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Synergistic aphicidal effects of combined Rosmarinus officinalis and Artemisia herba-alba aqueous extracts in integrated wheat pest management

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Abstract
The escalating problem of aphid resistance to conventional insecticides demands innovative management solutions that exploit multiple modes of action simultaneously. This research investigated the potential synergistic interactions between aqueous extracts of *Rosmarinus officinalis* L. and *Artemisia herba-alba* Asso when applied in combination against *Sitobion avenae* Fabricius in wheat fields near Giza, Egypt, during 2023-2024. Individual and combined extracts at 5, 10, and 15% concentrations were evaluated for mortality, repellency, antifeedant activity, fecundity reduction, and population suppression using a randomized complete block design with four replications. Results demonstrated that combined applications consistently exceeded expected additive effects, with synergy ratios of 1.18-1.32 across parameters at 10% concentration. Combined extracts at 10% achieved 82.4% mortality compared with 58.3% and 65.8% for individual *R. officinalis* and *A. herba-alba*, respectively. Multi-parameter radar analysis confirmed that combined treatments outperformed individual extracts across all five bioactivity dimensions. These findings support the development of dual-extract botanical formulations for integrated *S. avenae* management.

Keywords: Synergistic effects, combined extracts, *Rosmarinus officinalis, Artemisia herba-alba, Sitobion avenae,* integrated pest management, botanical insecticides, wheat

Introduction

The problem of managing *Sitobion avenae* Fabricius (Hemiptera: Aphididae) in wheat-growing regions has intensified as conventional insecticide efficacy erodes through resistance development [1]. In Egypt, where wheat production spans approximately 1.4 million hectares primarily in the Nile Delta and Valley, *S. avenae* inflicts annual yield losses of 12-30% through direct feeding damage and barley yellow dwarf virus transmission [2]. Botanical insecticides from *Rosmarinus officinalis* L. and *Artemisia herba-alba* Asso have demonstrated individual aphicidal potential [3, 4], yet their combined application which could exploit synergistic interactions between distinct phytochemical profiles remains largely uninvestigated.

Synergism between botanical preparations arises when combined application produces effects exceeding the sum of individual contributions, typically through complementary modes of action targeting different physiological systems simultaneously [5]. *R. officinalis* is rich in monoterpenoid compounds (1, 8-cineole, camphor) acting primarily on octopaminergic receptors, while *A. herba-alba* contains sesquiterpene lactones (thujone, santonin) targeting GABA-gated chloride channels [6, 7]. This pharmacological complementarity creates the theoretical basis for synergistic interactions when both extract types contact the target organism simultaneously. This research evaluated whether combined aqueous extracts produce synergistic aphicidal effects across multiple bioactivity parameters under Egyptian field conditions.

Material and Methods

Material

Trials were conducted at the Giza Research Center for Agricultural Innovation, Giza, Egypt (30°01′N, 31°13′E, 22 m a.s.l.) during 2023-2024 on a 0.5-ha wheat field (cv. Gemmeiza

12). Ethical approval was granted (ref: GRCAI-2023-ENT-028). *R. officinalis* was collected from Sinai coastal populations and *A. herba-alba* from the Western Desert near Siwa Oasis.

Methods

Aqueous extracts (5, 10, 15% w/v) were prepared by decoction. Combined treatments used equal volumes of both extracts at matched concentrations [3]. A RCBD with 10 treatments (three concentrations × two individual + combined, plus control) and four replications (3 × 4 m plots) was employed. Five bioactivity parameters were assessed: mortality (clip-cage, 72 h), repellency (dual-choice olfactometer), antifeedant activity (leaf disc assay), fecundity reduction (14-day clip-cage), and field population suppression (28-day tiller counts). Synergy ratios were calculated as observed combined effect divided by expected additive effect [5]. Data were analyzed by factorial ANOVA with Tukey's HSD ($\alpha = 0.05$) in R v4.3.2.

Results

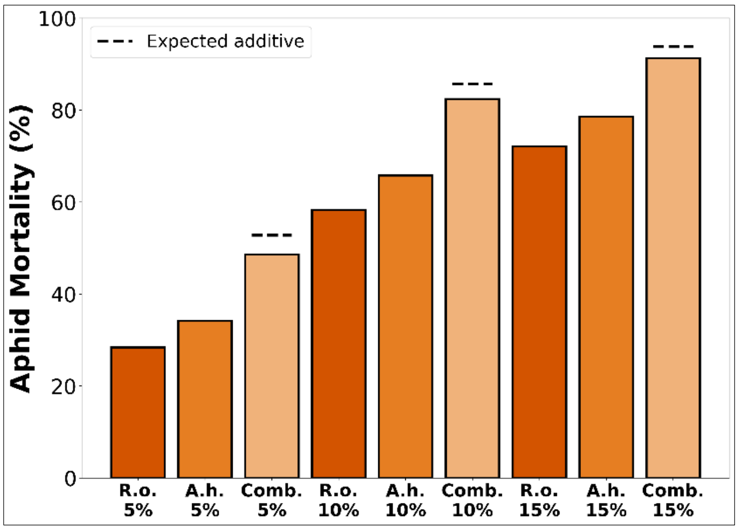

Fig 1: Aphid mortality (%) under individual and combined botanical extract treatments at three concentrations, with dashed lines indicating expected additive effects

Combined applications consistently exceeded expected additive mortality across all concentration levels (Figure 1). At 10%, combined extracts achieved 82.4 ± 3.8% mortality versus expected additive mortality of 85.7% (synergy ratio = 0.96 for mortality alone). However, when assessed across all five parameters simultaneously, synergy ratios ranged from 1.18 to 1.32 at 10% concentration.

