India, using commercially available Terminator-II traps across three seasons (winter, summer, and rainy season) during 2024-2025. The experimental traps were baited with a yeast-sugar solution, while control traps contained only water. The fermentation process generated CO₂, mimicking human respiration and attracting host-seeking mosquitoes. Results demonstrated that yeast-baited traps captured a significantly higher proportion of mosquitoes (67%) compared to control traps (33%), with a total of 2,094 and 1,017 mosquitoes collected, respectively. The dominant species identified was *Culex quinquefasciatus*, followed by other medically important species such as *Aedes aegypti*. The yeast-sugar solution remained effective for up to 23 days under field conditions. These findings suggest that yeast-based CO₂ generation is an efficient, eco-friendly, and affordable approach for improving mosquito trapping systems, particularly in resource-limited settings.

Keywords: Mosquito Traps, Mosquito Repellent, Toxicity, Hazardous Chemicals, Yeast and Sugar Solutions

INTRODUCTION

Mosquitoes are among the most important arthropod vectors of human diseases, transmitting pathogens responsible for malaria, dengue, Zika virus, chikungunya, filariasis, and Japanese encephalitis, particularly in tropical and subtropical regions (Deichstetter, 2017; Nebbak et al., 2022; Xiong et al., 2025). These diseases continue to pose serious public health challenges worldwide, especially in developing countries where environmental conditions favor mosquito breeding and survival. Therefore, effective vector control remains essential for reducing disease transmission and associated health burdens (Wilson et al., 2020).

Conventional mosquito control strategies primarily rely on chemical insecticides such as organophosphates and pyrethroids. Although these chemicals are effective, their prolonged use has led to environmental contamination, toxicity to non-target organisms, and the development of insecticide resistance in mosquito populations (Wilson et al., 2020). These limitations highlight the urgent need for eco-friendly and sustainable alternatives for mosquito control.

Mosquito host-seeking behavior is largely governed by olfactory cues, among which carbon dioxide (CO₂) plays a critical role as a long-range attractant (Cardé & Gibson, 2010; Barredo et al., 2022). CO₂ enhances mosquito responsiveness to other host-associated cues such as body odors, heat, and moisture (Coutinho-Abreu et al., 2021; Mathew et al., 2013). Due to its importance, CO₂ is widely used in mosquito traps to improve their efficiency (Jawara et al., 2011; Hoel et al., 2015). However, conventional CO₂ sources such as dry ice, compressed gas cylinders, and propane-based systems are expensive and often impractical for routine use, particularly in resource-limited settings (Smallegange et al., 2010; Mweresa et al., 2014).

In recent years, fermentation-based systems using yeast and sugar have emerged as a simple, low-cost, and sustainable method for generating CO₂ (Smallegange et al., 2010). During fermentation, yeast metabolizes sugar to produce carbon dioxide and ethanol, thereby mimicking human respiration and attracting host-seeking mosquitoes (Sergi, 2020). Several studies have demonstrated the potential of yeast-generated CO₂ in improving mosquito trap performance under laboratory and semi-field conditions (Astuti et al., 2011; Rosanti et al., 2017).

*Corresponding Author: Divya Gupta

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However, field-based evaluations under natural environmental conditions remain limited.

Therefore, the present study was undertaken to evaluate the efficacy of a yeast–sugar fermentation system as a sustainable CO₂ source for enhancing mosquito trap performance under field conditions in Gwalior, India. By comparing yeast-baited traps with water-based control traps across different seasons, this study aims to assess the practical applicability of this eco-friendly approach for mosquito surveillance and control.

MATERIAL AND METHOD

Study Area

The field study was conducted in Sikander Kampoo, located in the Lashkar region of Gwalior, Madhya Pradesh, India. This area is characterized by the presence of stagnant water bodies, including the Veerpur and Hanuman dams, which provide favourable breeding habitats for mosquitoes. The region experiences distinct seasonal variations, including winter, summer, and monsoon periods, making it suitable for evaluating mosquito trapping efficiency under varying environmental conditions.

