

Supplementary Materials: The following are available online: Table SI-1.1: Statistical analysis of fly fertility after exposure to Fe₃O₄NPs, Ch-Fe₃O₄NPs, and control; Table SI-1.2: Tukey's HSD test results for control, Fe₃O₄NPs, and Ch-Fe₃O₄NPs groups; Table SI-2.1: Linear fit, upper, and lower confidence limits (UCL and LCL) for each iron concentration within the studied ranges; Table SI-3.1: Iron concentrations in individual culture medium components.

SI-1: Fertility: Statistical analysis

Table SI-1.1 Statistical analysis of fly fertility after exposure to Fe₃O₄NPs, Ch-Fe₃O₄NPs, and control

| NP Concentration | Fe ₃ O ₄ NPs | | | | | | Ch-Fe ₃ O ₄ NPs | | | | | |
|------------------|------------------------------------|-------|----------------|-----|-------|----------------------------|---------------------------------------|-------|----------------|------|------|----------------------------|
| | N | Mean | Std. Deviation | F | Sig. | Duncan's test ^a | N | Mean | Std. Deviation | F | Sig. | Duncan's test ^a |
| 0 ppm | 10 | 119.7 | 37.256 | 1.5 | 0.208 | A | 10 | 119.7 | 32.511 | 22.9 | 0,00 | A |
| 100 ppm | 10 | 94.2 | 34.653 | | | A | 10 | 24.4 | 16.001 | | | B |
| 250 ppm | 10 | 91.4 | 34.1 | | | A | 10 | 34.2 | 20.357 | | | B |
| 500 ppm | 10 | 93 | 33.86 | | | A | 10 | 32.2 | 25.046 | | | B |
| 1000 ppm | 10 | 85.7 | 27.92 | | | B | 10 | 42.7 | 25.456 | | | B |

^a Means with the same letter are not significantly different (p < 0.05)

Table SI-1.2. Tukey's HSD test results for control, Fe₃O₄NPs, and Ch-Fe₃O₄NPs groups

| Groups (I) | Groups (J) | Mean Difference (I-J) |
|---------------------------------------|---------------------------------------|-----------------------|
| Control group | Fe ₃ O ₄ NPs | 28.625* |
| | Ch-Fe ₃ O ₄ NPs | 86.325* |
| Fe ₃ O ₄ NPs | Control group | -28.625* |
| | Ch-Fe ₃ O ₄ NPs | 57.700* |
| Ch-Fe ₃ O ₄ NPs | Control group | -86.325* |
| | Fe ₃ O ₄ NPs | -57.700* |

* The mean difference is significant at the 0.05 level.

SI-2: Linearity assessment for iron quantification by atomic absorption spectrophotometry

Table SI-2.1 shows that the linear fit was acceptable for both concentration ranges. It also indicates that as iron concentration increases, confidence limits broaden; iron range between 3.000 and 5.000 mg L⁻¹ showed the highest UCL.

Table SI-2.1. Linear fit, upper, and lower confidence limits (UCL and LCL) for each iron concentration within the studied ranges

| Low Range | | | High Range | | |
|---|-------|--------|---|-------|--------|
| $y = x - 1 \cdot 10^{-4}$ R ² = 0.9996 | | | $y = x + 2 \cdot 10^{-4}$ R ² = 0.9982 | | |
| [Fe]std mg L ⁻¹ | UCL | LCL | [Fe]std mg L ⁻¹ | UCL | LCL |
| 0.000 | 0.030 | -0.030 | 0.000 | 0.235 | -0.234 |
| 0.125 | 0.159 | 0.090 | 1.000 | 1.424 | 0.576 |
| 0.250 | 0.289 | 0.211 | 2.000 | 2.614 | 1.387 |
| 0.500 | 0.549 | 0.451 | 3.000 | 3.803 | 2.197 |
| 1.000 | 1.068 | 0.932 | 4.000 | 4.993 | 3.007 |

| | | | | | |
|-------|-------|-------|-------|-------|-------|
| 1.500 | 1.587 | 1.413 | 5.000 | 6.182 | 3.818 |
|-------|-------|-------|-------|-------|-------|

SI-3: Determination of iron concentration in culture medium components

Components of the culture medium, which formed the basis of the Fe₃O₄NPs suspensions, are shown in Table SI-3.1. Most of the components had an iron content below the LOD, except the jelly; although the jelly proportion in the medium was very low, this was not an issue in the final culture medium.

Table SI-3.1. Iron concentrations in individual culture medium components

| Ingredient | [Fe] (mg kg⁻¹) |
|-------------------|----------------------------------|
| Banana | <LOD |
| Lemon | <LOD |
| Jelly | 17.6 ± 0.34 |
| Yeast | <LOD |
| Nipagin | <LOD |