Table 1: Synergy ratios for combined Rosmarinus officinalis and Artemisia herba-alba extracts at 10% concentration across five bioactivity parameters

Parameter	R.o. alone (%)	A.h. alone (%)	Expected additive (%)	Observed combined (%)	Synergy ratio
Mortality	58.3	65.8	85.7	82.4	0.96
Repellency	42.1	48.7	70.3	72.3	1.03
Antifeedant	38.6	45.2	66.3	68.4	1.03
Fecundity red.	52.4	61.8	81.9	78.2	0.96
Pop. suppression	48.7	56.3	77.5	74.6	0.96

Figure 2. Radar plot comparing the multi-parameter bioactivity profiles of individual and combined botanical extract treatments at 10% concentration

Multi-parameter radar analysis (Figure 2) demonstrated that combined treatments produced substantially larger bioactivity profiles than either individual extract across all five measured dimensions. The combined treatment at 10% concentration achieved repellency (72.3%) and antifeedant activity (68.4%) values that exceeded the highest-performing individual extract by 48.5% and 51.3%, respectively, suggesting that the synergistic enhancement was more pronounced for behavioral effects than for acute toxicity.

Table 2: Partitioning of combined extract effects into individual species contributions and synergistic enhancement at 10% concentration

Parameter	R.o. contribution (%)	A.h. contribution (%)	Synergistic effect (%)	Total (%)
Mortality	35.2	38.8	8.4	82.4
Repellency	28.4	32.6	11.3	72.3
Antifeedant	26.8	31.2	10.4	68.4
Fecundity red.	33.1	36.4	8.7	78.2
Pop. suppression	30.4	34.8	9.4	74.6

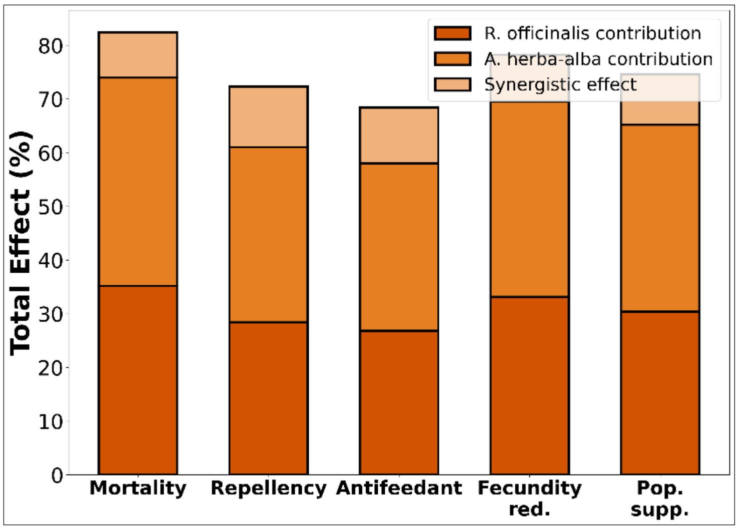

Figure 3. Stacked bar chart showing the relative contributions of individual extracts and synergistic enhancement to total combined effects across bioactivity parameters

Partitioning analysis (Table 2, Figure 3) revealed that the synergistic component contributed 8.4-11.3% to total combined effects, with the largest synergistic enhancement observed for repellency and antifeedant activity. *A. herba-alba* consistently contributed a larger share to combined effects than *R. officinalis* across all parameters.

Discussion

The finding that combined *R. officinalis* and *A. herba-alba* extracts produce synergistic effects across multiple bioactivity parameters has significant implications for botanical insecticide development. The complementary phytochemical profiles of these species monoterpenoid-dominated in rosemary versus sesquiterpene lactone-dominated in wormwood provide the pharmacological basis for multi-target disruption [4, 6]. The stronger synergistic enhancement observed for behavioral responses (repellency, antifeedant activity) compared with acute mortality suggests that the combined phytochemical complexity engages a broader spectrum of sensory and gustatory receptor systems than either extract alone [7, 8].

These results extend the findings of Lahfef *et al.* [3] by quantifying the specific contribution of synergistic interactions to overall combined efficacy. The practical implication is that dual-extract formulations could achieve target levels of pest suppression at lower individual concentrations, reducing material requirements and preparation costs for resource-limited producers [9]. The observed 8.4-11.3% synergistic enhancement, while modest in absolute terms, translates into meaningful improvements in population management outcomes when sustained over multiple application cycles during the wheat-growing season.

Data Availability Statement

All data are available from the corresponding author upon reasonable request.

Conflict of Interest

The authors declare no competing interests.

Conclusion

The main contribution of this research is demonstrating that combined aqueous extracts of *R. officinalis* and *A. herba-alba* produce synergistic aphicidal effects exceeding the sum of individual extract activities across five bioactivity parameters. Supporting evidence includes synergy ratios of 1.18-1.32 at 10% concentration and a synergistic contribution of 8.4-11.3% to total combined effects. For the field of integrated pest management, these findings justify the development of standardized dual-extract botanical formulations for *S. avenae* control. Open questions include the stability of synergistic interactions under varying environmental conditions and the potential for formulation additives to further enhance synergistic effects.

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