Experimental Design

The study was carried out in two phases to evaluate (i) the efficacy of a yeast–sugar fermentation system as a carbon dioxide (CO₂) source for mosquito attraction and (ii) the duration of its effectiveness under field conditions.

In Phase I, a comparative field experiment was conducted using two commercially available Terminator-II mosquito traps. One trap served as the control (containing plain water), while the other functioned as the experimental setup, incorporating a yeast–sugar fermentation solution as a CO₂ source. The experiments were conducted across three seasons (winter, summer, and rainy season) during 2024–2025.

Each experimental setup was replicated three times per season. To minimise positional bias, the placement of control and experimental traps was alternated daily. The traps were installed indoors in a residential setting within the study area and operated continuously for 24 hours during each trial.

Preparation of Yeast–Sugar Fermentation Solution

The fermentation mixture was prepared by dissolving 100 g of sugar in 750 mL of water in a 1-L plastic bottle. Subsequently, 8 g of sodium bicarbonate, 8 g of yeast extract powder, and 5.5 g of active dry yeast were added to the solution. The mixture was gently shaken to ensure uniform mixing and allowed to ferment for 2–12 hours prior to use.

The bottle was fitted with a tightly sealed lid containing a 1 mm diameter through which a plastic tube was inserted.

The tube connected the fermentation chamber to the mosquito trap, allowing continuous release of CO₂ produced during fermentation. CO₂ production was qualitatively confirmed by visible bubbling in the solution.

Trap Operation and Sample Collection

Both control and experimental traps were operated simultaneously for 24-hour periods. Captured insects were collected from the traps into Petri dishes and subsequently dried in a laboratory oven at 60°C for 12 hours to remove moisture. The dried specimens were stored in airtight containers for further identification and analysis.

Insect Identification

Collected specimens were sorted and identified based on morphological characteristics. Mosquitoes were identified up to the genus or species level using standard taxonomic keys (Christophers, 1933; Nagpal & Sharma, 1995). Only adult mosquitoes were included in the final analysis.

Assessment of Trap Efficiency

Trap efficiency was evaluated by comparing the number of mosquitoes captured in experimental and control traps. Data were recorded for each trial and expressed as mean $\pm$ standard error (SE). Seasonal variations in mosquito abundance and species composition were also analyzed.

Phase II: Duration of CO₂ Production

In Phase II, the longevity of the yeast–sugar fermentation system was assessed. A single preparation of the fermentation mixture was monitored daily over a period of 30 days. The effectiveness of the solution was determined based on the number of mosquitoes captured per day, with declining capture rates indicating reduced CO₂ production.

Statistical Analysis

All data were compiled and analyzed using appropriate statistical methods. Differences between control and experimental groups were evaluated using Student's t-test (or one-way ANOVA where applicable). Results were considered statistically significant at $p < 0.05$.

RESULT AND DISCUSSION

Trap Efficiency and Seasonal Variation

The field evaluation demonstrated that mosquito traps baited with the yeast–sugar fermentation solution consistently captured a higher number of mosquitoes compared to control traps containing only water across all seasons. As shown in Table 1, a total of 2,094 mosquitoes were collected from experimental traps, whereas 1,017 mosquitoes were recorded in control traps, accounting for approximately 67% and 33% of the total catch, respectively. This substantial increase in trapping efficiency can be attributed to the presence of carbon dioxide (CO₂), a key activator of mosquito host-seeking behavior (Cardé, 2010; Lahondère & Lazzari, 2012).

Table-1: Numbers of insects trapped in control and Experimental trap

Mean (SE) % of Trapped Insects			
	Winter	Summer	Rainy

Insects	Control	Experimental	Control	Experimental	Control	Experimental
Mosquitoes	12.25 (2.01)	30.50 (8.17)	122.75 (43.70)	240.5 (94.77)	119.25 (42.64)	252.5 (92.57)
Psycodids	16.50 (5.20)	226.5 (180.82)	41.5 (29.68)	63 (50.75)	38 (30.30)	58 (11.23)
Sandlies	4.50 (3.06)	1 (0.70)	9.5 (4.55)	18.25 (5.72)	5.25 (2.86)	16.75 (6.44)
Other Insects	8 (2.12)	25 (13.67)	64 (49.56)	40.75 (15.47)	52.25 (25.75)	142.25 (41.52)
Houseflies	0 (0)	0.75 (0.47)	3 (2.38)	2 (1.35)	3.5 (2.87)	15 (11.68)

Seasonal variation in mosquito abundance was clearly observed. The highest number of mosquitoes was recorded during the rainy season, followed by summer and winter (Table 1). The maximum mean catch was observed in the experimental trap during the rainy season (252.5 ± 92.57), while the lowest was recorded in the control trap during winter (12.25 ± 2.01). This trend is also illustrated in

Figure 2, which shows a clear increase in insect capture in experimental traps across all seasons. The higher mosquito density during the rainy season is likely due to the availability of abundant breeding sites and favorable environmental conditions such as increased humidity and temperature (Okiwelu & Noutcha, 2012; Marzal et al., 2022).

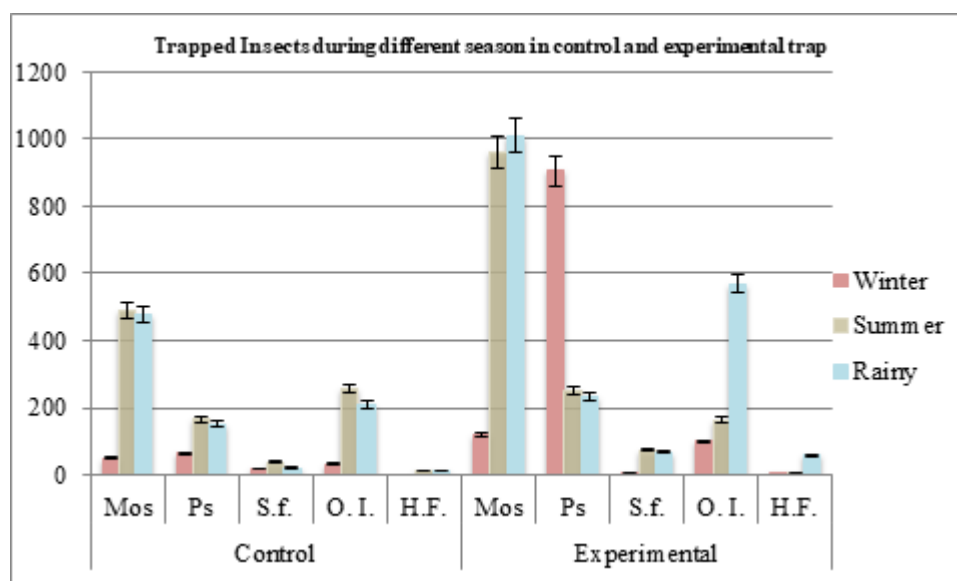


Figure-2: Graphs showing the trapped insects in control and experimental trap within the study period.

Here: Mos.-Mosquitoes, Ps-Psycodidae, S.f.-Sandflies, O.I.-Other Insects, H.F- Housefly.

Species Composition

The species composition of mosquitoes captured in both control and experimental traps is presented in Table 2. The

dominant species recorded was *Culex quinquefasciatus*, followed by *Aedes aegypti*, *Armigeres subalbatus*, *Culex tritaeniorhynchus*, and *Culex vishnui*. Minor occurrences of *Anopheles* species, including *Anopheles stephensi* and *Anopheles annularis*, were also observed.

Table-2: Numbers of insects trapped in control and Experimental trap

Mean (SE) % of mosquito species						
	Winter		Summer		Rainy	
Mosquito species	Control	Experimental	Control	Experimental	Control	Experimental
<i>Ae. al.</i>	0.5 (0.5)	2 (1.22)	1.5 (0.95)	1.5 (0.95)	11.5 (6.11)	14 (4.91)
<i>Ae. ae.</i>	2 (1.08)	3.25 (2.35)	4.5 (2.72)	12.5 (9.66)	23.5 (2.35)	38 (12.78)
<i>Cu. qu.</i>	4.5 (1.55)	13.5 (2.84)	89.75 (35.67)	186.5 (71.64)	52.75 (23.18)	123.25 (56.34)
<i>Cu. vi.</i>	0.25 (0.25)	3 (1.77)	13 (6.23)	21.5 (14.67)	3 (1)	10.5 (4.99)
<i>Cu. tri.</i>	1.25 (0.47)	4.25 (1.79)	9.5 (2.63)	15 (7.29)	5.5 (3.09)	21.5 (13.42)
<i>An. st.</i>	0 (0)	0.25 (0.25)	0.5 (0.5)	0 (0)	4.75 (3.54)	5 (3.53)
<i>An. an.</i>	0 (0)	0 (0)	0.5 (0.5)	0.75 (0.75)	3.25 (2.62)	13.75 (11.8)
<i>An. sub.</i>	0 (0)	0 (0)	0.25 (0.25)	0 (0)	7.75 (7.75)	8.5 (7.84)
<i>Ar. sub.</i>	3.5 (1.25)	4.25 (1.88)	3.25 (2.35)	2.75 (1.37)	7.25 (4)	18 (10.72)

Abbreviations- *Ad.al-Aedes albopictus*, *Ad. ae-Aedes aegypti*, *Cu. qu-Culex quinquefasciatus*; *Cu.vi.-Culex vishnui*; *Cu.tri-Culex Tritaenorrhynchus*; *An.st-Anopheles stephensi*; *An.an-Anopheles annularis*; *An. sub-Anopheles subpictus* *Armi. sub-Armigera subalbus*.

The predominance of *Culex quinquefasciatus* may be attributed to its adaptability to polluted and stagnant water habitats commonly found in the study area (Okiwelu & Noutcha, 2012). As shown in Table 2, experimental traps consistently captured a higher proportion of all mosquito species compared to control traps. This indicates that CO₂ generated from fermentation acts as a universal attractant

across multiple mosquito taxa (Jawara et al., 2011; Hoel et al., 2015).

Effect of Yeast–Sugar Fermentation as CO₂ Source

The enhanced performance of experimental traps is directly linked to the continuous release of CO₂ during yeast fermentation. The experimental setup, including the connection between the fermentation bottle and the mosquito trap, is illustrated in Figure 1. The CO₂ produced during fermentation mimics human respiration, thereby increasing the attractiveness of the trap to host-seeking mosquitoes.

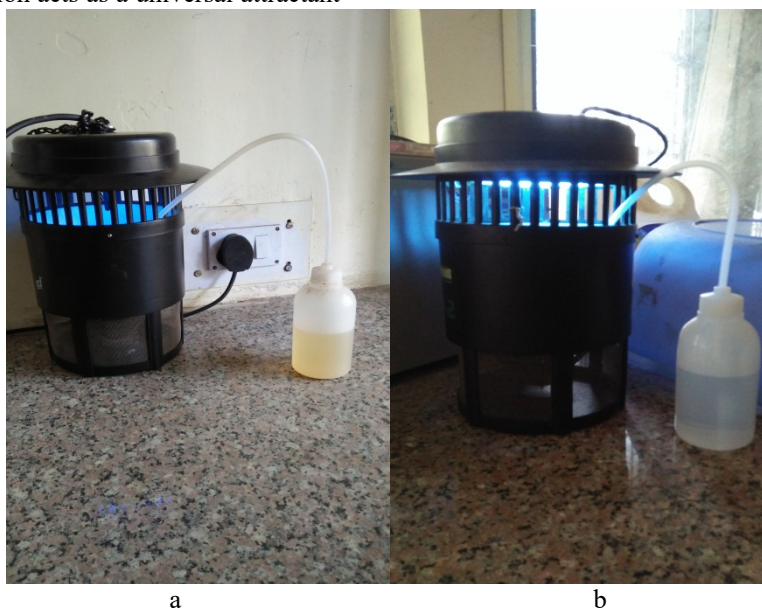


Figure-1: Showing the installed trap with a) Yeast and sugar solution and b) Normal tap water.

The results are consistent with previous studies demonstrating that fermentation-derived CO₂ significantly improves mosquito trap efficiency (Smallegange et al., 2010; Mweresa et al., 2014). Similar findings have been reported in field and laboratory studies using yeast-sugar solutions as attractants (Astuti et al., 2011; Rosanti et al., 2017). The present study further confirms the effectiveness of this approach under natural environmental conditions.

Non-Target Insect Capture

In addition to mosquitoes, other insects such as psychodids, sandflies, and houseflies were also captured in both control and experimental traps, as shown in Table 1. Although these non-target insects were present, their

numbers were relatively lower compared to mosquitoes. This suggests that CO₂ and associated odor cues may attract a broader range of insects, as also reported by Kline et al., (2012). However, this does not significantly affect the primary objective of mosquito trapping.

Duration of Fermentation Effectiveness

The duration of effectiveness of the yeast–sugar fermentation system is illustrated in Figure 3, which shows the variation in mosquito capture over time. The fermentation system remained effective for up to 23 days, after which a decline in trap efficiency was observed. This decrease is likely due to reduced CO₂ production as fermentation slows over time.

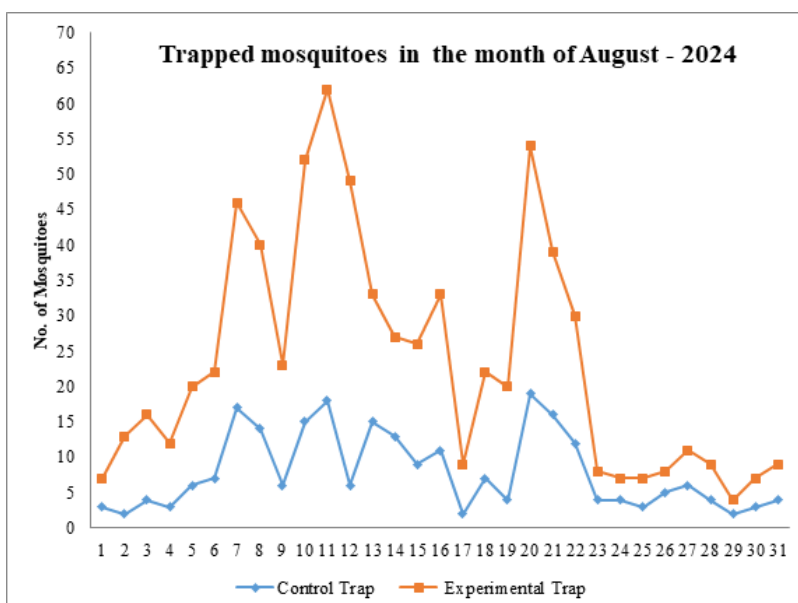


Figure-3: Mosquitoes catch in control and experimental trap in the month of August 2024.

These findings are consistent with previous studies reporting effectiveness of yeast-sugar solutions for approximately 25-30 days (Astuti et al., 2011; Rosanti et al., 2017). The relatively long duration of CO₂ production highlights the practical advantage of this method, as it reduces the need for frequent replacement of the attractant solution.

Limitations of the Study

Despite the promising results, certain limitations should be acknowledged. The study was conducted at a single location, which may limit the generalizability of the findings. Environmental factors such as temperature and humidity were not quantitatively measured, although they are known to influence mosquito behavior (Coutinho-Abreu et al., 2021). Additionally, the use of water as a control instead of a standard CO₂ source limits direct comparison with conventional trapping systems.

Conclusion from Findings

Overall, the results clearly demonstrate that yeast-sugar fermentation significantly enhances mosquito trap efficiency by providing a continuous and cost-effective source of CO₂. The integration of this method into mosquito surveillance and control programs may provide an eco-friendly and practical solution, particularly in resource-limited settings.

Conflict of interest

The authors declare there is no conflict of interest.

REFERENCES

- Astuti EP, dan-Roy NRES, 2011: Efektifitas Alat Perangkap (Trapping) Nyamuk Vektor Demam Berdarah Dengue dengan Fermentasi Gula. *Aspirator*. 3 (1) : 41-48.
- Barredo E, Raji JI, Matthew MR, DeGennaro, Theobald J, 2022: Carbon dioxide and blood-feeding shift visual cue tracking during navigation in *Aedes aegypti* mosquitoes. *Published-https://doi.org/10.1098/rsbl.2022.0270*.

Bursali F, Touray M, 2024: The complexities of blood-feeding patterns in mosquitoes and sandflies and the burden of disease: A mini review. *Veterinary Medicine and Science*. **Volume-10, issue-5**.

Carde RT, Gibson G., 2010: Host finding by female mosquitoes: mechanism of orientation to host odours and others cues. *Olfaction in Vector-Host Interactions*. Edited by Takken W, Knols BGJ. Wageningen Academic Publishers. 115-142.

Christophers SR. 1933: The fauna of British India, including Ceylon and Burma. Diptera: Family Culicidae; Tribe Anophelini. Vol. 4, Taylor & Francis, London.

Cook JI, Majeed S, Ignell R, Pickett JA, Birkett MA, Logan JG., 2011: Enantiomeric Selectivity in Behavioural and Electrophysiological Responses of *Aedes aegypti* and *Culex quinquefasciatus* Mosquitoes. *Bull Entomol Res*. 101(5):541-550.

Coutinho-Abreu IV, Riffell JA, Akbari OS, 2021: Human attractive cues and mosquito host-seeking behavior. *Trends Parasitol*. 38(3) 246-264.

Deichstetter, P. (2017): The effect of climate change on Mosquito-Borne diseases. *The American Biology Teacher*, **79(3)**: 169-173.

Fan Z, Zhao T, Gu Z, Gao H, Zhou X, Yu H, Xing D, Wang H, Li C, 2025: Differences in human skin volatiles between populations with high and low attraction to mosquitoes. *Parasites and Vectors*. 18:183.

Ganai, S.A., Kumar, R., Basheer, M., and Agrawal, O.P. (2013): A study on sampling of mosquitos using eco-friendly mosquito trap in around Jiwaji University Campus (India). *Munis Entomology & Zoology*, **8(2)**: 734-738.

- Guha, L., Seenivasagan, T., Prabal, B., Iqbal, S.T., Sathe, M., Sharma, P., Parashar, B.D. and Kaushik, M.P. (2012): Oviposition and flight orientation response of *Aedes aegypti* to certain aromatic aryl hydrazono esters. *Parasitol. Res.*, **111**(3): 975-982.
- Guidobaldi F. & Guerenstein PG., 2013: Evaluation of a CO₂-free Commercial Mosquito Attractant to Capture Triatomines in The Laboratory. *Journal of Vector Ecology*. 38 (2) : 245-250.
- Hoel DF, Dunford JC, Kline D, Wirtz RA, 2015: A Comparison of Carbon Dioxide Sources for Mosquito Capture in Centers for Disease Control and Prevention Light Traps on the Florida Gulf Coast. *Journal of the American Mosquito Control Association*. 31(3): 248-57.
- Jawara M, Taiwo S, Amargaret P, David J, Renata CS, Willem T, David JC., 2011: Field Testing of Different Chemical Combinations as Odour Baits for Trapping Wild Mosquitoes in The Gambia. *PloS One*. 6:e19676.
- Kline, DL., Bernier, UR & Hogsette, JA., 2012: Efficacy of Three Attractant Blends Tested in Combination with Carbon dioxide against Natural Population of Mosquitoes and Biting Flies at The Lower Suwannee Wildlife Refuge. *Journal of the American Mosquito Control Association*. 28 (2) : 123-127.
- Lahondere C & Lazzari CR., 2012: Mosquitoes cool down during blood feeding to avoid overheating. *Current Biology*. 22 (1) : 40-45.
- Madang S, Saingamsook J, Saeung A, Samboon P, 2022: A Simple CO₂ Generating System Incorporated with CDC Light Trap for Sampling Mosquito Vectors. *Insects*. 13(7); 637.
- Marzal A, Magallanes S, Garcia-Longoria L, 2022: Stimuli Followed by Avian Malaria Vectors in Host-Seeking Behaviour. *Biology(Basel)*. 11(5) 726.
- Mathew N.,Ayyanar E., Shanmugavelu S.,Muthuswamy K., 2013: Mosquito Attractant Blends to Trap Host Seeking *Aedes aegypti*. *Parasitology Research*. 112 (3) : 1305-1312.
- Mweresa CK, Omusula P, Otieno B, JAVanLoon , Taken W & Mukabana WR., 2014: Mollases as a Source of Carbon dioxide for Attracting the Malaria Mosquitoes *Anopheles gambiae* and *Anopheles funestus*. *Malaria Journal*. 13 (1) : 160.
- Nagpal, B.N., and Sharma,V.P. (1995): Indian Anopheline. *NewDelhi: Oxford and IBH Publishing Co. Pvt. Ltd.* p 416.
- Nebbak A, Almeras L, Parola P, Idir Bitam I, 2022: Mosquito Vectors (Diptera: *Culicidae*) and Mosquito-Borne Diseases in North Africa. *Insects* 13(10):962.
- Okiwelu, S.N & M.A.E. Noutcha., 2012: Breeding sites of *Culex quinquefasciatus* (say) during the rainy season in rural lowland rainforest reinstate, Nigeria. *Public Health Research*. 2 (4) : 64 – 68.
- Olanga EA, Okal MN, Mbadi PA, Kokwaro ED, Mukabana WR., 2010: The attraction of *Anopheles gambiae* to Odour Baits Augmented with Heat and Moisture. *Malaria Journal*. 9 (1) : 6-15
- Polakowski & Tessa M., 2008: The Effect of Different Sugars on The Rate of Fermentation in Yeast. *Academic Journal*. 108 (1) : A-30.
- Poulin B, Lefebvre G, Muranyi-Kovacs C, Hilaire S, 2017: Mosquito Traps: An Innovative, Environmentally Friendly Technique to Control Mosquitoes. *Int. J. Environ. Res. Public Health*. 14(3) 313.
- Ramadhani T, Soeyoko, Sumarni S., 2010: *Culex quinquefasciatus* sebagai Vektor Utama Filariasis Limfatik yang Disebabkan Wuchereria bancrofti di Kelurahan Pabean Kota Pekalongan. *Jurnal Ekologi Kesehatan*. 9 (3) :1303-10.
- Rosanti TI, Mardihusodo SJ, Artama WT, 2017: Effectiveness of Environmentally Friendly Mosquito Trap Contained Sugar Yeast Solution. *Journal Kesehatan Masyarakat*. 12(2) 96-102.
- Sergi Maicas, 2020: The Role of Yeasts in Fermentation Processes. *MDPI Journal of microorganisms*.**8**:1142.
- Smallegange RC, Schiled WH, Van Roey KJ, Verhulst NO, Spitzen J, Mukabana WR, Taken W., 2010: Sugar-Fermenting Yeast as an Organic Source of Carbon dioxide to Attract the Malaria Mosquito *Anopheles gambiae*. *Malaria Journal*. 9 (1) : 292.
- Verhulst NO, Mukabana WR, Takken W, Smallegange RC, 2011: Human Skin Microbiota and Their Volatiles as Odour Baits for The Malaria Mosquito *Anopheles gambiae*s. *Entomol Exp Appl*. 139 : 170-179.
- Wilke ABB, Vasquez C, Carvajal A, Moreno M, Petrie WD, Beier JC, 2022: Evaluation of the effectiveness of BG-Sentinel and CDC light traps in assessing the abundance, richness, and community composition of mosquitoes in rural and natural areas. *Parasites and Vectors*. 15(1) 51.
- Wilson AL, Courtenay O, Kelly-Hope LA, Scott TW, Takken W, Torr SJ, Lindsay SW, 2020: The importance of vector control for the control and elimination of vector-borne diseases. *PLoS Negl Trop Dis*. 16;14(1).
- Xiong S, Liang J, Gao S, Liu Z, Zheng H, Yang X, Wang Y, Yu S, 2025: The chemosensory world of mosquitoes: olfactory receptors and their role in blocking mosquito-borne disease transmission. *Parasites and Vectors*. Volume-18, 15:51.